

Controlled Release of Drugs for Management of Pulpitis

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Context: Biocompatible polymers are potentially effective for dental infections as delivery carriers of disinfectants or antibiotics into the root canal system (RCS). This study aimed to review polymeric microspheres enabling a controlled release of endodontic medicaments.

Evidence Acquisition: A literature search was carried out in the PubMed database (May 2013) using the following keywords: "poly lactic-co-glycolic acid or PLGA", "polymer microplate", "encapsulate", "drug delivery", "controlled release", "antibiotic", "gentamycin", and "amoxicillin". We intended to find articles on the application of polymer microparticles for delivery and release of drugs in dental infections or articles discussing factors affecting the properties of these materials.

Results: Seventeen articles were found evaluating the controlled release of the drugs for dental purposes; out of them, in 5 in vitro studies, polymer microspheres had been produced for root canal disinfection. Seven articles had investigated the properties of polymer microspheres and the factors influencing drug release by them.

Conclusions: Drug-loaded polymer microspheres may be used successfully as delivery carriers for controlled release of antibiotics into the root canal system. The efficacy and success rate of this method must be tested in animal models and then clinical trials.

Keywords: Poly Lactic acid -co- Glycolic acid; Microspheres; Delayed-Action Preparations; Anti-Bacterial Agents; Drug Delivery Systems; Root Canal Therapy; *Enterococcus faecalis*

1. Context

The main goal of root canal therapy (RCT) is to eliminate the microorganisms from the RCS by cleaning, and shaping the root canals accompanied by irrigation with antimicrobial agents. Endodontic treatment can be divided into three main phases: 1. Mechanical debridement to remove debris and eliminate bacteria from the RCS; 2. Administration of treatments aiming to target the metabolism of pathogenic microorganisms; and 3. Administration of treatments affecting the intracanal environment surrounding the microorganisms (1, 2).

Enterococcus faecalis (*E. faecalis*) is an anaerobic Gram-positive bacterium and part of normal flora of the mouth. It is found in small amounts in unprepared root canal systems. Its role in failure of endodontic treatments has yet to be fully understood. However, it is the most commonly isolated bacterium from the endodontically treated root canals with chronic apical periodontitis (3). *Enterococcus faecalis* has been introduced as a resistant pathogen to endodontic treatment due to its ability to invade dentinal tubules, resistance against different ecological conditions in the root canal system, and adaptation to unfavorable intracanal conditions (4).

Several studies have investigated the efficacy of amoxicillin (AMX), vancomycin, erythromycin, benzyl peni-

cillin and doxycycline against *E. faecalis* strains (5-7). Antibiotic therapy (both local and systemic) is now a well-accepted treatment modality in medicine and dentistry. In the local antibiotic therapy, side effects of the systemic administration of antibiotics are prevented and higher antibiotic concentrations can be achieved at the target site (8). Delivery carrier systems loaded with disinfectants or antibiotics can be used for the treatment of the infections or as a pharmaceutical adjunct to mechanical treatment for delivery of medications to the target site.

A new approach includes using local drug-delivery systems by applying microparticles made from biocompatible polymers (9, 10). These systems enable direct delivery of antimicrobial agents or other medications into the RCS causing the gradual release of constant concentrations of these medications. Adjunct use of different formulations of drug-loaded microparticles can decrease endodontic failure rate or even partially replace surgical or mechanical treatments, particularly in patients with resistant endodontic infections (11).

Biodegradable synthetic polymers include linear aliphatic polyesters, polyanhydrides, and poly (ortho esters); among them, linear aliphatic polyesters namely polylactic acid, polyglycolic acid and copolymers have extensive applications in local drug-delivery systems. Biological products such as glycolic and lactic acid mo-

meric units are naturally produced and then biodegraded in metabolic pathways of the human body. The difference lies only in their rate of degradability. Polyglycolic acid degrades more rapidly than polylactic acid (12). On the other hand, the behavior and the degradability of PLGA are controllable, which is the main advantage of this polymer and the reason for its vast application in medical and dental fields (13-15). Use of biodegradable polymers as microparticles loaded with medications is a suitable alternative to some complex medical and dental procedures. Many of the biocompatible synthetic microspheres are degradable and easily converted to three-dimensional matrixes of variable structures (12).

Several pharmacological and orthopedic studies have investigated the production of PLGA microspheres and have assessed the release of loaded antibiotics (16-20). In restorative dentistry, pulp-capping agents are used with the aim of eliminating microorganisms from the pulp chamber. Thus, it is particularly important to use antibiotic-loaded materials to minimize the microbial load as much as possible. Production of dental cements containing biodegradable polymer microspheres for controlled release of antibiotics without affecting the mechanical properties of these cements can be beneficial (21). In such conditions, the drug is loaded on the external surface or inside the synthetic microspheres of variable sizes (9, 10).

This study reviewed the polymer microspheres enabling a local, and controlled release of medications in the field of endodontics. Moreover, we evaluated the characteristics of drug-loaded polymer microparticles enabling delivery and gradual release of large amounts of medications, especially antibiotics into the root canal system.

2. Evidence Acquisition

We reviewed studies on the application of polymeric microparticles for delivery and release of medications for dental purposes. The under study drugs were either antibiotics or materials with innate antibiotic properties. No limitation was set for the methodology of studies. Studies on delivery of non-steroidal anti-inflammatory drugs (NSAIDs) or use of polymers for drug delivery for periodontal diseases were excluded. In addition to the application of drugs for root canal treatment, studies on factors influencing the quality and quantity of drug release by polymers were also reviewed.

A search was carried out in the PubMed database (May 2013) using the keywords "PLGA", "polymer", "microparticle", "encapsulate", "drug delivery", "controlled release", "antibiotic", "gentamycin", and "amoxicillin". English articles that met the inclusion criteria were selected and qualitatively reviewed.

3. Results

In the first step, title and abstracts of the searched articles were evaluated, and then full text of the articles was thoroughly read. Seven of the selected articles had studied the properties of polymer microspheres, including PLGA and factors affecting drug release by them (16-20, 22, 23). Seventeen studies had investigated the subject of controlled drug release in dentistry (Table 1); out of them, 5 studies (with in vitro design) had described production of polymer microspheres for elimination of *E. faecalis* from the root canal system or disinfection of the canal (24-28). The quality of all selected articles was thoroughly evaluated.

Table 1. List of Studies on the Application of Microspheres for Controlled Release of Drugs in Dentistry

Reference number	Year	Polymer	Drug	Descriptions
(29)	2011	PLGA/PCL	Doxycycline	Application for the treatment of chronic periodontitis in human
(28)	2010	PLGA/zein	Amoxicillin	Controlled drug release and antimicrobial activity against <i>E. faecalis</i>
(25)	2010	PLGA	Methylene blue (MB)	Antimicrobial activity against <i>E. faecalis</i>
(30)	2009	PLGA	Minocycline	Satisfactory drug release in human
(31)	2009	Chitosan CMC Chitosan	-	Biocompatibility of microspheres in rat
(27)	2009	Chitosan	-	Elimination of bacterial biofilm from the RCS
(32)	2008	PLGA	Minocycline	Adjunct to mechanical methods for treatment of peri-implantitis in human
(33)	2008	Gelatin	Propolis	Controlled release within 7 days
(34)	2008	PLGA	Minocycline	Adjunct to periodontal surgery in human
(26)	2008	Chitosan	-	Antimicrobial activity against <i>E. faecalis</i>
(35)	2007	PLGA/PCL	Doxycycline	Application for treatment of chronic periodontitis in human
(36)	2007	PLGA	Minocycline	Adjunct to scaling in smoker patients with chronic periodontitis
(37)	2005	Alginate/Chitosan	Minocycline	Controlled release within 7 days
(38)	2002	PLGA	Minocycline	Adjunct to scaling in human
(24)	2000	Ethyl cellulose	Chlorhexidine	Providing a model for controlled drug release in RCT
(39)	1999	Polyphosphazenes	Naproxen, Succinyl-sulfathiazole	Controlled release within 800 hours in rabbit
(40)	1997	L-PLA, D, L-PLA, DL-PLG	Tetracycline	Controlled release within 12 days

3.1. Polymeric Microspheres

Biodegradable microparticles are extensively used as delivery systems for many bioactive compounds such as the low-molecular weight drugs, macromolecules, polypeptides, DNA and antigens (41, 42). Synthetic or natural polymers are used for the preparation of microspheres for nasal, respiratory, oral or intravenous administration (43-45). Natural polymers such as collagen, chitosan, hyaluronic acid, alginate and cellulose have been used for production of microspheres due to their optimal biocompatibility. Biodegradable poly (D, L-lactide-co-glycolide) microspheres have long been used as drug-delivery systems in biomedical fields. These polymers are used for preparation of microspheres because their excellent biodegradability and biocompatibility and enabling controlled release of drugs (9, 10, 12).

Administration of drugs by using polymer microspheres has several advantages, including controlled and localized release of drug, maintaining the desired dosage of drug during a specific time period (and subsequently preventing systemic complications) and preventing early degradation and physiological elimination of the respective material (12). The main factors controlling drug release from the microspheres include physical and chemical properties of polymers such as size of particles, their chemical composition, morphology, and rate of degradation as well as the type of drug; these factors will be discussed later in this paper.

3.2. Application of PLGA for Drug Release

In the past two decades, PLGA has gained attention as a popular polymer for the production of the localized delivery systems for controlled drug release and also tissue engineering purposes. The PLGA is a biocompatible and biodegradable polymer. It has been approved by the American Food and Drug Administration (FDA) and because of having adjustable mechanical properties, numerous studies have evaluated its use for the fabrication of delivery carriers for controlled release of drugs made of small molecules, proteins or other macromolecules (9, 10).

Because of having optimal biodegradability, PLGA has extensive applications in the clinical setting and drug-delivery systems, including using for controlled release of different doses of drugs without the need for surgical procedures (9, 10). On the other hand, by changing the parameters like the molecular weight of the polymer, the ratio of lactide to glycolide and the drug concentration, we may be able to alter the physical properties of drug/polymer matrix. By doing so, the desired dosage of the medication may be achieved and the desired time intervals in between releases can be estimated according to the type of medication (46-48). However, the risk of drug toxicity should be thoroughly evaluated through meticulous investigations.

Considering the optimal biocompatibility and biodegradability, PLGA polymers are considered as suitable carriers for drug delivery and controlled release of medications, peptides and proteins. Generally, the degradation of PLGA polymers and release of drugs can be enhanced by increasing the hydrophilicity and chemical interactions among the hydrolytic groups, control of crystallinity and increasing the surface/volume ratio of the carrier. All these factors should be taken into account to confer biodegradability and induce drug release (10). Accordingly, for short-term (less than one month) controlled release of drugs, an amorphous polymer with high hydrophilicity is recommended. For long-term controlled release of medications, an amorphous polymer with high molecular weight is suggested. For controlled drug release over very long periods of time (more than six months), a semicrystalline polymer with a high degree of crystallinity can be used. It has been stated that PLGA can be formulated in different sizes (nanosphere, microsphere or millimeter scale implants) in different drug delivery systems. Using nanotechnology, many drugs, peptides and proteins can be encapsulated and released in a controlled manner at different time points (49).

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3.3. Influential Factors on Drug Release

The function of the controlled drug release systems depends on various factors such as the microstructure, surface to volume ratio, permeability, flexibility, percentage of porosity, matrix degradation, drug solubility, and drug-matrix interaction (50). Factors such as the type of encapsulated drug and polymer itself also affect the rate of drug release (51).

Drug release under in vitro condition depends highly on the size of microspheres (52). Larger microspheres measuring 100-300 μm enable slow release of medications over a longer period of time in comparison to smaller microspheres (53). Slower release of the drug by larger microspheres is due to the decreased path of drug diffusion inside the microspheres and decreased specific superficial areas in larger microspheres compared to smaller ones. In one previous study, drug release during 24 hours was related to the small size of microspheres (1.54 μm) (16). Drug release under in vitro condition also depends on drug diffusion within the microspheres.

3.4. Drug Release Rate

The initial drug release is highly correlated to the presence of pharmaceutical agents on the external surface of microspheres (54). In one study, gentamycin (GEN) was not only encapsulated into the microspheres, but also superficially absorbed on the external surface of microspheres. The mentioned study showed the initial release, including 60% of the entire encapsulated drug (16). In this study, slow and controlled release of drug occurred following the initial burst.

Stable release phase has also been observed when small amounts of the drug slowly diffuse into the micro-

spheres and release into the buffer. The duration of the stable release phase in polymer matrixes highly depends on the type of polymer used (55). It appears that polymers degrade via surface erosion, bulk degradation or both mechanisms. Homogenous surface erosion occurs following the increased penetration of water molecules into the polymer matrix causing bulk degradation. In a study by Francis et al., cumulative release of the entire drug occurred within 20 hours by 95.33%. Drug release did not appear to be due to the surface erosion or slow degradation of polymer. Drug release in the final phase most probably occurs through the aqueous canals via the penetration mechanism (16). It has been demonstrated that water sorption into the polymer is accelerated in the presence of hydrophilic drugs and polyvinyl alcohol (PVA). This factor has significant effects on the release of medications. Francis et al. evaluated the controlled release of GEN in poly-3-hydroxy butyrate microspheres and demonstrated that GEN was encapsulated in these microspheres, and drug was released via the mechanism of penetration and degradation. The initial burst was followed by a constant, slow release of the drug (16).

Blanco-Prieto et al. evaluated the drug release behavior of GEN-loaded microspheres. They reported that the release profile of GEN from microparticles had a biphasic pattern and after an initial burst, a continuous drug release was reported for up to 4 weeks (17). Evaluation of the characteristics and antibacterial activity of amoxicillin-loaded electrospun nano-hydroxyapatite/PLGA composite nanofibers revealed that the loaded amoxicillin within the n-HA/PLGA hybrid nanofibers had a sustained release profile, significantly reduced burst-release profile, had a good biocompatibility, improved mechanical durability and optimal activity to inhibit the growth of the model bacterium, i.e. *Staphylococcus aureus* (18).

Assessment of the controlled release of GEN from calcium phosphate-PLGA bone cement demonstrated that antibiotic release would increase by 30% with increasing drug loading of microspheres. Moreover, the drug burst of GEN in the microspheres was abolished (nearly zero) (19). In another study, Lu et al. evaluated the release profile of GEN in its carriers via incubation of its formulation in phosphate-buffered saline (PBS) solution at a pH of 7.4, at 37°C and reported that the release profile of GEN had a biphasic pattern and the released amount was greater in the first 24 hours. This finding may be due to the presence of antibiotic absorbed onto the surface of nanoparticles (22). After the initial burst release, the rate of drug release decreased and a continuous release pattern was observed for up to 10 weeks. It appears that the hydrophilic formulation of drug increases the solubility and erosion of polymer matrix and instead of extensive degradation in more hydrophilic formulations, it causes the extensive release of drug in the secondary phase. The high content of hydrophilic glycolic units in the copolymer increases the rate of degradation and consequently, enhances the release of encapsulated drugs from their carriers.

Imbuluzqueta et al. showed that by increasing the amount of GEN in the understudy formulations, the amount of released drug decreased as PLGA502H nanoparticles with 20 mg loading had a sustained release of the drug over 70 days, but the same nanoparticles loaded with 40 and 60 mg hydrophobic complex of GEN-anionic surfactant (GEN-AOT) released 87% and 70% of their contents, respectively (20). On the other hand, 752H nanoparticles loaded by 20, 40, and 60 mg released 67%, 61%, and 55% of their drug content, respectively. These findings were also reported by Mu et al. (23). They demonstrated that drug loading had no significant effect on the size of particles, and these findings were attributed to the formation of a more hydrophilic structure and compact internal structure of nanoparticles preventing water sorption (23). Moreover, coalesce of nanoparticles decreases the available and accessible surface and changes the drug release profiles. Consequently, drug release is decelerated. It should be noted that use of in vitro drug release models has some limitations in terms of physical and chemical conditions. For example, the Mueller-Hinton agar culture medium used in experiments contains nitrogen, vitamin, carbon, sulphur, and amino acids; these compounds are not present in PBS. Thus, different behaviors may be observed when different bacterial culture media are used and may result in increased release of antibiotics, especially GEN.

3.5. Controlled Release of Drugs in Root Canal Treatment

In RCT, canals are filled with materials of various shapes like conical, long intracanal posts, or screws. Many compounds have also been recommended to be placed in the RCS as temporary dressing for drug delivery and restoration. Antibiotic-loaded polymer matrixes have also been used by some researchers in endodontic treatments. Constant release systems such as uniform fibers made of acetate vinyl ethyl, and needle-shaped systems (56) have also been invented enabling the release of different amounts of drugs over time. Huang et al. prepared chlorhexidine-loaded systems using ethyl cellulose and a needle-shaped device for root canal therapy. Accordingly, a theoretical model of a controlled release device in the clinical setting was suggested based on the diffusion model by taking into account factors such as morphology, absorption and binding properties, limited capacity of the release environment and temporary seal of the root canals (24).

Attempts have been made to develop new constant drug release systems for RCT and several prototypes of biocompatible polymers have been invented and applied, including chitosan, PLGA and polymethyl methacrylate. They have been used for coating of absorbent paper points to determine the controlled release rate of chlorhexidine digluconate from loaded paper points (57). Luzardo-Alvarez et al. designed and employed amoxicillin-loaded collagen sponges (57). They reported the effects of cross-linker

on drug release, water sorption, mechanical characteristics and cytocompatibility of sponges made of collagen and Gantrez® loaded with amoxicillin compared with uncrosslinked sponges. Some other alternatives have also been invented and used for this purpose. Pagonis et al. used a combination of 150-200 nm diameter PLGA nanoparticles loaded with the photosensitizer methylene blue (MB) for treatment of infections caused by *E. faecalis* strains (25). They also performed photodynamic therapy under in vitro conditions using extracted human teeth infected with *E. faecalis*. Nanoparticles prepared by a solvent displacement procedure were placed into infected root canals and the survival fractions relative to the mean colony-forming units (CFU) for control levels (Mean CFU = 260.4) were 41.5% for MB-loaded nanoparticles and 15.2% for light with MB-loaded nanoparticles. Kishen et al. evaluated 60-120 nm unloaded chitosan nanoparticulates via tripolyphosphate ionic gelation and recorded a positive surface charge of 40 mV (26). Chitosan nanoparticulates showed high antibacterial activity against *E. faecalis* under in vitro condition and had high compatibility with the endodontic sealer. A significant reduction in the adherence of *E. faecalis* to nanoparticulate-treated dentin was also reported because of the electrostatic interactions between negatively charged dentin and cationic nanoparticles that significantly prevented bacterial adhesion to dentin substrate and inhibited the formation of bacterial biofilm. Although the obtained bacterial adhesion is probably weak, nanoparticles may directly react with microorganisms after entering the root canal. Chitosan and some of its derivatives prevent the microbial growth via different mechanisms (58-60).

Shrestha et al. used nanoparticles made of chitosan using high-intensity focused ultrasound (27). Bacterial adhesion was prevented by the formation of collapsing cavitation bubbles to deliver antibacterial nanoparticles into hard-to-reach dentinal tubules in conventional RCT. Recently, chitosan has become increasingly popular for the development of new types of root canal filling or pulp capping cements alone or in combination with other materials (61, 62).

Other studies have investigated microparticle-based formulations; which seem to be easily delivered to hard-to-reach areas in conventional RCT. Sousa et al. loaded PLGA microspheres with amoxicillin (28). Microparticles, measuring 5-38 µm, were prepared using a spray-drying technique and different drug-release patterns were observed among formulations. Drug composition played a significant role in the controlled release profile. The antibacterial activity of AMX continued even after its encapsulation against *E. faecalis* and as demonstrated by the antibiogram results; AMX had antimicrobial effects for 6 hours against *E. faecalis* strains. Spongy particles in contact with water enhance complete elimination of the formulation from the simulated RCS. However, this study, like the previous ones, was conducted in vitro; which has significant differences with the clinical setting.

4. Conclusions

Drugs are usually absorbed from the oral cavity in a short period of time; accordingly, duration of their activity will be short too. However, application of drugs into the tooth cavities or the RCS using drug-delivery systems, increases the durability of the medication. In the RCS, the drug-delivery system is limited to the root canal space. This system must be non-toxic and allow a constant drug release. It should be compatible with dentin and has low surface tension and optimal adaptation to the canal wall. It must be easily retrievable as well. It should be noted that tissue compatibility of the materials is particularly important in endodontic treatment; because they are in contact with the periapical tissue via the root apex.

Another advantage of using intracanal medicaments is that they are not easily washed away. The intracanal medicaments are surrounded by a tissue with low enzymatic activity, and the medicaments are in direct contact with the tooth structure. Furthermore, the need for long-term temporary dressing of tooth is eliminated. In this technique, these products are eliminated from the root canal system by the termination of treatment; therefore, hypersensitivity or allergic reactions would not occur. Considering all the above, the RCS is an important therapeutic route, particularly for teeth with endodontic failure.

Antibiotics are among the main medicaments used for elimination of bacterial infections (21, 63-65). Drug-delivery systems are placed into the canal prior to obturation. The drug-releasing microparticles may be injected into a prepared root canal by a syringe to prevent the bacterial growth. However, no animal model or human studies have been conducted in this respect yet.

The efficacy of root canal treatment or retreatment can be significantly enhanced by using microparticles. In endodontics, this treatment modality is taking its first steps and the required criteria for an ideal drug delivery system have yet to be defined for endodontic purposes. These systems should provide the desired concentrations of antibiotics or other drugs in the RCS. Researchers have set some criteria for these systems as well (11). These materials should be non-toxic and standard. Drug release should be continuous and precisely controlled. Ideally, these systems must be absorbed by the body after a while. Moreover, they must be small enough to enter the root canal system. They should also be sterilizable because they are primarily used for infection control.

Conventional management of endodontic failures includes the use of disinfecting irrigation solutions and mechanical procedures to remove the infected tissue and to prevent the re-growth of bacteria. Use of microsphere-based controlled drug-delivery systems provides constant release of the desired concentrations of drugs and increases the efficacy of conventional mechanical treatments. Controlled release of polymer nanoparticles enables drug delivery to the desired sites and improves the efficacy of medications by changing the physical

and chemical properties of microspheres or selection of different formulations. The efficacy of microspheres depends on the physical and chemical properties of the polymer, kinetic energy of release, site of infection, type of the pathogen, and the selected drug. Based on our results, drug-loaded microspheres can be used as carriers for controlled release of some active compounds such as antibiotics in areas like tooth cavities or root canals. Administration of simple formulations of nanoparticles in root canal cavities can decrease treatment sessions and may be an adjunct to surgical protocols to increase treatment success rate. However, the efficacy of loaded drugs and antibiotics must be systematically evaluated in animal model studies and clinical trials. Drug-loaded microspheres can provide new perspectives in endodontic treatments.

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