On the Move: The Commander IL-4 Leads the Cell Army in Collective Migration

The airway epithelium maintains an important barrier between the external and internal environments. Disruption of this barrier, which can be initiated by a range of insults such as noxious particles, house dust mites, and fungal allergens, allows the penetration of such particles or allergens through the barrier, and this has been shown to trigger inflammation and airway remodeling (1). The T-helper cell type 2 (Th2) cytokines IL-4 and IL-13 are key mediators in driving and exacerbating this allergic inflammation and airway remodeling (2, 3).

However, in addition to triggering an inflammatory response, disruption of the barrier has profound effects on epithelial cell biology including loss of tight- and adherens-junction integrity, and subsequent actin cytoskeleton reorganization. The actin cytoskeleton is critical for maintaining cell tension, which itself relies on interactions with both cell–cell adhesion and cell/extracellular matrix (ECM) complexes (4). Interestingly, IL-4 has also been shown to lead to reduced expression of the tight-junction proteins zonula occludens-1 (ZO-1) and occludin, along with reduced adhesion of epithelial cells to the ECM, both of which are necessary for cell migration (3).

In this issue of the *Journal*, Lee and colleagues (pp. 420–433) demonstrate the role of integrins $\alpha_v\beta_5$ and $\alpha_v\beta_6$ in IL-4–induced collective cell migration via the focal adhesion kinase (FAK) signaling pathway in well-differentiated primary human nasal epithelial cells (HNECs) (5). The authors use a combination of approaches, including live-cell imaging, a vertex model that links cell mechanics to cell shape and motility, and immunostaining of nasal mucosa specimens obtained from patients with allergic rhinitis. Integrins are transmembrane mechanoreceptors that connect the intracellular actin cytoskeleton to the ECM. These proteins regulate bidirectional mechanosignaling and play an important role in epithelial cell migration (6). First, the authors show that in HNECs, IL-4 treatment leads to disruption of tight junctions, adherens-junction remodeling, and actin cytoskeleton reorganization and cell shape changes resulting in the transition of cells to an "unjammed" state (7), all of which are gateways to epithelial collective cell migration. Collective cell migration is a particular form of migration that is extensively used during development and for repair after injury. In this process, groups of cells that are uniformly polarized migrate together by remaining attached to neighboring cells and forming adhesive structures (focal adhesions [FAs]) with the ECM through which traction forces can be exerted to drive the cells along (8).

Using live-cell imaging of *in vitro* wound healing after scratch injury of HNEC monolayers, the authors demonstrate that IL-4 increases cell migration. In addition, IL-4 treatment of HNECs leads to increased expression of αv, β5, and β6 integrin subunits, as well as FAK phosphorylation, with the majority of integrin-positive HNECs expressing the basal marker Keratin 5. Interestingly, the authors show that IL-4–induced collective cell migration and FAK phosphorylation can be significantly reversed by inhibiting the integrins αvβ5 or αvβ6 or by using decanoyl-RVKR-chloromethylketone, a pan proprotein convertase inhibitor that inhibits αv subunit processing, suggesting that αvβ5 and αvβ6 mediate IL-4–induced collective HNEC migration through FAK signaling. Moreover, a careful examination of αvβ5 and αvβ6 integrin localization by immunostaining in nasal brushings from control subjects and patients with allergic rhinitis indicates a correlation between these integrins and epithelial repair after injury. Thus, in keeping with the increasing implications of integrin dysregulation in various lung diseases (9), integrins are convincingly shown to be important for lung repair and regeneration.

Intriguingly, in their *in vitro* scratch assays, Lee and colleagues consistently observed an initial transient contraction of the cell monolayer away from the leading edge, with reduced expression of phosphorylated FAK in response to IL-4. Although these data are highly compelling, they lead to further questions regarding the mechanoregulatory events underlying this pivotal yet understated finding. In addition to regulating FA maturation and turnover, FAK is known to have a role in E-cadherin–based cell–cell contact (10). As such, questions arise as to whether the changes in phosphorylated FAK levels reflect or lead to any alterations in FAs themselves and/or in E-cadherin during the cell contraction period. Moreover, FA assembly and disassembly is a highly dynamic process, and thus live-cell imaging techniques, such as those used in this study, would be useful for revealing real-time changes in focal contact maturation and dynamics after IL-4 treatment. Furthermore, given that directional epithelial migration is driven by changes in cell–cell and cell–ECM adhesions, as well as internal actin traction forces (4), measurements of cellular traction force will be required to confirm the authors' speculation that the observed cell hypercontraction is due to an imbalance of cell–cell and cell–ECM adhesions.

Because abundant evidence has implicated Rho-family GTPases, transforming growth factor-β, and YAP/TAZ signaling as downstream effectors in mechanotransduction (6), in the future it would be informative to undertake transcriptomic analyses to determine whether these known mechanosensitive proteins/pathways are aberrant in IL-4–treated HNECs. Precisely how the transient cell contraction then leads to accelerated collective migration remains an important question. This is likely to be more challenging to determine; however, bioinformatics approaches may be useful in providing a better understanding of the complex feedback through the lens of mechanobiology pathways or interactome analyses.

Currently, it is unknown whether the role of IL-4– and integrin-mediated cell migration is specific to this particular

cytokine. Considering the high homology and redundant functionality of IL-4 and IL-13, and their common role in driving inflammation associated with allergy (11), as well as in regulating cell–ECM interactions (2), it would be interesting to investigate whether IL-13 plays a similar role, and if so, whether the same or alternative/additional integrins are involved.

Although further in-depth investigations will be required to fully define the relationship between IL-4 treatment and mechanosignaling in respiratory epithelial cells, this study provides an important interdisciplinary perspective that links a Th2 cytokine to integrin-mediated mechanotransduction and collective cell migration. Furthermore, it highlights the major roles played by biological events (such as mechanosignaling) that, in addition to inflammation, drive epithelial remodeling in allergic respiratory disease. ■

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

Sek-Shir Cheong, Ph.D.
Charlotte H. Dean, Ph.D.
National Heart and Lung Institute
Imperial College London
London, United Kingdom

ORCID ID: 0000-0002-8846-5472 (C.H.D.).

References

1. Lloyd CM, Robinson DS. Allergen-induced airway remodelling. *Eur Respir J* 2007;29:1020–1032.
2. Ramirez-Icaza G, Mohammed KA, Nasreen N, Van Horn RD, Hardwick JA, Sanders KL, *et al*. Th2 cytokines IL-4 and IL-13 downregulate paxillin expression in bronchial airway epithelial cells. *J Clin Immunol* 2004;24:426–434.
3. Georas SN, Rezaee F. Epithelial barrier function: at the front line of asthma immunology and allergic airway inflammation. *J Allergy Clin Immunol* 2014;134:509–520.
4. Lecuit T, Lenne P-F. Cell surface mechanics and the control of cell shape, tissue patterns and morphogenesis. *Nat Rev Mol Cell Biol* 2007;8:633–644.
5. Lee SN, Ahn JS, Lee SG, Park JA, Lee HS, Choi AMK, *et al*. Integrins $\alpha_v\beta_5$ and $\alpha_v\beta_6$ mediate IL-4–induced collective migration in human airway epithelial cells. *Am J Respir Cell Mol Biol* 2019;60:420–433.
6. Sun Z, Guo SS, Fässler R. Integrin-mediated mechanotransduction. *J Cell Biol* 2016;215:445–456.
7. Park JA, Kim JH, Bi D, Mitchel JA, Qazvini NT, Tantisira K, *et al*. Unjamming and cell shape in the asthmatic airway epithelium. *Nat Mater* 2015;14:1040–1048.
8. Ladoux B, Mège RM. Mechanobiology of collective cell behaviours. *Nat Rev Mol Cell Biol* 2017;18:743–757.
9. Teoh CM, Tan SS, Tran T. Integrins as therapeutic targets for respiratory diseases. *Curr Mol Med* 2015;15:714–734.
10. Mitra SK, Hanson DA, Schlaepfer DD. Focal adhesion kinase: in command and control of cell motility. *Nat Rev Mol Cell Biol* 2005;6:56–68.
11. Kau AL, Korenblat PE. Anti-interleukin 4 and 13 for asthma treatment in the era of endotypes. *Curr Opin Allergy Clin Immunol* 2014;14:570–575.