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Proresolving lipid mediators: potential for prevention and treatment of periodontitis

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Abstract

Aim: Periodontitis is an inflammatory disease initiated by microbial biofilm. The host response to the biofilm destroys the periodontium mediated by an overly robust inflammatory response in susceptible individuals. Whether the excessive host response is genetic, epigenetic or mediated by environment is unknown. New pathways of resolution of inflammation have been discovered. Resolution of inflammation is an active, agonist-mediated, programmed return to tissue homeostasis.

Materials and Methods: Various computer-based search engines were employed to identify papers relevant to resolution of inflammation.

Results: Recent data suggest that chronic inflammatory periodontal disease may be a failure of resolution pathways as well as overexpression of proinflammatory pathways. In this review, the biology of resolution of inflammation will be examined in normal tissues and periodontal disease. Anti-inflammatory pharmacologic agents [non-steroidal anti-inflammatory drugs (NSAIDs)] have been shown to prevent and slow the progression of periodontitis in animals and humans. However, the side effect profile of NSAIDS or other inhibitors or receptor antagonists preclude their use in periodontal therapy.

Conclusion: The isolation and characterization of proresolving lipid mediators that are receptor agonists has opened a new area of research for potential therapeutic agents for the management of inflammatory periodontitis.

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Key words: anti-inflammatory; lipoxins; *P. gingivalis*; periodontal disease; resolution agonists; resolvins

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The topic assigned for this review for the European Workshop to be held on 7–10 November 2010 is too new to be suitable for a systematic review as there are no human studies that would be applicable to the approach. The search terms used for this review included ''resolution of inflammation'', ''periodontal disease'', ''lipoxins (LX)'' and ''resolvins'' using the PubMed database. The references selected are original reports and reviews that define the principles of the subject. Most of the experimental work describes biochemical pathways and there are no conflicting reports to the translational work described. The goal of the author was to be inclusive, rather than exclusive, in an attempt to limit bias.

Conflict of interest and source of funding statement

Boston University is assigned patents on resolvins that are licensed for clinical development and are subject to consultant agreement for Thomas E. Van Dyke. This supplement was supported by an unrestricted grant from Colgate.

The development of a pathogenic biofilm is well-documented as the primary aetiologic agent for the development of periodontal disease (Van Dyke & Serhan 2003). However, the specific bacterium or bacteria that cause periodontitis is not established (Van Dyke 2007). Although cross-sectional studies have associated certain bacteria, such as *Porphyromonas gingivalis* (*P.g.*), *Tannerella forsythia and Treponema denticola,* the ''red complex'' bacteria (Socransky & Haffajee 1994), the cause-and-effect relationship of specific microbes remains to be determined in longitudinal studies (Van Dyke 2007). However, the host-mediated response to the biofilm in the susceptible individual destroys tissue and bone of the periodontium in the pathogenesis of the disease (Van Dyke & Serhan 2003, Serhan 2005). The acute inflammatory response is protective. There are several possible outcomes that follow the acute response; elimination of the pathogen and return to homeostasis is the optimum response; however, if this fails, abscess formation, repair with scarring or chronic inflammation are less desirable endpoints (Van Dyke & Serhan 2003).

The current therapeutic approach to control inflammation is to remove

aetiology. Because the bacterial challenge is probably related to overgrowth of commensal organisms and the oral cavity cannot be sterilized, the control of inflammation in susceptible people by this method is difficult and recurrence of disease is high (Socransky & Haffajee 2002). Pharmacologic approaches to control of inflammation in periodontitis are limited by available pharmacological agents such as non-steroidal anti-inflammatory drugs (NSAIDs) that antagonize proinflammatory pathways and/or signalling (Serhan et al. 2007). The use of these drugs in periodontics is limited by their side-effect profiles (Williams et al. 1988). More recently, new pathways and processes underlying resolution of inflammation have been discovered stimulating increased interest in proresolving lipid mediators of inflammation. The realization that resolution of inflammation is an active process, rather than a passive decay of proinflammatory signals, mediated by a new class of proresolving lipid mediator receptor agonists (Arita et al. 2005a, Serhan 2005, Chiang et al. 2006, Van Dyke 2007) has sparked interest in the potential us of proresolving molecules to neutralize and eliminate inflammatory leucocytes and prevent periodontal pathology (Van Dyke & Serhan 2003, Serhan 2005, Serhan et al. 2007).

Proresolving lipid mediator pathways are the subject of this review. The proresolving molecules include LX that are produced from the metabolism of endogenous arachidonic acid (AA) and resolvins that are derived from dietary omega-3 polyunsaturated fatty acids (ω-3 PUFA) (for review, see Serhan et al. 2008). The scope of ω-3 PUFA-derived molecules continues to grow as new discoveries are made and currently includes protectins and maresins (Hong et al. 2003, Serhan et al. 2004, Serhan et al. 2009). LX are receptor agonists that control the resolution phase of acute inflammation and promote healing of the lesion (Serhan 1994, Van Dyke 2007, Serhan & Chiang 2008). The resolvins and protectins, which share similar proresolving properties to LX, are also receptor agonists (different receptors, vide infra) that promote the resolution of inflammation (Serhan et al. 2008). This newly identified family of proresolving lipid mediators represents a new approach to control of inflammation; supplement or enhance the ''off signal'' rather than attempt to inhibit the ''on signal''. Importantly, such an approach offers the possibility driving resolution with positive, agonist/receptor-mediated cellular signals potentially without the multiple side-effects ascribed to traditional anti-inflammatory antagonist-based treatments (Hasturk et al. 2007, Serhan et al. 2008).

Physiological Processes Involved in the Restoration of Tissue Homeostasis

Resolution pathways are initiated following an acute inflammatory response. Release of AA from cell membranes generates lipid mediators of inflammation including the classic eicosanoids, prostanoids (e.g., prostaglandins, thromboxanes) and prostacyclins, as well as leucotrienes (LTs) (Fig. 1). The response is initiated through numerous pathways; in the case of periodontal tissues, there is a large body of evidence implicating pattern recognition receptors and pathogen-associated molecular patterns [for a recent review, see Darveau (2010)]. Agonist stimulation of G-protein receptors in the cell membrane triggers the release of AA from phosphatidylcholine by the enzyme phospholipase A_2 (reviewed by Kantarci et al. 2003). The free AA is metabolized by cyclooxygenase-1- (COX-1) and COX-2-dependent pathways that result in the generation of prostanoids, or a lipoxygenase (LO)-dependent pathway. The three LOs are cell specific; 5-LO is expressed by myeloid cells, 12-LO is expressed by platelets, and 15-LO is expressed by endothelium and epithelium. The end products of the LOs are 5, 12 or 15 hydroxyeicosatetraenoic acid (HETE; the 5-HETE is further metabolized to LTs, specifically LTB_4 (Fig. 1) (Kantarci et al. 2003). Prostanoids and LTs produce a wide range of pathophysiological responses associated with periodontal disease, including inflammatory cell recruitment, oedema, pain, collagen destruction and bone resorption (Pouliot et al. 2000).

The switch from proinflammation to proresolution is temporal and is linked to the expression of new genes. As the acute inflammatory lesion matures, there is an accumulation of cells containing LO and corresponding proinflammatory products such as prostaglandin E_2 (PGE_2) and the HETEs. In the normal resolution programme, a ''class switch'' occurs within neutrophils (Levy et al. 2001, Van Dyke 2007) giving rise to the synthesis of proresolving molecules through spatially and temporally distinct pathways. Proresolving lipid mediators are produced from AA through cell: cell interactions where 15-S-H(p)ETE is produced by oxidation of AA by 15-LO followed by 5-LO oxidation of the same molecule. The products include LX such as lipoxin A_4 (LXA_4) and lipoxin B_4 (Fig. 1) (Serhan 1994, Van Dyke & Serhan 2003). The LX produced bind to FPRL1 (Chiang et al.

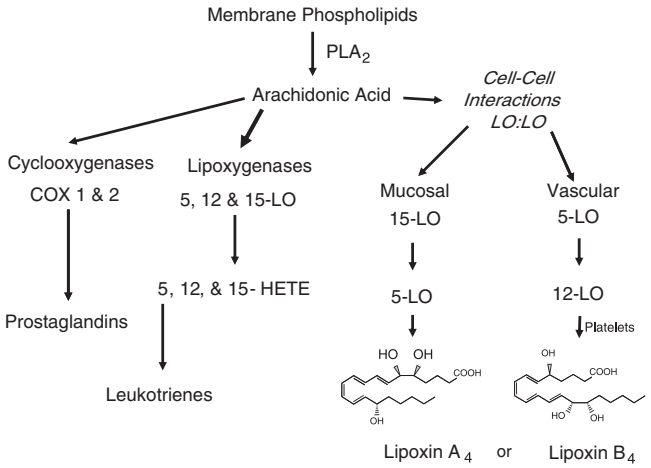

Fig. 1. Metabolism of arachidonic acid (AA) – AA is cleaved from the sn-2 position of membrane phospholipids and metabolized by cyclooxygenases (COX) or lipoxygenases (LO). The proinflammatory products are well known and include the prostaglandins from COX-1 and 2 and the leucotrienes and other hydroxyl acids (HETEs) from 5, 12 and 15-LO. Later in the inflammatory lesion, lipoxygenase interactions take place; a single AA molecule interacts with two different LOs yielding a new class of molecule called lipoxins.

2006), also known as the FMLP receptor, to stimulate the resolution of inflammation. The actions of LX include limitation of PMN migration into sites of inflammation with promotion of PMN apoptosis, and activation of monocytes to a non-phlogistic phenotype (e.g., non-inflammatory). The resulting non-phlogistic macrophage clears apoptotic PMN by phagocytosis and exhibits enhanced clearance of bacteria at mucosal surfaces (Serhan et al. 1993, Maddox & Serhan 1996, Maddox et al. 1997, Mitchell et al. 2002).

The actions of aspirin are quite distinct from other NSAIDs and aspirin plays an important role in the biology of resolution of inflammation. Aspirin has a unique property among the NSAIDs; it inactivates COX-2 by acetylation of the enzyme creating a new, active enzyme. Aspirin modified COX-2 is a 15R LO. The product of this so called ''aspirin-triggered'' pathway is 15R-H(p)ETE, which is further metabolized by PMN 5-LO to yield 15R-LXA_4, or aspirin-triggered lipoxin (ATL) (Claria & Serhan 1995). ATL are more bioactive and possess more powerful proresolving properties (Serhan et al. 1995, Claria, Lee & Serhan 1996, Van Dyke & Serhan 2003). Interestingly, statins also stimulate the production of 15R-LXA_4 through a nitrosylation reaction involving COX-2 perhaps explaining in part the anti-inflammatory actions of statins (Birnbaum et al. 2006).

While aspirin facilitated resolution pathways, other NSAIDs may actually be toxic to resolution of inflammation. The stimulus for induction of generation of LX or ATL that are triggered by proinflammatory lipid mediators such as PGE_2 may explain the potentially serious cardiovascular consequences of chronic use of selective COX-2 antagonists. Inflammation is a multifactor process, and a single, ''pan anti-inflammatory'' agent does not exist that antagonizes deleterious pathways while preserving resolution pathways (Serhan 2005, Serhan et al. 2007). As a result, the unfortunate consequence of antagonism of a single enzymatic pathway (COX-2) by NSAIDs (e.g., rofecoxib, valdecoxib and celecoxib) is the inhibition of resolution of inflammation. While inhibition of COX-2 by NSAIDs may attenuate signs of acute inflammation (pain, swelling), the reduction in PGE_2 would fail to generate proresolving LX that are required for restoring homeostasis in tissues (Levy et al. 2001, FitzGerald 2007). The resulting chronic low-grade vascular inflammation may explain the increased risk of cardiovascular disease in individuals receiving long-term treatment with selective COX-2 inhibitors (FitzGerald 2007).

The Potential of LX as Therapeutics

LX drive resolution of inflammation and restore tissue homeostasis. However, these short-lived autacoids are rapidly metabolized and unstable making them poor pharmacologic candidates in their native form. To circumvent this problem stable analogs of LX have been synthesized by total organic synthesis. Aspirin-triggered lipoxin analogs (ATLa) selectively interact with lipoxin receptors on neutrophils in vitro. The potential of ATLa as pharmacologic anti-inflammatory agents was examined in animal models of PMN-mediated tissue injury (Takano et al. 1997, Takano et al. 1998). In the mouse, intravenous injection of the proinflammatory mediator LTB_4 induces peripheral vasodilatation and leakage of blood from capillaries resulting in stasis of blood in tissues. The resulting cyanosis can be enhanced by intravenous blood dye that also leaks into the tissues rendering all exposed skin on the mouse (e.g., the ear) a distinctive blue. ATLa was topically applied to one ear and vehicle alone to the other immediately before injection of the LTB_4. The ATLa treated ear did not turn blue illustrating the potential of topically applied ATLa for preventing inflammation.

Resolution Agonists Derived from Dietary PUFA

While LX are produced from the endogenous ω-6 fatty acid substrate, AA, another family of proresolution agonists is metabolized by the same LO enzyme systems from dietary ω-3 PUFAs; the resolvins and protectins (for review, see Serhan et al. 2008). The resolvins and protectins and are derived from eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), the major ω-3 PUFA found in fish oil (Serhan et al. 2002). EPA is the substrate for resolvins of the E series and DHA the resolvin D series as well as protectins (or neuroprotectins in neural tissues) (reviewed in Van Dyke 2007, Serhan et al. 2008). Resolvins and protectins bind to distinct receptors to induce anti-inflammatory and proresolving pathways with unique mechanism of action that differs from the LX; for instance, RvE1 binds to BLT1 (an LTB_4 receptor) on neutrophils and chemR23 on monocyte/macrophages (Serhan et al. 2004, Van Dyke 2007). As with ATLa, aspirin modified COX-2 produces a class of resolvin molecule with the same increased stability and duration of action (Fig. 2) (Serhan et al. 2002).

Resolution of inflammation induced by resolvins and protectins is characterized by stopping neutrophil infiltration and driving neutrophil apoptosis, attracting non-phlogistic monocytes that differentiate into resolution macrophages. Resolution macrophages exhibit enhanced phagocytosis of apoptotic neutrophils and enhanced clearance of bacteria at mucosal surfaces promoting

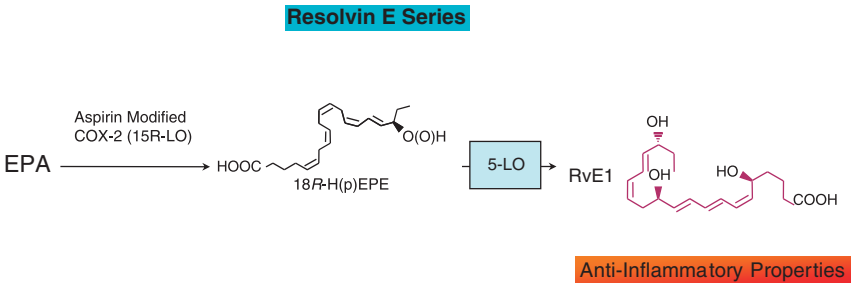

Fig. 2. Metabolism of omega-3 polyunsaturated fatty acids (ω-3 PUFA) – the same lipoxygenase (LO) enzyme system that generates lipoxins from AA metabolizes ω-3 PUFAs into resolvins and protectins. The example of Resolvin E1 (RvE1) generation is illustrated. The two major ω-3 PUFAs are EPA, which is metabolized into resolvins of the E series, and docosahexaenoic acid, which is metabolized into resolvins of the D series and the protectins. The role of aspirin-modified cyclooxygenases (COX)-2 is also indicated. In the presence of aspirin, COX-2 is acetylated and becomes a new active enzyme; a 15R-LR. Aspirin-modified COX-2 is responsible for the production of aspirin-triggered lipoxins, the 18R-resolvins of the E series and 17R-resolvins of the D series.

wound clearance and return to homeostasis (Bannenberg et al. 2005, Ariel et al. 2006, Schwab et al. 2007). The rapid resolution of acute inflammation induced by resolvins and protectins are, at least in part, the basis of the beneficial actions of dietary ω-3 PUFAs, EPA and DHA, in inflammatory diseases such as cardiovascular disease (DeCaterina et al. 1990, Albert et al. 2002).

Traditional Management of Inflammation in Periodontitis

The goal of treatment in an inflammatory disease such as periodontitis is the return of tissue homeostasis, which implies complete healing, with regeneration of lost or damaged tissue without scarring. The rapid and complete elimination of invading leucocytes from the lesion is, therefore, the ideal outcome following an inflammatory event (Van Dyke 2007). For periodontal disease, typical intervention procedures target removal of the aetiologic agents associated with inflammation to arrest further progression of periodontal disease. However, to date, therapeutic procedures do not restore tissue homeostasis; the outcome of periodontal therapy is repair with fibrosis and scarring resulting in a reduced periodontium. Periodontitis and the inability to reach homeostasis as defined above can be attributed to a failure of resolution of inflammation pathways (Van Dyke 2008).

Proresolving Therapy in an Experimental Rabbit Model of Periodontitis

To explore the potential of resolution agonists as therapeutics in periodontitis, an experimental model of *P.g.)* and ligature-induced periodontitis was established. Silk ligatures were tied bilaterally around the second premolars of both mandibular quadrants and *P.g.* was applied to ligatured sites three times a week (M, W, F) for 6 weeks (Hasturk et al. 2006). Inflammation and bone loss was induced with application of *P.g.*, which was inhibitable with systemic administration of metronidazole; ligature alone induced little inflammation and disease (Hasturk et al. 2006). Histological, morphometric and radiological evaluations revealed severe inflammation with profound loss of collagen and bone with infrabony (vertical bone loss) pockets. *P.g.*+ligature-induced $43.0 \pm 4.6\%$ bone loss over the 6-week period, whereas ligature alone resulted in $<10\%$ bone loss.

An evaluation of RvE1 for the prevention of inflammation and bone loss (periodontitis) in the model was explored (Hasturk et al. 2006). RvE1, prepared by total organic synthesis (Arita et al. 2005a, b) was topically applied directly to study teeth (4 μg/tooth, applied to the ligature with a micropipette) three-times weekly over the 6-week study period immediately after *P.g.* application. The control group received topical application of vehicle (ethanol); a third group received ligature alone (no *P.g.*, no therapy). Ligature alone did not produce much disease. However, animals receiving *P.g.*+ ligature with the ethanol as therapy exhibited marked inflammation and significant bone loss. Histology revealed a profound inflammatory infiltrate, loss of collagen and loss of bone. Osteoclasts were visible in resorption lacunae. Using RvE1 as a mono-therapy resulted in a complete absence of inflammation and complete prevention of gingivitis and periodontitis. There were no inflammatory changes, osteoclast formation, or bone loss observed. Radiographic assessment also demonstrated reduced bone loss with RvE1 treatment ($<5\%$) compared with vehicle controls ($>35\%$) or ligature alone ($\sim 12\%$) ($p<0.05$). In control experiments, it was clearly shown that resolvins have no intrinsic bactericidal activity (Hasturk et al. 2006, Hasturk et al. 2007).

Treatment of Experimental Periodontitis

Experiments were then undertaken to determine the potential for RvE1 treatment of established periodontitis (Hasturk et al. 2007). In this series of experiments, experimental periodontitis was induced for 6 weeks in three groups of rabbits as above with ligatures and topical application of *P.g.* The first group of animals was sacrificed at 6 weeks to determine the extent of periodontitis (baseline disease). In the other two groups, *P.g.* application ceased and animals received either RvE1 or vehicle treatment following the same regimen for an additional 6 weeks before sacrifice at week 12. Compared with baseline disease animals, vehicle-treated rabbits exhibited progression of disease and bone loss over the second 6 weeks of the experiment. Osteoclasts and osteoclast-like cells were clearly observed in the baseline disease and vehicle control groups. Treatment with RvE1 resulted in complete resolution of inflammation and restoration of both soft and hard tissues. There was no inflammation clinically or histologically with an absence of osteoclasts associated with bone (Fig. 3). The clinical appearance of the tissues resembled pre-disease, naive animals. There was a significant reduction in soft tissue pocket depth, bone loss and infrabony defect depth, compared with baseline disease and vehicle control groups ($p<0.01$). Histological assessment of undecalcified ground sections reveal complete regeneration of the periodontium with new cementum formation, connective tissue attachment and new bone suggesting regeneration of the natural architecture of the periodontal organ (Fig. 3) (Hasturk et al. 2007).

Inflammation and the Biofilm

The observations relating control of inflammation and progression of periodontitis, albeit in an animal model, challenge existing paradigms regarding the temporal association of biofilm composition and disease. In retrospect, the observation that pharmacologic control of inflammation can control progression of periodontitis is not new; only the agents are new. Experiments in the 1980s with flurbiprophen and the 1990's with tetracyclines clearly demonstrate the proof of principle that control of inflammation independent of plaque (biofilm) control can have a beneficial impact on the progression of periodontitis (for review, see Howell & Williams 1993). In the rabbit experiment, RvE1 treatment stimulates the resolution of inflammation and the restoration of bone loss; the temporal composition shifts in the microflora in the biofilm were also investigated (Hasturk et al. 2007). The dynamic nature of biofilm composition is well studied and significant changes in the balance of microbial species has been described (Marsh & Bradshaw 1990). A significant reduction in oral microflora due to the direct actions of RvE1 was not anticipated; previous studies failed to demonstrate significant antibacterial actions for RvE1 (Van Dyke 2007).

DNA–DNA checkerboard hybridization analyses were performed using

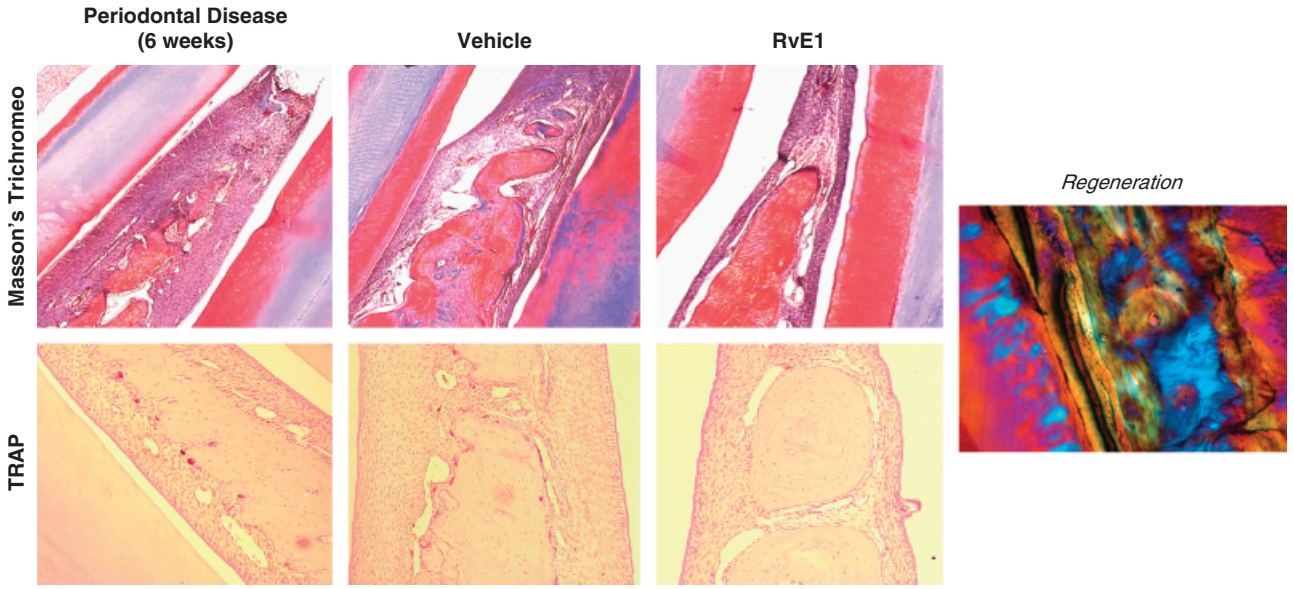

Fig. 3. Histological evaluation of treatment of rabbit periodontitis with RvE1 – representative sections from the rabbit periodontitis treatment experiment are shown; the degree of periodontal destruction after *Porphyromonas gingivalis* and ligature-induced periodontitis is illustrated in the left panels stained with Masson's Trichrome or tartrate resistant acid phosphatase (TRAP). Note the bone resorption and proliferation of osteoclastic cells. Treatment with placebo (ethanol alone for 6 weeks) illustrated in the centre panels reveals no improvement. Treatment with RvE1 for 6 weeks illustrated in the right panels reveals a restoration of the soft and hard tissues to pre-disease levels with a complete absence of clastic cells. To assess regeneration of the periodontal unit, undecalcified ground seconds viewed in polarized light (labelled *Regeneration*) reveal formation of new bundle bone, new periodontal ligament fibres inserting into newly formed cementum.

probes to human isolates. The microbial species present in the rabbit periodontal flora before ligature placement were relatively few and largely comprised *Actinomyces viscosus* and *Peptostreptococcus micros*-like organisms (Hasturk et al. 2007). Introduction of *P.g*. to the ligatures resulted in high numbers of this organism in 90% of the rabbit plaque samples that was apparent at 6 weeks and persisted to 12 weeks. A corresponding shift in the composition of the anaerobic flora occurred during this time with overgrowth of resident organisms (e.g., *A. viscosus, P. micros, Prevotella intermedia, Streptococcus intermedius and Eikenella corrodens*-like organisms*)* and the emergence of other pathogens such as *Aggregatibacter actinomycetemcomitans* (*A.a.*)-like and *Fusobacterium nucleatum*-like organisms, and an overall increase in bacterial load. Treatment with RvE1 reversed the changes in the microflora induced by development of periodontitis. *P.g* disappeared as did *A.a*. and the levels of other organisms returned to pre-disease levels.

The reason for the changes in biofilm composition with RvE1 treatment is unknown, because RvE1 has no direct antiseptic properties. A combinations of known resolution agonist properties probably comes into play; resolvin molecules promote the release of antimicrobial peptides, such as defensins and bactericidal/permeability-increasing protein (Levy et al. 2003); *P.g* is an asaccharolytic organism that requires essential amino acids as a food source derived from degradation of collagen fragments, such as those produced during an inflammatory response (Van Dyke 2007); increased bacterial clearance by resolution macrophages (Bannenberg et al. 2005). These observations suggest that an inflammatory site is an environment that favours bacterial growth. The implication is that inflammation precedes the emergence of pathogens in the periodontal pocket. This hypothesis is supported by the observations of Tanner et al. 2007 that demonstrate in a longitudinal study that gingivitis predicts future attachment loss, but specific bacteria or groups of bacteria do not.

Resolution of Inflammation in Human Periodontitis

None of the resolution agonists has, to date, been approved for clinical use, although several are in development. However, the biologic impact of dietary ω-3 PUFA on inflammation and inflammatory disease has been described (GISSI Prevenzione Investigators 1999). The question whether there is a clinical impact in periodontitis is beginning to be investigated. In a pilot trial (El-Sharkawy et al. 2010), 80 subjects with moderate to severe periodontitis were treated with either scaling and root planing followed by a regimen of 900 mg of EPA/DHA+81 mg aspirin daily for 6 months, or the same mechanical regiment and placebo tablets. Addition of the resolvin generating dietary supplement to standard periodontal therapy provided an added benefit reducing pocket depth and increasing clinical attachment, and providing measurable reductions in inflammatory mediators in saliva. These pilot data are not conclusive regarding mechanism of action, because the impact of aspirin alone and ω-3 PUFA alone was not evaluated. Nevertheless, the study provides preliminary proof of principle that the control of inflammation can have an impact on treatment of periodontitis. The mean pocket depth reduction, the usual measure of efficacy in such studies, was roughly the same magnitude as seen with topical, locally delivered antibiotics. There was a significant shift of pockets <4 mm requiring further treatment to pockets <4 mm in the treatment group compared with the placebo group.

Summary

The primary aetiology of periodontal disease is bacterial, but it appears that the host inflammatory response may be critical determinant in the pathogenesis of periodontitis. In fact, new data suggest that the inflammatory response (e.g., gingivitis) might actually precede the emergence and overgrowth of periodontal pathogens in the biofilm. Moreover, new data suggest that failure of resolution of inflammation pathways may play as important a role in disease as overproduction of proinflammatory mediators. Previously, the emphasis has been placed on prostanoids and LTs propagating the inflammatory response. A new appreciation of resolution agonists produced by previously unknown pathways such as LX, ATLa, resolvins and protectins, reveals important resolution pathways that drive restoration of tissue homeostasis. Experiments in animals and humans suggest an exciting new approach to the pharmacologic modulation of inflammation in disease; the use of receptor agonists of resolution of inflammation to actively regulate the inflammatory response. The potential is clear; resolution agonists are physiologic molecules, not inhibitors that will compromise host defense. The same lipid mediators that drive resolution of inflammation enhance microbial clearance at mucosal surfaces and improve bacterial clearance in infection (Spite et al. 2009). The potential for treating human disease with one or more of these specialized lipid mediators awaits thorough investigation.

Acknowledgements

Boston University is assigned patents on resolvins that are licensed for clinical development and are subject to consulting agreements for Dr. Van Dyke. This work was supported by USPHS Grants P50-DE16191 and DE19938.

References

GISSI Prevenzione Investigators (1999) Dietary supplementation with n-3 polyunsaturated fatty acids and vitamin E after myocardial infarction: results of the GISSI-Prevenzione trial. Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto miocardico. *Lancet* **354**, 447–455.

Albert, C. M., Campos, H., Stampfer, M. J., Ridker, P. M., Manson, J. E., Willett, W. C. & Ma, J. (2002) Blood levels of long-chain n-3 fatty acids and the risk of sudden death. *The New England Journal of Medicine* **346**, 1113–1118.

Ariel, A., Fredman, G., Sun, Y. P., Kantarci, A., Van Dyke, T. E., Luster, A. D. & Serhan, C. N. (2006) Apoptotic neutrophils and T cells sequester chemokines during immune response resolution through modulation of CCR5 expression. *Nature Immunology* **7**, 1209–1216.

Arita, M., Bianchini, F., Aliberti, J., Sher, A., Chiang, N., Hong, S., Yang, R., Petasis, N. A. & Serhan, C. N. (2005a) Stereochemical assignment, antiinflammatory properties, and receptor for the omega-3 lipid mediator resolvin E1. *The Journal of Experimental Medicine* **201**, 713–722.

Arita, M., Yoshida, M., Hong, S., Tjonahen, E., Glickman, J. N., Petasis, N. A., Blumberg, R. S. & Serhan, C. N. (2005b) Resolvin E1, an endogenous lipid mediator derived from omega-3 eicosapentaenoic acid, protects against 2,4,6-trinitrobenzene sulfonic acid-induced colitis. *Proceedings of the National Academy of Sciences of the USA* **102**, 7671–7676.

Bannenberg, G. L., Chiang, N., Ariel, A., Arita, M., Tjonahen, E., Gotlinger, K. H., Hong, S. & Serhan, C. N. (2005) Molecular circuits of resolution: formation and actions of resolvins and protectins. *Journal of Immunology* **174**, 4345–4355.

Birnbaum, Y., Ye, Y., Lin, Y., Freeberg, S. Y., Nishi, S. P., Martinez, J. D., Huang, M. H., Uretsky, B. F. & Perez-Polo, J. R. (2006) Augmentation of myocardial production of 15-epi-lipoxin-a4 by pioglitazone and atorvastatin in the rat. *Circulation* **114**, 929–935.

Chiang, N., Serhan, C. N., Dahlen, S. E., Drazen, J. M., Hay, D. W., Rovati, G. E., Shimizu, T., Yokomizo, T. & Brink, C. (2006) The lipoxin receptor ALX: potent ligand-specific and stereoselective actions in vivo. *Pharmacological Reviews* **58**, 463–487.

Claria, J., Lee, M. H. & Serhan, C. N. (1996) Aspirin-triggered lipoxins (15-epi-LX) are generated by the human lung adenocarcinoma cell line (A549)-neutrophil interactions and are potent inhibitors of cell proliferation. *Molecular Medicine* **2**, 583–596.

Claria, J. & Serhan, C. N. (1995) Aspirin triggers previously undescribed bioactive eicosanoids by human endothelial cell-leukocyte interactions. *Proceedings of the National Academy of Sciences of USA* **92**, 9475–9479.

Darveau, R. P. (2010) Periodontitis: a polymicrobial disruption of host homeostasis. *Nature Reviews Microbiology* **8**, 481–490.

DeCaterina, R., Giannessi, D., Mazzone, A., Bernini, W., Lazzerini, G., Maffei, S., Cerri, M., Salvatore, L. & Weksler, B. (1990) Vascular prostacyclin is increased in patients ingesting omega-3 polyunsaturated fatty acids before coronary artery bypass graft surgery. *Circulation* **82**, 428–438.

El-Sharkawy, H., Aboelsaad, N., Eliwa, M., Darweesh, M., Alshahat, M., Kantarci, A., Hasturk, H. & Van Dyke, T. E. (2010) Adjunctive treatment of chronic periodontitis with daily dietary supplementation with omega-3 Fatty acids and low-dose aspirin. *Journal of Periodontology* **81**, 1635–1643.

FitzGerald, G. A. (2007) COX-2 in play at the AHA and the FDA. *Trends in Pharmacology Sciences* **28**, 303–307.

Hasturk, H., Kantarci, A., Goguet-Surmenian, E., Blackwood, A., Andry, C., Serhan, C. N. & Van Dyke, T. E. (2007) Resolvin E1 regulates inflammation at the cellular and tissue level and restores tissue homeostasis in vivo. *Journal of Immunology* **179**, 7021–7029.

Hasturk, H., Kantarci, A., Ohira, T., Arita, M., Ebrahimi, N., Chiang, N., Petasis, N. A., Levy, B. D., Serhan, C. N. & Van Dyke, T. E. (2006) RvE1 protects from local inflammation and osteoclast-mediated bone destruction in periodontitis. *The Faseb Journal* **20**, 401–403.

Hong, S., Gronert, K., Devchand, P. R., Moussignac, R. L. & Serhan, C. N. (2003) Novel docosatrienes and 17S-resolvins generated from docosahexaenoic acid in murine brain, human blood, and glial cells. Autacoids in anti-inflammation. *The Journal of Biological Chemistry* **278**, 14677–14687.

Howell, T. H. & Williams, R. C. (1993) Nonsteroidal antiinflammatory drugs as inhibitors of periodontal disease progression. *Critical Review of Oral Biology and Medicine* **4**, 177–196.

Kantarci, A., Oyaizu, K. & Van Dyke, T. E. (2003) Neutrophil-mediated tissue injury in periodontal disease pathogenesis: findings from localized aggressive periodontitis. *Journal of Periodontology* **74**, 66–75.

Levy, B. D., Clish, C. B., Schmidt, B., Gronert, K. & Serhan, C. N. (2001) Lipid mediator class switching during acute inflammation: signals in resolution. *Nature Immunology* **2**, 612–619.

Levy, O., Canny, G., Serhan, C. N. & Colgan, S. P. (2003) Expression of BPI (bactericidal/permeability-increasing protein) in human mucosal epithelia. *Biochemical Society Transactions* **31**, 795–800.

Maddox, J. F., Hachicha, M., Takano, T., Petasis, N. A., Fokin, V. V. & Serhan, C. N. (1997) Lipoxin A4 stable analogs are potent mimetics that stimulate human monocytes and THP-1 cells via a G-protein-linked lipoxin A4 receptor. *Journal of Biological Chemistry* **272**, 6972–6978.

Maddox, J. F. & Serhan, C. N. (1996) Lipoxin A4 and B4 are potent stimuli for human monocyte migration and adhesion: selective inactivation by dehydrogenation and reduction. *The Journal of Experimental Medicine* **183**, 137–146.

Marsh, P. D. & Bradshaw, D. J. (1990) The effect of fluoride on the stability of oral bacterial communities in vitro. *Journal of Dental Research* **69** (Spec No), 668–671, discussion 682–663.

Mitchell, S., Thomas, G., Harvey, K., Cottell, D., Reville, K., Berlasconi, G., Petasis, N. A., Erwig, L., Rees, A. J., Savill, J., Brady, H. R. & Godson, C. (2002) Lipoxins, aspirin-triggered epi-lipoxins, lipoxin stable analogues, and the resolution of inflammation: stimulation of macrophage phagocytosis of apoptotic neutrophils in vivo. *Journal of the American Society of Nephrology* **13**, 2497–2507.

Pouliot, M., Clish, C. B., Petasis, N. A., Van Dyke, T. E. & Serhan, C. N. (2000) Lipoxin A(4) analogues inhibit leukocyte recruitment to *Porphyromonas gingivalis*: a role for cyclooxygenase-2 and lipoxins in periodontal disease. *Biochemistry* **39**, 4761–4768.

Schwab, J. M., Chiang, N., Arita, M. & Serhan, C. N. (2007) Resolvin E1 and protectin D1 activate inflammation-resolution programmes. *Nature* **447**, 869–874.

Serhan, C. N. (1994) Lipoxin biosynthesis and its impact in inflammatory and vascular events. *Biochimica et Biophysica Acta* **1212**, 1–25.

Serhan, C. N. (2005) Novel omega –3-derived local mediators in anti-inflammation and resolution. *Pharmacology and Therapeutics* **105**, 7–21.

Serhan, C. N., Brain, S. D., Buckley, C. D., Gilroy, D. W., Haslett, C., O'Neill, L. A., Perretti, M., Rossi, A. G. & Wallace, J. L. (2007) Resolution of inflammation: state of the art, definitions and terms. *The Faseb Journal* **21**, 325–332.

Serhan, C. N. & Chiang, N. (2008) Endogenous pro-resolving and anti-inflammatory lipid mediators: a new pharmacologic genus. *British Journal of Pharmacology* **153** (Suppl. 1), S200–215.

Serhan, C. N., Chiang, N. & Van Dyke, T. E. (2008) Resolving inflammation: dual anti-inflammatory and pro-resolution lipid mediators. *Nature Reviews Immunology* **8**, 349–361.

Serhan, C. N., Fiore, S., Brezinski, D. A. & Lynch, S. (1993) Lipoxin A4 metabolism by differentiated HL-60 cells and human monocytes: conversion to

novel 15-oxo and dihydro products. *Biochemistry* **32**, 6313–6319.

Serhan, C. N., Gotlinger, K., Hong, S. & Arita, M. (2004) Resolvins, docosatrienes, and neuroprotectins, novel omega-3-derived mediators, and their aspirin-triggered endogenous epimers: an overview of their protective roles in catabasis. *Prostaglandins and Other Lipid Mediators* **73**, 155–172.

Serhan, C. N., Hong, S., Gronert, K., Colgan, S. P., Devchand, P. R., Mirick, G. & Moussignac, R. L. (2002) Resolvins: a family of bioactive products of omega-3 fatty acid transformation circuits initiated by aspirin treatment that counter proinflammation signals. *The Journal of Experimental Medicine* **196**, 1025–1037.

Serhan, C. N., Maddox, J. F., Petasis, N. A., Akritopoulou-Zanze, I., Papayianni, A., Brady, H. R., Colgan, S. P. & Madara, J. L. (1995) Design of lipoxin A4 stable analogs that block transmigration and adhesion of human neutrophils. *Biochemistry* **34**, 14609–14615.

Serhan, C. N., Yang, R., Martinod, K., Kasuga, K., Pillai, P. S., Porter, T. F., Oh, S. F. & Spite, M. (2009) Maresins: novel macrophage mediators with potent antiinflammatory and proresolving actions. *The Journal of Experimental Medicine* **206**, 15–23.

Socransky, S. S. & Haffajee, A. D. (1994) Evidence of bacterial etiology: a historical perspective. *Periodontology 2000* **5**, 7–25.

Socransky, S. S. & Haffajee, A. D. (2002) Dental biofilms: difficult therapeutic targets. *Periodontology 2000* **28**, 12–55.

Spite, M., Norling, L. V., Summers, L., Yang, R., Cooper, D., Petasis, N. A., Flower, R. J., Perretti, M. & Serhan, C. N. (2009) Resolvin D2 is a potent regulator of leukocytes and controls microbial sepsis. *Nature* **461**, 1287–1291.

Takano, T., Clish, C. B., Gronert, K., Petasis, N. & Serhan, C. N. (1998) Neutrophil-mediated changes in vascular permeability are inhibited by topical application of aspirin-triggered 15-epi-lipoxin A4 and novel lipoxin B4 stable analogues. *Journal of Clinical Investigation* **101**, 819–826.

Takano, T., Fiore, S., Maddox, J. F., Brady, H. R., Petasis, N. A. & Serhan, C. N. (1997) Aspirin-triggered 15-epi-lipoxin A4 (LXA4) and LXA4 stable analogues are potent inhibitors of acute inflammation: evidence for anti-inflammatory receptors. *Journal of Experimental Medicine* **185**, 1693–1704.

Tanner, A. C., Kent, R. Jr., Kanasi, E., Lu, S. C., Paster, B. J., Sonis, S. T., Murray, L. A. & Van Dyke, T. E. (2007) Clinical characteristics and microbiota of progressing slight chronic periodontitis in adults. *Journal of Clinical Periodontology* **34**, 917–930.

Van Dyke, T. E. (2007) Control of inflammation and periodontitis. *Periodontology 2000* **45**, 158–166.

Van Dyke, T. E. (2008) The management of inflammation in periodontal disease. *Journal of Periodontology* **79**, 1601–1608.

Van Dyke, T. E. & Serhan, C. N. (2003) Resolution of inflammation: a new paradigm for the pathogenesis of periodontal diseases. *Journal of Dental Research* **82**, 82–90.

Williams, R. C., Jeffcoat, M. K., Howell, T. H., Reddy, M. S., Johnson, H. G., Hall, C. M. & Goldhaber, P. (1988) Topical flurbiprofen treatment of periodontitis in beagles. *Journal of Periodontal Research* **23**, 166–169.

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Clinical Relevance

Scientific rationale for study: This review was undertaken as part of a European Workshop.

Principle findings and practical implications: The principle findings are that pharmacotherapeutics aimed at modulating resolution of inflammation pathways offer promise as periodontal therapeutics in translational studies and suggest that a new paradigm for periodontal therapy may emerge in the future.