

Epigallocatechin gallate Mucoadhesive Gingival Patch as Potential Biomaterial to Regulate Macrophage and Lymphocyte Cells in Periodontitis: A Review

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ABSTRACT

Background: Periodontitis is a periodontal disease that affects more than 743 million people worldwide and causes damage to the periodontal ligament and alveolar bone. One of the bacteria that can cause periodontitis is *Porphyromonas gingivalis* (*P.gingivalis*). *P. gingivalis* has virulence factors that can damage the periodontal tissue. Treatment of periodontitis is in the form of non-surgical therapy such as scaling and root planning and some cases, doxycycline can be given as adjunctive therapy after scaling and root planing. Epigallocatechin gallate (EGCG) is one of the catechins found in green tea and has anti-bacterial properties. **Purpose:** The study aimed to describe the potency of the mucoadhesive gingival patch with EGCG green tea against the number of macrophage cells and lymphocyte cells during periodontitis through narrative review. **Review:** Mucoadhesive gingiva patch loaded with EGCG has the advantages such as maintaining drug bioavailability, non-invasive, and optimizing drug distribution. Using a mucoadhesive gingiva patch with EGCG can reduce macrophage and lymphocyte cells by inhibiting lipopolysaccharide, a virulence factor of *P. gingivalis*. Inhibited lipopolysaccharide will inhibit pro-inflammatory cytokines such as TNF- α and IL-6. Macrophage and lymphocyte cells will reduce due to the inhibition of pro-inflammatory cytokines. **Conclusion:** Mucoadhesive gingiva patch with EGCG green tea potentially to decreased macrophage and lymphocyte cells in periodontitis.

Keywords: Medicine; Dentistry; Epigallocatechin gallate; Tumor Necrosis Factor- α ; Periodontitis

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INTRODUCTION

Periodontal disease is an inflammatory condition in the periodontium and is one of the two biggest problems for oral health, with periodontitis being the sixth largest chronic disease that affects more than 743 million people worldwide. Periodontal disease can impair mastication, aesthetics, self-confidence, and quality of life.¹⁻³ Periodontitis is a common periodontal disease in which inflammatory conditions are caused by specific microorganisms. An inflammatory condition that occurs in the supporting tissues of the teeth causes gradual destruction of the periodontal ligament and alveolar bone.⁴ *P.gingivalis* is one of the microbes that can cause periodontitis and this microbe can be found in periodontal pockets in patients with periodontitis, where these microbes can interfere with tissue homeostasis by manipulating host signaling pathways.⁵ Green tea (*Camellia sinensis*) is one type of beverage that is well-known worldwide and contains various compounds such as amino acids, polysaccharides, vitamins, and polyphenols. Epigallocatechin gallate (EGCG) is one of

the polyphenols in green tea, which can help degrade the risk of cardiovascular disease and reduce the risk of developing periodontal disease.^{6,7} Administration of EGCG was able to inhibit lipopolysaccharide (LPS), which virulence factor of *P. gingivalis*. Hence, LPS could not activate pro-inflammatory cytokines. Therefore, increased inflammatory cells did not occur.⁸ Furthermore, this narrative review aims to explain the potency of the mucoadhesive gingival patch with EGCG green tea against the number of macrophage cells and lymphocyte cells in periodontitis.

Periodontitis

Periodontitis is an inflammatory condition that occurs in the supporting tissues of the teeth, which causes progressive damage. Periodontitis can occur due to interactions between bacteria, the environment, genetic and immune factors. One of the bacteria that can become pathogenic and is present in the accumulation of plaque on the teeth and the gingival surface is *P. gingivalis*. *P. gingivalis* is a group

of gram-negative bacteria and obligate anaerobes that are rod-shaped. *P. gingivalis* obtain metabolic energy from the fermentation of amino acids, heme, and vitamin K for growth. *P. gingivalis* can cause periodontitis and several systemic diseases such as Alzheimer's, cardiovascular, and rheumatoid arthritis. Lysine-specific gingipain (Kgp) and arginine-specific gingipain (RgpA/B) were produced by *P. gingivalis* for survival. These bacteria can create a dysbiosis that occurs between the host and dental plaque. In addition, *P. gingivalis* can trigger an immune response by increasing the concentration of pro-inflammatory mediators that cause an increase in periodontal destruction.^{4,9-11}

P. gingivalis has factors virulence that has a large role to defend itself in the host cell and increase the potential of *P. gingivalis* to cause disease, namely T9SS which plays a role in the secretory system, PAD or peptidyl arginine deiminase, type V pili, and Mfa which act as fimbriae, LPS which acts as protection, capsules as protection, and OMVs that act as extracellular vesicles.¹² LPS which is secreted by *P. gingivalis* can induce the production of pro-inflammatory cytokines. Other than that, LPS disrupts damage to the epithelium by affecting the junction between the gingival epithelial cells.¹³ The virulence ability of *P. gingivalis* LPS depends on the lipid A component. Host cells exposed to *P. gingivalis* LPS lipid A can cause an inflammatory response in the gingival tissue so that the environment around pathogenic bacteria becomes a good place for bacterial defense and increases the severity of the periodontal disease. Recent studies have shown that the specific binding between Toll Like Receptor (TLR) and LPS is determined by the structure of the lipid type A. The heterogeneity of lipid A in *P. gingivalis* LPS causes the innate immune response and production of different pro-inflammatory cytokines.¹⁴ Removing plaque and calculus is a form of periodontitis treatment. Plaque and calculus removal is done by scaling and root planing. Administration of antibiotics such as doxycycline as an adjunct therapy after scaling and root planing was considered effective.^{15,16}

Inflammation is the body's defense process that activates the innate immune system that occurs due to a microorganism infection or injury. Adhesion between the bacterial biofilm and the tooth surface can cause an inflammatory immune response. Lipopolysaccharide that binds to Toll-like receptors 4/2 will induce the formation of protein kinases that result in activated pro inflammatory transcription factors. The result of activated pro-inflammatory transcription factors is the release of mediators that will cause an immune response. The increased neutrophil in acute inflammatory conditions will lead to the formation of Interleukin-17 (IL-17), Cluster of Differentiation (CD) CD4+, and T helper (Th)17. Inflammation that has reached a chronic stage will activate the adaptive immune system. Lymphocytes in the adaptive immune system release inflammatory mediators that alter the balance of bone metabolism, thereby signaling the transition from gingivitis to periodontitis. Two types of signals are required for lymphocyte activation: signals induced by antigen receptors and costimulatory signals by Antigen Presenting Cells (APC). In periodontitis, the

predominant APCs are CD19+ and CD83+ B lymphocytes. Thus, activation of adaptive immunity affects bone loss in periodontitis and several studies have shown that B and T lymphocytes are activators of RANKL during periodontal inflammation.¹⁷

Macrophage Cells

Macrophages are proinflammatory molecules that have the potential to damage tissues such as Interleukin (IL)-1, Tumor necrosis factor (TNF)- α , Matrix Metalloproteinase (MMPs), and Prostaglandin E2 (PGE2) which are elevated in the gingiva and gingival crevicular fluid of patients with periodontitis. Macrophages contribute to the degradation of the collagen matrix in the periodontal connective tissue. Macrophages consist of M1 and M2 whereas M1 macrophages are induced by agents microbial such as LPS and show high phagocytosis and increased expression of cytokines proinflammatory. M2 macrophages are induced by Th2 cytokines and secrete IL-10 which plays a role in tissue regeneration. In periodontal inflammation, M1 macrophages are more dominant than M2 macrophages, so M1 macrophages are associated with periodontitis.¹⁷

Lymphocyte Cells

Lymphocytes are cells that play a role in the adaptive immune response. Lymphocyte cells consisting of natural killer (NK) cells, T lymphocyte cells, and B lymphocyte cells are derivatives of lymphoid progenitor cells. In the adaptive immune system, B cells are humoral components that can secrete antibodies and cytokines and act as APCs. T cells have a major role in cellular immunity and undergo maturation in the thymus and some in the tonsils. T cells have a role in producing pro-inflammatory cytokines, cytotoxic activity, and regulation of the other immune cells.¹⁸

Green Tea

Tea is one of the three most popular non-alcoholic beverages in the world and has many health benefits. According to the degree of oxidation, tea can be divided into green tea, oolong tea, and black tea. *Camellia sinensis* and *Camellia assamica* are two varieties of the tea plant that are cultivated and produce the most widely consumed tea commercially. The tea plant is thought to have originated in areas in the Yunnan, Guangxi, and Guizhou provinces of China.^{19,20} Green tea has white flowers and is fragrant. The leaves of green tea are dark green and oval and have jagged edges.^{21,22} Green tea polyphenols, namely catechins, are known to have anti-inflammatory effects. Several studies have shown that green tea catechins are effective in preventing periodontal inflammation through inhibition of cyclooxygenase-2 (COX-2) expression, negative regulation of PGE2 in

osteoblasts, and suppression of receptor activator nuclear kappa beta ligand (RANKL) expression with consequent inhibition of osteoclast activity and bone resorption. In addition, green tea catechins have the effect of inhibiting the expression of pro-inflammatory cytokines such as TNF-, IL-6, and IL-8.²³

Epigallocatechin-3-gallate (EGCG)

Catechins are the main components of polyphenols found in green tea. These catechins consist of epicatechin (EC), epicatechin gallate (ECG), epigallocatechin (EGC), and epigallocatechin-3-gallate (EGCG). Epigallocatechin-3-gallate is one of the polyphenols contained in green tea leaves by 50-80% and it's the most catechin in green tea and as anti-tumor, anti-viral, and anti-bacterial.²⁴⁻²⁶

EGCG can protect against the damaging effects caused by the invasion of *P. gingivalis*. Another effect of EGCG is the treatment of periodontitis due to its bactericidal effect on periodontal pathogens. The addition of ECG and EGCG can inhibit the expression of IL-6 and IL-8 when dental pulp cells are exposed to lipopolysaccharide and prostaglandin. EGCG inhibits the expression of several pro-inflammatory factors such as VEGF, COX-2, and PGE2 in human dental pulp stem cell (hDPSC) after stimulation with lipopolysaccharide.^{6,27}

EGCG has an anticancer effect by inhibiting the activity of carcinogens. Several studies suggest that EGCG can inhibit the IGF/IGF-1R axis so that diethylnitrosamine-induced obesity-related liver tumorigenesis can be prevented. EGCG can also play a role against angiogenesis so that tumor proliferation can be restrained. Besides being able to act as an anti-cancer, EGCG has a role as an antioxidant. Studies show EGCG can inhibit oxidative stress on blood platelets and improve mitochondrial function.²⁸

Mucoadhesive Gingival Patch

The patch is a preparation consisting of two layers of the main layer containing a mucoadhesive polymer coated with an impermeable backing layer.²⁹ Mucoadhesive is described as the ability to produce a bond from contact between the adhesive and the mucous membrane.³⁰ Various kinds of patches advantages are non-invasive, have minimal side effects, and can maintain drug bioavailability so that patients can improve patient compliance.² Mucoadhesive gingival patches consist of active ingredients, polymers, backing layers, plasticizers, and penetration enhancers. Mucoadhesive polymers aim to deliver the active substance to a specific location and optimize drug distribution. Optimizing drug distribution is done because of longer contact. Polymer is also a layer in contact with oral mucosa which is an important factor in the success of drug distribution. A group of polymers that are mucoadhesive and have potential and are economical among them are chitosan, hydroxyl propyl methylcellulose (HPMC), polyvinyl

pyrrolidone (PVP), and polyvinyl alcohol (PVA). These polymers are widely available so that able to be developed for a better drug distribution system.³¹

The backing layer serves to prevent the active ingredients from dissolving and being swallowed with saliva. In addition, the backing layer also provides the active ingredients with the direct flow to the layer mucosa. The backing layer is formed from a water-impermeable polymer.²⁹ One of the materials used in the backing layer is ethyl cellulose.³² Plasticizer is a component that functions to form a smooth thin film and flexible consisting of one type or a mixture of polymers. The plasticizer can also prevent the film from breaking, tearing, and peeling. The materials used as plasticizers include PEG-100, 400, propylene glycol.^{33,34} Penetration enhancers aim to help increase the penetration of active ingredients. One of the materials that can be used as a penetration enhancer is cyanoacrylate.^{32,33}

DISCUSSION

P. gingivalis is a gram-negative bacteria that can be pathogenic and cause periodontal disease. Virulence factors of *P. gingivalis* such as LPS cause *P. gingivalis* to become a keystone pathogen by causing dysbiosis and chronic inflammation.³⁴ Periodontitis is an inflammatory condition characterized by immune cells such as neutrophils, monocytes, and macrophages moving towards the site of inflammation. LPS possessed by bacteria can activate macrophage cells. Macrophage cells will phagocytize existing pathogens and induce the secretion of pro-inflammatory cytokines such as IL-1 β , IL-6, and TNF- α which will be secreted by activated macrophage cells. Excessive and prolonged production of pro-inflammatory cytokines can cause tissue damage.^{35,36}

Lymphocyte cells as a specific immune response appear when there is an injury during chronic inflammation. Lymphocytes induce receptor activator of nuclear factor- κ B ligand (RANKL) and bind to receptor activator of nuclear factor- κ B (RANK) resulting in osteoclast activation. The increasing number of osteoclasts without increasing the number of osteoblasts will cause bone resorption.³⁶ Using a patch loaded with EGCG is considered effective in reducing the number of macrophages and lymphocytes by inhibiting the lipopolysaccharide of *P. gingivalis*. Lipopolysaccharide can induce inflammatory cells to secrete pro-inflammatory cytokines such as TNF- α and IL-6. Pro-inflammatory cytokines can stimulate the activation of macrophages and lymphocytes. LPS inhibited by EGCG causes inhibition of these pro-inflammatory cytokines and inflammatory cells such as macrophages and lymphocytes will decrease.^{36,37} Reactive Oxygen Species (ROS) are induced by extracellular inflammatory stimuli such as lipopolysaccharide. NF- κ B activated by ROS can increase pro-inflammatory cells such as TNF- α , IL-1, and COX-2. EGCG can weaken the ability of ROS to produce NF- κ B by acting as an antioxidant.³⁸ Many studies suggest that catechins bind to amino acids possessed by bacteria through

hydrogen bonds. EGCG can provide an antibacterial effect by forming hydrogen peroxide, causing damage to bacterial cells and inhibiting the function of membrane proteins in the cell envelope.³⁹

CONCLUSION

From this narrative review can be concluded that mucoadhesive gingiva patch with Epigallocatechin-3-gallate (EGCG) green tea potentially reduces macrophage and lymphocyte cells in periodontitis.

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