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REVIEW

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Cranberry Proanthocyanidins: Natural Weapons against Periodontal Diseases†

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ABSTRACT: Cranberry (*Vaccinium macrocarpon*) is known to have a beneficial effect on several aspects of human health. Proanthocyanidins (PACs), the most abundant flavonoids extracted from red cranberry fruits, have been reported to possess antimicrobial, antiadhesion, antioxidant, and anti-inflammatory properties. Recent in vitro studies have shown that cranberry PACs may be potential therapeutic agents for the prevention and management of periodontitis, an inflammatory disease of bacterial origin affecting tooth-supporting tissues. After presenting an overview of cranberry phytochemicals and their potential for human health benefits, this review will focus on the effects of cranberry PACs on connective tissue breakdown and alveolar bone destruction, as well as their potential for controlling periodontal diseases. Possible mechanisms of action of cranberry PACs include the inhibition of (i) bacterial and host-derived proteolytic enzymes, (ii) host inflammatory response, and (iii) osteoclast differentiation and activity. Given that cranberry PACs have shown interesting properties in in vitro studies, clinical trials are warranted to better evaluate the potential of these molecules for controlling periodontal diseases.

KEYWORDS: bacteria, cranberry, cytokine, matrix metalloproteinase, periodontal disease, proanthocyanidins

INTRODUCTION

The American cranberry (*Vaccinium macrocarpon* Ait. Ericaceae) is widely consumed in the forms of juice, fresh fruits, dry fruits, and encapsulated powders. Phytochemicals in red cranberry fruit have been reported to exert various biological effects that are beneficial to human health. The efficacy of cranberry in the prevention of urinary tract infections has attracted a great deal of attention.[1,2] In recent years, investigators have brought evidence that cranberry phytochemicals may be of benefit in lowering risk factors and preventing certain types of cancers,[3] cardiovascular diseases,[4] neurological disorders,[5] and infectious diseases.[5] However, in most cases, there is little clinical evidence to support these findings. Although cranberry products are considered to be nontoxic and safe for humans,[6] concern has been expressed that cranberry components may interfere or interact with drugs. For example, it has been reported that cranberry juice may interfere with the anticoagulation properties of warfarin.[7] However, the jury is still out on whether cranberry juice affects warfarin therapy: normal serving size of cranberry juice drinks has been shown to be safe and does not affect the metabolism of warfarin,[8−10] and recent clinical trials have shown that consuming large amounts of cranberry juice does not alter the pharmacodynamics of warfarin.[11,12]

Cranberry proanthocyanidins (PACs) have shown promise for treating oral infections, especially dental caries. Cranberry PACs can inhibit the production of organic acids and the formation of biofilms by cariogenic bacteria.[13] Cranberry PACs may also be beneficial for periodontal health.[13] Periodontitis is a group of inflammatory conditions of infective etiology that lead to loss of tooth support. These diseases affect a large proportion of the population and may have systemic consequences.[14] Whereas bacteria are the primary factor in the etiology of periodontitis, an uncontrolled host immune response can lead to soft tissue destruction and alveolar bone resorption.[14] This review will present evidence to support the theory that cranberry PACs may be of value in the prevention and management of periodontitis.

ORIGIN AND PHYTOCHEMICAL COMPOSITION OF CRANBERRY

Cranberry plants are a group of evergreen dwarf shrubs or trailing vines that usually grow in the cooler regions of the northern hemisphere. Four species of cranberry have been described: *Vaccinium oxycoccos* (common cranberry or northern cranberry), *Vaccinium microcarpum* (small cranberry), *Vaccinium erythrocarpum* (southern mountain cranberry), and *Vaccinium macrocarpon* (large cranberry, American cranberry, or bearberry). This review will focus on *V. macrocarpon*, which has received the most attention in terms of its beneficial effects on human health.

The red cranberry fruit is pulpy and sour and is a rich source of various classes of potentially bioactive phenolic compounds.[5] Table 1 summarizes the major classes of phytochemicals in cranberry fruit and with which beneficial effects have been associated. Flavonoids, which are based on two parent structures (coumarin and chromone), are the most prominent phytochemicals in cranberry fruit. Three major classes of flavonoids have been identified in cranberry fruit: anthocyanins, flavonols, and flavan-3-ols.[5] The main cranberry anthocyanins, which are bound to different sugar moieties, are cyanidin, delphinidin, malvidin, perlagonidin, petunidin, and peonidin. Cranberry fruit is a major source of flavonols (kaempferol, myricetin, and quercetin), which are concentrated in the cortex. Flavonols are glycosylated,

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Table 1. Major Classes of Phytochemicals in Cranberry Fruit with Potential Beneficial Effects on Human Health

phytochemical class	examples
flavonoids	
anthocyanins	cyanidin, delphinidin, malvidin, peonidin, perlagonidin, petunidin[a]
flavonols	myricetin, quercetin, kaempferol[a]
flavan-3-ols	
monomers	catechin, epicatechin, epigallocatechin, epigallocatechin gallate
polymers (tannins)	proanthocyanidin A2
phenolic acids	benzoic acid, ellagic acid, hydroxycinnamic acid
stilbenes	resveratrol

[a] Linked to a sugar moiety.

with quercetin glycosides being the most common. Flavan-3-ols occur as aglycons of catechin, epicatechin, epigallocatechin, and epigallocatechin gallate. Cranberry contains flavan-3-ol monomers, oligomers, and polymers. The latter are also referred to as condensed nonhydrolyzable tannins or PACs. According to the USDA database,[15] PACs comprise many of the major flavonoids in cranberry.

PACs were discovered in the late 1940s by French researcher Jacques Masquelier, who assigned them the name "vitamin P" from Rusznyak, suggesting that plant flavonoids were vitamins.[16,17] PACs are widely distributed in the plant kingdom, especially in fruits, berries, nuts, seeds, trees, flowers, tubers, leaves, and vegetables.[18] The structural properties of PACs determine their bioactivity. Polyphenolic structures are responsible for the antioxidant properties, whereas neighboring hydroxyl groups are involved in metal binding. An important property of PACs is their ability to precipitate polypeptides and proteins, especially those with a high proline content.[19] PACs are flavan-3-ol polymers typically composed of 2−50 subunits. The subunits (catechin, epicatechin, epigallocatechin, epigallocatechin gallate) are most often linked via a single bond between C4 and C8 or C6 (B-type). B-type polymers are found in common food sources such as grapes and chocolate.[18] A-type polymers are less common (Figure 1) and possess at least one intermolecular bond between O7 and C2 in addition to the carbon−carbon bond. PACs isolated from cranberry are mainly composed of epicatechin subunits with at least one A-type bond.[20] It has been suggested that these A-type bonds are important for the antibacterial adhesion activity of PACs.[20]

OVERVIEW OF THE HEALTH BENEFITS OF CRANBERRY

Cranberry has received considerable attention for its putative human health benefits. Cranberry extracts as well as purified compounds may have potential for use as preventive/therapeutic agents regarding various human disorders. However, in most cases, there is little clinical evidence to support these findings.

Cancer. Cranberry phytochemicals may have anticancer properties based on their capacity to inhibit tumor cell growth. Seven flavonol glycosides (myricetin 3-α-arabinofuranoside, quercetin 3-xyloside, 3-methoxyquercetin 3-β-galactoside, 3-β-galactoside, quercetin 3-β-galactoside, quercetin 3-α-arabinofuranoside, and quercetin 3-α-rhamnopyranoside) isolated from whole fruit of

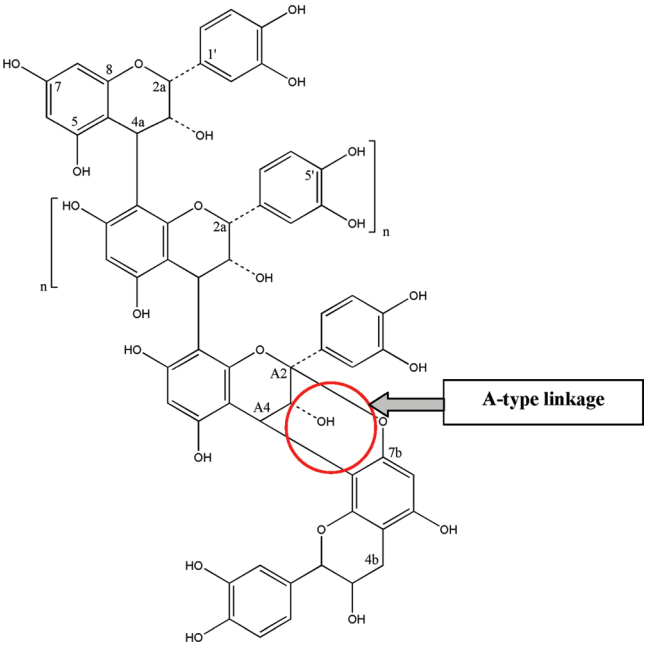

Figure 1. Structure of cranberry proanthocyanidins with A-linkage.

cranberry were shown to exhibit radical-scavenging activity.[21] Possible mechanisms of action include the induction of apoptosis in cancer cells, the reduction of invasion and metastasis by the inhibition of matrix metalloproteinase (MMP) expression and activity, and the inhibition of angiogenesis and inflammatory processes.[3,22] Flavonoids such as anthocyanins, flavonols, and proanthocyanidins; substituted cinnamic acids and stilbenes; and triterpenoids such as ursolic acid and its esters could contribute to these protective effects.[22] PAC-enriched and flavonol-enriched cranberry fractions have been reported to induce apoptosis in human prostate adenocarcinoma cells by the activation of caspases.[23]

Cardiovascular Diseases. A growing body of evidence suggests that cranberry flavonoids may decrease the risk of cardiovascular diseases by increasing the resistance of low-density lipoproteins (LDL) to oxidation, inhibiting platelet aggregation, reducing blood pressure, and inhibiting thrombosis and inflammation.[4,24] In addition, a clinical study showed that daily cranberry juice cocktail consumption is associated with an increase in plasma high-density lipoprotein (HDL)-cholesterol concentrations in abdominally obese men.[25]

Neurological Disorders. Cranberry may be of interest for treating neurological disorders such as Alzheimer's disease.[5] For example, a cranberry extract has been found to reduce a Ca^{2+} homeostasis deficit in dopamine- and amyloid-β-treated Alzheimer's disease model cells[26] and to enhance neural function, neuroprotective responses, and some motor functions in aged rats.[27] However, no clinical trials have been performed to confirm the neuroprotective effects of cranberry in humans.

Viral Infections. Several studies have investigated cranberry PACs as an alternative to currently available antiviral therapies, which are costly and only partially effective. Cranberry juice cocktail prevents the replication in monkey kidney host cells of simian rotavirus, the most common cause of diarrhea,[28] whereas high molecular weight nondialyzable material of cranberry juice completely inhibits hemagglutination.[29] In addition, Weiss et al. demonstrated that a PAC-rich cranberry fraction prevents the

adhesion of influenza virus to red blood cells by reacting with the viral hemagglutinin.[30] However, the antiviral activity of cranberry products and extracts in vivo has not been studied.

Bacterial Infections. Cranberry contains molecules that may contribute to the prevention of a number of bacterial infections, including urinary tract infections, gastric ulcers, and oral diseases. Urinary tract infections usually occur in the bladder (cystitis), renal parenchyma (pyelonephritis), or prostate (acute or chronic bacterial prostatitis).[2] *Escherichia coli* is considered to be a major etiologic agent of such infections.[2] The effect on urinary tract infections is certainly the best-known and most extensively documented health benefit of A-type cranberry PACs.[1,2] Several groups have reported that these compounds inhibit the adherence of P-fimbriated *E. coli* in in vitro models.[20,31,32]

Ulcers associated with *Helicobacter pylori* infections, which may lead to the onset of various gastric-related diseases, are a major problem in many parts of the world. Like uropathogenic *E. coli*, *H. pylori* can attach to various host surfaces via adhesins expressed on its surface. Once attached to the underlayer of epithelial cells, *H. pylori* can cause gastritis and promote the development of cancer. A PAC-rich cranberry fraction has been shown to inhibit the adherence of *H. pylori* to gastric mucus and epithelial cells, indicating that cranberry PACs may be able to prevent *H. pylori* infections.[33,34]

Dental caries is a multifactorial disease caused by specific acid-producing bacteria that are embedded in dental plaque biofilm and that ferment dietary carbohydrates such as sucrose.[35] When the pH at the surface of the tooth drops below 5.5, enamel demineralization occurs, resulting in tooth decay. Mutans streptococci such as *Streptococcus mutans* are considered to be the principal etiological agents of dental caries due to their aciduric, acidogenic, and adhesion properties.[35] PAC-containing cranberry fractions have been extensively investigated for their effect on the formation, persistence, and development of dental biofilm. The ability of cranberry PACs to prevent the sucrose-dependent biofilm formation has been attributed to their ability to inhibit the activity and production of fructosyltransferase (FTF) and glucosyltransferase (GTF), which are involved in the production of exopolysaccharides by *S. mutans*.[36] In addition, inhibition of the non-sucrose-dependent biofilm formation has been attributed to the ability of cranberry PACs to prevent bacterial coaggregation,[37] reduce bacterial hydrophobicity,[38] and alter cell surface molecules (M. Feldman, unpublished data). There is little in vivo evidence of a beneficial effect of cranberry PACs on dental caries. A preliminary human trial showed that the daily use (6 weeks) of cranberry-containing mouthwash reduces mutans streptococcal counts in saliva.[39] Cranberry PACs have also given promising results for the prevention and management of periodontal diseases. These findings will be discussed in detail in subsequent sections.

■ PERIODONTAL DISEASES

Periodontal diseases, which include gingivitis and periodontitis, are multifactorial chronic infections involving a specific group of Gram-negative anaerobic bacteria that interact with host immune cells. Gingivitis is the mildest form of periodontal disease, which manifests as red, swollen, and easily bleeding gingiva (gum). Gingivitis affects 50−90% of adults worldwide. In the United States, approximately 82% of adolescents suffer from gingivitis and gingival bleeding.[40] Although the prevalence of periodontitis is lower, 22% of American adults show signs of mild disease, whereas 13% suffer from moderate or severe forms of periodontitis. The disease is more frequent in men than women and in African- and Mexican-Americans than in Caucasians.[41] Smoking, diabetes, heredity, neutrophil dysfunction, and poor oral hygiene are significant risk factors for periodontitis.[42]

Two major etiological factors are involved in the pathogenesis of periodontitis. The first is microbial, notably the accumulation of periodontopathogenic bacteria in subgingival areas, where the toxins and proteinases produce damage to periodontal tissues.[43,44] *Porphyromonas gingivalis*, *Tannerella forsythia*, and *Treponema denticola* are associated with the chronic form of periodontitis, whereas *Aggregatibacter actinomycetemcomitans* is strongly associated with the aggressive form.[45] The second factor is the host response to periodontopathogens, notably the overproduction by resident and immune cells of inflammatory mediators (pro-inflammatory cytokines and prostanoids) and MMPs, which can modulate the progression and severity of periodontitis.[46,47]

Gingivitis can be reversed by dental treatments and adequate oral hygiene to remove plaque and calculus. If left untreated, gingivitis can progress to periodontitis, which is characterized by the formation of periodontal pockets and the destruction of tooth-supporting tissues, including connective tissue and adjacent alveolar bone. As the disease progresses, the pockets deepen and more gingival tissue and bone are destroyed, eventually leading to tooth loss.[48] Mechanical, chemical, and surgical methods are used to treat periodontal diseases. Nonsurgical conventional scaling and root-planing procedures to remove dental plaque and calculus combined with appropriate oral hygiene can help reduce tissue inflammation and pocket depth and improve clinical periodontal attachment.[49,50] Local and systemic antibiotics, anti-inflammatory drugs, and sub-antimicrobial low-dose doxycycline are also used to treat periodontitis.[51,52] In severe cases, periodontal surgery is required to provide access for the debridement of residual dental calculus and plaque, reduce the depth of periodontal pockets, and stimulate the regeneration of lost tissues by grafting with biomaterials.[14]

Recent studies have shown that there is an association between periodontal diseases and a variety of systemic complications. A systematic review and meta-analysis of seven cohort studies concluded that periodontal diseases are risk factors or markers for coronary heart disease (CHD).[53] This review estimated that there is a 24−35% increase in the risk of CHD in periodontal patients.[53] The relationship is bidirectional in regard to periodontal diseases and diabetes. Controlling periodontal diseases can help control glycemia in patients with type 2 diabetes, and improving glycemic control can contribute to a better control of periodontal diseases.[54] Bacteria colonizing the oral cavity can also have an impact on the initiation and progression of lung infections such as pneumonia and chronic obstructive pulmonary disease, particularly in hospital and nursing home patients.[55] On the basis of a systematic review, better oral hygiene and periodontal treatments using mechanical and/or topical chemical disinfection and antibiotics can reduce the incidence of nosocomial pneumonia up to 40%.[56] There may also be a correlation between periodontal diseases and the risk of preterm birth, low birth weight, and preeclampsia.[14] Repeated exposures to bacteremias and increases in inflammatory mediators during periodontal disease may trigger an inflammatory cascade in the uterus, leading to these complications.[57]

BACTERIA-MEDIATED PERIODONTAL CONNECTIVE TISSUE DESTRUCTION: TARGET 1 FOR CRANBERRY PROANTHOCYANIDINS

Bacterial attachment to host tissues is the first step toward tissue destruction. Our laboratory recently showed that cranberry PACs inhibit *P. gingivalis* adherence to human oral epithelial cells and Matrigel-coated surfaces.[58] We also showed that, although cranberry PACs do not interfere with the growth of *P. gingivalis*, they reduce bacterial biofilm formation, collagenase activity, and invasion.[58] The inhibitory effect on invasion of a basement membrane model is likely related to the anti-proteinase activity of the cranberry PACs since *P. gingivalis* proteinases may contribute to the penetration of this bacterium into the periodontium.[59] Bodet et al. demonstrated that a cranberry fraction rich in PACs dose-dependently inhibits the gingipain (both Arg- and Lys-gingipain) and dipeptidyl peptidase IV activities of *P. gingivalis*, the trypsin-like activity of *T. forsythia*, and the chymotrypsin-like activity of *T. denticola*.[60] It also blocks the ability of *P. gingivalis* to degrade native proteins, including type I collagen and transferrin.[60] This study suggested that this PAC-rich fraction has the potential to reduce the multiplication of *P. gingivalis*, *T. forsythia*, and *T. denticola* in periodontal pockets, because their growth relies on the availability of amino acids and peptides. The same fraction may also reduce the tissue destruction mediated by proteinases through its inhibitory effect on *P. gingivalis* gingipains and *T. denticola* chymotrypsin-like activities.[58,61] The toxicity of several periodontopathogen cell components toward epithelial cells may also contribute to the destruction of periodontal tissues. A PAC-rich cranberry fraction was shown to protect human oral epithelial cells from the cytotoxic effect of *Peptostreptococcus micros* cell wall components.[62] In summary, cranberry PACs can help prevent connective tissue destruction in the periodontium by inhibiting periodontopathogen attachment and invasion and by neutralizing periodontopathogen proteinases and cytotoxicity.

HOST-MEDIATED PERIODONTAL CONNECTIVE TISSUE DESTRUCTION: TARGET 2 FOR CRANBERRY PROANTHOCYANIDINS

MMPs are proteolytic enzymes released by major cell types found in the periodontium, including fibroblasts, neutrophils, and macrophages.[63] They are synthesized as latent proenzymes and are usually activated extracellularly or at the cell surface by tissue, plasma, and bacterial proteinases.[64] The enzymatic activity of MMPs is controlled by endogenous and specific tissue inhibitors called tissue inhibitor of matrix metalloproteinases (TIMPs).[65] Under normal circumstances, MMPs play a role in wound healing, angiogenesis, and gingival tissue remodeling.[66] However, when host cells are threatened by periodontal pathogens and their products such as lipopolysaccharides (LPS), MMP production increases dramatically, which disturbs the equilibrium between MMPs and TIMPs, leading to a high MMP/TIMP ratio. For example, *P. gingivalis* LPS is a relatively potent inducer of MMP-9 production by dendritic cells and a weak inducer of TIMP-1, its specific inhibitor.[67] MMPs are found in pathologically high levels in the gingival crevicular fluid and gingival tissue of periodontitis patients, leading to the suggestion that they may be used as markers of periodontal tissue destruction.[68] Because these enzymes have the capacity to degrade most components of the extracellular matrix (ECM), overexpression of MMPs leads to the destruction of periodontal tissues by the degradation of periodontal ligaments, the loss of gingival collagen, and the resorption of alveolar bone.[69] The inhibition of MMP production and activity may thus be an effective approach for treating periodontitis and other MMP-mediated diseases such as arthritis.[70] Interestingly, doxycycline, a well-documented MMP inhibitor, significantly improves periodontal healing when used as an adjunctive treatment to conventional scaling and root planning.[52] Some polyphenolic compounds have also been proposed as potential adjunctive treatments for inflammatory diseases due to their ability to inhibit MMP production and activity.[71] A PAC-enriched fraction prepared from cranberry juice inhibited both MMP-3 and MMP-9 production by gingival fibroblasts and macrophages stimulated with periodontopathogen LPS, whereas the same fraction inhibited MMP-3, MMP-9, and elastase activities, even at the low concentration tested (10 μg/mL).[72] Cranberry PACs have been shown to inhibit MMP-1, -3, -7, -8, -9, and -13 production by LPS-stimulated macrophages.[73] They are also effective in reducing the collagen and gelatin degradation activity of recombinant MMP-1 and MMP-9, respectively.[73] A recent study by our group showed that cranberry PACs inhibit the catalytic activity of *P. gingivalis* proteases,[58] which participate in MMP activation.[43] PACs also inhibit the phosphorylation of major macrophage intracellular signaling proteins induced by *A. actinomycetemcomitans* LPS, possibly by the inactivation of activator protein-1 (AP-1), which in turn reduces MMP production.[73] In addition, cranberry PACs inhibit the DNA-binding activity of NF-κB p65, another pathway involved in MMP production.[73]

Extracellular matrix metalloproteinase inducer (EMMPRIN) is a transmembrane glycoprotein expressed by various cell types.[74] *P. gingivalis* can induce the shedding of EMMPRIN from oral epithelial cells, which in turn can stimulate host cells to secrete MMPs.[75] Pretreating oral epithelial cells with cranberry PACs significantly reduces EMMPRIN shedding induced by *P. gingivalis* in a dose-dependent manner (M. Feldman and D. Grenier, unpublished data). This may contribute to reduce tissue destruction.

Macrophages and monocytes, which are found in higher numbers in active periodontal sites than in inactive sites,[76] play a crucial role in the host inflammatory response to periodontal pathogens.[77] The continuous high secretion of cytokines and chemokines such as interleukin-1β (IL-1β), interleukin-6 (IL-6), interleukin-8 (IL-8), and tumor necrosis factor-α (TNF-α) by host cells under inflammatory conditions modulates periodontal tissue destruction.[47] For example, IL-1 and TNF-α induce MMP release by gingival fibroblasts and periodontal ligament cells.[78,79] IL-1 also stimulates plasminogen activator, resulting in the production of plasmin, which in turn activates MMP production, thus aggravating periodontal tissue inflammation.[80] Previous studies have suggested that specific cytokines such as IL-1β be used as markers of the progression and severity of periodontal disease, as well as indicators of treatment outcomes.[81] Local inhibition of these pro-inflammatory mediators may be an interesting approach for controlling and reducing periodontal tissue and bone loss in periodontal diseases. A PAC-rich fraction from cranberry potently inhibits the secretion of IL-1β, IL-6, and TNF-α by macrophages stimulated with LPS from various periodontopathogens, including *A. actinomycetemcomitans*, *Fusobacterium nucleatum*, *P. gingivalis*, *T. denticola*, and *T. forsythia*.[82] This fraction also inhibits the secretion of IL-8 and chemokine (C−C motif) ligand 5 (CCL5), which play a role in directing the migration of neutrophils and monocytes to inflammation

sites.[82,83] In addition to their main function as collagen-producing cells, gingival fibroblasts, the main cell population of oral connective tissues, play an active role in modulating the inflammatory response.[84] In the presence of periodontopathogens, they can produce a wide variety of pro-inflammatory mediators.[85] A PAC-rich fraction from cranberry significantly reduces the secretion of IL-6, IL-8, and prostaglandin E_2 (PGE_2) by human gingival fibroblasts in response to *A. actinomycetemcomitans* LPS. Cyclooxygenase-2 expression by fibroblasts is also significantly reduced.[86]

■ ALVEOLAR BONE DESTRUCTION: TARGET 3 FOR CRANBERRY PROANTHOCYANIDINS

Bone resorption is the result of degradation of organic and inorganic phases controlled by mature osteoclasts. Receptor activator of nuclear factor kappa-B ligand (RANKL) and macrophage colony-stimulating factor (M-CSF) stimulate the production of preosteoclasts from hematopoietic monocyte/macrophage precursors.[87] Once activated, resorptive osteoclasts promote the dissolution of the inorganic phase of bone, exposing the organic matrix. Demineralization occurs by the acidification of the osteoclast extracellular microenvironment, which mobilizes bone minerals. The exposed organic component is then degraded by proteases such as cathepsin K and MMPs.[87]

The loss of alveolar bone is a typical hallmark of periodontitis. Gram-negative anaerobic bacteria in dental plaque can stimulate a host immune response, which in turn can lead to a destructive inflammatory process.[88] The production and release of pro-inflammatory mediators (cytokines and chemokines) propagate the inflammation throughout gingival tissues and then to the adjacent alveolar bone.[77,89] The accumulation of inflammatory cytokines enhances osteoclastogenesis by either stimulating osteoclast proliferation or promoting the differentiation and maturation of progenitor cells.[90,91]

The recognition that periodontitis involves an inflammatory component in addition to an alteration in bone metabolism has provided a new perspective on the management of the disease.[89] It has been suggested that bone loss due to periodontal diseases can be controlled by targeting the modulation of osteoclast function[91] and that this should serve as one of the therapeutic goals in preventing the progression of periodontal diseases. It was with this in mind that the potential of cranberry PACs to interfere with osteoclast maturation and function was assessed. Although cranberry PACs are not cytotoxic to osteoclast cells, they are capable of inhibiting their differentiation into bone-resorbing cells as well as decreasing MMP secretion, chemokine production, and bone resorption.[92] These observations provide support for the notion that cranberry PACs have potential as therapeutic agents for controlling the bone resorption process. Cranberry PACs are able to inhibit the maturation process of preosteoclastic cells, even in the presence of osteoclastogenesis mediators, suggesting that PACs may directly or indirectly interfere with those mediators involved in the process of osteoclastogenesis.[92] The impact of cranberry PACs on MMP production by osteoclasts is vital for inhibiting the resorption of the collagen-rich bone organic matrix,[92] which is susceptible to degradation by osteoclast-secreted proteases once the mineral scaffold has been dissolved. A significant decrease in production of helical peptide levels, a type I collagen byproduct released during bone degradation, has also been observed when mature osteoclastic cells in a human bone plate culture model are treated with cranberry PACs, which provides further evidence of the potential of these flavonoids to inhibit bone resorption.[92]

A comprehensive review of nutritional and physiological studies on polyphenols (the wider family of plant metabolites that contains the proanthocyanidins subtype) provided evidence of the ability of these compounds to interact with well-known osteoblast and osteoclast-related transcription factors such as Runx2, Osterix, AP-1, and NF-κB, and possibly with all bone morphogenetic proteins (BMP)-activated signaling pathways.[93] By interfering with these transcription factors, polyphenols and their derivatives may play a role in regulating osteoclastogenesis and bone formation. The intake of dietary flavonoids (a polyphenol subgroup that includes PACs) has also been shown to improve bone health, which confirms the positive relationship of these phytochemicals with bone physiology.[94]

Our research team is currently investigating the effects of cranberry PACs on osteoblasts and osteoblast bone-forming activity. We have observed an increase in the mineralization capacity of osteoblasts in the presence of cranberry PACs (unpublished data). A detailed investigation of the effects of PACs on osteoclasts and osteoblasts will lead to a better understanding of the mechanisms by which these plant metabolites interfere with bone homeostasis and how they can be used as preventive/therapeutic agents.

■ CONCLUSIONS

Biologically active compounds with the potential to modulate bacterial virulence and host responses are coming under considerable scrutiny because they have potential as new therapeutic agents for managing periodontal infections. Cranberry PACs are promising candidates for the development of such therapies due to their ability to inhibit periodontopathogen virulence factors and MMPs and to modulate the activities of the cells making up the periodontium. However, additional studies are required to identify the exact mechanisms by which PACs exert their beneficial properties.

It is unlikely that the consumption of cranberry juice on its own can benefit oral health given the short contact time between the oral surfaces (the teeth and gingiva) and the cranberry PACs. In addition, the sugar added to cranberry drinks and the acidity of these beverages may have a counterproductive effect by contributing to the demineralization of tooth enamel. It would be more appropriate to add purified PACs to oral hygiene products, which could then be tested for their potential for preventing oral diseases. These bioactive substances could also be applied locally to diseased periodontal sites by irrigation or by insertion of a resorbable fiber to modulate host responses by inhibiting the enzymes that destroy the ECM, reducing inflammation, and attenuating the virulence of the periodontopathogens. The use of cranberry PACs could eventually reduce the need for antibiotics and help prevent the development of bacterial resistance.

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REFERENCES

(1) Guay, D. R. Cranberry and urinary tract infections. *Drugs* **2009**, *69*, 775–807.
(2) Perez-Lopez, F. R.; Haya, J.; Chedraui, P. *Vaccinium macrocarpon*: an interesting option for women with recurrent urinary tract infections and other health benefits. *J. Obstet. Gynaecol. Res.* **2009**, *35*, 630–639.
(3) Neto, C. C. Cranberry and its phytochemicals: a review of in vitro anticancer studies. *J. Nutr.* **2007**, *137*, 186S–193S.
(4) Ruel, G.; Couillard, C. Evidences of the cardioprotective potential of fruits: the case of cranberries. *Mol. Nutr. Food Res.* **2007**, *51*, 692–701.
(5) Pappas, E.; Schaich, K. M. Phytochemicals of cranberries and cranberry products: characterization, potential health effects, and processing stability. *Crit. Rev. Food Sci. Nutr.* **2009**, *49*, 741–781.
(6) Dugoua, J. J.; Seely, D.; Perri, D.; Mills, E.; Koren, G. Safety and efficacy of cranberry (*Vaccinium macrocarpon*) during pregnancy and lactation. *Can. J. Clin. Pharmacol.* **2008**, *15*, e80–e86.
(7) Suvarna, R.; Pirmohamed, M.; Henderson, L. Possible interaction between warfarin and cranberry juice. *BMJ* **2003**, *327*, 1454.
(8) Lilja, J. J.; Backman, J. T.; Neuvonen, P. J. Effects of daily ingestion of cranberry juice on the pharmacokinetics of warfarin, tizanidine, and midazolam − probes of CYP2C9, CYP1A2, and CYP3A4. *Clin. Pharmacol. Ther.* **2007**, *81*, 833–839.
(9) Zikria, J.; Goldman, R.; Ansell, J. Cranberry juice and warfarin: when bad publicity trumps science. *Am. J. Med.* **2010**, *123*, 384–392.
(10) Hamann, G. L.; Campbell, J. D.; George, C. M. Warfarin−cranberry juice interaction. *Ann. Pharmacother.* **2011**, *45*, e17.
(11) Ansell, J.; McDonough, M.; Zhao, Y.; Harmatz, J. S.; Greenblatt, D. J. The absence of an interaction between warfarin and cranberry juice: a randomized, double-blind trial. *J. Clin. Pharmacol.* **2009**, *49*, 824–830.
(12) Mellen, C. K.; Ford, M.; Rindone, J. P. Effect of high-dose cranberry juice on the pharmacodynamics of warfarin in patients. *Br. J. Clin. Pharmacol.* **2010**, *70*, 139–142.
(13) Bonifait, L.; Grenier, D. Cranberry polyphenols: potential benefits for dental caries and periodontal disease. *J. Can. Dent. Assoc.* **2010**, *76*, a130.
(14) Pihlstrom, B. L.; Michalowicz, B. S.; Johnson, N. W. Periodontal diseases. *Lancet* **2005**, *366*, 1809–1820.
(15) USDA-ARS National Nutrient Database for Standard Reference (published in National Agricultural Library), 2004; p 17.
(16) Masquelier, J.; Michaud, J.; Laparra, J.; Dumon, M. C. [Flavonoids and pycnogenols]. *Int. J. Vitam. Nutr. Res.* **1979**, *49*, 307–311.
(17) Rusznyak, S. T.; Szentgyargyi, A. Vitamin P: flavonols as vitamins. *Nature* **1936**, *138*, 27.
(18) Gu, L.; Kelm, M. A.; Hammerstone, J. F.; Beecher, G.; Holden, J.; Haytowitz, D.; Gebhardt, S.; Prior, R. L. Concentrations of proanthocyanidins in common foods and estimations of normal consumption. *J. Nutr.* **2004**, *134*, 613–617.
(19) Bennick, A. Interaction of plant polyphenols with salivary proteins. *Crit. Rev. Oral Biol. Med.* **2002**, *13*, 184–196.
(20) Foo, L. Y.; Lu, Y.; Howell, A. B.; Vorsa, N. The structure of cranberry proanthocyanidins which inhibit adherence of uropathogenic P-fimbriated *Escherichia coli* in vitro. *Phytochemistry* **2000**, *54*, 173–181.
(21) Yan, X.; Murphy, B. T.; Hammond, G. B.; Vinson, J. A.; Neto, C. C. Antioxidant activities and antitumor screening of extracts from cranberry fruit (*Vaccinium macrocarpon*). *J. Agric. Food Chem.* **2002**, *50*, 5844–5849.
(22) Neto, C. C. Cranberry and blueberry: evidence for protective effects against cancer and vascular diseases. *Mol. Nutr. Food Res.* **2007**, *51*, 652–664.
(23) MacLean, M. A.; Scott, B. E.; Deziel, B. A.; Nunnelley, M. C.; Liberty, A. M.; Gottschall-Pass, K. T.; Neto, C. C.; Hurta, R. A. North American cranberry (*Vaccinium macrocarpon*) stimulates apoptotic pathways in DU145 human prostate cancer cells in vitro. *Nutr. Cancer* **2011**, *63*, 109–120.
(24) McKay, D. L.; Blumberg, J. B. Cranberries (*Vaccinium macrocarpon*) and cardiovascular disease risk factors. *Nutr. Rev.* **2007**, *65*, 490–502.
(25) Ruel, G.; Pomerleau, S.; Couture, P.; Lemieux, S.; Lamarche, B.; Couillard, C. Favourable impact of low-calorie cranberry juice consumption on plasma HDL-cholesterol concentrations in men. *Br. J. Nutr.* **2006**, *96*, 357–364.
(26) Joseph, J. A.; Fisher, D. R.; Carey, A. N. Fruit extracts antagonize Aβ- or DA-induced deficits in Ca^{2+} flux in M1-transfected COS-7 cells. *J. Alzheimers Dis.* **2004**, *6*, 403–411 (discussion 443−409).
(27) Shukitt-Hale, B.; Galli, R. L.; Meterko, V.; Carey, A.; Bielinski, D. F.; McGhie, T.; Joseph, J. A. Dietary supplementation with fruit polyphenolics ameliorates age-related deficits in behavior and neuronal markers of inflammation and oxidative stress. *Age* **2005**, *27*, 49–57.
(28) Lipson, S. M.; Sethi, L.; Cohen, P.; Gordon, R. E.; Tan, I. P.; Burdowski, A.; Stotzky, G. Antiviral effects on bacteriophages and rotavirus by cranberry juice. *Phytomedicine* **2007**, *14*, 23–30.
(29) Lipson, S. M.; Cohen, P.; Zhou, J.; Burdowski, A.; Stotzky, G. Cranberry cocktail juice, cranberry concentrates, and proanthocyanidins reduce reovirus infectivity titers in African green monkey kidney epithelial cell cultures. *Mol. Nutr. Food Res.* **2007**, *51*, 752–758.
(30) Weiss, E. I.; Houri-Haddad, Y.; Greenbaum, E.; Hochman, N.; Ofek, I.; Zakay-Rones, Z. Cranberry juice constituents affect influenza virus adhesion and infectivity. *Antiviral Res.* **2005**, *66*, 9–12.
(31) Howell, A. B.; Reed, J. D.; Krueger, C. G.; Winterbottom, R.; Cunningham, D. G.; Leahy, M. A-type cranberry proanthocyanidins and uropathogenic bacterial anti-adhesion activity. *Phytochemistry* **2005**, *66*, 2281–2291.
(32) Howell, A. B. Bioactive compounds in cranberries and their role in prevention of urinary tract infections. *Mol. Nutr. Food Res.* **2007**, *51*, 732–737.
(33) Burger, O.; Ofek, I.; Tabak, M.; Weiss, E. I.; Sharon, N.; Neeman, I. A high molecular mass constituent of cranberry juice inhibits *Helicobacter pylori* adhesion to human gastric mucus. *FEMS Immunol. Med. Microbiol.* **2000**, *29*, 295–301.
(34) Shmuely, H.; Burger, O.; Neeman, I.; Yahav, J.; Samra, Z.; Niv, Y.; Sharon, N.; Weiss, E.; Athamna, A.; Tabak, M.; Ofek, I. Susceptibility of *Helicobacter pylori* isolates to the antiadhesion activity of a high-molecular-weight constituent of cranberry. *Diagn. Microbiol. Infect. Dis.* **2004**, *50*, 231–235.
(35) Islam, B.; Khan, S. N.; Khan, A. U. Dental caries: from infection to prevention. *Med. Sci. Monit.* **2007**, *13*, RA196–RA203.
(36) Steinberg, D.; Feldman, M.; Ofek, I.; Weiss, E. I. Effect of a high-molecular-weight component of cranberry on constituents of dental biofilm. *J. Antimicrob. Chemother.* **2004**, *54*, 86–89.
(37) Weiss, E. I.; Lev-Dor, R.; Kashamn, Y.; Goldhar, J.; Sharon, N.; Ofek, I. Inhibiting interspecies coaggregation of plaque bacteria with a cranberry juice constituent [published errata appear in *J. Am. Dent. Assoc.* **1999**, *130* (1), 36; and **1999**, *130* (3), 332]. *J. Am. Dent. Assoc.* **1998**, *129*, 1719–1723.
(38) Yamanaka, A.; Kimizuka, R.; Kato, T.; Okuda, K. Inhibitory effects of cranberry juice on attachment of oral streptococci and biofilm formation. *Oral Microbiol. Immunol.* **2004**, *19*, 150–154.
(39) Weiss, E. I.; Kozlovsky, A.; Steinberg, D.; Lev-Dor, R.; Bar Ness Greenstein, R.; Feldman, M.; Sharon, N.; Ofek, I. A high molecular mass cranberry constituent reduces mutans streptococci level in saliva and inhibits in vitro adhesion to hydroxyapatite. *FEMS Microbiol. Lett.* **2004**, *232*, 89–92.
(40) Albandar, J. M.; Rams, T. E. Global epidemiology of periodontal diseases: an overview. *Periodontol 2000* **2002**, *29*, 7–10.
(41) Albandar, J. M.; Brunelle, J. A.; Kingman, A. Destructive periodontal disease in adults 30 years of age and older in the United States, 1988−1994. *J. Periodontol.* **1999**, *70*, 13–29.

(42) Stabholz, A.; Soskolne, W. A.; Shapira, L. Genetic and environmental risk factors for chronic periodontitis and aggressive periodontitis. *Periodontol 2000* **2010**, *53*, 138–153.

(43) Eley, B. M.; Cox, S. W. Proteolytic and hydrolytic enzymes from putative periodontal pathogens: characterization, molecular genetics, effects on host defenses and tissues and detection in gingival crevice fluid. *Periodontol 2000* **2003**, *31*, 105–124.

(44) O'Brien-Simpson, N. M.; Veith, P. D.; Dashper, S. G.; Reynolds, E. C. Antigens of bacteria associated with periodontitis. *Periodontol 2000* **2004**, *35*, 101–134.

(45) Feng, Z.; Weinberg, A. Role of bacteria in health and disease of periodontal tissues. *Periodontol 2000* **2006**, *40*, 50–76.

(46) Offenbacher, S.; Heasman, P. A.; Collins, J. G. Modulation of host PGE_2 secretion as a determinant of periodontal disease expression. *J. Periodontol.* **1993**, *64*, 432–444.

(47) Okada, H.; Murakami, S. Cytokine expression in periodontal health and disease. *Crit. Rev. Oral Biol. Med.* **1998**, *9*, 248–266.

(48) The American Academy of Periodontology. Epidemiology of periodontal diseases. *J. Periodontol.* **1996**, *67*, 935−945.

(49) Cobb, C. M. Clinical significance of non-surgical periodontal therapy: an evidence-based perspective of scaling and root planing. *J. Clin. Periodontol.* **2002**, *29* (Suppl. 2), 6–16.

(50) Suvan, J. E. Effectiveness of mechanical nonsurgical pocket therapy. *Periodontol 2000* **2005**, *37*, 48–71.

(51) Haffajee, A. D.; Socransky, S. S.; Gunsolley, J. C. Systemic antiinfective periodontal therapy. A systematic review. *Ann. Periodontol.* **2003**, *8*, 115–181.

(52) Preshaw, P. M.; Hefti, A. F.; Novak, M. J.; Michalowicz, B. S.; Pihlstrom, B. L.; Schoor, R.; Trummel, C. L.; Dean, J.; Van Dyke, T. E.; Walker, C. B.; Bradshaw, M. H. Subantimicrobial dose doxycycline enhances the efficacy of scaling and root planing in chronic periodontitis: a multicenter trial. *J. Periodontol.* **2004**, *75*, 1068–1076.

(53) Humphrey, L. L.; Fu, R.; Buckley, D. I.; Freeman, M.; Helfand, M. Periodontal disease and coronary heart disease incidence: a systematic review and meta-analysis. *J. Gen. Intern. Med.* **2008**, *23*, 2079–2086.

(54) Bascones-Martinez, A.; Matesanz-Perez, P.; Escribano-Bermejo, M.; Gonzalez-Moles, M. A.; Bascones-Ilundain, J.; Meurman, J. H. Periodontal disease and diabetes − review of the literature. *Med. Oral Patol. Oral Cir. Bucal* **2011**, *16* (6), e722–e729.

(55) Scannapieco, F. A.; Rethman, M. P. The relationship between periodontal diseases and respiratory diseases. *Dent. Today* **2003**, *22*, 79–83.

(56) Scannapieco, F. A.; Bush, R. B.; Paju, S. Associations between periodontal disease and risk for nosocomial bacterial pneumonia and chronic obstructive pulmonary disease. A systematic review. *Ann. Periodontol.* **2003**, *8*, 54–69.

(57) Gibbs, R. S. The relationship between infections and adverse pregnancy outcomes: an overview. *Ann. Periodontol.* **2001**, *6*, 153–163.

(58) La, V. D.; Howell, A. B.; Grenier, D. Anti-*Porphyromonas gingivalis* and anti-inflammatory activities of zA-type cranberry proanthocyanidins. *Antimicrob. Agents Chemother.* **2010**, *54*, 1778–1784.

(59) Andrian, E.; Grenier, D.; Rouabhia, M. *In vitro* models of tissue penetration and destruction by *Porphyromonas gingivalis*. *Infect. Immun.* **2004**, *72*, 4689–4698.

(60) Bodet, C.; Piche, M.; Chandad, F.; Grenier, D. Inhibition of periodontopathogen-derived proteolytic enzymes by a high-molecular-weight fraction isolated from cranberry. *J. Antimicrob. Chemother.* **2006**, *57*, 685–690.

(61) Yamanaka, A.; Kouchi, T.; Kasai, K.; Kato, T.; Ishihara, Y.; Okuda, K. Inhibitory effect of cranberry polyphenol on proteases of periodontopathic bacteria. *J. Dent. Res.* **2006**, *86*, Abstract 1078.

(62) La, V. D.; Labrecque, J.; Grenier, D. Cytoprotective effect of proanthocyanidin-rich cranberry fraction against bacterial cell wall-mediated toxicity in macrophages and epithelial cells. *Phytother. Res.* **2009**, *23*, 1449–1452.

(63) Sorsa, T.; Tjaderhane, L.; Salo, T. Matrix metalloproteinases (MMPs) in oral diseases. *Oral Dis.* **2004**, *10*, 311–318.

(64) Nagase, H. Activation mechanisms of matrix metalloproteinases. *Biol. Chem.* **1997**, *378*, 151–160.

(65) Kubota, T.; Nomura, T.; Takahashi, T.; Hara, K. Expression of mRNA for matrix metalloproteinases and tissue inhibitors of metalloproteinases in periodontitis-affected human gingival tissue. *Arch. Oral Biol.* **1996**, *41*, 253–262.

(66) Hannas, A. R.; Pereira, J. C.; Granjeiro, J. M.; Tjaderhane, L. The role of matrix metalloproteinases in the oral environment. *Acta Odontol. Scand.* **2007**, *65*, 1–13.

(67) Jotwani, R.; Eswaran, S. V.; Moonga, S.; Cutler, C. W. MMP-9/TIMP-1 imbalance induced in human dendritic cells by *Porphyromonas gingivalis*. *FEMS Immunol. Med. Microbiol.* **2010**, *58*, 314–321.

(68) Soell, M.; Elkaim, R.; Tenenbaum, H. Cathepsin C, matrix metalloproteinases, and their tissue inhibitors in gingiva and gingival crevicular fluid from periodontitis-affected patients. *J. Dent. Res.* **2002**, *81*, 174–178.

(69) Birkedal-Hansen, H. Role of matrix metalloproteinases in human periodontal diseases. *J. Periodontol.* **1993**, *64*, 474–484.

(70) Ramamurthy, N. S.; Xu, J. W.; Bird, J.; Baxter, A.; Bhogal, R.; Wills, R.; Watson, B.; Owen, D.; Wolff, M.; Greenwald, R. A. Inhibition of alveolar bone loss by matrix metalloproteinase inhibitors in experimental periodontal disease. *J. Periodontal Res.* **2002**, *37*, 1–7.

(71) Bellosta, S.; Dell'Agli, M.; Canavesi, M.; Mitro, N.; Monetti, M.; Crestani, M.; Verotta, L.; Fuzzati, N.; Bernini, F.; Bosisio, E. Inhibition of metalloproteinase-9 activity and gene expression by polyphenolic compounds isolated from the bark of *Tristaniopsis calobuxus* (Myrtaceae). *Cell. Mol. Life Sci.* **2003**, *60*, 1440–1448.

(72) Bodet, C.; Chandad, F.; Grenier, D. Inhibition of host extracellular matrix destructive enzyme production and activity by a high-molecular-weight cranberry fraction. *J. Periodontal Res.* **2007**, *42*, 159–168.

(73) La, V. D.; Howell, A. B.; Grenier, D. Cranberry proanthocyanidins inhibit MMP production and activity. *J. Dent. Res.* **2009**, *88*, 627–632.

(74) Gabison, E. E.; Hoang-Xuan, T.; Mauviel, A.; Menashi, S. EMMPRIN/CD147, an MMP modulator in cancer, development and tissue repair. *Biochimie* **2005**, *87*, 361–368.

(75) Feldman, M.; La, V. D.; Bedran, T.; Spolidorio, D.; Grenier, D. *Porphyromonas gingivalis*-mediated shedding of extracellular matrix metalloproteinase inducer (EMMPRIN) by oral epithelial cells: a potential role in inflammatory periodontal disease. *Microbes Infect.* **2011**, *13*, 1261–1269.

(76) Zappa, U.; Reinking-Zappa, M.; Graf, H.; Espeland, M. Cell populations and episodic periodontal attachment loss in humans. *J. Clin. Periodontol.* **1991**, *18*, 508–515.

(77) Kornman, K. S.; Page, R. C.; Tonetti, M. S. The host response to the microbial challenge in periodontitis: assembling the players. *Periodontol 2000* **1997**, *14*, 33–53.

(78) Meikle, M. C.; Atkinson, S. J.; Ward, R. V.; Murphy, G.; Reynolds, J. J. Gingival fibroblasts degrade type I collagen films when stimulated with tumor necrosis factor and interleukin 1: evidence that breakdown is mediated by metalloproteinases. *J. Periodontal Res.* **1989**, *24*, 207–213.

(79) Richards, D.; Rutherford, R. B. Interleukin-1 regulation of procollagenase mRNA and protein in periodontal fibroblasts in vitro. *J. Periodontal Res.* **1990**, *25*, 222–229.

(80) Mochan, E.; Armor, L.; Sporer, R. Interleukin 1 stimulation of plasminogen activator production in cultured gingival fibroblasts. *J. Periodontal Res.* **1988**, *23*, 28–32.

(81) Hou, L. T.; Liu, C. M.; Rossomando, E. F. Crevicular interleukin-1 β in moderate and severe periodontitis patients and the effect of phase I periodontal treatment. *J. Clin. Periodontol.* **1995**, *22*, 162–167.

(82) Bodet, C.; Chandad, F.; Grenier, D. Anti-inflammatory activity of a high-molecular-weight cranberry fraction on macrophages stimulated by lipopolysaccharides from periodontopathogens. *J. Dent. Res.* **2006**, *85*, 235–239.

(83) Luster, A. D. Chemokines − chemotactic cytokines that mediate inflammation. *N. Engl. J. Med.* **1998**, *338*, 436–445.

(84) Wassenaar, A.; Snijders, A.; Abraham-Inpijn, L.; Kapsenberg, M. L.; Kievits, F. Antigen-presenting properties of gingival fibroblasts in chronic adult periodontitis. *Clin. Exp. Immunol.* **1997**, *110*, 277–284.

(85) Dongari-Bagtzoglou, A. I.; Ebersole, J. L. Production of inflammatory mediators and cytokines by human gingival fibroblasts following bacterial challenge. *J Periodontal Res* **1996**, *31*, 90–98.

(86) Bodet, C.; Chandad, F.; Grenier, D. Cranberry components inhibit interleukin-6, interleukin-8, and prostaglandin E2 production by lipopolysaccharide-activated gingival fibroblasts. *Eur. J. Oral Sci.* **2007**, *115*, 64–70.

(87) Teitelbaum, S. L. Bone resorption by osteoclasts. *Science* **2000**, *289*, 1504–1508.

(88) Bodet, C.; Chandad, F.; Grenier, D. [Pathogenic potential of *Porphyromonas gingivalis*, *Treponema denticola* and *Tannerella forsythia*, the red bacterial complex associated with periodontitis]. *Pathol. Biol. (Paris)* **2007**, *55*, 154–162.

(89) Cochran, D. L. Inflammation and bone loss in periodontal disease. *J. Periodontol.* **2008**, *79*, 1569–1576.

(90) Birkedal-Hansen, H. Role of cytokines and inflammatory mediators in tissue destruction. *J. Periodontal Res.* **1993**, *28*, 500–510.

(91) McCauley, L. K.; Nohutcu, R. M. Mediators of periodontal osseous destruction and remodeling: principles and implications for diagnosis and therapy. *J. Periodontol.* **2002**, *73*, 1377–1391.

(92) Tanabe, S.; Santos, J.; La, V. D.; Howell, A. B.; Grenier, D. A-Type cranberry proanthocyanidins inhibit the RANKL-dependent differentiation and function of human osteoclasts. *Molecules* **2011**, *16*, 2365–2374.

(93) Trzeciakiewicz, A.; Habauzit, V.; Horcajada, M. N. When nutrition interacts with osteoblast function: molecular mechanisms of polyphenols. *Nutr. Res. Rev.* **2009**, *22*, 68–81.

(94) Hardcastle, A. C.; Aucott, L.; Reid, D. M.; Macdonald, H. M. Associations between dietary flavonoid intakes and bone health in a Scottish population. *J. Bone Miner. Res.* **2011**, *26*, 941–947.