Cell. Mol. Life Sci. 66 (2009) 553 – 558
1420-682X/09/040553-6
DOI 10.1007/s00018-009-8506-y

Cellular and Molecular Life Sciences

Visions & Reflections (Minireview)

Chronic granulomatous disease

B. H. Segal[a, *], L. Romani[b] and P. Puccetti[b]

[a] Departments of Medicine and Immunology, Division of Infectious Diseases, Roswell Park Cancer Institute, Elm & Carlton Streets, Buffalo, NY 14263 (USA), Fax: +1-716-845-5777, e-mail: brahm.segal@roswellpark.org
[b] Department of Experimental Medicine, University of Perugia, Perugia (Italy)

Received 18 August 2008; received after revision 29 November 2008; accepted 16 December 2008
Online First 5 February 2009

Abstract. Chronic granulomatous disease is an inherited disorder of the NADPH oxidase characterized by severe bacterial and fungal infections and disordered inflammation. We propose that NADPH oxidase has a key role in regulating acute neutrophilic and T cell responses, which in turn restrains fungal growth and calibrates the inflammatory response to minimize injury and allergy. In this model, superoxide-induced activation of indoleamine 2,3-dioxygenase (IDO) is a central mechanism by which the optimal balance of antifungal host defense and immune tolerance occurs. This model is based on studies in mice and requires correlation in humans.

Keywords. Chronic granulomatous disease, NADPH oxidase, inflammation, Interleukin-17, indoleamine 2,3-dioxygenase.

Chronic granulomatous disease: A disorder of host defense and excessive inflammation

Chronic granulomatous disease (CGD) is an inherited disorder of the NADPH oxidase complex in which phagocytes are defective in generating the superoxide anion and downstream reactive oxidant intermediates (ROIs), hydrogen peroxide (H_2O_2), hydroxyl anion, and hypohalous acid. As a result of the defect in this key host defense pathway, CGD patients suffer from recurrent life-threatening bacterial and fungal infections [1]. CGD is also characterized by abnormally exuberant inflammatory responses leading to granuloma formation, such as granulomatous enteritis, genitourinary obstruction, and poor wound healing and dehiscence (pathological opening up of a surgical wound) [2, 3]. "Mulch pneumonitis" is a recently described life-threatening complication in CGD characterized by rapid-onset life-threatening pulmonary inflammation following mould exposure, and treated with systemic antifungals and prolonged corticosteroids [4].

Activated NADPH oxidase is responsible for the respiratory burst – a rapid consumption of oxygen – that occurs following neutrophil stimulation. The phagocyte NADPH oxidase functions to rapidly generate superoxide anion by transferring electrons from NADPH to molecular O_2 in response to physiological stimuli such as ligation of specific pathogen recognition receptors (Fig. 1A). The cytochrome, composed of $gp91^{phox}$ (phox, phagocyte oxidase) and $p22^{phox}$, is embedded in membranes. In neutrophils, approximately 85% of the cytochrome is in the membranes of specific granules or gelatinase-containing granules, and the remainder is present in the plasma membrane and in secretory granules [5, 6]. The NADPH binding site is located on the cytoplasmic side of membranes. Upon activation of the oxidase, the cytoplasmic subunits $p47^{phox}$, $p67^{phox}$,

* Corresponding author.

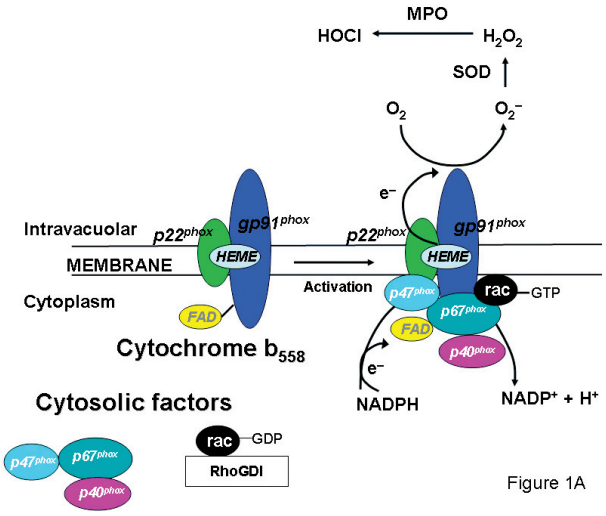

Figure 1. (*A*) Schematic of NADPH oxidase activation. NADPH oxidase activation requires translocation of the cytoplasmic phox constituents and rac to the membrane-bound flavocytochrome. Superoxide anion is the direct product of NADPH oxidase activation. Superoxide dismutase (SOD) converts superoxide anion to H_2O_2. Myeloperoxidase (MPO) produces hypohalous acid from H_2O_2 and halide anions (*e.g.*, chloride) during the neutrophil's respiratory burst.

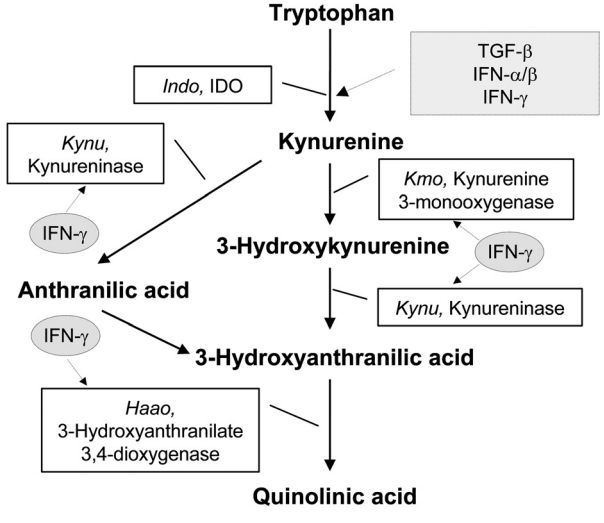

Figure 1. *(continued)* (*B*) Schematic representation of how superoxide anion regulates indoleamine 2,3-dioxygenase (IDO)-dependent counter-inflammatory homeostatic responses in potentially harmful inflammation (for full mechanistic molecular insight, see Macchiarulo et al. [40]). Tryptophan catabolism along the so-called kynurenine pathway leads to tryptophan starvation and the production of immunoregulatory kynurenines, the combined effects of which result in an arrest in T cell proliferation, induction of T-helper cell apoptosis, reversible impairment of T cell activity through down-regulation of T cell receptor ζ-chain, and the generation or activation of IL-10-producing regulatory T (Treg) cells. In addition to IDO-encoding *Indo*, all kynurenine pathway enzyme genes (indicated) are activated transcriptionally by pro-inflammatory cytokines.

and $p40^{phox}$ appear to translocate *en bloc* to the membrane-bound cytochrome. Activation of *rac*, a member of the low molecular weight GTP-binding proteins, and translocation of *rac* to the membrane-bound cytochrome are also critical for NADPH oxidase activation [7, 8]. In the unstimulated state, *rac* is bound in the cytoplasm to RhoGDI, the GDP dissociation inhibitor of Rho.

NADPH is oxidized to $NADP^+$, and electrons are transported down a reducing potential gradient to FAD and then possibly to two non-identical heme groups. On the vacuolar or extracellular side of the membrane, the final step in the electron transport chain occurs when oxygen accepts an electron and is converted to superoxide anion. CGD results from disabling mutations in genes encoding $gp91^{phox}$, $p22^{phox}$, $p47^{phox}$, or $p67^{phox}$.

How does NADPH oxidase mediate host defense?

Neutrophil-mediated killing of pathogens has been segregated into ROI-dependent and ROI-independent pathways. The ROI products of NADPH oxidase activation have direct antimicrobial properties. Myeloperoxidase produces microbicidal hypohalous acid from H_2O_2 and halide anions (*e.g.*, chloride) during the neutrophil's respiratory burst. ROIs can also interact with reactive nitrogen intermediates to produce cytotoxic microbicidal products (*e.g.*, peroxynitrite anion).

In addition to the direct antimicrobial effects of ROIs, neutrophil NADPH oxidase activation is linked to key intracellular signaling events critical to host defense. Reeves et al. [9] showed that activation of the NADPH oxidase in neutrophils leads to an influx of reactive oxidant species into the endocytic vacuole, resulting in an accumulation of anionic charge. To maintain electrogenic neutrality, K^+ ions cross the membrane in a pH-dependent manner. The rise in ionic strength leads to the release of cationic granule proteins, including elastase and cathepsin G, which are bound to the anionic proteoglycan matrix in the inactivated state. Knockout mice deficient in the cationic granule proteins have a similar phenotype to CGD mice regarding susceptibility to experimental bacterial and fungal infections [9, 10]. Taken together, activation of preformed granular proteases is likely to be a principal pathway for NADPH oxidase-mediated destruction of pathogens.

Invasive aspergillosis in CGD

Invasive aspergillosis is a major cause of morbidity and mortality in CGD [11, 12]. Although NADPH oxidase-independent pathways are inadequate to prevent invasive pulmonary aspergillosis, they are able to damage hyphae and protect against vascular invasion in genetically engineered CGD mice [13]. However, even a low virulent mutant strain of *Aspergillus nidulans* caused mortality in pulmonary aspergillosis in CGD mice due to excessive inflammation [14], emphasizing the unique pathophysiological features of aspergillosis in CGD. Indeed, administration of heat-killed hyphae [15] or branched beta-glucans (fungal cell wall constituents) [16] stimulated robust pulmonary inflammation in CGD, but not wild-type, mice. This study shows that NADPH oxidase has a dual role as both a mediator of innate host defense and as an anti-inflammatory pathway.

Taken together, invasive aspergillosis in CGD is characterized by two interrelated features: impaired ability to damage hyphae and dysregulated inflammation. Knowledge of the immunopathogenesis of aspergillosis in CGD illuminates mechanisms by which NADPH oxidase defends against *Aspergillus* species while also restraining excessive inflammation induced by fungal cell wall constituents.

Defective tryptophan catabolism in CGD mice

Using the CGD mouse model, Romani et al. showed that the NADPH oxidase, specific pathogen recognition receptors, and regulatory T cells (Tregs) have key interactive roles in orchestrating inflammation and antifungal resistance [17]. Tryptophan metabolism regulates both antimicrobial resistance and protective tolerance to pro-inflammatory microbial motifs. Indoleamine 2,3-dioxygenase (IDO) and the other kynurenine pathway enzymes represent not only effector host defense pathways, but also, paradoxically, a means of generating Tregs with anti-inflammatory, tolerogenic activity [18, 19]. Tregs and IL-17-producing T cells mediate opposing responses. IL-17 stimulates production of G-CSF, GM-CSF, TNF-α, and chemokines that regulate myelopoiesis and neutrophil recruitment to inflammatory sites [20–22]. The IL-23/IL-17 (IL-23 expands Th17 cells) axis mediates several experimental autoimmune disorders, including colitis, collagen-induced arthritis, and experimental autoimmune encephalomyelitis, and is a promising target for drug development [23].

Since superoxide is a co-factor of IDO, the rate-limiting enzyme in tryptophan degradation along the kynurenine pathway (Fig. 1B), we hypothesized that the absence of NADPH oxidase-derived superoxide in CGD would lead to an augmentation of IL-17-driven inflammation and a reduction in Treg lymphocyte responses. Montagnoli et al. previously showed that naturally occurring Tregs are recruited early in the inflammatory response to *Aspergillus* in wild-type mice, and are capable of suppressing inflammation [24]. IL-17 augmented inflammation, but, paradoxically, impaired neutrophil antifungal host defense in mice [25]. Romani et al. subsequently demonstrated that IDO-mediated tryptophan metabolism along the kynurenine pathway is defective in CGD mice [17]. When challenged with intratracheal *Aspergillus fumigatus*, unrestrained $\gamma\delta$ T cell and $\alpha\beta$ Th17 expansion, defective regulatory Treg activity, and acute inflammatory lung injury leading to mortality occurred in CGD mice. Although beneficial effects were induced by IL-17 neutralization or depletion of $\gamma\delta$ T cells (which produce IL-17) in CGD mice, complete protection from invasive fungal disease and reversal of the hyperinflammatory phenotype were achieved by recombinant interferon-γ and administration of a natural kynurenine distal to the IDO blockade. Effective therapy restored production of downstream immunoactive metabolites and enabled the emergence of Treg responses.

Similar to *Aspergillus* challenge, intratracheal zymosan (a sterile fungal cell wall constituent that ligates Toll-like receptor 2 and dectin-1), caused dramatically augmented lung inflammation and IL-17 production and diminished Treg responses in CGD compared to wild-type mice, emphasizing the intrinsic regulatory role of the NADPH oxidase on inflammation (manuscript in preparation). George-Chandy et al. [26] recently showed that gp91$^{phox-/-}$ dendritic cells (DCs) enhance induction of both Th17 and Th1 responses *in vitro* compared to wild-type DCs. T cell development was dependent exclusively on the NADPH oxidase status of the activating DCs rather than the responding T cells, which also harbor a functional NADPH oxidase [27].

These results provide a model illuminating the basis of both the impaired antifungal resistance and excessive inflammation in CGD (Fig. 2). NADPH oxidase activation has multiple signaling roles, including activation of granular proteases and IDO. Defective IDO activation in CGD mice exacerbates both the impaired antifungal host defense and excessive inflammation in aspergillosis, by skewing T cell responses toward the Th17 phenotype and impairing the development of tolerogenic responses. This imbalance in T cell responses can be remedied by either depletion of IL-17 or administration of kynurenine, a tryptophan metabolite distal to the IDO blockade. Kynurenine products resulting from IDO-mediated

tryptophan catabolism drive Treg development and restrain IL-17 production, likely by inducing apoptosis of IL-17-producing lymphocyte subsets [17].

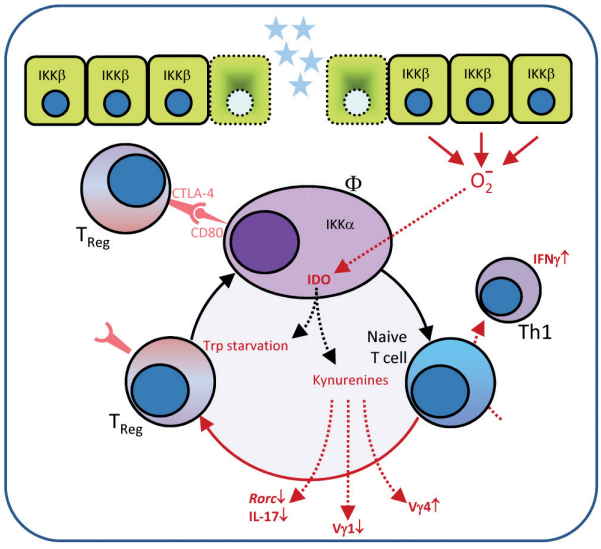

Figure 2. Hypothetical model of how superoxide-driven induction of IDO is essential for the maintenance of immune homeostasis in the airways, a mechanism apparently deficient in chronic granulomatous disease (CGD) mice. A degree of inflammation (epitomized here by activation of the canonical pathway of NF-κB, requiring IKKβ kinase) is probably required for local antimicrobial protection, but over-reacting response are prevented by superoxide anion, which activates the noncanonical NF-κB-dependent (IKKα) function of IDO in phagocytes and accessory cells (Φ). In addition to general effects mentioned in the legend to Figure 1B, the immunoregulatory activity of kynurenines may further result in transcriptional suppression of IL-17-encoding *Rorc*, contraction of an IL-17-producing $V\gamma1^+$ T cell population, expansion of a regulatory $V\gamma4^+$ component, and development of a T-helper type 1 (Th1) response, which produces IL-17-opposing interferon γ (IFN-γ). Newly generated Treg cells locally reinforce the counter-inflammatory response through CTLA-4/CD80-dependent 'reverse signaling'. Trp, tryptophan.

Prior studies have shown that IL-17 up-regulates the expression of antimicrobial peptides in target cells. IL-17 and IL-22 promote the production of antimicrobial peptides in human keratinocytes [28, 29]. IL-17 up-regulates β-defensin-2 expression in human airway epithelium [30]. The antibacterial activity of defensins is enhanced by ROIs in lung tissue [31]. These studies raise a paradox: if the effect of IL-17 appears to augment the expression of antimicrobial peptides, why then in our studies does IL-17 hamper neutrophil-mediated antifungal host defense?

Cystic fibrosis (CF) and CGD, although completely different genetic diseases, have some common features that may provide insight. They are both associated with aspergillosis. CF is commonly associated with persistent airway colonization with *Aspergillus* species, leading to allergic (rather than invasive) aspergillosis [32]. *Staphylococcus aureus* and *Burkholderia cepacia* infections are also common in both diseases. Both CF and CGD are associated with an abundance of lung inflammation during infection and impaired host defense. In CF, dysfunction of neutrophils independent of salt sensitivity occurs within the lung but not peripherally, arguing that the inflammatory milieu in the lung depresses neutrophil function [33, 34]. In addition, very high levels of human neutrophil peptides (HNP; α-defensins) in sputum have been reported in CF patients [35], despite the antimicrobial properties of airway surface fluid being compromised [34]. Finally, there is a growing appreciation of IL-17 in the pathogenesis of CF [36]. It is unknown if IL-17 directly or indirectly regulates HNP production. The data on CF and our data in CGD mice support the notion of locally produced factors in the lung that disable host defense; IL-17 may play a complex role by enhancing host defense against some microbes and disabling defense against others.

Therapeutic options in CGD

Stem cell transplantation is curative in CGD, but transplant-related mortality is significant, particularly in cases of donor/recipient HLA antigen disparity [37]. Since CGD is a disorder of myeloid stem cells, gene therapy is an attractive option, but maintaining a stable population of myeloid gene-corrected cells has been a persistent challenge [38]. The strategy of non-myeloablative conditioning followed by gene therapy facilitates the expansion of gene-corrected cells [39]. Additional novel therapeutic options are necessary for this rare disorder. Our studies suggest that shifting the balance between pro-inflammatory IL-17-driven responses to IDO-induced Treg responses is a potential therapeutic strategy in CGD. We emphasize that these studies were performed in CGD mice and results cannot be extrapolated to patients.

1 Segal, B. H., Leto, T. L., Gallin, J. I., Malech, H. L. and Holland, S. M. (2000) Genetic, biochemical, and clinical features of chronic granulomatous disease. Medicine (Baltimore) 79, 170–200
2 Rosenzweig, S. D. (2008) Inflammatory manifestations in chronic granulomatous disease (CGD). J. Clin. Immunol. 28 Suppl 1, S67–72
3 Marciano, B. E., Rosenzweig, S. D., Kleiner, D. E., Anderson, V. L., Darnell, D. N., Anaya-O'Brien, S., Hilligoss, D. M., Malech, H. L., Gallin, J. I. and Holland, S. M. (2004) Gastrointestinal involvement in chronic granulomatous disease. Pediatrics 114, 462–468
4 Siddiqui, S., Anderson, V. L., Hilligoss, D. M., Abinun, M., Kuijpers, T. W., Masur, H., Witebsky, F. G., Shea, Y. R., Gallin, J. I., Malech, H. L. and Holland, S. M. (2007) Fulminant mulch pneumonitis: An emergency presentation of chronic granulomatous disease. Clin. Infect. Dis. 45, 673–681

5 Babior, B. M. (1999) NADPH oxidase: An update. Blood 93, 1464–1476
6 Thrasher, A. J., Keep, N. H., Wientjes, F. and Segal, A. W. (1994) Chronic granulomatous disease. Biochim. Biophys. Acta 1227, 1–24
7 Abo, A., Pick, E., Hall, A., Totty, N., Teahan, C. G. and Segal, A. W. (1991) Activation of the NADPH oxidase involves the small GTP-binding protein p21rac1. Nature 353, 668–670
8 Knaus, U. G., Heyworth, P. G., Evans, T., Curnutte, J. T. and Bokoch, G. M. (1991) Regulation of phagocyte oxygen radical production by the GTP-binding protein Rac 2. Science 254, 1512–1515
9 Reeves, E. P., Lu, H., Jacobs, H. L., Messina, C. G., Bolsover, S., Gabella, G., Potma, E. O., Warley, A., Roes, J. and Segal, A. W. (2002) Killing activity of neutrophils is mediated through activation of proteases by K^+ flux. Nature 416, 291–297
10 Tkalcevic, J., Novelli, M., Phylactides, M., Iredale, J. P., Segal, A. W. and Roes, J. (2000) Impaired immunity and enhanced resistance to endotoxin in the absence of neutrophil elastase and cathepsin G. Immunity 12, 201–210
11 Segal, B. H., DeCarlo, E. S., Kwon-Chung, K. J., Malech, H. L., Gallin, J. I. and Holland, S. M. (1998) *Aspergillus nidulans* infection in chronic granulomatous disease. Medicine (Baltimore) 77, 345–354
12 Winkelstein, J. A., Marino, M. C., Johnston, R. B. Jr, Boyle, J., Curnutte, J., Gallin, J. I., Malech, H. L., Holland, S. M., Ochs, H., Quie, P., Buckley, R. H., Foster, C. B., Chanock, S. J. and Dickler, H. (2000) Chronic granulomatous disease: Report on a national registry of 368 patients. Medicine (Baltimore) 79, 155–169
13 Dennis, C. G., Greco, W. R., Brun, Y., Youn, R., Slocum, H. K., Bernacki, R. J., Lewis, R., Wiederhold, N., Holland, S. M., Petraitiene, R., Walsh, T. J. and Segal, B. H. (2006) Effect of amphotericin B and micafungin combination on survival, histopathology, and fungal burden in experimental aspergillosis in the p47phox-/- mouse model of chronic granulomatous disease. Antimicrob. Agents Chemother. 50, 422–427
14 Bignell, E., Negrete-Urtasun, S., Calcagno, A. M., Arst, H. N. Jr., Rogers, T. and Haynes, K. (2005) Virulence comparisons of *Aspergillus nidulans* mutants are confounded by the inflammatory response of p47phox-/- mice. Infect. Immun. 73, 5204–5207
15 Morgenstern, D. E., Gifford, M. A., Li, L. L., Doerschuk, C. M. and Dinauer, M. C. (1997) Absence of respiratory burst in X-linked chronic granulomatous disease mice leads to abnormalities in both host defense and inflammatory response to *Aspergillus fumigatus*. J. Exp. Med. 185, 207–218
16 Schappi, M., Deffert, C., Fiette, L., Gavazzi, G., Herrmann, F., Belli, D. and Krause, K. H. (2008) Branched fungal beta-glucan causes hyperinflammation and necrosis in phagocyte NADPH oxidase-deficient mice. J. Pathol. 214, 434–444
17 Romani, L., Fallarino, F., De Luca, A., Montagnoli, C., D'Angelo, C., Zelante, T., Vacca, C., Bistoni, F., Fioretti, M. C., Grohmann, U., Segal, B. H. and Puccetti, P. (2008) Defective tryptophan catabolism underlies inflammation in mouse chronic granulomatous disease. Nature 451, 211–215
18 Romani, L., Zelante, T., De Luca, A., Bozza, S., Bonifazi, P., Moretti, S., D'Angelo, C., Giovannini, G., Bistoni, F., Fallarino, F. and Puccetti, P. (2008) Indoleamine 2,3-dioxygenase (IDO) in inflammation and allergy to *Aspergillus*. Med. Mycol. 12, 1–8
19 Puccetti, P. and Grohmann, U. (2007) IDO and regulatory T cells: A role for reverse signalling and non-canonical NF-kappaB activation. Nat. Rev. Immunol. 7, 817–823
20 Gaffen, S. L., Kramer, J. M., Yu, J. J. and Shen, F. (2006) The IL-17 cytokine family. Vitam. Horm. 74, 255–282
21 Ley, K., Smith, E. and Stark, M. A. (2006) IL-17A-producing neutrophil-regulatory Tn lymphocytes. Immunol. Res. 34, 229–242
22 Romani, L., Zelante, T., De Luca, A., Fallarino, F. and Puccetti, P. (2008) IL-17 and therapeutic kynurenines in pathogenic inflammation to fungi. J. Immunol. 180, 5157–5162
23 Kastelein, R. A., Hunter, C. A. and Cua, D. J. (2007) Discovery and biology of IL-23 and IL-27: Related but functionally distinct regulators of inflammation. Annu. Rev. Immunol. 25, 221–242
24 Montagnoli, C., Fallarino, F., Gaziano, R., Bozza, S., Bellocchio, S., Zelante, T., Kurup, W. P., Pitzurra, L., Puccetti, P. and Romani, L. (2006) Immunity and tolerance to *Aspergillus* involve functionally distinct regulatory T cells and tryptophan catabolism. J. Immunol. 176, 1712–1723
25 Zelante, T., De Luca, A., Bonifazi, P., Montagnoli, C., Bozza, S., Moretti, S., Belladonna, M. L., Vacca, C., Conte, C., Mosci, P., Bistoni, F., Puccetti, P., Kastelein, R. A., Kopf, M. and Romani, L. (2007) IL-23 and the Th17 pathway promote inflammation and impair antifungal immune resistance. Eur. J. Immunol. 37, 2695–2706
26 George-Chandy, A., Nordstrom, I., Nygren, E., Jonsson, I. M., Postigo, J., Collins, L. V. and Eriksson, K. (2008) Th17 development and autoimmune arthritis in the absence of reactive oxygen species. Eur. J. Immunol. 38, 1118–1126
27 Jackson, S. H., Devadas, S., Kwon, J., Pinto, L. A. and Williams, M. S. (2004) T cells express a phagocyte-type NADPH oxidase that is activated after T cell receptor stimulation. Nat. Immunol. 5, 818–827
28 Wilson, N. J., Boniface, K., Chan, J. R., McKenzie, B. S., Blumenschein, W. M., Mattson, J. D., Basham, B., Smith, K., Chen, T., Morel, F., Lecron, J. C., Kastelein, R. A., Cua, D. J., McClanahan, T. K., Bowman, E. P. and de Waal Malefyt, R. (2007) Development, cytokine profile and function of human interleukin 17-producing helper T cells. Nat. Immunol 8, 950–957
29 Liang, S. C., Tan, X. Y., Luxenberg, D. P., Karim, R., Dunussi-Joannopoulos, K., Collins, M. and Fouser, L. A. (2006) Interleukin (IL)-22 and IL-17 are coexpressed by Th17 cells and cooperatively enhance expression of antimicrobial peptides. J. Exp. Med. 203, 2271–2279
30 Kao, C. Y., Chen, Y., Thai, P., Wachi, S., Huang, F., Kim, C., Harper, R. W. and Wu, R. (2004) IL-17 markedly up-regulates beta-defensin-2 expression in human airway epithelium *via* JAK and NF-kappaB signaling pathways. J. Immunol. 173, 3482–3491
31 Porro, G. A., Lee, J. H., de Azavedo, J., Crandall, I., Whitehead, T., Tullis, E., Ganz, T., Liu, M., Slutsky, A. S. and Zhang, H. (2001) Direct and indirect bacterial killing functions of neutrophil defensins in lung explants. Am. J. Physiol. Lung Cell. Mol. Physiol. 281, L1240–1247
32 Stevens, D. A., Moss, R. B., Kurup, V. P., Knutsen, A. P., Greenberger, P., Judson, M. A., Denning, D. W., Crameri, R., Brody, A. S., Light, M., Skov, M., Maish, W., Mastella, G., Participants in the Cystic Fibrosis Foundation Consensus Conference (2003) Allergic bronchopulmonary aspergillosis in cystic fibrosis – State of the art: Cystic Fibrosis Foundation Consensus Conference. Clin. Infect. Dis. 37 Suppl 3, S225–264
33 Bals, R., Weiner, D. J., Meegalla, R. L., Accurso, F. and Wilson, J. M. (2001) Salt-independent abnormality of antimicrobial activity in cystic fibrosis airway surface fluid. Am. J. Respir. Cell. Mol. Biol. 25, 21–25
34 Moraes, T. J., Plumb, J., Martin, R., Vachon, E., Cherepanov, V., Koh, A., Loeve, C., Jongstra-Bilen, J., Zurawska, J. H., Kus, J. V., Burrows, L. L., Grinstein, S. and Downey, G. P. (2006) Abnormalities in the pulmonary innate immune system in cystic fibrosis. Am. J. Respir. Cell. Mol. Biol. 34, 364–374
35 Soong, L. B., Ganz, T., Ellison, A. and Caughey, G. H. (1997) Purification and characterization of defensins from cystic fibrosis sputum. Inflamm. Res. 46, 98–102
36 Dubin, P. J., McAllister, F. and Kolls, J. K. (2007) Is cystic fibrosis a TH17 disease? Inflamm. Res. 56, 221–227
37 Horwitz, M. E., Barrett, A. J., Brown, M. R., Carter, C. S., Childs, R., Gallin, J. I., Holland, S. M., Linton, G. F., Miller, J. A., Leitman, S. F., Read, E. J. and Malech, H. L. (2001) Treatment of chronic granulomatous disease with nonmyeloablative conditioning and a T-cell-depleted hematopoietic allograft. N. Engl. J. Med. 344, 881–888

38 Malech, H. L., Maples, P. B., Whiting-Theobald, N., Linton, G. F., Sekhsaria, S., Vowells, S. J., Li, F., Miller, J. A., DeCarlo, E., Holland, S. M., Leitman, S. F., Carter, C S., Butz, R. E., Read, E. J., Fleisher, T. A., Schneiderman, R. D., Van Epps, D. E., Spratt, S. K., Maack, C. A., Rokovich, J. A., Cohen, L. K. and Gallin, J. I. (1997) Prolonged production of NADPH oxidase-corrected granulocytes after gene therapy of chronic granulomatous disease. Proc. Natl. Acad. Sci. USA. 94, 12133–12138

39 Ott, M. G., Schmidt, M., Schwarzwaelder, K., Stein, S., Siler, U., Koehl, U., Glimm, H., Kühlcke, K., Schilz, A., Kunkel, H., Naundorf, S., Brinkmann, A., Deichmann, A., Fischer, M., Ball, C., Pilz, I., Dunbar, C., Du, Y., Jenkins, N. A., Copeland, N. G., Lüthi, U., Hassan, M., Thrasher, A. J., Hoelzer, D., von Kalle, C., Seger, R. and Grez, M. (2006) Correction of X-linked chronic granulomatous disease by gene therapy, augmented by insertional activation of MDS1-EVI1, PRDM16 or SETBP1. Nat. Med. 12, 401–409

40 Macchiarulo, A., Camaioni, E., Nuti, R. and Pellicciari, R. (2008) Highlights at the gate of tryptophan catabolism: A review on the mechanisms of activation and regulation of indoleamine 2,3-dioxygenase (IDO), a novel target in cancer disease. Amino Acids [Epub ahead of print]