Bioactive lipids in antiviral immunity
Lipids may influence viral entry, replication, and clearance and modulate immune responses

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Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has brought focus to attempts to limit viral replication and manage the immunological response to infection. Lipids modulate host receptor binding, facilitate viral fusion, and fuel viral replication; thus, modulation of viral-host lipid interactions may have therapeutic utility (1). Indeed, the spike (S) glycoprotein on the surface of SARS-CoV-2 tightly binds the free fatty acid linoleic acid, stabilizing it and reducing its interaction with the host angiotensin-converting enzyme 2 (ACE2) receptor that facilitates viral cell entry (2). However, in the case of many viral infections, including COVID-19, it is the overexuberant host immune response that results in life-threatening consequences of infection. Therefore, therapies that modulate bioactive lipids that regulate the host immune response to respiratory viral infections may be beneficial.

Viruses may release lipids as they transiently disrupt membrane integrity during cell entry or by causing cell lysis. Oxidized phospholipids (oxPLs) are generated enzymatically—for example, by 15-lipoxygenase in immune cells—yielding specific enantiomers (3), or they are produced nonenzymatically when reactive oxygen species react with unsaturated fatty acids in membrane phospholipids (see the figure). The effects of oxPLs are context- and site-specific. They can function as damage associated molecular patterns (DAMPs) to amplify the host innate immune response and contribute to the pathogenesis of acute lung injury by activating Toll-like receptor 4 (TLR4) signaling (4) with a potency similar to that of endotoxin (produced by Gram-negative bacteria). However, lower concentrations of oxPLs can have protective effects by enhancing pulmonary endothelial barrier function and inducing expression of antioxidant genes. Autoantibodies directed against PLs and PL binding proteins may worsen the severity of COVID-19 because they may stimulate the formation of neutrophil extracellular traps, which are extruded DNA “nets” that can promote venous thrombosis (blood clots). Cleavage of isoprostanes—free radical oxidized prostaglandin (PG) isomers—from membrane lipids affords an accessible index in biological fluids as biomarkers of lipid peroxidation.

Sphingosine-1-phosphate (SIP) is formed by phosphorylation of the lipid sphingosine by one of two sphingosine kinases (SK1 and SK2), and its concentrations in blood are tightly regulated through irreversible cleavage by SIP lyase (SPL) and lipid phosphate phosphatases (LPPs). It acts intracellularly or through one of five membrane G protein–coupled receptors (SIPR, to SIPR) in immune cells, resulting in diverse effects on innate immune function. SIPR antagonists are approved for treatment of autoimmune diseases. SIP signaling in the lung decreases dendritic cell (DC) migration and expansion and recruitment of CD8+ T cells in response to influenza A virus (IAV) infection. Antigen presentation to lymphocytes by DCs thus links the innate and adaptive immune systems. SIPR activation in endothelial cells early (i.e., 1 hour) in IAV infection in mice tempers the hyperinflammatory response and improves survival, whereas deletion of Sip1 in endothelial cells decreases survival and worsens lung pathology after IAV infection (5). It remains to be seen if SIPR agonists would be similarly beneficial if administered at later stages of infection, as would be more common in clinical settings. SK2 is also a rational drug target because it facilitates viral persistence by restraining T cell immune pathology during lymphocytic choriomeningitis virus clone 13 infection in mice (6).

Eicosanoids are metabolites of arachidonic acid, the fatty acid present in membrane phospholipids. Eicosanoids include PGs and thromboxane—among them, prostaglandins formed by the cyclooxygenases COX-1 and COX-2; the leukotrienes formed by the lipoxigenase (LOX) enzymes; and epoxyeicosatrienoic acids (EETs) and 20-hydroxy-eicosatetraenoic acid (20-HETE) formed by cytochrome P450 (CYP) enzymes. These bioactive lipids can modulate antiviral immune responses differently.

The first step in prostaglandin synthesis is the conversion of arachidonic acid to prostaglandin H2 (PGH2) by COX-1 or COX-2. PGH2 is further metabolized to the bioactive prostaglandins prostaglandin E2 (PGE2), prostaglandin D2 (PGD2), prostacyclin (PGI2), prostaglandin F2α (PGF2α), and thromboxane A2 (TXA2), which can then bind to their receptors to elicit biological effects. The role of COX-1 in viral infections has not been well described, although mortality after H3N2 IAV infection was higher in mice in which COX-1 was deleted or inhibited. By contrast, COX-2 expression is up-regulated in response to viral infection and suppression of COX-2–derived PGs or deletion of Ptgs2 (which encodes COX-2) before H3N2 IAV infection in mice tempers the inflammatory response, attenuates weight loss, and increases survival. This is coincident with a reduction in the numbers of neutrophils, macrophages, and the cytokines tumor necrosis factor–α (TNF-α), interleukin-1β (IL-1β), IL-6, and interferon-γ (IFN-γ) in bronchoalveolar lavage fluid (BALF) (7).

Treatment of mice with COX inhibitors before IAV infection similarly reduced inflammation and improved survival. COX-2 inhibition may also enhance the response to viral infection in the setting of a chronic inflammatory state such as that induced by obesity (8). Obese mice have higher amounts of cytokines and chemokines in lung homogenates at baseline and display poor induction of the type 1 IFN response that is critical to viral clearance. PG suppression restores type 1 IFN induction and improves survival in response to infection in obese, but not in lean, mice (8). Overall, genetic deletion and pharmacological inhibition of the COXs give concordant results where available, but these enzymes produce prostaglandins with contrasting effects on immune function and hemostasis.

Downstream of the COXs, attention has focused on PGE2, given its well-recognized immunomodulatory effects. Deletion or blockade of microsomal PGE synthase 1 (mPGES-1), the major source of PGE2, or of the E prostaglandin receptors, EP2 or EP4, suppresses the immune response to viral infection. Ptges deletion improves survival, lowers viral load, and promotes pulmonary infiltration of macrophages, monocytes, and DCs after H1N1 IAV infection in mice (9). Furthermore, Ptges deletion enhanced adaptive immune responses, with higher numbers of CD4+ and CD8+ T cells in lymph nodes, BALF, and lungs of mice. Pharmacologically induced inhibition of mPGES-1 or EP2 or EP4 signaling in vivo improves...
survival after IAV infection, and this benefit is lost in mice lacking the type I IFN-α/β receptor.

PGD₂ is the predominant COX-2 product of mast cells and acts through its two receptors, Drp1 and Drp2. Drp1 signaling delays migration of DCs to lung and lymph nodes by down-regulating the expression of C-C chemokine receptor type 7 (CCR7) on respiratory DCs in response to infection. Drp1 inhibition enhances DC migration and, in turn, T cell proliferation, which increased survival in older but not younger mice after SARS-CoV infection (10). In a neonatal mouse model of severe respiratory syncytial virus (RSV)-induced bronchiolitis, treatment with a Drp2 inhibitor decreased viral load and improved morbidity by up-regulating IFN-γ expression. This effect was recapitulated by treatment with a Drp1 agonist, suggesting that these two receptors for PGD₂ have opposing roles in the regulation of the antiviral response (11).

PGF₂α, prostanycin, and TxA₂ may contribute to the systemic effects of respiratory viral infection. For example, PGF₂α and prostanycin promote or restrain, respectively, pulmonary fibrotic reactions to viral infection in mice. Antagonism of the thromboxane receptor (TPr) prevents acute respiratory distress syndrome (ARDS) in an endotoxin model in sheep (12). Suppression of TxA₂ formation may have the added benefit of inhibiting platelet-dependent thrombosis. Thus, early administration of well-tolerated TPr antagonists may limit progress to severe COVID-19.

Of the leukotriene eicosanoids, the expression of LTD₄ and cysteinyl leukotrienes (cysLTs) is up-regulated in nasal or lung lavage fluids from animals and humans infected with viruses. They promote inflammation, vascular leakiness, and broncho- and vasoconstriction. However, evidence for modulation by leukotrienes of the immune response to viral infection is conflicting. Administration of zileuton, an inhibitor of 5-lipoxygenase (5-LO, which produces a precursor of LTD₄), before RSV infection in mice improved virus-infected airway constriction and reduced inflammatory cells in the lungs and weight loss (13).

However, leukotriene signaling may support tolerance to IAV infection. In the lungs of IAV-infected mice, intercellular macromolecules deficient in LTD₄ receptor signaling failed to produce type I IFN to suppress activated inflammatory monocyte-derived macrophages, which was one of the main drivers of tissue injury and death from uncontrolled immune responses (14). Similarly, CysLT1 receptor blockade reduced the susceptibility of alveolar macrophage-deficient mice to IAV and decreased airway hyper-responsiveness in mice challenged with RSV. However, treatment of respiratory symptoms of post-RSV bronchiolitis in humans with montelukast, a CysLT receptor antagonist, was inconclusive.

CYP-derived eicosanoids also possess immunomodulatory properties, but their role in regulating the host response to viral infection has not been studied. The EETs attenuate cytokine-induced nuclear factor κB (NF-κB) activation and leukocyte adhesion to the vascular wall. Soluble epoxide hydrolase (sEH) rapidly hydrolyzes EETs to the less biologically active dihydroxyeicosatetraenoic acids (DHETs), and inhibition of sEH has been studied as a therapeutic strategy to decrease inflammation in vivo. Conversely, 20-HETE activates NF-κB signaling and induces expression of cellular adhesion molecules and cytokines, thereby promoting inflammation.

The role of bioactive lipids in inflammation is complex, and additional studies in humans and model organisms are essential to elucidate their importance in regulating the immune response to viral infection, particularly SARS-CoV-2. Further, genomic, transcriptomic, and serial lipidomic analyses in individuals with COVID-19 may reveal specific lipid pathways that contribute to pathology, as well as identify whether these pathways predict or mediate the heterogeneous response to viral infection.

In the current pandemic, there may be utility in targeting eicosanoids and other bioactive lipids with existing drugs. These approaches would likely be most effective early in the disease before the development of ARDS, where cytokines and chemokines dominate. However, even in that setting, leukotrienes and related compounds may serve as important amplifying signals, which has prompted suggestions to combine approved 5-LO inhibitors with CysLT antagonists in the treatment of severe COVID-19 (15). Given the thrombotic complications of COVID-19, early intervention in patients with increased hemostatic markers with low-dose aspirin (which inhibits platelet COX1), TPr antagonists, or prostanycin receptor (IPr) agonists are among the therapeutic possibilities that might be explored. Although nonsteroidal anti-inflammatory drugs (NSAIDs, such as ibuprofen) should be avoided in patients with renal compromise in ARDS owing to the importance of vasodilator PGs in the maintenance of renal blood flow, more discrete modulation of selective, downstream biosynthetic enzymes and/or receptors—for example, with mPGES-1 inhibitors or EPr antagonists—may have therapeutic utility in the treatment of viral disease.

**REFERENCES AND NOTES**


10.1126/science.aba6526

238 15 JANUARY 2021 • VOL 371 ISSUE 6526
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Science 371 (6526), 237-238.
DOI: 10.1126/science.abf3192