ABSTRACTS

R1.1

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Effect of soluble *S. mutans* toxins on cultured human pulp derived (hpd) cells

Aim To investigate the influence of *Streptococcus mutans* toxins on proliferation and expression of genes known to be involved in anabolic hard tissue metabolism on cultured human pulp derived (hpd) cells.

Methodology Human pulp derived cells were obtained from two caries free impacted wisdom teeth from a female donor, aged 19 years. They were pre-cultivated in an incubator up to transfer phase (T) 2. In T3 they were seeded on glass sheets (Menzel, Germany) and transferred into a perfusion cell culture system (Minucells and Minutissue, Germany). Afterwards each of the two cell lines was divided into four groups. DMEM supplemented with 8% (v/v) foetal calf serum and 1% (v/v) penicillin-streptomycinsolution (Invitrogen, Germany) was used as base medium (bm). Group 1: 5% (v/v) of sterile filtrate of the centrifuged S. mutans suspension was added to bm. Group 2: 5% of sterile filtrate of ultrasonic treated and centrifuged S. mutans suspension was added to bm. Group 3: 5% of sterile Schaedler broth as bacterial medium control was added to bm. Group 4: bm without additives served as the cell culture medium control. Sampling of the hpd cells was carried out after the first, third and sixth week of cultivation. Analysis was undertaken by cell counting (Casy, Schärfe, Germany) and quantitative RT-PCR (iCycler, BioRad, Germany). Different noncollagenous proteins playing a role in biomineralization processes were quantitatively determined (Osteonectin, Osteopontin, Alkaline Phosphatase, Fibronectin 1, Bone Morphogenic Protein-1, and -3). Results No statistical significant differences could be found in the hpd cell counts in all four groups, nor in gene expression of the tested genes.

Conclusions The soluble *S. mutans* toxins used had no significant stimulating effect on hpd cells in connection with proliferation and expression of genes relevant for biomineralization.

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R1.2

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The evaluation of matrix metalloproteinase (MMP)-2 and -9 in acute and chronic periapical inflammation

Aim To evaluate the presence and levels of matrix metalloproteinase (MMP)-2 and -9 in acute and chronic periapical inflammation.

Methodology Twelve samples of root canal exudates were collected from teeth with clinically and radiographically verified acute or chronic periapical inflammation during root canal treatment. MMP-2 and MMP-9 levels were assayed by utilizing substrate gel zymography. MMP activity was visualized as clearance zones in the stained gels. The intensity of molecular forms of MMP-2 and MMP-9 were analysed by an image analysis system. Statistical analysis was performed using the Mann–Whitney *U* test at P = 0.05.

Results The root canal samples demonstrated marked differences in MMP-9 levels between acute and chronic periapical inflammation (P < 0.05). MMP-2 was not found in samples with chronic periapical inflammation. The levels of MMP-9 were significantly higher than the MMP-2 levels in samples with acute periapical inflammation (P < 0.05).

Conclusions These findings suggest that MMP-2 may have a specific role in acute periapical inflammation.

R1.3

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Effect of an antibacterial root canal cleanser (MTAD) on vascular responses in carotid artery tissue

Aim To investigate the effects of an antibacterial root canal cleanser (MTAD, Dentsply-Tulsa, OK, USA) on isolated rat carotid artery.

Methodology Following ethical committee approval, surgicallyremoved sections of the left carotid arteries were cut into transverse sections of 2–3 mm length, keeping the luminal surface intact. The rings (n = 5 per group) were suspended under 500 mg resting tension in a tissue bath containing Tyrode solution, gassed with 95% O₂ and 5% CO₂. The temperature of the bath (MAY STBSO5) was kept constant at 37°C and tissues were equilibrated for 30 min. About 300 and 500 μL of MTAD (doses determined after pilot tests) were applied on each ring and isometric muscle contractions were recorded to a PC using a variable force transducer (Biopac Inc., CA, USA). Between and after each measurement interval, the bathing medium was replaced with 15 mL tyrode solution. By the end of measurements, three different doses of epinephrine and papaverine were administered in the same manner and the contraction/relaxation forces were recorded, oneway ANOVA and t-tests were used for statistical analysis of contraction forces (P = 0.05).

Results Application of 500 μ L MTAD on the vessels resulted in a rapid collapse of the vessels after which MTAD-induced relaxation could not be recovered by subsequent epinephrine application. Application of 300 μ L MTAD caused relaxation in the smooth muscle, but recovery of tissue was confirmed by dose-dependent contractions and relaxations obtained following administration of adrenalin and papaverine, respectively.

Conclusions When forced inadvertently outside the root canal, MTAD can lead to a dose-dependent collapse in periapical vascular tissue. At higher dose, the collapse may be irreversible.

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R1.4

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Influence of an antibacterial root canal cleanser (MTAD) on compound nerve action potentials

Aim To investigate the possible neurotoxic effects of an antibacterial root canal cleanser (MTAD, Dentsply–Tulsa, OK, USA) on periapical nerve tissue, when forced inadvertently beyond the root apex.

Methodology Following ethical committee approval, rat sciatic nerves (n = 5/group) were isolated surgically and placed between two platinum electrodes in a bath containing Tyrode solution, which allowed for stimulation and recording of compound action potentials (CAP). Each nerve was repeatedly stimulated with an electrical stimulus (3 V amplitude, 0.05 ms pulse width). Raw data were processed digitally by amplifying with ×500 gain and 2 kHz low pass, Q = 0.070 filtering. Biopac LabPro V.3.7 software (Biopac Inc., CA, USA) was used in recording and analyzing the data. The minimal threshold required for evoking a compound action potential was recorded. The experiments were performed at supramaximal stimulation. The nerves were stimulated for at least 60 min prior to experiments. Thereafter, MTAD was brought into contact with the nerves and the evoked compound action potentials (CAP) were recorded.

Results The MTAD caused total inhibition of the CAP within an average time of 20 min. All CAPs in this group were blocked irreversibly.

Conclusions As a possible complication of root canal irrigation, long-term or permanent paraesthesia may be observed if MTAD is injected accidentally beyond the root apex.

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R1.5

Abstract Withdrawn.

R1.6

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Comparison of dentine matrix components extracted by the soluble products of white and grey MTA

Aim To compare the soluble products of white and grey mineral trioxide aggregate (MTA) and investigate the potentially bioactive dentine matrix proteins (DMPs) extracted from human dentine by these solutions.

Methodology Soluble products of set/setting ProRoot MTA (Tulsa Dentsply, USA) [white (pH 11.7) and grey (pH 11.7)], or 0.02M Ca(OH)2 (pH 11.9) were analyzed using Atomic Absorption Spectroscopy (AAS). Dissected powdered sound human dentine was subsequently treated with these solutions over a period of 14 days and solubilized DMPs were exhaustively dialyzed against water and lyophilized. The extracts were analyzed for the concentration of

non-collagenous proteins (NCPs) and glycosaminoglycans (GAGs), the diversity of which was assessed using 1D-polyacrylamide gel electrophoresis (1D-PAGE). The proteins extracted were further analyzed using sandwich ELISA for TGF- β 1 and adrenomedullin (ADM).

Results Comparable amounts of aluminium, calcium, potassium and sodium ions were liberated by both white and grey MTA solutions. NCPs and GAGs were found within the extracted DMP complex of all solutions. White and grey MTA extracted more NCPs and GAGs than Ca(OH)2. The 1D-PAGE analysis indicated that Ca(OH)2, white and grey MTA solubilized different DMP profiles. ELISAs demonstrated that there was significantly more TGF- β 1 in white MTA extracted DMPs than grey MTA extracts (P < 0.001) which had significantly more than calcium hydroxide extracted DMPs (P < 0.001). ADM ELISA demonstrated that grey MTA solubilized significantly more ADM than white MTA (P < 0.005). **Conclusions** This data suggests that although the soluble products of white and grey MTA are similar, these solutions release differential profiles of dentine matrix components which may have important clinical ramifications.

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R1.7

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The effect of MTA and rhBMP-2 on the mineralizing potential of human cementum-derived cells

Aim To investigate the effect of MTA on the mineralizing potential of human cementum-derived cells (HCDCs) and evaluate whether this effect is affected by the rhBMP-2.

Methodology Human cementum-derived cells used in this study were isolated and characterized as described previously and human osteoblastic bone marrow stromal cells (BMSCs, courtesy of Dr. Grzesik) were used for comparison. Cells were grown in DMEM/F12 media with 10% foetal bovine serum and antibiotics. Standard amounts of MTA (50 mg) was mixed under aseptic condition and placed on the bottom of 24-well tissue culture plates and allowed to set for 24 h in a humidified incubator. Culture plates without MTA were used as controls. About 20000 cells in culture medium were seeded into each well and incubated for 24 h and then the medium was switched to mineralizing medium with or without rhBMP-2 for 9 days. The supernatant was collected and subjected to sandwich ELISA technique to determine the concentration of osteocalcin. The cell layer was lysed with 0.1% SDS solution and alkaline phosphatase (ALPase) activity was assessed from cell lysate using p-nitrophenol phosphate.

Results In both cell groups, the concentrations of osteocalcin produced by the cells grown in the presence of MTA were comparable to the cells grown without MTA. However, when rhBMP was added, the levels of osteocalcin secreted by HCDCs were increased while an addition of rhBMP-2 to BMSCs group did not make any differences. HCDCs produced a lower level of ALPase activity in the presence of MTA than control cells while BMSCs produced similar levels of ALPase activity irrelevant to the presence of MTA. In both cell groups, rhBMP-2 slightly increased the levels of ALPase activity.

Conclusions The behaviour of HCDCs on MTA was different from that of BMSCs. The mineralizing potential of HCDCs was hindered by MTA and rhBMP-2 reversed this phenomenon.

R1.8

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Proliferation of pulp cells after direct pulp capping with a dentine adhesive *in vivo*

Aim To evaluate the proliferation of pulp cells 1, 3, and 7 days after direct pulp capping with the dentine adhesive Gluma Comfort Bond (GCB) in comparison to calcium hydroxide (CH) using light microscopy (LM).

Methodology In 72 caries-free maxillary molars (36 Wistar-rats) an occlusal cavity was prepared with a cylindrical diamond bur (ISO 008). Then the pulp chambers were perforated with a sharp probe and GCB (Heraeus, Germany) or CH was placed onto the exposed pulp tissue. The cavities were filled with composite (Tetric flow, Vivadent, Germany), successively. 1, 3, and 7 days after pulp capping the animals were sacrificed. For immunohistology 1 h prior to scarification 5-Bromo-2'-desoxyuridine (BrdU, Sigma-Aldrich, Germany) was injected into the intraperitoneal cavity. BrdU was incorporated into the cell nucleus during the S-phase of the cell cycle. Using an antibody staining (APAAP) proliferating cells could be marked and counted. Three animals (=6 molars) served as control and were not further treated. A statistical evaluation was performed using the nonparametric Mann-Whitney-U-test. For routine LM the crown pulp was histologically evaluated and separately recorded according to following scores: bacterial infection, inflammatory cells, and necrosis. The extent of the observed alterations were scored with 1 = absent, 2 = slight, 3 = moderate, 4 = profound. Statistical evaluation was performed using the ANOVA-test for non-parametrical data.

Results The marked cells were identified as fibroblasts, endothelial cells (after 1, 3, 7 days) and Höhl cells (after 7 days). After 1 day capping significantly more cells were stained in the GCB than in the CH group (P < 0.05). After 3 days significantly more cells were stained in the GCB than in the CH and the control group (P < 0.016). The scores for bacterial infection (after 1, 3, 7 days) and inflammatory cells (after 1 day and 7 days) were significantly higher for GCB than for CH (P < 0.05).

Conclusions Direct pulp capping with GCB led to significantly more granulation tissue due to bacterial infection and inflammation.

R1.9

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Procedural errors during root canal treatment performed by undergraduate students in Athens: a prospective study

Aim To detect procedural errors during root canal treatment in an undergraduate dental clinic.

Methodology A prospective study was undertaken over an 18-month period among patients who received root canal treatment by fifth year undergraduate students at the Dental School of Athens, Greece. All treatments were performed in two visits, using a step-back technique by means of hand stainless steel files. Iatrogenic errors that were detected included ledges, root perforations, furcation perforations, strip perforations and instrument separation. **Results** The final sample consisted of 323 teeth, that is 152 anterior (109 maxillary and 43 mandibular), 98 premolars (46 maxillary and 52 mandibular), and 73 molars (34 maxillary and 39 mandibular). Ledge formation was by far the most frequently encountered error. In total 52 teeth were ledged (16%), that is eight

anterior (15.4%), 17 premolars (32.7%) and 27 molars (51.9%). The frequency of ledged root canals was significantly greater in molars than in anterior teeth. In 23 teeth (8%) a root perforation was detected. The most common location was the apical third (82.6%), whereas only one (4.3%) was located at the middle third and three (13.1%) at the cervical third. Strip perforation occurred in one and furcation perforation in two cases. Instrument separation occurred in only two cases.

Conclusions Ledge formation, especially in molars and premolars, as well as perforations of the apical third of the root was the most common procedural errors during root canal treatment. Their relatively high incidence imply that modifications to the endodontic undergraduate curriculum might be necessary, in order to achieve a better quality of root canal treatment.

R1.10

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Radiographic evaluation of root canal fillings performed by undergraduate students at the University of Athens

Aim To evaluate radiographically the quality of root canal fillings performed by undergraduate students at the Dental School of Athens.

Methodology All root canal treatments completed by fifth year undergraduates over a 12-month period were included in the study. At least three periapical radiographs (pre-operative, working length determination and post-operative) were available for each root filled tooth examined. All treatments were performed in two visits, using a step-back technique by means of hand stainless steel files. The quality of root fillings was evaluated regarding (a) the length in relation to the radiographic apex and (b) the presence of voids laterally. Moreover, the presence of ledge formations was also recorded. Statistical evaluation was performed using random effects logistic regression models.

Results The final sample consisted of 258 root filled teeth with 392 root canals in 154 patients. The length of the root filling was acceptable (0–2 mm from the apex) in 299 (76.3%) root canals, short (>2 mm from the apex) in 53 (13.5%) and overfilled in 40 (10.2%) root canals. About 77% of the root filled canals had no voids laterally. Both acceptable length and no voids laterally were recorded in 236 (60.2%) root canals whereas, only 27 (6.9%) of the root canals treated presented unacceptable length (either short or over-filled) together with voids. Molar teeth had significantly poorer quality of root fillings as well as 47% greater possibility of a ledge being present than the other tooth types.

Conclusions The quality of root fillings performed by undergraduate students at the Dental School of Athens was better than the technical standard of root fillings reported by other dental clinics.

R1.11

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Technical quality of root fillings completed by undergraduate students

Aim To assess radiographically the technical quality of root fillings performed by undergraduate students in Riga Stradins University, Riga, Latvia.

Methodology All root canal treatments completed by second clinical year undergraduate students during the period 2003–2004 were included in the study. A total of 210 root-filled teeth with 407 roots were evaluated: 43 undergraduate students were involved in the treatment of these cases. The two examiners were calibrated for radiographic quality of root fillings according to criteria modified from Petersson and co-workers (Petersson *et al.* 1986). Inter-examiner agreement was measured by Cohen's kappa (k) values. Teeth with readable post-operative periapical radiographs that had received primary treatment were examined for quality of root fillings, and categorized as complete, incomplete apical, incomplete lateral, incomplete apical and lateral or not assessable. Data were entered into an Excel databases and statistical tests were carried out using an SPSS statistical package.

Results The k-value for inter-examiner variability was 0.6. Of the 210 teeth four were re-treatments, seven had no post-operative radiograph and 14 were not assessable in one or more areas; these teeth were excluded from the statistical analysis. Of the remaining 185 teeth, 49 were anterior teeth, 71 were premolars and 65 were molars. Of the 389 root canals, 287 (74%) were filled completely, with 102 (26%) having voids either apically and/or laterally.

Conclusions The technical quality of root canal treatment performed by undergraduate dental students was acceptable in 74% of cases. This demonstrates a substantial improvement when compared with a previous report from Riga Stradins University (Kundzina *et al.* 2003) that included the study period 1996–1999.

R1.12

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Outcome of root canal treatment performed by undergraduate students at Riga Stradins University, Riga, Latvia

Aim To assess outcome of root canal treatment performed by undergraduate students.

Methodology A total of 108 patients received 137 root canal treatments at the Department of Conservative Dentistry between September 2002 and May 2003. Before commencing the study two examiners were calibrated for the PAI index (periapical condition) and Ryge criteria (restoration quality). All 108 patients were recalled after 3 to 4 years when post-operative periapical radiographs were taken of all root filled teeth. During the recall patients were examined for clinical signs and symptoms, quality of existing restorations, technical quality of root fillings (modified from Petersson *et al.* 1986), periapical condition (PAI index) and radiographic quality of restorations. Apical periodontitis was recorded if the PAI scores were 3, 4 or 5. Data were analyzed using SPSS 11 computer software program.

Results Sixty-two patients with 78 root-filled teeth (57%) did not attend the follow-up investigation: seven were unwell, 11 refused to attend and 44 could not be contacted. In total 59 teeth were examined (recall rate 43%). Three teeth had been extracted because of periodontal disease and were excluded from the statistical analysis. A healthy periapical region (91% for all teeth) was found significantly more often associated with teeth treated without initial apical periodontitis (95%) than with initial apical periodontitis (77%). Acceptable radiographic quality of root canal fillings was noted in 75% of cases. Acceptable clinical and radiographic quality of restorations was 86%. There were no significant correlations between restoration type or quality and PAI score or between quality of root filling and PAI score.

Conclusions A healthy outcome following root canal treatment by undergraduate students in the Department of Riga Stradins University occurred in 91% of teeth examined after 3 to 4 years. Pre-operative apical periodontitis had a negative impact on the outcome.

R1.13

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Requirements in terms of root canal treatment of undergraduates in the European Union

Aim To gather information from across the European Union about current requirements of undergraduates in the area of root canal treatment.

Methodology An online survey was created and representatives at 192 EU dental schools were emailed and asked to complete the survey. The survey contained both open and closed format questions and it inquired about how root canal treatment is assessed, whether students have to complete a minimum number of root canal treatments, whether they have to be in both anterior and posterior teeth, and if experience of root canal retreatment and endodontic surgery are compulsory for undergraduates. The final question asked how competence in root canal treatment can be described.

Results Responses came from 44 dental schools in 19 countries. Results showed wide variation in assessment methods for root canal treatment. Overall, 95% of the sample schools insist on students performing root canal treatment in both anterior and posterior teeth. Eighty percent ensure their students perform a minimum number of root filled teeth with a variation in what that minimum number is. Eighty-four percent require undergraduates to perform retreatment cases, but just 5% said endodontic surgery is a requirement. Descriptions of competence in root canal treatment varied greatly, though some common themes were present.

Conclusions Despite convergence of the undergraduate curriculum being endorsed by many and movement of dentists within the EU escalating, there is still inconsistency in the output of dental schools with regard to root canal treatment. This sample demonstrated diversity in assessment methods, requirements and deviation from the ESE 2001 Undergraduate Curriculum Guidelines for Endodontology.

R1.14

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The prevalence and management of instrument fracture by postgraduate residents: a retrospective study

Aim To investigate the prevalence and management of instrument fracture during root canal instrumentation by postgraduate residents and to determine the percentage of endodontic cases referred to and managed by postgraduate residents of the Endodontic program with separated instruments.

Methodology A retrospective study was conducted by reviewing the dental notes of 1367 patients (2180 endodontic cases – 4897 root canals) treated between October 2001 and June 2006 by endodontic postgraduate residents at the Dental School of Athens. Type of tooth and canal, type and length of fractured segments, level of instrument fracture and management that followed were recorded.

Results The prevalence of instrument fracture during root canal preparation was 1.83%. However, the prevalence of endodontic cases with separated instruments referred to the Endodontic postgraduates was 4.31%; this difference was statistically significant (P < 0.001). The prevalence of hand and rotary NiTi instrument separation by postgraduate residents were 0.55% and 1.28%, respectively (P < 0.001). The prevalence of instruments fractured in the apical third (52.5%) was significantly higher when compared with coronal (12.5%) and middle (27.5%) thirds of the canals. The retrieval or by-pass of fractured instruments was the most successful in the coronal (100%) and middle (45.4%) thirds when compared with the apical third (37.5%) of canals.

Conclusions (1) The prevalence of endodontic instrument fracture by the postgraduate residents was relatively low when compared with the prevalence of fractured instruments in endodontic cases referred for management to the endodontic residents. (2) The prevalence of fracture of rotary NiTi instruments was more frequent than that of hand instruments. (3) Retrieval or by-pass of the fractured instrument in the apical third was less successful.

R1.15

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Radiological assessment of endodontic treatment needs in an adult population in the Lodz Region of Poland

Aim To radiographically evaluate endodontic treatment needs in an adult population in the Lodz Region of Poland.

Methodology A total of 439 panoramic radiographs from patients (18–86 years), reporting for the first time to the Institute of Dentistry, Medical University of Lodz, were evaluated. The radiographs were randomly selected amongst patients admitted to the Institute within one calendar year. The following dental treatment needs were considered: 1) root canal treatment of non-root filled teeth with periapical pathosis or with visible carious cavities reaching the pulp cavity; 2) root canal treatment of root filled teeth with periapical pathosis; 3) extraction of teeth not suitable for root canal treatment. Patients with less than 10 remaining teeth were excluded from the study.

Results Amongst the teeth with endodontic treatment needs, 74.4% required root canal treatment and 25.6% extraction (*P* < 0.0005). Endodontic treatment needs were more frequently observed in root filled (86.8%) than non-root filled teeth (57.0%) (*P* < 0.00005), whereas extraction was more often necessary in non-root filled (43.0%) than in root filled teeth (13.2%) (*P* < 0.00005). Necessity of endodontic retreatment was more frequently reported in multi-rooted than in single-rooted teeth (74.3% versus 58.6%) (*P* = 0.0003), while single-rooted teeth (41.4%) more frequently required endodontic treatment than multi-rooted (25.7%) (*P* = 0.0003). More than two-thirds of endodontic treatment needs concerned teeth that were root filled and one-third related to teeth that were to be treated for the first time.

Conclusions Amongst the population of the Lodz Region, endodontic treatment needs result predominantly from the necessity to retreat teeth with inadequately filled root canals and periapical pathosis. Improvement in the quality of root canal treatment may reduce patients' needs in this field.

R1.16

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Prevalence of apical periodontitis and quality of endodontic treatment in the Slovenian population

Aim To determine radiographically the prevalence of apical periodontitis (AP) as well as the prevalence and quality of root fillings in an adult Slovenian population.

Methodology A total of 217 orthopantomograms of adult patients attending general practice for the first time were examined. The number of teeth, number of root filled teeth (RFT), and number of teeth with AP were recorded. The adequacy of root canal treatment was determined using the guidelines of the European Society of Endodontology. The type of coronal restoration was recorded. Relations between the apical status, presence, and quality of root fillings were statistically evaluated using the chi-square test.

Results Overall, 78% of the patients examined (aged 38.8 ± 11.0 years) had RFT and 80% had at least one tooth with AP. Of the 5710 teeth examined, 7.8% were root filled. AP was found in 8.4% of all teeth. In 45% of RFT, the filling ended within 2 mm from the radiographic apex, 50% were underfilled, 5% were overfilled, and 47% were homogenous. Significantly more AP was observed in RFT as compared with non-root filled teeth (55% versus 4%; P < 0.001). Adequately filled teeth showed significantly less AP than those inadequately filled (23% versus 69%; P < 0.001). RFT were restored with plastic materials in 54%, post and crown in 29%, and crown in 13%; 5% had no restoration.

Conclusions The prevalence of AP and RFT were similar to those reported in previous methodologically comparable European studies. AP was more likely in RFT. The presence of an adequate root filling had a positive impact on the apical status.

R1.17

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Periapical health of an urban population in a recent historic period in the region of Slavonia, Croatia

Aim To evaluate the periapical health of an archaeological sample from a recent historic period, found in the cathedral of the town of Pozega, Slavonia, Croatia.

Methodology Forty well preserved skulls excavated from the archaeological findings in the church crypt were examined radiographically. According to the church records, those individuals were buried between 1760 and 1868. Their age was determined by the Lovejoy method and their gender by their craniofacial osseal characteristics. A total of 960 teeth present in their skulls were analyzed, with an additional 116 teeth which were lost after death and 204 lost before death. Caries was assessed using a dental explorer and radiographically. Periapical health was evaluated by the presence of periapical radiolucency, root resorption or condensing osteitis on panoramic radiographs. The Periapical index (PAI) was used to determine the presence of periapical periodontitis. Any score equal to or greater than three was categorized as a diseased periapex. Root resorption was established by the radiographic appearance of the roots by comparing their length to adjacent teeth. Condensing osteitis was identified as a radiopacity in the periapical area.

Results Caries was found in 112 teeth (12%). Periapical pathosis was found in 49 (5%) teeth, of which 44 (4.5%) had periapical periodontitis and five (0.5%) had root resorption. Condensing osteitis was found in the periapical region of five teeth. Periapical pathological changes were found in 20 (50%) skulls, of which, 74% were associated with caries.

Conclusions Periapical periodontitis was a common disease in the urban Slavonian population of a recent historic period. Most of the periapical periodontitis was associated with caries.

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R1.18

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Interappointment flare-ups: incidence and related factors

Aim To evaluate the incidence of interappointment flare-ups and to examine their possible correlation to several clinical factors.

Methodology A prospective study was set-up over an 18-month period among patients who received root canal treatment by fifth year undergraduate students at the Dental School of Athens, Greece. All treatments were performed in two visits, using a step back technique. Flare-ups were defined as the occurrence of severe pain and/or swelling following cleaning and shaping, requiring an unscheduled visit and active treatment. The following factors were recorded: age, gender, tooth group, arch, pulp vitality, presence of periapical lesion, as well as treatment factors (initial treatment or retreatment).

Results The final sample consisted of 323 teeth in 150 patients, 69 males and 81 females, 9–73 and 13–73-years-old respectively. The overall incidence of flare-ups was 5.9%, including seven anterior teeth (six maxillary, one mandibular), five mandibular premolars and 7 molars (two maxillary, five mandibular). Flare-ups occurred in eight male patients, mainly between 50 and 59 years, and 10 females mainly between 40 and 49 years. Pulp diagnosis was non-vital in 17 cases (89.5%) and irreversible pulpitis in two cases of multi-rooted teeth. In 14 cases the treated teeth were related to periapical pathosis (73.7%). Root canal treatment was completed for the first time in 11 cases (57.9%), whereas the remaining eight were retreatment cases.

Conclusions The incidence of interappointment flare-ups is relatively low. Their occurrence was related to pulp necrosis and presence of periapical pathosis.

R1.19

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Effect of the digital image display monitor on the evaluation of the quality of root fillings

Aim To compare a liquid crystal display (LCD) monitor with a high resolution cathode ray tube (CRT) monitor for the evaluation of length and homogeneity of root fillings.

Methodology Root canals of 17 extracted permanent mandibular incisors were filled. With the teeth placed in their jaws radiographic images were obtained with the Accu-I-Tomo Limited Cone Beam CT (LCBCT) and Digora Optime image plate systems (SPP). Six observers rated the quality of all images displayed on the two monitor types in terms of their ability to evaluate root filling quality. Results were compared using McNemar's test (P < 0.05). Cochran's Q was used to determine the reliability among the raters' scores. Differences among LCBCT and SPP images were determined by binomial test. Kappa was used to measure inter-observer agreement.

Results No significant difference was found between image quality ratings (P > 0.05) for images displayed on CRT and LCD monitors. However, in the LCD images the inter-observer agreement was significantly better (P < 0.05). SPP and LCBCT images were rated significantly different (P < 0.05) with the former being rated highest.

Conclusions The quality of LCD images was similar to that of CRT images with respect to evaluating length and homogeneity of root canal fillings in mandibular incisors. Images from the SPP system were significantly better.

R1.20

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Accuracy of limited cone-beam CT, storage phosphor and conventional film radiography for the detection of periapical lesions

Aim To compare the diagnostic accuracy of Accu-I-Tomo (Morita Co Ltd, Tokyo, Japan), Digora Optime (Soredex Corporation, Helsinki, Finland) image plate system and F-speed film (Eastman Kodak, Rochester, NY, USA) for the detection of chemically created periapical lesions in dry skulls.

Methodology Lesions were created by placing a solution of 70% perchloric acid at the bottom of tooth sockets of extracted teeth in 12 human jaw specimens. The acid was applied in three time periods (1 h, 1.5 h and 2 h). Teeth were replaced in their sockets and radiographed with three imaging modalities - Accu-I-Tomo limited cone-beam CT (LCBCT), Digora Optime storage phosphor plate (SPP) system and F-speed film. Six observers scored the images for the presence/absence of lesions using a 5-graded scale. Diagnostic accuracy, expressed as the area under the ROC curve (Az), for each observer and modality was compared using Kruskall-Wallis analysis. Kappa (k) was used to measure inter-observer agreement.

Results Significantly greater accuracy was noted for LCBCT images (Az = 0.862) compared with both SPP (Az = 0.752) and F-speed film (Az = 0.726) for all acid durations. Pair-wise comparisons showed significant differences between LCBCT and film images, and LCBCT and SPP images, respectively, only for 1.5 h of acid application. k ranged between fair to moderate for LCBCT and between slight to fair for SPP and film.

Conclusions Overall, in LCBCT images, observers were better in detecting chemically induced apical lesions. However, the detectability was similar for all imaging modes for shorter (smaller lesions) and longer (larger lesions) acid applications.

R1.21

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Absolute and relative quantification of known and novel endodontic pathogens

Aim To quantify selected endodontic pathogens such as *Treponema* spp., *Prevotella intermedia* and *Porphyromonas gingivalis* as well as recently reported novel endodontic phylotypes from the bacterial division *Synergistes* and from the domain *Archaea*.

Methodology Thirty-two necrotic teeth with radiographic evidence of apical periodontitis and primary endodontic infections were analysed using real-time quantitative PCR targeting taxaspecific 16S rRNA genes regions.

Results Eight of the 32 cases were found to be positive for *Archaea* and seven for *Synergistes*. In addition *Treponema* was found in 18 cases, while *P. intermedia* and *P. gingivalis* were found in five and nine cases, respectively. The size of the *Archaea* population ranged from $1.3 \times E5$ to $6.8 \times E5$; and of *Synergistes* from $4.5 \times E4$ to $1.5 \times E6$ 16S rRNA gene target molecules. In contrast, the range of *Treponema* was higher, ranging from $2.9 \times E4$ to $7.8 \times E7$ target molecules. The median proportion of *Archaea*, *Synergistes* and *Treponema* relative to the total bacterial community (determined by a universal 16S rDNA-based RTQ PCR assay) was comparable with 0.87%, 0.79%, and 1.48%, respectively. However, the median proportion of *P. intermedia* and *P. gingivalis* was substantially lower (below 0.01%).

Conclusions The prevalence and quantity of *Archaea* and *Syner*gistes was within the range of the other pathogens analyzed, suggesting their clinical relevance in endodontic infections.

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R1.22

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Bacteria associated with post treatment disease and the antimicrobial susceptibility of *E. faecalis* isolated from the filled root canals

Aim To (i) detect the presence of *Enterococcus* spp., *Staphylococcus* spp., *Candida* spp. and Enterobacteria in saliva, (ii) to identify the microorganisms around the coronal restoration and in the canals of 30 root filled teeth with periradicular pathosis and (iii) to test the antibiotic susceptibility of *E. faecalis* isolated from the root filled canals.

Methodology Transport media, specific culture media and adequate gaseous requirements were used to isolate as many strict and facultative anaerobes as possible. Selected media were used to isolate *Enterococcus* spp., Enterobacteria and yeasts. Antibiotic susceptibility of *Enterococcus faecalis* isolates was accomplished with the *E*-test system.

Results From the 114 species of microorganisms isolated from the root canals. 81.5% were facultative anaerobes and 18.5% strict

anaerobes, 86% were Gram-positive and 14% Gram-negative bacteria. The most frequently recovered bacterial genera from the root canals were: *Staphylococcus* (15/30), *Streptococcus* (11/30), *Actinomyces* (12/30), *Enterococcus*, *Gemella* (8/30), *Propionibacterium*, *Clostridium* (4/30), *Peptostreptococcus*, *Bifidobacterium*, *Eubacterium* (3/30), *Lactobacillus*, *Prevotella* (2/30) and *Candida* (1/30). *Enterococcus* spp., *Staphylococcus* spp., *ad* Enterobacteria were isolated in 26.7%, 50%, 3.3% and 16.7% of the canals, in 20%, 40%, 6.7% and 13.3% of the crown and in 43.3%, 56.7%, 50% and 26.7% of saliva, respectively. *Enterococcus faecalis* (n = 8) were 100% susceptible to amoxicillin combined with clavulanate and to gentamicin; 87.5% to amoxicillin and moxifloxacin; and 60% to ciprofloxacin. However, 100% of the *E. faecalis* isolates were resistant to clindamycin and metronidazol; 87.5% to azithromycin and rifampicin.

Conclusions *Enterococcus* spp., *Staphylococcus* spp., *Candida* spp. and Enterobacteria could be detected in the saliva, crowns and the root canals of the same patients. The effective antibiotics for *E. faecalis* were amoxicillin combined with clavulanate and gentamicin.

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R1.23

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Microbial profile before and after chemo-mechanical preparation of root canals of teeth with endodontic and periodontal diseases

Aim To investigate microbiologically whether the chemo-mechanical preparation of infected root canals with adjacent periodontal pockets would affect the periodontal microbial population.

Methodology Fifteen teeth with endodontic and periodontal diseases were evaluated. Samples were taken before and after chemo-mechanical preparation of the root canals. Transport medium, culture media and adequate gaseous requirements favouring the growth of strict anaerobic microorganisms were used. Colony forming (CFU/mL) were counted and the microorganisms identified by biochemical tests.

Results Initially, the microorganisms recovered from the root canals ranged from 10 to 1.7×10^7 CFU/mL; while in the periodontal pockets they ranged from 4.0×10^5 to 6.0×10^8 CFU/mL. The microorganisms most frequently found in the root canals were *Actinomyces meyeri* and *Fusobacterium nucleatum*, while in the periodontal pockets were *Prevotella intermedia* and *Fusobacterium nucleatum*. Post-chemo-mechanical preparation samples showed approximately 100% reduction in cultivable bacteria in the root canals but no reduction in the periodontal pockets.

Conclusions Chemo-mechanical preparation reduced the number of microorganisms in the root canal; however, it did not affect immediately the periodontal population in the adjacent pockets.

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R1.24

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Relationship of pain of endodontic origin with the presence of microorganisms and endotoxins in infected root canals and the antimicrobial susceptibility of selected anaerobic bacteria

Aim To analyze the microbiota isolated from infected root canals of symptomatic or asymptomatic teeth, to quantify the presence of endotoxins in infected root canals; to correlate the presence of specific bacteria and the amount of endotoxins with endodontic symptomatology; and to investigate the antibiotic susceptibility of anaerobic bacteria isolated from infected teeth with periapical lesions against eight antibiotics through the *E*-test.

Methodology Microbial samples were taken from 90 root canals of teeth with necrotic dental pulps, and analysed using standard culture procedures. A further 50 samples were collected from infected symptomatic or asymptomatic root canals and analysed using a chromogenic test for quantification of endotoxin. Statistical analysis included the use of Pearson ×2 test and a one-sided Fisher's Exact test, as appropriate.

Results A total of 400 cultivable isolates were recovered from 69 different microbial species and 22 different genera. Root canals from symptomatic teeth harboured more obligate anaerobes and a larger number of bacterial species than the asymptomatic teeth. More than 80% of the total bacterial isolates were strict anaerobes. Relationships were found between specific microorganisms, especially gram-negative anaerobes and the presence of pain or history of pain, tenderness to percussion, pain to palpation and swelling. High concentrations of endotoxins were found in root canals of symptomatic teeth and there was a positive correlation between endodontic signs and symptoms and the concentration of endotoxins in infected root canals. Amoxicillin, amoxicillin + clavulanate and cephaclor were effective against all the strains tested.

Conclusions Specific bacteria and endotoxins were associated with endodontic symptoms from infected teeth. The majority of the anaerobic species tested were susceptible to all antibiotics studied. **Acknowledgements** This work was supported by the Brazilian agencies FAPESP, CNPq and CAPES.

R1.25

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Detection of pathogenic microorganisms in primary endodontic infections using a polymerase chain reaction assay

Aim To evaluate the prevalence of 15 bacteria in primary endodontic infections using a polymerase chain reaction (PCR) assay. **Methodology** Specimens were selected from adult patients (ages ranging from 18–60 years) who had been referred for root canal treatment to the Department of Endodontics, Faculty of Dentistry, Ankara University, Ankara, Turkey. Samples were obtained from 62 teeth having carious lesions, necrotic pulps and different forms of periradicular diseases. Samples were then transferred to cryotubes containing TSB-DMSO and frozen at -20° C. DNA was extracted from the samples, and a 16S rDNA-based polymerase chain reaction assay was used to identify target microbial species. **Results** Microorganisms detected in 62 cases were as follows: *P. gingivalis* 62.9%, *Strep.* spp. 53.2%, *P. endodontalis* 53.2%, *M. timidum* 45.1%, *S. exigua* 30.6%, *B. forsythus* 25.8%, *T. denticola* 25.8%, *P nigrescens* 24.1%, *P. micros* 19.3%, *A. actinomycetemcomitans* 19.3 %, *F. nucleatum* 12.9%, *A. israeli* 9.6%, *E. Saphenum* 6.4%, *P. intermedia* 4.8%.

Conclusions PCR technology appears to be a valuable, rapid and reliable method for the detection of anaerobic microorganisms.

R1.26

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Prevalence and elimination of microorganisms in root filled teeth associated with periradicular lesions

Aim To isolate and identify microorganisms in root filled teeth associated with periradicular lesions and to assess a disinfecting regimen including the use of NaOCl, EDTA and CHX irrigation and Ca(OH)2 dressing.

Methodology After removal of the root filling material, specimens of twenty cases undergoing retreatment were sampled. Moreover, the canals were sampled after each step of the disinfecting regimen. Serial dilutions were plated on Columbia Blood Agar (CBA) and on Yeast-Cystein Blood Agar (HCB). Bile Esculin Agar was used to cultivate enterococci. The bacteria were identified by morphological and biochemical analysis. Additionally, identification by PCR and 16S rRNA-gene sequencing was applied.

Results Prevalence of microorganisms was 60% by culture (12 of 20 teeth) and 65% by PCR (13 of 20 teeth). In four of those samples (31%) DNA of *Enterococcus faecalis* was found. The colony forming units/mL (CFU/mL) was in the range of 10^3 – 10^7 on CBA and of 10^3 – 10^9 on HCB, respectively. Species of *Actinomyces, Atopobium, Bacteroides, Bulleidia, Campylobacter, Dialister, Enterococcus, Flexistipes, Fusobacterium, Klebsiella, Megasphaera, Olsenella, Peptostreptococcus, Vagococcus and Veillonella were detected. In samples of two patients <i>E. faecalis* was the only species isolated. After further root canal preparation and irrigation using NaOCl and EDTA microorganisms could not be detected in any of the teeth. Thus, CHX and Ca(OH)2 did not show further disinfection. In contrast, microorganisms were found in two teeth after the interappointment dressing.

Conclusions Microorganisms were not isolated from all teeth. The majority of positive samples revealed a mixed culture of two to nine species. *E. faecalis* was the only detected species in two patients underlining its suggested important role in endodontic infections. It may be concluded that proper root canal preparation and irrigation using NaOCl and EDTA are sufficient for decontamination of the root canal system during root canal retreatment.

R1.27

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Clinical efficacy of bioactive glass S53P4 and calcium hydroxide in root canal and dentine disinfection

Aim To compare bioactive glass S53P4 (BAG (Vivoxid Ltd., Turku, Finland)) as an interappointment dressing to conventional calcium hydroxide.

Methodology Twelve generally healthy subjects with apical periodontitis were treated. The baseline microbial growth was examined with a microbiological sample taken immediately after access to the root canals. After chemomechanical preparation with 0.5% sodium hypochlorite, the subjects were randomly allocated into the test group (BAG, n = 6) and the control group (calcium hydroxide, n = 6) according to the interappointment dressing used. After 14 days, the canals were cleansed and left empty but sealed for 7 days prior to the second microbiological sampling. Rubber dam and a strict aseptic technique were applied in all clinical procedures. The samples were analysed using conventional clinical laboratory culture and identification methods. Bacterial growth in the samples was judged as negative, sparse or moderate/heavy.

Results All teeth had moderate/heavy bacterial growth at baseline. After treatment, bacterial growth was still detected in five (two sparse, three moderate/heavy) of the six samples taken from the test group, and in four (all sparse) out of six samples from the control group.

Conclusions The number of subjects was small and statistically significant differences were not found. However, this study did not indicate a better clinical efficacy of BAG S53P4 in comparison to calcium hydroxide.

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R1.28

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Adherence and biofilm formation affect susceptibility to calcium hydroxide

Aim To compare in vitro the susceptibility to calcium hydroxide of six microbial species in planktonic suspensions, as adherent monospecies cells or as established multi-species biofilm on dentine. Methodology Enterococcus faecalis ATCC29212, Streptococcus sobrinus OMZ176, Sreptococcus oralis OMZ607 and Candida albicans ATCC90028; and Actinomyces naeslundii ATCC12104 and Fusobacterium nucleatum ATCC10953 were grown according to the specific requirements of each species. Individual cultures were used as planktonic cells, or allowed to adhere on dentine for 24 h; mixed cultures were used to form a multispecies biofilm under anaerobic conditions with a serum-enriched nutrient supply at 37°C for 5 days. Conventional direct exposure test to saturated aqueous calcium hydroxide solution (pH 12.5) for 10 and 100 min was applied. In addition, planktonic microorganisms were also exposed to calcium hydroxide solution with an adjusted pH of 12.2. Physiologic saline served as control. After exposure the cells were harvested, suspensions diluted and cultured. Colonies were counted and the survival rates were analyzed by one-way ANOVA. Each experiment was repeated five times.

Results In general, planktonic microorganisms were most susceptible, only *E. faecalis* and *C. albicans* survived in saturated solution for 10 min, the latter also for 100 min. The pH adjustment to 12.2 significantly improved the survival of each species. In comparison to planktonic suspensions *E. faecalis* and *A. naeslundii* were significantly more resistant as adherent cells. The highest resistance of each species was observed in multispecies biofilms. *E. faecalis* demonstrated significant differences between each test method. Time dependency and dose (pH) dependency (planktonic microorganims) were found throughout the study.

Conclusions Adherence on dentine and interactions between microbial species in a biofilm appear to modulate their sensitivity to calcium hydroxide.

R1.29

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Antibacterial activity of new root canal filling materials against *Enterococcus faecalis*

Aim To investigate the antimicrobial activity against *Enterococcus faecalis* of new root canal sealers, Epiphany (Pentron, USA) used with Resilon, EndoREZ (Ultradent, USA), RC Sealer (Sun Medical, Japan), Acroseal (Septodont, France), GuttaFlow (Coltène/Whaledent, Germany) in comparison with established sealers AH Plus (DeTrey Dentsply, Germany), Apexit (Vivadent, Liechtenstein) and RoekoSeal (Coltène/Whaledent, Germany), used with conventional gutta-percha.

Methodology Filter paper discs were immersed in spectrophotometrically standardized *E. faecalis* suspensions ($OD_{540} = 0.400$) and exposed to freshly mixed sealers and standardized gutta-percha specimens inserted in teflon wells for 30 min, without or with a filter membrane (Whatman, UK, 0.22 µm pore size) placed between filter paper discs and sealers (direct contact and membrane restricted contact test, respectively). Activ Point, Resilon, and Hygenic gutta-percha were also tested in the absence of sealers. After exposure, the filter paper discs were transferred to vials containing phosphate-buffered saline and glass beads, and vortexed vigorously. About 25 µL droplets from serial dilutions were seeded on tryptone soy agar plates. The plates were incubated ($37^{\circ}C$, 24 h) and colony-forming units were counted. The data obtained were analyzed using Student's *t*-test (P < 0.05).

Results In the direct contact test, the materials displayed antibacterial activity in the following, decreasing order: RC Sealer, AH Plus, Epiphany, Acroseal, EndoREZ, Hygenic gutta-percha, Activ Point, GuttaFlow, RoekoSeal, Resilon, Apexit. RC Sealer was significantly more active than Epiphany and the materials with less average activity. In the membrane-restricted test, the materials displayed antibacterial activity in the following, decreasing order: RC Sealer, AH Plus, RoekoSeal, Hygenic gutta-percha, Resilon, Activ Point, EndoREZ, GuttaFlow, Epiphany, Acroseal, and Apexit. RC Sealer was significantly more potent than AH Plus and the materials with less average activity.

Conclusions RC Sealer and AH Plus sealers were most and Apexit was least effective in reducing *E. faecalis*. Activity of Epiphany was reduced when the *E. faecalis* cells were separated by a membrane. **Acknowledgements** The financial support of the Research Council of Norway and NIOM-Nordic Institute of Dental Materials is gratefully acknowledged.

R1.30

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Effect of different vehicles and barium sulfate on pH and calcium-ion release of calcium hydroxide

Aim To evaluate the effect of different vehicles and barium sulfate on pH and calcium-ion release of calcium hydroxide.

Methodology Calcium hydroxide mixtures were prepared with either bidistilled water (Group 1), chlorhexidine (Group 2), physiological saline solution (Group 3), propylene glycol (Group 4) or bidistilled water and barium sulfate added as radiopacifier (1 : 8, by weight, added to calcium hydroxide powder) (Group 5). The materials were placed in polyethylene tubes (length 1 cm, diameter 4 mm). The tubes were then immersed in a glass flask containing 10 mL bidistilled water (n = 10), which was sealed hermetically to eliminate the effects of environmental factors until the measurement time and stored at 37°C. The control group contained bidistilled water with empty tubes (n = 10). At pre-determined time intervals (24 h, 96 h, and 7, 10, 14, and 28 days) the pH of the bidistilled water was evaluated with a pH meter and released calcium assessed using spectrophotometry. Statistical analysis was performed by ANOVA for each time interval studied; individual comparisons were performed using Turkey HSD Test.

Results There was no significant difference in the pH and calcium ion release of different mixtures at 24 h. Addition of barium sulfate decreased significantly the pH level and calcium ion release of calcium hydroxide in the other test periods (P < 0.05). The use of different vehicles had no significant effect on the pH levels and calcium ion release of different calcium hydroxide mixtures (P > 0.05).

Conclusions Barium sulfate had an adverse effect on pH level and calcium ion release of calcium hydroxide mixtures. Further studies should evaluate the optimum amount of barium sulfate that does not compromise pH levels or calcium ion release.

R1.31

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Association of serum IgA, IgG levels and presence of *Treponema denticola* with human periapical lesions

Aim To compare serum immunoglobulin A (IgA), immunoglobulin G (IgG) levels and the presence of *Treponema denticola* in the root canals of a group of teeth with/without periapical radiolucency.

Methodology A total of 66 children aged 8–13 years with endodontic treatment need were involved in this study. Ethical approval was granted by Ege University (no. 05–11.2/8). About 5 mL of blood were collected at the beginning of the treatment to detect the serum IgA and IgG levels using a nephalometric technique. Sixty-six root canal samples from different teeth were also obtained to determine the presence of *Treponema denticola* using a polymerase chain reaction method.

Results The difference between the serum IgG, IgA levels and periapical lesion was not statistically significant (P > 0.05). The

presence of *Treponema denticola* in the groups with periapical lesions and without was significantly different (P = 0.026). The correlation between serum IgG, IgA levels and the presence of *Treponema denticola* was also significant (P = 0.023 and 0.038, respectively). **Conclusions** This study tends to support the hypothesis that the presence of *Treponema denticola* in root canals is primarily related with periapical lesions; the higher levels of serum IgG and IgA levels may provide protection against *Treponema denticola*.

R1.32

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Influence of an induced apical periodontitis on the level of systemic inflammation markers - a pilot study

Aim To examine the occurrence of systemic inflammatory response during chronic apical periodontitis in a rat model.

Methodology Dental pulps in six male Dark Agouti rats (four as control) without a history of inflammatory conditions were exposed and auto-infected to induce chronic apical periodontitis. Two blood samples were drawn 1 week pre-operatively (first time period) and 8 weeks (second time period) post-infection. Relative gene expression was assessed by measuring the level of Gamma interferon (IFN- γ) Interleukin-1 β (IL-1 β), tumour necrosis factor alpha (TNF- α), Interleukin-10 (IL-10), and transforming growth factor beta 1 (TGF- β 1). The mean and standard deviation of the level of each marker was calculated and logarithmic transformed. The differences in the level of each marker between the control and infected group(s) and the changes over time were tested by ANOVA for repeated measures.

Results An up-regulation over the first time period was observed for relative expressions of IFN- γ (P = 0.012). The differences in the changes of the IL-1 β level in the rats over the first and the second time period were borderline significant (P = 0.050 and P = 0.069, respectively). IL-10 increased significantly over the second time period (P = 0.016). For TGF- β 1, a difference in changes was found during the second time period (P = 0.002). TNF- α did not respond. **Conclusions** Chronic apical periodontitis was associated with increased levels of systemic inflammatory markers. The present model may help investigate the interaction of disease susceptibility, periapical pathogenesis, and systemic inflammation.

SESSION 2: FRIDAY 7 SEPTEMBER 2007

ABSTRACTS

R2.33

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Ultrastructure of young and old root canal dentine following 17% EDTA irrigation: an image analysis study

Aim To evaluate and compare the effects of 1 and 10 min irrigation with 17% EDTA on the ultrastructure of root canal dentine in young and old extracted teeth.

Methodology Human anterior teeth were extracted from young (< 30 years) and old (> 60 years) patients. The root canals were enlarged to an apical size 60, after which the crowns and apical thirds were removed. The remaining mid-root portions were bisected longitudinally. Each half of the root was treated with 17% EDTA for 1 or 10 min, followed by NaOCl irrigation (2.5%, 10 mL). The samples were then prepared for scanning electron microscopic evaluation and the dentine surfaces photographed digitally at three equidistant (coronal-middle-apical) regions at 900× and standardized magnifications. In each specimen, the area of exposed dentinal tubules was quantified using an open-source image analysis toolkit (Image], National Institute of Health, Maryland, USA). The Mann-Whitney U test was used to compare the area of exposed dentinal tubules between the two age groups and the two durations of EDTA treatment. Results Extended application of EDTA treatment resulted in erosion of the periphery of dentinal tubules, regardless of dentine type (young or old). In young dentine, the 10 min EDTA treatment did not result in significantly (P > 0.05) larger total area of exposed dentinal tubules than 1 min EDTA treatment. In old dentine, 10 min treatment with EDTA resulted in a significantly larger total area of exposed dentinal tubules. There was no significant difference between young and old dentine with regard to the treatment times (P > 0.05). Conclusions In young dentine, the effects of 1 or 10 min treatment with 17% EDTA solution on the dentine ultrastructure were not significantly different. In old dentine, extending the treatment time resulted in a significant increase in dentinal tubule opening at the expense of dentine erosion.

R2.34

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Effect of different irrigation solutions on the microhardness of root dentine

Aim To evaluate the effect of various chelating solutions (7.5% EGTA, 7.5% EDTA, and 7.5% CDTA) used in combination with 2.5% NaOCl or 2.5% ethylenediamine (ED) solutions on the microhardness of root dentine.

Methodology The crowns of 25 maxillary canine teeth were removed, the roots sectioned longitudinally into two segments, embedded in polyester, and polished. Before treatment with any solution, Vickers microhardness values of the specimens were obtained as the control value. A total of fifty specimens were divided into five groups and treated as follows. Group 1: CDTA and NaOCI; Group 2: EDTA and ED; Group 3: EDTA + ED mixture (1/1 volume) and then EDTA + ED mixture (1/1 volume); Group 4: EGTA and NaOCI; Group5: EDTA and NaOCI. The specimens were soaked in

50 mL of each solution for 1 min. After each soaking procedure, the microhardness of root dentine was re-assessed and compared with the control values. Data were analyzed statistically using ANOVA. When significance was detected, covariance analysis and Bonferroni test were used to determine statistical differences among the solutions (P = 0.05).

Results The results indicated that all solutions significantly decreased microhardness values of root dentine in all thirds (P < 0.05). Soaking with 7.5% EGTA resulted in a greater reduction of dentine hardness (P < 0.05). Treatment with 2.5% NaOCl gave the lowest values as final solution (P < 0.05).

Conclusions Irrigation with EDTA + ED (1/1 volume) produced minimal changes on the microhardness of root dentine compared with the other solutions.

R2.35

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Time-dependent effects of 5.25% NaOCI and MTAD on the calcium content of root dentine

Aim To evaluate time-dependent effects of 5.25% NaOCl with or without subsequent MTAD (Dentsply, Tulsa Dental, OK, USA) application on the calcium content of root canal dentine.

Methodology Crowns of single-rooted teeth were removed and the roots bisected longitudinally. The root halves (n = 35) were covered with two layers of nail varnish, leaving the root canal surface exposed. The specimens were then dehumidified at 120°C for 45 min. Except for one group (5 min MTAD treatment alone), all specimens were immersed in either 5.25% NaOCl or distilled water for 5, 10 or 15 min. Specimens treated with three different application times of 5.25% NaOCl were further exposed to MTAD treatment (5 min). The amount of calcium release from dentine following single (MTAD, NaOCl or distilled water alone) and combined (NaOCl + MTAD) treatment regimens were determined by flame photometry. Kruskal–Wallis analysis of variance and Wilcoxon signed ranks tests were used for statistical analysis of data (P = 0.05).

Results Regardless of treatment time, all 'single' and 'combined' treatment regimens removed significantly more calcium than that of the control (P < 0.05). 5.25 % NaOCl + MTAD combined treatment groups removed significantly more calcium than when NaOCl was used alone, regardless of treatment time (P < 0.05).

Conclusions Subsequent irrigation with MTAD significantly increased the liberation of calcium ions from root canal dentine treated with 5.25 % NaOCl for 5, 10 and 15 minutes.

R2.36

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The effect of a new root canal irrigation device on dentine debris

Aim To evaluate the effect of the new root canal irrigation device RinsEndo (Dürr Dental, Germany) on debris in simulated root canal irregularities compared with conventional rinsing techniques.

Methodology Six root canals from single rooted human teeth were prepared to an apical size 45 using a crown-down technique. All teeth were split longitudinally into two halves. To simulate uninstrumented canal irregularities three standard depressions were cut on the canal wall of each half at 2, 4 and 6 mm from the apex. Dentine debris was produced by mixing dentine shavings with 1% NaOCl. The amount of dentine debris was standardized and placed into the depressions. The two halves of each tooth were then reassembled and fixed in an impression material. Four irrigation techniques were compared; group 1: syringe irrigation; group 2: syringe irrigation with a NaviTip FX needle; group 3: irrigation with RinsEndo (Dürr Dental GmbH, Germany) and group 4: ultrasonic irrigation using Endo Soft Instruments (EMS, Switzerland). The same six teeth were used for every group to standardize the experimental conditions in each group. About 5 mL of 1% solution of sodium hypochlorite was used in each group. Images were taken before and after irrigation with a digital camera connected to a microscope. A scoring system (Lee et al. 2004) was used to evaluate the amount of remaining debris (scores 1-5: '1' = complete debris removal and '5' = no removal).

Results All irrigation techniques reduced the amount of debris. Ultrasonic irrigation (group 5) had significantly lower scores than groups 1 and 2 and completely removed debris in all cases (Score 1). RinsEndo also achieved a score of 1 other than in two cases (score 2). There was no significant difference between groups 3 and 4.

Conclusions Under the conditions of this study irrigation with RinsEndo and an ultrasonic device using 1% solution of sodium hypochlorite was more effective in removing artificial dentine debris from root canal walls.

R2.37

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Cleanliness of root canal walls – laser versus irrigation: a micro-Raman spectroscopic and SEM analysis

Aim To determine chemical and structural modifications of root canal dentine surfaces after laser treatment (Nd:YAG, KTP) or irrigation with 2.5% NaOCl and 17% EDTA.

Methodology Fifty-six extracted single rooted teeth were assigned into seven groups of eight teeth. Group 1: Root canal dentine – untreated (control). Group 2: root canal treated and irrigation with 2.5% NaOCl (3 mL) after each file size. Group 3: protocol of Group 2 with final rinse of 17% EDTA (3 mL, 3 min). Group 4: protocol Group 2 and Nd:YAG irradiation (Fidelis, Herzele, Belgium) (1.5 W, 15 Hz, 4 × 5 s and 20 s interval). Group 5: protocol of Group 2, vertical root splitting and Nd:YAG irradiation direct on the root canal wall. Groups 6 and 7: protocol Groups 4–5 but with KTP laser (DEKA, Calenzano, Italy) (1.5 W, continuous mode, 4 × 5 s and 20 s interval). Five roots per group were selected for microRaman analysis, three were kept for SEM analysis.

Results MicroRaman analysis revealed local decreases or disappearance of the phosphate peak after use of EDTA. The collagen peaks were modified after Nd : YAG and KTP irradiation: changes in the collagen bands revealed degradation of the collagen up to the formation of amorphous carbon, depending on the exposure intensity of the laser irradiation. SEM analysis confirmed these ultramorphological changes of the root canal wall dentine after laser irradiation. Irrigation with EDTA resulted in the removal of smear layer and the

presence of open dentine tubules. The sole use of NaOCl did not result in smear layer removal. The removal of smear layer was significantly higher with Nd : YAG than with KTP.

Conclusions Depending on the degree of laser irradiation exposure a broad peak due to collagen degradation or creation of amorphous carbon was observed. Nd : YAG was more effective in removing smear layer than KTP-laser. EDTA removed smear layer most efficiently.

R2.38

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A SEM study of smear layer removal in association with NaOCI and EDTA formulations

Aim To determine ultramorphologically (SEM) the *ex vivo* effect of EDTA paste (19%), EDTA rinse (17%) and NaOCl (2.5%) on intracanal smear layer.

Methodology Thirty-six single rooted canines were prepared to a size 45 K-file and divided into six groups. Groups 1 and 4 were irrigated using NaOCl (3 mL) after each file. Groups 2 and 5 underwent the same protocol as group 1, but with a final rinse of EDTA (3 mL, 3 min, Salvizol, Pierre Rolland, France). Groups 3 and 6 underwent the same protocol as group 1, but with a final rinse of EDTA (3 mL, 3 min) and NaOCl (3 mL, 2 min). In Groups 4, 5, 6 an EDTA paste (File Eze, Ultradent, USA) was used with each file. All teeth were split longitudinally and observed under SEM at 1, 3, 6, 8, 12 mm from the physiological apex for smear layer evaluation, counting the number of dentine tubules and measuring their diameters. Values were statistically analyzed using non-parametric tests.

Results The use of EDTA as a single final rinse resulted in significant differences in smear layer removal between groups 1 and 2 at 6, 8 and 12 mm from the apex (P < 0.05). Also, significantly more tubules were observed in group 2 than in group 1 at 3, 6, 8 and 12 mm (P < 0.05). The use of EDTA paste and the use of NaOCl after the EDTA rinse did not significantly affect the smear layer, nor the amount of exposed tubules. Comparing tubules diameters, there were no significant differences amongst all groups except at the 6 mm level.

Conclusions Ethylenediaminetetraacetic acid as a final rinse was efficient in removing smear layer and exposing tubules. The use of NaOCl after EDTA rinse had no additional effect, neither had EDTA paste during preparation.

R2.39

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Four methods for cleaning simulated lateral extensions of curved root canals – a SEM evaluation

Aim To evaluate the cleaning efficiency of four techniques to remove artificially-placed dentine chips from simulated apical groves and depressions within curved human root canals.

Methodology Forty curved root canals of human molars were prepared to size 80, 0.02 taper. Teeth were split longitudinally. A groove serving as a non-instrumented lateral extension (length: 4 mm, width: 0.2 mm, depth: 0.5 mm) was created 2–6 mm from the apex in the wall of one-half of each root canal. Three depressions serving as canal irregularities (diameter: 0.3 mm, depth: 0.5 mm) were prepared in the other half at 2, 4 and 6 mm from the apex (Lee *et al.* 2004). Grooves and depressions were packed with dentine debris under a stereo-microscope (50×), reassembled and fixed in putty material. Four irrigation methods were performed (n = 10): group 1: citric acid (40%) and NaOCl (5%) using syringe irrigation (Endoneedle Buquet, Vedefar, Belgium), group 2: citric acid/sonic activation (Sonic Air, Micromega, France) and NaOCl/syringe irrigation, group 3: citric acid/ultrasonic activation (Piezon Master 400, EMS, Switzerland) and NaOCl/syringe irrigation, group 4: citric acid/endobrush activation (Coltène Whaledent, Germany) and NaOCl/syringe irrigation. Images of specimens were taken using a SEM. Grooves and depressions were evaluated for remaining debris using a 4-step scoring system.

Results Ultrasonic irrigation reduced debris significantly better from grooves and depressions than syringe irrigation (Mann– Whitney-test: P < 0.05), followed by endobrush and sonic activation. The groups differed significantly (Kruskal–Wallis-test: P < 0.05). The greatest amount of the debris remained in grooves and depressions without activation (syringe irrigation only). The groups with activation showed a significant difference compared with syringe irrigation (Kruskal–Wallis-test: P < 0.05). No significant difference was found among groups with different methods of activation (P > 0.05).

Conclusions Treatment with sonic, ultrasonic and endobrush activation improved the removal of debris from simulated canal extensions and irregularities in the apical portions of curved root canals.

R2.40

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Root canal cleanliness after instrumentation with modern nickel titanium instruments – a SEM study

Aim To evaluate the quality of root canal instrumentation using three newly developed nickel titanium instrument systems in comparison to ProFile instruments.

Methodology Root canals of 48 premolars were randomly assigned to one of the following four groups: group 1: ProFile (Dentsply Maillefer, Switzerland), group 2: Liberator (Miltex, USA), group 3: Alpha System (Brasseler, Germany), group 4: Mtwo (VDW, Germany). Instruments were used in the sequence recommended by the manufacturers. In order to avoid an interference of irrigants on the cleaning process, the root canals were rinsed only with distilled water. Following instrumentation, the roots were dried and longitudinally cracked. Specimens were prepared for SEM analysis and each root half was photographed in the apical, middle, and coronal section of the root canal. SEM images were scored for debris, smear layer and root canal surface by two calibrated investigators according to Hülsmann *et al.* (1997). Statistics were computed using SPSS Win 14.0 (Kruskal–Wallis test, Friedman test, Mann–Whitney test).

Results Kruskal–Wallis test yielded significant differences among the groups (P < 0.001). Debridement of apical portions of the root canals was performed significantly better by Liberator, Mtwo and Alpha System than by ProFile instruments (Mann–Whitney test, P < 0.05). Liberator instruments produced less smear layer than ProFile and Alpha System instruments (Mann–Whitney test P < 0.05). No significant differences were found between Liberator and Mtwo instrumentation (Mann–Whitney test, P > 0.05) as well as for different regions within the root canal (Friedman test, P > 0.05).

Conclusions Modern nickel titanium instruments revealed better cleaning efficiency than ProFile instruments. Using these instru-

ments, root canal instrumentation with less smear layer creation and smooth root canal walls can be achieved. However, due to their design, ProFile instruments yielded the smoothest root canal walls of all instruments.

R2.41

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Accuracy of root canal length determination with the impedance ratio method

Aim To determine optimal measuring parameters of the impedance ratio method for root canal length determination and to evaluate its accuracy.

Methodology Electrical impedance was measured *ex vivo* on 14 extracted human teeth with single canals using a QuadTech 1920 precision impedance analyzer. A file electrode was inserted into the root canal; the second electrode was placed in the saline solution surrounding the tooth. Measurements were performed in a frequency range from 20Hz up to 1 MHz, and were repeated with different distances of the file tip from the apical foramen (from 8 mm inside the canal to 2 mm beyond the foramen). The measured impedances were analyzed as a function of distance of the file tip to the apical foramen. Parameters (e.g. measurement frequencies, impedance ratio value) that would result in optimal determination of the root canal length were evaluated.

Results The optimal root canal length determination was obtained at frequencies of 5 and 1 kHz for the impedance ratio 0.79. The average measured distance from apical foramen (\pm SD) was 0.07 mm beyond the apical foramen (\pm 1.04 mm). The strict clinically acceptable limit of \pm 0.5 mm was attained in 57% of all measurements. Standard deviations decreased as the average measured distance from the apical foramen went further beyond the apical foramen.

Conclusions The impedance ratio method determined the apical foramen with clinically acceptable accuracy and consistency. A trade-off between minimal average measured distance from apical foramen and the standard deviation is necessary.

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R2.42

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Ex vivo evaluation of the accuracy of three apex locators in simulated clinical conditions

Aim To evaluate the accuracy of three apex locators: DentaPort zx (Morita, Japan), ProPex (Dentsply, USA) and LAEC (Castellini, Italy; part of a dental unit), in simulated clinical situations.

Methodology Freshly extracted human teeth (n = 52) were stored in sterile saline, and cleaned to remove periodontal fibres and calculus. The crowns were sectioned at the cemento-enamel junction using a diamond disc. The actual lengths (AL) of the canals were measured with a size 10 K-file under a microscope (×24). The canals were then instrumented using K3 NiTi instruments (Sybron Endo, USA) to an apical size 25 and a 0.06 taper. The specimens were mounted in an electro-conductive model consisting of a polystyrene foam cup with a plastic support for the teeth, a hole for the labial clip of the apex locator, and a solution of Agar agar. The teeth were randomly allocated into five groups and filled with human blood (group A); hydrogen peroxide (group B); substitute saliva gel (group C); paracetic acid (group D); or 5.25% NaOCl (group E). The length of each canal was determined with a size 25 K-file attached to one of the three apex locators. The data were recorded and analysed using the error evaluation; the Friedman's test at P < 0.001 and the Newman Keuls test at P < 0.01.

Results In the presence of blood, NaOCl, or hydrogen peroxide, the canal lengths determined by all the apex locators were different from the AL by 0.5-1 mm. The DentaPort zx gave a significantly better estimate of the canal length than ProPex apex locator.

Conclusions All apex locators were affected by the presence of the different fluids in the canal; however, they gave an acceptable level of canal length estimation, including the LAEC that is incorporated into the dental unit.

R2.43

Abstract Withdrawn.

R2.44

Abstract Withdrawn.

R2.45

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Histological examination of the accuracy of three apex locators using an accurate mounting model

Aim To evaluate the accuracy of apex locators in determining the apical constriction.

Methodology Three apex locators (Root ZX, RayPex[®]4 and ApexPointer) were tested in 38 anterior teeth. In order to eliminate operator bias during the measuring procedure, teeth were fixed in a Mounting Model. The Mounting Model allowed a semi-automated positioning of the measuring file and provided a real-time digital accurate reading of the working length. Moreover, the Mounting Model facilitated the use of different apex locators in the same root canal without changing the relative position between tooth and file. Working length determination was performed at display level '0.5' of the apex locators. All 38 root canals were measured with each of the three apex locators. Finally, the measuring file was fixed in the pulp chamber using a resin (at the display level '0.5' of Root ZX). The apical portion of each root was exposed histologically and photographed at 30× magnification. The extent of the apical constriction was defined and the frequency of localizing the tip of the measuring file within the apical constriction was recorded (with a precision of ±0.5 mm). The accuracy of each device was determined by calculating the percentage of localizing the file tip within the apical constriction. A 95% confidence interval was calculated to compare the apex locators.

Results Statistically there was no significant difference between the accuracy of the three apex locators. The percentage of each device detecting the apical constriction was: Root ZX (79%, 30/38 root canals), RayPex[®]4 (82%, 31/38) and ApexPointer (79%, 30/ 38). In all three apex locators no measurement was beyond the apical foramen or short of the apical constriction.

Conclusions Measurements taken from these apex locators were either at the apical constriction or between the apical constriction and foramen. In nearly 80% of the measurements the apical constriction was accurately determined.

R2.46

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The accuracy of working length measurements using apex locators, conventional film and digital radiography

Aim To compare the accuracy of apex locators, conventional film and digital radiography in determining working length (WL) in root canals.

Methodology Thirty canals from single-rooted teeth that had been scheduled for extraction were used. The coronal portion of each canal was prepared using Gates-Glidden drills, and a H-file was then inserted into the canal until the apex locator (Root ZX, J. Morita, Japan) indicated that the file had reached the apical constriction. After cementing the file in the root canal using resin composite, standardized images of the teeth were obtained with E-speed film and Schick CDR (Schick Industries, Long Island City, USA) digital system. Following extraction, the apical 4 mm of each root canal was exposed along the long axis of the tooth and viewed using a stereomicroscope at 40× magnification. To determine the actual canal length, the minor foramen was detected. The distance between the minor foramen and the file tip was measured and the actual length was calculated accordingly. Four examiners viewed each radiograph or image and determined the radiological WL. Pearson correlation and *t*-test was used for the statistical analysis (P = 0.05). Results Pearson test revealed significant correlations between the actual length and the measurements of the apex locator, film and digital radiography (r = 0.971 - 0.979). Apex locator measurements were longer than the actual length (P < 0.05). No significant differences were observed between the actual length and radiographic measurements (P > 0.05).

Conclusions The apex locator did not correctly determine the position of the minor foramen and were on average longer than the actual length.

R2.47

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Cylindrical representation and microtomography: a new methodology to assess root canal geometry and changes after endodontic preparation

Aim To propose a modified version of 3D reconstruction methodology to assess root canal geometry before and after canal preparation. Methodology Two steps have been used to obtain root canal images: image acquisition and three-dimensional (3-D) reconstruction. Acquisition: the specimens (freshly extracted human maxillary and mandibular molars) were scanned, before and after root canal shaping with HeroShaper instruments (Micro-Mega, Besançon, France) following the manufacturers' guidelines. Acquisitions were carried out using a high resolution microtomography scanner, Skyscan1072 (Skyscan, Belgium). Each acquisition contained 470 slices with a thickness of 38.57 µm. 3-D reconstruction: data was transferred and 3-D image segmentation and registration was performed using commercial software, Amira (Mercury Computer Systems, Richmond, TX, USA). Filtering and measurement were carried out through a newly developed research platform. Assessment of the anatomical changes was achieved using a mathematical method, i.e., cylindrical representation.

Results The 3-D images of the root canals were obtained. Volumes and surfaces area, cylindrical representations, and canal axis for

each canal before and after preparation were calculated. Curvilinear abscissas were calculated for each canal before and after canal shaping with endodontic instruments.

Conclusions The 3-D reconstruction methodology was adequate and thus a promising methodology to explore root canal geometry and anatomical changes.

R2.48

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A preliminary micro-computed tomographic analysis of apical enlargement obtained using Mtwo NiTi rotary apical files

Aim To evaluate apical preparation obtained using Mtwo size 25, 0.06 taper (VDW GmbH, München, Germany) and Mtwo apical files (Mtwo A; VDW GmbH) assessed by micro-computed tomography. Methodology A micro-computed tomography scanner (SkyScan 1072, Assing SPA, Belgium) was used to analyse 15 root canals in five extracted maxillary molars. Specimens were scanned before root canal preparation, after shaping using Mtwo basic sequence (size 10, 0.04 taper, 15, 0.05 taper, 20, 0.06 taper, 25, 0.06 taper) and after apical enlargement using Mtwo apical files. These instruments have a greater taper in the apical 3 mm (Mtwo A1, tip size 20, 0.15 taper; Mtwo A2, tip size 25, 0.15 taper; Mtwo A3, tip size 25, 0.20 taper), while the rest of the coronal portion is a 0.02 taper. Mesio-buccal and disto-buccal canals were prepared to the Mtwo A2, palatal canals were prepared to the Mtwo A3. Threedimensional root canal models were reconstructed and evaluated for volume of each of the apical 3 mm and for root canal diameters 3 mm, 2 mm and 1 mm from the apex. The mean volume and diameters change were determined at the three levels analysed. Roundness of apical preparation was further evaluated (diameter ratio) and a qualitative evaluation of apical preparation was performed. Comparisons were made using repeated-measures ANOVA (P < 0.05).

Results No instruments fractured or deformed in the study. No obvious procedural errors were detected after canal preparation with Mtwo and Mtwo A. Repeated-measures ANOVA revealed that preparation significantly increased canal volumes and diameters (P < 0.05). The root canal diameter ratio revealed round preparation after the use of both Mtwo size 25, 0.06 taper and Mtwo A, with larger preparations obtained using Mtwo A.

Conclusions Under the conditions of this study, Mtwo apical files effectively enlarged the apical 3 mm of root canals in maxillary molars after preparation using a size 25, 0.06 taper file.

R2.49

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Micro-computed tomographic analysis of molars prepared using ProFile and AET endodontic instruments

Aim To evaluate root canal morphology after instrumentation with Endo-Eze AET stainless-steel instruments (Ultradent Inc., South Jordan, UT, USA) and ProFile Ni-Ti rotary instruments (Dentsply Maillefer, Baillagues, Switzerland) assessed by micro-computed tomography.

Methodology A micro-computed tomography scanner (SkyScan 1072, Assing SPA, Belgium) was used to analyse 10 maxillary and 10 mandibular first molars. Specimens were scanned before and after root canal preparation using Endo-Eze AET and ProFile. Each system was used to prepare five maxillary and five mandibular molars. Three-dimensional root canal models were reconstructed and evaluated for volume. The total volume of dentine removed and the volume of the coronal, middle and apical third of each root canal were calculated. The mean volume change before and after instrumentation was determined for the entire root canal as well as each section and the values compared. A qualitative evaluation of root canal preparation was performed. Student *t*-test was used to determine the difference between the two experimental groups (P < 0.05).

Results No instruments fractured in the study. Deformation occurred in one ProFile size 30, 0.04 taper instrument. No statistically significant differences were noted between the groups in the volume of dentine removed after root canal preparation in all root canals of mandibular and maxillary molars, except for the total volume (P = 0.04) and the volume of the apical third (P = 0.03) of the disto-buccal canal of maxillary molars, where ProFile instruments produced significantly less enlarged canal volume than the Endo-Eze AET (P = 0.04). Qualitative evaluation of the preparations showed that both ProFile and Endo-Eze AET were able to prepare root canals in mandibular and maxillary molars with little or no procedural error.

Conclusions Under the conditions of this study, both the systems analysed were able to prepare molar teeth with similar amount of dentine removal and low risk of procedural errors.

R2.50

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Apical adaptation of four different instruments: An *ex vivo* study

Aim To evaluate the adaptation of both hand and rotary filing instruments inside the root canal at 1 mm short of the apical foramen.

Methodology Forty extracted mandibular incisors with one canal and single apical foramen were selected. The teeth were de-coronated at the cemento-enamel junction, pulpal chambers were accessed and pulp tissue removed. A size 8 K-file was introduced to the foramen in the canal of each tooth to confirm apical patency. Working lengths (WL) were determined 1 mm short of the apical foramen. Following flaring of the coronal and middle third of the canals, the apical foramen was prepared with either ProTaper, K-file, ProFile or Hedström files (10 teeth for each type of instrument). When the apical preparation was completed the instruments were fixed in the root canals using acrylic resin. The apical 1 mm of each root tip was ground on wet sandpaper in order to expose the canal and the instrument at the WL. Apical debris was removed with an ultrasonic cleaner and the apical region of each tooth was examined with a stereomicroscope. The stereoscopic images of the teeth were digitized and the differences between the areas of root canals and file tips were determined using a image analysis software. Data were analyzed using one way ANOVA test. Results ProTaper had the best adaptation with the root canal walls, followed by K-file, ProFile, and Hedström files. However, differences were not statistically significant (P > 0.05).

Conclusions None of the instruments were perfectly adapted to the root canal walls at the WL in mandibular incisors. Therefore, total removal of debris from the canal walls may not be achieved when these instruments are used in the apical region.

R2.51

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Comparison of root canal preparation using FlexMaster NiTi and AET stainless steel instruments

Aim To compare various parameters of root canal preparation using FlexMaster (VDW, Munich, Germany) rotary nickel-titanium and AET (Ultradent Products, South Jordan, UT, USA) oscillating stainless steel instruments.

Methodology Fifty extracted mandibular molars with mesial root canal curvatures between 20° and 40° were embedded in a muffle system and prepared using FlexMaster instruments with the EndoStepper motor (VDW, Munich, Germany) and AET instruments with an EndoEze handpiece (Ultradent Products, South Jordan, UT, USA). Preparation was performed in a crown-down-sequence as recommended by the manufacturers with a final apical preparation size of 45 for all teeth. The following parameters were analysed: straightening of curvature, post-operative root canal cross-section, cleanliness of canal walls, safety issues, and working time.

Results The mean degree of straightening was significantly less for FlexMaster (mb 0.3° , mL 0.3°), respectively than for AET (mb 3.9° , mL 5.4°) (Mann–Whitney test). Overall, 84% (FlexMaster) and 86% (AET) of the post-operative cross-sections were acceptable showing a round or oval diameter; differences were not significant (Fisher's exact test). Neither of the systems completely eliminated debris and smear layer, but FlexMaster left significantly less smear layer (Mann–Whitney test). No losses of working length or instrument fractures occurred with either of the systems. Mean working time was significantly shorter for FlexMaster (mb 93 s, mL 95 s) than for AET (mb 330 s, mL 315 s).

Conclusions FlexMaster NiTi instruments respected original root canal curvature well, whereas preparation with AET instruments resulted in unacceptable straightening. Both systems were safe to use. Owing to insufficient cleaning of canals an effective irrigation regime is necessary. AET cannot be recommended for preparation of curved root canals.

R2.52

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Efficiency of Mtwo[®] and ProTaper[®] in preparing oval root canals

Aim To assess the preparation of greater taper instruments in oval root canals.

Methodology Ninety oval root canals were integrated in a reassembly model to take pre- and post-preparation photographs. Samples were divided into three groups (n = 30) according to the preparation method: Mtwo[®], ProTaper[®] and NiTi-Hand-Files. Roots were sectioned at two levels in the middle third of the root (approximately 4 mm and 7 mm from the apex) and pre-preparation photos of the sectioned root surfaces were taken at $30 \times$ magnification. Roots were reassembled and conventional endodontic access cavities were performed. The working length was determined to be 1 mm short of the apical foramen. In addition to the normal sequence of each system and to the Balanced-force-technique for NiTi-Hand-Files, a circumferential filling of the middle third of the root canal was performed with each instrument/file used. In Mtwo[®] and ProTaper[®] groups, freely rotating instruments were pressed against the root canal walls and displaced in coronal

direction. Preparation procedures were performed under simulated clinical conditions on a dental mannequin. Post-preparation photos were taken and superimposed on the pre-preparation photos. The percentage of prepared root canal outline was measured and the number of sections with less than 0.5 mm remaining dentine-wall thickness was recorded. For each group the mean and the 95% confidence interval (CI) were calculated.

Results Considering the percentage of prepared root canal outline, statistically there was no significant difference between Mtwo[®] 75.8% [CI: 70.3%; 81.3%] and ProTaper[®] 73.5% [CI: 68.0%; 79.0%], but both groups were significantly different from the NiTi-Hand-Files group 64.1% [CI: 58.6%; 69.6%]. The minimal dentine-wall thickness was less than 0.5 mm in eight root canals of ProTaper[®] (27%) and in five root canals of Mtwo[®] (17%).

Conclusions None of the tested systems was able to completely prepare oval root canals.

R2.53

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Shaping ability of Mtwo and ProTaper rotary instruments in simulated root canals

Aim To compare the shaping ability of Mtwo (VDW, Germany) rotary nickel-titanium instruments with ProTaper (Dentsply Maillefer Switzerland) rotary nickel-titanium instruments.

Methodology Simulated curved canals (n = 40) were prepared using Mtwo instruments (n = 20) or ProTaper instruments (n = 20) running at 250 rpm according to manufacturers' instructions. Half of the canals in each group were prepared by one of the two operators. One of operators was familiar with the ProTaper system and the other with the Mtwo system. Both operators became familiar with the respective new rotary system by using it on clinics prior to the test. The canals were prepared up to apical size 25. The pre- and post-instrumentation images of the canals were recorded and assessment of the canal shape was completed with a computer image analysis program. The amount of material removed was measured at 18 measuring points, beginning 0.5 mm short of the end-point of preparation. Incidence of canal transportation, total preparation time and instrument failures were also recorded. The data were analysed using two-way analysis of variance with simple effects assessment.

Results In the apical and middle thirds of the canals, Mtwo instruments removed more material from the outer canal wall and less from the inner canal wall than ProTaper instruments. In the coronal third of the canals, Mtwo instruments removed less material from the outer canal wall and more from the inner canal wall than the ProTaper instruments. Preparation with ProTaper instruments resulted in ledges in three canals whereas Mtwo instruments did not cause any ledges. All these differences were statistically significant at the 5% level.

Conclusions Mtwo instruments tended to straighten canals in the middle and apical thirds. ProTaper instruments created ledges in the apical part of some canals.

R2.54

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Effects of anticurvature filing on danger zone width in curved root canals

Aim To compare the effects of anticurvature filing with stainless steel K-files versus nickel-titanium rotary ProFiles in the shaping of mesial root canals of extracted mandibular molars.

Methodology A total of 60 canals from 30 mesial roots of mandibular molar teeth were randomly assigned to three groups (n = 20 each). They were prepared with different instruments and methods: The first group with stainless steel K-files and circumferential filing, the second with pre-curved stainless steel K-files and anticurvature filing and the third with ProFiles (0.06 taper) and anticurvature filing. Using a micro-computed tomography system (Skyscan-1076, Skyscan, Antwerp, Belgium), pre-and post-operative specimens were scanned. Subsequently, canal images were superimposed and changes in root dentine thickness were measured along distal side (danger zone) of the canal. The data was analyzed using a one-way ANOVA and the comparison of means was conducted using a post hoc multiple comparison Tukey test.

Results There were significant differences in the thickness of root dentine removed at the 7.5–8.5 mm level between groups 1–3. Changes in root dentine thickness at the 5–6 mm level of danger zones in Group 1 were more than those in Group 3 and there were more changes in Group 2 at the 3.5–6 mm level than those in Group 3 significantly (n = 20, P < 0.05).

Conclusions Anticurvature filing was effective for not only stainless steel K-file but also NiTi ProFile to minimize the possibility of strip perforation in the danger zone of mandibular molars.

R2.55

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A comparison of the efficacy of Hero 642 versus hand instruments for removing gutta-percha root fillings

Aim To assess the efficacy of Hero 642 rotary instruments in removal of root fillings during re-treatment in mandibular molar teeth.

Methodology The canals of forty human mandibular molar teeth were instrumented using hand files and obturated with Diaket and gutta-percha. Apical, middle and coronal thirds of each root were obtained by sectioning the roots with a low speed precision saw (Isomet, Buehler, IL, USA). Following reorientation of the root segments, an individual muffle was created for every tooth. Re-treatment was performed using hand instruments only (Group 1), hand instruments + solvent (Group 2), Hero 642 instruments only (Group 3) and Hero 642 instruments + solvent (Group 4). The solvent used was chloroform and ethyl ether mixture (1:8). The root canal cross-sections were inspected before and after re-treatment. The remaining root canal filling material was quantified microscopically using LUCIA software (Version 4.21, Laboratory Imaging Ltd., Czechoslovakia). The data were analyzed using Kruskal-Wallis and Mann Whitney-U, Friedman and Wilcoxon tests and Chi-square tests (P = 0.05).

Results Re-treatment using Hand instruments + solvent resulted in significantly less residual gutta-percha filling than Hero 642 (P = 0.018) or Hero 642 + solvent (P = 0.016). There was no significant difference between the hand instruments and hand instruments + solvent groups (P = 0.639) or between the Hero 642 and Hero 642 + solvent groups (P = 0.990). In the apical third, perforations were observed in 47.5% of teeth. The use of solvent reduced the prevalence of perforations in the apical segments regardless of re-treatment method, but the effect was not statistically significant (P > 0.05). No hand or rotary instrument breakage was observed in any groups.

Conclusions Hand instruments resulted in less residual guttapercha following re-treatment and solvent use decreased the potential of apical perforation.

Acknowledgements Assoc Prof. Timur KOSE, Prof. Sema BELLI.

R2.56

Abstract Withdrawn.

R2.57

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An *ex vivo* comparative study of apically extruded debris and irrigants resulting from conventional and three rotary (RaCe, K3, FlexMaster) instrumentation techniques

Aim To evaluate the weight of debris and irrigant volume extruded apically from extracted teeth *ex vivo* after endodontic instrumentation using a stepback technique or three engine-driven techniques utilizing nickel–titanium instruments (RaCe, K3, FlexMaster).

Methodology Sixty single-rooted premolars were divided into four groups of 15 teeth each. The root canals of group 1 were prepared with RaCe, Group 2 - K3, Group 3 - FlexMaster, Group 4 - stainless steel files. During canal preparation teeth were placed in glass vials with rubber stoppers to evaluate the amount of debris and volume of liquid that extruded from the apical foramina. A bent 27 gauge needle was forced alongside the rubber stopper to serve as a drainage cannula. The vial was vented with this needle during insertion to equalize the air pressure inside and outside the vial. The needle was also used as an electrode for the electronic working length determination during canal instrumentation. All canal preparations and working length measurements were completed using an Endomaster (EMS, SA, Switzerland) endodontic handpiece. Extruded irrigant was collected from the drainage cannula into a disposable plastic insulin syringe. The amount of the extruded debris collected in the vial was determined by means of an analytic balance at 10^{-5} gram precision, after evaporating the fluid in the vial and subtracting the weight of the vial and salt deposits.

Results There was a statically significant difference for liquid extrusion between K3 and the stepback group (P < 0.05). Differences between other groups were not statically significant (P > 0.05). Most liquid extruded apiacally was seen in the group that used K type stainless steel instruments with the stepback technique. Most apical debris extrusion was seen in the group that used K-files used in a stepback technique. There was no significant difference for extruded debris between the groups (P > 0.05).

Conclusions The engine-driven nickel-titanium systems were associated with less apical extrusion and irrigant.

R2.58

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Evaluation of the success rate of a minimal-invasive technique for removal of fractured instruments

Aim To evaluate the success rate of a minimal-invasive technique using the operating microscope and ultrasonic tips for removal of fractured instruments.

Methodology Retrospectively, the radiographs and records of 170 consecutive cases (147 teeth in 145 patients) with fractured instruments were evaluated. All teeth had been treated in a private practice by one experienced endodontist using a standardized technique. The majority of cases were referrals from general dental practitioners. All removal attempts were performed using an

operating microscope. Following preparation of an access cavity ultrasonic steel instruments (Endosonore, DeTrey/Dentsply, Ballaigues, Switzerland) were used to loosen and remove the fragments. **Results** From a total of 170 fragments 162 (95%) were removed, eight cases failed (5%) including one perforation and one case in which the fragment was only bypassed. No differences could be detected with regard to type, length and material (NiTi versus SS) of the separated instrument, the degree of curvature or type of tooth, and the relation of fragment to the curvature.

Conclusions Using a standardized technique including use of a microscope and ultrasonic instruments more than 90% of fractured instruments were removed successfully. The risk of perforation during removal attempts was low. The results were clearly superior to those from a previous study (Hülsmann & Schinkel 1999) on 113, cases which were treated without a standardized technique (no microscope, different techniques and devices) and resulted in 49% removal, additional 19% bypassing, and 11% perforations. The success rate was similar to that reported by Suter *et al.* (2005) who achieved successful removal in 87% of 97 cases.

R2.59

Turkey

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Determining the corrosion rates of rotary Ni-Ti instruments in different irrigating solutions

Aim To evaluate and compare the electrochemical corrosion rate of nickel-titanium rotary instruments when immersed in four different irrigating solutions. The corrosion area on the surface of the files under SEM was also investigated.

Methodology The corrosion rates of the nickel-titanium rotary files immersed in 2% chlorhexidine gluconate, 5% NaOCl, 2.5% NaOCl and 15% EDTA irrigating solutions were determined electrochemically by the Tafel extrapolation method. Data was obtained using a combined system containing a potentiostat, a voltage scan generator and a recorder. The cutting flutes of files were immersed in solutions for 20 min and used as an electrode. A saturated calomel electrode was used as a reference and a platinum plate was used as a counter electrode. In order to determine corrosion rates, the linear region of anodic currents obtained from electrochemical current-potential curves, was extrapolated to corrosion potentials. Seven experiments were completed for each solution, and statistical analysis of the data was performed using the one way Kruskal–Wallis test.

Results The corrosion rates of nickel-titanium rotary instruments from the highest to the lowest were: 5% NaOCl > 15% EDTA > 2.5% NaOCl > 2% chlorhexidine gluconate. The differences between the corrosion rates of files were statistically significant for all solutions (P < 0.001). Average corrosion rates determined by the Tafel extrapolation method, were confirmed by SEM findings.

Conclusions About 20 min immersion in 5% NaOCl or 15% EDTA caused severe corrosion on the surface of selected Ni-Ti instruments.

SESSION 3: SATURDAY 8 SEPTEMBER 2007

ABSTRACTS

R3.60

Abstract Withdrawn.

R3.61

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Antibacterial activity of root canal sealers

Aim To determine the *in vitro* antibacterial activity of the root canal sealers in direct contact with *Escherichia coli, Serratia marcescens, Pseudomonas aeruginosa* and *Staphylococcus aureus.*

Methodology The antibacterial effect of GuttaFlow (Roeko, Coltene/Whaledent GmbH + Co.KG, Germany), Epiphany (Pentron, Wallingford, CT, USA), Diaket (3M ESPE, Germany), IRM (Dentsply, International INC, Milford, USA), SuperEBA (Bosworth Company, Skokie, Illinois, USA) and Hermetic (Pharma Gmbh + Co.KG. Dettenhausen. Germany) was tested using the Direct contact test-DCT. The sealers were prepared according to the manufacturers instructions under aseptic conditions. An increment of each sealer was placed on the side wall of microtiter plate wells and 10 μ l of bacterial suspension (10⁶ bacteria) was placed onto its surface. Each bacterial species was in direct contact with each sealer for 1 h at 37°C. Brain heart infusion (BHI) broth (250 µL) was then added and the growth of each strain was measured after 1, 6, 20 and 24 h. The results of the viable bacteria were expressed as log10 of the mean colony forming units (CFU) per millilitre. The results were compared, using the Student's t-test.

Results The IRM and Epiphany had the greatest antibacterial efficacy against *E. coli*, *P. aeruginosa*, *S. aureus* and *S. marcescens*. Diaket had limited antibacterial efficacy on *S. aureus* but a strong effect on *P. aeruginosa*. SuperEBA and GuttaFlow did not display any antibacterial effect on *E. coli* and *S. marcescens*. Hermetic did not display a bactericidal effect on any of the microorganism.

Conclusions Of the tested materials, IRM and Epiphany had the greatest antibacterial activity *ex vivo*, Diaket, SuperEBA and Guttaflow had limited activity against some species, whilst Hermetic did not exhibit any antibacterial activity.

R3.62

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Evaluation of the degree of conversion of resin based endodontic sealers using the DSC technique

Aim To determine the conversion Degree (CD) of two different resin based endodontic sealers.

Methodology The sealers tested were: EndoREZ (ER) (Ultradent, USA): EndoREZ with catalyst (ERC) (Ultradent, USA): and Real Seal (Sybron Endo, USA). Two LED light units were used to activate the sealers: Ultralume 5 LED (Ultradent, USA); Miniled (Novaxa, France). Two 4.0 mg samples of each group (one for each lamp) were prepared and analyzed with a DSC 7 (Perkin Elmer, USA) calorimeter, in isothermal mode at 32°C (305°K) under flowing argon. The calorimeter cell was calibrated with pure In, Zn and Kr2Cr2O7 standard samples. Temperature measurements were carried out using aluminium pans with a constant heating rate of 10°C. Each lamp was positioned at a fixed distance from the specimen and used for 20 s according to the manufacturer's instructions to irradiate each specimen four times. The software supplied with Perkin Elmer DSC7 device monitored and recorded the heatflow (J/g) developed during treatment. The Kinetic curves and the conversion degree were calculated from the values of heat developed during each polymerization. Data were elaborated using specific software and statistically analyzed with an ANOVA test (P < 0.001).

Results All samples demonstrated almost complete conversion after the first irradiation; prolonged irradiation time increased the degree of polymerization. There was a significant difference between the degree of conversion among the groups: EndoRez with catalyst had the higher values with both lamps.

Conclusions The greater polymerization rate in resin sealers was obtained with the addition of a catalyst.

R3.63

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Adhesion of root canal sealer to dentine and Soft-Core gutta-percha - a SEM evaluation

Aim To evaluate the influence of smear layer on the adhesion of the sealing materials to root canal dentine walls.

Methodology The teeth were aggregated into three groups using the irrigation protocol and instrumentation technique as criterion. Group 1, samples instrumented by hand (K-file Edenta, Switzerland), irrigated with distilled water, filled with Sealapex (Kerr Corporation, Ca. USA) and Soft-Core gutta-percha (3rd Generation Endodontic Obturator, Soft-Core Dental production, Denmark); Group II, samples also instrumented by hand, but irrigated with 5.25% NaOCl (ADD Vision, Germany) and filled with Sealapex and Soft-Core gutta-percha; Group III, samples were prepared with Pro-File rotary instruments (Series 29, 06 taper, Tulsa Dental Products, USA) using the crown-down technique with the Endo-It hand piece (Aseptico Inc., USA) at 300 rpm clockwise rotations, 240 gr cm⁻². To remove the smear layer, 10 mL 17% EDTA (Calcinase, Lege Artis, Germany) followed by 10 mL 5.25% NaOCl were used. Irrigating solutions were delivered via a 23 gauge needle inserted into the canal without binding into the walls. The canals were irrigated with distilled water and dried with paper points and sealed with Sealapex and Soft-Core gutta-percha. All teeth were micro photographed with a SEM (JEOL, JSM-6335F, Japan).

Results The contacts at the sealer/dentine interface in group I were weak revealing gaps. Group II samples revealed a better contact between the gutta-percha and sealer interface compared with the sealer and dentine walls. Maximum contact at the sealer/ dentine interface was achieved in Group III.

Conclusions Penetration of the sealer material into the dentinal tubules when the smear layer was removed was not associated with higher bond strength.

R3.64

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Influence of four different obturation techniques on the quality of gutta-percha root fillings

Aim To compare the homogeneity and adaptation of gutta-percha to root canal walls using four obturation methods.

Methodology One hundred and twenty root canals (34 molars and 30 anterior teeth) were prepared to an apical size of 40–60 using ProTaper® and FlexMaster® systems in a hybrid technique. Using block randomization samples were divided into four groups (n = 30) according to the filling technique: TF (ThermaFil®), VC (vertical condensation), GF (GuttaFlow®) and AC (vertical condensation after fitting a 3–5 mm apical cone). Preparation and filling were performed in a dental mannequin to simulate clinical conditions. The apical 2/3 of the root canals was sectioned at five levels (1–1.5mm apart) and photographed at 30× magnification. In each section the area of filling and voids was measured and the percentage of filled area (PFA) calculated. In addition, the percentage of sections with no voids in each group was calculated. Mean values and the 95% confidence interval (CI) were calculated for comparison.

Results The mean of the PFA in the ThermaFil® group (96.2%, [95% CI: 94.9%; 97.6%]) was statistically significantly lower than the other groups: (VC 99.1% [95% CI: 98.5%; 99.7%], AC 98.4% [95% CI: 97.7%; 99.0%] and GF 98.4% [95% CI: 98.0%; 98.8%]). The highest percentage of sections with no voids was in the TF (81%) compared with VC, AC and GF (75%, 59% and 35% respectively).

Conclusions Vertical condensation, AC and GF provided similar results regarding the parameter PFA. By contrast TF had significantly lower values although voids were found far less frequently compared with GF and AC. From a clinical point of view there is little difference between the four obturation techniques with regard to PFA.

R3.65

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Evaluation of the voids in different root filling materials using computed tomography

Aim To evaluate void volumes in the teeth root filled with Resilon/Epiphany, Resilon/AH Plus, gutta-percha/Epiphany, gutta-percha/AH Plus, Resilon and gutta-percha using three dimensional (3D) analysis of teeth images scanned in a computed tomography (CT).

Methodology Eighty extracted human maxillary and mandibular anterior teeth, excluding mandibular incisors, with single, straight roots were selected. The crowns of all teeth were removed at the CEJ allowing access to the canal. The canals were instrumented using a standard crown-down technique to a size 50, 0.04 taper ProFile (Tulsa Dental, OK, USA) instrument at the established working length. The teeth were irrigated with 5.25% NaOCI followed by 17% EDTA and distilled water (10 mL each). The specimens were randomly divided into six groups and obturated with Resilon/Epiphany (n = 15), Resilon/AH Plus (n = 15), guttapercha/Epiphany (n = 15), gutta-percha/AH Plus (n = 15), Resilon (control, n = 10) or gutta-percha (control, n = 10), using the cold lateral condensation technique. After storing the samples for 1 week in 100% humidity at 37°C, the teeth were scanned in a CT and 3D images were obtained for each sample to evaluate the volume of the voids in mm³ between the root canal wall and the filling material.

Results Voids were consistently found in every specimen of each group. Gutta-percha/Epiphany (0.01 ± 0.01) demonstrated significantly less voids than Resilon/AH Plus (0.3 ± 0.02), Resilon (control) (0.02 ± 0.01) and gutta-percha (control) (0.03 ± 0.01) groups (P < 0.05) whereas it was no statistically different from Resilon/Epiphany (0.02 ± 0.01) and gutta-percha/AH Plus (0.02 ± 0.01) (P > 0.05).

Conclusions Computed tomography allows detailed 3D observation of root fillings and may play an important role in the analysis of their quality.

R3.66

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Bond strength to root canal dentine following different irrigation protocols using a new testing method

Aim To evaluate the bond strength of root canal sealers to dentine using a pull-out test.

Methodology Root canals of 240 premolars were instrumented to size 60, 0.02 taper, decoronated to a remaining root canal length of 8 mm and embedded in epoxy-resin. Teeth were divided into six groups according to the sealers used: group 1: AH Plus (DeTrey Dentsply, Germany), group 2: EndoREZ (Ultradent, USA), group 3: Epiphany (Pentron, USA), group 4: FibreFill (Pentron, USA), group 5: Maxcem (KerrHawe, Switzerland), group 6: RelyX Unicem (3M Espe, Germany). Groups were separated into four subgroups according to different irrigation protocols (subgroup, irrigation sequence): a) NaOCl 5%; b) citric acid 40%/NaOCl 5%; c) citric acid 40%/NaOCl 5%/Alcohol 70%; d) citric acid 40%/NaOCl 5%/sodium ascorbate 10%/distilled water. Spreader blanks adjusted to size 60, 0.02 taper, were silica-coated and inserted into the root canal following placement of the sealers. After setting of sealers (7days/ 37°C/100% humidity) the spreaders were pulled out of the root canal using a universal testing machine (Zwick, Germany; cross hea! d speed: 1 cm/min). Statistics were computed using SPSS Win 14.0 (Kolmogorov-Smirnov test, ANOVA, multifactorial ANOVA (MFA). t-test).

Results Fracture modes were predominantly adhesive at the sealerdentine interface or cohesive. Bond strength values were (MPa/SD): group 1a: (2.2/1.3); group 1b: (4.7/2.4); group 1c: (3.3/1.5); group 1d: (5.8/2.5); group 2a: (6.0/3.6); group 2b: (10.3/4.3); group 2c: (10.7/3.7); group 2d: (8.7/3.6); group 3a: (5.6/3.6); group 3b: (4.0/2.2); group 3c: (6.1/2.4); group 3d: (4.7/2.0); group 4a: (8.0/ 6.3); group 4b: (5.8/3.2); group 4c: (7.9/5.9); group 4d: (11.3/3.8); group 5a: (10.9/2.7); group 5b: (11.7/5.6); group 5c: (10.1/7.5); group 5d: (6.6/1.5); group 6a: (11.6/4.6); group 6b: (11.4/ 4.0); group 6c: (12.6/4.6); group 6d: (9.9/3.3). Pull-out bond strength significantly depended on the sealer (MFA, P < 0.001) and the combination sealer/irrigation (MFA, P < 0.05) but not on the irrigation itself (MFA, P > 0.05). **Conclusions** Sealers had different pull-out bond strengths to dentine; the irrigation protocol was a confounding influence.

R3.67

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Shear bond strength of a self-etching, dual-curable sealer: MetaSEAL

Aim To test shear bond strength (SBS) of three resin based sealers (AH Plus, Epiphany, and MetaSEAL) to dentine and to evaluate if a dual-cure adhesive could improve bond strength.

Methodology Occlusal enamel of seventy-two extracted human third molars was removed, dentine surfaces were exposed (#600 SiC paper), treated with 5.25% NaOCl and 17% EDTA and randomly divided into six groups (n = 12). About 3 mm long sections of polyethylene tubing were filled with one of the sealers: AH Plus (Dentsply, Germany), Epiphany (Pentron, USA), and MetaSEAL (Parkell, USA) and placed on the dentine surfaces (Group 1–3). Bond It (Pentron) was used before placing the sealers in Groups 4, 5 and 6. The specimens were stored in an incubator (37° C, 1 month) and subjected to a shear load (0.5 mm min⁻¹). The SBS values were calculated (MPa) and statistically analyzed (two-way ANOVA, Tukey HSD tests).

Results Shear bond strength values (Mean ± SD) were as follows respectively: Group 1 (AH Plus): 2.04 ± 0.93 ; Group 2 (Epiphany): 2.15 ± 1.07 ; Group 3 (MetaSEAL): 0.56 ± 0.36 ; Group 4 (AH Plus + Bond It): 4.13 ± 1.1 ; Group 5 (Epiphany + Bond It): 3.23 ± 0.94 ; Group 6 (MetaSEAL + Bond It): 4.68 ± 1.19 . Epiphany and AH Plus sealers had significantly higher bond strength values than MetaSEAL (P < 0.05). MetaSEAL had better bond strength values than Epiphany and values were comparable to AH Plus when all the sealers used with Bond It (P < 0.05). When failure modes were evaluated, Epiphany revealed mostly mixed failure; AH Plus had 100% and MetaSEAL had mostly cohesive failure.

Conclusions The use of the dual-cure dentine bonding agent Bond It improved the adhesive performance of Epiphany, AH Plus and MetaSEAL sealers.

R3.68

Abstract Withdrawn.

R3.69

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Chemical analysis of MTA before and after hydration with water and a water/serum mixture

Aim To determine and compare the major constituents and crystalline structures present in commercial Mineral Trioxide Aggregates (MTAs) and Portland cements before and after hydration with water or a water/serum mixture.

Methodology Materials examined: Grey and white MTA (Angelus and ProRoot), a trial MTA and grey and white Portland cements (Lafarge, Chilton, Oxford, UK).

Methods: The elemental content of dry powder, and cured samples after mixing with water or a 50:50 water : defibrinated horse serum mixture was determined under SEM with Energy Dispersive Analysis by X-ray (EDAX). Additional samples (n = 2) of each material were analyzed by X-ray diffraction (XRD) to determine the major compounds and phases present.

Results White MTAs and white Portland cement were composed primarily of calcium, silicon, aluminum and oxygen. Bismuth was present only in MTAs. Grey MTAs and Portland cement had similar composition, but contained more iron and magnesium than white versions. There was no difference in major elemental composition between dry powder and hydrated material. The addition of serum had no detectable influence on composition. XRD analysis showed that all types of MTA and Portland cement were composed mainly of tricalcium silicate, dicalcium silicate and tricalcium aluminate with bismuth oxide detected only in MTAs. Calcium hydroxide was found in all the set materials. The presence of serum had no discernable effect on the crystal phases found within cured specimens of MTAs and Portland cements.

Conclusions Incorporation of defibrinated horse serum had no effect on the major elemental composition or crystalline structure of hydrated MTAs and Portland cements. All types of MTA were shown to have similar constituents to Portland cements except for the addition of bismuth oxide. White MTAs and white Portland cement did not contain iron and magnesium. Calcium hydroxide was found in the cured materials.

R3.70

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Contact angle measurements of amalgam restorative material following bleaching

Aim To detect the effect of bleaching on the wettability of amalgam *ex vivo*.

Methodology Thirty-six samples of amalgam packed into silicon moulds and smoothed with fine polishing disks, were treated with either 10% carbamide peroxide (CP), 10% hydrogen peroxide (HP) or 0.1 M phosphate buffer-control for 1 or 3 days. Measurements were performed underwater by measuring the contact angles of air on surfaces of amalgam samples using a goniometer (Olympus, VS-IV, Tokyo, Japan) with captive bubble technique. Differences between the means of each group were analyzed statistically using Mann–Whitney *U* test at 95% level of significance.

Results Both bleaching materials and the buffer control decreased significantly the contact angle levels of amalgam samples after 1 and 3 days incubation (P < 0.05). When compared with the control group, there was a significant difference in contact angle levels following 1 and 3 days incubation with the bleaching materials (P < 0.05).

Conclusions Bleaching materials increased the wettability of amalgam.

R3.71

Abstract Withdrawn.

R3.72

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Fracture resistance of roots filled with new resin based sealers

Aim To test the effect of three different resin based sealers on resistance to root fracture and to evaluate whether using guttapercha (GP) or Resilon (R) has an effect on fracture strength.

Methodology Eighty extracted human maxillary incisors were randomly divided into eight groups (n = 10), prepared (size 50) and filled using cold lateral condensation except groups 7 and 8 as follows: Group 1: AH Plus (Dentsply) + GP; Group 2: AH Plus + R; Group 3: Epiphany (Pentron) + GP; Group 4: Epiphany + R; Group 5: CZ-S2000 (MetaSEAL,Parkell) + GP; Group 6: CZ-S2000 (MetaSEAL) + R; Group 7: (non-prepared), Group 8: (prepared, kept unfilled). After storing 1 week (100% humidity, 37°C), the coronal lingual walls were reduced by 2 mm, placed into acrylic blocks so that 10 mm of buccal roots were exposed and then placed in a specially designed steel pad at an angle of 15 degrees to the long axis of the root. Load was applied (0.5 mm min⁻¹) at the junction of the buccal wall and root canal. The force required to fracture each root was recorded as kg and statistically analyzed (one way ANOVA, Tukey HSD tests).

Results Mean fracture values were as follows: Group 1: 48.75 ± 7.12b, Group 2: 54.52 ± 5.91bc, Group 3: 60.45 ± 5.42cd, Group 4: 67.96 ± 5.91de, Group 5: 57.85 ± 4.03c, Group 6: 65.47 ± 4.70de, Group 7: 70.70 ± 5.24e, Group 8: 26.65 ± 4.71a (Different letters indicate significantly different groups, P < 0.001). Instrumentation of the root canals significantly weakened the tooth structure. All the sealers increased resistance to fracture. Epiphany or CZ-S2000 (MetaSEAL) in combination with Resilon groups had similar fracture resistance to an unprepared root.

Conclusions Preparation of root canals weakened roots. However, initial strength can be gained with the use of Epiphany or CZ-S2000 (MetaSEAL) sealers in combination with Resilon.

R3.73

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Effect of different restorative materials on the fracture resistance of root filled teeth

Aim To evaluate the fracture resistance of root filled mandibular molars with mesioocclusodistal (MOD) cavity preparations restored with three different restorative materials.

Methodology Fifty sound extracted mandibular molars were randomly divided into five groups (n = 10) and prepared as follows: Group 1: Intact teeth; Group 2: MOD cavities were prepared, root canals were filled and no restoration was placed; Group 3: Teeth were prepared as Group 2 and restored conventionally with amalgam; Group 4: Teeth were prepared as Group 2 and restored with composite resin (Clearfil Photoposterior, Kuraray, Tokyo, Japan); Group 5: Teeth were prepared as Group 2 and restored with direct ceramic inlay material (Estenia, Kuraray, Tokyo, Japan). All specimens except Group 2 were loaded to failure by the chewing simulation device ($60.000 \times 50 \text{ N}$, 1.3 Hz) in an artificial environment at 37°C. The compressive load to each tooth was applied with a constant speed of 1 mm min⁻¹ until

fracture occurred. The forces required to fracture teeth were recorded as Newton and statistically analyzed using Kruskal–Wallis and Mann–Whitney U tests.

Results Mean fracture values were as follows: Group 1: 2485.28 ± 193.89a, Group2: 533.85 ± 59.36b, Group 3: 1705.83 ± 135.69c, Group 4: 2033.34 ± 137.63d, Group 5: 2121.30 ± 156.54d. (Different letters indicate significantly different groups, *P* < 0.05). The amalgam restored group (Group 3) had the lowest resistance to fracture when compared with composite resin (Group 4) and inlay groups (Group 5). No statistically significant differences were found between the composite resin and the inlay groups. Although composite resin and inlay groups gave high fracture strength values, their values were still less than intact teeth.

Conclusions Teeth restored with direct intra-coronal adhesive restorations were significantly stronger than those teeth restored with conventional amalgam. However, none of these materials was able to restore completely the fracture resistance of root filled molar teeth with MOD cavity preparation.

R3.74

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Ex vivo structural resistance of immature teeth after different root reinforcements

Aim To compare strengthening ability of light-cured posterior composite resin with or without fibre reinforcement in immature roots restored with two types of posts.

Methodology Seventy-two maxillary central incisor teeth were divided into six groups of 12 teeth each. Teeth in all groups except the negative controls (group 1) were prepared with no. 6 Peeso drills (1.7 mm) through the apex in order to simulate immature roots. After smear layer removal, root canals were dried and a mixture of antibiotic drugs was placed. After 21 days, the antibacterial dressing was removed and root-ends were filled with a 5 mm barrier of MTA Angelus (Angelus, Brazil). In group 2 the canal walls were reinforced with Clearfil Photo Posterior composite (Kuraray, Japan), cured with the help of a translucent curing post (Luminex, Dentatus, Sweden) and restored with Everstick posts (Stick Tech., Finland). Group 3 was reinforced as in group 2 with the addition of Ribbond under the composite. Group 4 was reinforced with posterior composite and restored with Fibre post (GC, Japan). Group 5 was restored as in group 4 with the addition of Ribbond under the composite. Group 6 received no treatment (positive control). Teeth were submitted to compressive forces at an angle of 45 degrees and a speed of 1 mm min⁻¹. The peak load to fracture was recorded in Newton and statistically analyzed (One way ANOVA and Tukey HSD).

Results Fracture resistance values (Mean \pm SD) were as follows: group 1: 1431.74 \pm 237.06; group 2: 1212.88 \pm 426.14; group 3: 1184.68 \pm 287.61; group 4: 1139.93 \pm 422.11; group 5: 1191.60 \pm 306.01; group 6: 336.61 \pm 219.04. There was no significant difference among the groups (*P* > 0.05) except group 6. **Conclusions** Reinforcement with posterior composite, fibre and different posts significantly increased fracture resistance of simulated immature teeth.

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R3.75

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Effect of cusp reduction on the fracture resistance of endodontically accessed premolars restored with MOD composite restoration

Aim To evaluate the effect of cusp reduction on the fracture resistance of composite-restored premolars.

Methodology Eighty extracted human premolars with similar dimensions were selected. Endodontic-access cavities were prepared in 60 teeth and divided into four test groups (n = 15). Mesialocclusal-distal (MOD) cavities were prepared up to 1 mm from the cemento-enamel-junction. In groups R1, R2 and R3 the mandibular buccal cusps and the maxillary palatal cusps were reduced to a height of 3.5 mm from the cavity floor. Additionally, the cusp wall thickness was reduced to be: R1 = (1-1.5 mm), R2 = (1.5-2 mm)and R3 = (2-3 mm). In group NR (non-reduced cusp), the cusp wall thickness was 2-3 mm. MOD cavities and the reduced cusps were restored with Tertic EvoCeram® (Ivoclar Vivadent, Lichtenstein) and Optibond FL® (Kerr, CA, USA). Ten intact premolars and 10 unrestored MOD-prepared premolars served as positive and negative controls, respectively. Specimens were submitted to 1.2 Million cycles with 45-60 N loading forces in a masticationsimulation-machine. The mandibular buccal walls and the maxillary palatal walls were subjected to compressive force till fracture using a Universal-testing-machine (Loading angle = 30° , cross head speed = 0.5 mm min^{-1}). The load to fracture was recorded and for each group the mean and the Confidence Interval (CI) were calculated.

Results There was a statistically significant difference between the mean fracture resistance of the non-reduced cusp group NR: 305 N [95% CI 264: 347] compared with the three reduced cusp groups; R1: 603 N [95% CI 475; 731], R2: 712 N [95% CI 591; 833] and R3: 697 N [95% CI 583; 812].

Conclusions Cusp reduction significantly increased the fracture resistance of endodontically-accessed premolars restored with MOD-composite, no statistically significant difference to intact teeth was found. MOD-composite restoration without cusp reduction did not completely restore the fracture resistance of premolars.

R3.76

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Micro-push-out bond strengths of four fibre-reinforced composite post systems and two luting cements

Aim To compare the bond strengths of two types of dual luting agents used for cementation of four different fibre reinforced composite (FRC) posts using push-out test and to evaluate the failure modes of these systems.

Methodology Eighty human maxillary central incisors were divided into eight groups (n = 10), decoronated, root filled and restored with one of the following post systems: Groups 1, 2, 3 and 4:Translucent quartz FRC posts (DT Light and DT Light SL, VDW, Munich, Germany); Groups 5, 6: Opaque glass FRC post (FRC Postec Plus, Ivoclar Vivadent, Schaan, Liechtenstein) and Groups 7, 8: individually formed electrical glass fibre post (Everstick,

StickTech Ltd, Turku, Finland). Cementation was performed with two types of dual-polymerizing resin luting agents: Variolink II (Ivoclar Vivadent, Schaan, Liechtenstein) (Groups 1, 3, 5, 7) and a new self adhesive resin cement RelyX Unicem (3M ESPE, Seefeld, Germany) (Groups 2, 4, 6, 8). Slices with a thickness of 1.00 ± 0.05 mm were prepared from the coronal third of each root using a low speed saw (Minitom, Struers, Germany). Push-out tests were performed at a cross-head speed of 1 mm min⁻¹ using a universal testing machine (Testometric, UK) and the data was statistically analyzed (Kruskal–Wallis and Tamhane tests, P < 0.05). Fracture modes were evaluated at ×40 magnification.

Results A significant difference was found among the groups (P < 0.05). FRC posts luted with Variolink II showed higher bond strengths and the groups ordered as 5, 1, 3, 7, 6, 2, 4 and 8 with the values (MPa) (means/SD): 13.80/4.99, 13.77/3.78, 12.20/4.79, 9.39/2.48, 9.21/7.76, 7.25/2.71, 3.89/4.41, and 3.77/1.20, respectively. Adhesive failures between dentine and cement were observed more than cohesive failures in cement or post.

Conclusions Push-out bond strengths can be affected by luting agent and post type. Variolink II and fibre post combinations resulted in high bond strength values.

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R3.77

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Cement thickness and presence of voids using two different fibre-reinforced composite posts in oval canals

Aim To measure the cement layer thickness and the number of voids after luting preformed or anatomically formed fibre-reinforced composite (FRC) posts in teeth with oval root canals.

Methodology Ten freshly extracted human premolars, intact and completely formed were selected on the basis of similar root and canal dimensions. The root canals were prepared and filled and prepared for the insertion of a fibre post. Glass-fibre preformed posts were luted with a dual-cure flowable composite resin in five specimens (group 1). In the other five specimens, experimental FRC posts were modelled in order to give them a shape as close as possible to the anatomy of the root canals, using a cast previously created as a guide. The specimens were analyzed with microcomputed tomography (Skyscan 1072, Skyscan, Belgium). The cross-sections corresponding to the cemento-enamel junction (CEJ) and 3 and 6 mm apically to the CEJ were analyzed to measure the mesio-distal and the bucco-lingual cement thickness and the number of voids within the luting material or at its interfaces. A Student t-test was performed to determine significance differences between groups with levels of significance set at P < 0.05.

Results The layer of cement was significantly thinner in group B at the CEJ and 3 mm apically to the CEJ in the bucco-lingual dimension (P < 0.05), and at the CEJ and 6 mm apically to the CEJ in the mesio-distal dimension (P < 0.05). The number of voids within the cement layer was significantly lower in teeth from group B (P < 0.05).

Conclusions As a result of the adaptation of the fibre post, the thickness of the cement layer was consistently lower in Group B and the formation of voids was less likely.

R3.78

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Effect of Nd : YAG laser irradiation on root canal walls and microleakage of root filled teeth

Aim To evaluate the effect of Nd : YAG laser irradiation on root canal walls as compared with the use of 17% EDTA irrigant after root canal preparation and irrigation with 2.5% NaOCl, and the degree of microleakage after canal filling *ex vivo*.

Methodology Thirty single-rooted teeth were instrumented (crown-down/step-back) up to a size 40 K-file (rinsed with 3 mL 2.5% NaOCl between each file) and then divided into three groups of 10 teeth: group 1 with EDTA final rinse (3 mL, 3 min) and additional 2.5% NaOCl rinse, group 2 with EDTA final rinse and Nd : YAG laser irradiation (1.5 W, 15 Hz, four times 5 s with a 20 s interval) (Fidelis Plus, Herzele, Belgium), group 3 without final rinse and only Nd : YAG laser irradiation. All 30 teeth were filled with cold lateral condensation of gutta-percha and AH26 (Dentsply De Trey, Germany). After storage in an environment of 95–100% humidity for 48 h the teeth were submitted to capillary flow porometry (PMI, USA) in order to assess the minimal, mean and maximal through-pore diameters of each experimental tooth. The results of the tests were statistically evaluated using non-parametric tests. The level of significance was set at 0.05.

Results No statistically significant differences were demonstrated for the minimal, mean and maximum pore diameters between the three groups. There was evidence of melted and evaporated smear layer in group 3, the typical melting effects were less pronounced in groups 1 and 2.

Conclusions There was no statistically significant influence of the final EDTA rinses, of Nd:YAG laser irradiation, and the combined use of EDTA and Nd : YAG laser on microleakage in root filled teeth. The use of EDTA whether or not prior to Nd : YAG laser irradiation had a significant influence on root canal wall cleanliness.

R3.79

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Longitudinal study on the influence of Nd : YAG laser irradiation on microleakage of two filling techniques

Aim To evaluate apical and coronal microleakage of filled root canals *ex vivo* after root canal treatment with and without Nd : YAG laser irradiation and comparing cold lateral condensation and hybrid gutta-percha condensation.

Methodology Three hundred single-rooted teeth were instrumented to a size 40 K-file and divided into a group of 240 teeth (DP) and a group of 60 teeth (FTM). Each group was then divided into four equal subgroups according to filling technique (cold lateral condensation – CLC; hybrid condensation – HC) and whether or not Nd : YAG lased (Nd). The Nd : YAG laser (Fidelis Plus, Herzele, Belgium) was operated at 1.5 W, 15 Hz, four times for 5 s with a 20 s interval. Leakage was assessed with a dye penetration technique (Indian ink) (DP) or with the fluid transport model (FTM), and as a function of time after 48 h, 1, 6 and 12 months. For the DP-group further

division into four groups of 15 teeth each was needed for each evaluation time. Coronal and apical dye leakage were assessed in the same experimental teeth after splitting them longitudinally. Through-and-through leakage (L in microL/day) was measured for 48 h under a pressure of 1.2 atm using the fluid transport model and recorded as L = 0 (L1), 0 < L < or = 10 (L2), L > 10 (L3).

Results Apical and coronal dye leakage was observed in all groups. Statistically significant differences were found between 'HC' and 'HC + Nd' for apical leakage at 1, 6 and 12 months; and between 'CLC' and 'CLC + Nd' at 6 and 12 months. No significant differences were found between laser irradiated and non-laser irradiated groups with FTM. All apical foramina remained patent whether Nd : YAG lased or not.

Conclusions Pulsed Nd : YAG laser irradiation following root canal preparation reduced apical leakage in teeth with root fillings placed for over 6 months especially in association with hybrid gutta-percha condensation.

R3.80

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Sealing ability of CZ-S2000 (MetaSEAL) in conjunction with different canal filling techniques

Aim To evaluate the apical sealing ability of CZ-S2000 (MetaSEAL, Parkell, USA) in conjunction with different filling techniques.

Methodology Seventy-two extracted human mandibular straight single-rooted teeth with mature apices were prepared using a stepback technique and divided into four experimental groups (n = 15). An additional six teeth were prepared as controls (three for positive and three for negative controls). The experimental groups were root filled with CZ-S2000 (MetaSEAL) using either cold lateral condensation, vertical condensation, Thermafil or the Ultrafil techniques. Fluid movement along the filled canals was measured using a fluid filtration method. Measurements were made at 2 min intervals for 8 min. The data was calculated as μ L/min and statistically analyzed using one-way ANOVA and Duncan test.

Results The mean and standard deviation for each group were respectively as follows: Cold lateral condensation: 0.512 ± 0.162 ; Vertical condensation: 0.625 ± 0.241 ; Thermafil: 3.225 ± 0.958 ; Ultrafil: 2.377 ± 1.221 . Cold lateral and vertical condensation had significantly less fluid movement than the Thermafil and Ultrafil groups. Thermafil group had the highest fluid movement values when compared with the other groups.

Conclusions CZ-S2000 (MetaSEAL) had less fluid movement with cold lateral and vertical condensation techniques when compared with Thermafil and Ultrafil techniques.

R3.81

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Assessment of the apical sealing ability of Resilon/Epiphany using a fluid filtration technique

Aim To compare the sealing ability of Resilon/Epiphany and guttapercha with Diaket sealer. Methodology Eighty single-rooted mandibular premolar teeth with one canal and having similar dimensions were selected. All canals were prepared with the 0.06 ProFile rotary system to size 40. There were four groups of 20 teeth each. Group 1: filled with guttapercha and Diaket after the final irrigation using EDTA, NaOCl, EDTA, distilled water. Group 2: filled with Resilon /Epiphany after the final irrigation using EDTA, NaOCl, EDTA, distilled water. Group 3: filled with gutta-percha and Diaket after the final irrigation using EDTA, NaOCl, EDTA, ascorbic acid. Group 4: filled with Resilon / Epiphany after the final irrigation using EDTA, NaOCl, EDTA, ascorbic acid. Positive and negative control groups were also prepared. Following the irrigation and filling of the root canals using the lateral compaction technique fluid movement along the specimen was measured using the fluid filtration technique. Results were evaluated statistically with one way and univariate analysis of variance.

Results Gutta-percha groups allowed significantly more fluid movement then the Resilon groups. The ascorbic acid groups revealed less fluid movement then the distilled water groups. The Resilon groups with self-etched primer and Epiphany resin root canal sealer were significantly more resistant to fluid movement. Furthermore the use of different irrigants had an effect on the fluid movement. Using ascorbic acid instead of distilled water after conventional irrigation solutions reduced leakage in both guttapercha Diaket sealer and Resilon/Epiphany groups.

Conclusions The Resilon groups with self etched pirmer and Epiphany resin root canal sealer were significantly more resistant to fluid movement then gutta-percha and Diaket sealer groups. Furthermore, the use of different irrigants had an effect on fluid movement. Long term clinical studies are needed to determine if outcome studies support the use of this material.

R3.82

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Evaluation of glucose penetration along gutta-percha and thermoplastic synthetic polymer-based root filling systems

Aim To compare penetration of glucose along gutta-percha and thermoplastic synthetic polymer-based root filling systems.

Methodology Premolar teeth (n = 156) instrumented to an apical size 40, 0.06 taper were divided into twelve experimental (n = 12) and two control groups (n = 6). The root canals were filled with either gutta-percha (groups 1–6) or resilon (groups 7–12) core points combined with AH Plus (De-Trey-Dentsply, Germany) (groups 1, 4, 7 and 10), Ketac-Endo (Espe Gmbh, Germany) (groups 2, 5, 8 and 11) or Epiphany (Pentron Clinical Technologies, Wallingford, CT, USA) (groups 3, 6, 9 and 12) using cold lateral compaction (groups 1–3, groups 7–9) or System B (SybronEndo, USA) with Obtura II (Obtura Spartan, Fenton, MO, USA) (groups 4–6, groups 10–12). The concentration of glucose in the apical reservoir was measured spectrophotometrically at 1, 8, 15, 22 and 30 days. The percentage of each group that allowed glucose penetration was also calculated. The data was recorded as mmol/L and statistically analyzed with Kruskal–Wallis and Mann–Whitney U tests (P = 0.05).

Results Throughout the experimental period the Resilon/Ketac-Endo/cold lateral compaction group (group 8) were associated with the highest mean cumulative glucose penetration (mmol/L) (17.27 \pm 10.32), whereas resilon/AH Plus/cold lateral compaction group (group 7) had the least (3.36 \pm 4.65) (*P* < 0.05). After 30 days fewer specimens in the gutta percha/Epiphany/cold lateral compaction (group 3) leaked compared with the other groups. Glucose penetration was affected by the filling technique in some groups: Ketac-Endo either with gutta-percha (group 2) or Resilon (group 8) had significantly less glucose penetration with warm techniques, whereas Gutta-percha/Epiphany (group 3) had significantly less glucose penetration with cold techniques (P < 0.05).

Conclusions All material/technique combinations allowed glucose penetration. Gutta-percha/AH Plus combinations allowed similar patterns of glucose penetration to Resilon/Epiphany combinations. **Acknowledgements** The authors thank Assist Prof. Sual Ozkorucuklu, for the design of glucose penetration apparatus, Assist Prof Hikmet Orhon, for his help with the statistically analysis and to Research Council of Suleyman Demirel University for the support in project no: 1180-D-05.

R3.83

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An assessment of the sealing ability of three root canal sealers using different instrumentation techniques

Aim To evaluate the sealing ability of AH Plus (DeTrey Dentsply, Germany), Epiphany (Jeneric/Pentron, USA) and EZ Fill Epoxy Root Canal Cement (EDS, USA) when used with different filling techniques. Methodology Sixty extracted single-rooted teeth were randomly divided into six groups of 10 teeth each. The thirty teeth were handfiled to a size 40 master K-file (Mani, Japan) using a step-back technique and filled using lateral condensation with gutta-percha and AH Plus (group 1), gutta-percha and EZ Fill Epoxy Root Canal Cement (group 2) and Resilon/Epiphany (group 3). Another thirty teeth were prepared to the size 35, 0.06 taper by rotary FlexMaster nickel-titanium instruments (VDW, Germany) and filled using a single cone technique with 0.06 tapered gutta-percha and AH Plus (group 4), 0.06 tapered gutta-percha and EZ Fill Epoxy Root Canal Cement (group 5) and 0.06 tapered Resilon/Epiphany (group 6-ten teeth). Microleakage was evaluated using a dye penetration test (5% solution of methylene blue) for 48 h. The teeth were sectioned longitudinally and examined under stereo-microscope to assess dye penetration.

Results Linear dye penetration (mm) (means/SD): group 1 (1.38/ 2.14); group 2 (1.94/2.22); group 3 (8.97/4.56); group 4 (1.41/ 1.55); group 5 (1.79/1.99); group 6 (11.06/3.66). Tamhane's test showed that the Resilon/Epiphany allowed significantly greater leakage throughout the experimental period (groups 3 and 6). No significant differences (P > 0.05) existed between gutta-percha with AH Plus (groups 1 and 4) and gutta-percha with EZ Fill Epoxy Root Canal Cement (groups 2 and 5). No significant difference in leakage (P > 0.05) existed between the lateral condensation groups and single cone tapered 0.06 groups.

Conclusions Resilon/Epiphany showed gross leakage in all groups. The cold laterally compacted method and single cone method provided similar sealing ability.

R3.84

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An *ex vivo* comparison of the sealing ability of three endodontic sealers

Aim To compare the sealing ability of Real Seal, AH Plus and Gutta Flow.

Methodology Canals of 33 extracted maxillary incisors and canines were prepared chemo-mechanically with NiTi Tee (Poldent, Poland) up to size 45, 0.04 taper and irrigated with 5.25% NaOCl, 15% EDTA and sterile water. All teeth were randomly divided into three groups (n = 11). For root fillings the following techniques were used: Real Seal obturation system (Real Seal primer, sealer, and a single cone size 40, 0.04 taper) (SybronEndo, Orange, USA) (group 1), AH Plus (Dentsply De Trey, Germany) and a single guttapercha cone size 40, 0.04 taper (group 2), Gutta Flow (Coltene Whaledent, Germany) and a single gutta-percha cone size 40, taper 0.04 (group 3). The sealers were introduced into the canals with a lentulo spiral. Each tooth was coated with two layers of nail varnish except for the apical root-end. The specimens were further stored in a 2% methylene blue for 72 h at 37°C and cross sectioned in regular (1 mm) intervals. Depth of linear penetration was measured under a stereo microscope at 20× magnification. The extent of leakage among the groups was compared using the non-parametric Kruskal-Wallis analysis of variance and Mann-Whitney U test (P < 0.05).

Results Median leakage [mm] in groups 1, 2 and 3 and were 4.0, 7.0 and 1.0, respectively. Gutta Flow had significantly different distribution of leakage penetration than Real Seal (P < 0.02), and AH Plus (P < 0.02). There were no differences in leakage distribution between Real Seal and AH Plus.

Conclusions Gutta Flow had better sealing ability than Real Seal and AH Plus. The sealing ability of Real Seal and AH Plus were comparable.

R3.85

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Resilon-Epiphany as a root-end filling: the seal evaluated by CFP

Aim To compare the root-end sealing ability of the Resilon-Epiphany system (Pentron, USA) (RE) to warm Gutta Percha (Obtura II, Obtura-Spartan, USA) with AH26 (Dentsply De Trey, Germany) (GP) and Pro Root MTA Tooth-Colored Formula (Dentsply, USA) (MTA) in standard bovine root sections.

Methodology Twenty standard bovine root sections were prepared each 3 mm high with a diameter of 7 mm and an internal diameter of 2.5 mm. These sections were filled with RE. The filled sections were stored in an environment of 37° C and 95-100% humidity for 24 h, then exposed to distilled water and 24 h later submitted to capillary flow porometry (CFP) (PMI, USA) in order to assess the minimum, mean and maximum through-pore diameters of each root section. These sections were compared with sections filled with GP or MTA using the same methodology. The results of the tests were statistically evaluated using non-parametric tests. The level of significance was set at 0.05.

Results Significant differences were demonstrated for the minimum, mean and maximum pore diameters. Between GP and MTA no significant differences were demonstrated, but significant differences were shown between RE and GP and between RE and MTA for minimum, mean and maximum pore diameters. Pores in samples filled with RE proved smaller than pores in samples filled with GP or MTA.

Conclusions Under the conditions of this study, RE at 48 h demonstrated a better seal than GP and MTA. Future measurements at 1 month and 6 months will demonstrate whether these results are duplicated as a function of time.

R3.86

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Bacterial leakage in filled root canals using four root canal sealers

Aim To evaluate the sealing properties of four root canal sealers using a bacterial penetration model.

Methodology Root canals of 50 premolars were prepared to size 45, 0.04 taper and randomly assigned to one of the following groups: group 1 (AH Plus/gutta-percha); group 2 (Epiphany/ Resilon); group 3 (FibreFill/gutta-percha); group 4 (GuttaFlow/ gutta-percha). Five teeth served as positive controls, five as negative controls. Teeth were decoronated resulting in a canal length of 10 mm. Canals were filled and allowed to set for 7 days (100% humidity/37°C). After filling, teeth were mounted in a two-chamber set-up according to Torabinejad *et al.* (1990) and gamma sterilized. *Lactobacillus rhannosus* was used for the penetration test. Bacterial leakage was recorded when turbidity within the apical chamber was recognized. Statistical analyses (Kolmogorov–Smirnov test, Kruskal–Wallis test, Mann–Whitney test) were computed using SPSS Win 14.0.

Results Bacterial penetration was observed as follows [mean penetration time (days)/SD]: group 1 (21.2/1.9); group 2 (2.1/0.3); group 3 (16.7/0.9); group 4 (13.8/1.0); positive control group (1/0). None of the negative controls showed turbidity at the end of the observation period (>30 days). Epiphany had significantly faster bacterial penetration than the other sealers (Mann–Whitney test, P < 0.001). All possible pairs of groups showed significant differences (Mann–Whitney test, P < 0.01). All penetration longer than the other sealers.

Conclusions Epiphany allowed a penetration of *Lactobacillus rhamnosus* to the apex in the shortest time when used in combination with Resilon cones. The other sealers prevented bacterial leakage to the apex for at least 2 weeks.

R3.87

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Coronal microleakage of two root-end filling materials using a polymicrobe marker

Aim To evaluate bacterial coronal microleakage of MTA and amalgam as root-end fillings of roots orthogradly filled with guttapercha and three root canal sealers.

Methodology A total of 108 single rooted teeth were randomly divided into three groups of 32 teeth each and positive and negative control groups of six teeth. Following root canal preparation the canals were filled using cold lateral compaction of gutta-percha and either Diaket, AH Plus or Ketac Endo. Each of the three groups was further divided into two subgroups of 16 teeth and the apical root ends were re-sected, cavities prepared and filled with either MTA or zinc free amalgam. The samples were incorporated in a model system so that the apical part of the sample was dipped into uncontaminated buyon, while 0.5 mL of the buyone contaminated with a polymicrobial marker of five facultative anaerobes

(Streptococcus Mutans 6715 WT, Streptotoccus Mitis, Lactobacillus Species NCTC1723, Staphylococcus Aureus ATCC 29213, Enterococcus Faecalis ATCC 29212) was placed on the coronal part of each sample. The teeth were checked for bacterial leakage daily for 90 days and the exact day of buyon contamination was recorded for every sample. The obtained data were analyzed using descriptive statistics, variance analysis, Sheffe post hoc test and Kaplan Meier test.

Results The longest period before leakage was observed with combination of Diaket sealer and MTA root-end filling (76.9 ± 14.8 days) followed by AH Plus and MTA (66.1 ± 18.7), Diaket and amalgam (60.0 ± 23.1), AH Plus and amalgam (56.9 ± 22.1), Ketac Endo and MTA (42.1 ± 17.8) while the shortest period before leakage was observed in the Ketac Endo and amalgam group (40.0 ± 17.24). Groups sealed with Diaket and AH Plus leaked after a significantly longer time (P < 0.05) than groups with Ketac Endo. Groups filled with MTA leaked after a significantly longer time (P < 0.05) than amalgam.

Conclusions MTA in combination with Diaket were associated with the longest period of time prior to leakage.

R3.88

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Comparison of the sealing of different dentine bonding adhesives as root-end filling materials: a bacterial leakage study

Aim To compare the root-end sealing of the different dentine bonding adhesives using a bacterial leakage system.

Methodology One hundred extracted single-rooted teeth were instrumented and root-ends were resected and ultrasonically prepared. All teeth were then randomly assigned to six experimental groups (n = 15) and two control groups (positive and negative control, n = 5) for root-end filling with iBond (Heraeus Kulzer, Hanau, Germany), G-bond (GC Co., Tokyo, Japan), Xeno III Bond (Dentsply/Caulk, Milford, DE, USA), Adhese Bond (Ivoclar Vivadent, Schaa, Liechenstein), Clearfil Protect Bond (Kuraray, Osaka, Japan), Clearfil Tri-S Bond (Kuraray). All preparations were further restored with a resin composite (Renew; Bisco, Schaumburg, IL, USA). Nail varnish was applied to all external root surfaces to the level of the resected root-ends to prevent lateral microleakage. Samples were sterilized in an ethylene oxide sterilizer for 12 h. The apical 3-4 mm of the roots were immersed in BHI culture medium with phenol red indicator within culture chambers. The coronal access of each specimen was inoculated every 48 h with a suspension of Enterococcus faecalis. Bacterial leakage was monitored every 24 h for 4 weeks. The data obtained were analyzed using the chi-square test, with P = 0.05 as the level for statistical significance.

Results All positive controls leaked within 24 h; none of the negative controls leaked. Under the conditions of the study, despite some variations, there were no statistically significant differences in the rate of bacterial leakage among the experimental test materials at 1-4 weeks (P > 0.05).

Conclusions The type of dentine bonding adhesive had no influence in bacterial leakage of the root-end fillings.

R3.89

Abstract Withdrawn.

R3.90

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Secondary protective seal of Clearfil SE Bond over root fillings using AH Plus sealer

Aim To evaluate microleakage of a secondary protective seal of Clearfil SE Bond and Tetric flow following root filling using AH Plus sealer with different methods of cleaning the access cavity.

Methodology Forty extracted maxillary and mandibular molars were used for the study. All root canals were prepared to size 60, 0.02 taper and filled with AH Plus (DeTrey Dentsply, Germany) and a single size 55, 0.02 taper gutta percha cone. Teeth were randomly divided into four groups of 10 teeth each. After removal of excess sealer with an ethanol-moistured foam pellet, different pretreatments were performed: no further pre-treatmernt (group 1), preparation with an abrasive water-cooled diamond bur (group 2), and etching with 37% H3PO4-gel for 10 s (group 3) or bur preparation followed by etching (group 4). Clearfil SE Bond (Kuraray, Japan) and two thin layers of Tetric flow (Ivoclar Vivadent, Liechtenstein) were applied. A dye penetration test (5% methylene blue dye) was carried out using entrifugation for 3 min at 30 G followed by examination under a stereo microscope (40×) with an image processing software. Linear dye penetration and dye penetration area were evaluated. Statistical tests (Kolmogorov-Smirnov-tests, Kruskal-Wallis-tests (KWT) and Mann-Whitneytests (MWT)) were carried out using SPSS Win 14.0 (SPSS Corp., USA).

Results Linear dye penetration (mm)/dye penetration area (mm²) (Mean; SD): group 1 (1.1; 1.4)/(1.4; 2.6); group 2 (0.7; 0.9)/(1.0; 1.8); group 3 (0.4; 0.5)/(0; 0); group 4 (2.1; 0.6)/(3.1; 2.8). Significant differences were apparent (KWT; P < 0.01). Group 4 had significantly more leakage than groups 2 and 3 (MWT; P < 0.05) for Linear dye penetration and dye penetration area. For group 3, dye penetration less than 1 mm occurred in all specimen. **Conclusions** Within the limits of this study, when using Clearfil SE bond for a secondary protective seal acid etching or bur preparation of dentine following the use of AH plus sealer is recommended.

R3.91

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Ex vivo evaluation of leakage of immature teeth after different root reinforcing restorations

Aim To evaluate the leakage of simulated immature teeth after restoration with apical plugs of MTA-Angelus (Angelus, Brazil), light-cured posterior composite resin in combination with or without fibre reinforcement (Ribbond, USA) and with two different types of posts.

Methodology Forty-eight central incisor teeth were divided into five groups (n = 12), prepared with no. 6 Peeso drills (1.7 mm) through the apex in order to simulate immature roots. After smear

layer removal, root canals were dried and a mixture of antibiotic drugs was placed. After 21 days, the antibacterial dressing was removed and a 5 mm of apical MTA plug was placed. In group 1, the canal walls were reinforced with Clearfil Photo Posterior composite (Kuraray, Japan), cured with the help of a translucent curing Luminex post (Dentatus, Sweden) and then restored with EverStick posts (Stick Tech., Finland) and Panavia F (Kuraray, Japan). Group 2 was reinforced as in group 1 with the addition of Ribbond under the composite. Group 3 was reinforced with posterior composite and restored with fibre posts (GC, Japan). Group 4 was restored as in group 3 with the addition of Ribbond under the composite. One additional tooth was prepared and not restored (positive control). Sealing ability of the samples was evaluated using a fluid transport system. The data was calculated as Lp and statistically analyzed (One way ANOVA and Tukey HSD). **Results** Leakage values (Mean ± SD) were as follows: Group 1: 0.0052 ± 0.003 ; Group 2: 0.014 ± 0.0037 ; Group 3: 0.0072 ± 0.0026 ; Group 4: 0.0074 ± 0.0032 . There was no significant difference among the groups (P > 0.05) except group 2. Conclusions Reinforcement of simulated immature teeth with or without fibre reinforcement and with two different types of posts provided an effective seal except the fibre reinforced EverStick group. When immature teeth are restored with composite resin and EverStick posts, additional fibre reinforcement is not necessary.

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R3.92

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Coronal leakage analysis of adhesively luted glass fibre posts

Aim To evaluate the coronal sealing properties of different luting composites for adhesive post cementation.

Methodology Root canals of 60 premolars were instrumented to size 60, 0.02 taper and obturated with AH Plus (DeTrey Dentsply, Germany) and gutta-percha. After sealer setting for 7 days (100% humidity/37°C) post space was created using a size 2 NTI FiberMaster post space bur (NTI, Germany). Roots were decoronated to a post space length of 9 mm. Posts were luted using the following six luting cements: group 1: Embrace (Pulpdent, USA), group 2: Maxcem (KerrHawe, Switzerland), group 3: Multicore Flow (Ivoclar Vivadent, Liechtenstein), group 4: Panavia F 2.0 (Kuraray, Japan), group 5: Paracem (Coltène-Whaledent, Switzerland), group 6: RelyX Unicem (3M Espe, Germany) (n = 10). Materials were used according to the manufacturers' instructions. Posts were coronally reduced and completely coated with two layers of nail varnish except a small area along the luting zone. A dye penetration test (methylene blue dye 5%, centrifugation for 3 min at 30G) was used to evaluate linear dye penetration and dye penetration area. Statistics were computed using SPSS Win 12.0 (Kolmogorov–Smirnov test, ANOVA, t-test). **Results** Linear dye penetration (mm); dye penetration area (mm²) (Mean/SD): group 1 (1.7/0.7); (4.5/2.8), group 2 (8.9/2.0); (32.1/ 7.6), group 3 (1.4/0.7); (0.7/1.6), group 4 (5.1/2.9); (13.3/12.9), group 5 (3.2/2.9); (5.2/6.2), group 6 (6.4/2.2); (22.6/9.4). Significant differences were found between groups (ANOVA, P < 0.001). No significant differences were observed between Embrace, Multicore Flow and Paracem (t-test, P > 0.05). Maxcem revealed significantly higher dye penetration values than the other luting cements (*t*-test, P < 0.05).

Conclusions Within the limits of this study the adhesive insertion of NTI FiberMaster glass fibre posts using the multi-step systems Multicore Flow and Paracem and the self-adhesive Embrace had least dye penetration.

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