Periapical disease is the result of bacteria, their product, and the host response to them. Early histological studies of diseased periapical tissue have not been able to demonstrate viable bacteria in the lesions studied. Recent reports indicate that many of periapical lesions are indeed infected before and after endodontic treatment. The validity and applicability of the microbial delivery overcome many disadvantages that we see with systemic drugs. In this case report we presented a novel approach of managing chronic diffuse periapical lesion of lower molar based on specific selection of intracanal medicament in combination with direct periapical injection. We used bacterial culturing and antibiotic sensitivity test to select specific intracanal medicament, in addition we presented an intraosseous injection technique to locally deliver the selected medicament directly into the periapical lesion. Our findings are encouraging and promising. The validity and applicability of the technique needs to be tested in a well controlled clinical trial.

KEY WORDS: periapical disease, root canal therapy, microbiology, intraosseous injection, local antimicrobial delivery, clindamycin.
INTERROGATION

Periapical disease is the result of bacteria, their product, and the host response to them. Early histological studies of diseased periapical tissue have not been able to demonstrate viable bacteria in the lesions studied (1). More recent reports indicate that many of these lesions are indeed infected before and after endodontic treatment. In 1992 Wayman studied 58 cases of periapical lesion (2). He cut these lesions in half and examined one half histological and cultured the other half. In only 8 of 58 cases could he demonstrate bacteria histologically. However, when the other half lesion was cultured 51 of 58 cases were positive. He found 133 isolates, of which 87 were strict anaerobes, 37 were facultative anaerobes and only 9 were aerobes. Iwu showed that 88% or 1 of 16 periapical granulomas were positive for bacteria when they were cultured. Barkhodar and Desouza (3) also found bacteria in granulomas and cysts. For many clinicians calcium hydroxide is the most commonly used intracanal medicament, although the effectiveness of many other intracanal antimicrobials, such as metronidazole antibacterial gel (4), ledermix with tetracycline (5), clindamycin-impregnated fiber (6), antibiotic mixtures of ciprofloxacin, metronidazole, minocycline (7), has been proven effective. Calcium hydroxide as an intracanal medicament has many advantages (8,9), but latest evidence demonstrated that enterococci and fungi, commonly found in cases of endodontic failure are highly resistant to calcium hydroxide (10).

AIM

The primary aim of this case report is to evaluate the effectiveness of using microbial culturing and antibiotic sensitivity testing to select specific antimicrobial medications to manage endodontic infections. A secondary aim is to evaluate the effectiveness of the selected antibiotic as an intracanal medicament in combination with intraosseous injection directly into the periapical lesion was infected as a result of microleakage and consequently dental decay and bacterial flora was transferred from root canal towards periapical tissue.

SPECIMEN COLLECTION AND MICROBIOLOGICAL PROCEDURE

After clinical and X-ray diagnose of asymptomatic left mandibular second molar with diffuse periapical lesion we did access preparation by sterile burs, restoration and complete removal of soft, carious dentin using sterile burs without water spray. ISO 15-25 K-files used to scale off dentin debris from the root canal walls. Specimen collected by K-files is directly inoculated in liquid thioglycolate broth with paraffin oil, for further primary and secondary microbiological procedure (Figure 1.). These procedures included; direct microscopy examination of the specimen, showing size, shape, morphotype, and Gram staining of the present bacteria. During aerobic procedure specimen from thioglycolate broth is inoculated in the blood agar, McConcey and Sabourad (Figure 2.). For anaerobic procedure another agar plate is necessary with 4% hemin and placed discs. Anaerobic plates were in pot with Gas-pac system, closed for 48-72 hours at 37°C in incubator (Figure 3.). Findings of bacteria around discs and far from discs determined the result of antibiotic susceptibility test. All isolated Streptococcus spp. during microbiological procedure were sensitive to clindamycin.

CONVENTIONAL ENDOdontIC TREATMENT

After completing mechanical instrumentation of two root canals of mandibular second molar we used oxidizing agent, 3% hydrogen peroxide as root canal irrigant. According to the antimicrobial susceptibility test obtained, as an intracanal medicament a sterile pharmaceutical formulation (with the proper rheological properties) for the root canal treatment was prepared and used. Antimicrobial agent clindamycin phosphate, as an intracanal medicament, was used in concentration of 3%, incorporated in the formulation containing benzyl alcohol, sodium hydroxide, hydroxyethylcellulose, propylene glycol and water for injection. The temporary filling was intact, without leaking, or contamination, sealed hermetically, without exudate, or foul odor from the root canals. Next step was complete obturation of the root canals using cement sealer and gutta-percha cone. All procedures were done with respect to ethical standards regulated by Helsinki Declaration.
With assigned form of patient consent we proceeded with the treatment and local antimicrobial drug delivery.

**The use of intraosseous injection**

In our next appointment patient received regional anesthesia (Figure 4). The point of intraosseous penetration was attached gingiva, area between the two roots of the second lower molar. We used special equipment, the perforator attached to a slow-speed hand piece was advanced through anesthetized gingiva and bone until penetration through the cancellous bone is experienced (Figure 5) (11, 12). The perforator was removed, the short 8 mm needle was inserted through the perforation into the cancellous space and 1.8 ml of clindamycin phosphate solution was delivered slowly (Figure 6).

**CLINICAL OUTCOME**

We monitored response to treatment on sequential periapical radiographs using a portable transmission densitometer (Portable densitometer X-Rite 331, X-Rite Ltd., United Kingdom). We took periapical images at the beginning of the endodontic treatment, during obturation of the root canals two- and six months later. A change in periapical lesion was detected by X-Rite 331 densitometer showing improvement in periapical healing process. Measured optical density units at the beginning of the endodontic treatment and two months later showed difference of 0.16 ODU, suggesting improvement in healing process of the affected tissue. Figures 7, 8 and 9 present periapical images obtained at the beginning of the endodontic treatment, after completed obturation and six months later.

**DISCUSSION**

In this case report we used bacterial culturing and antibiotic sensitivity to select specific intracanal medicament, in addition we presented an intraosseous injection technique to deliver the selected medicament directly into the periapical lesion. The combined use of the selected antibiotic as intracanal medicament as well as direct injection into the periapical area enhanced the healing and resolution of the lesion as demonstrated on the sequential periapical radiographs. The intervention is completely painless since we had to apply conventional mandibular block, before we used supplemental intraosseus injection technique for local application of clindamycin phosphate. Clindamycin is derived from Strepto-
myces lincolnensis (13). The mechanism of action, like the macrolide class, is inhibition of protein synthesis by binding to the 50S ribosomal subunit. It has a wide distribution, but does not cross the blood-brain barrier. The drug is concentrated in the bone, respiratory tissues, mucus, and saliva. At lower concentrations the drug is bacteriostatic, but bacteriocidal at higher concentration. Resistance is mediated by decreased permeability of the drug into the cell and alteration of binding sites on the 50S ribosome subunit. The spectrum of action is primarily against gram-positive and gram-negative bacteria and anaerobes (especially B. fragilis). Clindamycin
phosphate has been shown to be effective, among others in the treatment of periodontal abscess, periodontitis, and bone infections when caused by susceptible anaerobic bacteria or susceptible strains of gram-positive bacteria such as Streptococci, Staphylococci and Pneumococci. It possesses both a bacteriostatic and a bactericidal action. Clindamycin-affected microbial cultures show biphasic steady-state generation curves. An initial (phase I) generation of the clindamycin-affected microbes is followed by an ultimate (phase II) generation at the same dose level. The dependence of the apparent generation rate constant on drug concentration yields a sigmoidal curve, which is coincident by a potency factor for the phase I and phase II generations of clindamycin-affected microbes and suggests a common mechanism of action for both generation phases.

CONCLUSIONS

In previously reported cases of conventional endodontic treatment of lower molars with chronic diffuse periapical pathology the outcome of treatment has been slow and questionable. In this case report we presented a novel approach of managing these lesions based on specific selection of intracanal medicament in combination with direct periapical injection. Our findings are encouraging and promising. The validity and applicability of the technique needs to be tested in a well controlled clinical trial.

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