ANTIMICROBIAL PEPTIDES IN ENDODONTICS. A REVIEW

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Abstract

Antimicrobial drugs are facing a significant problem as a result of the sharp rise in endodontic infections that are drug-resistant. The urgent need to create new control agents is highlighted by the inability of the strongest medicines to eradicate "superbugs." Antimicrobial peptides (AMPs), a rising class of natural and synthetic peptides with a wide range of targets including viruses, bacteria, fungi, and parasites, are reviewed here along with recent advancements in the field.

Keywords: antimicrobial peptides, endodontic biofilm, entercoccus faecalis

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Introduction:

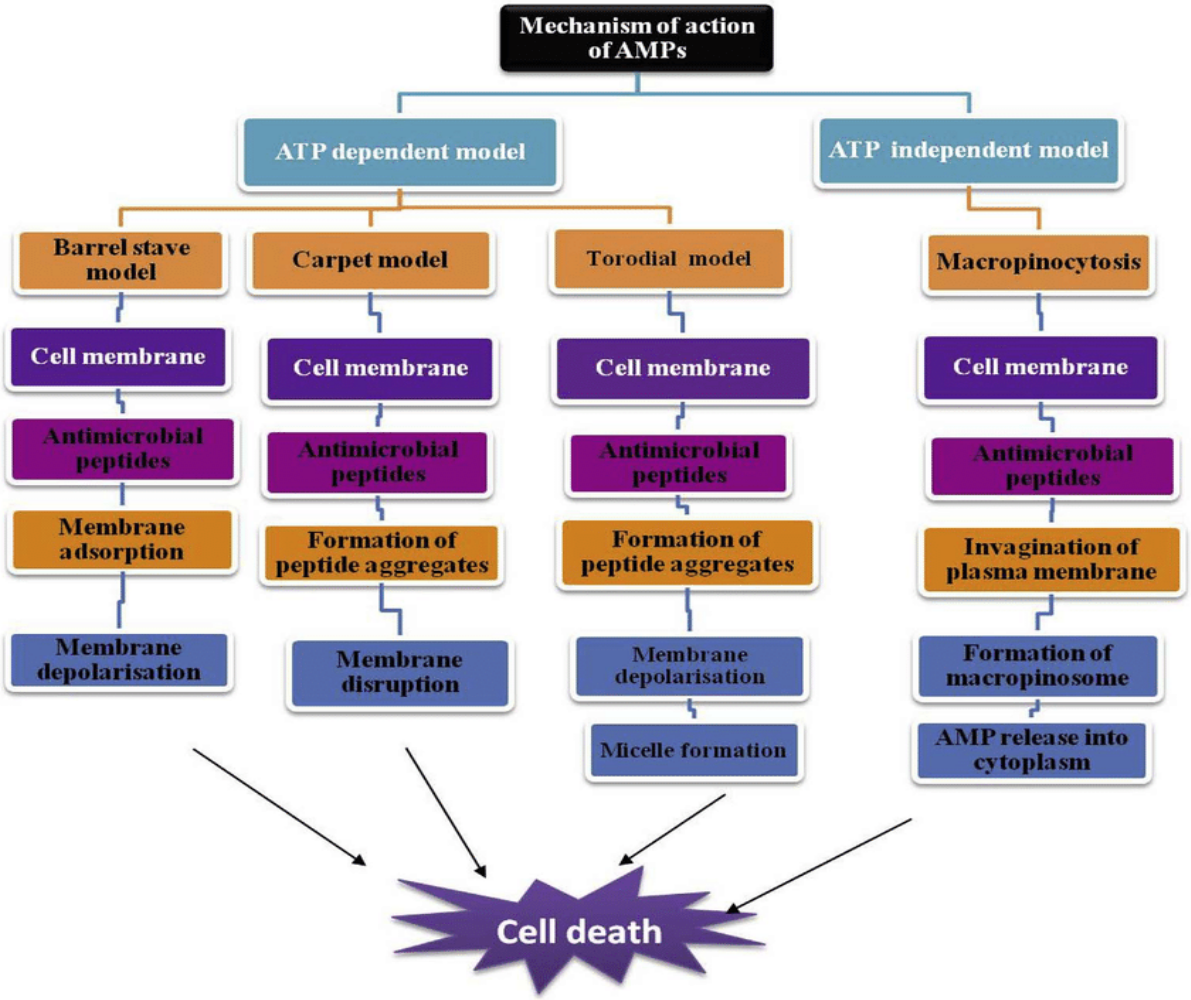
The tooth's developing organ is the pulp. The most frequent cause of pulpal infection is dental caries, and the microbe's virulence speeds up the infection's course, causing an irreversible pulp infection. In order to remove resistant microbes and encourage tissue repair, the current treatment for endodontic infection involves access cavity preparation, cleaning, and shaping, intracanal medication placement, and irrigation of the root canal with potent irrigants like chlorhexidine and sodium hypochlorite.

As a result, the search for novel agents capable of eradicating such microorganisms becomes critical. The research and launch of new endodontic remedies was proposed, including the use of a bio tool called antimicrobial peptides. Antimicrobial peptides are endogenous macromolecules that are created early in the course of an infection to keep the host-pathogen connection healthy.

In endodontic therapy, their concentration and tissue repair abilities operate as an intracanal medicament. This review focuses on naturally occurring and synthetic peptides, as well as their prospective use as an intracanal medicament and a regenerative agent, as well as the advantages of the applications and treatment outcomes [1].

Mechanism of action

Gram-negative and Gram-positive bacteria, drug-resistant strains, and even fungi are all killed by antibiofilm peptides [16,17]. The majority of AMPs permeate bacterial cell membranes, resulting in extensive harm or tiny flaws that allow the transmembrane potential to deplete and ultimately cause cell death [16,18]. Particularly, pore and nonpore models aid in the explanation of the mechanisms of action.



There are two pore model theories:[18] the barrel stave pore model and the toroidal pore model. In the barrel stave pore model[19], AMPs interact with the bacterial cell membrane to produce a hydrophilic channel. In the toroidal pore model[20], AMPs have an effect on the membrane's curvature. Models for the nonpore theory have included the carpet model, the detergent model, the molecular

shape model, and others. The carpet model, which depicts parallel AMP deposition on the cell membrane and results in bilayer instability[21], is the model that is most frequently utilised. According to each of these hypotheses about how antibiofilm peptides work, a certain level of peptide concentration in the cell membrane is necessary for disruption. These peptides that target the cell membrane are well suited for use in medicine as well as in dentistry.

In addition to the models mentioned above, a number of special peptides have a varied impact on antibiofilm performance. It has been demonstrated that these peptides prevent the synthesis of bacterial cell walls, degrade DNA or RNA in cell plasma, fragment or destroy intracellular proteins, activate autolysin, and impair enzyme activity[7].

Antibiofilm peptides can have additional qualities, such as mineralization and anti-resorptive effects, in addition to their direct antibacterial activity. These additional qualities may be important when treating the complex oral microbial populations using peptides. According to Kraus et al.[22], human-defensins (HBDs) can affect the maturation and proliferation of osteoblast-like cells.

Osteoclastogenesis, a process critical for the healing of oral wounds, is decreased by several AMPs generated by host cells[23]. Typically, peptides from the cathelicidin family bacteria protect against infection-induced osteoclastic bone resorption. [24.]

Natural antimicrobial peptides: (table 1)

Peptide	**Microorganism**	**Minimum inhibitory concentration**	**Reference**
Human beta-defensin (HBD) 3	Fusobacterium nucleatum , Enterococcus faecalis , Tannerella forsythia , Eikenella corrodens, Candida albicans	5 10 >25 2 10	Song et al. (2009) Song et al. (2009) Ji et al. (2007) Ji et al. (2007) Song et al. (2009)
SMAP29	Fusobacterium nucleatum	1	Weistroffer et al. (2008)
Human neutrophil defensin 1	Prevotella intermedia	2	Gursoy et al. (2013)
SMAP14A	Peptostreptococcus micros	2	Weistroffer et al. (2008)
LL- 37	Treponema denticola, Enterococcus faecalis, Porphyromonas gingivalis, Tannerella forsythia , Eikenella corrodens	19	Rosen et al. Thennarasu et al. (2010) Thennarasu et al. (2010) Ji et al. (2007) Ji et al. (2007
HBD1	Enterococcus faecalis	10	Lee and Baek (2012)
HBD2	Enterococcus faecalis	5	Lee and Baek (2012
HBD4	Enterococcus faecalis	3	Lee and Baek (2012
Clavanin A	Enterococcus faecalis	1	Lee and Baek (2012
Cecropin B	Prevotella nigrescens	25	Devine et al. (1999)
AR23	Prevotella melaninogenica	3	Urban et al. (2007)
Mellitin	Candida albicans	6	Urban et al. (2007)
SMAP28	Actinomyces naeslundii , Actinobacillus actinomycetemcomitans, Actinomyces israelii	1 1 1	Weistroffer et al. (2008) Weistroffer et al. (2008) Weistroffer et al. (2008)
Meta-phenylene ethynylene (mPE)	Actinomyces viscosus, Actinobacillus actinomycetemcomitans , Porphyromonas gingivalis	2 1 4	Beckloff et al. (2007) Beckloff et al. (2007) Beckloff et al. (2007)
Chrysophsin 1	Enterococcus faecalis , Actinomyces naeslundii, Streptococcus gordonii	3 3 3	Wang et al. (2012) Wang et al. (2012) Wang et al. (2012)
Nisin	Streptococcus gordonii Enterococcus faecalis	150 150	Turner et al. (2004) Turner et al. (2004)

TABLE 1

Most living things, including the human body, can produce short chains of amino acids called monomers to fight bacterial infection. [4]. Micronized compounds known as antimicrobial peptides (AMPs) or host-defense peptides (HDPs) are found in the innate immune systems of animals and plants as well as in bacteria and fungus. [5]

One of the earliest cationic peptides discovered was nisin, which was isolated from Lactobacillus lactis in 1928.

Now marketed for use in food, polylysine is a natural AMP produced from Streptomyces albulus 346.

Nisin and polylysine use in dentistry was found to be synergistic for in vitro suppression of Streptococcus mutans [8].

Natural AMPs have recently been used as templates to construct synthetic peptide libraries [9].

Synthetic antimicrobial peptides: (table 2)
In recent years, natural peptide templates have been modified in terms of biological activity and size in order to improve their antibiofilm function [10]. A recent study found that removing four N-terminal amino acids from the antimicrobial peptide B1CTcu5 produced by frogs has bactericidal effects [11].
Another study discovered that a dermaseptin S4 peptide could reduce bacteria connected to oral problems by replacing fatty acids for amino acids. [12]. Pseudomonas aeruginosa biofilm growth has been shown to be severely inhibited by certain D-enantiomeric peptides (DJK-5 and 6), and preformed biofilms of numerous more Gram-negative bacteria, both wild-type and antibiotic-resistant, have also been shown to be destroyed [13].

Another technique for lowering the length of expensive AMPs is sequence truncation. The effectiveness, stability, and safety of the modified peptides can be increased by properly truncating certain parts of the original long-length peptide [14]. Additionally, two or more proteins can be combined to create antibiofilm peptides.

According to one study, a hybrid peptide's antifungal impact on Candida albicans is achieved via disrupting the yeast cell's plasma membrane [15]. High-throughput screening matrixes can be used to create next-generation peptides with increased biological activity relative to parent peptides [9].

Product	**Test phase**	**Active peptide**	**Drug presentation**	**Target**	**Reference**
Toraymyxin	Phase 3	PolymyxinB	Fiber	Gram-negative bacteria	Shimokawa et al. (2012), www.fda.gov, www.patft.uspto.gov
Cubicin	Clinically approved/ marketed	Daptomycin	Injection	Gram-positive bacteria	Cafini et al. (2007), www.fda.gov, www.patft.uspto.gov
Forteo	Clinically approved/ marketed	Teriparatide	Injection	Osteoporosis	Yamaguchi and Sugimoto (2009), www.fda.gov, www.patft.uspto.gov
Byetta/ exenatide	Clinically approved/ marketed	GLP-1	Injection	Type 2 diabetes Mellitus	Holscher (2010), www.fda.gov, www.patft.uspto.gov
Fuzeon	Clinically approved/ marketed	Enfuvirtide	Injection	HIV	Yu et al. (2013), www.fda.gov, www.patft.uspto.gov
Firmagon	Clinically approved/ marketed	Degarelix	Injection	Prostate cancer	Clyne (2014), www.fda.gov, www.patft.uspto.gov
Mepact	Phase 3	Mifamurtide	Injection	Osteosarcoma	Ando et al. (2011), www.fda.gov, www.patft.uspto.gov
Natrecor	Clinically approved/ marketed	Nesiritide	Injection	Cardiovascular Diseases	Lyu et al. (2014), www.fda.gov, www.patft.uspto.gov
Zoladex	Clinically approved/ marketed	Goserelin	Injection	Cancer	Kawakami and Morales (2013), www.fda.gov, www.patft.uspto.gov
Copaxone	Clinically approved/ marketed	Glatiramer	Injection	Multiple sclerosis	Towfic et al. (2014), www.fda.gov, www.patft.uspto.gov
Sandostatin	Clinically approved/ marketed	Octreotide	Injection	Neuroendocrine tumor	Banck and Beutler (2014), www.fda.gov, www.patft.uspto.gov

Somatuline	Clinically approved/ marketed	Lanreotide	Injection	Acromegaly	Caron et al. (2014), www.fda.gov, www.patft.uspto.gov
Firazyr	Clinically approved/ marketed	Icatibant	Injection	Hereditary Angioedema	Bas et al. (2013), www.fda.gov, www.patft.uspto.gov
Prialt	Clinically approved/ marketed	Ziconotide	Injection	Pain	Strigenz and Ziconotide (2014), www.fda.gov, www.patft.uspto.gov
Symlin	Clinically approved/ marketed	Pramlintide	Injection	Diabetes mellitus	Tomabechi (2013), www.fda.gov,
Nplate	Phase 3	Romiplostim	Injection	Dyscrasias Thrombocytopenic	Seidel et al. (2014), www.fda.gov, www.patft.uspto.gov
Recaldent	Clinically approved/ marketed	CPP-ACP	Toothpaste; chewing gum; tooth mousse	Tooth remineralization	Zhou et al. (2014), www.fda.gov, www.patft.uspto.gov
Iseganan	Phase 3	Iseganan	Mouthwash	Mucositis	Giles et al. (2002), www.fda.gov, www.patft.uspto.gov

TABLE 2

Antimicrobial peptides application in endodontics:
Endodontic pathology has a well-established role in the onset and progression of biofilm [25]. Free-floating microorganisms in the root-canal region can link with one another and create a biofilm [26] as a microbial community. It has been demonstrated that the biofilm's maturity affects its resistance to antibiofilm drugs [27]. Bacteria in established biofilms can tolerate irrigation and are very challenging to eradicate [28]. Given that specific antibiofilm peptides have been created and used in numerous other medical and dental-related aspects, endodontics should be viewed as a potential application area.

Biofilm grown in an open-culture model

The bulk of endodontic biofilms in the root-canal system develop on the dentin walls. For the replication of biofilm infection, various in vitro models have been developed. biofilms can reportedly grow on the bottom of 96-well plates or on collagen-coated hydroxyapatite discs, both of which provide simple access for cleaning solutions.[29]

Enterococcus faecalis is a frequent isolate that has been linked to chronic endodontic infections [30]. E. faecalis' innate antimicrobial resistance and ability to adapt to harsh environmental changes may play a role in the development of these infections[31]. In endodontics, irrigation solutions such as chlorhexidine have long been utilised as an intra-canal medicament.

However, in actual practise, the killing and penetration efficacy of various endodontic antimicrobial drugs appear to be less than optimum [32]. To maximise the eradication and biofilm disruption of therapy-resistant microorganisms, different intra-canal medicaments should be investigated [33]. Antibiofilm peptides have been employed in several investigations to assess their efficiency against microorganisms often present in endodontic diseases. In both planktonic and biofilm cultures, Liu et al. [33] investigated the antibacterial effectiveness of a casein peptide against E. faecalis.E. faecalis planktonic growth was strongly reduced by both glycosylated and non-glycosylated versions of the kappa-casein peptide. The glycosylated version of the kappa-casein peptide effectively reduced E. faecalis biofilm development, suggesting that it could improve the efficacy of standard antiseptics [33]. On a 3-day old E. faecalis biofilm, researchers examined the antibiofilm activity of four synthetic lipopeptides. The lipopeptide synthesised in the biohybrid polymer medium displayed some antibiofilm activity against E. faecalis, according to confocal microscopy and MIC data[34].

Oral multi-species plaque biofilm formation was successfully prevented both in the presence and absence of saliva, indicating that the peptide was resistant to inhibition or degradation by salivary factors [35]. Furthermore, when low-concentration peptide 1018 (10 g/mL) and 2% chlorhexidine were used together, the antibiofilm action was boosted compared to when they were used separately, resulting in >50% biofilm bacteria killing [35]. On

single- and multi-species oral biofilms, a recent study compared the antibiofilm efficiency of a new D-enatiometic peptide (DJK-5) with peptide 1018 and found that DJK-5 was more effective in biofilm inhibition than peptide 1018 [36]. Lee et al. [37] tested the antibiofilm efficiency of the HBD3 peptide against mixed species biofilms produced by four different species (Actinomyces naeslundii, L. salivarius, S. mutans, and E. faecalis); HBD3 had stronger bactericidal activity than calcium hydroxide on 3-week-old biofilms.

Dentin biofilm model

The difficulty of removing biofilm germs is exacerbated by disinfecting chemicals' antibacterial action being inhibited, for example, by numerous charged molecules and substances in the chemical environment of the root canal [38].Dentin has been found in previous research to reduce the antibacterial efficacy of numerous endodontic disinfectants [39,40]. As a result, the bacteria's survival could be linked to their invasion into dentinal tubules, where they are protected from endodontic medicaments [41].

For the evaluation of dentin disinfection in endodontics, many dentin infection models have been created[38]. HBD3's antibiofilm efficacy against 3-week-old E. faecalis biofilms was assessed in a recent study using the culturing procedure to create standardised dentin blocks[33]. The peptide outperformed calcium hydroxide and chlorhexidine gel in terms of antibacterial effectiveness. HBD3 decreased E. faecalis biofilm development considerably[33].

Challenges of antimicrobial peptides in clinical application:

Despite all of the benefits that antibiofilm peptides provide, there are certain limitations to their therapeutic efficacy [18]. The antibiofilm activity of peptides can be dramatically diminished in the presence of biological fluids such as plasma, serum, or saliva when compared to their performance in non-physiological settings[42]. Furthermore, antibiofilm peptides are difficult and expensive to produce in high numbers[43], owing to the complicated extraction, isolation, and purification techniques required. Antibiofilm peptides are also challenging to provide by parenteral route because they are quickly eliminated by the kidney [44].Furthermore, some antibiofilm peptides are toxic to host cells and can trigger undesired pro-inflammatory responses [45].

Conclusion:

Antibiofilm peptides have been reported to be ineffective against oral biofilms associated with caries, endodontic, periodontal, and fungal diseases. But more investigation into novel peptides that are simpler to produce and that might alleviate some of the drawbacks of the current peptides is still required.

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