Nature Reviews Drug Discovery | AOP, published online 16 May 2014; doi:10.1038/nrd4344

LUNG DISEASE

Resetting the redox balance in lung fibrosis

Fibrotic disorders such as idiopathic pulmonary fibrosis (IPF) are strongly associated with ageing, but the underlying mechanisms are unclear. Hecker and colleagues now report that a loss of redox homeostasis in aged mice contributes to the persistence of senescent — but apoptosis-resistant — myofibroblasts, which may explain the reduced ability of these animals to resolve fibrosis. Moreover, the authors show that inhibition of NADPH oxidase 4 (NOX4) — an enzyme that generates reactive oxygen species — reverses this myofibroblast phenotype and prolongs survival in an aged mouse model of persistent lung fibrosis.

> “collagen levels in the lungs of GKT137831-treated animals were similar to uninjured mice”

Image Zoo/Alamy

Bleomycin-induced lung injury in young (2-month-old) mice triggers a normal fibrotic response, which peaks at 3 weeks post-injury and then self-resolves, in part through apoptosis of myofibroblasts. However, in aged (18-month-old) mice the fibrotic response persists for months and lung myofibroblasts continue to accumulate and produce extracellular matrix, leading to IPF-like symptoms.

Using this ageing model of persistent fibrosis, the authors showed that after bleomycin injury, lung fibroblasts from aged mice exhibit increased levels of senescence markers and B cell lymphoma 2 (BCL-2), an anti-apoptotic marker, compared with fibroblasts from young mice. Moreover, the authors demonstrated that the senescent, apoptosis-resistant myofibroblast phenotype observed in aged mice was mediated by a redox imbalance. This imbalance was associated with deficient activation of the transcription factor NFE2-related factor 2 (NRF2) — a master regulator of antioxidant genes — and sustained activation of NOX4. This previously unknown profibrotic mechanism may help to explain why IPF develops more frequently in older individuals.

Hecker *et al.* then investigated NOX4 as a possible therapeutic target by treating aged mice with intranasal NOX4-targeting small interfering RNA (siRNA) on alternate days between weeks 3 and 6 after bleomycin injury. Compared with lung fibroblasts from controls, fibroblasts from anti-NOX4 siRNA-treated animals exhibited markedly reduced levels of BCL-2 and senescence markers, and produced less collagen α1 (a major component of fibrotic scar tissue).

Finally, animals were treated daily in weeks 3 to 6 after bleomycin exposure with oral doses (40 mg per kg) of the NOX1 and NOX4 dual inhibitor GKT137831. Compared with controls, lungs harvested from animals treated with GKT137831 6 weeks after bleomycin injury exhibited fewer senescent myofibroblasts. Furthermore, unlike the vehicle-treated controls, GKT137831-treated mice recovered their baseline body weight within the 6-week observation period. In addition, collagen levels in the lungs of GKT137831-treated animals were similar to uninjured mice and, importantly, more animals treated with GKT137831 survived the 6 weeks following bleomycin injury than did vehicle-treated animals (~95% compared with ~75%).

Taken together, these data indicate that NOX4 inhibition using compounds such as GKT137831 may reset the redox balance and thus promote the susceptibility of senescent myofibroblasts to apoptosis. Therefore, NOX4 inhibitors such as GKT137831 (which is currently in Phase II trials for diabetic nephropathy) might be useful against age-associated fibrotic diseases such as IPF.

Natasha Bray

ORIGINAL RESEARCH PAPER Hecker, L. *et al.* Reversal of persistent fibrosis in aging by targeting Nox4-Nrf2 redox imbalance. *Sci. Trans. Med.* **6**, 231ra47 (2014)