

Correspondence

Reply

To the Editor:

We thank Hervier et al¹ for providing comments on our article² characterizing natural killer (NK) cells in the human lung. Based on analysis of CD69 expression, our results suggest that most CD56^{bright} NK cells are tissue-resident, whereas most CD56^{dim} NK cells in the lung circulate.² Our results also indicate that the total lung NK cell population is largely hypofunctional. Hervier et al¹ regret the fact that we studied NK cell functionality only as a whole, that is, regardless of the cells' status as tissue-resident (CD69-expressing) or circulating.¹ Furthermore, they express that it also would have been of interest to study NK cell function following stimulation with IL-15, which is locally produced in lung. We believe that these are important questions worth considering in more detail.

In our study, we showed that both CD56^{bright} and CD56^{dim} NK cells in lung tissue are hypofunctional compared with the corresponding subsets in peripheral blood. Sorted CD56^{bright} lung NK cells produced less TNF and GM-CSF after stimulation with PMA/ionomycin. Similarly, sorted CD56^{dim} NK cells responded less well in response to stimulation with K562 cells and rituximab-coated 721.221 cells, as measured with assays for both degranulation and production of TNF. Overall, our results showed that lung CD56^{dim} and CD56^{bright} NK cells are hyporesponsive to several distinct stimuli and by multiple functional readouts.² Given that most CD56^{bright} NK cells express CD69, our results indicate that the lung-resident NK cells are also hyporesponsive. In preliminary experiments, lung NK cells responded to IL-15 by upregulation of Ki67, a marker of proliferation, and increased expression of granzyme B (unpublished data, Marquardt et al, 2017). However, more extensive comparative studies are required to draw more firm conclusions whether there are distinct effects of IL-15 on tissue-resident NK cells.

Several recent studies of tissue-resident lymphocytes, including NK cells, have revealed that there is considerable phenotypic and functional heterogeneity among tissue-resident lymphocyte subsets.³ In addition to CD69, tissue-resident lymphocytes can be characterized by the expression of adhesion molecules, for example, CD49a and CD103. These molecules facilitate retention in specific parts of tissues. For example, CD103 binds to E-cadherin, which is predominantly expressed on epithelial cells and, as such, identifies intraepithelial tissue-resident lymphocytes.^{4,5} Although the expression of CD69 on

NK cells in tissues provides a good indication of tissue-residency, CD69 is also upregulated upon NK cell activation. Therefore, analysis of CD69 in combination with other markers of tissue-residency, *for example*, CD103 and CD49a, would be desirable to further dissect the characteristics of *bona-fide* human lung-resident NK cells.

The recent findings by Hervier et al of increased frequencies of NKp46⁺ cells in lungs of patients with antisyndetase syndrome,⁶ as well as reports on increased CD69 expression on intratumoral NK cells,⁷ further highlight the need for more detailed studies of human lung-resident NK cells, in health and various settings of human disease. Indeed, a better understanding of these cells could lead to improved insights into disease pathogenesis, as well as promoting the identification of new drug targets or diagnostic tools in lung-associated conditions.

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