**Invited JII**

**Innate immunity of the lung: basic mechanisms and disease implications**

Hartl D1,2, Tirouvanziam R3, Laval J1, Greene C4, Habiel D5, Sharma L6, Yildirim OE7, Dela Cruz CS6, Hogaboam CM5

Affiliations:

1Children’s Hospital, University of Tübingen, Tübingen, Germany.

2Roche Pharma Research & Early Development (pRED), Immunology, Inflammation and Infectious Diseases (I3) Discovery and Translational Area, Roche Innovation Center Basel, Switzerland

3Department of Pediatrics, Emory University School of Medicine, Atlanta, GA 30322; Center for Cystic Fibrosis and Airways Disease Research, Children's Healthcare of Atlanta, Atlanta

4Department of Medicine, Royal College of Surgeons in Ireland Education and Research Centre, Beaumont Hospital, Dublin, Ireland

5Division of Pulmonary and Critical Care Medicine, Department of Medicine, Cedars-Sinai Medical Center, Los Angeles, CA, 90048

6Section of Pulmonary, Critical Care and Sleep Medicine, Department of Internal Medicine and Microbial Pathogenesis, Yale University School of Medicine, New Haven, CT 06520, USA

7Comprehensive Pneumology Center, Institute of Lung Biology and Disease, Helmholtz Zentrum München, Member of the German Center for Lung Research, Neuherberg, Germany

Correspondence:

Prof. Dr. Dominik Hartl

Children’s Hospital, University of Tübingen, Tübingen, Germany

E-Mail: dominik.hartl@med.uni-tuebingen.de

Phone: +49707129-81460

Fax: +49707129-4449

**ABSTRACT**

The respiratory tract is daily faced with 10,000 litres of inhaled air. While the majority of air contains harmless environmental components, the pulmonary immune system has also to cope with harmful microbial or sterile threats and react rapidly to protect the host at this intimate barrier zone. The airways are endowed with a broad armamentarium of cellular and humoral host defense mechanisms, most of them belonging to the innate arm of the immune system. The complex interplay between resident and infiltrating immune cells and secreted innate immune proteins shapes the outcome of host-pathogen, host-allergen and host-particle interactions within the mucosal airway compartment. Here, we summarize and discuss recent findings on pulmonary innate immunity and highlight key pathways relevant for biomarker and therapeutic targeting strategies for acute and chronic diseases of the respiratory tract.

**MANUSCRIPT**

*The lung as innate immune sentinel*

The human respiratory tract is daily faced with 10,000 litres of inhaled air, containing harmless environmental components, but also potentially airborne pathogens. This constant exposure requires a fine-tuned and rapidly acting pulmonary immune system in order to immediately sense harmful microbial or sterile threats and to protect the host at this intimate contact zone [1-3]. For that purpose, the airways are endowed with a broad armamentarium of cellular and humoral host defense mechanisms, most of them belonging to the innate arm of the immune system [1,3]. The complex interplay between resident (airway epithelial cells) and infiltrating immune cells acting in concert with secreted innate immune proteins, such as defensins, mucins or collectins, shapes the outcome of host-pathogen, host-allergen and host-particle interactions within the airway microenvironment. Airway epithelial cells, dendritic cells and (in the lower airways) alveolar macrophages are the initial checkpoints that encounter inhaled antigens and trigger pro-inflammatory or tolerogenic/anti-inflammatory downstream immune responses. Major mediators communicating between airway sentinel cells and bone marrow-derived immune cells are chemoattractants, such as lipid mediators (prototypically eicosanoids/leukotrienes) and chemokines [4-7]. Of particular importance for the airway microenvironment are the CXC chemokines CXCL1-8 and CXCL12 and the CC chemokines CCL2, CCL17 (TARC), CCL18 (PARC) and CCL20. The biological effect of chemokines is supported by distinct cytokines that are released by local airway cells and induce microenvironmentally-tailored immune contexts, particularly IL-1-alpha and IL-1-beta, IL-10, IL-17, IL-23, IL-25, IL-33 and TSLP. Through the integrated action of pro-inflammatory lipid mediator, chemokine and cytokine mediators, different immune cell populations are sequentially recruited into the bronchoalveolar and lung parenchymal compartments. Initially, neutrophils are attracted and localize mainly in the bronchoalveolar space where they engage in short-term host-pathogen interactions [8], followed by monocyes and lymphocytes for more sustained/chronic host defense functions, with the latter having a clear tissue predominance for infiltrating the lung parenchyma rather than the bronchoalveolar space [9,10]. Within the pulmonary tissue, lymphocytes form organized ectopic tertiary lymphoid organs, termed bronchus-associated lymphoid tissue (BALT) or inducible BALT (for humans) [11]. Besides these more established components of the pulmonary immune response, recent studies highlight novel immune cells and mediators, such as innate lymphoid cells (ILCs), mucosal-associated invariant T cells (MAITS), chitinase-like proteins and others. Here we aim to summarize the different components of the innate pulmonary immune response with an emphasis on novel directions for translational research and drug development.

*Airway epithelial cells as first line of innate immune sensing*

Pathogens are sensed by a variety of receptors expressed by airway epithelial cells. These include various families of pattern recognition receptors (PRRs) represented, for example, by Toll-like receptors (TLRs), RIG-I-like receptors (RLRs), Protease-activated receptors (PARs), Nod-like receptors (NLRs), C-type lectin receptors (CLRs), and the Bitter and Sweet taste receptors. TLR expression and function in airway epithelium has been widely studied. These PRRs are expressed by epithelial cells throughout the respiratory tract and respond rapidly to local microbial and host-derived factors present as a result of infection or tissue damage. Following ligand recognition TLRs then activate intracellular downstream signalling cascades leading to changes in gene expression associated with typical innate immune responses and activation of adaptive immunity. Tengroth *et al.* [12] recently described the importance of the nucleic acid-sensing TLRs (TLR3, TLR7, TLR9) and the RLRs, RIG-I and MDA-5, which recognise replicating RNA viruses in infected cells, in nasal epithelium. These studies using nasal biopsies and primary human nasal epithelial cells (HNECs) stimulated with artificial TLR and RLR agonists demonstrated how these receptors generate robust IL-6, IL-8 and IFN-β responses and act as a first line of defence against viruses invading the respiratory tract. In another study reactive oxygen species (ROS) produced via Duox2 in influenza A virus-infected nasal epithelial cells were shown to increase RIG-I and MDA-5 mRNA in a process believed to resist IAV infection [13]. Kim *et al.* also evaluated inflammatory and antiviral responses to human virus infection in HNECs from chronic rhinosinusitis patients, wherein decreased MDA-5 and IFN-β expression were evident, and human rhinovirus clearance was slightly impaired [14].

Another type of PRR essential for innate immunity in the pulmonary environment are CLRs that sense fungal patterns. On bronchial epithelial cells this is mainly orchestrated by dectin-1 which mediates recognition of β-glucan motifs in *Aspergillus fumigatus* and house dust mite (HDM) [15] leading to secretion of the dendritic cell chemokine CCL20. Other non-fungal allergens, and specifically those with proteolytic properties such as Derp1 and cockroach allergen, can elicit allergic airway inflammation via PAR-2 when administered through the mucosa [16]. Interestingly according to Post *et al.* [17] who studied allergic sensitisation and HDM-induced allergic airway inflammation, whilst PAR-2 contributes to HDM-induced IgE responses it is dispensable for the induction of pro-inflammatory cytokine by HDM. In another study others demonstrated that in addition to producing cytokines, allergen-activated airway epithelial cells can also release uric acid [18], thereby promoting TH2 sensitisation and amplifying allergic inflammation (reviewed in [19]).

Among NLRs, NOD1, NOD2 and NLRP3 are expressed by airway epithelial cells. Expression of NOD1 has been shown to be down-regulated during pollen season among patients with allergic rhinitis [20] and its normal activation can reduce airway hyper-responsiveness accompanied by a reduction of allergen-specific T-cell proliferation in allergen-induced lung inflammation [21]. NLRP3 mediates cellular responses to inhaled particular matter e.g. PM10, and has recently been elegantly shown to have an important role in innate, but not adaptive immune responses in airway epithelial cells [22]. A novel NLR termed NLRX-1 has been identified in nasal epithelium that is activated by double stranded RNA and participates in rhinoviral-mediated disruption of polarised airway epithelial cell barrier function [23].

One particularly exciting new finding on PRR expression and function in airway epithelial cells in recent years has been the discovery of the G-protein coupled Bitter and Sweet Taste receptors (T2R and T1R, respectively) in respiratory epithelia (reviewed in [24]). Extraoral taste receptors have been detected in human bronchial epithelial cells and specialised solitary ‘sinonasal chemosensory cells’ (SCCs) in the upper respiratory tract [25,26]. Bitter taste receptors are activated by quorum-sensing molecules whereas sweet receptors respond to sugars. For example the bitter taste receptor T2R38 is activated by homoserine lactones from *Pseudomomas aeruginosa*, causing an increase in nitric oxide and enhanced mucociliary clearance [27]. Lee *et al.* [28] have studied the regulation of bitter and sweet taste receptors in the upper airways and found that antimicrobial peptide induction by T2R, is repressed by activation of sweet taste receptor T1R2/3. Thus activation of T12/3 and repression of T2R due to increased intranasal glucose may be responsible for chronic rhinosinusitis.

Beyond these major families of PRR, new roles in innate immunity have emerged recently for other receptors expressed by airway epithelium. Selected examples include (i) the short-chain fatty acid receptor GPR41 which is present at higher than normal levels in cystic fibrosis bronchial epithelium and is responsible for exaggerated IL-8 induction in these cells in response to SCFAs from anaerobic bacteria in the lung [29], (ii) the fraktalkine receptor CX3CR1 which was recently reported to mediate respiratory syncytial virus induced cytokine production in primary human airway epithelial cells [30], and (iii) an isoform of the coxsackievirus and adenovirus receptor (CAR), CAREx8a, expressed on the apical surface of airway epithelial cell in culture, that enhances adenovirus entry into cells by co-opting neutrophils [31].

*Innate immune effector cells: neutrophils and macrophages*

Neutrophils are the most abundant subset of leukocytes patrolling in blood in search of potential injuries and stimuli (e.g. PAMPs, DAMPs, chemokines, cytokines and lipid mediators), which induce their rapid recruitment to peripheral sites of inflammation. The primary role fulfilled by neutrophils is pathogen killing and removal. Neutrophils are conventionally thought of as short-living cells with limited capacity for *de novo* mRNA transcription and protein production. However, recent evidence show that they are endowed with extensive adaptive abilities, which underlie an equally extensive functional complexity [8].

Human blood neutrophil lifespan was previously estimated to be shorter than a day, however recent studies suggested an upper limit of closer to 5 days, even in the absence of inflammation [32]. This topic remains controversial and requires further investigation. In addition to circulating freely in blood, a large fraction of neutrophils is tethered to the lining of the liver, spleen, bone marrow and lung vasculature, and referred to as the “marginated” pool [33]. Within the lung alone, this pool constitutes the most prominent reservoir of neutrophils in the systemic circulation (approximately 40% of total body neutrophils) [34]. Key protective mechanisms have been attributed to the lung marginated pool, because it enables rapid neutrophil recruitment inside of the tissue following injury and/or infection. Additionally, the lung vasculature has been involved in neutrophil “de-priming” via its ability to filter and inactivate primed cells before they are re-released in the blood stream [35]. However, this mechanism of neutrophil retention is impaired in acute respiratory distress syndrome (ARDS) patients, resulting in the exposure of peripheral organs to primed systemic neutrophils, leading to severe complications [36].

As the first circulating immune subset recruited into the lung upon infection or sterile inflammation, neutrophils orchestrate both pro- and anti-inflammatory actions therein, as required to efficiently kill pathogens and resolve inflammation. In fulfilling these tasks, neutrophils undergo profound phenotypic and functional changes, delineating distinct fates. Indeed, various neutrophil fates can be observed in blood and tissue under both homeostatic and pathological conditions such as infection, autoimmunity and cancer [37]. Within the airways, recruited neutrophils display an activated phenotype (CD16high CD62Ldim CD11bhigh) under both healthy and inflamed conditions [38,39]. When crossing into the airway lumen, neutrophils further modulate surface expression of chemokine and Toll-like receptors, which together mediate core inflammatory signaling. These changes in neutrophil phenotypes include lower expression level of receptors for the CXCL8 (IL8) chemokine, CXCR1 and CXCR2, than in blood. By contrast, a large fraction of neutrophils isolated from the bronchoalveolar lavage fluid (BALF) of patients suffering from chronic airway inflammation upregulate CCR1, CCR2, CCR3, CCR5, CXCR3 and CXCR4 (as observed in CF, COPD and asthma), as well as TLR2, TLR4, TLR5 and TLR9 (as observed in CF and non CF-bronchiectasis), when compared to airway neutrophils from healthy subjects. These phenotypic changes are linked to neutrophil respiratory burst activity and/or bacterial killing and are concomitant to *de novo* protein synthesis, granule exocytosis and later induction of apoptosis [40-42]. While typical surface receptors are altered in neutrophils during chronic lung inflammation, further studies suggested more profound modulation of their metabolism and function. In particular, recent studies showed that upon recruitment into CF airways, neutrophils undergo significant activation of both cAMP Response Element Binding protein (CREB) and mechanistic (or mammalian) Target Of Rapamycin (mTOR) pathways, linked to surface upregulation of functional receptors (e.g., the Receptor for Advanced Glycation End-products -RAGE-) and metabolite transporters (e.g., glucose and amino acid transporters, Glut1 and ASCT2) [43-45]. Since CF airways are enriched in inflammatory mediators and nutrient such as glucose and amino acids, these results suggest the ability of neutrophils to adapt to specific microenvironments, through coordinated stress responses and metabolic reprogramming (reviewed in [46,47]. A hallmark of neutrophil activity within inflamed airways is the extracellular release of proteases and oxidases (e.g., neutrophil elastase -NE- and myeloperoxidase -MPO-) following granule mobilization to the cell surface. This process, well recognized in airway diseases such as CF and COPD, induces an upregulation of characteristic markers for secondary and primary granules (CD66b and CD63, respectively) on the surface of airway neutrophils, and results in high oxidative and proteolytic activities. The latter was shown to be responsible for the cleavage as key receptors involved in neutrophil antibacterial activities such as CXCR1, CD16 and CD14 [43,46-48]. Further studies revealed a role for airway neutrophils in the regulation of the adaptive immune system, further emphasizing the multidimensional importance of neutrophil plasticity [37]. For example, strong immunosuppressive function was identified in CF airway neutrophils, which can downregulate T-cell activity through the release and activation of arginase I, concomitant with granule exocytosis [49]. Activated neutrophils may also impact T-cells positively within CF airways but also within the lymphatic compartment, by displaying antigen-presenting cell capabilities (e.g., expression of CD80, CD86 and MHC II). Interestingly, CXCR4 expression, usually characteristic of immature or senescent neutrophils, is strongly upregulated on activated airway neutrophils and could relate to their acquired ability to egress from inflammatory tissues into lymphatic vessels [40,43,50]. Neutrophil transit toward lymph nodes has been associated with T-cell proliferation and therefore participates in the positive regulation of the adaptive immune response by neutrophils [50,51]. In some cases, lymphatic neutrophils could be used a “Trojan horses” by intracellular pathogens, which can use them as a vehicle to spread through the body [52,53].

Until recently, neutrophils have been primarily recognized as professional killers critical to the initial immune response that leads to pathogen clearance, using both intracellular killing by phagocytosis and extracellular killing by exocytosis of granule components. In the past decade, a third effector mechanism used by neutrophils has been described, which consists in the release of extracellular complexes or “traps” formed by decondensed nuclear DNA (e.g., after histone citrullination), histones, and cationic effectors such as NE and MPO. The deployment of neutrophil extracellular traps (NETs), termed “NETosis”, is believed to enable immobilization and possibly killing of pathogens, while precipitating neutrophil death [54,55]. While NETs have been associated with numerous pathologies including autoimmune disorders, and infectious diseases related to bacteria, fungi, as well as viruses, their role in acute and chronic inflammation has not yet been fully elucidated [8,54]. In the context of airway diseases, studies focusing on NETs have shown both beneficial and harmful effects of these structures (reviewed in [56]). The ability of NETs to spread out and trap microbes could trigger increased killing efficiency and reduction of pathogen burden. Mitigating these positive effects of NETs are the facts that pathogens such as *Staphylococcus aureus* were shown to developed NET-evasion strategies, and that the extracellular presence of host DNA, histones, NE, and MPO can cause direct or indirect cell toxicity and subsequent lung injury [57-59], as well as airway obstruction via an increase in mucus viscosity [60-62].

Taken together, numerous studies in the past decade have highlighted the adaptability of neutrophils in acute and chronic immune responses, contradicting the conventional view as “simple” cells, incapable of protein synthesis, with limited lifespan, and primarily relying on oxidative and proteolytic killing to carry out their function. The identification of novel neutrophil functions and regulatory mechanisms highlighted their role in balancing pro- and anti-inflammatory signaling, in order to promote a swift return to homeostasis and limit tissue damage and disease progression. The concept of “neutrophil heterogeneity” originated by Gallin et al in 1984 [63] has now emerged in full bloom, encompassing the formation of distinct subsets in both blood and peripheral tissues, and raising particular interest in the functional characterization of these subsets for novel neutrophil-targeted therapies for CF, COPD, neutrophilic asthma and other chronic airway diseases.

# Macrophages, first discovered by Ilya Metchnikoff, belong to the  [mononuclear phagocyte system](https://en.wikipedia.org/wiki/Mononuclear_phagocyte_system%22%20%5Co%20%22Mononuclear%20phagocyte%20system) and represent potent anti-microbial innate immune cells that are found in all tissues across the human body. Macrophages in the pulmonary compartment are classified and termed according to their anatomical location in the lung as alveolar or interstitial macrophages [64]. Since interstitial macrophages are less defined and more heterogeneous depending on the pulmonary subcompartment, the species studied and the disease model investigated, we will focus here on alveolar macrophages. The alveolar macrophage, 15 to 50 μm in diameter, is mainly located in the alveolar space and represents the predominant phagocytic and antigen-presenting cell in the human respiratory tract [65]. Under homeostatic/healthy conditions, alveolar macrophages are the most abundant cellular fraction within BAL fluids, while under acute or chronic inflammatory conditions other leukocyte populations, prototypically neutrophils (acute infections, CF or ARDS) and lymphocytes (sarcoidosis, allergic alveolitis/EAA), accumulate and shift this balance. Distinct to other tissue macrophage subsets, alveolar macrophages feature a remarkably high phenotypic, metabolic and functional plasticity [65-67]. Metabolically, alveolar macrophages exhibit a high basal glucose consumption and respiratory rate, yet a low respiratory burst activity. Phenotypically, alveolar macrophages directly reflect the alveolar host-environment interterface zone and contain granules of exogenous material, as exemplified in chronic smokers where alveolar macrophages accumulate in the BAL, are larger in diameter, activated and stain dark on cytospin preparations. By taking up inhaled environmental particles, pollutants, allergens and airborne microbes, alveolar macrophages serve as airway scavengers to keep the respiratory system clean and homeostatic. Another function related to their potent phagocytic capacity is the maintainance of surfactant homeostasis by engulfing and catabolizing local surrounding surfactant proteins, a functionality that is impaired in a severe disorder called pulmonary alveolar proteinosis (PAP), characterized by a dysfunction of the GM-CSF pathway and related to alveolar macrophage functions [68,69]. By sensing bronchoalveolar PAMP/MAMP and DAMP signals, alveolar macrophages integrate various microbial- and host-derived signals and respond with the concerted release of various pro- or anti-inflammatory mediators into the alveolar space, mainly cytokines such as IL-1-beta, IL-6, IL-10 or TNF-alpha. Recent studies further highlight that alveolar macrophages can have immunoregulatory roles. In murine asthma model systems, pulmonary macrophages were found to generate regulatory T cells to induce airway tolerance [70]. Alveolar macrophages were further found to attach to the alveolar wall (become “sessile”) and communicate with the alveolar epithelium to induce a immunosuppressive/-modulatory signal in order to dampen lung inflammation [71].

# Ontologically, tissue macrophages derive from either circulating/recruited monocytes or are established during embryogenesis (yolk sac and foetal liver) independently of monocytes, as supported by recent murine studies, and populate different organs as long-lived tissue-resident macrophages [64,72]. The precise origin of alveolar macrophages, however, across species remains a matter of intense ongoing investigations. Previous studies showed that alveolar macrophages develop from fetal monocytes that differentiate into long-lived mononuclear cells in the first week of life mediated through GM-CSF [73]. In murine fate-mapping studies, yolk sac derived erythro-myeloid progenitors have been identified as origins for several tissue-resident macrophage populations, including alveolar macrophages [74]. The airway environment/niche seems to be essential as well to shape the unique phenotype and functional characteristics of alveolar macrophages *in vivo* [75]. For a more detailed and comprehensive review on the ontogeny of alveolar macrophages we refer to a recent dedicated review [64].

# Macrophages have a remarkable functional and phenotypic diversity and can be dichotomized into M1 (IFN-gamma/classically activated) and M2 (IL-4/alternatively activated) polarization phenotypes with different roles in cancer, infection, allergy and fibrosis. Despite compelling evidence *in vitro*, the *in vivo* relevance of this classification has been challenged and is continuously refined given the high plasticity and heterogeneity of macrophages depending on the respective tissue compartment and disease model analyzed. For a more detailed review on macrophage activation, plasticity and M1/M2 polarization we refer to recent reviews in that field [76-78]. Overall, the complex multifunctionality of alveolar macrophages, interacting as APCs with T cells, clearing apoptotic and necrotic cells through efferocytosis, killing pathogens and releasing both pro- and anti-inflammatory factors, renders a good vs bad “cop” assignment for these cells for the majority of lung diseases challenging. *In vivo* macrophage depletion studies revealed different outcomes depending on the lung disease model and the time point of intervention used [64,65,79,80]. IPF is a prototypic example of a complex chronic lung disease where the role of pulmonary macrophages remains controversial. In brief, alveolar macrophages seem to play a dual role in lung fibrosis given their release of profibrotic factors (such as TGF-beta1 and PDGF) and thereby driving disease progression and, on the other hand, their potency to liberate proteases (MMPs) that can digest extracellular matrix and thereby act anti-fibrotic/fibrinolytic. For a more thorough discussion of macrophage phenotypes and functions in IPF we refer to a recent review in that field [79].

*Innate lymphoid cells*

ILCs are a recently identified group of heterogeneous innate immune cells belonging to the lymphoid lineage but lacking antigen-specific receptors [81]. Mechanistically, ILCs are involved in protective anti-microbial responses, particularly at mucosal barrier organs, but have also pathogenic roles in inflammation, autoimmunity, allergy and fibrosis within tissues [81,82]. ILCs are distinguished from canonical T and B cells by their ontogeny / development and the expression of a distinct set of cellular markers [81]. Of note, ILCs do not express RAG (recombination-activating gene) genes, implying that ILCs can be, in contrast to T and B cells, activated directly [83]. ILCs regularly express IL-2Rα (CD25), IL-7Rα (CD127) and IL-7Rγ (CD132) [84]. Depending on their ability to synthesize and release cytokines and their transcription factor profile, ILCs are divided into three distinct subsets. ILC1s, which include NK cells, are defined, in analogy to Th1 cells, by their ability to release interferon-γ (IFN-γ) and tumor necrosis factor (TNF) and the expression of the T-box transcription factor T-bet or eomesodermin (Eomes) [85]. Functionally, NK cells require only Eomes and IL-5, but not IL-7 or T-bet. Across tissues, ILC1 cells are found mostly in liver, thymus, uterus, skin, lung, secondary lymphoid tissue, spleen and gut. ILC1s have been demonstrated to accumulate in response to viral and bacterial infections in mice [86]. This has probable relevance for chronic human disease conditions, as circulating ILCs were increased in COPD patients upon exacerbations [87]. Type 2 ILC (ILC2s), the counterpart of Th2 cells, produce IL-4, IL-5, IL-9 and IL-13 in response to IL-25, IL-33 and TSLP (Thymic stromal lymphopoietin) and express high levels of the Th2-signature transcription factor GATA-3, which is necessary for their functional maturation and maintenance [88,89]. ILC2s are localized mostly in the healthy skin, lung, and adipose tissue of mice and humans and have been mainly involved in tissue inflammation, remodeling and fibrosis. Interestingly, ILC2s are the main cell type among ILCs found in the murine lung, however, representing overall only 0.4-1% of total live lung cells. A recent study demonstrated, using single-cell RNA sequencing of lung-resident ILCs, that in the context of allergic lung inflammation the alarmin cytokines IL-25 and IL-33 were linked to the activation and expansion of lung-resident ILC2s [88]. *In vivo* activation by IL-25 induced high expression of the Neuromedin U (NMU) receptor (NMUR1) in ILC2s. Interestingly, the combined treatment with IL-25 and NMU synergistically promoted allergic inflammation. In line with previous studies showing increased numbers of ILC2s in the blood of asthma patients [90], this study suggests that ILC2s could represent a future and potentially predictive biomarker for patients with asthma. Type 3 ILCs contain natural cytotoxicity receptor positive (NCR+) ILC3 and NCR- LTi cells, which depend on the transcription factor RORγt and produce either IL-22 (NCR+ ILC3 or ILC22 cells) or both IL-17 and IL-22 (NCR- LTi or ILC17 cells) in response to IL-23 [82,91]. LTi cells were reported to contribute to the development of secondary lymphoid tissue, where they activate stromal cells through the lymphotoxin-α1β2-mediated LTβR signaling pathway. This triggers the expression of adhesion molecules (e.g. ICAM, VCAM) and chemokines (CXCL13, CCL19 and CCL21) required for the formation of lymphoid follicles [92]. We have shown previously that B-cell-dependent inducible bronchus associated lymphoid tissue (iBALT) formation is essential for emphysema development in the cigarette smoke-induced COPD mouse model [93]. Therefore, when viewed in combination, it is tempting to speculate that LTi cells may play a key role in the development of COPD immune pathogenesis. Although the mechanistic and functional contribution of ILCs in animal models of asthma and COPD has been interrogated, there is limited evidence for a potential role in human disease conditions, requiring further studies in that field. With regards to lung fibrosis, the potential role of ILCs is discussed in a comprehensive review [94]. In brief, ILC2s and ILC3s have been involved in the pathogenesis of lung fibrosis. IL-17A plays a key role in pulmonary inflammation and fibrosis and has been implicated in the pathomechanisms underlying and driving asthma, COPD, IPF and CF [95-97]. Given ILC3s as essential source of IL-17 at mucosal sites, these innate tissue cells are suggested as early orchestrators of lung tissue remodeling and fibrogenesis. The potential role of ILCs in CF lung disease is elusive, yet Moretti and coworkers showed recently that a mast cell - ILC2 - Th9 pathway promotes pulmonary inflammation in murine infection models of CF-like lung disease [98].

*Mucosal-Associated Invariant T cells*

Mucosal Associated Invariant T cells (MAITs) are a group of innate-like T lymphocytes, that are highly aboundant in human blood (~1-10%; [99]) and in mucosal tissues including intestines, lungs and livers [100-103]. These cells are characterized by their expression of the invariant T cell recpeor (TCR) α chain, TRAV1-2 joined with TRAJ33 and a limited range of TCRß chains and abundant expression of CD161 and CD218 (IL18Rα) [101,104,105]. Recently, studies have shown that MAIT cells can recognize Vitamin B metabolic byproducts produced by a range of micro organisms presented by the ubiquitously expressed, MHC1 related protein, MR1 [106-109]. Activated MAIT cells have been observed to generate high levels of IFN-γ, TNF-α, IL-17 and cytotoxic/cytolytic perforin and Granzymes A, B and K[110-115].

MAIT cells frequencies in normal human lungs are variable, ranging from 2-20% of all T cells in the lungs [103,116]. Given their abundance in the blood, many studies have studied peripheral blood MAIT cells in various respiratory infections and maladies. There were no changes in MAIT cell frequency in the blood of inhaled corticosteroid (ICS) naïve COPD relative to healthy donors, however, their frequency was significantly reduced in ICS treated COPD patient’s blood and bronchial biopsies [103]. Reduced MAIT cell frequency in the blood of was more pronounced in moderate to severe COPD patients and was associated with elevated serum C-reactive protein levels and reduced lung function (as assessed via FEV1/FVC ratio) [117]. Consistent with a role of ICS in modulating MAIT cell numbers, these cells were also observed to be significantly reduced in ICS treated asmatic patient’s blood and lung tissue relative to normal individuals [118,119]. This was especially evident in severe asthmatic patients [119]. These studies suggest that MAIT cells are sucipitable to ICS treatment and that their deficiency is associated with severe respiratory pathology.

Experimental evidence suggested that MAIT cells can be activated with epithelial cells-infected with multiple bacterial strains [110]. Indeed, an important role of these cells in anti-bacterial immunity is supported by studies showing a significant inverse correlation between peripheral blood MAIT cell numbers and CF disease severity and inverse association with *Pseudomonas aeruginosa* infection and disease exacerbation [120]. Consistently, circulating MAIT cells were also observed to be decreased in the peripheral blood of patients with tuberculosis (TB) and nontuberculous mycobacterial infection [121,122] and their deficiency was correlated with disease severity [121]. *In vitro* stimulation of freshly isolated MAIT cells using PMA, ionomycin, IL-15, anti-CD28 antibodies and/or mycobacterial lysates suggested a severe functional deficiency in TB patient relative to healthy donor derived MAIT cells, as assessed by the induction of IFN-γ and IL-17 [121,123]. Various studies have shown an important, non redudntant, role for IL-12, IL-18, and IL-2 signaling in MAIT cell activation [113,115,124-126], and a significant reduction of, IL-12 and/or IL-2 receptors in *in vitro* stimulated peripheral blood MAIT cells from TB patients [123,126]. Finally, two reports have suggested that functionally deficient MAIT cells from the peripheral blood of TB patients and HIV+TB patients have elevated levels of cell surface PD-1 expression relative to those from healthy donors [127,128], suggesting that immune checkpoint pathways may functionally regulate these cells during infection. Collectively, these studies suggest that MAIT cells are functionally important in controlling bacterial infections, and defeciencies in these cells is observed in patients with active bacterial infecitons.

Identification of mechanisms propagated by MAIT cells in pulmonary immunity were hindered hindered due to the low abundance of these cells in germ free laboratory mice; however, with the recent development of iVα19 TCRα [129] transgenic and B6-MAITCAST [130] and the commercial availability of murine MR1 tetramers, many studies have been performed characterizing the role of these cells in models of pulmonary inflammation and infection. Utilizing iVα19-transgenic MR1 sufficient or iVα19-trangenic MR1 deficient mice to study the role of MAIT cells in anti-bacterial immunity, Le Bourhis et al. [122] observed that MR1 sufficient mice, with more activated MAIT cells, had a lower bacterial burden relative to MR1 deficient mice. In a model of pulmonary bacterial infection, one study have shown evidence for the importance of MAIT cells in protecting against *Francisella tularenis* (LVS)pulmonary infection [131]. Utilizing C57BL6 *MR1+* or *MR1-* mice, this group observed an MR1 dependent recrutement and/or expansion of MAIT cells in LVS infected mice, and a lower bacterial burden in the lungs of the MR1-sufficient relative to MR1-deficient murine lungs. *In vitro* coculture studies utilizing MAIT cells and LVS infected macrophages indicated that MAIT cell derived IL-12p40, TNF and IFN-γ were indespensible in controlling intracellular bacterial growth in macrophages. Finally, this study have observed delayed recruitment of effector CD4+ helper and CD8+ cytotixic T cells in MR1-deficient mice relative to their MR1 sufficent counterparts. In a subsequent report, this group have identified MAIT cell derived GM-CSF to be required for the differentiation of CCR2+ monocytes into dendritic cells and the subsequent recruitment of effector helper and cytotixic T cells for efficient bacterial clearance [132]. Collectively, these studies suggest that MAIT cells elaborate a protective effect against bacterial infection via multiple mechanisms, including macrophage activation and monocyte to dendritic cell differentiation and subsequent briding of the innate and adaptive immune mechanisms.

Given the identification of bacterial and fungal Vitamin B metabolites as MR1 presented antigens [106-109], MAIT cells were thought not to play a role in viral infection. However, a recent study have observed higher MAIT cell numbers in H7N9 influenza patients who recover from infection, relative to those who succumbed to the infection [125]. Utilizing an *in vitro* co-culture system of influenza infected airway epithelial cells line (A549) with human peripheral blood mononuclear cells, this group have observed an MR1 independent, CD14+ cell and IL18 dependent activation of MAIT cells as assessed by intracellular elevation of IFN-γ and granzyme B proteins [125]. Indeed, this was confirmed in another study, where MAIT cells were observed to be activated in an MR1 independent and IL-18, IL-12 and IL-15 dependent manner in response to various viral infections, including dengue, HCV and influenza viruses [133]. These studies suggest that MAIT cells may play an important role in viral infections.

Collectively, these studies suggest that MAIT cells are indespensible in the immune protection of the respiratory system against viral and bacterial infections. There is mounting evidence for changes in bacterial composition in exacerbated COPD, where there was more abundant microbial spiecies commonly observed in exacerbated COPD [134,135]. Further, severe asthmatic patients and asthmatic exacerbations are known to occur after respiratory viral infections (reviewed in [136,137]). Given the potential role of MAIT cells in controlling bacterial and viral infections, their deficiency in severe ICS resistant COPD and asthma may reflect enhanced sucipitability of these patients to pulmonary infections and may contribute to the severity of the disease.

*Novel mediators/Chitinase-like proteins*

There are many novel mediators that participate in lung innate immunity. These mediators include chitinase and chitinase like proteins (CLPs) that are conserved group of proteins that belong to the 18-glycosyl hydrolase family [138]. Although mammals do not synthesize chitins, the presence of chitinases and CLPs suggest their role in digestions of chitin-containing food or protection against chitin containing pathogens. This hypothesis was based on the initial epidemiologic evidences showing increased expression of these proteins in populations that are exposed to more chitin-containing pathogens and food products [139,140]. However, later studies casted doubts on this hypothesis indicating limited correlation between Chit1 deficiency and exposure to chitin containing parasites as well food [141-143].

CLPs have high binding capacity to chitin but lacks enzymatic activity to cleave chitin suggesting a limited direct role against chitin-containing pathogens or in digestion of chitin-containing food. Due to the conserved presence without any obvious roles, recent advances have shown that CLPs as well as chitinases play important roles in immune-related pathophysiology. Elevated levels of chitinase activity has been observed in lysosomal storage disease such as Gaucher’s disease or lung diseases where inflammatory response plays a major role in the pathogenesis such as chronic obstructive pulmonary disease (COPD), asthma, sarcoidosis, cystic fibrosis, etc [144-147]. Acidic mammalian chitinase (Chit2), one of two true chitinase present in mammals, have been shown to mediate type 2 inflammation and pathology in mouse model of asthama [147]. Similar role for chitotriosidase (Chit1) was observed during fungal lung infection, where cleavage of fungal chitin by chitotriosidase meadiates pathological responses [148]. In both the asthma models and fungal infection models, better outcomes and survival were observed in mice lacking AMCase and Chit1 [147,148]. To put in perspective, a substantial human population (3-20%) lacks true chitinase activity due to a 24-bp mutation in Chit1 gene (the major contributor of chitinase activity in humans), suggesting their non-essential but potential harmful effects during pathological challenges [149-151].

On the other hand, CLPs are well conserved but have high divergence in mammals (YKL-39 & YKL-40 in humans and BRP-39, Ym1 & Ym2 in mice), suggesting their important protective roles to the host [138]. The divergence in CLPs among mammals have been attributed to the difference in microbial threat faced by each individual species [138]. Chil1 (BRP-39 in mice and YKL 40 in humans) is one of the most prevalent CLP present in humans. Chil1 is highly expressed in immune cells including macrophages and neutrophils suggesting a immune-specific roles. Indeed; studies using bacterial lung infections indicated their important roles in inflammasome regulation *in-vivo* and *in-vitro* [152,153]. Absence of BRP-39 in mice during lung infection with Gram positive *Streptococcus* or Gram negative *Pseudomonas* leads to exaggerated inflammasome activation. BRP-39 limits macrophage pyroptosis during bacterial infection to give advantage to the host by limiting bacterial growth and exuberant inflammation leading to lung injury that improes survival [152,153]. BRP-39 has been shown to bind IL-13 receptor α involving the protein TMEM to exert its effect [154,155].

The other members of CLPs in mice include Ym1 and Ym2 [138]. These two proteins are hightly expressed in the lung and their expression is increased during the induction of type 2 inflammation such as nematode infection and house dust mite (HDM) allergen model of asthma [156,157]. Ym1 overexpression leads to increase in lung neutrophilia and blocking Ym1 using monoclonal antibody resulted in decreased neutrophil accumulation in lung during HDM model in mice. γδ T cells are the major target of CLPs where overexpression of CLPs result in increase production of IL-17A [156]. These experiments suggest important roles played by Ym1 in mediating inflammation in the lung. Overall, chitinase and CLPs are associated with many inflammatory diseases and their causal role in various diseases has been established using mouse models. These proteins might provide novel diagnostic and therapeutic targets to understand, treat and moniter therapeutic efficacies in many inflammatory and infectious diseases.

*Summary and disease implications*

The multidimensional nature of pulmonary innate immunity can be subdivided into three categories: (i) cellular vs non-cellular components, (ii) protective vs harmful mechanisms and (iii) translational disease relevance for biomarker and therapeutic targeting approaches.

To summarize on (i): innate immune responses at mucosal sites in general comprise two arms: cellular [158] and non-cellular/humoral components. Particular to the pulmonary compartment is the mucociliary escalator as a physical innate host defense barrier in conjunction with airway epithelial cell-derived innate effector proteins, prototypically anti-microbial proteins, such as defensins and collectins, that act anti-microbial. Among innate immune cells, alveolar macrophages represent a potent phagocytic immune cell sentinel in the lower airway compartment equipped with a broad cellular armamentarium to protect the airspace from microbial and non-microbial (dust, cigarette smoke) airborne exposures and serving as a rheostat to maintain alveolar heomeostasis. While alveolar macrophages dominate in the lower/alveolar space, neutrophils accumulate in the more proximal/bronchial airway compartments. Lymphocytes, in turn, are mainly found in lung tissue/parenchyma [10], while they are scarce in the bronchoalveolar space, which might be due to suppressive neutrophil-T cell interactions [49]. Novel innate immune cell types, such as MDSCs, MAIT cells and ILCs add to the emerging complexity of several layers of innate host defense shields in the pulmonary mucosal environment. While our understainding on their regulatory and pathophysiological role in murine model systems is increasing, their relevance for human pathologies remains poorly defined.

To summarize on (ii): protective vs harmful activities of innate immune cells and proteins mainly depend on the spatiotemporal context: intracellular enzymes, such as elastase or MMPs, act protective by degrading phagocytosed microbial proteins, while the same enzymes liberated into the extracellular microenvironment can cause severe tissue injury by degrading extracellular matrices, such as elastin, and thereby remodeling the fragile pulmonary architecure. Temporally, innate immune effector responses are mostly protective in the titial phases of infection and inflammation, but become harmful and auto-inflammatory if they fail to resolve and perpetuate, exemplified in chronic lung diseases [42], such as CF, IPF and COPD. From a cellular perspective, pulmonary innate immune cells can be further subdivided into two simplified categories: granulocytic cells (neutrophils, eosinophils and basophils) that are short-lived and bear a higher acute pathogenic potential by rapidly releasing their toxic and tissue-damaging ingrendients (proteases, oxidants) upon local airway activation [8,46], and mononuclear cells (monocytes, alveolar/interstitial macrophages and dendritic cells) that live longer, are more robust and controlled in terms of enzyme release and mainly serve as APCs and protective scavengers of apoptotic bodies/debris and invading microbes. Short-lived innate immune cells play a major role in the early phase of acute respiratory conditions, such as neutrophils in lung infections or ARDS, whereas long-lived innate immune cells, such as macrophages and ILCs, probably orchestrate the chronic outcome of tissue inflammation and remodeling in chronic pulmonary conditions like lung fibrosis/IPF and COPD. Notably, different macrophage subtypes, M1 vs M2, have been proposed to differentially affect pulmonary disease outcome in asthma, COPD, asthma and CF.

To summarize on (iii): therapeutic approaches targeting innate immune cells and particularly innate immune cell-derived proteases are reasonable given the surplus of these enzymes in the pulmonary microenvironment (particularly in CF and COPD), yet clinical studies were so far either limited by safety (for small molecule approaches targeting MMPs) or by efficacy (for supplementation of anti-proteases, prototypically alpha-1 antitrypsin) [159,160]. Therapeutic strategies targeting rarer innate immune cell subpopulations, such as ILCs or MAIT cells, are hampered by (i) the early/poor understanding of their protective vs harmful potential in human disease conditions and (ii) the lack of knowledge how to target these cell types specifically. Recently, novel innate immune mediators have been emerged as potential biomarkers and/or drug targets for respiratory diseases. Of note, alarmins/DAMPs (such as S100 proteins, ATP or HMGB1) seem to play a role in lung immunity and bind to pattern recognition receptors like TLR4 and RAGE. Targeting these innate inflammation-amplifying loops while leaving bacterial sensing intact will pose a challenge to future drug development approaches in that space. Collectively, innate immunity of the lung is multi-faceted and multi-layered given the anatomical complexity (upper versus lower airways, bronchoalveolar space versus lung parenchyma), the mucosal barrier interactions with microbial colonizers and the spatiotemporal dynamics of infiltrating (e.g. neutrophils) and resident (e.g. alveolar macrophages or ILC subtypes) immune cells. The key challenge for biomarker and therapeutic success will be to define beneficial vs harmful innate immune cell subsets across species and to identify ways to selectively target respective cell types or their released mediators.

**ACKNOWLEDGEMENTS**

This work was supported by funds of the DFG CRC/SFB685 to D.H. (University of Tübingen, Germany).

**REFERENCES**

1 Hiemstra PS, McCray PB, Jr., Bals R: The innate immune function of airway epithelial cells in inflammatory lung disease. The European respiratory journal 2015;45:1150-1162.

2 Hasenberg M, Stegemann-Koniszewski S, Gunzer M: Cellular immune reactions in the lung. Immunol Rev 2013;251:189-214.

3 Chaudhuri N, Sabroe I: Basic science of the innate immune system and the lung. Paediatr Respir Rev 2008;9:236-242.

4 Sabroe I, Lloyd CM, Whyte MK, Dower SK, Williams TJ, Pease JE: Chemokines, innate and adaptive immunity, and respiratory disease. EurRespirJ 2002;19:350-355.

5 Panina-Bordignon P, D'Ambrosio D: Chemokines and their receptors in asthma and chronic obstructive pulmonary disease. CurrOpinPulm Med 2003;9:104-110.

6 Owen C: Chemokine receptors in airway disease: Which receptors to target? Pulm PharmacolTher 2001;14:193-202.

7 Gerard C, Rollins BJ: Chemokines and disease. NatImmunol 2001;2:108-115.

8 Kruger P, Saffarzadeh M, Weber AN, Rieber N, Radsak M, von Bernuth H, Benarafa C, Roos D, Skokowa J, Hartl D: Neutrophils: Between host defence, immune modulation, and tissue injury. PLoS Pathog 2015;11:e1004651.

9 Tang AC, Turvey SE, Alves MP, Regamey N, Tummler B, Hartl D: Current concepts: Host-pathogen interactions in cystic fibrosis airways disease. Eur Respir Rev 2014;23:320-332.

10 Regamey N, Tsartsali L, Hilliard TN, Fuchs O, Tan HL, Zhu J, Qiu YS, Alton EW, Jeffery PK, Bush A, Davies JC: Distinct patterns of inflammation in the airway lumen and bronchial mucosa of children with cystic fibrosis. Thorax 2011

11 Pabst R: Is balt a major component of the human lung immune system? Immunol Today 1992;13:119-122.

12 Tengroth L, Millrud CR, Kvarnhammar AM, Kumlien Georen S, Latif L, Cardell LO: Functional effects of toll-like receptor (tlr)3, 7, 9, rig-i and mda-5 stimulation in nasal epithelial cells. PLoS One 2014;9:e98239.

13 Kim HJ, Kim CH, Kim MJ, Ryu JH, Seong SY, Kim S, Lim SJ, Holtzman MJ, Yoon JH: The induction of pattern-recognition receptor expression against influenza a virus through duox2-derived reactive oxygen species in nasal mucosa. Am J Respir Cell Mol Biol 2015;53:525-535.

14 Kim JH, Kim YS, Cho GS, Kim NH, Gong CH, Lee BJ, Jang YJ: Human rhinovirus-induced proinflammatory cytokine and interferon-beta responses in nasal epithelial cells from chronic rhinosinusitis patients. Allergy Asthma Immunol Res 2015;7:489-496.

15 Nathan AT, Peterson EA, Chakir J, Wills-Karp M: Innate immune responses of airway epithelium to house dust mite are mediated through beta-glucan-dependent pathways. J Allergy Clin Immunol 2009;123:612-618.

16 Page K, Ledford JR, Zhou P, Dienger K, Wills-Karp M: Mucosal sensitization to german cockroach involves protease-activated receptor-2. Respir Res 2010;11:62.

17 Post S, Heijink IH, Petersen AH, de Bruin HG, van Oosterhout AJ, Nawijn MC: Protease-activated receptor-2 activation contributes to house dust mite-induced ige responses in mice. PLoS One 2014;9:e91206.

18 Kool M, Willart MA, van Nimwegen M, Bergen I, Pouliot P, Virchow JC, Rogers N, Osorio F, Reis e Sousa C, Hammad H, Lambrecht BN: An unexpected role for uric acid as an inducer of t helper 2 cell immunity to inhaled antigens and inflammatory mediator of allergic asthma. Immunity 2011;34:527-540.

19 Lambrecht BN, Hammad H: The airway epithelium in asthma. Nat Med 2012;18:684-692.

20 Bogefors J, Rydberg C, Uddman R, Fransson M, Mansson A, Benson M, Adner M, Cardell LO: Nod1, nod2 and nalp3 receptors, new potential targets in treatment of allergic rhinitis? Allergy 2010;65:1222-1226.

21 Tabeling C, Scheer H, Schonrock SM, Runge F, Gutbier B, Lienau J, Hamelmann E, Opitz B, Suttorp N, Mayer K, Behrens GM, Tschernig T, Witzenrath M: Nucleotide oligomerization domain 1 ligation suppressed murine allergen-specific t-cell proliferation and airway hyperresponsiveness. Am J Respir Cell Mol Biol 2014;50:903-911.

22 Hirota JA, Gold MJ, Hiebert PR, Parkinson LG, Wee T, Smith D, Hansbro PM, Carlsten C, VanEeden S, Sin DD, McNagny KM, Knight DA: The nucleotide-binding domain, leucine-rich repeat protein 3 inflammasome/il-1 receptor i axis mediates innate, but not adaptive, immune responses after exposure to particulate matter under 10 mum. Am J Respir Cell Mol Biol 2015;52:96-105.

23 Unger BL, Ganesan S, Comstock AT, Faris AN, Hershenson MB, Sajjan US: Nod-like receptor x-1 is required for rhinovirus-induced barrier dysfunction in airway epithelial cells. J Virol 2014;88:3705-3718.

24 Lee RJ, Cohen NA: Bitter and sweet taste receptors in the respiratory epithelium in health and disease. J Mol Med (Berl) 2014;92:1235-1244.

25 Barham HP, Cooper SE, Anderson CB, Tizzano M, Kingdom TT, Finger TE, Kinnamon SC, Ramakrishnan VR: Solitary chemosensory cells and bitter taste receptor signaling in human sinonasal mucosa. Int Forum Allergy Rhinol 2013;3:450-457.

26 Shah AS, Ben-Shahar Y, Moninger TO, Kline JN, Welsh MJ: Motile cilia of human airway epithelia are chemosensory. Science 2009;325:1131-1134.

27 Lee RJ, Cohen NA: The emerging role of the bitter taste receptor t2r38 in upper respiratory infection and chronic rhinosinusitis. Am J Rhinol Allergy 2013;27:283-286.

28 Lee RJ, Kofonow JM, Rosen PL, Siebert AP, Chen B, Doghramji L, Xiong G, Adappa ND, Palmer JN, Kennedy DW, Kreindler JL, Margolskee RF, Cohen NA: Bitter and sweet taste receptors regulate human upper respiratory innate immunity. J Clin Invest 2014;124:1393-1405.

29 Mirkovic B, Murray MA, Lavelle GM, Molloy K, Azim AA, Gunaratnam C, Healy F, Slattery D, McNally P, Hatch J, Wolfgang M, Tunney MM, Muhlebach MS, Devery R, Greene CM, McElvaney NG: The role of short-chain fatty acids, produced by anaerobic bacteria, in the cystic fibrosis airway. American journal of respiratory and critical care medicine 2015;192:1314-1324.

30 Chirkova T, Lin S, Oomens AG, Gaston KA, Boyoglu-Barnum S, Meng J, Stobart CC, Cotton CU, Hartert TV, Moore ML, Ziady AG, Anderson LJ: Cx3cr1 is an important surface molecule for respiratory syncytial virus infection in human airway epithelial cells. J Gen Virol 2015;96:2543-2556.

31 Kotha PL, Sharma P, Kolawole AO, Yan R, Alghamri MS, Brockman TL, Gomez-Cambronero J, Excoffon KJ: Adenovirus entry from the apical surface of polarized epithelia is facilitated by the host innate immune response. PLoS Pathog 2015;11:e1004696.

32 Pillay J, den Braber I, Vrisekoop N, Kwast LM, de Boer RJ, Borghans JA, Tesselaar K, Koenderman L: In vivo labeling with 2h2o reveals a human neutrophil lifespan of 5.4 days. Blood 2010;116:625-627.

33 Mauer AM, Athens JW, Ashenbrucker H, Cartwright GE, Wintrobe MM: Leukokinetic studies. Ii. A method for labeling granulocytes in vitro with radioactive diisopropylfluorophosphate (dfp). J Clin Invest 1960;39:1481-1486.

34 Zhang P, Summer WR, Bagby GJ, Nelson S: Innate immunity and pulmonary host defense. Immunol Rev 2000;173:39-51.

35 Summers C, Chilvers ER, Peters AM: Mathematical modeling supports the presence of neutrophil depriming in vivo. Physiol Rep 2014;2:e00241.

36 Summers C, Singh NR, White JF, Mackenzie IM, Johnston A, Solanki C, Balan KK, Peters AM, Chilvers ER: Pulmonary retention of primed neutrophils: A novel protective host response, which is impaired in the acute respiratory distress syndrome. Thorax 2014;69:623-629.

37 Scapini P, Marini O, Tecchio C, Cassatella MA: Human neutrophils in the saga of cellular heterogeneity: Insights and open questions. Immunol Rev 2016;273:48-60.

38 Williams AE, Chambers RC: The mercurial nature of neutrophils: Still an enigma in ards? Am J Physiol Lung Cell Mol Physiol 2014;306:L217-230.

39 Fortunati E, Kazemier KM, Grutters JC, Koenderman L, Van den Bosch v J: Human neutrophils switch to an activated phenotype after homing to the lung irrespective of inflammatory disease. Clin Exp Immunol 2009;155:559-566.

40 Hartl D, Krauss-Etschmann S, Koller B, Hordijk PL, Kuijpers TW, Hoffmann F, Hector A, Eber E, Marcos V, Bittmann I, Eickelberg O, Griese M, Roos D: Infiltrated neutrophils acquire novel chemokine receptor expression and chemokine responsiveness in chronic inflammatory lung diseases. J Immunol 2008;181:8053-8067.

41 Koller B, Kappler M, Latzin P, Gaggar A, Schreiner M, Takyar S, Kormann M, Kabesch M, Roos D, Griese M, Hartl D: Tlr expression on neutrophils at the pulmonary site of infection: Tlr1/tlr2-mediated up-regulation of tlr5 expression in cystic fibrosis lung disease. J Immunol 2008;181:2753-2763.

42 Koller B, Bals R, Roos D, Korting HC, Griese M, Hartl D: Innate immune receptors on neutrophils and their role in chronic lung disease. Eur J Clin Invest 2009;39:535-547.

43 Tirouvanziam R, Gernez Y, Conrad CK, Moss RB, Schrijver I, Dunn CE, Davies ZA, Herzenberg LA: Profound functional and signaling changes in viable inflammatory neutrophils homing to cystic fibrosis airways. Proc Natl Acad Sci U S A 2008;105:4335-4339.

44 Makam M, Diaz D, Laval J, Gernez Y, Conrad CK, Dunn CE, Davies ZA, Moss RB, Herzenberg LA, Herzenberg LA, Tirouvanziam R: Activation of critical, host-induced, metabolic and stress pathways marks neutrophil entry into cystic fibrosis lungs. Proc Natl Acad Sci U S A 2009;106:5779-5783.

45 Laval J, Touhami J, Herzenberg LA, Conrad C, Taylor N, Battini JL, Sitbon M, Tirouvanziam R: Metabolic adaptation of neutrophils in cystic fibrosis airways involves distinct shifts in nutrient transporter expression. J Immunol 2013;190:6043-6050.

46 Ralhan A, Laval J, Lelis F, Ballbach M, Grund C, Hector A, Hartl D: Current concepts and controversies in innate immunity of cystic fibrosis lung disease. J Innate Immun 2016;8:531-540.

47 Laval J, Ralhan A, Hartl D: Neutrophils in cystic fibrosis. Biol Chem 2016;397:485-496.

48 Hartl D, Latzin P, Hordijk P, Marcos V, Rudolph C, Woischnik M, Krauss-Etschmann S, Koller B, Reinhardt D, Roscher AA, Roos D, Griese M: Cleavage of cxcr1 on neutrophils disables bacterial killing in cystic fibrosis lung disease. Nat Med 2007;13:1423-1430.

49 Ingersoll SA, Laval J, Forrest OA, Preininger M, Brown MR, Arafat D, Gibson G, Tangpricha V, Tirouvanziam R: Mature cystic fibrosis airway neutrophils suppress t cell function: Evidence for a role of arginase 1 but not programmed death-ligand 1. J Immunol 2015;194:5520-5528.

50 Hampton HR, Bailey J, Tomura M, Brink R, Chtanova T: Microbe-dependent lymphatic migration of neutrophils modulates lymphocyte proliferation in lymph nodes. Nat Commun 2015;6:7139.

51 Duffy D, Perrin H, Abadie V, Benhabiles N, Boissonnas A, Liard C, Descours B, Reboulleau D, Bonduelle O, Verrier B, Van Rooijen N, Combadiere C, Combadiere B: Neutrophils transport antigen from the dermis to the bone marrow, initiating a source of memory cd8+ t cells. Immunity 2012;37:917-929.

52 Abadie V, Badell E, Douillard P, Ensergueix D, Leenen PJ, Tanguy M, Fiette L, Saeland S, Gicquel B, Winter N: Neutrophils rapidly migrate via lymphatics after mycobacterium bovis bcg intradermal vaccination and shuttle live bacilli to the draining lymph nodes. Blood 2005;106:1843-1850.

53 Coombes JL, Charsar BA, Han SJ, Halkias J, Chan SW, Koshy AA, Striepen B, Robey EA: Motile invaded neutrophils in the small intestine of toxoplasma gondii-infected mice reveal a potential mechanism for parasite spread. Proc Natl Acad Sci U S A 2013;110:E1913-1922.

54 Brinkmann V, Zychlinsky A: Beneficial suicide: Why neutrophils die to make nets. Nature reviews Microbiology 2007;5:577-582.

55 Brinkmann V, Reichard U, Goosmann C, Fauler B, Uhlemann Y, Weiss DS, Weinrauch Y, Zychlinsky A: Neutrophil extracellular traps kill bacteria. Science 2004;303:1532-1535.

56 Cortjens B, van Woensel JB, Bem RA: Neutrophil extracellular traps in respiratory disease: Guided anti-microbial traps or toxic webs? Paediatr Respir Rev 2017;21:54-61.

57 Cheng OZ, Palaniyar N: Net balancing: A problem in inflammatory lung diseases. Front Immunol 2013;4:1.

58 Yipp BG, Kubes P: Netosis: How vital is it? Blood 2013;122:2784-2794.

59 Sorensen OE, Borregaard N: Neutrophil extracellular traps - the dark side of neutrophils. J Clin Invest 2016;126:1612-1620.

60 Grabcanovic-Musija F, Obermayer A, Stoiber W, Krautgartner WD, Steinbacher P, Winterberg N, Bathke AC, Klappacher M, Studnicka M: Neutrophil extracellular trap (net) formation characterises stable and exacerbated copd and correlates with airflow limitation. Respir Res 2015;16:59.

61 Wright TK, Gibson PG, Simpson JL, McDonald VM, Wood LG, Baines KJ: Neutrophil extracellular traps are associated with inflammation in chronic airway disease. Respirology 2016;21:467-475.

62 Cortjens B, de Boer OJ, de Jong R, Antonis AF, Sabogal Pineros YS, Lutter R, van Woensel JB, Bem RA: Neutrophil extracellular traps cause airway obstruction during respiratory syncytial virus disease. J Pathol 2016;238:401-411.

63 Gallin JI: Human neutrophil heterogeneity exists, but is it meaningful? Blood 1984;63:977-983.

64 Garbi N, Lambrecht BN: Location, function, and ontogeny of pulmonary macrophages during the steady state. Pflugers Arch 2017;469:561-572.

65 Hussell T, Bell TJ: Alveolar macrophages: Plasticity in a tissue-specific context. Nat Rev Immunol 2014;14:81-93.

66 Fels AO, Cohn ZA: The alveolar macrophage. J Appl Physiol (1985) 1986;60:353-369.

67 du Bois RM: The alveolar macrophage. Thorax 1985;40:321-327.

68 Trapnell BC, Carey BC, Uchida K, Suzuki T: Pulmonary alveolar proteinosis, a primary immunodeficiency of impaired gm-csf stimulation of macrophages. Curr Opin Immunol 2009;21:514-521.

69 Trapnell BC, Whitsett JA, Nakata K: Pulmonary alveolar proteinosis. The New England journal of medicine 2003;349:2527-2539.

70 Soroosh P, Doherty TA, Duan W, Mehta AK, Choi H, Adams YF, Mikulski Z, Khorram N, Rosenthal P, Broide DH, Croft M: Lung-resident tissue macrophages generate foxp3+ regulatory t cells and promote airway tolerance. J Exp Med 2013;210:775-788.

71 Westphalen K, Gusarova GA, Islam MN, Subramanian M, Cohen TS, Prince AS, Bhattacharya J: Sessile alveolar macrophages communicate with alveolar epithelium to modulate immunity. Nature 2014;506:503-506.

72 van de Laar L, Saelens W, De Prijck S, Martens L, Scott CL, Van Isterdael G, Hoffmann E, Beyaert R, Saeys Y, Lambrecht BN, Guilliams M: Yolk sac macrophages, fetal liver, and adult monocytes can colonize an empty niche and develop into functional tissue-resident macrophages. Immunity 2016;44:755-768.

73 Guilliams M, De Kleer I, Henri S, Post S, Vanhoutte L, De Prijck S, Deswarte K, Malissen B, Hammad H, Lambrecht BN: 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.

74 Gomez Perdiguero E, Klapproth K, Schulz C, Busch K, Azzoni E, Crozet L, Garner H, Trouillet C, de Bruijn MF, Geissmann F, Rodewald HR: Tissue-resident macrophages originate from yolk-sac-derived erythro-myeloid progenitors. Nature 2015;518:547-551.

75 Guth AM, Janssen WJ, Bosio CM, Crouch EC, Henson PM, Dow SW: Lung environment determines unique phenotype of alveolar macrophages. Am J Physiol Lung Cell Mol Physiol 2009;296:L936-946.

76 Sica A, Mantovani A: Macrophage plasticity and polarization: In vivo veritas. J Clin Invest 2012;122:787-795.

77 Mantovani A, Biswas SK, Galdiero MR, Sica A, Locati M: Macrophage plasticity and polarization in tissue repair and remodelling. J Pathol 2013;229:176-185.

78 Murray PJ, Allen JE, Biswas SK, Fisher EA, Gilroy DW, Goerdt S, Gordon S, Hamilton JA, Ivashkiv LB, Lawrence T, Locati M, Mantovani A, Martinez FO, Mege JL, Mosser DM, Natoli G, Saeij JP, Schultze JL, Shirey KA, Sica A, Suttles J, Udalova I, van Ginderachter JA, Vogel SN, Wynn TA: Macrophage activation and polarization: Nomenclature and experimental guidelines. Immunity 2014;41:14-20.

79 Kolahian S, Fernandez IE, Eickelberg O, Hartl D: Immune mechanisms in pulmonary fibrosis. Am J Respir Cell Mol Biol 2016;55:309-322.

80 Bruscia EM, Bonfield TL: Cystic fibrosis lung immunity: The role of the macrophage. J Innate Immun 2016

81 Spits H, Cupedo T: Innate lymphoid cells: Emerging insights in development, lineage relationships, and function. Annu Rev Immunol 2012;30:647-675.

82 Sonnenberg GF, Artis D: Innate lymphoid cells in the initiation, regulation and resolution of inflammation. Nat Med 2015;21:698-708.

83 Spits H, Artis D, Colonna M, Diefenbach A, Di Santo JP, Eberl G, Koyasu S, Locksley RM, McKenzie AN, Mebius RE, Powrie F, Vivier E: Innate lymphoid cells--a proposal for uniform nomenclature. Nat Rev Immunol 2013;13:145-149.

84 Guo L, Junttila IS, Paul WE: Cytokine-induced cytokine production by conventional and innate lymphoid cells. Trends Immunol 2012;33:598-606.

85 Klose CSN, Flach M, Mohle L, Rogell L, Hoyler T, Ebert K, Fabiunke C, Pfeifer D, Sexl V, Fonseca-Pereira D, Domingues RG, Veiga-Fernandes H, Arnold SJ, Busslinger M, Dunay IR, Tanriver Y, Diefenbach A: Differentiation of type 1 ilcs from a common progenitor to all helper-like innate lymphoid cell lineages. Cell 2014;157:340-356.

86 Silver JS, Kearley J, Copenhaver AM, Sanden C, Mori M, Yu L, Pritchard GH, Berlin AA, Hunter CA, Bowler R, Erjefalt JS, Kolbeck R, Humbles AA: Inflammatory triggers associated with exacerbations of copd orchestrate plasticity of group 2 innate lymphoid cells in the lungs. Nat Immunol 2016;17:626-635.

87 Bal SM, Bernink JH, Nagasawa M, Groot J, Shikhagaie MM, Golebski K, van Drunen CM, Lutter R, Jonkers RE, Hombrink P, Bruchard M, Villaudy J, Munneke JM, Fokkens W, Erjefalt JS, Spits H, Ros XR: Il-1beta, il-4 and il-12 control the fate of group 2 innate lymphoid cells in human airway inflammation in the lungs. Nat Immunol 2016;17:636-645.

88 Wallrapp A, Riesenfeld SJ, Burkett PR, Abdulnour RE, Nyman J, Dionne D, Hofree M, Cuoco MS, Rodman C, Farouq D, Haas BJ, Tickle TL, Trombetta JJ, Baral P, Klose CSN, Mahlakoiv T, Artis D, Rozenblatt-Rosen O, Chiu IM, Levy BD, Kowalczyk MS, Regev A, Kuchroo VK: The neuropeptide nmu amplifies ilc2-driven allergic lung inflammation. Nature 2017;549:351-356.

89 Bernink JH, Germar K, Spits H: The role of ilc2 in pathology of type 2 inflammatory diseases. Curr Opin Immunol 2014;31:115-120.

90 Jia Y, Fang X, Zhu X, Bai C, Zhu L, Jin M, Wang X, Hu M, Tang R, Chen Z: Il-13+ type 2 innate lymphoid cells correlate with asthma control status and treatment response. Am J Respir Cell Mol Biol 2016;55:675-683.

91 Gronke K, Kofoed-Nielsen M, Diefenbach A: Innate lymphoid cells, precursors and plasticity. Immunol Lett 2016

92 Mebius RE: Organogenesis of lymphoid tissues. Nat Rev Immunol 2003;3:292-303.

93 John-Schuster G, Hager K, Conlon TM, Irmler M, Beckers J, Eickelberg O, Yildirim AO: Cigarette smoke-induced ibalt mediates macrophage activation in a b cell-dependent manner in copd. Am J Physiol Lung Cell Mol Physiol 2014;307:L692-706.

94 Hams E, Bermingham R, Fallon PG: Macrophage and innate lymphoid cell interplay in the genesis of fibrosis. Front Immunol 2015;6:597.

95 Tan HL, Rosenthal M: Il-17 in lung disease: Friend or foe? Thorax 2013;68:788-790.

96 Chan YR, Chen K, Duncan SR, Lathrop KL, Latoche JD, Logar AJ, Pociask DA, Wahlberg BJ, Ray P, Ray A, Pilewski JM, Kolls JK: Patients with cystic fibrosis have inducible il-17+il-22+ memory cells in lung draining lymph nodes. J Allergy Clin Immunol 2013;131:1117-1129, 1129 e1111-1115.

97 Tan HL, Regamey N, Brown S, Bush A, Lloyd CM, Davies JC: The th17 pathway in cystic fibrosis lung disease. American journal of respiratory and critical care medicine 2011;184:252-258.

98 Moretti S, Renga G, Oikonomou V, Galosi C, Pariano M, Iannitti RG, Borghi M, Puccetti M, De Zuani M, Pucillo CE, Paolicelli G, Zelante T, Renauld JC, Bereshchenko O, Sportoletti P, Lucidi V, Russo MC, Colombo C, Fiscarelli E, Lass-Florl C, Majo F, Ricciotti G, Ellemunter H, Ratclif L, Talesa VN, Napolioni V, Romani L: A mast cell-ilc2-th9 pathway promotes lung inflammation in cystic fibrosis. Nat Commun 2017;8:14017.

99 Martin E, Treiner E, Duban L, Guerri L, Laude H, Toly C, Premel V, Devys A, Moura IC, Tilloy F, Cherif S, Vera G, Latour S, Soudais C, Lantz O: Stepwise development of mait cells in mouse and human. PLoS Biol 2009;7:e54.

100 Dusseaux M, Martin E, Serriari N, Peguillet I, Premel V, Louis D, Milder M, Le Bourhis L, Soudais C, Treiner E, Lantz O: Human mait cells are xenobiotic-resistant, tissue-targeted, cd161hi il-17-secreting t cells. Blood 2011;117:1250-1259.

101 Treiner E, Duban L, Bahram S, Radosavljevic M, Wanner V, Tilloy F, Affaticati P, Gilfillan S, Lantz O: Selection of evolutionarily conserved mucosal-associated invariant t cells by mr1. Nature 2003;422:164-169.

102 Leeansyah E, Loh L, Nixon DF, Sandberg JK: Acquisition of innate-like microbial reactivity in mucosal tissues during human fetal mait-cell development. Nat Commun 2014;5:3143.

103 Hinks TS, Wallington JC, Williams AP, Djukanovic R, Staples KJ, Wilkinson TM: Steroid-induced deficiency of mucosal-associated invariant t cells in the chronic obstructive pulmonary disease lung. Implications for nontypeable haemophilus influenzae infection. Am J Respir Crit Care Med 2016;194:1208-1218.

104 Porcelli S, Yockey CE, Brenner MB, Balk SP: Analysis of t cell antigen receptor (tcr) expression by human peripheral blood cd4-8- alpha/beta t cells demonstrates preferential use of several v beta genes and an invariant tcr alpha chain. J Exp Med 1993;178:1-16.

105 Tilloy F, Treiner E, Park SH, Garcia C, Lemonnier F, de la Salle H, Bendelac A, Bonneville M, Lantz O: An invariant t cell receptor alpha chain defines a novel tap-independent major histocompatibility complex class ib-restricted alpha/beta t cell subpopulation in mammals. J Exp Med 1999;189:1907-1921.

106 Mak JY, Xu W, Reid RC, Corbett AJ, Meehan BS, Wang H, Chen Z, Rossjohn J, McCluskey J, Liu L, Fairlie DP: Stabilizing short-lived schiff base derivatives of 5-aminouracils that activate mucosal-associated invariant t cells. Nat Commun 2017;8:14599.

107 McWilliam HE, Birkinshaw RW, Villadangos JA, McCluskey J, Rossjohn J: Mr1 presentation of vitamin b-based metabolite ligands. Curr Opin Immunol 2015;34:28-34.

108 Patel O, Kjer-Nielsen L, Le Nours J, Eckle SB, Birkinshaw R, Beddoe T, Corbett AJ, Liu L, Miles JJ, Meehan B, Reantragoon R, Sandoval-Romero ML, Sullivan LC, Brooks AG, Chen Z, Fairlie DP, McCluskey J, Rossjohn J: Recognition of vitamin b metabolites by mucosal-associated invariant t cells. Nat Commun 2013;4:2142.

109 Kjer-Nielsen L, Patel O, Corbett AJ, Le Nours J, Meehan B, Liu L, Bhati M, Chen Z, Kostenko L, Reantragoon R, Williamson NA, Purcell AW, Dudek NL, McConville MJ, O'Hair RA, Khairallah GN, Godfrey DI, Fairlie DP, Rossjohn J, McCluskey J: Mr1 presents microbial vitamin b metabolites to mait cells. Nature 2012;491:717-723.

110 Gold MC, Cerri S, Smyk-Pearson S, Cansler ME, Vogt TM, Delepine J, Winata E, Swarbrick GM, Chua WJ, Yu YY, Lantz O, Cook MS, Null MD, Jacoby DB, Harriff MJ, Lewinsohn DA, Hansen TH, Lewinsohn DM: Human mucosal associated invariant t cells detect bacterially infected cells. PLoS Biol 2010;8:e1000407.

111 Gibbs A, Leeansyah E, Introini A, Paquin-Proulx D, Hasselrot K, Andersson E, Broliden K, Sandberg JK, Tjernlund A: Mait cells reside in the female genital mucosa and are biased towards il-17 and il-22 production in response to bacterial stimulation. Mucosal Immunol 2017;10:35-45.

112 Gracey E, Qaiyum Z, Almaghlouth I, Lawson D, Karki S, Avvaru N, Zhang Z, Yao Y, Ranganathan V, Baglaenko Y, Inman RD: Il-7 primes il-17 in mucosal-associated invariant t (mait) cells, which contribute to the th17-axis in ankylosing spondylitis. Ann Rheum Dis 2016;75:2124-2132.

113 Kurioka A, Ussher JE, Cosgrove C, Clough C, Fergusson JR, Smith K, Kang YH, Walker LJ, Hansen TH, Willberg CB, Klenerman P: Mait cells are licensed through granzyme exchange to kill bacterially sensitized targets. Mucosal Immunol 2015;8:429-440.

114 Leeansyah E, Svard J, Dias J, Buggert M, Nystrom J, Quigley MF, Moll M, Sonnerborg A, Nowak P, Sandberg JK: Arming of mait cell cytolytic antimicrobial activity is induced by il-7 and defective in hiv-1 infection. PLoS Pathog 2015;11:e1005072.

115 Sattler A, Dang-Heine C, Reinke P, Babel N: Il-15 dependent induction of il-18 secretion as a feedback mechanism controlling human mait-cell effector functions. Eur J Immunol 2015;45:2286-2298.

116 Gold MC, Napier RJ, Lewinsohn DM: Mr1-restricted mucosal associated invariant t (mait) cells in the immune response to mycobacterium tuberculosis. Immunol Rev 2015;264:154-166.

117 Kwon YS, Jin HM, Cho YN, Kim MJ, Kang JH, Jung HJ, Park KJ, Kee HJ, Kee SJ, Park YW: Mucosal-associated invariant t cell deficiency in chronic obstructive pulmonary disease. COPD 2016;13:196-202.

118 Hinks TS, Zhou X, Staples KJ, Dimitrov BD, Manta A, Petrossian T, Lum PY, Smith CG, Ward JA, Howarth PH, Walls AF, Gadola SD, Djukanovic R: Innate and adaptive t cells in asthmatic patients: Relationship to severity and disease mechanisms. J Allergy Clin Immunol 2015;136:323-333.

119 Ishimori A, Harada N, Chiba A, Harada S, Matsuno K, Makino F, Ito J, Ohta S, Ono J, Atsuta R, Izuhara K, Takahashi K, Miyake S: Circulating activated innate lymphoid cells and mucosal-associated invariant t cells are associated with airflow limitation in patients with asthma. Allergol Int 2016

120 Smith DJ, Hill GR, Bell SC, Reid DW: Reduced mucosal associated invariant t-cells are associated with increased disease severity and pseudomonas aeruginosa infection in cystic fibrosis. PLoS One 2014;9:e109891.

121 Kwon YS, Cho YN, Kim MJ, Jin HM, Jung HJ, Kang JH, Park KJ, Kim TJ, Kee HJ, Kim N, Kee SJ, Park YW: Mucosal-associated invariant t cells are numerically and functionally deficient in patients with mycobacterial infection and reflect disease activity. Tuberculosis (Edinb) 2015;95:267-274.

122 Le Bourhis L, Martin E, Peguillet I, Guihot A, Froux N, Core M, Levy E, Dusseaux M, Meyssonnier V, Premel V, Ngo C, Riteau B, Duban L, Robert D, Huang S, Rottman M, Soudais C, Lantz O: Antimicrobial activity of mucosal-associated invariant t cells. Nat Immunol 2010;11:701-708.

123 Jiang J, Yang B, An H, Wang X, Liu Y, Cao Z, Zhai F, Wang R, Cao Y, Cheng X: Mucosal-associated invariant t cells from patients with tuberculosis exhibit impaired immune response. J Infect 2016;72:338-352.

124 Ussher JE, Bilton M, Attwod E, Shadwell J, Richardson R, de Lara C, Mettke E, Kurioka A, Hansen TH, Klenerman P, Willberg CB: Cd161++ cd8+ t cells, including the mait cell subset, are specifically activated by il-12+il-18 in a tcr-independent manner. Eur J Immunol 2014;44:195-203.

125 Loh L, Wang Z, Sant S, Koutsakos M, Jegaskanda S, Corbett AJ, Liu L, Fairlie DP, Crowe J, Rossjohn J, Xu J, Doherty PC, McCluskey J, Kedzierska K: Human mucosal-associated invariant t cells contribute to antiviral influenza immunity via il-18-dependent activation. Proc Natl Acad Sci U S A 2016;113:10133-10138.

126 Jiang J, Chen X, An H, Yang B, Zhang F, Cheng X: Enhanced immune response of mait cells in tuberculous pleural effusions depends on cytokine signaling. Sci Rep 2016;6:32320.

127 Jiang J, Wang X, An H, Yang B, Cao Z, Liu Y, Su J, Zhai F, Wang R, Zhang G, Cheng X: Mucosal-associated invariant t-cell function is modulated by programmed death-1 signaling in patients with active tuberculosis. Am J Respir Crit Care Med 2014;190:329-339.

128 Saeidi A, Tien Tien VL, Al-Batran R, Al-Darraji HA, Tan HY, Yong YK, Ponnampalavanar S, Barathan M, Rukumani DV, Ansari AW, Velu V, Kamarulzaman A, Larsson M, Shankar EM: Attrition of tcr valpha7.2+ cd161++ mait cells in hiv-tuberculosis co-infection is associated with elevated levels of pd-1 expression. PLoS One 2015;10:e0124659.

129 Kawachi I, Maldonado J, Strader C, Gilfillan S: Mr1-restricted v alpha 19i mucosal-associated invariant t cells are innate t cells in the gut lamina propria that provide a rapid and diverse cytokine response. J Immunol 2006;176:1618-1627.

130 Cui Y, Franciszkiewicz K, Mburu YK, Mondot S, Le Bourhis L, Premel V, Martin E, Kachaner A, Duban L, Ingersoll MA, Rabot S, Jaubert J, De Villartay JP, Soudais C, Lantz O: Mucosal-associated invariant t cell-rich congenic mouse strain allows functional evaluation. J Clin Invest 2015;125:4171-4185.

131 Meierovics A, Yankelevich WJ, Cowley SC: Mait cells are critical for optimal mucosal immune responses during in vivo pulmonary bacterial infection. Proc Natl Acad Sci U S A 2013;110:E3119-3128.

132 Meierovics AI, Cowley SC: Mait cells promote inflammatory monocyte differentiation into dendritic cells during pulmonary intracellular infection. J Exp Med 2016;213:2793-2809.

133 van Wilgenburg B, Scherwitzl I, Hutchinson EC, Leng T, Kurioka A, Kulicke C, de Lara C, Cole S, Vasanawathana S, Limpitikul W, Malasit P, Young D, Denney L, consortium S-H, Moore MD, Fabris P, Giordani MT, Oo YH, Laidlaw SM, Dustin LB, Ho LP, Thompson FM, Ramamurthy N, Mongkolsapaya J, Willberg CB, Screaton GR, Klenerman P: Mait cells are activated during human viral infections. Nat Commun 2016;7:11653.

134 Huang YJ, Boushey HA: The sputum microbiome in chronic obstructive pulmonary disease exacerbations. Ann Am Thorac Soc 2015;12 Suppl 2:S176-180.

135 Groenewegen KH, Wouters EF: Bacterial infections in patients requiring admission for an acute exacerbation of copd; a 1-year prospective study. Respir Med 2003;97:770-777.

136 Busse WW, Lemanske RF, Jr., Gern JE: Role of viral respiratory infections in asthma and asthma exacerbations. Lancet 2010;376:826-834.

137 Pelaia G, Vatrella A, Gallelli L, Renda T, Cazzola M, Maselli R, Marsico SA: Respiratory infections and asthma. Respir Med 2006;100:775-784.

138 Bussink AP, Speijer D, Aerts JM, Boot RG: Evolution of mammalian chitinase(-like) members of family 18 glycosyl hydrolases. Genetics 2007;177:959-970.

139 Musumeci M, Malaguarnera L, Simpore J, Barone R, Whalen M, Musumeci S: Chitotriosidase activity in colostrum from african and caucasian women. Clin Chem Lab Med 2005;43:198-201.

140 Malaguarnera L, Simpore J, Prodi DA, Angius A, Sassu A, Persico I, Barone R, Musumeci S: A 24-bp duplication in exon 10 of human chitotriosidase gene from the sub-saharan to the mediterranean area: Role of parasitic diseases and environmental conditions. Genes Immun 2003;4:570-574.

141 Manno N, Sherratt S, Boaretto F, Coico FM, Camus CE, Campos CJ, Musumeci S, Battisti A, Quinnell RJ, Leon JM, Vazza G, Mostacciuolo ML, Paoletti MG, Falcone FH: High prevalence of chitotriosidase deficiency in peruvian amerindians exposed to chitin-bearing food and enteroparasites. Carbohydr Polym 2014;113:607-614.

142 Hall AJ, Quinnell RJ, Raiko A, Lagog M, Siba P, Morroll S, Falcone FH: Chitotriosidase deficiency is not associated with human hookworm infection in a papua new guinean population. Infect Genet Evol 2007;7:743-747.

143 Lee P, Waalen J, Crain K, Smargon A, Beutler E: Human chitotriosidase polymorphisms g354r and a442v associated with reduced enzyme activity. Blood Cells Mol Dis 2007;39:353-360.

144 Hector A, Chotirmall SH, Lavelle GM, Mirkovic B, Horan D, Eichler L, Mezger M, Singh A, Ralhan A, Berenbrinker S, Mack I, Ensenauer R, Riethmuller J, Graepler-Mainka U, Murray MA, Griese M, McElvaney NG, Hartl D: Chitinase activation in patients with fungus-associated cystic fibrosis lung disease. J Allergy Clin Immunol 2016;138:1183-1189 e1184.

145 Letuve S, Kozhich A, Humbles A, Brewah Y, Dombret MC, Grandsaigne M, Adle H, Kolbeck R, Aubier M, Coyle AJ, Pretolani M: Lung chitinolytic activity and chitotriosidase are elevated in chronic obstructive pulmonary disease and contribute to lung inflammation. Am J Pathol 2010;176:638-649.

146 Gavala ML, Kelly EA, Esnault S, Kukreja S, Evans MD, Bertics PJ, Chupp GL, Jarjour NN: Segmental allergen challenge enhances chitinase activity and levels of ccl18 in mild atopic asthma. Clin Exp Allergy 2013;43:187-197.

147 Zhu Z, Zheng T, Homer RJ, Kim YK, Chen NY, Cohn L, Hamid Q, Elias JA: Acidic mammalian chitinase in asthmatic th2 inflammation and il-13 pathway activation. Science 2004;304:1678-1682.

148 Wiesner DL, Specht CA, Lee CK, Smith KD, Mukaremera L, Lee ST, Lee CG, Elias JA, Nielsen JN, Boulware DR, Bohjanen PR, Jenkins MK, Levitz SM, Nielsen K: Chitin recognition via chitotriosidase promotes pathologic type-2 helper t cell responses to cryptococcal infection. PLoS Pathog 2015;11:e1004701.

149 Kanneganti M, Kamba A, Mizoguchi E: Role of chitotriosidase (chitinase 1) under normal and disease conditions. J Epithel Biol Pharmacol 2012;5:1-9.

150 Piras I, Melis A, Ghiani ME, Falchi A, Luiselli D, Moral P, Varesi L, Calo CM, Vona G: Human chit1 gene distribution: New data from mediterranean and european populations. J Hum Genet 2007;52:110-116.

151 Boot RG, Renkema GH, Verhoek M, Strijland A, Bliek J, de Meulemeester TM, Mannens MM, Aerts JM: The human chitotriosidase gene. Nature of inherited enzyme deficiency. J Biol Chem 1998;273:25680-25685.

152 Marion CR, Wang J, Sharma L, Losier A, Lui W, Andrews N, Elias JA, Kazmierczak BI, Roy CR, Dela Cruz CS: Chitinase 3-like 1 (chil1) regulates survival and macrophage-mediated interleukin-1beta and tumor necrosis factor alpha during pseudomonas aeruginosa pneumonia. Infect Immun 2016;84:2094-2104.

153 Dela Cruz CS, Liu W, He CH, Jacoby A, Gornitzky A, Ma B, Flavell R, Lee CG, Elias JA: Chitinase 3-like-1 promotes streptococcus pneumoniae killing and augments host tolerance to lung antibacterial responses. Cell Host Microbe 2012;12:34-46.

154 Lee CM, He CH, Nour AM, Zhou Y, Ma B, Park JW, Kim KH, Dela Cruz C, Sharma L, Nasr ML, Modis Y, Lee CG, Elias JA: Il-13ralpha2 uses tmem219 in chitinase 3-like-1-induced signalling and effector responses. Nat Commun 2016;7:12752.

155 He CH, Lee CG, Dela Cruz CS, Lee CM, Zhou Y, Ahangari F, Ma B, Herzog EL, Rosenberg SA, Li Y, Nour AM, Parikh CR, Schmidt I, Modis Y, Cantley L, Elias JA: Chitinase 3-like 1 regulates cellular and tissue responses via il-13 receptor alpha2. Cell Rep 2013;4:830-841.

156 Sutherland TE, Logan N, Ruckerl D, Humbles AA, Allan SM, Papayannopoulos V, Stockinger B, Maizels RM, Allen JE: Chitinase-like proteins promote il-17-mediated neutrophilia in a tradeoff between nematode killing and host damage. Nat Immunol 2014;15:1116-1125.

157 Nair MG, Gallagher IJ, Taylor MD, Loke P, Coulson PS, Wilson RA, Maizels RM, Allen JE: Chitinase and fizz family members are a generalized feature of nematode infection with selective upregulation of ym1 and fizz1 by antigen-presenting cells. Infect Immun 2005;73:385-394.

158 Gasteiger G, D'Osualdo A, Schubert DA, Weber A, Bruscia EM, Hartl D: Cellular innate immunity: An old game with new players. J Innate Immun 2017;9:111-125.

159 Gaggar A, Hector A, Bratcher PE, Mall MA, Griese M, Hartl D: The role of matrix metalloproteinases in cystic fibrosis lung disease. The European respiratory journal 2011;38:721-727.

160 Griese M, Kappler M, Gaggar A, Hartl D: Inhibition of airway proteases in cystic fibrosis lung disease. European Respiratory Journal 2008;32:783-795.