Check for updates

Innate Immune Reprogramming in Chronic Obstructive Pulmonary Disease

New Mechanisms for Old Questions

The alveolar macrophage is an innate immune cell best known for its phagocytic functions, especially in the context of infection. It is often overlooked because immunology is a rapidly advancing field, with mediators like IL-36 (1) and cells like type 2 innate lymphoid cells (2) being relatively recent discoveries. However, the lung macrophage family members are critical innate immune cells that can drive pathophysiological processes in respiratory diseases and, in particular, have been demonstrated as integral to the pathobiology of chronic obstructive pulmonary disease (COPD).

Macrophage biology is complex. In the lung, there are two types of resident macrophages: alveolar macrophages and interstitial macrophages. Alveolar macrophages can originate from local fetal monocyte progenitors that seed the lungs during development (3–5). In contrast, interstitial macrophages represent a mixed phenotype that can be self-renewing or derived from circulating monocytes. Interestingly, during infectious or environmental challenges to the lung, recruited monocytes can differentiate into cells similar to resident alveolar macrophages, further complicating what should be a simple question regarding the origin of the cell. The quote by Douglas Adams, "if it looks like a duck and quacks like a duck, we have at least to consider the possibility that we have a small aquatic bird of the family Anatidae on our hands," comes to mind here, but even though macrophages may look like ordinary macrophages in people with COPD, asthma, or cystic fibrosis, the intrinsic properties of these cells can be radically different. For example, mediator release is increased (6) and efferocytosis is defective (7) in macrophages in COPD. However, the mechanisms driving this immune reprogramming are poorly understood.

In this issue of the *Journal*, Salih and colleagues (pp. 511–522) present data that begin to shed light on the complex innate immune reprogramming of macrophages in COPD (8). They first transcriptionally profile bone marrow–derived macrophages (BMDMs) in an *in vivo* model of COPD. This is an important experimental nuance that needs to be considered. The differentiation of monocytes into macrophages *ex vivo* occurs between 7 and 14 days. Therefore, it is unlikely that, at this time point, they are simply measuring the response to *in vivo* stimulation given that maximal inflammatory responses in this cell type occur approximately 24–48 hours after stimulation, followed by a return to baseline. Perhaps unsurprisingly, given the inflammatory nature of COPD, BMDMs from cigarette smoke–exposed mice displayed a proinflammatory phenotype. This demonstrates that systemic responses to cigarette smoke exposure may alter intrinsic leukocyte properties and that the altered macrophage phenotype observed in the human lung in COPD may be the result of different factors acting on local and bone marrow–derived immune cells.

The authors demonstrate that several genes were alternatively spliced following *in vivo* cigarette smoke exposure. Alternative splicing of genes can enhance or suppress the activity of the gene product depending on the translated product. Although 47–425 splicing events were found, none occurred in inflammatory genes. Network analysis identified that the Notch 2 signaling pathway may be dysregulated, suggesting a potential mechanism by which cigarette smoke exposure could affect developmental processes.

Long noncoding RNAs (lncRNAs), which function as important genomic regulators, can also be alternatively spliced, and the authors, therefore, sought to profile the production of lncRNA. Only three lncRNA species were upregulated following *in vivo* smoke exposure, one of which was lncRNA (long non-coding RNA)-Cox2 (which regulates *Ptgs2*, the gene that encodes the enzyme cyclooxygenase [COX]-2). Macrophages self-regulate cytokine production via the autocrine action of the eicosanoid prostaglandin E2 (PGE2), a mechanism first described almost 35 years ago (9). PGE2 is a lipid mediator that is produced in response to upregulation of COX-2. The regulation of *Ptgs2* is therefore critical for normal functioning of macrophages.

To understand the sequelae of upregulation of lncRNA-Cox2, a knockout mouse model was used, and cytokine production in serum and BAL fluid was evaluated. Unsurprisingly, the lack of lncRNA-Cox2 resulted in the dysregulation of several inflammatory mediators in both body compartments, which could be rescued via the overexpression of lncRNA-Cox2. Overexpression was achieved by cross-breeding lncRNA-Cox2–deficient and lncRNA-Cox2–overexpressing mice. To further corroborate these observations, the authors performed *ex vivo* experiments involving LPS stimulation of BMDMs following cigarette smoke exposure. Interestingly, constitutive cytokine release was not affected in either genotype; however, LPS stimulation revealed cytokine dysregulation with enhanced production of several proinflammatory mediators.

This is a robust and well-controlled study, but it does have some limitations. The first is that the use of a global knockout or global overexpression model to interrogate the function of an individual cell type for a ubiquitously expressed gene makes it difficult to draw definitive conclusions. Future studies may include the use of conditional under- and/or overexpression models in

This article is open access and distributed under the terms of the Creative Commons Attribution Non-Commercial No Derivatives License 4.0. For commercial usage and reprints, please e-mail Diane Gern.

Originally Published in Press as DOI: 10.1165/rcmb.2023-0030ED on February 16, 2023

macrophages or macrophage depletion *in vivo* followed by adoptive transfer of the appropriate cell type. The second limitation of this study was the lack of investigation of eicosanoids. There is a complex relationship between the production and biological activity of lipid mediators that depends on receptor expression and differential and context-dependent activities/roles of PGE2 in different cell types. Simplistically, eicosanoids can be divided into antiinflammatory prostaglandins and proinflammatory leukotrienes, both derived from arachidonic acid. Prostaglandins are produced via the actions of COX-2, whereas leukotrienes are produced by arachidonate 5-lipoxygenase activity. Inhibition of COX-2 may result in the shunting of arachidonic acid metabolism into the production of leukotrienes (10). Therefore, in this study in which, presumably, and most likely, prostaglandin production is decreased, there may be a consequential increase in leukotrienes, which will drive different inflammatory processes. Furthermore, in our own studies on the roles of PGE2, we discovered marked differences in the amounts of PGE2 that are released by different lung cell types (11). The profiling of lipid mediators and the use of receptor antagonists can answer these questions in future studies.

Overall, this study provides new clues to the old question of why and how macrophage function is dysregulated in COPD. The study raises interesting questions around the permanency of changes to the bone marrow niche, i.e., is this a transitory effect of systemic inflammation in smokers and COPD, and which population of macrophages is most affected? Several RNA sequencing and genome-wide association studies of circulating inflammatory cells in COPD have been performed, so it will be interesting to see if the same genetic switch and upregulation of lncRNA-Cox2 can be observed in circulating leukocytes. ■

Author disclosures are available with the text of this article at www.atsjournals.org.

Richard Y. Kim, Ph.D.
School of Life Sciences
University of Technology Sydney
Sydney, Australia

Brian Oliver, Ph.D.
School of Life Sciences
University of Technology Sydney
Sydney, Australia

and

Woolcock Institute of Medical Research
The University of Sydney
Sydney, Australia

References

1. Baker JR, Fenwick PS, Koss CK, Owles HB, Elkin SL, Fine J, *et al.* IL-36 receptor agonist and antagonist imbalance drives neutrophilic inflammation in COPD. *JCI Insight* 2022;7:e155581.
2. Liu W, Wang S, Wang J, Zheng R, Wang D, Yu R, *et al.* Neuromedin U induces pulmonary ILC2 activation via the NMUR1 pathway during acute RSV infection. *Am J Respir Cell Mol Biol* [online ahead of print] 13 Oct 2022; DOI: 10.1165/rcmb.2022-0123OC.
3. Allen DE, Donohue KC, Cadwell CR, Shin D, Keefe MG, Sohal VS, *et al.* Fate mapping of neural stem cell niches reveals distinct origins of human cortical astrocytes. *Science* 2022;376:1441–1446.
4. Hashimoto D, Chow A, Noizat C, Teo P, Beasley MB, Leboeuf M, *et al.* Tissue-resident macrophages self-maintain locally throughout adult life with minimal contribution from circulating monocytes. *Immunity* 2013; 38:792–804.
5. Guilliams M, De Kleer I, Henri S, Post S, Vanhoutte L, De Prijck S, *et al.* Alveolar macrophages develop from fetal monocytes that differentiate into long-lived cells in the first week of life via GM-CSF. *J Exp Med* 2013;210:1977–1992.
6. Russell RE, Culpitt SV, DeMatos C, Donnelly L, Smith M, Wiggins J, *et al.* Release and activity of matrix metalloproteinase-9 and tissue inhibitor of metalloproteinase-1 by alveolar macrophages from patients with chronic obstructive pulmonary disease. *Am J Respir Cell Mol Biol* 2002;26:602–609.
7. Hodge S, Hodge G, Ahern J, Jersmann H, Holmes M, Reynolds PN. Smoking alters alveolar macrophage recognition and phagocytic ability: implications in chronic obstructive pulmonary disease. *Am J Respir Cell Mol Biol* 2007;37:748–755.
8. Salih MM, Robinson EK, Malekos E, Perez E, Capili A, Kim K, *et al.* *LincRNA-Cox2* regulates smoke-induced inflammation in murine macrophages. *Am J Respir Cell Mol Biol* 2023;68:511–522.
9. Kunkel SL, Spengler M, May MA, Spengler R, Larrick J, Remick D. Prostaglandin E2 regulates macrophage-derived tumor necrosis factor gene expression. *J Biol Chem* 1988;263:5380–5384.
10. Ganesh R, Marks DJ, Sales K, Winslet MC, Seifalian AM. Cyclooxygenase/lipoxygenase shunting lowers the anti-cancer effect of cyclooxygenase-2 inhibition in colorectal cancer cells. *World J Surg Oncol* 2012;10:200.
11. Van Ly D, Faiz A, Jenkins C, Crossett B, Black JL, McParland B, *et al.* Characterising the mechanism of airway smooth muscle β2 adrenoceptor desensitization by rhinovirus infected bronchial epithelial cells. *PLoS One* 2013;8:e56058.