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Bioceramic materials in endodontics

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During the past two decades, a number of major advances have been made in the field of bioactive ceramics used for endodontic treatment. This article reviews the physico-chemical and biological properties of bioceramic materials and the application of bioceramic technology to endodontics. Bioceramic materials, with their biocompatible nature and excellent physico-chemical properties, are widely used in endodontic applications. They can function as cements, root repair materials, root canal sealers and filling materials, which have the advantages of enhanced biocompatibility, potential increased root strength following obturation, antibacterial properties and sealing ability. New bioceramic materials have demonstrated the ability to overcome some of the significant limitations of earlier generations of endodontic materials. Most bioceramic materials have been shown to be biocompatible and have good physico-chemical characteristics, therefore having a potential use in clinical endodontics. Although *in vitro* studies on the use of bioceramic materials in endodontics have given encouraging results, randomized and double-blind clinical studies of sufficient length with these materials are needed to confirm long-term success following their use.

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Introduction

Most endodontic failures occur as a result of leakage of bacterial invasion from treated and filled root canals into the periapical tissues (1). The leakage can occur between the root filling, sealer or repair materials and dentin after root canal treatment. Endodontic materials that are placed permanently in the root canal should block the pathways of communication between the root canal system and its surrounding tissues to ensure long-term endodontic success (2). Hence, the criteria for the ideal material for use in endodontics has become comprehensive and includes the following characteristics: non-toxic, insoluble in tissue fluids, dimensionally stable, antibacterial, hard tissue conductive, biocompatible, radiopaque and easy to handle (3,4). However, the existing root filling and restorative/reparative materials currently used in endodontics do not possess all these desired characteristics.

Bioceramic materials are biocompatible ceramics suitable for use in the human body (5). Bioceramic-based materials were introduced to endodontics in the 1990s, first as retrograde filling materials and then as root repair cements, root canal sealers, and coatings for gutta-percha cones (6,7). The potential advantages of bioceramic materials in endodontics are related to their physico-chemical and biological properties. Bioceramics are biocompatible, non-toxic, non-shrinking, and usually chemically stable within the biological environment (8). A further advantage of these materials is their ability to form hydroxyapatite and ultimately create a bond between dentin and the material (9).

Following the introduction of bioceramic materials into clinical endodontics, mineral trioxide aggregate (MTA) has become recognized as the gold-standard material for a variety of clinical situations and is perhaps closest to the ideal reparative material, due to its excellent physico-chemical and biological properties (8,10). Recently, a class of new bioceramic materials have been introduced, which possess many similar and some different *in vitro* and *in vivo* characteristics, in an effort to

develop the ideal materials for use in endodontic applications.

In this review, focus will be placed upon updating the previously published information by presenting a comprehensive list of literature from 1981 to 2015, summarizing the physico-chemical and biological properties of current bioceramic materials and their clinical applications, and finally estimating the future perspectives of the bioceramic materials in endodontics.

Characteristics and identification of an optimal material for endodontic use

Physico-chemical properties

Short setting time

Setting time is the length of time for a material to transition from a fluid state into a hardened state. The presence of moisture is usually required for bioceramic materials to set (11). A short setting time can help facilitate a tight seal between the root canal system and the periodontium, while a long setting time may result in difficulties with maintaining consistency of the mixture (12).

Several methods have been used to evaluate the setting of dental materials. One of them is based on the principle in ISO 9917, which evaluates the resistance of a needle penetrating the surface of the material as a reflection of the setting time (13). An alternative method is to use a rheometer to measure the constraint transmitted by the sample material when a sinusoidal strain is applied, and thereby measure the evolution of the elastic modulus of the material without any modification to the material structure (14). The final setting time is the time at which a material achieved less than 95% of its maximum displacement (10,15).

High mechanical strength

The compressive strength of a material is the value of uniaxial compressive stress reached when the material fails completely. It has been reported that high compressive strength of a root repair material could enable it to withstand loads tending to deformation and shrinkage (16). Samples for measuring compressive strength are made in cylinders and tested in compression at an appropriate loading rate using a testing jig attached to a console (17).

Flexural strength is another type of mechanical strength which is defined as a material's ability to resist deformation under a load (18). The transverse bending test is most frequently employed where a rod specimen, having either a circular or rectangular cross-section, is bent until fracture using a three point flexural test technique (19). The three point bending test is essential when a material is being used for Class I, II and IV cavities. The higher flexural strength the material has, the lower the risk of fracture in clinical use (18).

Push-out strength is an important attribute for materials used in perforation repair, because tooth movement in function may dislodge the material after perforation repair (20). The push-out strength value is measured with a universal testing machine, which evaluates samples placed on a metal slab with a central hole to allow for the free motion of the plunger (21). The compressive load is applied by exerting a download pressure on the surface of the test material with the probe moving at a constant speed (22). The push-out strength is calculated by dividing the force by the surface area of the test material.

Finally, hardness can be defined as the resistance to plastic deformation of the surface of a material after indentation or penetration. The reported microhardness values for natural dentin are in the range of 60-90 VHN (23). It would be optimal if the surface hardness of a bioceramic material could reach the same range as dentin.

High alkaline pH and calcium ion release

It has been suggested, by both *in vitro* and *in vivo* studies, that the mechanism of pulp wound healing by the deposition of mineralized apatite depends on pH and the ability of calcium ion-release (24,25). Moisture facilitates the hydration reactions of calcium silicates to produce calcium silicate hydrogel and calcium hydroxide, which partially react with the phosphate to form hydroxyapatite and water (26, 27).

Calcium hydroxide has been advocated as an intracanal medicament in the treatment of external inflammatory root resorption (28,29). The effect of calcium hydroxide in the root canal on external

resorption may be a consequence of its antibacterial activity against bacteria, which are necessary for continuing external inflammatory root resorption (30,31). Several studies have reported that a high alkaline pH may induce limited necrosis of resorptive cells on the root surface and abolish the biological activity of lipopolysaccharides (32). Therefore, an alkaline pH and calcium ion release are desired during the setting reaction of any material that is permanently sealed in the root canal.

High radiopacity, moderate flow, low porosity and solubility

Radiopacity is an essential physical property which allows the viewing of endodontic filling materials by radiographic examination, in order to check the obturation quality (16). An ideal root canal filling and sealing material should have a certain degree of radiopacity to be clearly visible on radiographs (33). The radiopacity measurement is performed according to ISO 6876/2001 recommendations. Aluminum is used as the control material to evaluate the radiopacity of a tested material. International standards require a minimal radiopacity equivalent to 3 mm thick (16).

Flow is the ability of a cement to penetrate into the irregularities and accessory canals of the root canal system and is considered to be an important physical property. The greater the flow, the better the ability to penetrate into irregularities (34). The flow ability is also influenced by the size of the particles—the smaller the particles, the greater the flow (35). However, if the flow is excessive, the risk of material extravasation to the periapical area is increased, which could damage periodontal tissues and compromise healing (34,35). Thus, a moderate flow is preferred for the cement to access the areas that need to be filled. The flow test is performed according to ISO 6876/2001 specifications, which state that the minimal flow required for cements is 20 mm (35).

The mechanical resistance of calcium silicate-based materials is partially dependent on their low level of porosity (36). The amount of porosity in mixed cement is related to the amount of water added to make a cement paste, entrapment of air bubbles during the mixing procedure, and the environmental acidic pH value (37,38). The lower the porosity, the higher the mechanical strength.

Solubility is another factor in assessing the suitability of materials used as restorative materials in dentistry. Lack of solubility is a desired characteristic for root-end filling materials and materials used for perforation repair (16).

Biological properties

Biocompatibility

Materials used in endodontics are frequently placed in intimate contact with the pulp or periodontium and thus must be non-toxic and biocompatible with host tissues (Fig. 1). There are different *in vitro* and *in vivo* tests to evaluate the biocompatibility of dental materials. The *in vitro* tests include evaluating the cytotoxicity profile of potential materials using different cell lines (DC-27, MDPC-23, Od-21 etc.) and the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay (3), flow cytometry using cell viability staining (39), and tests for the ability of cells to grow and populate on the surface of a material. The *in vivo* tests comprise of usage tests in experimental animals, according to accepted clinical protocols, followed by histological examination (40,41).

Stimulation of biomineralization

An optimal material used for endodontic purposes, such as pulp capping, perforation repair, or root-end filling, not only provides an effective seal, but also induces chemical bond formation and apatite precipitation in dentin over time (42). Biomineralization is

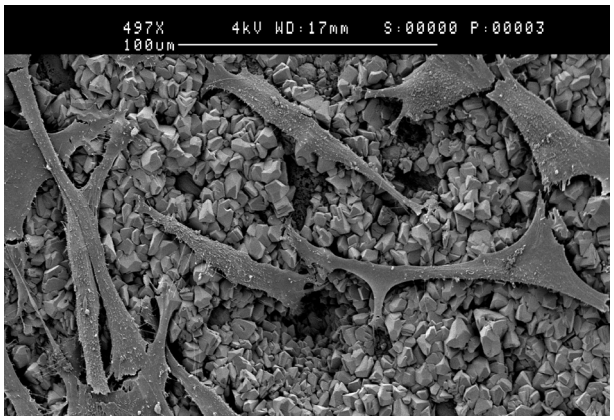

Fig. 1. SEM micrograph of human gingival fibroblasts on the surface of BC Sealer after culturing in DMEM for 24 hours.

likely to facilitate healing at the material-tissue interface, resulting in the elevation of local pH, the release of mineral ions and formation of apatite-like structures (9). The apatite crystals grow within collagen fibrils, promoting controlled mineral nucleation on dentin and triggering the formation of an interfacial layer at the material-dentin interface (43,44).

An ideal biomaterial used in endodontics should stimulate and modulate the biomineralization process to properly seal the margin of a tooth defect, so that the newly formed barrier of mineralized tissue can protect the root canal from bacteria and toxins.

The phenomenon of biomineralization can be identified by various microscopic techniques, including scanning electron microscopy (SEM) attached with energy dispersive X-ray analysis (EDX) (45), transmission electron microscopy (TEM), with selected area electron diffraction (SAED) (46); spectroscopic techniques including Fourier transform infrared spectroscopy (FTIR), Raman microscopy, X-ray diffraction (XRD) (47, 48) and histochemical methods including alizarin red staining (49).

Induction of pulp cell differentiation

Severe pulp exposure and the destruction of the underlying odontoblast layer may, under specific conditions, initiate regeneration of the pulp-dentin complex through progenitor cell recruitment and differentiation into secreting cells and the stimulation of reparative dentinogenesis (42,50). The main objective in using a bioactive material for this reparative and healing process is to form a barrier of mineralized tissue to protect the pulp from further leakage (51). Clinically, the objectives of treatments such as direct pulp capping and pulpotomy are to seal the pulp wound, induce odontoblast-like cell differentiation and stimulate dentin secretion in order to build a dentin bridge (52). Hence, in order to improve the clinical outcome, new biomaterials should be developed that can optimally and specifically stimulate odontoblast differentiation pathways. The differentiation of odontoblasts can be identified by the relative gene expression.

Antibacterial activity

Root canal treatment reduces but usually does not eliminate all microbes. The persistence of microorganisms in dentinal tubules, lateral canals and apical ramifications after root canal treatment have been reported (53,54). Therefore, it is considered beneficial if endodontic materials that are permanently sealed in the root canal have long-lasting antibacterial activity (55).

Agar diffusion test (ADT) assays have been used in earlier studies as a semi-quantitative technique to evaluate the antibacterial activity of various dental materials (56,57). However, it is problematic to compare bacterial inhibition data because of the lack of control of a large number of variables (58). The results of ADT are also highly influenced by the solubility and diffusion of the test agent through the agar, but these interactions are not well understood (58). Therefore, the value of ADT in predicting the antimicrobial effectiveness of various materials *in vivo* is poor, and ADT is no longer recommended for this purpose. The ADT has since been replaced by the direct contact test (DCT) which is based on measuring the effect of direct and close contact between microorganisms and the tested material on microbial outgrowth (59). DCT better reflects the true antimicrobial potential of the various sealers in standardized settings. Although many DCT studies have found several endodontic sealers to be effective against planktonic culturing bacteria, there is little evidence so far of their effectiveness against the microorganisms in biofilms and infected dentin. Recently, a new dentin infection model was introduced to establish a standardized and dense presence of bacteria deep in the dentinal tubules (Fig. 2) (60). This model has

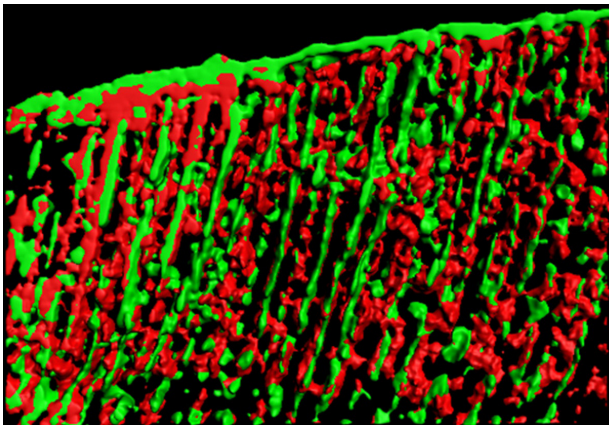

Fig. 2. Confocal laser scanning microscope image of bacteria in dentinal tubules. Specimen is stained with viability staining; red stain indicates severe cell wall damage, green stain intact microbial cells. In this dentin infection model bacteria are forced into dentin canals by centrifugation.

been used to measure the effectiveness of disinfecting solutions against *E. faecalis* biofilms in dentin, and has produced reproducible results in a standardized setting using viability staining and confocal laser scanning microscopy (CLSM) (61,62). So far, little evidence is available showing that bioceramic materials used for endodontic purposes can kill bacteria in infected dentin canals. However, the standardized dentin infection model has great potential to reveal the antibacterial activity of endodontic bioceramics against dentin infection.

Bioceramic cements

Biodentine

Physico-chemical properties

Biodentine (Septodont, Saint-Maur-des-Fosses, France) is a novel bioceramic cement (Fig. 3) claimed by the manufacturer to possess the benefits but not the drawbacks of other conventional cements (63). Biodentine contains tricalcium silicate, calcium carbonate, zirconium oxide, and a water-based liquid-containing calcium chloride as the setting accelerator. Zirconium oxide is the radiopaque agent allowing identification on radiographs. According to the ISO 6876/2001, Biodentine displays a radiopacity greater than 3 mm thickness of aluminum (64), which makes it suitable e.g. in the endodontic indications of perforation repair.

According to the manufacturer's instructions, Biodentine is a fast-setting (around 10-12 minutes) calcium silicate-based restorative material and is recommended for use as a dentin substitute which can be used both as a coronal restoration material, for perforation repair, and as a pulp-capping material in direct contact with the pulp (63). Grech et al. (64) recently compared the setting times of Biodentine, zirconium replaced tricalcium silicate cement (TCS-20-Z), and BioAggregate. BioAggregate is a new bioceramic material intended for perforation repair and as a retrograde filling material (65), which contains calcium phosphate and silicon dioxide (66).

Fig. 3. Biodentine is the only bioceramic cement where the powder and liquid are mixed using a mixing device. Final mixing and adjustment of suitable water content are done manually to obtain the desired consistency for each case (bottom right).

Biodentine had the shortest setting time among tricalcium silicate cements (ProRoot MTA, MTA Angelus etc.) and is used in endodontics because of its different setting kinetics (67), which is an improvement to conventional calcium silicate materials. Another advantage of such novel calcium silicate-based materials over MTA is that they exhibited clinically perceptible color changes, which is one of the reasons why calcium silicate materials are considered as alternatives for use in esthetically critical regions in the clinic (68).

The pH value of the Biodentine leachate has been reported to be 11.7 after 1 day immersion in Hank's balanced salt solution and demonstrates no significant change over the next 28-day period (69). Several studies have compared the calcium release of Biodentine with other bioceramic materials (70). Biodentine showed a higher level of calcium ion release than MTA, EndoSequence BC Sealer (BC Sealer), BioAggregate, TCS-Zr, and intermediate restorative material (IRM) (69,71). The incorporation of calcium and silicate was detected deep in Biodentine-treated root dentin after immersion in phosphate-buffered saline (PBS) for 90 days (71).

As a dentin substitute, Biodentine exhibited high mechanical properties in terms of compressive strength, push-out strength and microhardness (22, 64). A recent experiment reported significantly higher compressive strength for Biodentine over Bio-Aggregate, TCS-20-Z and IRM under a loading rate of 50 N/min. Moreover, Biodentine had a three times higher Vickers microhardness value than the other three materials (64), even though a recent study reported the value to be slightly reduced after acid etching (72). Another recent investigation evaluated the push-out strength of Biodentine in comparison with contemporary root perforation repair materials (22). Almost all Biodentine samples revealed a cohesive bond failure, whereas MTA showed adhesive bond failure. The different failure types of Biodentine and MTA may be explained by the particle size of these materials, which affects the penetration of the cement into the dentinal tubules in tag-like structures leading to a micromechanical anchor (73). The smaller particle size and uniform components of Biodentine might have a role in its ability to interlock better with the dentin (6). As a result, Biodentine showed significantly higher push-out bond strength than MTA. Furthermore, the push-out bond strength of amalgam, IRM, and Biodentine was not significantly different when immersed in sodium hypochlorite (NaOCl), chlorhexidine (CHX), and saline solutions, whereas MTA lost strength when exposed to CHX (22). However, a latest study reported that the push-out bond strength values can be significantly reduced if the smear layer is removed, which is detrimental to the interlock between Biodentine and dentin (74).

Although Biodentine can facilitate the remineralization of dentin, prolonged contact of dentin with Biodentine has been reported to have an adverse effect on the integrity of the dentin collagen matrix (46). Another study reported that Biodentine altered material toughness more than the strength and stiffness of dentin collagen after aging in 100% relative humidity (75). However, the authors suggested that the amount of collagen extracted was limited to the contact surface and therefore should not affect the use of Biodentine in endodontic procedures (46).

Biological properties

The biocompatibility of Biodentine has been evaluated using different types of cells. One flow cytometry study showed that cell viabilities were highest with extracts from Biodentine and MTA at all extract concentrations, without a significant difference between each other, whereas cells exposed to extracts from glass ionomer cement displayed the lowest viabilities (39). Other studies using different pulp cell lines reported different effects of Biodentine on the cell proliferation (76,77). Pseudo-odontoblastic cells (MDPC-23) cultured in the presence of Biodentine have lower levels of viability than those cultured in the presence of MTA on day seven. For another type of pulp cell (OD-21), proliferation rates on day 7 were significantly lower in the presence of both Biodentine and MTA compared to the control. One recent study reported that the stimulation on odontoblastic differentiation by Biodentine is due to the activation of MAPK and CaMKII pathways (78,79).

Moreover, Biodentine has been reported to induce osteoblast differentiation in different stem cells. Messenger RNA level of osteogenic genes, including ALP, osteocalcin and bone sialoprotein, was increased after exposure to Biodentine for 3 days (80). Zanini et al. (42) confirmed, with the exposure

to Biodentine, the differentiation of pulp cells into odontoblasts by examining the expression of Msx2, Runx2, Osx, and Dlx5 genes as biomolecular markers. Alizarin red staining results showed the nodules of biomineralization induced by Biodentine containing medium for 11 days.

The rats pulp injury model was used to evaluate the capacity of different calcium silicate-based restorative cements to induce pulp healing (81,82). Their results revealed the formation of a homogeneous and continuous dentin bridge in Biodentine and MTA groups at the injury site on day 14. In the case of direct pulp capping and pulpotomy in pigs (83, 84), histological sections of the teeth were done after 1, 4, and 12 weeks of treatment by Biodentine, MTA, and formocresol. Results showed that Biodentine and MTA promoted beneficial calcification after one week, whereas formocresol induced necrosis and inflammation.

Above all, it has been recently stated that Biodentine could be considered as an alternative to MTA due to comparable bioactivity by producing an interfacial layer on the root canal dentin (85–88).

Clinical studies

Koubi et al. (4) conducted a clinical study on a total of 397 cases and found that Biodentine restorations in posterior teeth were clinically sound and symptom free for up to 6 months. When Biodentine was retained as a dentin substitute after pulp vitality control, it was covered systematically with the composite Z100. This procedure yielded restorations that were clinically sound and symptom free, and the resistance to marginal discoloration was superior with Biodentine compared to composite Z100.

Handling of Biodentine is somewhat different than that of MTA. When mixed wet, Biodentine sticks well to instruments and dentin, resembling glass ionomer. A drier mix of Biodentine does not stick right away to dentin and can therefore be pushed down to the apical canal with paper points, using a similar technique as MTA. A recent investigation compared the response of the pulp-dentin complex in human teeth after direct capping with Biodentine and MTA (89). Results showed complete dentinal bridge formation and the absence of an inflammatory pulp response for both materials. Layers of well-arranged odontoblast and odontoblast-like cells were found to form tubular dentin under the osteodentin. Because Biodentine had a similar efficacy in the clinical setting, it may be considered an alternative to MTA in pulp-capping treatment during vital pulp therapy.

MTA

Physico-chemical properties

The original formulation of MTA was developed in the 1990s, and is manufactured by Dentsply International (Dentsply-Tulsa Dental, Johnson City, USA). MTA materials are a mixture of dicalcium silicate, tricalcium silicate, tricalcium aluminate, gypsum, tetracalcium aluminoferrite, and bismuth oxide. MTA is currently marketed in two forms: gray (GMTA) and white (WMTA) (Fig. 4). WMTA was developed a few years later than the original GMTA because of the potential of dentin discoloration by GMTA (90). Investigations have shown lower amounts of iron, aluminum and magnesium in WMTA compared to GMTA (91–93). The component bismuth oxide has also been used to produce the radiopacifier and enhance physical properties in other calcium silicate cement (94).

Since its introduction into the endodontic market, MTA has gained widespread use as a retrograde filling material in pulp capping, pulpotomy, apical barrier formation, apexification and perforation repair (95). MTA is prepared by mixing the powder with sterile water in a 3:1 powder-to-liquid ratio (96). The mean setting time of MTA has been reported to be approximately 165 minutes, which is longer than amalgam, Super EBA and IRM (16,97). GMTA has significantly higher initial and final setting times than WMTA (98).

The pH value of MTA is 10.2 after mixing and rises up to 12.5 after 3 hours (16). WMTA displays a significantly higher pH value 60 minutes after mixing compared to GMTA (99). The high pH value is attributed to the constant release of calcium ions from MTA and the formation of calcium hydroxide (71,100,101).

The results of the degree of solubility of MTA have been contradictory between different studies (16,98,102,103). Most investigations reported low or no solubility for MTA (102–104). However,

Fig. 4. MTA is often regarded as the golden standard of bioceramic cements against which other products are compared. Both gray and white MTA are powders which are manually mixed with water.

increased solubility has been reported in a long-term study (100).

The porosity of MTA cement is related to the entrapment of air bubbles during the mixing procedure and the environmental acidic pH. A trend has been observed that the more acidic the environment, the more extensive the porosity of the specimens (105). Present data also shows that low humidity, low pH values, the presence of a chelating agent and high condensation forces may adversely affect MTA microhardness (106–109).

The mean radiopacity for MTA has been reported at 7.17 mm of an equivalent thickness of aluminum (16). WMTA was more radiopaque than GMTA in two studies (98,99). Because a similar amount of bismuth oxide is used to produce radiopacity for both materials, the presence of other substances in WMTA may explain the difference between the two (10).

Several factors may influence the compressive strength of MTA, including the type of MTA (110), the liquid that is mixed with the material (111), condensation pressure on the material (108), the pH value of the liquid used for mixing (107), and the conditions of MTA storage (112). A recent study revealed that the compressive strength values of MTA were significantly greater than those of MTA Angelus (Angelus, Londrina, PR, Brazil) (110). Another investigation reported significantly lower compressive strength for WMTA when the material was etched by phosphoric acid, compared to the unetched control group (107). Moreover, the compressive strength of MTA was reported to be significantly less than that of amalgam, IRM and Super EBA after 24 hours. However, after 3 weeks, no significant difference was observed between these materials (16).

It has been suggested that placing a moist cotton pellet over MTA for the first 24 hours can increase its flexural strength (113). Recent studies have focused on the effects of bioceramic materials on the flexural strength of dentin, showing that when exposed to MTA the flexural strength decreased significantly after 3 months (75,114).

The force needed for displacement of MTA, in neutral pH, was reported to be stable at around 7.28-7.68 MPa (115,116). However, the push-out bond strength of MTA could be influenced by different acidic and alkaline pH values (116). The force needed for the displacement of MTA was significantly lower in samples stored at lower pH values (115), while the highest push-out strength was achieved at pH 8.4 (116). MTA stored in butyric acid at pH 7.4 showed needle-like crystals formed in the inter-grain spaces between small cubic particles (Fig. 5). The needle-like structures appeared to form in bundles as part of the framework of the hydrated MTA (Fig. 5). Most of the jagged clusters and interlinking needle-like ettringite crystals disappeared when the MTA specimens were exposed to an acidic environment of pH 5.4 (Fig. 5). Reyes-Carmona et al. (117) indicated that the biomineralization process

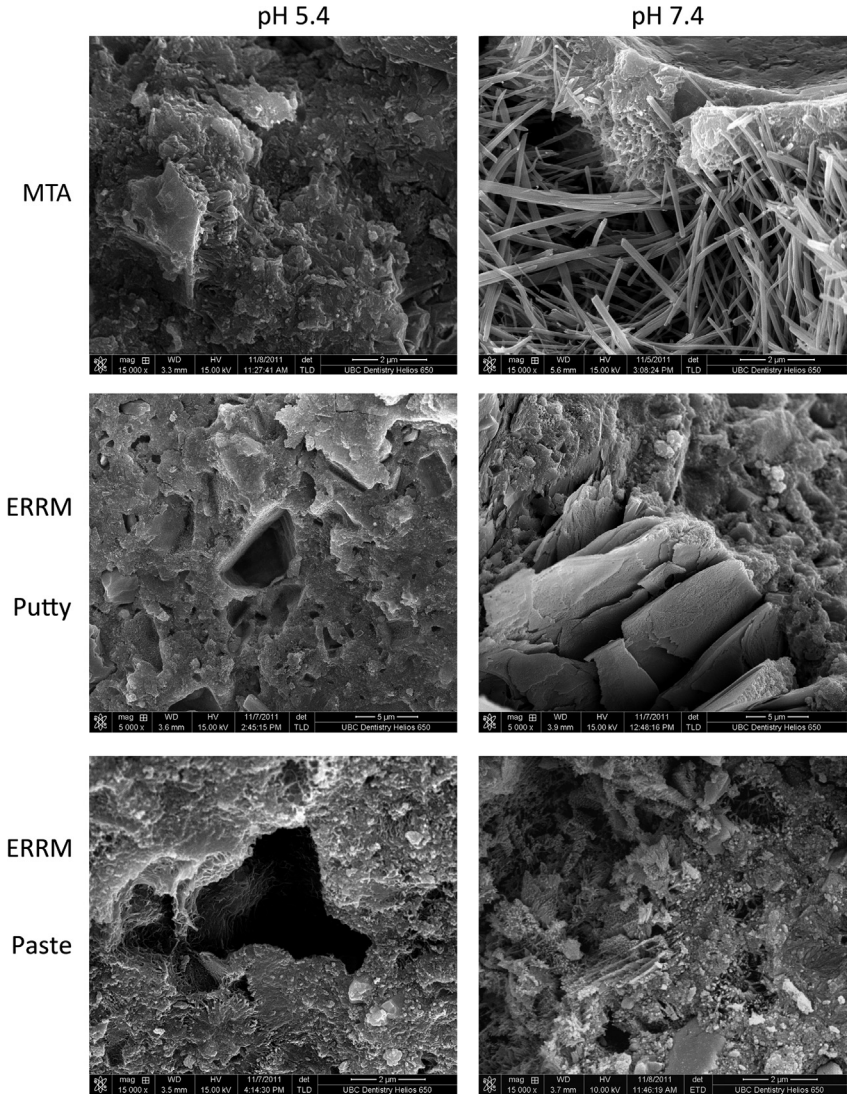

Fig. 5. SEM images of cross-sections of MTA, ERRM Putty and ERRM Paste exposed to butyric acid at pH 5.4 and 7.4 after 7 days of setting.

positively influenced the push-out bond strength of the MTA and several MTA-based materials. The biomineralization ability of MTA, most likely through the formation of tags (Fig. 6), may be the reason of the dislodgement resistance.

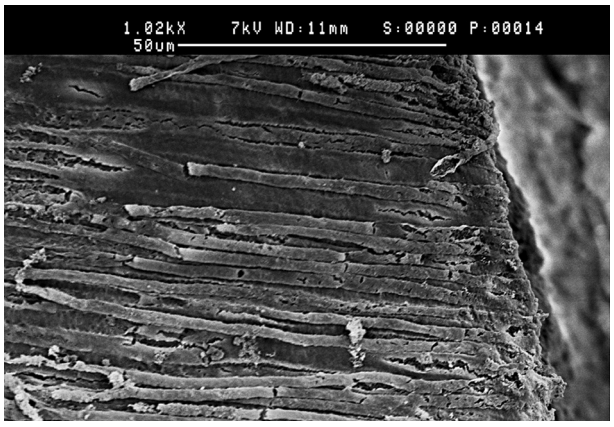

Fig. 6. SEM micrographs of biomineralization in dentinal tubules induced by MTA.

Biological properties

A number of biocompatibility studies have shown that MTA is a biocompatible material. Cytotoxicity and cell attachment studies with various cell cultures showed better results with MTA than with amalgam (118–121), Super EBA (118,122,123), IRM (119,124), various types of glass ionomers

(118,123,125,126), and gutta-percha (123). MTA shows similar biocompatibility as the new generation bioceramic endodontic materials, including Biodentine (39), EndoSequence root repair material (3,127,128) and BC Sealer (or iRoot SP) (129). However, there is some disagreement regarding the relative cytotoxicity of WMTA and GMTA among studies (130–132).

Biomineralization and stimulation of cell differentiation are another two biological characteristics of MTA. It has been reported that MTA could induce osteogenic phenotype which reflects the up-regulation of alkaline phosphatase, osteonidogen, osteonectin, and osteopontin expression (133). Moghaddame-Jafari et al. (134) showed the proliferation of murine odontoblast-like and undifferentiated pulp cells induced by WMTA. Moreover, cells cultured with WMTA revealed heat-shock protein 25 as a marker for odontoblast differentiation during pulp healing (135). More recent investigations reported that MTA, with or without enamel matrix derivative, improves human dental pulp cell differentiation and mineralization (136,137).

The antibacterial properties of MTA have been extensively evaluated with somewhat conflicting reports (36,65,138–141). Several studies reported that MTA has a limited antimicrobial effect against some microorganisms (140–142). The antibacterial effect of MTA was detected on some facultative bacteria, but there was no effect on strictly anaerobic species (141). Recently, Zhang et al. (65) reported that dentin enhances MTA's antibacterial activity, and Heyder et al. (143) indicated that the antibacterial behavior of MTA could only be detected when freshly mixed. Conflicting results on the antibacterial activities of MTA may be attributed to the species of the microorganisms and the amount and type of MTA used (10).

Clinical studies

Numerous clinical studies have evaluated MTA as the material for root-end filling (144), pulp capping (89,145–147), pulpotomy for primary teeth (148–150), apical barrier formation for teeth with necrotic pulps and open apexes (151), perforation repair (152,153), and apexification (154,155). Meta-analysis studies have concluded that MTA has a high clinical success rate, provides an adequate seal, shows excellent biocompatibility and promotes tissue regeneration (156,157).

Despite its many advantages MTA also has some less than optimal properties, such as difficult handling, long setting time and potential discoloration of teeth with GMTA (158–160). Efforts have been made to overcome these shortcomings; however, adding or removing various elements to alleviate these shortcomings may affect MTA's otherwise excellent characteristics (161). Introducing new formulations should therefore await comprehensive *in vitro* and *in vivo* evaluations.

MTA Angelus

Physico-chemical properties

MTA Angelus from Brazil (Angelus, Londrina, PR, Brazil) is a MTA-based cement. MTA Angelus is composed of 80% Portland cement and 20% bismuth oxide, with no addition of calcium sulfate in an attempt to reduce setting time (162). The setting time of MTA Angelus is approximately 14 minutes (12), which is considerably less than WMTA and GMTA (16,99).

XRD analysis from a previous study compared the powder composition form of GMTA and MTA Angelus, and found GMTA contains a greater amount of bismuth oxide and magnesium phosphate (163). In contrast, the amount of calcium carbonate, calcium silicate and barium zinc phosphate in MTA Angelus was greater than in GMTA. Another study using EDX showed that the amount of calcium in MTA Angelus is higher than in GMTA, whereas the amounts of carbon, oxygen, bismuth and silica are higher in GMTA (10,164). In addition, a study on the particle size of MTA-based cements concluded that MTA Angelus had a higher variation in particle size than MTA (165).

The results on the pH and calcium ion release of MTA Angelus are conflicting (166). Duarte et al. (162) showed that MTA Angelus produced a higher pH value and calcium ion release than GMTA within 168 hours after mixing. However, Reyes-Carmona et al. (9) reported that pH and calcium release was lower in MTA Angelus than in MTA. Parirokh & Torabinejad (10) concluded that the pH and calcium ion release between MTA and MTA Angelus was not significantly different. The differences between

these studies may be attributed to the length of time to measurement after mixing the material (99,167,168) and the effect induced by changing the solution for material incubation (98,162). In addition, the microhardness of MTA Angelus was reported to be increasing with incubation time and influenced by the technique of mixing (169).

Several dye leakage studies have compared the quality of the seal by MTA Angelus, zinc-free amalgam, Vitremer (a resin-modified glass ionomer cement), and Super EBA, with conflicting reports. Pereira et al. (170) reported that MTA Angelus gave the best seal against root dentin among all of the tested materials. In contrast, another study found more leakage with MTA Angelus and Vitremer compared to Super EBA in apical sections (171). However, no significant difference could be found between MTA Angelus and Super EBA in other tooth sections. Controversy also exists between MTA Angelus and MTA. One study showed no significant difference in dye penetration between them, whereas GMTA showed less dye leakage when used as a perforation repair material in another investigation (170,171). When an internal matrix was used for MTA Angelus, it demonstrated a better seal. The possible reason for the conflicting results can be the differences in experimental design between studies.

MTA Angelus has also shown to have a lower radiopacity than WMTA and GMTA (10,33,99).

Biological properties

Ribeiro et al. (172–174) published a series of biocompatibility studies showing gray and white MTA Angelus are not cytotoxic or genotoxic in concentrations of 1 to 1000 μg/mL on Chinese hamster ovary cells and lymphoma cells. Another three studies on materials used in pulp capping and pulpotomy reported both MTA and MTA Angelus were less cytotoxic on fibroblast and pulp cell lines than formocresol, calcium hydroxide, Vitrebond, Super EBA and ferric sulfate (175–177). A flow cytometry investigation compared the cytotoxicity and genotoxicity of MTA Angelus and a new bioceramic material containing castor bean oil. None of the three materials showed cytotoxic effects on transfected human pulp cells (178).

Reduction in cell viability has also been reported by several studies when using MTA Angelus. De Deus et al. (179) demonstrated that MTA Angelus significantly reduced endothelial cell viability in the first 24 hours. However, no significant difference in cell viability was found between control and experimental groups at 48 and 72 hours. Two other studies reported that MTA Angelus and MTA have cytotoxic effects on V79 fibroblasts (123) and human gingival fibroblasts (180).

Histological sections have been used to evaluate tissue reactions in several pulp capping studies. Accorinte et al. (147) performed pulp capping in 40 permanent premolars with MTA Angelus or calcium hydroxide and extracted the teeth after 30 or 60 days. The 60-day follow-up histologic results showed a similar histological response, with the formation of a hard tissue bridge in almost all cases with low inflammatory infiltrate.

Rat alveolar bone and the subcutaneous response occurring after implantation of experimental MTA Angelus, a Portland cement modified sealer and a light-cured MTA has been evaluated in *in vivo* animal studies (181,182). The results showed that light-cured and non-light-cured MTA Angelus produced a similar inflammatory response and bone healing. However, dystrophic mineralization was observed only with MTA Angelus, which stimulated mineralization in subcutaneous connective tissue of the rats (182).

Compared with MTA, the number of studies on the antibacterial ability of MTA Angelus is low. Sipert et al. (183) compared the antimicrobial properties of MTA Angelus, Sealapex, Fill Canal, Portland cement, and EndoRez on various species of microorganisms including *E. faecalis, Escherichia coli*. The results indicated that both MTA Angelus and Portland cement were effective against all the microorganisms except for *E. coli*. Another study comparing GMTA, MTA Angelus, and Portland cements showed that all these materials had antimicrobial activity against the microorganisms tested, including *E. coli* (138). Other studies have reported similar antibacterial activity against *Streptococcus* species, *Micrococcus luteus, Staphylococcus aureus, Pseudomonas aeruginosa, Enterococcus faecalis* and *Candida albicans* by MTA Angelus and MTA (184,185).

Clinical studies

Jacobovitz & de Lima (186) reported a clinical case with inflammatory internal resorption treated by

MTA Angelus; the 20-month follow-up radiographs demonstrated the maintenance of a functional tooth. Marginal gingiva discoloration has been found after using gray MTA Angelus for a root perforation case (159). Replacing gray MTA Angelus with white MTA Angelus produced satisfactory esthetic results.

A randomized controlled trial study compared the clinical and radiographic performance of MTA Angelus and Portland cement as pulp dressing agents in 30 carious primary teeth (187). None of the treated teeth showed radiographic or clinical signs of pathology, or any other indications of failure, up to 24 months after treatment.

ERRM Putty, ERRM Paste, and iRoot FS

Physico-chemical properties

EndoSequence root repair material (ERRM) (Brasseler USA, Savannah, GA) is a new bioceramic material delivered as a premixed moldable putty (Fig. 7) (also labeled as iRoot BP Plus) or as a preloaded paste in a syringe with delivery tips for intracanal delivery (188, 189). Both materials are composed mainly of calcium silicates, zirconium oxide, tantalum oxide and calcium phosphate monobasic. Ma et al. (3) used EDX to confirm the chemical composition of the two materials; the results revealed that the ERRM putty and paste crystals have similar elements (calcium, silicate, phosphate, carbon and oxygen) but different overall composition to GMTA (tricalcium silicate, dicalcium silicate, bismuth oxide, and small proportions of tricalcium aluminate and calcium sulfate). Moreover, ERRM putty and paste each have a similar crystallographic surface structure to GMTA, showing hexagonal-shaped crystals varying in size, appearing both discretely and in aggregates (3). Another recent study analyzed the structure and composition variations of ERRM over a longer period of time (190). Results showed the precipitation of apatite and an increase of calcium and phosphate on the surface of the material after a 2-month immersion in PBS. ERRM has similar compressive strength to MTA (191). Although there are no studies so far showing the bonding strength of ERRM. The adhesion of ERRM to dentin and its formation of tag-like structures inside the dentinal tubules could possibly act as a micromechanical anchor to dentin (Fig. 8).

According to the manufacturer ERRM has a working time of more than 30 minutes and approximately a 4-hour setting time. Presence of moisture is required for the material to harden. The pH value of ERRM has been reported be as high as 12.4, which is probably responsible for its antibacterial properties during the setting reaction. Hansen et al. (192) compared the pH changes in simulated root resorption defects filled with MTA and ERRM, and concluded that intracanal placement of MTA resulted in a higher pH than with ERRM. The pH value of both ERRM and MTA treated canals declined to the levels of the negative control after a 4-week incubation in saline. A recent study showed that the microhardness values of ERRM Putty and ERRM Paste can be reduced in an acidic environment, and resulted in these materials having more porous and less crystalline microstructures (193). ERRM Putty

Fig. 7. ERRM bioceramic cement is a ready to use material with "putty" consistency; it sets slowly when in moist environment, e.g. inside a pulp chamber or in a root canal.

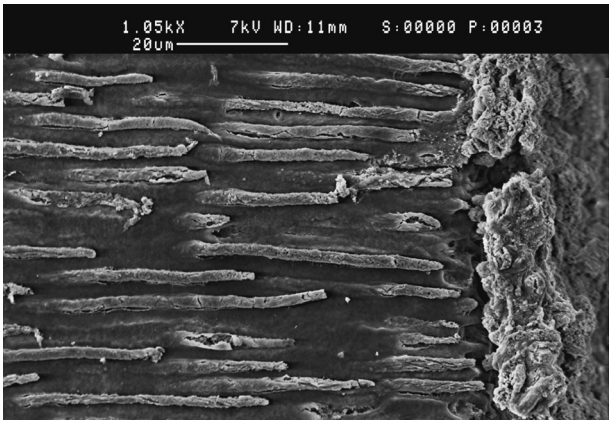

Fig. 8. SEM micrographs of biomineralization in dentinal tubules induced by ERRM.

samples at pH 7.4 showed a flake-like structure (Fig. 5). The edge of the flake-like structure was well-defined at pH 7.4. Empty pores were observed at pH 5.4 in ERRM Putty samples (Fig. 5). Similarly, more porous structures were found at pH 5.4 and more crystallized structures were observed at pH 7.4 for ERRM Paste (Fig. 5).

iRoot FS is one of the iRoot series bioceramic materials developed for permanent root canal repair (Fig. 9). iRoot FS has improved handling properties and shorter setting times (Brasseler USA, Savannah, GA). iRoot FS completely solidifies within 1 hour at 37°C in 100% humidity with an extra load of 500 g (194).

Biological properties

The cytotoxicity of ERRM has been evaluated in several recent studies. Using the MTT assay and different cell lines, the majority of studies concluded that ERRM has minimal *in vitro* cytotoxicity, similar to MTA and MTA Angelus (3,127,128,195). Gingival fibroblasts spread and attach on the surface of an ERRM disk forming a matrix-like overlay observed with SEM (3). Similar cytokine expression of IL-1β, IL-6, IL-8, and minimal TNF-α was detected from human osteoblasts seeded on ERRM and MTA (128). Only one study reported lower cell viability and alkaline phosphatase activity of human Saos-2 osteoblast-like cells when treated with extracts from ERRM compared to MTA (196). The differences between the results of this study and others may be related to the different cell lines and extract dilutions used. iRoot BP Plus was also found to be non-toxic to human dental pulp cells and was able to induce mineralization and odontoblastic differentiation-associated gene expression (197,198). *In vitro* and *in vivo* studies showed no significant difference in pulp response to iRoot BP Plus or MTA (189), consequently iRoot BP Plus might be used as a pulp capping material for vital pulp therapy (199). iRoot FS showed similar biocompatibility with iRoot BP Plus and ProRoot MTA, but it displayed the best cell adhesion capacity in both L929 and MG63 cells among iRoot BP Plus, ProRoot MTA and Super-EBA (194). Recently, an *in vivo* study was performed comparing the healing effect following root-end surgery with MTA and ERRM Putty, as root-end filling material, in an animal model using CBCT and micro-CT (200). Results showed that ERRM Putty achieved a better tissue healing response adjacent to the resected root-end surface histologically and a superior healing tendency compared to MTA, when detected by CBCT and micro-CT (200). The superior performance by ERRM Putty may be due to its better mineralized tissue inductive/conductive properties, thus accelerating the deposition of cementum-like tissue on the root-end surface, accompanied by periodontal ligament-like tissue and bone. However, no superior healing effect of ERRM Putty over MTA could be detected when using periapical radiographs (200).

In a study of the antibacterial effect of the cements, ERRM and MTA had a similar effect on

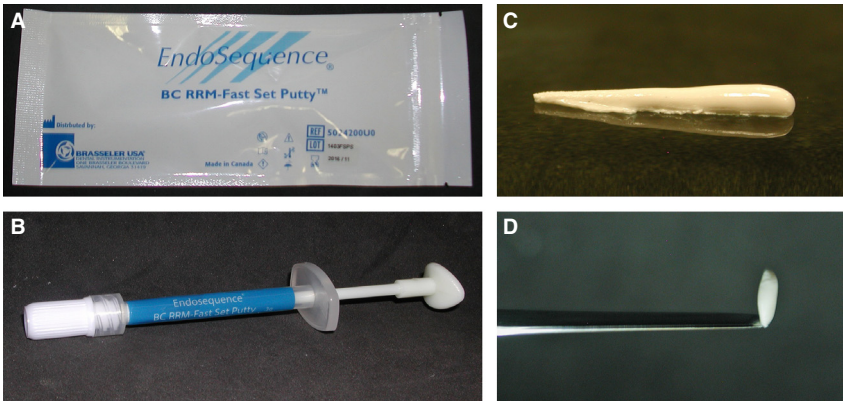

Fig. 9. iRoot FS (Fast Set Putty). (A) Package of iRoot FS. (B) A ready-to-use syringe of the Fast Set Putty. (C) A small portion of Fast Set putty material shaped on a glass plate. (D) iRoot FS on an instrument tip, ready to be placed in a retrograde cavity.

planktonic *E. faecalis* cells (188). Another recent study reported ERRM and MTA have comparable antifungal biofilm activity (201).

Bioceramic sealers

BC Sealer and iRoot SP

Physico-chemical properties

EndoSequence BC Sealer (Brasseler USA, Savannah, GA; also known as iRoot SP root canal sealer, Innovative BioCeramix Inc., Vancouver, Canada) is a premixed bioceramic endodontic sealer that contains zirconium oxide, tricalcium silicate, dicalcium silicate, colloidal silica, calcium silicates, calcium phosphate monobasic, and calcium hydroxide (202). Zirconium oxide is added as the radiopacifier (203). The radiopacity value of BC Sealer is 3.83 mm Al which is greater than the minimal requirement (3.00 mm Al), but significantly lower than AH Plus (6.936 mm Al) (34). The inorganic and radiopacifier components of the sealer are premixed with water-free liquid-thickening carriers and water is required for the sealer to reach its final set. Both the absorbed water, derived from the external environment, and the water produced by the reaction between calcium phosphates and calcium hydroxide participates are used to generate a calcium silicate hydrate phase (203). Hydroxyapatite is precipitated simultaneously within the calcium silicate hydrate phase and reinforces a bond between the dentinal wall and the sealer (26). Several retreatment studies indicated that the complete removal of BC Sealer or iRoot SP from root canals is difficult (202,204,205).

Moisture in dentinal tubules helps BC Sealer to initiate and complete its setting reaction (206–208). According to the manufacturer, the working time can be more than 4 hours at room temperature, and the setting time may range from 4 hours to more than 10 hours in very dry canals. However, in the *in vitro* condition, more time may be required to achieve a final set. Loushine et al. (203) found it required 72 hours to achieve the initial set and 240 hours to reach the final set of a BC Sealer disk in 100% relative humidity condition using the Gillmore needle method. When the set sealer was exposed to water, the microhardness of BC Sealer declined significantly (203). Moreover, a concordance was observed between pH and the amount of calcium ion released in BC Sealer. BC Sealer demonstrated higher pH, calcium ion release and flow values than AH Plus (203), but less calcium ion release than Biodentine (71).

Variability exists in the bonding behavior of BC Sealer in different studies. A recent study showed that iRoot SP presented the highest bond strength when compared with AH Plus, Epiphany, and MTA Fillapex (206,209). However, another three separate investigations showed iRoot SP has a similar bond strength to AH Plus (20,210,211), and a higher bond strength than MTA Fillapex (20). Only one study reported iRoot SP having a significantly lower bond strength than AH Plus (212). Discrepancies among studies may be explained on the basis of differences in experimental design, e.g. obturation technique, sealer brands, and compositions used. Regardless of the sealer type, the push-out bond strength of the sealer alone is significantly higher than with a matching single-cone technique (213). This may probably explain the relatively higher push-out bond values of calcium silicate-based sealers when used alone (210,214). When using the cold lateral condensation root-filling method, iRoot SP showed a similar push-out bond strength to AH Plus (20). The forces applied during filling probably provided a closer adaptation of the core material to the dentin walls and consequently, more frictional resistance (212). Besides the push-out bond strength, promising fracture resistance of roots obturated with iRoot SP and ActiV GP have been reported (215).

It is still unclear whether the solubility of sealers can influence the sealing properties of BC Sealer. A study has shown that iRoot SP recorded higher solubility than AH Plus, MTA Fillapex, and MTA Angelus (216), whereas a more recent investigation reported no significant difference in the solubility values between iRoot SP and AH Plus (217). Despite the discrepancies in solubility results between these studies, they consistently showed similar apical leakage with iRoot SP and AH Plus by fluid filtration test (27,217).

Biological properties

The cytotoxicity of BC Sealer (or iRoot SP) was found to be time and concentration dependent. Fresh iRoot SP was moderately cytotoxic and

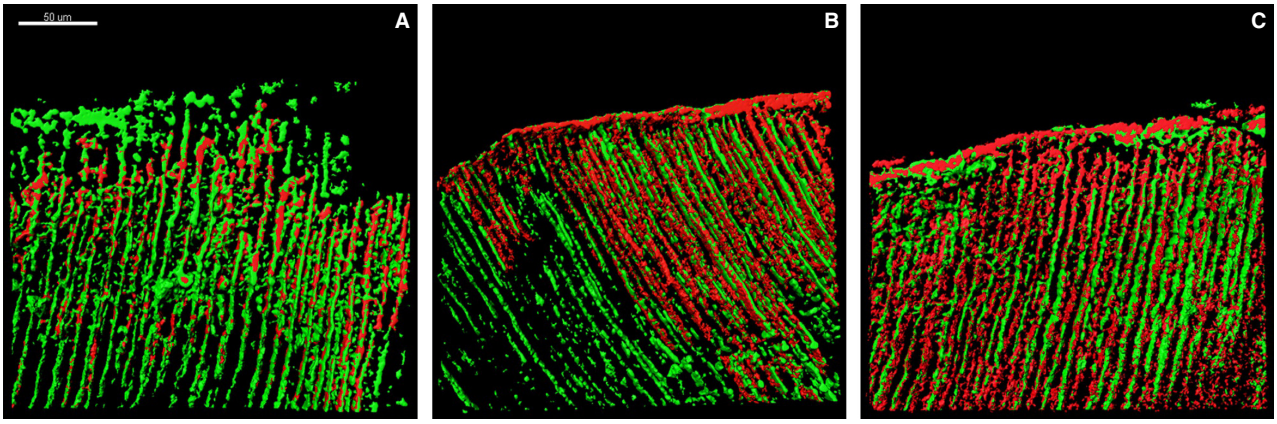

Fig. 10. Confocal laser scanning microscope images of 3-week-old *E. faecalis* infected dentin after exposure to BC Sealer and viability staining. (A) 1-day sealer exposure, (B) 7-day exposure, (C) 30-day exposure.

became slightly toxic after setting, and non-cytotoxic after long-term setting (7 days) or when highly diluted (1/50, 1/100) (129,218,219). The formation of calcium hydroxide during the setting reaction probably accounts for the cell death upon exposure to fresh sealer. However, human gingival fibroblasts can attach to and spread over the surface of a BC Sealer disk (set for 72 hours) after a 24-hour culture (Fig. 1). The dilution of the pure sealer must be in consistency with the clinical condition, where the material becomes diluted with the surrounding body fluid (219). Recent studies have compared the cytotoxicity of BC Sealer (or iRoot SP) with AH Plus and another bioceramic sealer, BioAggregate. BioAggregate was more toxic than iRoot SP, possibly due to the presence of silicon oxide and tantalum oxide and its high solubility (219), while one recent study showed the osteogenesis effect of BioAggregate on mesenchymal stem cells (80). A recent flow cytometry study showed significantly higher viability of human gingival fibroblast in the BC Sealer extract group compared to other sealer groups (220). Epoxy resin component, released during the setting of AH Plus, caused severe cytotoxicity (129,221). The opposite result was reported by Loushine et al. (203), who found the cytotoxicity of BC Sealer to be higher than AH Plus, with no reduction over 6 weeks. The difference may be related to the different types of cell line and the method of exposing the cells to the extract.

Guven et al. (222) revealed that iRoot SP and MTA could induce human tooth germ stem cells to differentiate into odontoblast-like cells, and induce biomineralization as evidenced by mineralization-related gene and protein expression (223). iRoot SP showed less inductive potential compared to MTA, but higher hard tissue deposition capability than AH Plus (223).

iRoot SP possessed low contact angle to sterile water, and displayed potent antibacterial effect against *E. faecalis* by using modified DCT (224). The antibacterial effect of iRoot SP might be a combination of high pH, hydrophilicity, and active calcium hydroxide release. The antibacterial effect continued for 3 days; after 7 days no antibacterial activity could be detected (224). In the meantime, a novel dentin infection model has been introduced to establish a standardized and deep penetration of bacteria into dentinal tubules (60). The effectiveness of endodontic sealers against bacteria inside dentinal tubules were investigated (225). Results showed that BC Sealer had antibacterial effects against *E. faecalis* in the dentinal tubules by killing 18%, 35% and 45% bacteria in 1, 7 and 30 days respectively (Fig. 10). BC Sealer and AH Plus had superior antibacterial effects compared to zinc oxide–eugenol sealer (225).

MTA Fillapex

Physico-chemical properties

MTA Fillapex (Angelus Solucoes Odontologicas, Londrina, PR, Brazil) is a recently introduced calcium silicate-based bioceramic sealer (21). MTA Fillapex was created in an attempt to combine the physico-chemical properties of a resin-based root

canal sealer with the biological properties of MTA (35). The composition of MTA Fillapex after mixture is mineral trioxide aggregate, salicylate resin, natural resin, bismuth, and silica (Fig. 11). According to the manufacturer, MTA Fillapex has an adequate working time, high radiopacity, and is easy to handle.

Similar to iRoot SP, MTA Fillapex has a high solubility, pH and calcium ion release (216,226). One study compared the pH and solubility of MTA Fillapex with several other endodontic sealers, 2 days after manipulation, and demonstrated a pH increased to 10.06 after 15 hours of immersion in deionized water; the solubility was the highest among all tested sealers (226). This high pH and solubility may benefit the antibacterial activity of MTA Fillapex.

MTA Fillapex has lower radiopacity (227), but significantly higher flow values than AH Plus (35), as measured according to the ISO 6786/2001 recommendations. The difference in the compositions of the two sealers is considered to be the main factor for their different flow values.

EDX analysis revealed high levels of calcium and carbon at the surface of MTA Fillapex (216). After 7 days there was a reduction in the amount of carbon, probably caused by the degradation of the polymer (228). As a consequence, this degradation may facilitate water sorption and calcium ion release. The occurrence of zirconium in MTA Fillapex was also demonstrated by EDX, suggesting that radiopacifiers used in AH Plus and iRoot SP sealers, such as calcium tungstate and zirconium oxide, may be present

Fig. 11. MTA Fillapex is a "hybrid" bioceramic sealer, which also has a resin component. Delivery syringe facilitates correct mixing.

(216). However, EDX analysis was unable to detect the bismuth trioxide probably because of its low amount.

The chemical composition of this MTA-based sealer may also influence its bonding behavior. The bond strength of MTA Fillapex has been widely reported to be significantly lower than other endodontic sealers e.g. AH Plus, iRootSP (20,21,206,212,229,230). In a recent study, the reason for the low bond strength of MTA Fillapex was claimed to be the low adhesion capacity of the tag-like structures (20). The resin component of MTA Fillapex may affect its bonding to dentin (21). Exposing MTA Fillapex specimens to a solubility test in deionized water for 7 days has shown porosities and cracks in the resin matrix under SEM (216).

A dye penetration study using methylene blue for 72 hours on single-rooted teeth reported that MTA Fillapex has significantly higher microleakage values than MTA and AH Plus (231). Although another investigation using Rhodamine B for 24 hours showed acceptable dye leakage in the MTA Fillapex group (232).

Biological properties

Most previous studies on the cytotoxicity of MTA Fillapex have reported significant reduction in cell viability, which may be caused by lead released from the set sealer (220). Bin et al. (233) showed that MTA Fillapex has severely reduced the cell survival rates, even when they were exposed to 1:4 dilution of the sealer extract. However, after 48 hours the cytotoxicity was greatly reduced and a higher level of cell viability was detected. Scelza et al. (234) also found that MTA Fillapex was highly cytotoxic to human primary osteoblast cells. Two other separate investigations revealed that MTA Fillapex was more cytotoxic to 3T3 fibroblast cells than AH Plus during a 4-week incubation time (35,235). In contrast, one study found only an initial cytotoxicity effect existed during the setting of MTA Fillapex, and cell viability increased after 3 days (49). This study also showed that MTA Fillapex has the ability to stimulate nucleation sites for the formation of apatite crystals in human osteoblast-like cell culture. Different results reported by different studies could be due to different experimental treatment conditions (e.g. concentration and time), cell lines and viability

measurements. One possible explanation for the relatively high cytotoxicity of MTA Fillapex is the presence of salicylate resin in the material (233).

Although the resin released from MTA Fillapex reduces the biocompatibility of the material, it could also play a role in killing *E. faecalis* which can survive in alkaline environments (236). MTA Fillapex has been reported to have antibiofilm activity against *E. faecalis* in dentin block when investigated using the direct contact test (DCT) 7 days post-manipulation (226). Another study using the agar diffusion test demonstrated the antibacterial action of MTA Fillapex against *E. faecalis*, however, no antibacterial action was found when the DCT method was used (237).

Besides the influence on the antibacterial activity of MTA Fillapex, cytotoxicity also influences the tissue reaction. In two studies, MTA Fillapex and several other endodontic sealers were implanted into the subcutaneous connective tissue of rats for several months and found a severe inflammatory reaction at 60 and 90 days with the presence of macrophages and lymphocytes by MTA Fillapex (41,238). In contrast, Gomes-Filho et al. (40) reported favorable reactions of the connective tissue with MTA Fillapex, therefore demonstrating biocompatibility and mineralization properties in a rat animal model. Assmann et al. (239) reported a decreased inflammatory response in bone tissue when using MTA Fillapex. The conflicting results between studies are probably related to different details in the experimental procedures and the classification of the host response intensity (238).

MTA Plus

Physico-chemical properties

MTA Plus (Avalon Biomed Inc., Bradenton, FL, USA) is a finer powder, lower-cost product that has a composition similar to tooth-colored ProRoot MTA, and can be mixed with a liquid or a gel (240). This tricalcium and dicalcium silicate-based material can be used as a root canal sealer when mixed with gel, which also improves the handling properties and washout resistance (241). By using a gel and varying the powder to gel ratio, different setting times and physical-rheologic properties can be obtained (240).

MTA Plus showed improved reactivity and prolonged capability to release calcium and increase the local pH to alkaline values compared to ProRoot MTA. The ion-releasing property is interlinked with its fine powder, porosity, water sorption, and solubility and the formation of calcium phosphate minerals.

One recent study evaluated the push-out bond strengths of MTA Plus and EndoSequence BC Sealer when they are used in a thermoplastic technique (241). MTA Plus and BC Sealer showed favorable bond strengths when used in a single-cone technique, whereas when using continuous wave technique MTA Plus exhibited significantly lower bond strengths than using single-cone technique (241). However, this difference was not observed in BC Sealer group, which probably indicates that the performance of the sealers were affected by their different composition. Nevertheless, a recent *in vitro* study found minimal chemical changes in MTA Plus when subjected to warm vertical compaction (242).

Regarding compressive strength, MTA Plus was reported to have similar values to ERRM and MTA, but higher values than QuickSet (Avalon Biomed Inc., Bradenton, FL, USA).

Biological properties

Both the gray and white versions of MTA Plus were found to be negligible in cytotoxic risks to rat odontoblast-like cells (MDPC-23) when using XTT assay and flow cytometry (243). The cytotoxic effects imposed by MTA Plus on MDPC-23 cells are both time and concentration dependent. Moreover, gray MTA Plus might favor more cell growth and viability compared with white MTA Plus (243).

When used as a capping agent after pulpotomy, dentinal bridging could be detected at 30 and 60 days when applied with MTA Plus, and the pulps were still vital 60 days after capping (244). MTA Plus showed equivalent properties to ProRoot MTA which is used for endodontic pulp-capping procedures.

Bioceramic gutta-percha

Unlike traditional gutta-percha, EndoSequence BC gutta-percha (Brasseler USA, Savannah, GA) is subjected to a proprietary process of impregnating and coating each cone with bioceramic nanoparticles.

Table 1: Summary of reported physico-chemical and biological properties of bioceramic materials used in endodontics

	Biodentine	MTA	MTA Angelus	ERRM (or iRoot BP Plus)	BC Sealer (or iRoot SP)	MTA Fillapex	MTA Plus
pH	11.7–12.4 (69)	9.0–12.5 (16,162)	7.3–9.6 (162,167,168)	7.3–8.9 (192)	10.3–11.1 (34)	9.7–10.5 (35,226,237)	8.3–11.7 (243)
Calcium release (mg/L)	14.7–34.0 (71)	9.7–24.0 (10,71)	0.8–122.3 (162,167,168,216)	179.6 (216)	2.5–11.3 (34,71)	144.4 (216)	7.7–43.4 (243)
Flow rate (mm)	–	–	–	–	26.9 (34)	31.0 (216)	–
Porosity (%)	6.8 (63)	30.3–38.4 (37)	28.0 (166)	–	–	–	40.3 (243)
Solubility (%)	< 0.0 (64)	1.7–2.8 (37)	-1.2–6.4 (167,168,216)	–	20.6 (216)	14.8–16.1 (216,226)	18.5 (243)
Radiopacity (mm Al)	3.3–4.1 (63,64)	7.1 (16)	5.3–6.9 (167)	–	3.8 (34)	7.0 (35)	
Setting time (h)	0.1–0.7 (63,64)	6.9 (16,97)	0.2–5.3 (12,167,168)	>24.0 (3)	72.0–240.0 (34,203)	>12.0 (233)	0.9 (243)
Microhardness (VHN or KHN)	48.4–130.0 VHN (63,64)	53.2–60.0 VHN (105,108), 46.6–52.3 KHN (109)	36.3–84.3 VHN (169)	–	>15.0 KHN (203)	–	–
Compressive strength (N)	67.1–316.4 (63,64)	60.0–101.7 (108,110)	53.4–81.3 (110)	41.0–43.0 (191)	–	–	32.0–47.0 (191)
Push-out bond strength (MPa)	6.47–7.64 (22,74)	3.0–9.4 (22,115–117)	–	–	0.8–3.4 (206,212,241)	0.2–3.0 (21,117, 206,212)	0.98–2.3 (241)
Flexural strength (MPa)	34.0 (63)	10.7–14.2 (113)	–	–	–	–	–
Cell viability (%)	60.0–100.0 (39,63)	55.0–110.0 (3,127,177)	88.9–105.4 (127,177,195)	40.0–110.0 (3,127,195)	>90.0–100.0 (221)	35.0–95.0 (35)	>80.0 (243)

Data are collected in ranges from different experimental conditions in various studies. Data are shown in values (references).

However, there are not enough studies about the physico-chemical and biological properties of the EndoSequence BC gutta-percha points for any conclusions to be drawn.

According to the manufacturer's instructions, the bioceramic cones strictly follow the ISO 6786 recommendations, and are laser verified for tip and taper accuracy. Each EndoSequence BC gutta-percha cone has undergone a unique stiffening process making them easy to work with inside the canal. When used with BC Sealer, the manufacturer claims that a monobloc can be achieved by the chemical and mechanical bonding to both dentin and the EndoSequence BC Points. BC Sealer and BC Points allow a "three-dimensional" bonded obturation with no shrinkage.

Bioactive glass

Bioactive glass (BAG), calcium sodium phosphosilicate, is an extensively studied bioceramic material used in the field of cariology, restorative dentistry, and periodontology due to its high biocompatibility and remarkable bioactive capability in forming apatite-like structure (45,245).

BAG is currently used in the dental clinic mainly for air polishing procedures, and is incorporated into desensitizing toothpastes, bonding materials, and bone regeneration materials (45,245,246). Although BAG has not been extensively applied in clinical endodontics, recent studies have shown some potential for its use in endodontic treatments. Mohn et al. (247) mixed BAG particles with 50 wt% bismuth oxide as a potential root canal filling material, and revealed radiopacity with an equivalent of 4.94 mm aluminum and high pH value. Another study incorporated BAG with composite materials and found the immediate sealing ability was improved (248).

BAG has a directly and an indirectly pH-related antibacterial effect (249). The effect which is not directly linked to pH is a result of ion release (250). Nanometric bioactive glass releases more alkaline species, and consequently displayed a stronger antimicrobial effect against clinical isolates of enterococci than the micron-sized BAG (251). When dentin is incorporated in the root canal disinfection it may increase BAG dissolution, causing elevated local pH and silica levels, therefore increasing the killing efficacy of BAG (252).

One study evaluated the use of BAG (PerioGlas) after retrograde filling with Super EBA cement in the treatment of periapical bone destruction (253). At the short-term follow-up of 9 months to 2 years, the frequency of positive healing outcome was higher in the EBA + PerioGlas group than among those treated by EBA alone.

Outlook

A summary of the physico-chemical and biological properties of bioceramic materials in endodontics is shown in Table 1. The application of bioceramic technology has changed both surgical and non-surgical endodontic treatment, providing a promising direction for the preservation of patients' teeth. Most of the current bioceramic materials have rapidly gained acceptance in clinical applications for their physico-chemical and biological properties. However, limitations still exist when compared to the criteria for an ideal material used for endodontic purposes. Indeed, it is expected that the presently available bioceramic materials will be further modified and developed to overcome the few remaining challenges.

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References

1. Haapasalo M, Endal U, Zandi H, Coil J. Eradication of endodontic infection by instrumentation and irrigation solutions. *Endod Topics* 2005: **10**: 77–102.
2. Darvell BW, Wu RC. "MTA"-an Hydraulic Silicate Cement: review update and setting reaction. *Dent Mater* 2011: **27**: 407–422.
3. Ma J, Shen Y, Stojicic S, Haapasalo M. Biocompatibility of two novel root repair materials. *J Endod* 2011: **37**: 793–798.
4. Koubi G, Colon P, Franquin J-C, Hartmann A, Richard G, Faure M-O, Lambert G. Clinical evaluation of the performance and safety of a new dentine substitute, Biodentine, in the restoration of posterior teeth—a prospective study. *Clin Oral Invest* 2013: **17**: 243–249.
5. Veljović D, Colić M, Kojić V, Bogdanović G, Kojić Z, Banjać A, et al. The effect of grain size on the biocompatibility, cell-materials interface, and mechanical properties of microwave-sintered bioceramics. *J Biomed Mater Res A* 2012: **100**: 3059–3070.

6. Atmeh AR, Chong EZ, Richard G, Festy F, Watson TF. Dentin-cement interfacial interaction: calcium silicates and polyalkenoates. *J Dent Res* 2012: **91**: 454–459.
7. Koch KA, Brave DG. Bioceramics, part I: the clinician's viewpoint. *Dent Today* 2012: **31**: 130–135.
8. Camilleri J, Pitt Ford TR. Mineral trioxide aggregate: a review of the constituents and biological properties of the material. *Int Endod J* 2006: **39**: 747–754.
9. Reyes-Carmona JF, Felippe MS, Felippe WT. Biomineralization ability and interaction of mineral trioxide aggregate and white Portland cement with dentin in a phosphate-containing fluid. *J Endod* 2009: **35**: 731–736.
10. Parirokh M, Torabinejad M. Mineral trioxide aggregate: a comprehensive literature review—Part I: chemical, physical, and antibacterial properties. *J Endod* 2010: **36**: 16–27.
11. Persson C, Engqvist H. Premixed calcium silicate cement for endodontic applications: injectability, setting time and radiopacity. *Biomatter* 2011: **1**: 76–80.
12. Santos AD, Araujo EB, Yukimitu K, Barbosa JC, Moraes JC. Setting time and thermal expansion of two endodontic cements. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2008: **106**: e77–79.
13. Miyamoto Y, Ishikawa K, Fukao H, Sawada M, Nagayama M, Kon M, Asaoka K. *In vivo* setting behaviour of fast-setting calcium phosphate cement. *Biomaterials* 1995: **16**: 855–860.
14. Ørstavik D. Physical properties of root canal sealers: measurement of flow, working time, and compressive strength. *Int Endod J* 1983: **16**: 99–107.
15. Algera TJ, Kleverlaan CJ, Prahl-Andersen B, Feilzer AJ. The influence of environmental conditions on the material properties of setting glass-ionomer cements. *Dent Mater* 2006: **22**: 852–856.
16. Torabinejad M, Hong CU, McDonald F, Pitt Ford TR. Physical and chemical properties of a new root-end filling material. *J Endod* 1995: **21**: 349–353.
17. Gancedo-Caravia L, Garcia-Barbero E. Influence of humidity and setting time on the push-out strength of mineral trioxide aggregate obturations. *J Endod* 2006: **32**: 894–896.
18. Moshaverinia A, Brantley WA, Chee WW, Rohpour N, Ansari S, Zheng F, et al. Measure of microhardness, fracture toughness and flexural strength of N-vinylcaprolactam (NVC)-containing glass-ionomer dental cements. *Dent Mater* 2010: **26**: 1137–1143.
19. Adabo GL, dos Santos Cruz CA, Fonseca RG, Vaz LG. The volumetric fraction of inorganic particles and the flexural strength of composites for posterior teeth. *J Dent* 2003: **31**: 353–359.
20. Sagsen B, Ustun Y, Demirbuga S, Pala K. Push-out bond strength of two new calcium silicate-based endodontic sealers to root canal dentine. *Int Endod J* 2011: **44**: 1088–1091.
21. Assmann E, Scarparo RK, Bottcher DE, Grecca FS. Dentin bond strength of two mineral trioxide aggregate-based and one epoxy resin-based sealers. *J Endod* 2012: **38**: 219–221.
22. Guneser MB, Akbulut MB, Eldeniz AU. Effect of various endodontic irrigants on the push-out bond strength of Biodentine and conventional root perforation repair materials. *J Endod* 2013: **39**: 380–384.
23. Lai YL, Yang ML, Lee SY. Microhardness and color changes of human dentin with repeated intracoronal bleaching. *Oper Dent* 2003: **28**: 786–792.
24. Okabe T, Sakamoto M, Takeuchi H, Matsushima K. Effects of pH on mineralization ability of human dental pulp cells. *J Endod* 2006: **32**: 198–201.
25. Holland R, de Souza V, Nery MJ, Faraco Júnior IM, Bernabé PF, Otoboni Filho JA, et al. Reaction of rat connective tissue to implanted dentin tube filled with mineral trioxide aggregate, Portland cement or calcium hydroxide. *Braz Dent J* 2001: **12**: 3–8.
26. Yang Q, Troczynski T, Liu DM. Influence of apatite seeds on the synthesis of calcium phosphate cement. *Biomaterials* 2002: **23**: 2751–2760.
27. Zhang W, Li Z, Peng B. Assessment of a new root canal sealer's apical sealing ability. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2009: **107**: e79–82.
28. Tronstad L, Andreasen JO, Hasselgren G, Kristerson L, Riis I. pH changes in dental tissues after root canal filling with calcium hydroxide. *J Endod* 1981: **7**: 17–21.
29. Fuss Z, Tsesis I, Lin S. Root resorption–diagnosis, classification and treatment choices based on stimulation factors. *Dent Traumatol* 2003: **19**: 175–182.
30. McHugh CP, Zhang P, Michalek S, Eleazer PD. pH required to kill *Enterococcus faecalis in vitro. J Endod* 2004: **30**: 218–219.
31. Stuart CH, Schwartz SA, Beeson TJ, Owatz CB. *Enterococcus faecalis*: its role in root canal treatment failure and current concepts in retreatment. *J Endod* 2006: **32**: 93–98.
32. Trope M, Yesilsoy C, Koren L, Moshonov J, Friedman S. Effect of different endodontic treatment protocols on periodontal repair and root resorption of replanted dog teeth. *J Endod* 1992: **18**: 492–496.
33. Hwang YC, Lee SH, Hwang IN, Kang IC, Kim MS, Kim SH, et al. Chemical composition, radiopacity, and biocompatibility of Portland cement with bismuth oxide. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2009: **107**: e96–102.
34. Candeiro GT, Correia FC, Duarte MA, Ribeiro-Siqueira DC, Gavini G. Evaluation of radiopacity, pH, release of calcium ions, and flow of a bioceramic root canal sealer. *J Endod* 2012: **38**: 842–845.
35. Silva EJ, Rosa TP, Herrera DR, Jacinto RC, Gomes BP, Zaia AA. Evaluation of cytotoxicity and physicochemical properties of calcium silicate-based endodontic sealer MTA Fillapex. *J Endod* 2013: **39**: 274–277.
36. Holt DM, Watts JD, Beeson TJ, Kirkpatrick TC, Rutledge RE. The anti-microbial effect against *Enterococcus faecalis* and the compressive strength of two types of mineral trioxide aggregate mixed with sterile water or 2% chlorhexidine liquid. *J Endod* 2007: **33**: 844–847.

37. Fridland M, Rosado R. Mineral trioxide aggregate (MTA) solubility and porosity with different water-to-powder ratios. *J Endod* 2003: **29**: 814–817.
38. Coomaraswamy KS, Lumley PJ, Hofmann MP. Effect of bismuth oxide radioopacifier content on the material properties of an endodontic Portland cement-based (MTA-like) system. *J Endod* 2007: **33**: 295–298.
39. Zhou H, Shen Y, Wang Z, Li L, Zheng Y, Häkkinen L, et al. *In vitro* cytotoxicity evaluation of a novel root repair material. *J Endod* 2013: **39**: 478–483.
40. Gomes-Filho JE, Watanabe S, Lodi CS, Cintra LT, Nery MJ, Filho JA, et al. Rat tissue reaction to MTA FILLAPEX(R). *Dent Traumatol* 2012: **28**: 452–456.
41. Zmener O, Martinez Lalis R, Pameijer CH, Chaves C, Kokubu G, Grana D. Reaction of rat subcutaneous connective tissue to a mineral trioxide aggregate-based and a zinc oxide and eugenol sealer. *J Endod* 2012: **38**: 1233–1238.
42. Zanini M, Sautier JM, Berdal A, Simon S. Biodentine induces immortalized murine pulp cell differentiation into odontoblast-like cells and stimulates biomineralization. *J Endod* 2012: **38**: 1220–1226.
43. Tay FR, Pashley DH, Rueggeberg FA, Loushine RJ, Weller RN. Calcium phosphate phase transformation produced by the interaction of the Portland cement component of white mineral trioxide aggregate with a phosphate-containing fluid. *J Endod* 2007: **33**: 1347–1351.
44. Liu Y, Kim YK, Dai L, Li N, Khan SO, Pashley DH, et al. Hierarchical and non-hierarchical mineralisation of collagen. *Biomaterials* 2011: **32**: 1291–1300.
45. Wang Z, Jiang T, Sauro S, Wang Y, Thompson I, Watson TF, et al. Dentine remineralization induced by two bioactive glasses developed for air abrasion purposes. *J Dent* 2011: **39**: 746–756.
46. Leiendecker AP, Qi Y-P, Sawyer AN, Niu L-N, Agee KA, Loushine RJ, et al. Effects of calcium silicate-based materials on collagen matrix integrity of mineralized dentin. *J Endod* 2012: **38**: 829–833.
47. Wang Z, Jiang T, Sauro S, Pashley DH, Toledano M, Osorio R, et al. The dentine remineralization activity of a desensitizing bioactive glass-containing toothpaste: an *in vitro* study. *Aust Dent J* 2011: **56**: 372–381.
48. Wang Z, Jiang T, Sauro S, Wang Y, Xing W, Liang S, et al. Nerve-targeted desensitizing toothpastes occlude dentin tubules and induce mineral precipitation. *Am J Dent* 2012: **25**: 26–30.
49. Salles LP, Gomes-Cornélio AL, Guimarães FC, Herrera BS, Bao SN, Rossa-Junior C, et al. Mineral trioxide aggregate-based endodontic sealer stimulates hydroxyapatite nucleation in human osteoblast-like cell culture. *J Endod* 2012: **38**: 971–976.
50. Smith AJ, Cassidy N, Perry H, Bègue-Kirn C, Ruch JV, Lesot H. Reactionary dentinogenesis. *Int J Dev Biol* 1995: **39**: 273–280.
51. Cox CF, Subay RK, Ostro E, Suzuki S, Suzuki SH. Tunnel defects in dentin bridges: their formation following direct pulp capping. *Oper Dent* 1996: **21**: 4–11.
52. Schröder U. Effects of calcium hydroxide-containing pulp-capping agents on pulp cell migration, proliferation, and differentiation. *J Dent Res* 1985: **64**(Spec No): 541–548.
53. Nair PN, Henry S, Cano V, Vera J. Microbial status of apical root canal system of human mandibular first molars with primary apical periodontitis after "one-visit" endodontic treatment. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2005: **99**: 231–252.
54. Spångberg L, Haapasalo M. Rationale and efficacy of root canal medicaments and root filling materials with emphasis on treatment outcome. *Endod Topics* 2002: **2**: 35–58.
55. Siqueira JF Jr, Gonçalves R. Antibacterial activities of root canal sealers against selected anaerobic bacteria. *J Endod* 1996: **22**: 79–80.
56. Cobankara FK, Altinöz HC, Ergani O, Kav K, Belli S. *In vitro* antibacterial activities of root-canal sealers by using two different methods. *J Endod* 2004: **30**: 57–60.
57. Fuss Z, Charniaque O, Pilo R, Weiss E. Effect of various mixing ratios on antibacterial properties and hardness of endodontic sealers. *J Endod* 2000: **26**: 519–522.
58. Pizzo G, Giammanco GM, Cumbo E, Nicolosi G, Gallina G. *In vitro* antibacterial activity of endodontic sealers. *J Dent* 2006: **34**: 35–40.
59. Weiss EI, Shalhav M, Fuss Z. Assessment of antibacterial activity of endodontic sealers by a direct contact test. *Endod Dent Traumatol* 1996: **12**: 179–184.
60. Ma J, Wang Z, Shen Y, Haapasalo M. A new noninvasive model to study the effectiveness of dentin disinfection by using confocal laser scanning microscopy. *J Endod* 2011: **37**: 1380–1385.
61. Wang Z, Shen Y, Haapasalo M. Effectiveness of endodontic disinfecting solutions against young and old *Enterococcus faecalis* biofilms in dentin canals. *J Endod* 2012: **38**: 1376–1379.
62. Wang Z, Shen Y, Ma J, Haapasalo M. The effect of detergents on the antibacterial activity of disinfecting solutions in dentin. *J Endod* 2012: **38**: 948–953.
63. Biodentine scientific file. Active Biosilicate Technology™, Septodont. Saint-Maur-des-Fossés Cedex, France: R&D Department. 2010: www.septodont.fr.
64. Grech L, Mallia B, Camilleri J. Investigation of the physical properties of tricalcium silicate cement-based root-end filling materials. *Dent Mater* 2013: **29**: e20–28.
65. Zhang H, Pappen FG, Haapasalo M. Dentin enhances the antibacterial effect of mineral trioxide aggregate and BioAggregate. *J Endod* 2009: **35**: 221–224.
66. Camilleri J, Sorrentino F, Damidot D. Characterization of un-hydrated and hydrated BioAggregate and MTA Angelus. *Clin Oral Investig* 2015: **19**: 689–698.
67. Setbon HM, Devaux J, Iserentant A, Leloup G, Leprince JG. Influence of composition on setting

kinetics of new injectable and/or fast setting tricalcium silicate cements. *Dent Mater* 2014: **30**: 1291–1303.
68. Keskin C, Demiryurek EO, Ozyurek T. Color stabilities of calcium silicate-based materials in contact with different irrigation solutions. *J Endod* 2015: **41**: 409–411.
69. Grech L, Mallia B, Camilleri J. Characterization of set Intermediate Restorative Material, Biodentine, BioAggregate and a prototype calcium silicate cement for use as root-end filling materials. *Int Endod J* 2013: **46**: 632–641.
70. Natale LC, Rodrigues MC, Xavier TA, Simoes A, de Souza DN, Braga RR. Ion release and mechanical properties of calcium silicate and calcium hydroxide materials used for pulp capping. *Int Endod J* 2015: **48**: 89–94.
71. Han L, Okiji T. Bioactivity evaluation of three calcium silicate-based endodontic materials. *Int Endod J* 2013: **46**: 808–814.
72. Camilleri J. Investigation of Biodentine as dentine replacement material. *J Dent* 2013: **41**: 600–610.
73. Han L, Okiji T. Uptake of calcium and silicon released from calcium silicate-based endodontic materials into root canal dentine. *Int Endod J* 2011: **44**: 1081–1087.
74. EL-Ma'aita AM, Qualtrougha AJ, Watts DC. The effect of smear layer on the push-out bond strength of root canal calcium silicate cements. *Dent Mater* 2013: **29**: 797–803.
75. Sawyer AN, Nikonov SY, Pancio AK, Niu LN, Agee KA, Loushine RJ, et al. Effects of calcium silicate-based materials on the flexural properties of dentin. *J Endod* 2012: **38**: 680–683.
76. Pérard M, Clerc J, Meary F, Pérez F, Tricot-Doleux S, Pellen-Mussi P. Spheroid model study comparing the biocompatibility of Biodentine and MTA. *J Mater Sci Mater Med* 2013: **24**: 1527–1534.
77. Luo Z, Li D, Kohli MR, Yu Q, Kim S, He WX. Effect of Biodentine on the proliferation, migration and adhesion of human dental pulp stem cells. *J Dent* 2014: **42**: 490–497.
78. Jung JY, Woo SM, Lee BN, Koh JT, Nor JE, Hwang YC. Effect of Biodentine and BioAggregate on odontoblastic differentiation via mitogen-activated protein kinase pathway in human dental pulp cells. *Int Endod J* 2015: **48**: 177–184.
79. Luo Z, Kohli MR, Yu Q, Kim S, Qu T, He WX. Biodentine induces human dental pulp stem cell differentiation through mitogen-activated protein kinase and calcium-/calmodulin-dependent protein kinase II pathways. *J Endod* 2014: **40**: 937–942.
80. Lee BN, Lee KN, Koh JT, Min KS, Chang HS, Hwang IN, et al. Effects of 3 endodontic bioactive cements on osteogenic differentiation in mesenchymal stem cells. *J Endod* 2014: **40**: 1217–1222.
81. Tran XV, Gorin C, Willig C, Baroukh B, Pellat B, Decup F, et al. Effect of a calcium-silicate-based restorative cement on pulp repair. *J Dent Res* 2012: **91**: 1166–1171.
82. Mori GG, Teixeira LM, de Oliveira DL, Jacomini LM, da Silva SR. Biocompatibility evaluation of Biodentine in subcutaneous tissue of rats. *J Endod* 2014: **40**: 1485–1488.
83. Shayegan A, Jurysta C, Atash R, Petein M, Abbeele AV. Biodentine used as a pulp-capping agent in primary pig teeth. *Pediatr Dent* 2012: **34**: e202–208.
84. Tziafa C, Koliniotou-Koumpia E, Papadimitriou S, Tziafas D. Dentinogenic responses after direct pulp capping of miniature swine teeth with Biodentine. *J Endod* 2014: **40**: 1967–1971.
85. Kim JR, Nosrat A, Fouad AF. Interfacial characteristics of Biodentine and MTA with dentine in simulated body fluid. *J Dent* 2015: **43**: 241–247.
86. Malkondu O, Karapinar Kazandag M, Kazazoglu E. A review on Biodentine, a contemporary dentine replacement and repair material. *Biomed Res Int* 2014: **2014**: 160951.
87. Corral Nunez CM, Bosomworth HJ, Field C, Whitworth JM, Valentine RA. Biodentine and mineral trioxide aggregate induce similar cellular responses in a fibroblast cell line. *J Endod* 2014: **40**: 406–411.
88. Chang SW, Lee SY, Ann HJ, Kum KY, Kim EC. Effects of calcium silicate endodontic cements on biocompatibility and mineralization-inducing potentials in human dental pulp cells. *J Endod* 2014: **40**: 1194–1200.
89. Nowicka A, Lipski M, Parafiniuk M, Sporniak-Tutak K, Lichota D, Kosierkiewicz A, et al. Response of human dental pulp capped with Biodentine and mineral trioxide aggregate. *J Endod* 2013: **39**: 743–747.
90. Kratchman SI. Perforation repair and one-step apexification procedures. *Dent Clin North Am* 2004: **48**: 291–307.
91. Camilleri J, Montesin FE, Brady K, Sweeney R, Curtis RV, Ford TR. The constitution of mineral trioxide aggregate. *Dent Mater* 2005: **21**: 297–303.
92. Asgary S, Parirokh M, Eghbal MJ, Stowe S, Brink F. A qualitative X-ray analysis of white and grey mineral trioxide aggregate using compositional imaging. *J Mater Sci Mater Med* 2006: **17**: 187–191.
93. Asgary S, Eghbal MJ, Parirokh M, Ghoddusi J, Kheirieh S, Brink F. Comparison of mineral trioxide aggregate's composition with Portland cements and a new endodontic cement. *J Endod* 2009: **35**: 243–250.
94. Saghiri MA, Gutmann JL, Orangi J, Asatourian A, Sheibani N. Radiopacifier particle size impacts the physical properties of tricalcium silicate-based cements. *J Endod* 2015: **41**: 225–230.
95. Roberts HW, Toth JM, Berzins DW, Charlton DG. Mineral trioxide aggregate material use in endodontic treatment: a review of the literature. *Dent Mater* 2008: **24**: 149–164.
96. Torabinejad M, Watson TF, Pitt Ford TR. Sealing ability of a mineral trioxide aggregate when used as a root end filling material. *J Endod* 1993: **19**: 591–595.
97. Gandolfi MG, Iacono F, Agee K, Siboni F, Tay F, Pashley DH, et al. Setting time and expansion in different soaking media of experimental accelerated

calcium-silicate cements and ProRoot MTA. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2009: **108**: e39–45.
98. Islam I, Chng HK, Yap AU. Comparison of the physical and mechanical properties of MTA and Portland cement. *J Endod* 2006: **32**: 193–197.
99. Chng HK, Islam I, Yap AU, Tong YW, Koh ET. Properties of a new root-end filling material. *J Endod* 2005: **31**: 665–668.
100. Fridland M, Rosado R. MTA solubility: a long term study. *J Endod* 2005: **31**: 376–379.
101. de Vasconcelos BC, Bernardes RA, Cruz SM, Duarte MA, Padilha Pde M, Bernardineli N, et al. Evaluation of pH and calcium ion release of new root-end filling materials. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2009: **108**: 135–139.
102. Poggio C, Lombardini M, Alessandro C, Simonetta R. Solubility of root-end-filling materials: a comparative study. *J Endod* 2007: **33**: 1094–1097.
103. Danesh G, Dammaschke T, Gerth HU, Zandbiglari T, Schäfer E. A comparative study of selected properties of ProRoot mineral trioxide aggregate and two Portland cements. *Int Endod J* 2006: **39**: 213–219.
104. Shie MY, Huang TH, Kao CT, Huang CH, Ding SJ. The effect of a physiologic solution pH on properties of white mineral trioxide aggregate. *J Endod* 2009: **35**: 98–101.
105. Namazikhah MS, Nekoofar MH, Sheykhrezae MS, Salariyeh S, Hayes SJ, Bryant ST, et al. The effect of pH on surface hardness and microstructure of mineral trioxide aggregate. *Int Endod J* 2008: **41**: 108–116.
106. Lee YL, Lin FH, Wang WH, Ritchie HH, Lan WH, Lin CP. Effects of EDTA on the hydration mechanism of mineral trioxide aggregate. *J Dent Res* 2007: **86**: 534–538.
107. Kayahan MB, Nekoofar MH, Kazandağ M, Canpolat C, Malkondu O, Kaptan F, et al. Effect of acid-etching procedure on selected physical properties of mineral trioxide aggregate. *Int Endod J* 2009: **42**: 1004–1014.
108. Nekoofar MH, Adusei G, Sheykhrezae MS, Hayes SJ, Bryant ST, Dummer PM. The effect of condensation pressure on selected physical properties of mineral trioxide aggregate. *Int Endod J* 2007: **40**: 453–461.
109. Lee YL, Lee BS, Lin FH, Yun Lin A, Lan WH, Lin CP. Effects of physiological environments on the hydration behavior of mineral trioxide aggregate. *Biomaterials* 2004: **25**: 787–793.
110. Basturk FB, Nekoofar MH, Günday M, Dummer PM. The effect of various mixing and placement techniques on the compressive strength of mineral trioxide aggregate. *J Endod* 2013: **39**: 111–114.
111. Formosa LM, Mallia B, Camilleri J. Mineral trioxide aggregate with anti-washout gel—properties and microstructure. *Dent Mater* 2013: **29**: 294–306.
112. Formosa LM, Mallia B, Camilleri J. The effect of curing conditions on the physical properties of tricalcium silicate cement for use as a dental biomaterial. *Int Endod J* 2012: **45**: 326–336.
113. Walker MP, Diliberto A, Lee C. Effect of setting conditions on mineral trioxide aggregate flexural strength. *J Endod* 2006: **32**: 334–336.
114. Sahebi S, Nabavizadeh M, Dolatkhah V, Jamshidi D. Short term effect of calcium hydroxide, mineral trioxide aggregate and calcium-enriched mixture cement on the strength of bovine root dentin. *Iran Endod J* 2012: **7**: 68–73.
115. Shokouhinejad N, Nekoofar MH, Iravani A, Kharrazifard MJ, Dummer PM. Effect of acidic environment on the push-out bond strength of mineral trioxide aggregate. *J Endod* 2010: **36**: 871–874.
116. Saghiri MA, Shokouhinejad N, Lotfi M, Aminsobhani M, Saghiri AM. Push-out bond strength of mineral trioxide aggregate in the presence of alkaline pH. *J Endod* 2010: **36**: 1856–1859.
117. Reyes-Carmona JF, Felippe MS, Felippe WT. The biomineralization ability of mineral trioxide aggregate and Portland cement on dentin enhances the push-out strength. *J Endod* 2010: **36**: 286–291.
118. Osorio RM, Hefti A, Vertucci FJ, Shawley AL. Cytotoxicity of endodontic materials. *J Endod* 1998: **24**: 91–96.
119. Zhu Q, Haglund R, Safavi KE, Spångberg LS. Adhesion of human osteoblasts on root-end filling materials. *J Endod* 2000: **26**: 404–406.
120. Pistorius A, Willershausen B, Briseño Marroquin B. Effect of apical root-end filling materials on gingival fibroblasts. *Int Endod J* 2003: **36**: 610–615.
121. Pelliccioni GA, Ciapetti G, Cenni E, Granchi D, Nanni M, Pagani S, et al. Evaluation of osteoblast-like cell response to Proroot MTA (mineral trioxide aggregate) cement. *J Mater Sci Mater Med* 2004: **15**: 167–173.
122. Huang TH, Yang CC, Ding SJ, Yan M, Chou MY, Kao CT. Biocompatibility of human osteosarcoma cells to root end filling materials. *J Biomed Mater Res B Appl Biomater* 2005: **72**: 140–145.
123. Souza NJ, Justo GZ, Oliveira CR, Haun M, Bincoletto C. Cytotoxicity of materials used in perforation repair tested using the V79 fibroblast cell line and the granulocyte-macrophage progenitor cells. *Int Endod J* 2006: **39**: 40–47.
124. Yoshimine Y, Ono M, Akamine A. *In vitro* comparison of the biocompatibility of mineral trioxide aggregate, 4META/MMA-TBB resin, and intermediate restorative material as root-end-filling materials. *J Endod* 2007: **33**: 1066–1069.
125. Vajrabhaya LO, Korsuwannawong S, Jantarat J, Korre S. Biocompatibility of furcal perforation repair material using cell culture technique: Ketac Molar versus ProRoot MTA. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2006: **102**: e48–e50.
126. Gorduysus M, Avcu N, Gorduysus O, Pekel A, Baran Y, Avcu F, et al. Cytotoxic effects of four different endodontic materials in human periodontal ligament fibroblasts. *J Endod* 2007: **33**: 1450–1454.
127. Damas BA, Wheater MA, Bringas JS, Hoen MM. Cytotoxicity comparison of mineral trioxide

aggregates and EndoSequence bioceramic root repair materials. *J Endod* 2011: **37**: 372–375.
128. Ciasca M, Aminoshariae A, Jin G, Montagnese T, Mickel A. A comparison of the cytotoxicity and pro-inflammatory cytokine production of EndoSequence root repair material and ProRoot mineral trioxide aggregate in human osteoblast cell culture using reverse-transcriptase polymerase chain reaction. *J Endod* 2012: **38**: 486–489.
129. Zhang W, Li Z, Peng B. *Ex vivo* cytotoxicity of a new calcium silicate-based canal filling material. *Int Endod J* 2010: **43**: 769–774.
130. Camilleri J, Montesin FE, Papaioannou S, McDonald F, Pitt Ford TR. Biocompatibility of two commercial forms of mineral trioxide aggregate. *Int Endod J* 2004: **37**: 699–704.
131. Pérez AL, Spears R, Gutmann JL, Opperman LA. Osteoblasts and MG-63 osteosarcoma cells behave differently when in contact with ProRoot MTA and White MTA. *Int Endod J* 2003: **36**: 564–570.
132. Oviir T, Pagoria D, Ibarra G, Geurtsen W. Effects of gray and white mineral trioxide aggregate on the proliferation of oral keratinocytes and cementoblasts. *J Endod* 2006: **32**: 210–213.
133. Bonson S, Jeansonne BG, Lallier TE. Root-end filling materials alter fibroblast differentiation. *J Dent Res* 2004: **83**: 408–413.
134. Moghaddame-Jafari S, Mantellini MG, Botero TM, McDonald NJ, Nor JE. Effect of ProRoot MTA on pulp cell apoptosis and proliferation *in vitro*. *J Endod* 2005: **31**: 387–391.
135. Masuda-Murakami Y, Kobayashi M, Wang X, Yamada Y, Kimura Y, Hossain M, et al. Effects of mineral trioxide aggregate on the differentiation of rat dental pulp cells. *Acta Histochem* 2010: **112**: 452–458.
136. Min KS, Yang SH, Kim EC. The combined effect of mineral trioxide aggregate and enamel matrix derivative on odontoblastic differentiation in human dental pulp cells. *J Endod* 2009: **35**: 847–851.
137. Woo SM, Hwang YC, Lim HS, Choi NK, Kim SH, Kim WJ, et al. Effect of nifedipine on the differentiation of human dental pulp cells cultured with mineral trioxide aggregate. *J Endod* 2013: **39**: 801–805.
138. Tanomaru-Filho M, Tanomaru JM, Barros DB, Watanabe E, Ito IY. *In vitro* antimicrobial activity of endodontic sealers, MTA-based cements and Portland cement. *J Oral Sci* 2007: **49**: 41–45.
139. Asgary S, Kamrani FA. Antibacterial effects of five different root canal sealing materials. *J Oral Sci* 2008: **50**: 469–474.
140. Miyagak DC, de Carvalho EM, Robazza CR, Chavasco JK, Levorato GL. *In vitro* evaluation of the antimicrobial activity of endodontic sealers. *Braz Oral Res* 2006: **20**: 303–306.
141. Torabinejad M, Hong CU, Pitt Ford TR, Kettering JD. Antibacterial effects of some root end filling materials. *J Endod* 1995: **21**: 403–406.
142. Yasuda Y, Kamaguchi A, Saito T. *In vitro* evaluation of the antimicrobial activity of a new resin-based endodontic sealer against endodontic pathogens. *J Oral Sci* 2008: **50**: 309–313.
143. Heyder M, Kranz S, Völpel A, Pfister W, Watts DC, Jandt KD, et al. Antibacterial effect of different root canal sealers on three bacterial species. *Dent Mater* 2013: **29**: 542–549.
144. Song M, Kim E. A prospective randomized controlled study of mineral trioxide aggregate and super ethoxy-benzoic acid as root-end filling materials in endodontic microsurgery. *J Endod* 2012: **38**: 875–879.
145. Nair PN, Duncan HF, Pitt Ford TR, Luder HU. Histological, ultrastructural and quantitative investigations on the response of healthy human pulps to experimental capping with Mineral Trioxide Aggregate: a randomized controlled trial. 2008. *Int Endod J* 2009: **42**: 422–444.
146. Hilton TJ, Ferracane JL, Mancl L. Comparison of CaOH with MTA for direct pulp capping: a PBRN randomized clinical trial. *J Dent Res* 2013: **92**: 16S–22S.
147. Accorinte ML, Loguercio AD, Reis A, Carneiro E, Grande RH, Murata SS, et al. Response of human dental pulp capped with MTA and calcium hydroxide powder. *Oper Dent* 2008: **33**: 488–495.
148. Kumar V, Chawla A, Logani A, Shah N. Mineral trioxide aggregate pulpotomy: an ideal treatment option for management of talon cusp. *Contemp Clin Dent* 2012: **3**: 491–493.
149. Subay RK, Ilhan B, Ulukapi H. Mineral trioxide aggregate as a pulpotomy agent in immature teeth: long-term case report. *Eur J Dent* 2013: **7**: 133–138.
150. Sushynski JM, Zealand CM, Botero TM, Boynton JR, Majewski RF, Shelburne CE, et al. Comparison of gray mineral trioxide aggregate and diluted formocresol in pulpotomized primary molars: a 6- to 24-month observation. *Pediatr Dent* 2012: **34**: 120–128.
151. Mooney GC, North S. The current opinions and use of MTA for apical barrier formation of non-vital immature permanent incisors by consultants in paediatric dentistry in the UK. *Dent Traumatol* 2008: **24**: 65–69.
152. Brito-Junior M, Quintino AF, Camilo CC, Normanha JA, Faria-e-Silva AL. Nonsurgical endodontic management using MTA for perforative defect of internal root resorption: report of a long term follow-up. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2010: **110**: 784–788.
153. Bains R, Bains VK, Loomba K, Verma K, Nasir A. Management of pulpal floor perforation and grade II furcation involvement using mineral trioxide aggregate and platelet rich fibrin: a clinical report. *Contemp Clin Dent* 2012: **3**: S223–227.
154. Chala S, Abouqal R, Rida S. Apexification of immature teeth with calcium hydroxide or mineral trioxide aggregate: systematic review and meta-analysis. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2011: **112**: e36–42.
155. Jeeruphan T, Jantarat J, Yanpiset K, Suwannapan L, Khewsawai P, Hargreaves KM. Mahidol study 1:

comparison of radiographic and survival outcomes of immature teeth treated with either regenerative endodontic or apexification methods: a retrospective study. *J Endod* 2012: **38**: 1330–1336.
156. Fernandez-Yanez Sanchez A, Leco-Berrocal MI, Martinez-Gonzalez JM. Metaanalysis of filler materials in periapical surgery. *Med Oral Patol Oral Cir Bucal* 2008: **13**: E180–185.
157. Tsesis I, Rosen E, Taschieri S, Telishevsky Strauss Y, Ceresoli V, Del Fabbro M. Outcomes of surgical endodontic treatment performed by a modern technique: an updated meta-analysis of the literature. *J Endod* 2013: **39**: 332–339.
158. Parirokh M, Torabinejad M. Mineral trioxide aggregate: a comprehensive literature review–Part III: clinical applications, drawbacks, and mechanism of action. *J Endod* 2010: **36**: 400–413.
159. Bortoluzzi EA, Araujo GS, Guerreiro Tanomaru JM, Tanomaru-Filho M. Marginal gingiva discoloration by gray MTA: a case report. *J Endod* 2007: **33**: 325–327.
160. Mestres G, Aguilera FS, Manzanares N, Sauro S, Osorio R, Toledano M, et al. Magnesium phosphate cements for endodontic applications with improved long-term sealing ability. *Int Endod J* 2014: **47**: 127–139.
161. Oliveira IR, Andrade TL, Jacobovitz M, Pandolfelli VC. Bioactivity of calcium aluminate endodontic cement. *J Endod* 2013: **39**: 774–778.
162. Duarte MA, Demarchi AC, Yamashita JC, Kuga MC, Fraga Sde C. pH and calcium ion release of 2 root-end filling materials. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2003: **95**: 345–347.
163. Song JS, Mante FK, Romanow WJ, Kim S. Chemical analysis of powder and set forms of Portland cement, gray ProRoot MTA, white ProRoot MTA, and gray MTA-Angelus. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2006: **102**: 809–815.
164. Oliveira MG, Xavier CB, Demarco FF, Pinheiro AL, Costa AT, Pozza DH. Comparative chemical study of MTA and Portland cements. *Braz Dent J* 2007: **18**: 3–7.
165. Komabayashi T, Spångberg LS. Comparative analysis of the particle size and shape of commercially available mineral trioxide aggregates and Portland cement: a study with a flow particle image analyzer. *J Endod* 2008: **34**: 94–98.
166. Oliveira IR, Pandolfelli VC, Jacobovitz M. Chemical, physical and mechanical properties of a novel calcium aluminate endodontic cement. *Int Endod J* 2010: **43**: 1069–1076.
167. Cavenago BC, Pereira TC, Duarte MA, Ordinola-Zapata R, Marciano MA, Bramante CM, et al. Influence of powder-to-water ratio on radiopacity, setting time, pH, calcium ion release and a micro-CT volumetric solubility of white mineral trioxide aggregate. *Int Endod J* 2014: **47**: 120–126.
168. Vivan RR, Zapata RO, Zeferino MA, Bramante CM, Bernardineli N, Garcia RB, et al. Evaluation of the physical and chemical properties of two commercial and three experimental root-end filling materials. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2010: **110**: 250–256.
169. Nekoofar MH, Aseeley Z, Dummer PM. The effect of various mixing techniques on the surface microhardness of mineral trioxide aggregate. *Int Endod J* 2010: **43**: 312–320.
170. Pereira CL, Cenci MS, Demarco FF. Sealing ability of MTA, Super EBA, Vitremer and amalgam as root-end filling materials. *Braz Oral Res* 2004: **18**: 317–321.
171. Xavier CB, Weismann R, de Oliveira MG, Demarco FF, Pozza DH. Root-end filling materials: apical microleakage and marginal adaptation. *J Endod* 2005: **31**: 539–542.
172. Ribeiro DA, Sugui MM, Matsumoto MA, Duarte MA, Marques ME, Salvadori DM. Genotoxicity and cytotoxicity of mineral trioxide aggregate and regular and white Portland cements on Chinese hamster ovary (CHO) cells *in vitro*. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2006: **101**: 258–261.
173. Ribeiro DA, Matsumoto MA, Duarte MA, Marques ME, Salvadori DM. *Ex vivo* biocompatibility tests of regular and white forms of mineral trioxide aggregate. *Int Endod J* 2006: **39**: 26–30.
174. Ribeiro DA, Matsumoto MA, Duarte MA, Marques ME, Salvadori DM. *In vitro* biocompatibility tests of two commercial types of mineral trioxide aggregate. *Braz Oral Res* 2005: **19**: 183–187.
175. Miranda RB, Fidel SR, Boller MA. L929 cell response to root perforation repair cements: an *in vitro* cytotoxicity assay. *Braz Dent J* 2009: **20**: 22–26.
176. de Menezes JV, Takamori ER, Bijella MF, Granjeiro JM. *In vitro* toxicity of MTA compared with other primary teeth pulpotomy agents. *J Clin Pediatr Dent* 2009: **33**: 217–221.
177. Koulaouzidou EA, Economides N, Beltes P, Geromichalos G, Papazisis K. *In vitro* evaluation of the cytotoxicity of ProRoot MTA and MTA Angelus. *J Oral Sci* 2008: **50**: 397–402.
178. Camargo SE, Camargo CH, Hiller KA, Rode SM, Schweikl H, Schmalz G. Cytotoxicity and genotoxicity of pulp capping materials in two cell lines. *Int Endod J* 2009: **42**: 227–237.
179. De Deus G, Ximenes R, Gurgel-Filho ED, Plotkowski MC, Coutinho-Filho T. Cytotoxicity of MTA and Portland cement on human ECV 304 endothelial cells. *Int Endod J* 2005: **38**: 604–609.
180. Guven G, Cehreli ZC, Ural A, Serdar MA, Basak F. Effect of mineral trioxide aggregate cements on transforming growth factor beta1 and bone morphogenetic protein production by human fibroblasts *in vitro*. *J Endod* 2007: **33**: 447–450.
181. Gomes-Filho JE, Rodrigues G, Watanabe S, Estrada Bernabe PF, Lodi CS, Gomes AC, et al. Evaluation of the tissue reaction to fast endodontic cement (CER) and Angelus MTA. *J Endod* 2009: **35**: 1377–1380.
182. Gomes-Filho JE, Watanabe S, Bernabé PFE, de Moraes Costa MT. A mineral trioxide aggregate sealer stimulated mineralization. *J Endod* 2009: **35**: 256–260.

183. Sipert CR, Hussne RP, Nishiyama CK, Torres SA. *In vitro* antimicrobial activity of Fill Canal, Sealapex, Mineral Trioxide Aggregate, Portland cement and EndoRez. *Int Endod J* 2005: **38**: 539–543.
184. Tanomaru JM, Tanomaru-Filho M, Hotta J, Watanabe E, Ito IY. Antimicrobial activity of endodontic sealers based on calcium hydroxide and MTA. *Acta Odontol Latinoam* 2008: **21**: 147–151.
185. Luczaj-Cepowicz E, Pawinska M, Marczuk-Kolada G, Leszczynska K, Waszkiel D. Antibacterial activity of two mineral trioxide aggregate materials *in vitro* evaluation. *Ann Acad Med Stetin* 2008: **54**: 147–150.
186. Jacobovitz M, de Lima RK. Treatment of inflammatory internal root resorption with mineral trioxide aggregate: a case report. *Int Endod J* 2008: **41**: 905–912.
187. Sakai VT, Moretti AB, Oliveira TM, Fornetti AP, Santos CF, Machado MA, et al. Pulpotomy of human primary molars with MTA and Portland cement: a randomised controlled trial. *Br Dent J* 2009: **207**: E5.
188. Lovato KF, Sedgley CM. Antibacterial activity of EndoSequence root repair material and ProRoot MTA against clinical isolates of *Enterococcus faecalis*. *J Endod* 2011: **37**: 1542–1546.
189. Shi S, Bao ZF, Liu Y, Zhang DD, Chen X, Jiang LM, et al. Comparison of *in vivo* dental pulp responses to capping with iRoot BP Plus and mineral trioxide aggregate. *Int Endod J* 2015: in press.
190. Shokouhinejad N, Nekoofar MH, Razmi H, Sajadi S, Davies TE, Saghiri MA, et al. Bioactivity of EndoSequence root repair material and BioAggregate. *Int Endod J* 2012: **45**: 1127–1134.
191. Walsh RM, Woodmansey KF, Glickman GN, He J. Evaluation of compressive strength of hydraulic silicate-based root-end filling materials. *J Endod* 2014: **40**: 969–972.
192. Hansen SW, Marshall JG, Sedgley CM. Comparison of intracanal EndoSequence Root Repair Material and ProRoot MTA to induce pH changes in simulated root resorption defects over 4 weeks in matched pairs of human teeth. *J Endod* 2011: **37**: 502–506.
193. Wang Z, Ma J, Shen Y, Haapasalo M. Acidic pH weakens the microhardness and microstructure of three tricalcium silicate materials. *Int Endod J* 2015: **48**: 323–332.
194. Jiang Y, Zheng Q, Zhou X, Gao Y, Huang D. A comparative study on root canal repair materials: a cytocompatibility assessment in L929 and MG63 cells. *Scientific World Journal* 2014: **2014**: 463826.
195. Hirschman WR, Wheater MA, Bringas JS, Hoen MM. Cytotoxicity comparison of three current direct pulp-capping agents with a new bioceramic root repair putty. *J Endod* 2012: **38**: 385–388.
196. Modareszadeh MR, Di Fiore PM, Tipton DA, Salamat N. Cytotoxicity and alkaline phosphatase activity evaluation of EndoSequence root repair material. *J Endod* 2012: **38**: 1101–1105.
197. Zhang S, Yang X, Fan M. BioAggregate and iRoot BP Plus optimize the proliferation and mineralization ability of human dental pulp cells. *Int Endod J* 2013: **46**: 923–929.
198. De-Deus G, Canabarro A, Alves GG, Marins JR, Linhares AB, Granjeiro JM. Cytocompatibility of the ready-to-use bioceramic putty repair cement iRoot BP Plus with primary human osteoblasts. *Int Endod J* 2012: **45**: 508–513.
199. Liu S, Wang S, Dong Y. Evaluation of a bioceramic as a pulp capping agent *in vitro* and *in vivo*. *J Endod* 2015: **41**: 652–657.
200. Chen I, Karabucak B, Wang C, Wang HG, Koyama E, Kohli MR, et al. Healing after root-end microsurgery by using mineral trioxide aggregate and a new calcium silicate-based bioceramic material as root-end filling materials in dogs. *J Endod* 2015: **41**: 389–399.
201. Alsalleeh F, Chung N, Stephenson L. Antifungal activity of EndoSequence root repair material and mineral trioxide aggregate. *J Endod* 2014: **40**: 1815–1819.
202. Hess D, Solomon E, Spears R, He J. Retreatability of a bioceramic root canal sealing material. *J Endod* 2011: **37**: 1547–1549.
203. Loushine BA, Bryan TE, Looney SW, Gillen BM, Loushine RJ, Weller RN, et al. Setting properties and cytotoxicity evaluation of a premixed bioceramic root canal sealer. *J Endod* 2011: **37**: 673–677.
204. Ersev H, Yilmaz B, Dincol ME, Daglaroglu R. The efficacy of ProTaper Universal rotary retreatment instrumentation to remove single gutta-percha cones cemented with several endodontic sealers. *Int Endod J* 2012: **45**: 756–762.
205. Ma J, Al-Ashaw AJ, Shen Y, Gao Y, Yang Y, Zhang C, et al. Efficacy of ProTaper Universal Rotary Retreatment system for gutta-percha removal from oval root canals: a micro-computed tomography study. *J Endod* 2012: **38**: 1516–1520.
206. Nagas E, Uyanik MO, Eymirli A, Cehreli ZC, Vallittu PK, Lassila LV, et al. Dentin moisture conditions affect the adhesion of root canal sealers. *J Endod* 2012: **38**: 240–244.
207. Shokouhinejad N, Hoseini A, Gorjestani H, Raoof M, Assadian H, Shamshiri AR. Effect of phosphate-buffered saline on push-out bond strength of a new bioceramic sealer to root canal dentin. *Dent Res J (Isfahan)* 2012: **9**: 595–599.
208. Shokouhinejad N, Hoseini A, Gorjestani H, Shamshiri AR. The effect of different irrigation protocols for smear layer removal on bond strength of a new bioceramic sealer. *Iran Endod J* 2013: **8**: 10–13.
209. DeLong C, He J, Woodmansey KF. The effect of obturation technique on the push-out bond strength of calcium silicate sealers. *J Endod* 2015: **41**: 385–388.
210. Ersahan S, Aydin C. Dislocation resistance of iRoot SP, a calcium silicate-based sealer, from radicular dentine. *J Endod* 2010: **36**: 2000–2002.
211. Ozcan E, Capar ID, Cetin AR, Tuncdemir AR, Aydinbelge HA. The effect of calcium silicate-based sealer on the push-out bond strength of fibre posts. *Aust Dent J* 2012: **57**: 166–170.

212. Amin SA, Seyam RS, El-Samman MA. The effect of prior calcium hydroxide intracanal placement on the bond strength of two calcium silicate-based and an epoxy resin-based endodontic sealer. *J Endod* 2012: **38**: 696–699.
213. Jainaen A, Palamara JE, Messer HH. Push-out bond strengths of the dentine-sealer interface with and without a main cone. *Int Endod J* 2007: **40**: 882–890.
214. Huffman BP, Mai S, Pinna L, Weller RN, Primus CM, Gutmann JL, et al. Dislocation resistance of ProRoot Endo Sealer, a calcium silicate-based root canal sealer, from radicular dentine. *Int Endod J* 2009: **42**: 34–46.
215. Ghoneim AG, Lutfy RA, Sabet NE, Fayyad DM. Resistance to fracture of roots obturated with novel canal-filling systems. *J Endod* 2011: **37**: 1590–1592.
216. Borges RP, Sousa-Neto MD, Versiani MA, Rached-Junior FA, De-Deus G, Miranda CE, et al. Changes in the surface of four calcium silicate-containing endodontic materials and an epoxy resin-based sealer after a solubility test. *Int Endod J* 2012: **45**: 419–428.
217. Ersahan S, Aydin C. Solubility and apical sealing characteristics of a new calcium silicate-based root canal sealer in comparison to calcium hydroxide-, methacrylate resin- and epoxy resin-based sealers. *Acta Odontol Scand* 2013: **71**: 857–862.
218. Willershausen I, Callaway A, Briseno B, Willershausen B. *In vitro* analysis of the cytotoxicity and the antimicrobial effect of four endodontic sealers. *Head Face Med* 2011: **7**: 15.
219. Mukhtar-Fayyad D. Cytocompatibility of new bioceramic-based materials on human fibroblast cells (MRC-5). *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2011: **112**: e137–142.
220. Zhou HM, Du TF, Shen Y, Wang ZJ, Zheng YF, Haapasalo M. *In vitro* cytotoxicity of calcium silicate-containing endodontic sealers. *J Endod* 2015: **41**: 56–61.
221. Zoufan K, Jiang J, Komabayashi T, Wang YH, Safavi KE, Zhu Q. Cytotoxicity evaluation of Gutta Flow and Endo Sequence BC sealers. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2011: **112**: 657–661.
222. Guven EP, Tasli PN, Yalvac ME, Sofiev N, Kayahan MB, Sahin F. *In vitro* comparison of induction capacity and biomineralization ability of mineral trioxide aggregate and a bioceramic root canal sealer. *Int Endod J* 2013: **46**: 1173–1182.
223. Zhang W, Li Z, Peng B. Effects of iRoot SP on mineralization-related genes expression in MG63 cells. *J Endod* 2010: **36**: 1978–1982.
224. Zhang H, Shen Y, Ruse ND, Haapasalo M. Antibacterial activity of endodontic sealers by modified direct contact test against *Enterococcus faecalis*. *J Endod* 2009: **35**: 1051–1055.
225. Wang Z, Shen Y, Haapasalo M. Dentin extends the antibacterial effect of endodontic sealers against *Enterococcus faecalis* biofilms. *J Endod* 2014: **40**: 505–508.
226. Faria-Júnior NB, Tanomaru-Filho M, Berbert FL, Guerreiro-Tanomaru JM. Antibiofilm activity, pH and solubility of endodontic sealers. *Int Endod J* 2013: **46**: 755–762.
227. Xuereb M, Vella P, Damidot D, Sammut CV, Camilleri J. *In situ* assessment of the setting of tricalcium silicate-based sealers using a dentin pressure model. *J Endod* 2015: **41**: 111–124.
228. Ali SA, Zhong SP, Doherty PJ, Williams DF. Mechanisms of polymer degradation in implantable devices. I. Poly(caprolactone). *Biomaterials* 1993: **14**: 648–656.
229. Tanalp J, Dikbas I, Malkondu O, Ersev H, Gungor T, Bayirli G. Comparison of the fracture resistance of simulated immature permanent teeth using various canal filling materials and fiber posts. *Dent Traumatol* 2012: **28**: 457–464.
230. Sonmez IS, Sonmez D, Almaz ME. Evaluation of push-out bond strength of a new MTA-based sealer. *Eur Arch Paediatr Dent* 2013: **14**: 161–166.
231. Sonmez IS, Oba AA, Sonmez D, Almaz ME. *In vitro* evaluation of apical microleakage of a new MTA-based sealer. *Eur Arch Paediatr Dent* 2012: **13**: 252–255.
232. Gomes-Filho JE, Moreira JV, Watanabe S, Lodi CS, Cintra LT, Dezan Junior E, et al. Sealability of MTA and calcium hydroxide-containing sealers. *J Appl Oral Sci* 2012: **20**: 347–351.
233. Bin CV, Valera MC, Camargo SE, Rabelo SB, Silva GO, Balducci I, et al. Cytotoxicity and genotoxicity of root canal sealers based on mineral trioxide aggregate. *J Endod* 2012: **38**: 495–500.
234. Scelza MZ, Linhares AB, da Silva LE, Granjeiro JM, Alves GG. A multiparametric assay to compare the cytotoxicity of endodontic sealers with primary human osteoblasts. *Int Endod J* 2012: **45**: 12–18.
235. da Silva EJ, Santos CC, Zaia AA. Long-term cytotoxic effects of contemporary root canal sealers. *J Appl Oral Sci* 2013: **21**: 43–47.
236. Evans M, Davies JK, Sundqvist G, Figdor D. Mechanisms involved in the resistance of *Enterococcus faecalis* to calcium hydroxide. *Int Endod J* 2002: **35**: 221–228.
237. Morgental RD, Vier-Pelisser FV, Oliveira SD, Antunes FC, Cogo DM, Kopper PM. Antibacterial activity of two MTA-based root canal sealers. *Int Endod J* 2011: **44**: 1128–1133.
238. Tavares CO, Bottcher DE, Assmann E, Kopper PM, de Figueiredo JA, Grecca FS, et al. Tissue reactions to a new mineral trioxide aggregate-containing endodontic sealer. *J Endod* 2013: **39**: 653–657.
239. Assmann E, Bottcher DE, Hoppe CB, Grecca FS, Kopper PM. Evaluation of bone tissue response to a sealer containing mineral trioxide aggregate. *J Endod* 2015: **41**: 62–66.
240. Gandolfi MG, Siboni F, Primus CM, Prati C. Ion release, porosity, solubility, and bioactivity of MTA Plus tricalcium silicate. *J Endod* 2014: **40**: 1632–1637.
241. DeLong C, He J, Woodmansey KF. The effect of obturation technique on the push-out bond strength of calcium silicate sealers. *J Endod* 2015: **41**: 385–388.

242. Viapiana R, Baluci CA, Tanomaru-Filho M, Camilleri J. Investigation of chemical changes in sealers during application of the warm vertical compaction technique. *Int Endod J* 2015: **48**: 16–27.
243. Eid AA, Gosier JL, Primus CM, Hammond BD, Susin LF, Pashley DH, et al. *In vitro* biocompatibility and oxidative stress profiles of different hydraulic calcium silicate cements. *J Endod* 2014: **40**: 255–260.
244. Kramer PR, Woodmansey KF, White R, Primus CM, Opperman LA. Capping a pulpotomy with calcium aluminosilicate cement: comparison to mineral trioxide aggregates. *J Endod* 2014: **40**: 1429–1434.
245. Vollenweider M, Brunner TJ, Knecht S, Grass RN, Zehnder M, Imfeld T, et al. Remineralization of human dentin using ultrafine bioactive glass particles. *Acta Biomater* 2007: **3**: 936–943.
246. Blaker JJ, Gough JE, Maquet V, Notingher I, Boccaccini AR. *In vitro* evaluation of novel bioactive composites based on Bioglass-filled polylactide foams for bone tissue engineering scaffolds. *J Biomed Mater Res A* 2003: **67**: 1401–1411.
247. Mohn D, Zehnder M, Imfeld T, Stark WJ. Radioopaque nanosized bioactive glass for potential root canal application: evaluation of radiopacity, bioactivity and alkaline capacity. *Int Endod J* 2010: **43**: 210–217.
248. Mohn D, Bruhin C, Luechinger NA, Stark WJ, Imfeld T, Zehnder M. Composites made of flame-sprayed bioactive glass 45S5 and polymers: bioactivity and immediate sealing properties. *Int Endod J* 2010: **43**: 1037–1046.
249. Zehnder M, Soderling E, Salonen J, Waltimo T. Preliminary evaluation of bioactive glass S53P4 as an endodontic medication *in vitro*. *J Endod* 2004: **30**: 220–224.
250. Gubler M, Brunner TJ, Zehnder M, Waltimo T, Sener B, Stark WJ. Do bioactive glasses convey a disinfecting mechanism beyond a mere increase in pH? *Int Endod J* 2008: **41**: 670–678.
251. Waltimo T, Brunner TJ, Vollenweider M, Stark WJ, Zehnder M. Antimicrobial effect of nanometric bioactive glass 45S5. *J Dent Res* 2007: **86**: 754–757.
252. Zehnder M, Waltimo T, Sener B, Soderling E. Dentin enhances the effectiveness of bioactive glass S53P4 against a strain of *Enterococcus faecalis*. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2006: **101**: 530–535.
253. Pantchev A, Nohlert E, Tegelberg A. Endodontic surgery with and without inserts of bioactive glass PerioGlas—a clinical and radiographic follow-up. *Oral Maxillofac Surg* 2009: **13**: 21–26.