REVIEW

Chelating agents in root canal treatment: mode of action and indications for their use

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Abstract

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Chelating agents were introduced into endodontics as an aid for the preparation of narrow and calcified root canals in 1957 by Nygaard-Østby. A liquid solution of ethylenediaminetetraacetic acid (EDTA) was thought to chemically soften the root canal dentine and dissolve the smear layer, as well as to increase dentine permeability. Although the efficacy of EDTA preparations in softening root dentine has been debated, chelator preparations have regained popularity recently. Almost all manufacturers of nickel–titanium instruments recommend their use as a lubricant during rotary root canal preparation. Additionally, a final irrigation of the root canal with 15–17% EDTA solutions to dissolve the smear layer is recommended in many textbooks. This paper reviews the relevant literature on chelating agents, presents an overview of the chemical and pharmacological properties of EDTA preparations and makes recommendations for their clinical use.

Keywords: chelators, dentine hardness, dentine permeability, EDTA, root canal treatment, smear layer.

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Historical development of chelators

The term 'chelate' originates from the Greek word 'chele' (crab claw). Chelates are particularly stable complexes of metal ions with organic substances as a result of ring-shaped bonds. This stability is a result of the bond between the chelator, which has more than one pair of free electrons, and the central metal ion (Grossman *et al.* 1988, Zeeck *et al.* 1992; Fig. 1). The ability of chelators to bind and inactivate metallic ions is widely exploited in medicine. Chelators can be used to bring about excretion of dangerous ions in the case of metal poisoning or in the treatment of copper metabolism disturbances (Zeeck *et al.* 1992).

In 1951, the first reports on the demineralizing effect of ethylenediaminetetraacetic acid (EDTA) on dental hard tissues were published (Hahn & Reygadas 1951, Screebny & Nikiforuk 1951). Chelators were first introduced to endodontics by Nygaard-Østby (1957), who recommended the use of a 15% EDTA solution (pH 7.3) with the following composition:

- Disodium salt of EDTA (17.00 g)
- Aqua dest. (100.00 mL)
- 5 M sodium hydroxide (9.25 mL)

A few years later, a detergent was added in order to increase the cleaning and bactericidal potential of the EDTA solution, the new composition being known as EDTAC (Von der Fehr & Nygaard-Østby 1963). EDTAC is produced when EDTA is mixed with 0.84 g of a quarternary ammonium compound (Cetavlon; Goldberg & Abramovich 1977). This addition is aimed at reducing the surface tension of the irrigant, facilitating the wetting of the entire root canal wall and thereby increasing the ability of the chelators to penetrate the dentine. EDTA in its pure form already has a lower surface tension than 1 or 5% sodium hypochlorite (NaOCl), saline solution or distilled water (Tasman *et al.* 2000). Furthermore, EDTAC should have a greater antimicrobial effect than

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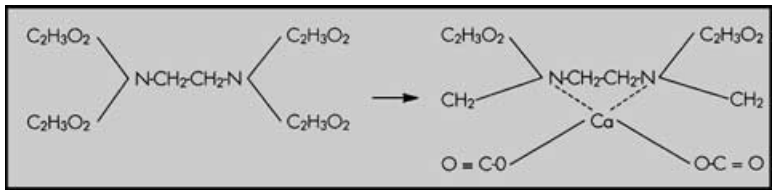

Figure 1 Chemical structure and mechanism of EDTA binding.

EDTA, although it also causes greater inflammatory reactions in soft tissue (Weine 1988). In contrast to this, no difference in the effectiveness of EDTAC and EDTA has been reported (Weinreb & Meier 1965).

Initially, chelators were used as liquids for irrigation during mechanical instrumentation of the root canal system. In 1969, Stewart *et al.* presented RC-Prep (premier Dental; Philadelphia, PA, USA), probably the best known paste-type chelating agent. Although the efficacy of liquid and paste-type EDTA preparations in softening root dentine has been a point of controversy, chelator preparations have been advocated frequently as adjuncts for root canal preparation, especially in narrow and calcified root canals (Serene 1976, Stock & Nehammer 1985, Stewart 1986, 1995, Weine 1988, Lovdahl & Gutmann 1997), and for removal of the smear layer (McComb & Smith 1975, Goldman *et al.* 1985, Berg *et al.* 1986, Baumgartner & Mader 1987, Ciucchi *et al.* 1989, Aktener & Bilkay 1993, Garberoglio & Becce 1994, Hottel *et al.* 1999, Çalt & Serper 2000, Di Lenarda *et al.* 2000, O'Connell *et al.* 2000, Scelza *et al.* 2000). Recently, paste-type chelators have regained popularity as almost all manufacturers of nickel–titanium instruments recommend their use as a lubricant during rotary root canal preparation, presumably to reduce the risk of instrument separation.

Chelator preparations

Liquid chelators

The most common liquid chelator preparations and their main ingredients are:

- Calcinase (lege artis, Dettenhausen, Germany) is a liqiud chelator preparation and contains 17% sodium edetate, sodium hydroxide as a stabilizor and purified water.
- REDTA (Roth International, Chicago, IL., USA) is a 17% EDTA solution with the addition of 0.84 g Cetyl-tri-methyl ammonium bromide (Cetrimide) to reduce surface tension. The other ingredients are 9.25 mL 5 M sodium hydroxide and 100 mL distilled water.
- EDTAC and DTPAC are solutions of EDTA (15%) and diethyl-triamine-penta acetic acid (DTPA) at pH 8. When 0.75 g of the detergent Cetyl-tri-methyl ammonium bromide is added to 100 mL of these solutions, respectively, two new solutions named EDTAC and DTPAC are produced (Pawlicka *et al.* 1981, 1982).
- EDTA-T (Formula & Açao Farmacia, Sao Paulo, Brazil) consists of 17% EDTA plus sodium lauryl ether sulfate (Tergentol) as a detergent (Scelza *et al.* 2000).
- EGTA (Sigma, St Louis, MO, USA) is a chelator whose main component is ethylene glycol bis (β-amino-ethyl ether)-*N,N,N′,N′*-tetra acetic acid. It is reported to bind Ca^{+} more specifically than EDTA (Çalt & Serper 2000).
- CDTA (experimental solution) is a 1% solution of cyclohexane-1,2-diaminetetraacetic acid (Cruz-Filho *et al.* 2001).
- Largal Ultra (Septodont, Paris, France) contains a 15% EDTA solution as a disodium salt, 0.75% Cetyl-tri-methyl ammonium bromide (Cetrimide) and sodium hydroxide to adjust the pH value to 7.4.
- Salvizol (Ravens, Konstanz, Germany) is based on a 5% aminoquinaldinumdiacetate in propylene glycol and has a pH of 6.6 (Kaufman *et al.* 1978).
- Decal (Veikko Auer, Helsinki, Finland) has a pH value of 3.4 and is composed of 5.3% oxyl-acetate, 4.6% ammonium oxyl-acetate and 0.06% Cetyl-tri-methyl ammonium bromide (Cetrimide), thereby combining the effects of a chelator complex and dissolution by an acid component.
- Tubulicid Plus (Dental Therapeutics, Nacka, Sweden) contains 1.5 g Amphoteric-2 (38%), 0.5 g benzalkonichloride, 3 g disodium EDTA dihydrate, phosphate buffer solution pH 7.3 q.s., 100 g distilled water and 50% citric acid.
- Hypaque (experimental solution) is composed of 5% NaOCl, 17% EDTA and hypaque, a high-contrast injectable dye for angiography and arteriography (Scarfe *et al.* 1995). Hypaque is an aqueous solution of two iodine salts, diatrizoate meglumine and sodium iodine. It is water soluble and has a pH of 6.7–7.7. This agent is intended to visualize the complexity of the root canal system and thus to combine the solving potential of EDTA and NaOCl and the radiopacity of the contrast solution (Ruddle 2002).

Paste-type chelators

Whilst the literature reports predominantly on the mode of action of liquid chelator solutions for root canal irriga-

tion, the chelators recommended for use during rotary root canal preparation have a paste or gel consistency. The best known paste chelators include the following substances:

- Calcinase slide (lege artis, Dettenhausen, Germany) contains 15% sodium EDTA and 58–64% water, but no peroxides, colourants or preservatives (self-preserving). The preparation has an alkaline pH value of 8–9, which remains stable under clinical conditions. According to the manufacturer, no EDTA precipitation occurs in combination with the commonly used irrigants. Furthermore, the gel can be mixed with water and therefore can be easily rinsed out of the root canal system. Because of its thixotropic nature, the gel is firm at room temperature and develops a creamy consistency when agitated. In this way, the EDTA preparation not only adheres well to the working instrument, but also disperses well inside the root canal. The manufacturers do not claim a pharmacological effect as such, because the complex effect of EDTA is only seen after the removal of the EDTA-softened surface dentine layer using mechanical instrumentation. Intermittent use in combination with a NaOCl solution is recommended.
- RC-Prep (Premier Dental, Philadelphia, PA, USA) is a combination of 10% urea peroxide, 15% EDTA and glycol in an aqueous ointment base. Oxygen is set free by the reaction of RC-Prep with a NaOCl irrigant so that pulpal remnants and blood coagulates can be easily removed from the root canal wall (Stewart *et al*. 1969). The manufacturer claims that discoloured teeth can be bleached in this way. Urea peroxide retains its antibacterial action in the presence of blood (Stewart *et al*. 1961). The glycol component of RC-Prep serves as a lubricant for instruments and is thought to inhibit the oxidation of EDTA by urea peroxide.
- Glyde file (DeTrey Dentsply, Konstanz, Germany) is composed of 15% EDTA and 10% urea peroxide in aqueous solution, and its viscosity is dependent on storage conditions. It was developed for use with NaOCl irrigants, because the oxygen release from urea peroxide caused effervescence and this is claimed to facilitate the removal of dentine particles and pulpal remnants. In addition to this, internal bleaching is claimed to take place.
- FileCare EDTA (VDW Antaeos, Munich, Germany) also is composed of 15% EDTA and 10% urea peroxide.
- File-EZE (Ultradent Products, South Jordan, UT, USA) is a chelating agent in an aqueous water-soluble solution containing 19% EDTA.

It should be noted that some of the chelator preparations listed above are distributed under different names in some countries.

Demineralization

Nygaard-Østby (1957) used the principle of a constant solubility product to explain the demineralization of dental hard tissue by EDTA and its sodium salt. An equilibrium is established between the saturated salt solution and the consolidated precipitate because ions from the precipitate constantly go into solution, whilst at the same time, ions from the solution are precipitated as solids. The concentration of the salt remains constant, and therefore the product of the concentrations of the ions in solution at a given temperature (the solubility product) remains stable.

According to Nygaard-Østby (1957), even lyophobic substances such as dentine, the mineral components of which are mainly phosphate and calcium, are soluble in water. When the disodium salt of EDTA is added to this equilibrium, calcium ions are removed from the solution. This leads to the dissolution of further ions from dentine so that the solubility product remains constant. Thus, chelators cause decalcification of dentine. A normal concentration of EDTA can remove 10.5 g from 100 g calcium (Pawlicka *et al*. 1981).

In the first study of Nygaard-Østby (1957), the EDTA (15% (pH 7.3))-treated samples were analysed using polarized microscopy. The root canal lumen was encircled by a clearly defined zone of demineralized dentine. The extension of the demineralized zone was dependent on the working time (20 min – 96 h). These experiments demonstrated that EDTAC had a rapid demineralizing effect. EDTAC is thought to have a lower surface tension than EDTA because of the addition of a detergent, thereby increasing the ability to penetrate deeper into the dentine. A 20–30-μm demineralized zone was apparent after 5 min. This increased to 30–40 μm after 30 min and to 50 μm after a working time of 24–48 h. This layer was separated from the deeper unchanged dentine by a clearly defined smooth demarcation line. Therefore, the solution did not permeate diffusely into the dentine and the effect was thought to be self-limiting because the demineralization did not extend beyond 50 μm even after a relatively long working time (Nygaard-Østby 1957).

Chelators such as EDTA form a stable complex with calcium. When all available ions have been bound, an equilibrium is formed and no further dissolution takes place. Using gravimetrical analyses, Seidberg & Schilder (1974) showed that the properties of EDTA were self-limiting. This limitation is thought to be because of pH changes during demineralization of dentine. Under neutral conditions, most chelators have a pH near the neutral value,

99% of the EDTA is present as $EDTAHNa_3$. The exchange of calcium from the dentine by hydrogen results in a subsequent decrease in pH. Because of the release of acid, the efficiency of EDTA decreases with time; on the other hand, the reaction of the acid with hydroxyapatite affects the solubility of dentine.

Chemically, two coexisting reactions can be distinguished: complex formation (Eqn. 1) and protonation (Eqn. 2; Perez *et al.* 1989):

$$EDTAH^{3-} + Ca^{2+} = EDTACa^{2-} + H \quad (1)$$

$$EDTAH^{3-} + H = EDTAH_2{}^{2-} \quad (2)$$

As this reaction proceeds, acid accumulates and protonation of EDTA prevails (Eqn. 2), thus decreasing the rate of demineralization. EDTA has four carboxyl groups, and the dissociation takes place in four steps each with its own dissociation constant (pK), ranging from $pK_1 = 2.0$ for the first to $pK_4 = 10.26$ for the fourth step. This means that the dissolution of EDTA takes place over a broad range of different pH values (Sand 1961).

In contrast, Patterson (1963) concluded from his studies that EDTA induced decalcification, which was not self-limiting and proceeded for up to 5 days, although the maximum penetration was 28 μm. The demineralization proceeds until all chelators have formed complexes with calcium. Dentine demineralization is observed at pH values of 4–5, but enamel is not affected. The difference in solubility can be explained by the differences in apatite crystal size, the presence of tubules in dentine and the favourable proportion of calcium to EDTA. Although dentine demineralization was thought not to be pH-dependent (Seidberg & Schilder 1974, Schmidt 1968), a neutral or alkaline EDTA solution gives an optimal effect (Screebny & Nikiforuk 1951, Nikiforuk & Screebny 1953, Rubin *et al.* 1979, Serper & Çalt 2002). This is supported by a study showing that the optimum pH for demineralization of dentine is between 5.0 and 6.0 (Cury *et al.* 1981). The demineralizing effect, measured as the amount of released phosphorus is stronger for solutions with a pH of 7.5 as compared to solutions with a pH of 9.0 (Serper & Çalt 2002). In the coronal and middle parts of the root canal, EDTA, with a neutral pH, dissolves significantly more calcium and phosphorus than RC-Prep (Verdelis *et al.* 1999). The authors concluded from their study that RC-Prep mainly decalcified and removed the loosely attached part of the superficial smear layer, but was not able to modify the subsurface dentine. Besides the low pH of RC-prep, insufficient wetting of the dentine and possible side interactions are discussed as possible reasons.

Further studies have shown that mechanical preparation in combination with EDTA could remove more calcium than instrumentation with physiological saline, but slightly less than preparation with 20% hydrochloric acid (Heling *et al.* 1965).

In a recent study (Hülsmann & Heckendorff 2002), the weight loss of dentine discs was measured after 3, 6 and 9 min of application of the chelator pastes Calcinase slide, RC-Prep and Glyde File Prep. There were significant differences between the control group (no chelating agent) and the chelator pastes, and between the different application times. No significant difference was found between the chelator pastes after 3 min. Calcinase slide caused greater mineral loss than RC-Prep after 6 and 9 min and Glyde File Prep was superior to RC-Prep after 6 min. Measuring the amount of liberated phosphorus at different intervals after exposure to EDTA solutions (1–15 min) with different concentrations (10 and 17%) and pH (7.5 and 9.0), the pH did not play any significant role, whereas time of exposure and concentration significantly influenced the demineralization of root dentine. Nevertheless, solutions with a pH of 7.5 performed more efficiently than those with a pH of 9.0 (Serper & Çalt 2002). root canal dentine showed severe peritubular and intratubular erosions after 10 min irrigation with a liquid EDTA chelator (17%), whereas a 1-min exposure was effective in removing the smear layer (Çalt & Serper 2002). After 3, 10 and 15 min of exposure, no differences in the amount of extracted Ca^{++} could be found between 17% EDTA and 10% citric acid, whereas EDTA-T showed worse results (Scelza *et al.* 2003). Following irrigation with 15% EDTA for 2 or 3 min and subsequent irrigation with 6% NaOCl for 2 or 3 min, erosion was found to be more pronounced than following irrigation with EDTA alone, suggesting that 6% NaOCl accelerates erosion of the dentinal tubules (Niu *et al.* 2002).

More recent results have shown that a neutral EDTA solution reduces the mineral and noncollagenous protein (NCP) component of dentine, leading to surface softening but not to erosion of the surface dentine layer (Kawasaki *et al.* 1999, Verdelis *et al.* 1999). EDTA can remove not only calcium ions but also water-soluble NCP and phosphoproteins at a neutral pH (Kuboki *et al.* 1979). Thus, not only calcium ions but also calcium bonded to the extracted fractions of NCPs is removed by EDTA. As the content of noncollagenous organic matrix decreases in the apical part of the root dentine, this may explain the lower degree of decalcification in this part of the root.

The use of EDTA followed by NaOCl irrigation significantly changes the calcium and phosphate content of

root dentine in contrast to irrigation with EDTA and RC-Prep alone, whereas the magnesium content increases (Dogan & Çalt 2001). Although not definitely clarified, the authors suggest that magnesium replaces calcium in dentine. On the other hand, earlier clinical, experimental and histological investigations cast doubt on the efficiency of EDTA for dentine demineralization under clinical conditions (Wandelt 1961, 1965, Ram 1980, Dow 1984). Fraser (1974) calculated that 0.02 mL of EDTA decalcified only about 0.35 mm^2 of dentine. EDTA solution seems to be limited in its ability to demineralize because each relatively large chelator molecule can only bind a single calcium ion. When all molecules are bound, the reaction stops. Wandelt (1961, 1965) stated that the desired effect can only be achieved when a sufficiently large amount of active substance for the respective surface area and enough time are available to allow the complex formation to take place. The author concluded from the results of his studies that the effect of chelators depends on the width of the root canal and that only an insufficient amount of active substance can be introduced into narrow canals.

Changes in dentine hardness

The hardness value of unaffected root dentine is between 40 and 75 kg mm^{-2} (Vickers hardness; Patterson 1963, Komiya & Kröncke 1968). Dentine hardness increases characteristically from the root canal lumen towards the cemento-dentinal junction, whereas the values in the apical-third are lower than in the middle and cervical sections of the root (Patterson 1963). In contrast, the hardness of the root canal wall is almost constant with a Vickers hardness of 88.78 kg mm^{-2} at the entrance to the root canal and 94.68 kg mm^{-2} at the apex (Fromme *et al.* 1970, Pawlicka *et al.* 1981).

Pawlicka (1982) reported that chelators can change the root dentine hardness by about 20 HV (Vickers hardness), whereby the greatest differences are to be found in dentine immediately adjacent to the root canal lumen. The effect of the chelator is already apparent after 5 min and cannot be significantly increased by extending the working time to 24 h. No difference in the change could be found between the chelators used: EDTA, EDTAC, DTPA and DTPAC. This was confirmed further by studies (Weinreb & Meier 1965) when it was demonstrated that chelators have the ability to soften dentine. The hardness of untreated dentine was 25 Knoop hardness number (KHN) near to the dentine–cement junction, reached a maximum of over 70 KHN and decreased again to 42 KHN in the immediate area of the root canal lumen. After 9 min treatment with EDTAC, the dentine hardness decreased from 60 to 45 KHN and after 24 h, the average hardness of the treated dentine was 7 KHN (Patterson 1963). EGTA solutions (1, 3 and 5%) significantly reduce dentine hardness when compared to distilled water; the degree of softening is dependent on the concentration of the chelating agent (Cruz-Filho *et al.* 2002). A comparative study of 15% EDTAC, 1% CDTA and 1% EGTA using the same technique revealed no significant differences between the three solutions investigated (Cruz-Filho *et al.* 2001). Fromme *et al.* (1970) used the chelator preparation Largal Ultra and found that the reduction in hardness took place in wide sections of the canal and at the canal entrance and not at the root tip or in narrow sections of the root. In general, these authors concluded that chelators showed a demineralizing effect on dental hard tissue, but were ineffective in narrow sections of the root. They believed that this was because of the difficulty in providing sufficient volume of material and exchanging used material within roots having a narrow lumen. Fraser (1974) also doubted the usefulness of chelators during root canal preparation. Whilst Fromme *et al.* (1970) applied chelators through the root canal entrance, the chelators used by Fraser (1974) – RC-Prep, Largal Ultra and Decal – were applied directly to the canal wall for 15 min. Whilst chelators have been shown to have a softening effect on dentine in the cervical- and middle-third of the root (20–40 μm deep depending on the preparation used), little or no effect has been shown in narrow areas of the apical section of the root (Hampson & Atkinson 1964, Wandelt 1965, Fromme *et al.* 1970, Fraser 1974). This is not only because of the difficulty in providing a sufficient amount of chelator to this part of the root canal, but also reflects the differences in structure between the middle, coronal and apical dentine (Pashley *et al.* 1985, Mjör *et al.* 2001).

Changes in dentine permeability

The diameter of dentine tubules decreases from 1.2 μm at the pulp–dentine junction to 0.4 μm at the cemento-dentinal junction (Pashley 2002). The number of tubules per square millimeter is also greater near to the pulp (58000 mm^{-2}) than further away from the pulp (10000 mm^{-2}; Mjör *et al.* 2002). As the tubule density reduces towards the apex, so does the dentine permeability (Fraser 1974). Furthermore, root dentine is not uniformly mineralized. Apical dentine is more frequently sclerosed, and is more mineralized (Vasiliadis *et al.*

1983). Intratubular mineralization can lead to narrowing of the dentine canal lumen (Schroeder 1992).

Dentine permeability is directly dependent on the area of the tubule lumina and in reverse proportion to the wall thickness of the root canal (Reeder *et al.* 1978). After mechanical preparation, the wall thickness of the root canal is reduced whilst the surface area of the lumen is increased. Additionally, the smear layer acts as a diffusion barrier, reducing dentine permeability by 25–49% (Pashley 1984, Pashley & Depew 1986, Pashley *et al.* 1988, Fogel & Pashley 1990).

Dentine permeability is increased (Cohen *et al.* 1970, Brännström 1984, Guignes *et al.* 1996) and a reduction in microleakage between the definitive root canal filling and the canal wall dentine is achieved (Cergneux *et al.* 1987, Petschelt *et al.* 1987, Wennberg & Ørstavik 1990, Behrend *et al.* 1996) after smear layer removal with the aid of EDTA. In addition, it is possible to obturate a greater number of lateral canals (Goldberg *et al.* 1986). SEM investigations show that the use of EDTA during preparation leads to enlargement of the dentinal tubule openings (Goldberg & Abramovich 1977, Hottel *et al.* 1999). Furthermore, EDTA produces an increase in root dentine permeability, which in turn, results in an increase in the activity of endodontic medicaments (Hampson & Atkinson 1964).

Tao *et al.* (1991) point out the importance of root cementum in dentine permeability. Instrumentation of the root canal in cases with intact root cementum did not lead to a change in permeability. Therefore, root canal instrumentation does not necessarily result in an increase in permeability in cases where external root resorption has not taken place. Dissolution of the smear layer by EDTA had no significant effect on permeability (Tao *et al.* 1991). The rate of diffusion fell initially after smear layer removal and increased significantly again after storing the extracted teeth for 2 months in deionized water (Galvan *et al.* 1994). The authors presumed that crystals formed during the dissolution of the smear layer by EDTA caused a precipitation of calcium phosphate crystals inside the dentinal tubules. After storage in water, these calcium phosphate crystals appeared to have dissolved, and the rate of diffusion increased again.

Removal of smear layer

A 1–5-μm thick smear layer is the result of the direct action of the endodontic instrument on the root canal wall (Mader *et al.* 1984, Goldman *et al.* 1985, Koçkapan 1995). McComb & Smith (1975) were the first to describe the smear layer on instrumented root canal walls. The depth of the dentinal tubule plug can be between 6 and 40 μm (Mader *et al.* 1984, Petschelt & Oberschachtsiek 1985). The smear layer consists of ground dentine and predentine, pulpal remnants, odontoblast processes, remnants of the irrigant and, in the case of infected teeth, bacteria (McComb & Smith 1975, Mader *et al.* 1984, Koçkapan 1995, Sen *et al.* 1995, Torabinejad *et al.* 2002; Fig. 2a–c). There is controversy as to whether the smear layer should be removed or not before obturation of the root canal. Microorganisms can remain in or migrate into dentine despite thorough chemomechanical preparation (Shovelton 1964, Brännström & Johnson 1974, Byström & Sundqvist 1981, 1983). Some authors propose that the smear layer acts as a barrier to bacterial metabolites, preventing the bacterial invasion of the dentinal tubules, and not as a preferred site for bacterial colonization (Vojinovic *et al.* 1973, Michelich *et al.* 1980, Diamond & Carrel 1984). However, bacteria not only remain, but also survive and multiply in the smear layer (Brännström & Nyborg 1973, Baker *et al.* 1975, Yamada *et al.* 1983, Brännström 1984) and can also penetrate into dentinal tubules (Olgart *et al.* 1974, Akpata & Blechman 1982, Williams & Goldman 1985, Meryon *et al.* 1986, Meryon & Brook 1990). The antimicrobial action of medicaments in the dentinal tubules can be delayed or hindered by the smear layer (Goldberg & Abramovich 1977, Brännström & Nordervall 1978, Wayman *et al.* 1979, Yamada *et al.* 1983, Brännström 1984, Baumgartner & Mader 1987, Ørstavik & Haapasalo 1990, Drake *et al.* 1994). This would appear to make smear layer removal advisable. Moreover, the ability of the sealer to penetrate the dentinal tubules and thereby the adaptation of the root canal filling to the root canal wall is much improved after removal of the smear layer (Diamond & Carrel 1984, White *et al.* 1984, 1987, Wennberg & Ørstavik 1990, Oksan *et al.* 1993). Some investigators found that root filling impermeability is significantly higher after smear layer removal (Kennedy *et al.* 1986, Cergneux *et al.* 1987, Petschelt *et al.* 1987), whilst others showed no differences (Madison & Krell 1984, Goldberg *et al.* 1985, Evans & Simon 1986).

Electron microscopy has shown that the smear layer contains both organic and inorganic substances (Yamada *et al.* 1983, Pashley 1984, Koçkapan 1987). It appears, however, to consist mostly of inorganic components as root canal irrigation with NaOCl has little effect on removal of this layer. Partial if not complete smear layer removal is achieved only with the aid of acids and chelators (Yamada *et al.* 1983, Koçkapan 1987). As the components of the smear layer are small particles with a large surface/mass ratio, they are highly soluble in

acids (Pashley 1992). Numerous studies have reported that irrigation with a 17% EDTA solution has a good cleaning effect on the root canal walls (McComb & Smith 1975, Goldberg & Abramovich 1977, Goldman *et al.* 1985, Berg *et al.* 1986, Baumgartner & Mader 1987, Cergneux *et al.* 1987, Meryon *et al.* 1987, Ciucchi *et al.* 1989, Aktener & Bilkay 1993, Garberoglio & Becce 1994, Hottel *et al.* 1999, Çalt & Serper 2000, Di Lenarda *et al.* 2000, O'Connell *et al.* 2000, Scelza *et al.* 2000). Following smear layer removal, the root canal walls are clean and the dentinal tubules are clearly recognizable (McComb & Smith 1975, Goldberg & Abramovich 1977, Ram 1977, Pawlicka *et al.* 1981, Goldman *et al.* 1985, Cergneux *et al.* 1987, Aktener & Bilkay 1993, Liolios *et al.* 1997, Çalt & Serper 2000; Fig. 3a–b). The tubule orifices are enlarged because of dissolution of peritubular dentine (Goldberg & Abramovich 1977, Cergneux *et al.* 1987, Meryon *et al.* 1987, Hottel *et al.* 1999, Çalt & Serper 2000; Fig. 4). Some studies even detected erosion of the dentinal tubules (Çalt & Serper 2002, Niu *et al.* 2002, Torabinejad *et al.*

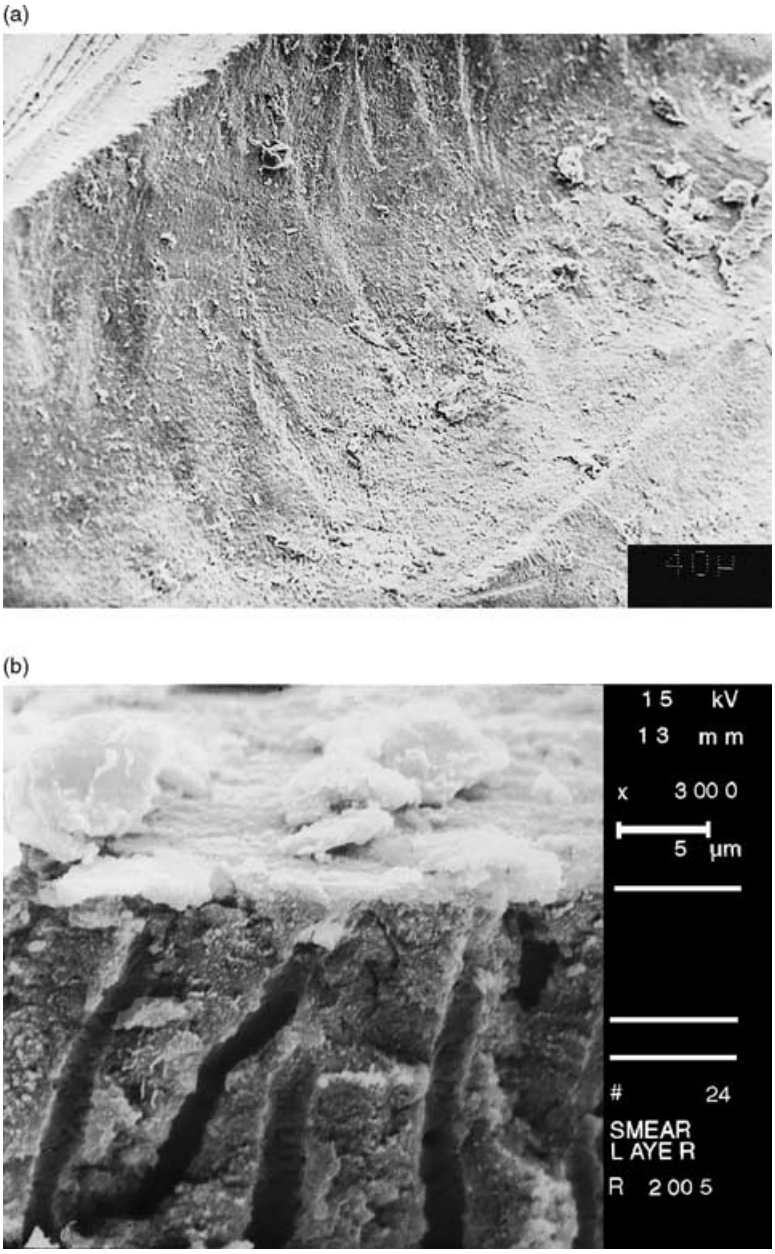

Figure 2 (a) Following instrumentation, the root canal wall is covered by debris and smear layer (magnification 250×). (b) Following instrumentation of the root canal, the dentine surface is covered with smear layer (magnification 3000×). (c) The smear layer is pressed into the dentinal tubules (magnification 8500×).

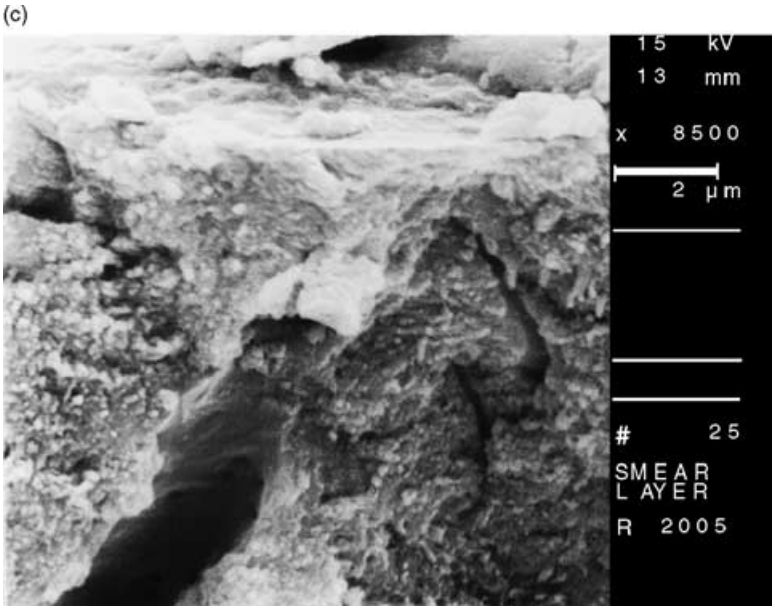

Figure 2 continued

2002; 2003a,b; Fig. 5). Whilst Pawlicka *et al.* (1981) reported that the root canal wall is clean along its entire length after use of EDTA preparations, other authors found that the cleaning action is reduced towards the apex and therefore more efficient in the coronal- and middle-third of the root (Baker *et al.* 1975, McComb & Smith 1975, Chow 1983, Ciucchi *et al.* 1989, Abbott *et al.* 1991, Aktener & Bilkay 1993, O'Connell *et al.* 2000, Scelza *et al.* 2000, Hülsmann & Heckendorff 2002, Lim *et al.* 2003). The number of visible tubule openings reduces from coronal to apical (Scelza *et al.* 2000). In a comparative study, a carbon dioxide or Er:YAG laser removed the smear layer more efficiently than EDTA (Takeda *et al.* 1999). A new irrigant, composed of tetracycline isomer, an acid and a detergent (MTAD) showed similar results but less erosion than EDTA (Torabinejad *et al.* 2003a,b).

Combined use of EDTA, sodium hypochlorite and ultrasonics

Because EDTA acts by dissolving the inorganic components of the smear layer, several authors have recommended its use in combination with NaOCl (0.5–5.25%) in order to remove organic remnants (Goldman *et al.* 1982, Yamada *et al.* 1983, Baumgartner & Mader 1987, Çengiz *et al.* 1990, Abbott *et al.* 1991, Stewart 1998, Tatsuta *et al.* 1999, Brandt *et al.* 2001). Both the cleaning action (Baumgartner & Mader 1987, Stewart 1998, Lim *et al.* 2003, Yamashita *et al.* 2003) and the antimicrobial effect (Byström & Sundqvist 1985) are greater when these irrigants are used in combination rather than alone. It has been shown that EDTA retained its ability to chelate calcium in the presence of NaOCl, whereas the tissue-dissolving ability of NaOCl was reduced (Grawehr *et al.* 2003). The content of available chlorine was drastically reduced from 0.50 to 0.06% when EDTA was added to a NaOCl solution; nevertheless, the antibacterial efficacy against *Candida albicans* and *Enterococcus faecalis* was the same for 8.5% EDTA and a 17% EDTA/1% NaOCl solution (Grawehr *et al.* 2003). The authors concluded that both solutions therefore should be used separately. Ultrasonically supported irrigation with EDTA does not improve the cleaning effect of EDTA (Ciucchi *et al.* 1989, Abbott *et al.* 1991). Possibly, ultrasound waves produced by the vibrating instrument reduce the demineralizing effect of the chelator by reducing the working time, as EDTA only develops its full effectiveness after a certain working time (Abbott *et al.* 1991). In contrast, a superior root canal cleanliness is achieved following ultrasonically agitated irrigation with NaOCl (1–4%) and EDTAC when compared to ultrasonically agitated irrigation with distilled water or NaOCl alone (Cameron 1995, Guerisoli *et al.* 2002).

Pawlicka *et al.* (1981) compared the cleaning effect of different chelator preparations and found that the efficiency of EDTA and DTPA could be further improved by the addition of a detergent (EDTAC and DTPAC). The observation that Salvizol can dissolve both organic and inorganic components of the smear layer and has a

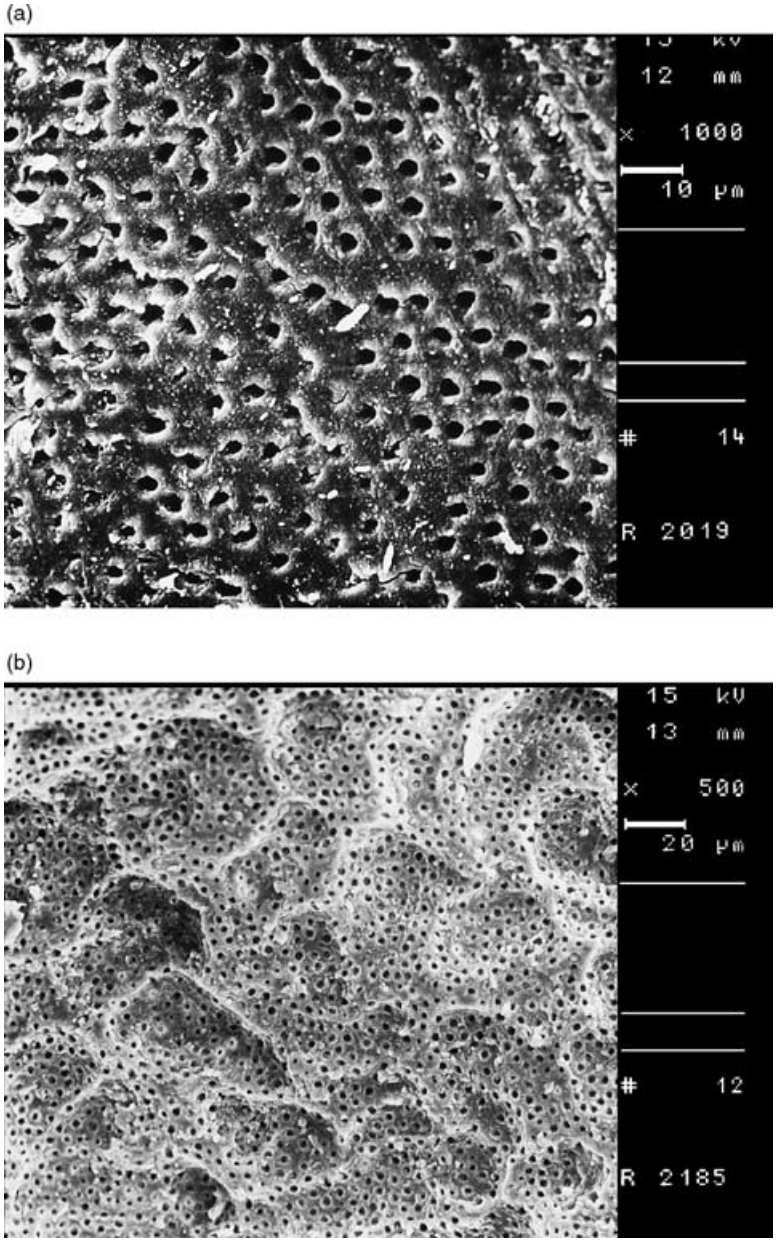

Figure 3 (a) root canal wall after removal of smear layer using RC-Prep (magnification 1000×). (b) root canal wall after removal of smear layer using Calcinase slide (magnification 500×).

superior cleaning action than EDTAC (Kaufman *et al.* 1978, Spångberg *et al.* 1978), however, could not be confirmed (Velvart 1987). Neither Salvizol, Largal Ultra nor Decal were able to dissolve both organic and inorganic substances (Koskinen *et al.* 1980).

Tubulicid Plus and Largal Ultra both removed the smear layer resulting in a surface with open dentinal tubules (Liolios *et al.* 1997). A comparison of EDTA, RC-Prep and Salvizol showed that EDTA was the most effective solution to remove the smear layer (Ram 1977). A neutral solution of EDTA had a better cleaning action than RC-Prep and allowed visualization of the dentinal tubules (Verdelis *et al.* 1999). EGTA has been recommended as an alternative to EDTA for dissolution of the smear layer because it does not cause erosion of the dentinal tubules unlike EDTA (Çalt & Serper 2000).

The cleaning effect of Calcinase Slide, RC-Prep and Glyde File Prep after 5 min application in alternating rinses with H_2O_2 and NaOCl were compared in a recent study (Hülsmann & Heckendorff 2002). Calcinase Slide cleaned significantly better than the other preparations in the coronal- and middle-third of the canal. No differ-

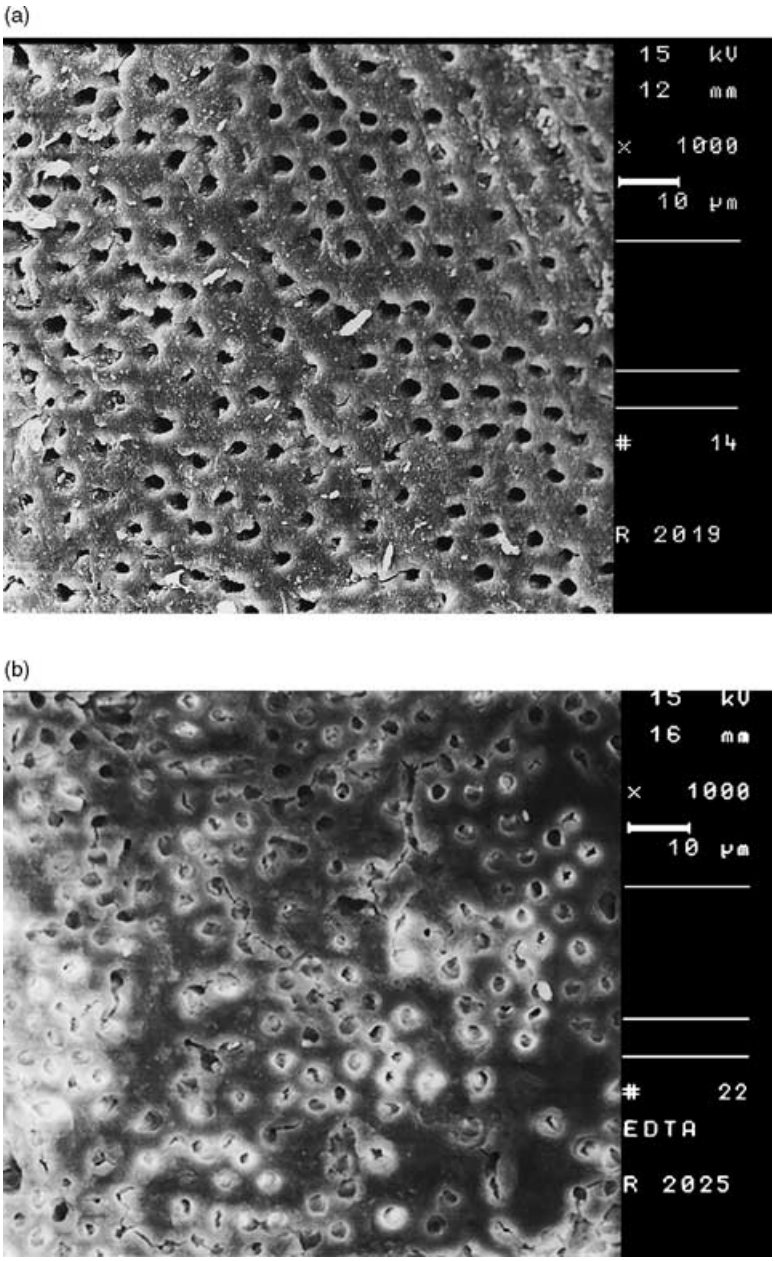

Figure 4 (a) Enlarged tubule openings following the use of a paste-type chelating agent (magnification 1000×). (b) Some tubule openings are surrounded by a decalcified zone of peritubular dentine (magnification 1000×).

ence could be found between the preparations in the apical-third of the canal. A reduction in root canal-wall cleanliness towards the apex was also observed.

Irrigation with 17% EDTA resulted in better root canal cleanliness than the use of Glyde File during rotary preparation with Lightspeed NiTi instruments (Ahn & Yu 2000). The use of Glyde File or irrigation with 17% EDTA removed the smear layer more effectively than NaOCl (Lim *et al.* 2003). Comparing root canal preparation with rotary ProFile NiTi instruments, root canal cleanliness proved to be superior following preparation with the paste-type chelator Glyde File Prep and 2.5% NaOCl as an irrigant when compared to preparation with NaOCl or physiological solution alone (Grandini *et al.* 2002). Nevertheless, complete removal of the smear layer could not be achieved, which is confirmed by several studies on NiTi preparation in combination with a paste-type chelator and NaOCl (Peters *et al.* 1997, Peters & Barbakow 2000, Hülsmann *et al.* 2001, Schäfer & Lohmann 2002, Versümer *et al.* 2002).

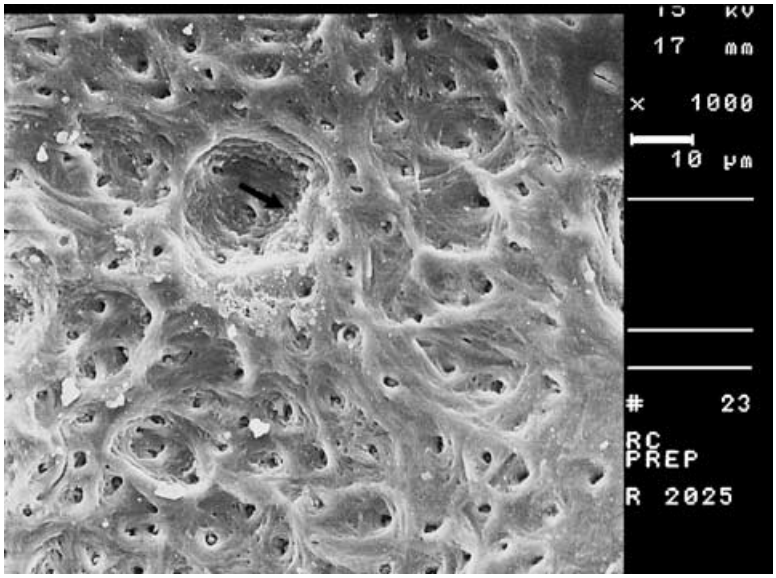

Figure 5 Following the use of a chelator paste for 5 min, some dentinal tubules showed erosion of peritubular dentine (arrow; magnification 1000×).

Comparing different sequences of irrigation, the sequence EDTAC > NaOCl > EDTAC proved to be more efficient in smear layer removal than the sequence NaOCl > EDTAC > NaOCl (Abbott *et al.* 1991). The additional use of ultrasound did not enhance cleaning ability of these irrigants.

Working time of chelators

Still, the optimal working time of chelating agents is unknown. A certain cleaning effect is achieved after chelator application for a few minutes. According to Goldberg & Spielberg (1982), the optimal cleaning effect is only achieved after 15 min. In contrast, McComb & Smith (1975) were able to show a better effect when the chelator preparation was left in the root canal for 14 h. In a study using autoradiographic tracings of ^{45}Ca-labelled EDTA, no significant difference in penetration depth could be found after 15-min and 24-h working time (Nicholson *et al.* 1968). Additionally, quantity of smear layer removal is related both to the pH and to the length of time for which the chelating agent has been exposed (Morgan & Baumgartner 1997). Several studies confirmed that mineral loss, changes in dentine hardness and cleanliness of the root canal walls depend on the working time (Nygaard-Østby 1957, Hülsmann & Heckendorff 2002, Serper & Çalt 2002).

Several studies have reported a good cleaning efficacy of liqiud or paste-type EDTA after working times between 1 and 5 min (Yamada *et al.* 1983, Cergneux *et al.* 1987, Çalt & Serper 2000; 2002, Hülsmann & Heckendorff 2002, Scelza *et al.* 2003). In a recent study, 1-min exposure to 10 mL EDTA was sufficient to remove the smear layer, whereas an exposure for 10 min caused excessive peritubular and intratubular erosion (Çalt & Serper 2002). This kind of erosion has been proposed because of the result of the combined use of EDTA and NaOCl rather than EDTA alone (Niu *et al.* 2002). Nevertheless, at present, no definite recommendation can be given on the optimal amount and working time for a paste or liquid chelator under clinical conditions.

Biocompatibility of chelating agents

There is much discussion as to whether and what degree of inflammatory tissue reaction can be caused by chelating agents passing through the apical foramen. Nygaard-Østby (1957) investigated the effect of a 15% EDTA solution (pH 7.3) on human periapical tissue as well as on pulpal tissue under clinical conditions in cases with vital and necrotic pulps. No periapical tissue damage could be detected after a period of action of up to 14 months, even though EDTA was intentionally forced through the apical constriction using a file. The histological examination revealed normally regenerated alveolar bone and new functional periodontal ligament fibres. In addition, clinical studies showed that placement of EDTA for up to 28 days after pulpotomy failed to produce any pulpal tissue necrosis.

In an investigation of the tissue reaction in rats after intramuscular implantation and injection of EDTA and EDTAC (15%), the latter caused much greater tissue irritation after implantation and after injection than

10% EDTA (Patterson 1963). No periapical tissue irritation or damage of any kind occurred in 200 clinical cases where EDTA was used as an irrigant. Acute exacerbation did not seem to occur more frequently than with other irrigants. Further studies have indicated that EDTA is not capable of destroying collagen (Lindemann *et al.* 1985). When the dentine is intact, the effect on the pulp seems to be negligible (Lindemann *et al.* 1985) so that EDTA can also be recommended as a conditioning agent prior to application of dentine bonding (Cao *et al.* 1992).

In contrast to previous findings, Collet *et al.* (1981) concluded that a 15% sodium (Na)-EDTA solution has toxic effects *in vitro.* Complete prevention of cell growth was detected after *in vitro* use of EDTA-T (Scelza *et al.* 2001). Additionally, 15% solutions of EDTA and EDTAC at pH 7.3 have the potential to cause severe irritation (Koulaouzidou *et al.* 1999). These authors found that 15 and 17% EDTA solutions and 2.25% NaOCl solutions produce severe cytotoxic effects, whilst 1% solutions of both agents evoked only moderate reactions.

Extrusion of even a low concentration of EDTA solution through the apical constriction results not only in an irreversible decalcification of periapical bone, but can also have consequences for neuroimmunological regulatory mechanisms (Segura *et al.* 1996). These authors investigated the effect of EDTA and EGTA on the binding of vasoactive intestinal peptides (VIP) to macrophages. VIPs act not only as vasoactive substances, but also play an important role as neuropeptides in the communication between nerves and immune cells in the pulp and periapical tissue by modifying the macrophage function. EDTA inhibits VIP binding to macrophages even in lower concentrations than those used in endodontics (10%). EDTA can prevent the adhesion of macrophages to substrate; this is time- and concentration-dependent (Segura *et al.* 1997). EDTA concentrations measurable in the periapical tissues are capable of reducing binding by 50%. The degree to which VIP and substrate control of macrophage function effects the healing process is not clear. On one hand, changes in macrophage activity can cause the inflammatory reaction to be more easily initiated; on the other hand, reduced capacity of phagocytosis can result. In addition, it has been found that EDTA improves plasma extravasation and mediator action (Segura *et al.* 1997).

In an investigation of the effects of dental etchants and chelators on nerve compound action potentials (Çehreli *et al.* 2002), RC-Prep and File-EZE were shown to reduce the compound action potentials after an application time of 160 min by 61.8 and 62.4%, respectively.

Comparing the cytotoxic effects of the three irrigants, EDTA provoked more cytotoxic effects than oxidative potential water or NaOCl (Serper *et al.* 2001). Salvizol was found to be less cytotoxic than EDTAC (Spångberg *et al.* 1978). In the light of these results, extrusion of EDTA into the periapical tissue during chemomechanical root canal preparation should be avoided.

Antibacterial effects of EDTA

Ethylenediaminetetraacetic acid has a certain, albeit limited, antibacterial effect (Patterson 1963). This is thought to be because of the chelation of cations from the outer membrane of bacteria. The use of a 10% EDTA solution results in the formation of a zone of inhibition of bacterial growth similar to creosote. Lower concentrations of EDTA solution (0.03–1%) produce a reduced effect or no effect at all (Russell 1999). The antibacterial properties of EDTA depend on concentration and pH (Kotula & Bordacova 1969). The antibacterial effect of Na-EDTA is only maintained as long as the chelators have not formed bonds with metal ions (Hendershot *et al.* 1960, Kotula & Bordacova 1969).

Bacteriological investigations (Pawlicka & Nowacka 1982) have shown that whilst chelators have an antibacterial effect, it is much less than that of paramonochlorophenol. No differences could be shown between the effect of EDTA and DTPA, even after the addition of a detergent (EDTAC and DTPAC).

In a clinical study, Yoshida *et al.* (1995) examined the effectiveness of 15% EDTA solution used in combination with ultrasound. No intracanal dressing was placed between treatment sessions. After 1 week, no bacteria could be found in the root canal in 93 of 129 cases. EDTA proved to have a greater antimicrobial effect than saline. The authors concluded that a negative bacteria culture could only be expected after removal of the smear layer using EDTA. Furthermore, the combined use of EDTA and 5% NaOCl has a greater antimicrobial effect than NaOCl alone (Byström & Sundqvist 1985).

REDTA can only inhibit the growth of some anaerobes after 60 min to a week. *Porphyromonas gingivalis* is the exception, because an effect is seen with a 10% solution after only 1 min (Ohara *et al.* 1993). Salvizol is thought to have fungicidal and broad-spectrum antibiotic activity (Nawrath *et al.* 1960), with lower tissue toxicity than EDTAC (Spångberg *et al.* 1978). Further studies show that RC-Prep has a limited antibacterial effect, depending on the microorganisms used (Heling & Chandler 1998, Buck *et al.* 1999, Steinberg *et al.* 1999). RC-Prep is more

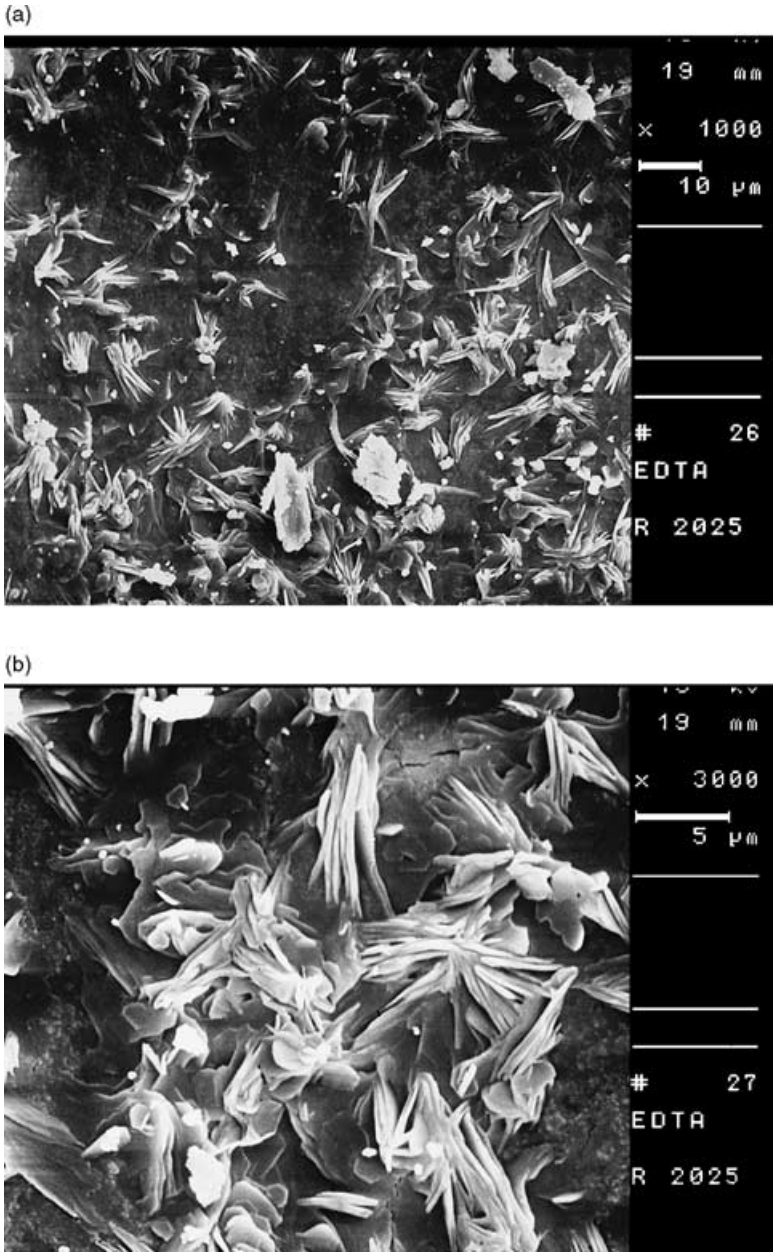

Figure 6 Crystals on the root canal wall following the use of a paste-type chelator and irrigation with NaOCl. These crystals could be observed only in a limited number of specimens (magnification 1000×).

effective against Gram-negative than Gram-positive aerobes (Heling & Chandler 1998).

Increasing the incubation period from 10 to 45 min increases the antimicrobial effect of RC-Prep on *Staphylococcus aureus* and *Streptococcus peltzer* in dentinal tubules (Heling *et al.* 1999). The urea peroxide (10%) content of RC-Prep is thought to be mainly responsible for its antibacterial effect. Urea peroxide is an oxidizing antibacterial agent (Block 1991), which retains its effectiveness in the presence of blood (Stewart *et al.* 1961). Investigations of more than 100 patients after irrigation with EDTA, urea peroxide and NaOCl resulted in a negative bacterial culture in 97.2% of cases at the end of the first appointment. At the beginning of the second appointment, 94.4% of the teeth showed no bacterial growth, although no canal dressing had been used (Stewart *et al.* 1969).

Steinberg *et al.* (1999) investigated the bacteriostatic and bactericidal effect of the individual components of

RC-Prep on *S. sobrinus*. The minimum concentration to yield an inhibitory effect was 0.125% for EDTA, 0.25% for urea peroxide and 30% for glycol, whilst the minimum concentration for a bactericidal effect was 0.25% for EDTA, 0.5% for urea peroxide and 50% for glycol. In contrast to this, Ørstavik & Haapasalo (1990) questioned the antibacterial effect and the tubular disinfection produced by a 17% EDTA solution. Whilst the number of *S. sanguis* at a depth of 100–300 μm in the dentinal tubules could be reduced after 5 min incubation with NaOCl, no disinfection could be achieved using EDTA.

Following irrigation with NaOCl (3.5%) and 0.5 M EDTA, Yang & Bae (2002) found significantly less bacteria (*Prevotella nigrescens*) adhering to the root dentine than in root canals where the smear layer had not been removed or removed after irrigation with only NaOCl (3.5%).

Effect of EDTA on the quality of root canal obturation

An increased number of obturated accessory canals is found after final irrigation with NaOCl (6%) alone or in combination with EDTA than after no irrigation or irrigation with distilled water (Villegas *et al.* 2002). The root canals in that study were obturated using the System B (Analytic Technology, San Diego, CA, USA) and Obtura II (Texceed Corp., Fenton, MO, USA).

The dentine adhesion of endodontic sealers can be improved by dentine pretreatment with EDTAC, although this effect is more pronounced after ER:YAG laser pretreatment (Pecora *et al.* 2001, Picoli *et al.* 2003). The highest increase in adhesiveness was found for Sealer 26 (Dentsply, Petropolis, Brazil); for calcium hydroxide-containing sealers such as Sealapex (Kerr, Romulus, Mich, USA), Apexit (Vivadent, Schaan, Liechtenstein) and CRCS (Hygienic, Mahwah, NJ, USA), only a slight increase was found (Picoli *et al.* 2003). On the other hand, both NaOCl and RC-Prep significantly reduced the bond strength of resin cement to root dentine (Morris *et al.* 2001). This reduction can be completely reversed by application of 10% ascorbic acid or 10% sodium ascorbate. Dental adhesives bound significantly better to calcified dentine than to decalcified dentine pretreated with EDTA (Perdigao *et al.* 2001).

Although some studies suggest that removal of the smear layer reduces apical leakage after obturation (Kennedy *et al.* 1986, Cergneux *et al.* 1987, Petschelt *et al.* 1987), the treatment with EDTA may leave a chelated layer of dentine at the dentine–root filling interface. Residual EDTA inside the dentinal tubules, which was measured to be up to 3.8% of the originally applied volume (Zurbriggen *et al.* 1975), may contribute additionally to ongoing demineralization, resulting in further increase of apical leakage (Cooke *et al.* 1976, Biesterfeld & Taintor 1980). Residual EDTA also may interact with the sealer, which has been demonstrated for zinc oxide–eugenol-containing sealers (Biesterfeld & Taintor 1980). However, Madison & Krell (1984), who compared NaOCl, REDTA and a combination of both, could not detect any influence of the irrigation solution on the apical seal.

Bleaching effect

In some commercially available EDTA preparations, urea peroxide is added to the EDTA. The release of oxygen results in some effervescence that is not only expected to enhance the cleaning efficiency, but also is claimed to have a bleaching effect. No scientific evidence exists to support the bleaching effect of such chelators. Regarding the working time of a chelator under clinical conditions, no bleaching should be expected as a visible effect of internal bleaching procedures using urea peroxide can be observed only after 48 h (Matis *et al.* 1998, Attin *et al.*, 2003).

Additional findings

1 In some studies, it has been mentioned that crystals could be observed on the dentine surface following the use of chelators (Schmidt 1968, Koçkapan 1987, Behrens & Sierra 1992, Liolios *et al.* 1997, Hülsmann & Heckendorff 2002; Fig. 6a,b). Electron dispersive spectrometer analysis revealed that these crystals contained mainly calcium and phosphorus (Schwarze & Geurtsen 1995, Liolios *et al.* 1997). Storage in formaldehyde-containing liquids has been proposed as one possible reason for the presence and growth of such crystals (Schwarze & Geurtsen 1995).

2 In an *in vitro* study, irrigation with EDTA did not affect the accuracy of electronic length determination using the Root ZX (Morita, Tokyo, Japan; Jenkins *et al.* 2001).

3 The use of EDTA during root canal preparation caused less corrosion to stainless steel files than NaOCl (5.25%; Mueller 1982, Öztan *et al.* 2002). However, other studies could not detect any corroding effect of EDTA on steel instruments (Aten 1993). In contrast, the cutting efficacy of steel files is reduced significantly by EDTA as a result of slight corrosion but less than following exposure to NaOCl (Neal *et al.* 1983).

4 The release of mercury from amalgam fillings was lower when EDTA was used as an irrigant in combina-

tion with NaOCl than with NaOCl alone (Rotstein *et al.* 2001).

5 When curved root canals were instrumented with NiTi instruments to size 30, maintenance of the original curvature was better in the group in which no EDTA was used (Bramante & Betti 2000). Nevertheless, the number of teeth per group in this study was too small to draw definite conclusions.

6 It has been hypothesized that the use of EDTA would substantially increase the retention of posts, but no significant effect has been found (Burns *et al.* 1993). Although smear layer removal would be beneficial for increased retention, the decalcification of the dentine could actually reduce retention. In contrast, a significantly increased retention was observed after irrigation with 17% EDTA and 5.25% NaOCl (Goldman *et al.* 1984a,b).

Conclusions and clinical recommendations

The critical examination of the extensive literature on the effectiveness of chelator preparations presented here indicates that the use of chelators is recommended during root canal preparation. Chelator preparations can reduce the extent of the smear layer produced during preparation. The effectiveness of these preparations depends more on the length of application time than the specific product chosen and clearly decreases from the canal orifice towards the apex. With careful use, the risk of damage to the periapical tissues is low. On the other hand, the antibacterial effect of chelator preparations is low, and fluid chelators should not replace NaOCl as a standard irrigant, although they may improve the ability of NaOCl to penetrate into the dentine and increase its antibacterial effect. The degree to which chelators actually facilitate negotiation and preparation of calcified and narrow root canals is unknown. As the effectiveness of chelating agents is dependent not only on the concentration and working time but also on the relationship between the amount of available chelator solution and the canal-wall surface area, skepticism is indicated.

The following clinical recommendations are made based on the previous discussion:

1 root canal preparation can be carried out with the aid of a chelator paste. This may be introduced into the root canal with the preparation instrument. The canals should first have been flooded with NaOCl to dissolve vital or necrotic tissue.

2 A chelator in paste form serves as a lubricant for files and may reduce the risk of instrument fracture in the canal, although there is no experimental evidence for this claim. In relation to this, the NiTi instrument manufacturers' recommendations should be followed, even though there are no clinical or experimental studies available on this matter.

3 Preferably, a NaOCl solution should be used during preparation because of its superior antibacterial and tissue-dissolving properties.

4 A final intensive rinse with a 17% chelator solution reduces the extent of the smear layer remaining, which, in turn, results in a cleaner canal wall and better adaptation of the root filling to the canal wall. The order in which the NaOCl and the chelator should be used has not yet been defined.

5 EDTA-containing agents should be used between 1 and 5 min.

6 A liquid EDTA solution may be introduced into the pulp chamber (pipette, cotton pellet) to identify the entrance to calcified canals.

7 The differences in certain properties and modes of action of individual chelators found in the few comparative studies do not allow the recommendation of any particular chelator preparation.

8 EDTA pretreatment may reduce bond strength of adhesive materials and obturation materials.

9 There is no evidence for a bleaching effect when using EDTA preparations containing urea peroxide.

10 Apical extrusion of chelators should be avoided.

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