

## EDITORIAL

# Neutrophil-Mediating Lung Diseases and Integrin $\alpha$ subfamily, $\alpha 4$ and $\alpha 9$

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## ABSTRACT

Neutrophil-mediating acute lung injury like aspiration pneumonia is a serious problem in the elderly. Although integrin  $\beta 2$  plays an important role in neutrophil-mediating lung diseases, integrin  $\beta 2$ -independent neutrophil transmigration pathways have also been reported. Several members of the integrin  $\alpha$  subfamily ( $\alpha 2\beta 1$ ,  $\alpha 3\beta 1$ ,  $\alpha 4\beta 1$ ,  $\alpha 5\beta 1$ ,  $\alpha 6\beta 1$  and  $\alpha 9\beta 1$ ) are known to be expressed on neutrophils, and up-regulated by activation. And these integrins on

systemic neutrophils might mediate migration into the site of pulmonary inflammation. Especially, integrin  $\alpha 9\beta 1$  and integrin  $\alpha 4\beta 1$  have been also shown to mediate adhesion to vascular cell adhesion molecule (VCAM)-1. In this draft, author focused lung diseases and the involvement of integrin  $\alpha 9$  and  $\alpha 4$ . And the future direction as the therapeutic strategy against neutrophil-mediating lung injury was also discussed.

**Key words:** Integrin  $\alpha 9\beta 1$ ; Integrin  $\alpha 4\beta 1$ ; Neutrophil

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## NEUTROPHIL-MEDIATING LUNG DISEASES

Integrins are  $\alpha\beta$  heterodimeric, transmembrane proteins that mediate both cell-substrate and cell-cell interactions<sup>[1]</sup>. As one of their functions, integrins mediate cell migration. Neutrophils play an important role in various kinds of lung diseases<sup>[2]</sup> including aspiration pneumonia<sup>[3,4]</sup>, chronic obstructive lung disease, idiopathic pulmonary fibrosis, and drug-induced lung injury<sup>[1]</sup>, and circulating neutrophils are recruited into the alveolar space by transmigrating across vascular endothelial cells. Integrin  $\beta 2$  is known to play a central role in extravasation of leukocytes, but integrin  $\beta 2$ -independent neutrophil transmigration pathways have also been reported in certain conditions including animal models, which neutrophil emigration is induced by bacterial pneumonia<sup>[5]</sup>. Although adhesion molecules that mediate the integrin  $\beta 2$ -independent pathways still remained uncertain, integrin  $\alpha$  subfamily is one of the candidates. Several members of the integrin  $\alpha$  subfamily ( $\alpha 2\beta 1$ ,  $\alpha 3\beta 1$ ,  $\alpha 4\beta 1$ ,  $\alpha 5\beta 1$ ,  $\alpha 6\beta 1$  and  $\alpha 9\beta 1$ ) are known to be expressed on neutrophils, and up-regulated by activation<sup>[4,5]</sup>. The  $\alpha 9$  subunit cDNA sequence is 41% identical to the integrin  $\alpha 4$  subunit sequence, but no more than 27% identical to any other integrin subunit, showing  $\alpha 9$  and  $\alpha 4$  as specific members of a

subfamily of integrin  $\alpha$  subunits<sup>[6]</sup>. Furthermore, both  $\alpha 4\beta 1$  and  $\alpha 9\beta 1$  has a receptor against vascular cell adhesion molecule-1 (VCAM-1) as a ligand<sup>[4]</sup>. In this draft, author focused lung diseases and the involvement of integrin  $\alpha 9$  and  $\alpha 4$ . And I discussed the future direction as the therapeutic strategy against neutrophil-mediated lung injury.

## INTEGRIN $\alpha 4\beta 1$

Integrin  $\beta 1$  and  $\beta 7$  are known as the partners of integrin  $\alpha 4$ . Integrin  $\alpha 4\beta 1$  constitutively expressed on the cell surface of lymphocytes, monocytes, eosinophils, and neutrophils, and is known to be up-regulated by stimuli including inflammatory cytokines<sup>[4,7]</sup>. Since the expression level of integrin  $\alpha 4\beta 1$  on human neutrophil is very weak comparing with that of mouse, human neutrophil was supposed not to express integrin  $\alpha 4\beta 1$  before. After that, many papers have been reported about the expression of  $\alpha 4\beta 1$  on human neutrophils<sup>[4,5]</sup>. In animal model, integrin  $\alpha 4\beta 1$  involved in mice pneumonia by *Streptococcus pneumoniae*<sup>[8]</sup>. Integrin  $\alpha 4\beta 1$  on neutrophils has a role in adhesion and migration to move into the site of inflammation, and integrin  $\alpha 4\beta 1$  mediate integrin  $\beta 2$ -independent neutrophil accumulation<sup>[7]</sup>. Not only in animal model, but also human septic cases showed up-regulation of integrin  $\alpha 4\beta 1$  on neutrophils<sup>[9,10]</sup>. On the other hand, increased expression of integrin  $\alpha 4\beta 1$  on neutrophils was not recognized in early stage of acute respiratory distress syndrome (ARDS) cases in another paper<sup>[11]</sup>. They discussed that increased expression level of adhesion molecule might be delay, several hours later. Although literatures about molecular mechanism of involvement of integrin  $\alpha 4\beta 1$  on neutrophils were limited and not fully elucidated, these evidences showed the possibility of integrin  $\alpha 4\beta 1$  mediating human neutrophilic pulmonary inflammation.

## INTEGRIN $\alpha 9\beta 1$

Integrin  $\alpha 9\beta 1$  also expressed on human neutrophils and VCAM-1 on the surface of vascular endothelial cells, and integrin  $\alpha 9\beta 1$  plays an important role in the extravasation of neutrophils to the site of inflammation<sup>[4]</sup>. Author reported increased expression levels of integrin  $\alpha 9\beta 1$  and CD11b on circulating neutrophils and elevated serum IL-17A in elderly aspiration pneumonia cases<sup>[12]</sup>. Previously, the possibility of cross-talking signaling pathways playing a role in modifying migration between integrin  $\beta 2$  and integrin  $\beta 1$  has been already reported<sup>[13]</sup>, but the signal pathway of integrin  $\alpha 9\beta 1$  is not well understood. Recently, it was reported that integrin  $\alpha 9\beta 1$ /VCAM-1 interaction could be trigger to the activation of signal pathway to modulate neutrophil apoptosis and mediating inflammation<sup>[14,15]</sup>. And during the process, integrin  $\alpha 9\beta 1$  on human neutrophils depends on the activated mitogen-activated protein kinase<sup>[14]</sup> and phosphoinositol 3-kinase<sup>[15]</sup> pathway. Although mechanism of signal transduction of integrin  $\alpha 4\beta 1$  may be different from that of integrin  $\alpha 4\beta 1$ , integrin  $\alpha 9\beta 1$  on human neutrophils also mediate lung injury in a certain condition.

## INTEGRIN, LUNG CANCER AND LUNG FIBROSIS

Notable characteristics of integrin  $\alpha$  subfamily are diversity and multiplicity. For example, integrin  $\alpha 9\beta 1$  expressed on cancer cells including lung cancer, and promoted tumor growth<sup>[16]</sup>. Gupta SK *et al*<sup>[16]</sup> reported that endogenous  $\alpha 9\beta 1$  mediated epithelial-mesenchymal transition in lung carcinoma cells and increased cell migration and invasiveness. Furthermore, integrin  $\beta 1$  is involved

in lung fibrosis<sup>[17]</sup>. Zhao X *et al*<sup>[17]</sup> demonstrated that platelet derived growth factor BB (PDGF-BB) induces focal adhesion kinase (FAK) activation and lung fibroblast migration after receiving PDGF-BB stimulation. FAK associated with integrin  $\beta 1$  and PDGF-BB stimulated lung fibroblasts, then FAK activation and  $\beta 1$  integrin mediated fibroblast migration. About pulmonary fibrosis, activating transforming growth factor (TGF)  $\beta$  regulated by integrin  $\alpha v\beta 6$  is also well known<sup>[18]</sup>. Various kinds of integrin related cancer invasion and lung fibrosis, and further detailed examination is necessary.

## FUTURE DIRECTION

From the point of above description, inhibiting integrin  $\alpha 4$ /VCAM-1 and/or integrin  $\alpha 9$ /VCAM-1 interaction might attenuate neutrophilic lung inflammation at least in a certain condition. As the therapeutic options to inhibit integrin  $\alpha 4/\alpha 9$ , functional blocking anti-integrin  $\alpha 4\beta 1$  or  $\alpha 9\beta 1$  monoclonal antibody, disintegrin, and small molecules inhibiting intracellular signal transduction are supposed to be candidates. As anti-monoclonal antibody inhibiting integrin  $\alpha 4\beta 1$ , commercial-based drug, natalizumab (Biogen-Idec, Durham, NC, USA) is already available against multiple sclerosis in many countries<sup>[19]</sup>. The major adverse effect of natalizumab is immunosuppression. Specific inhibition of neutrophilic lung injury without immunosuppression should be expected, when against severe pneumonia patients for their treatment. In this meaning, natalizumab is not sufficient for treatment of sepsis cases. As disintegrin derived from venomous vipers, Marcinkiewicz C *et al* reported that functional inhibitor of integrin  $\alpha 9$ , named EC3 and EC6 inhibited  $\alpha 9\beta 1$ -mediated adhesion to VCAM-1 and of neutrophil migration *in vitro* experiment<sup>[20]</sup>. And the peptide containing a novel MLDG motif shared by both of these disintegrins also inhibited  $\alpha 9\beta 1$ - and  $\alpha 4\beta 1$ -mediated adhesion to VCAM-1. They also reported that disintegrin might provide a basis for the development of potent drugs that could be used to inhibit emigration of neutrophils into pulmonary inflammation<sup>[19]</sup>. Furthermore, paxillin inhibitors are also promising candidate. Integrin  $\alpha 4$  cytoplasmic domain enhances cell migration and spreading, which effects depend on interaction with the adaptor protein, paxillin, and to migrate circulating neutrophils into the site of inflammation<sup>[21,22]</sup>. This interaction is critical for integrin dependent cell adhesion, spreading, and migration as it provides a needed connection to the actin cytoskeleton<sup>[18]</sup>. And Rose DM discussed that  $\alpha 4$ -paxillin interaction was a potentially therapeutic target in controlling leukocyte trafficking<sup>[18]</sup>.

So far, mechanism between integrin  $\alpha 4/\alpha 9$  and neutrophil migration is not well elucidated, and the further study would be necessary. But author supposes they are promising therapeutic strategy against neutrophil-mediated lung injury in the future.

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