Fine-tuning inflammation-resolution programs: focus on atherosclerosis

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Purpose of review
Nonresolving inflammation is now considered the underpinning of several prevalent human diseases, including atherosclerosis. The resolution of inflammation is a highly coordinated program to counterbalance proinflammatory signals for a swift return to tissue homeostasis. This process is controlled in part by endogenous specialized proresolving lipid mediators (SPMs). Emerging evidence has revealed that the balance of SPMs and proinflammatory mediators during acute inflammation regulates the duration of the inflammatory response and the timing of tissue resolution. Moreover, an imbalance between SPMs and proinflammatory mediators has been linked to several prevalent chronic inflammatory diseases in humans, including atherosclerosis.

Recent findings
Lipid mediator imbalances have recently been linked to atherosclerotic plaque instability. Administration of key SPMs restored this imbalance and led to plaque stability. SPMs have also recently been shown to be protective in other cardiovascular disease models including myocardial infarction, stroke and neointimal hyperplasia.

Summary
The current review highlights recent work that supports the concept of dysregulated inflammation-resolution in atherosclerosis with a particular focus on mechanisms and therapeutic opportunities associated with SPM receptors and lipid mediator imbalances. This article is based on experimental studies.

Keywords
atherosclerosis, inflammation-resolution, specialized proresolving mediators

INTRODUCTION

Atherosclerosis is driven by the persistent subendothelial retention of Apolipoprotein B lipoproteins and is widely regarded as a condition in which immune cells ultimately cause damage to the vessel wall [1–7]. This damage is in part attributed to impaired clearance of dead cells, persistence of inflammation and failed resolution programs [4,8,9]. The current therapy for atherosclerosis is LDL cholesterol–lowering by drugs like statins and is beneficial for a wide range of patients [10]. However, some statin-treated patients continue to suffer from life-threatening vascular events (an issue referred to as 'residual risk' in the clinical literature) [10], and thus cardiovascular diseases (CVD) still remain the leading cause of death worldwide. This concept of 'residual risk' raises the question as to what is needed to better manage this disease [10]. One aspect of reducing this risk is to aggressively lower LDL, as is the case with PCSK9 inhibitors [10]. The other risk to consider is the inflammatory component of the disease [1,8,10–12]. Ongoing trials like Cardiovascular Inflammation Reduction Trial (CIRT)and Canakinumab Anti-inflammatory Thrombosis Outcomes Study (CANTOS) will better aid in our understanding of how certain aspects of the inflammatory component of this disease can be regulated [13,14]. This review will highlight new mechanisms and therapeutic strategies that not only temper inflammation but also enhance tissue repair [15,16]. In this regard, tissue reparative processes are largely accomplished through a highly

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KEY POINTS

- Impaired inflammation-resolution is the underpinning of several prevalent human diseases, including atherosclerosis.
- Imbalances in the ratio of SPMs : proinflammatory mediators drive atherosclerosis progression.
- Stimulating inflammation-resolution with the use of proresolving ligands promotes plaque stability and is thus protective in experimental atherosclerosis.
- A better understanding as to how individuals can mount and resolve acute inflammatory responses will likely yield important insight into the pathological basis of several nonresolving diseases.

coordinated program called the resolution of inflammation (or inflammation-resolution) [15,17▪▪]. Atherosclerotic lesions are like nonresolving vascular wounds [1]; therefore, understanding how to boost inflammation-resolution and thus tissue reparative processes may yield very important findings for the development of future therapeutics [4,5,9,18].

INFLAMMATION-RESOLUTION IS A PROTECTIVE RESPONSE

The acute inflammatory response, when coupled with a timely resolution program, is a protective process that is necessary to neutralize pathogens and repair injury [19]. Uncontrolled or nonresolving inflammation can lead to unintended host-mediated tissue damage [15,20]. In this regard, a failure of the inflammation-resolution response is now thought to be the underpinning of numerous chronic inflammatory diseases [15]. Inflammation-resolution is controlled by several endogenous mediators that include specialized proresolving mediators (SPMs) such as lipoxins, resolvins, protectins and maresins [15] and other lipid mediators like 15-deoxy-delta-12,14-prostaglandin J2 (15d-PGJ2) [19]; protein/peptide mediators such as annexin A1 [21,22] and IL-10; and gases such as carbon monoxide and hydrogen sulfide [23]. SPMs each have distinct chemical structures and bind/activate select G-protein coupled receptors (GPCRs) [15]. Significantly, SPMs are actively synthesized at the onset of acute inflammation to counteract proinflammatory signals [15]. How exactly SPMs act at the receptor level to counterbalance proinflammatory signals *in vivo* is largely unknown, and recent key initial findings underlying this concept will be highlighted below [24].

Emerging evidence has revealed that the balance of SPMs and proinflammatory mediators during acute inflammation regulates the duration of the inflammatory response and the timing of resolution [25–27]. Moreover, an imbalance between SPMs and proinflammatory mediators has been linked to a number of diverse chronic inflammatory diseases in humans, including, chronic obstructive pulmonary disease, asthma [28], aggressive periodontitis [29] and atherosclerosis [30,31▪]. In this regard, it is no surprise that administration of SPMs are protective in several disease models including sepsis, asthma, periodontitis, arthritis, injury-induced neointimal hyperplasia, myocardial infarction, stroke and atherosclerosis [9,31▪,32▪▪,33▪,34], to name a few [5,15,17▪▪].

SPECIALIZED PRORESOLVING LIPID MEDIATORS ARE PROTECTIVE IN EXPERIMENTAL ATHEROSCLEROSIS

SPMs are agonists that bind and activate specific GPCRs [15]. Deletion of key SPM receptors such as lipoxin receptor/formyl peptide receptor 2 (ALX/FPR2) or G-protein coupled receptor 18 (GPR18) leads to exacerbated inflammation and defective resolution [18,35,36]. These findings provide evidence that SPMs are essential endogenous mediators of the inflammation-resolution response. With regard to atherosclerosis, engagement of the ALX/FPR2 receptor with its proresolving ligands (Annexin A1 or Ac2–26) led to improved plaque stability [37▪] and decreased atherogenesis [38]. In the absence of exogenous ligand, the role of ALX/FPR2 in atheroprogression is a bit more complex, likely because ALX/FPR2 is expressed on multiple cell types and can bind several ligands [39]. Significantly, the proresolving protein IL-10, when encapsulated into plaque-targeted nanoparticles, promoted plaque stability in part through a reduction in necrosis and increase in fibrous cap thickness [40]. In addition to proresolving proteins and peptide, exogenous administration of SPMs to atherosclerotic mice and rabbits has recently been explored. Resolvin E1 (RvE1), which is a ligand for the GPCR called ChemR23, reduced atherogenesis in cholesterol-fed rabbits [32▪▪]. Another study highlighted that RvE1, when administered in combination with atorvastatin, led to a significantly greater decrease in plaque size compared with RvE1 alone [34]. These results suggest that a cotreatment strategy, as alluded to above, to both lower cholesterol and enhance inflammation-resolution, may be particularly beneficial for thwarting plaque progression.

More recent studies focused on SPMs and plaque progression. Specifically, atheroprogression was associated with SPM : proinflammatory mediator imbalances in human [31▪] and murine plaques

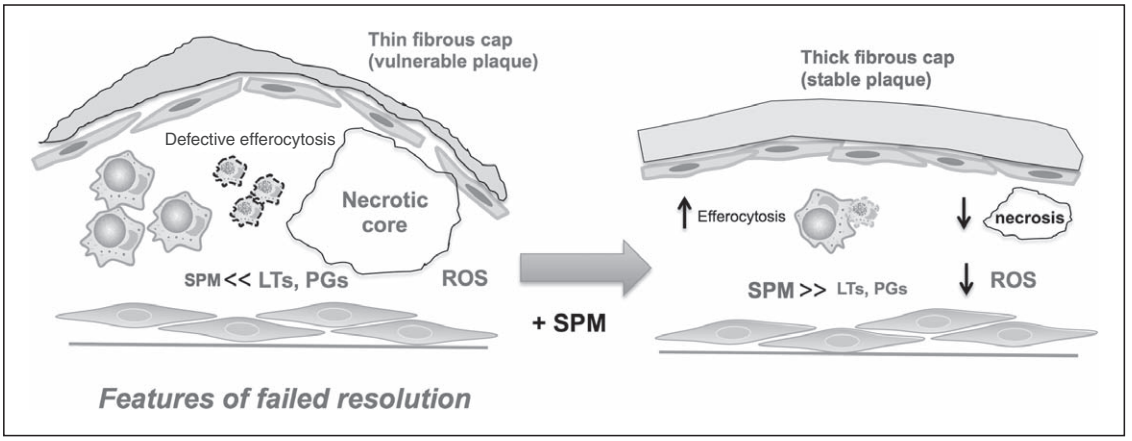

FIGURE 1. Specialized proresolving lipid mediators are protective in advanced murine atherosclerosis. Advanced atherosclerotic lesions have features of failed resolution including an imbalance in specialized proresolving lipid mediators : proinflammatory leukotrienes, defective efferocytosis, large necrotic cores, thin fibrous caps and increased reactive oxygen species. Restoration of key defective specialized proresolving lipid mediators such as resolvin D1, resolvin D2 or maresin 1 to already established atherosclerotic lesions promote lesion stability largely in part through reduced necrotic cores and thicker fibrous caps. Resolvin D1 also increases lesional efferocytosis and decreases reactive oxygen species.

[31▪,33▪]. Restoration of key defective SPMs, like Resolvin D1 (RvD1), which is an ALX/FPR2 ligand or a combination of Resolvin D2 (RvD2, a GPR18 ligand) and maresin 1, led to decreased plaque necrosis and increased fibrous cap thickness (Fig. 1) [31▪,33▪]. Moreover, RvD1 administration increased lesional efferocytosis, which is a process known to promote tissue repair. Together, these results suggest that large necrotic cores, defective efferocytosis and thin caps are all features of failed inflammation resolution [41,42]. The above findings strongly suggest that lipid mediator imbalances promote plaque instability. New mechanisms that underpin plaque necrosis [43] and efferocytosis are emerging [44], and how lipid mediator imbalances impact these new processes are of unknown.

Gaps in our understanding of SPM biology in atherosclerosis remain. First, it would be informative to know the cellular targets of SPMs. In this regard, generation and validation of ALX/FPR2, GPR18 and ChemR23 floxed mice would be invaluable tools for the field and would help identify the cell type(s) that evoke the above protective actions. Secondly, a deeper understanding of the SPM-GPCR interaction and signaling is important for future translation to humans because we will not only gain more insight into proresolution mechanisms but we can begin to determine how SPMs may interact with other drugs [22], the latter being especially important for precision medicine [45].

In this regard, the Perretti laboratory has uncovered many new mechanisms associated with ALX/FPR2 signaling [24,46]. As an example, GPCRs can form dimers that affect their activation and downstream signaling [47]. Although dimerization is not required for ligand recognition, emerging evidence suggests that dimerization may play a key role in fine-tuning host responses [47]. For example, Cooray *et al.* [24] found that Annexin A1 initiated ALX/FPR2 homodimerization, which led to a proresolution feed-forward circuit and an increase in IL-10. Ac2–26 is a bioactive peptide from Annexin A1 that has been shown to have anti-inflammatory and proresolving properties [22]. Ac2–26 initiates a heterodimer complex with ALX/FPR2 and FPR1, the latter is a GPCR, which is thought to play roles in proinflammatory signaling. This heterodimer complex initiated by Ac2–26 binding neutralizes the actions of a proinflammatory ligand called serum amyloid A [24]. These results suggest that proresolution ligands beget proresolution signaling to counterbalance proinflammatory signals at the level of the receptor. How these processes occur *in vivo* and in atherosclerosis remain of immense interest. Overall, an improved understanding of SPM-initiated signaling pathways will unravel new mechanisms that may give rise to novel therapeutics and may reveal rapid molecular readouts for assessing resolution responses in humans.

MECHANISMS ASSOCIATED WITH LIPID MEDIATOR IMBALANCES AND BEYOND

Acute inflammatory processes include vascular leakage and edema formation, mobilization of circulating leukocytes into tissues, phagocyte removal of dead cells and debris from tissue, eventual phagocyte clearance and mobilization of cells from the bone marrow [15,20]. These processes are required

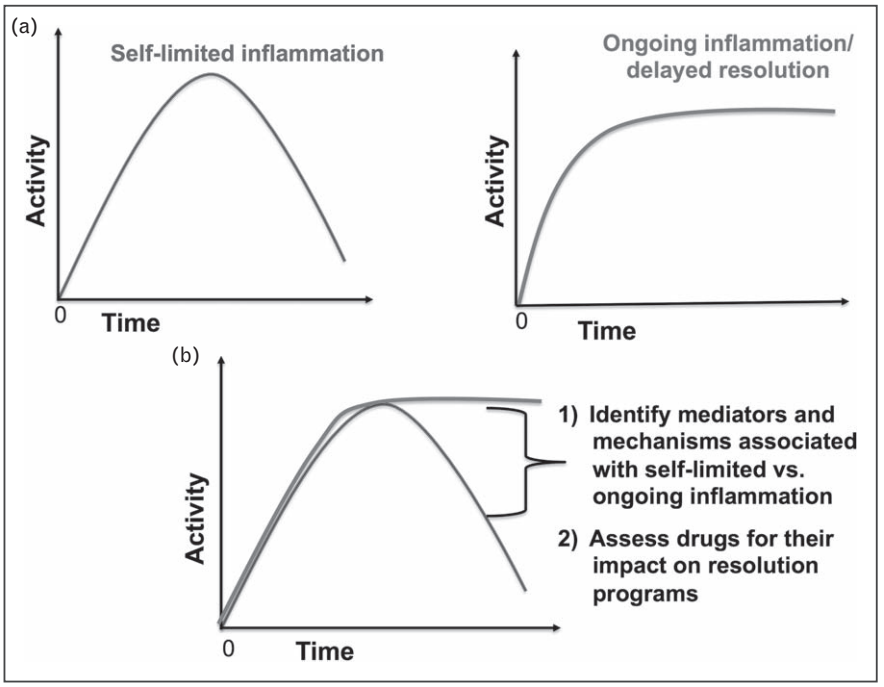

FIGURE 2. Self-limited versus ongoing inflammation from mouse to human. Low-dose zymosan A–induced peritonitis yields a self-limited inflammatory response, whereas high zymosan A reveals a response that represents ongoing inflammation and delayed resolution (a). A direct comparison of the low-dose versus high-dose zymosan peritonitis reactions has uncovered mediators and mechanisms associated with healthy versus dysregulated resolution (b). Furthermore, the low-dose versus high-dose zymosan peritonitis model provides a means to accurately assess the impact of particular drugs on inflammation and resolution programs. Translating this approach to humans will likely yield important ways to assess how particular drugs impact inflammation-resolution programs.

for neutralizing pathogens and tissue repair. With that said, a dysregulation of any one (or a combination) of these processes is associated with several chronic inflammatory diseases [27]. The challenge we are facing is how to target and temper the immune response without completely abolishing these host-protective pathways [15,16].

Simple proof-of-concept models like zymosan A–induced peritonitis simulate an acute inflammatory response and encompass all of the processes mentioned above. The dose of zymosan dictates the magnitude and extent of inflammation and resolution. Low-dose zymosan initiates a healthy, self-limited inflammatory reaction (i.e. inflammation that naturally resolves), and high-dose zymosan yields an outcome of ongoing inflammation and impaired resolution (Fig. 2a) [48,49]. The exact nature of inflammation incited by zymosan is not identical to the several nonresolving diseases mentioned above. Nevertheless, high-dose zymosan represents inflammation in its ongoing state and offers an approach to identify mediators involved in this maladaptive process (Fig. 2b). With this simple model, the Serhan lab uncovered that inflammation-resolution is an active process, which is in part regulated by SPMs [15]. In addition, the Serhan laboratory established resolution indices [25,48,50], which provide a quantitative measure of the host's ability to resolve inflammation. Furthermore, these indices provide a means to accurately assess the impact of particular drugs, cells, diet and others on inflammation and resolution processes. The causative role for SPMs in resolution was shown when exogenously administered SPMs led to a swifter resolution response [48]. Also, administration of key proresolving proteins and peptides like Annexin A1, Ac2–26 and chemerin also leads to swifter resolution response [51].

Fine-tune immune responses through mechanisms associated with lipid mediator imbalances

Exudates from ongoing inflammation (i.e. high-dose zymosan) had a significant imbalance in the SPM to proinflammatory leukotriene ratio compared with exudates from self-limited or resolving inflammation (i.e. low-dose zymosan) [25]. This imbalance, as mentioned above, has been associated with several human diseases and is evidence that this model can translate endpoints that are relevant to complex diseases. In the case of atherosclerosis, possible mechanisms

underlying SPM : leukotriene imbalances include factors that affect the subcellular localization of a key biosynthetic SPM and leukotriene enzyme called five-lipoxygenase (5-LOX), such as oxysterols/oxidative stress and SPMs [27]. During oxidative stress or inflammation, 5-LOX is phosphorylated and translocates to the nuclear membrane, which favors the biosynthesis of the proinflammatory mediator, leukotriene B_4 (LTB_4) [31▪,52]. The SPM RvD1 promotes nuclear exclusion of 5-LOX and thereby suppresses LTB_4 and enhances the proresolving molecule, lipoxin A_4 in macrophages [27]. Subcellular localization of five-lipoxygenase activating protein [53,54] and regulation of leukotriene A_4 hydrolase may also play important roles in SPM formation and in SPM : leukotriene balances [53,55]. Other processes that can affect SPM production and possibly play roles in atherosclerosis and other nonresolving diseases include transcellular biosynthesis between interacting cells [15,30] and the release of lipoxin precursors from cellular phospholipids [56]. Further understanding of mechanisms underlying SPM : leukotriene imbalances will yield new ways to fine tune mammalian host responses and may be an attractive way to approach treatment of long-term diseases, such as atherosclerosis. In this regard, mer proto-oncogene tyrosine kinase (MerTK), a well known efferocytosis receptor, is emerging as a key target in the regulation of SPM : leukotriene ratios [57▪]. MerTK is regulated by an ADAM17-mediated cleavage process that disables the receptor and produces a fraction called soluble Mer [58]. In this regard, MerTK cleavage was shown to dismantle SPM biosynthesis, which led to a marked imbalance in SPM : leukotrienes [57▪]. Thus, the resolution cycle is broken when MerTK is cleaved and the system loses both efferocytosis and the proper balance of proresolving and proinflammatory lipid mediators. Earlier work demonstrated that MerTK, a key efferocytosis receptor on macrophages, is a critical regulator of plaque necrosis and lesion stability [59,60]. It is possible that these new findings could apply to atherosclerosis, in which defects in both efferocytosis and inflammation-resolution underpin the disorder.

Together, these findings also suggest that SPM : leukotriene ratios and/or MerTK cleavage may be important readouts for whether inflammation is in its ongoing state or whether inflammation will be swiftly resolved.

Consider paths toward precision medicine

As mentioned above, the low-dose and high-dose zymosan peritonitis model provides a means to accurately assess the impact of particular drugs on inflammation and resolution processes [48,49,61]. Navarro-Xavier *et al.* [49] tested several anti-inflammatory drugs and SPMs in the peritonitis model and found that each drug had a distinct fingerprint for its impact on the inflammation-resolution response.

The next challenge is to translate these concepts to humans [45]. Because inflammation-resolution is required for host defense against pathogens and for tissue repair, understanding the impact of a particular drug on this program is extremely important. For example, cyclooxygenase-2 (COX-2) inhibitors and anti-TNFα therapy are considered 'inflammation-resolution toxic' because they impinge on host defense and tissue repair mechanisms [16,19]. Although the benefit of these drugs outweighs the risks in debilitative diseases like arthritis or colitis, the benefit : risk ratio for progressive diseases like atherosclerosis is low [16]. Significantly, statins have been shown to boost SPMs in certain contexts [62], but whether statins are capable of exerting this action in CVD patients is unknown.

The acute inflammatory response in humans can be assessed by the systemic inflammatory reaction following endotoxin [63,64] or the local inflammatory reaction following a cantharidin-induced skin blister [65]. Recent findings in humans indicate that SPMs are present in the plasma [66–68] and can possibly serve as temporal biomarkers of inflammation-resolution after endotoxin challenge. In support of this, plasma from sepsis patients revealed specific lipid mediator signatures associated with clinical outcomes [69▪▪]. The blister model is particularly intriguing because it provides a controlled induction and termination of local inflammation. Using this approach, Morris *et al.* [70] uncovered that humans broadly fall into two categories, those who resolved their acute inflammatory response swiftly and those who showed a more delayed or prolonged inflammatory response. The magnitude and duration of the acute inflammatory response was controlled by endogenous epimeric lipoxins and ALX/FPR2 expression [70]. Overall, these simple and noninvasive human tests can lead to a better understanding of how an individual can mount and resolve an acute inflammatory response and may be an ideal platform to test whether particular drugs impinge on an individual's ability to resolve inflammation. The latter will be particularly important for people who have long-term chronic diseases.

CONCLUSION

Current treatments for atherosclerosis have made major improvements in patient outcomes, but even so, CVD still remains the leading cause of death worldwide. Therefore, in addition to lipid lowering, we need to also seriously consider the inflammatory

processes of the disease. Atherosclerosis is a long-term progressive disease, and so anti-inflammatory drugs that dampen host defense mechanisms or that also disrupt vascular homeostasis are not ideal [16,71,72]. Inflammation-resolution pathways not only quell excessive or persistent inflammation without disrupting necessary host defense programs but also stimulate tissue repair and regeneration [15]. Moreover, the very recent preclinical literature that is summarized in this review supports the atheroprotective action of proresolving mediators and suggests that a new way to treat CVD may be through the activation of tissue reparative programs.

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Conflicts of interest

There are no conflicts of interest. This article is exclusively based on experimental studies.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ■ of special interest
- ■■ of outstanding interest

1. Ross R. Rous–Whipple Award Lecture. Atherosclerosis: a defense mechanism gone awry. Am J Pathol 1993; 143:987–1002.
2. Tabas I, Williams KJ, Boren J. Subendothelial lipoprotein retention as the initiating process in atherosclerosis: update and therapeutic implications. Circulation 2007; 116:1832–1844.
3. Tabas I, Bornfeldt KE. Macrophage phenotype and function in different stages of atherosclerosis. Circ Res 2016; 118:653–667.
4. Libby P, Tabas I, Fredman G, Fisher EA. Inflammation and its resolution as determinants of acute coronary syndromes. Circ Res 2014; 114:1867–1879.
5. Sansbury BE, Spite M. Resolution of acute inflammation and the role of resolvins in immunity, thrombosis, and vascular biology. Circ Res 2016; 119:113–130.
6. Tall AR, Yvan-Charvet L. Cholesterol, inflammation and innate immunity. Nat Rev Immunol 2015; 15:104–116.
7. Moore KJ, Sheedy FJ, Fisher EA. Macrophages in atherosclerosis: a dynamic balance. Nat Rev Immunol 2013; 13:709–721.
8. Tabas I, Garcia-Cardena G, Owens GK. Recent insights into the cellular biology of atherosclerosis. J Cell Biol 2015; 209:13–22.
9. Merched AJ, Ko K, Gotlinger KH, *et al.* Atherosclerosis: evidence for impairment of resolution of vascular inflammation governed by specific lipid mediators. FASEB J 2008; 22:3595–3606.
10. Ridker PM. Residual inflammatory risk: addressing the obverse side of the atherosclerosis prevention coin. Eur Heart J 2016; 37:1720–1722.
11. Libby P, Hansson GK. Inflammation and immunity in diseases of the arterial tree: players and layers. Circ Res 2015; 116:307–311.
12. Libby P, Nahrendorf M, Swirski FK. Leukocytes link local and systemic inflammation in ischemic cardiovascular disease: an expanded 'cardiovascular continuum'. J Am Coll Cardiol 2016; 67:1091–1103.
13. Everett BM, Pradhan AD, Solomon DH, *et al.* Rationale and design of the cardiovascular inflammation reduction trial: a test of the inflammatory hypothesis of atherothrombosis. Am Heart J 2013; 166:199–207.e15.
14. Ridker PM, Thuren T, Zalewski A, Libby P. Interleukin-1beta inhibition and the prevention of recurrent cardiovascular events: rationale and design of the Canakinumab Anti-inflammatory Thrombosis Outcomes Study (CANTOS). Am Heart J 2011; 162:597–605.
15. Serhan CN. Pro-resolving lipid mediators are leads for resolution physiology. Nature 2014; 510:92–101.
16. Tabas I, Glass CK. Anti-inflammatory therapy in chronic disease: challenges and opportunities. Science 2013; 339:166–172.
17. ■■ Serhan CN, Chiang N, Dalli J. The resolution code of acute inflammation: novel pro-resolving lipid mediators in resolution. Semin Immunol 2015; 27:200–215.

Charles Serhan discovered specialized proresolving lipid mediators (SPMs), their biosynthetic pathways, structures and several G-protein coupled receptors to which they bind.

18. Gobbetti T, Coldewey SM, Chen J, *et al.* Nonredundant protective properties of FPR2/ALX in polymicrobial murine sepsis. Proc Natl Acad Sci U S A 2014; 111:18685–18690.
19. Buckley CD, Gilroy DW, Serhan CN. Proresolving lipid mediators and mechanisms in the resolution of acute inflammation. Immunity 2014; 40:315–327.
20. Kotas ME, Medzhitov R. Homeostasis, inflammation, and disease susceptibility. Cell 2015; 160:816–827.
21. Sugimoto MA, Sousa LP, Pinho V, *et al.* Resolution of inflammation: what controls its onset? Front Immunol 2016; 7:160.
22. Perretti M, Leroy X, Bland EJ, Montero-Melendez T. Resolution pharmacology: opportunities for therapeutic innovation in inflammation. Trends Pharmacol Sci 2015; 36:737–755.
23. Wallace JL, Ianaro A, Flannigan KL, Cirino G. Gaseous mediators in resolution of inflammation. Semin Immunol 2015; 27:227–233.
24. Cooray SN, Gobbetti T, Montero-Melendez T, *et al.* Ligand-specific conformational change of the G-protein-coupled receptor ALX/FPR2 determines proresolving functional responses. Proc Natl Acad Sci U S A 2013; 110:18232–18237.
25. Fredman G, Li Y, Dalli J, *et al.* Self-limited versus delayed resolution of acute inflammation: temporal regulation of pro-resolving mediators and microRNA. Sci Rep 2012; 2:639.
26. Chiang N, Fredman G, Backhed F, *et al.* Infection regulates pro-resolving mediators that lower antibiotic requirements. Nature 2012; 484:524–528.
27. Fredman G, Ozcan L, Spolitu S, *et al.* Resolvin D1 limits 5-lipoxygenase nuclear localization and leukotriene B_4 synthesis by inhibiting a calcium-activated kinase pathway. Proc Natl Acad Sci U S A 2014; 111:14530–14535.
28. Basil MC, Levy BD. Specialized pro-resolving mediators: endogenous regulators of infection and inflammation. Nat Rev Immunol 2016; 16:51–67.
29. Freire MO, Van Dyke TE. Natural resolution of inflammation. Periodontol 2000 2013; 63:149–164.
30. Brezinski DA, Nesto RW, Serhan CN. Angioplasty triggers intracoronary leukotrienes and lipoxin A_4. Impact of aspirin therapy. Circulation 1992; 86:56–63.
31. ■ Fredman G, Hellmann J, Proto JD, *et al.* An imbalance between specialized pro-resolving lipid mediators and pro-inflammatory leukotrienes promotes instability of atherosclerotic plaques. Nat Commun 2016; 7:12859.

Quantified lipid mediator imbalances in human and murine atherosclerotic plaques and found that these imbalances led to plaque vulnerability.

32. ■■ Hasturk H, Abdallah R, Kantarci A, *et al.* Resolvin E1 (RvE1) attenuates atherosclerotic plaque formation in diet and inflammation-induced atherogenesis. Arterioscler Thromb Vasc Biol 2015; 35:1123–1133.

This was the first report to show that administration of a key SPM called resolvin E1 thwarted atherogenesis.

33. ■ Viola J, Lemnitzer P, Jansen Y, *et al.* Resolving lipid mediators maresin 1 and resolvin D2 prevent atheroprogression in mice. Circ Res 2016; 119:1030–1038.

Quantified lipid mediator imbalances in murine atherosclerotic plaques and found that these imbalances led to plaque vulnerability.

34. Salic K, Morrison MC, Verschuren L, *et al.* Resolvin E1 attenuates atherosclerosis in absence of cholesterol-lowering effects and on top of atorvastatin. Atherosclerosis 2016; 250:158–165.
35. Dufton N, Hannon R, Brancaleone V, *et al.* Anti-inflammatory role of the murine formyl-peptide receptor 2: ligand-specific effects on leukocyte responses and experimental inflammation. J Immunol 2010; 184:2611–2619.
36. Chiang N, Dalli J, Colas RA, Serhan CN. Identification of resolvin D2 receptor mediating resolution of infections and organ protection. J Exp Med 2015; 212:1203–1217.
37. ■ Fredman G, Kamaly N, Spolitu S, *et al.* Targeted nanoparticles containing the proresolving peptide Ac2–26 protect against advanced atherosclerosis in hypercholesterolemic mice. Sci Transl Med 2015; 7:275ra20.

The first to report atherosclerotic plaque-targeted proresolving nanomedicines in mice.

38. Drechsler M, de Jong R, Rossaint J, *et al.* Annexin A1 counteracts chemokine-induced arterial myeloid cell recruitment. Circ Res 2015; 116:827–835.
39. Petri MH, Laguna-Fernandez A, Gonzalez-Diez M, *et al.* The role of the FPR2/ALX receptor in atherosclerosis development and plaque stability. Cardiovasc Res 2015; 105:65–74.
40. Kamaly N, Fredman G, Fojas JJ, *et al.* Targeted interleukin-10 nanotherapeutics developed with a microfluidic chip enhance resolution of inflammation in advanced atherosclerosis. ACS Nano 2016; 10:5280–5292.
41. Soehnlein O. (Re)solving atherosclerosis. Sci Transl Med 2015; 7:275fs7.

42. Viola J, Soehnlein O. Atherosclerosis – a matter of unresolved inflammation. Semin Immunol 2015; 27:184–193.

43. Karunakaran D, Geoffrion M, Wei L, *et al.* Targeting macrophage necroptosis for therapeutic and diagnostic interventions in atherosclerosis. Sci Adv 2016; 2:e1600224.

44. Kojima Y, Volkmer JP, McKenna K, *et al.* CD47-blocking antibodies restore phagocytosis and prevent atherosclerosis. Nature 2016; 536:86–90.

45. Fullerton JN, Gilroy DW. Resolution of inflammation: a new therapeutic frontier. Nat Rev Drug Discov 2016; 15:551–567.

46. Thompson D, McArthur S, Hislop JN, *et al.* Identification of a novel recycling sequence in the C-tail of FPR2/ALX receptor: association with cell protection from apoptosis. J Biol Chem 2014; 289:36166–36178.

47. Filep JG. Biasing the lipoxin A4/formyl peptide receptor 2 pushes inflammatory resolution. Proc Natl Acad Sci U S A 2013; 110:18033–18034.

48. Bannenberg GL, Chiang N, Ariel A, *et al.* Molecular circuits of resolution: formation and actions of resolvins and protectins. J Immunol 2005; 174:4345–4355.

49. Navarro-Xavier RA, Newson J, Silveira VL, *et al.* A new strategy for the identification of novel molecules with targeted proresolution of inflammation properties. J Immunol 2010; 184:1516–1525.

50. Schwab JM, Chiang N, Arita M, Serhan CN. Resolvin E1 and protectin D1 activate inflammation-resolution programmes. Nature 2007; 447:869–874.

51. Cash JL, Norling LV, Perretti M. Resolution of inflammation: targeting GPCRs that interact with lipids and peptides. Drug Discov Today 2014; 19:1186–1192.

52. Radmark O, Werz O, Steinhilber D, Samuelsson B. 5-Lipoxygenase, a key enzyme for leukotriene biosynthesis in health and disease. Biochim Biophys Acta 2015; 1851:331–339.

53. Lehmann C, Homann J, Ball AK, *et al.* Lipoxin and resolvin biosynthesis is dependent on 5-lipoxygenase activating protein. FASEB J 2015; 29:5029–5043.

54. Elias I, Ferre T, Vila L, *et al.* Alox5ap overexpression in adipose tissue leads to LXA_4 production and protection against diet-induced obesity and insulin resistance. Diabetes 2016.

55. Haeggstrom JZ, Funk CD. Lipoxygenase and leukotriene pathways: biochemistry, biology, and roles in disease. Chem Rev 2011; 111:5866–5898.

56. Norris PC, Gosselin D, Reichart D, *et al.* Phospholipase A2 regulates eicosanoid class switching during inflammasome activation. Proc Natl Acad Sci U S A 2014; 111:12746–12751.

57. ▪ Cai B, Thorp EB, Doran AC, *et al.* MerTK cleavage limits proresolving mediator biosynthesis and exacerbates tissue inflammation. Proc Natl Acad Sci U S A 2016; 113:6526–6531.

First to report that MerTK signaling regulates the SPM : proinflammatory mediator ratio.

58. Thorp E, Vaisar T, Subramanian M, *et al.* Shedding of the Mer tyrosine kinase receptor is mediated by ADAM17 protein through a pathway involving reactive oxygen species, protein kinase Cdelta, and p38 mitogen-activated protein kinase (MAPK). J Biol Chem 2011; 286:33335–33344.

59. Ait-Oufella H, Pouresmail V, Simon T, *et al.* Defective mer receptor tyrosine kinase signaling in bone marrow cells promotes apoptotic cell accumulation and accelerates atherosclerosis. Arterioscler Thromb Vasc Biol 2008; 28:1429–1431.

60. Thorp E, Cui D, Schrijvers DM, *et al.* Mertk receptor mutation reduces efferocytosis efficiency and promotes apoptotic cell accumulation and plaque necrosis in atherosclerotic lesions of $apoe^{-/-}$ mice. Arterioscler Thromb Vasc Biol 2008; 28:1421–1428.

61. Chiang N, Schwab JM, Fredman G, *et al.* Anesthetics impact the resolution of inflammation. PLoS One 2008; 3:e1879.

62. Dalli J, Chiang N, Serhan CN. Elucidation of novel 13-series resolvins that increase with atorvastatin and clear infections. Nat Med 2015; 21:1071–1075.

63. Patel PN, Shah RY, Ferguson JF, Reilly MP. Human experimental endotoxemia in modeling the pathophysiology, genomics, and therapeutics of innate immunity in complex cardiometabolic diseases. Arterioscler Thromb Vasc Biol 2014; 35:525–534.

64. Fullerton JN, Segre E, De Maeyer RP, *et al.* Intravenous endotoxin challenge in healthy humans: an experimental platform to investigate and modulate systemic inflammation. J Vis Exp 2016; doi: 10.3791/53913. [Epub ahead of print]

65. Jenner WJ, Gilroy DW. Assessment of leukocyte trafficking in humans using the cantharidin blister model. JRSM Cardiovasc Dis 2012; 1. doi: 10.1258/cvd.2012.012009.

66. Markworth JF, Kaur G, Miller EG, *et al.* Divergent shifts in lipid mediator profile following supplementation with n-3 docosapentaenoic acid and eicosapentaenoic acid. FASEB J 2016; 30:3714–3725.

67. Elajami TK, Colas RA, Dalli J, *et al.* Specialized proresolving lipid mediators in patients with coronary artery disease and their potential for clot remodeling. FASEB J 2016; 30:2792–2801.

68. Colas RA, Shinohara M, Dalli J, *et al.* Identification and signature profiles for pro-resolving and inflammatory lipid mediators in human tissue. Am J Physiol Cell Physiol 2014; 307:C39–C54.

69. ▪▪ Dalli J, Colas RA, Quintana C, *et al.* Human sepsis eicosanoid and proresolving lipid mediator temporal profiles: correlations with survival and clinical outcomes. Crit Care Med 2016. [Epub ahead of print]

Identified lipid mediator profiles in plasma from surviving versus nonsurviving sepsis patients. This report suggests that SPM and proinflammatory mediators in the plasma may be useful biomarkers for clinical outcomes.

70. Morris T, Stables M, Colville-Nash P, *et al.* Dichotomy in duration and severity of acute inflammatory responses in humans arising from differentially expressed proresolution pathways. Proc Natl Acad Sci U S A 2010; 107:8842–8847.

71. van der Valk FM, van Wijk DF, Lobatto ME, *et al.* Prednisolone-containing liposomes accumulate in human atherosclerotic macrophages upon intravenous administration. Nanomedicine 2015; 11:1039–1046.

72. van der Valk FM, Schulte DM, Meiler S, *et al.* Liposomal prednisolone promotes macrophage lipotoxicity in experimental atherosclerosis. Nanomedicine 2016; 12:1463–1470.