REVIEW

Targeting inducible epigenetic reprogramming pathways in chronic airway remodeling

Allan R Brasier MD1, Istvan Boldogh DM&B, PhD2

1Institute for Clinical and Translational Research, University of Wisconsin-Madison School of Medicine and Public Health, 4246 Health Sciences Learning Center, 750 Highland Ave, Madison, WI 53705, USA; 2Department of Microbiology and Immunology, University of Texas Medical Branch at Galveston, 301 University Blvd, Galveston, TX 77555, USA

Abstract

Allergic asthma is a chronic inflammatory airway disease whose clinical course is punctuated by acute exacerbations from aeroallergen exposure or respiratory virus infections. Aeroallergens and respiratory viruses stimulate toll-like receptor (TLR) signaling, producing oxidative injury and inflammation. Repetitive exacerbations produce complex mucosal adaptations, cell-state changes, and structural remodeling. These structural changes produce substantial morbidity, decrease lung capacity, and impair quality of life. We will review recent systems-level studies that provide fundamental new insights into how repetitive activation of innate signaling pathways produce epigenetic ‘training’ to induce adaptive epithelial responses. Oxidative stress produced downstream of TLR signaling induces transient oxidation of guanine bases in the regulatory regions of inflammatory genes. The epigenetic mark 8-oxoG is bound by a pleiotropic DNA repair enzyme, 8-oxoguanine DNA glycosylase (OGG1), which induces conformational changes in adjacent DNA to recruit the NFκB-bromodomain-containing protein 4 (BRD4) complex. The NFκB-BRD4 complex not only plays a central role in inflammation, but also triggers mesenchymal transition and extracellular matrix remodeling. Small molecule inhibitors of OGG1-8-oxoG binding and BRD4–acetylated histone interaction have been developed. We present studies demonstrating efficacy of these in reducing airway inflammation in preclinical models. Targeting inducible epigenetic reprogramming pathway shows promise for therapeutics in reversing airway remodeling in a variety of chronic airway diseases.

Keywords: 8-oxoguanine DNA glycosylase (OGG1), airway remodeling, bromodomain-containing protein 4 (BRD4), epigenetics, mesenchymal transition, myofibroblast.

Citation

Brasier AR, Boldogh I. Targeting inducible epigenetic reprogramming pathways in chronic airway remodeling. Drugs in Context 2019; 8: 2019-8-3. DOI: 10.7573/dic.2019-8-3

Introduction

Allergic asthma (AA) is a chronic, relapsing disease that affects ~339 million people worldwide.1 Heterogeneous in nature, AA is typically characterized by Th2- and Th17-polarized lymphocytic inflammation in the airway and variable degrees of bronchial hyperreactivity. The clinical course of AA is punctuated by intercurrent acute exacerbations (AEs). AEs are episodes of obstructive symptoms, including shortness of breath, wheezing, coughing, and mucous production. Mechanistically, these clinical deteriorations are due to inflammation-induced small airway constriction and edema, decreasing expiratory airflow and producing mucous plugging, resulting in ball-valve small airway obstruction.2 Epidemiological studies show that AEs are provoked by environmental interactions, including aeroallergen exposure, viral upper respiratory tract infections, or environmental oxidants.3 AEs produce substantial clinical impact. AEs are responsible for unscheduled visits that produce significant healthcare costs. In the United States alone, AEs account for 15 million outpatient visits, 2 million emergency room (ER) visits, and 500,000 hospitalizations annually.4 Moreover, AEs diminish the quality of life in patients and their families.5 In addition to acute worsening of disease, prospective observational studies indicate that AAs with frequent AEs are a distinct phenotype, frequently in association with glucocorticoid resistance.6 Notably, this phenotype is prone to structural remodeling, producing a functional decline in
l lung function. Mechanistically, AEs are the result of toll-like receptor (TLR)-induced inflammation, producing remodeling through complex mucosal adaptions through epigenetic reprogramming. In this review, we will give an overview of the presence of mucosal environmental-inducible epigenetic changes and mechanistic pathways controlling them that influence airway remodeling and inform potential therapeutic strategies. This is a literature review using PubMed searches for allergic asthma, airway remodeling, epigenetics, and innate inflammation.

**AEs and airway remodeling**

AA is a highly heterogeneous disease in its etiology, triggers, and clinical course. In particular, a subset of ‘exacerbation-prone’ AAs can be identified that exhibit differences in the course of their disease. Most strikingly, AAs with a history of recent severe exacerbation requiring an ER visit or hospitalization in the past 3 months are at significantly increased risk of having recurrent, future exacerbations. This relationship was confirmed in a 3-year multicenter observational study of difficult-to-treat asthmatics – The Epidemiology and Natural History of Asthma: Outcomes and Treatment Regimens (TENOR) Study – and a prospective study involving mild–moderate asthmatics presenting to the ER. Emerging data indicate that the exacerbation-prone phenotype is more likely to have substantial reduction in pulmonary function. For example, in the abovementioned TENOR study, the frequency of exacerbations was linked to reductions in pulmonary function. In the US Severe Asthma Research Program (SARP) study, exacerbation-prone subjects had a greater frequency of irreversible airflow limitation; this association of exacerbations and reduced airflow has been reproduced in the European Network for Understanding Mechanisms of Severe Asthma (ENFUMOSA) cohort. Similarly, lower respiratory tract infections in early life are associated with reduced lung function and increased airway reactivity (wheezeing) that persists for as much as a decade after the infection. A 20-year follow-up study of respiratory syncytial virus-induced lower respiratory tract infection (LRTI) in infancy found that LRTI was an independent risk factor for decreased lung mechanics. These essential findings have been replicated in an independent 18-year follow-up study in a Swedish cohort as well as the Dutch Avon Longitudinal Study of Parents and Children (ALSPAC) study. Moreover, the Tucson Children’s Respiratory Study identified reduced pulmonary function in children at school age who had respiratory syncytial virus (RSV) bronchiolitis before the age of 3 years. This finding is significant because long-term follow-up studies of reduced lung function in childhood are predictive of adult chronic obstructive pulmonary disease (COPD) and asthma–COPD overlap syndrome. Even repetitive methacholine-induced bronchoconstriction produces enhanced extracellular matrix deposition and remodeling. Collectively, these data indicate AEs of any type trigger airway remodeling.

Reductions in pulmonary function are the consequence of airway remodeling. This term refers to a constellation of structural changes of the cellular components and their supporting extracellular matrix in the pulmonary tree. These changes include collagen deposition in the subepithelial basement membrane, disruption of the epithelial barrier, epithelial cell-state change (mucous metaplasia and/or mesenchymal transition), and smooth muscle hypertrophy. Collectively, airway remodeling narrows the small airways, producing obstruction and reducing lung compliance, and is associated with hyperreactivity to nonspecific stimuli.

**Epithelial innate inflammation**

Epithelial cells represent the initial surface that responds to viruses and aeroallergens in the process of provoking an AE. Not solely a passive barrier, the epithelial cell dynamically responds to environmental exposures, through signal transduction pathways affecting the expression of homeostatic gene- and protein expression programs. These dynamic responses are determined by the type of exposure and location of the cell in the respiratory tree. Of particular focus here, environmental signals trigger innate signaling pathways through families of TLRs. TLRs play a central role in AEs of lung disease by producing mucin, stimulating leukocytic infiltration, and mesenchymal transition that drives fibrosis and remodeling. In addition, the epithelial innate response shapes the evolution of downstream Th2- and Th17-type adaptive immunity characteristic of asthma. In AA, aeroallergens induce a robust small airway epithelial transforming growth factor-beta 1 (TGFβ) response, important in activation of interleukin 13 (IL-13)-producing innate lymphoid type 2 (ILC2) cells and initiating an allergic response. Epithelial gene expression programs – and consequently secreted chemokines – produced by innate signaling in the upper airway are overlapping, but they are functionally distinct from programs produced by the lower airway epithelium in response to the same stimuli. These cell-type distinctions have been observed in gene expression and protein expression studies. In particular, unbiased proteomics studies showed that, compared to proximal (tracheal) epithelial cells, bronchiolar-derived epithelial cells produce over 106 distinct proteins in response to viral infections. These factors include a subset of NF-κB-dependent Th2-polarizing chemokines, including chemokine (C-C motif) ligand 20 (CCL20)/macrophage-inducible protein 3α, thymic stromal lymphopoietin (TSLP), IL6, and CCL3-like 1 that are functionally and immunologically relevant to the pathogenesis of AA. To provide greater insight into the functional role of the small airway epithelial cell in viral-induced inflammation, we examined the response of a conditional knockout of the nuclear factor kappa B (NFκB)/RelA transcription factor subunit in small airway bronchiolar cells. Interestingly, these animals are protected from TLR3-induced leukocytic inflammation and RSV-induced airway obstruction. These findings indicate that a special type of
bronchiolar epithelial cells derived from the secretoglobin expressing-small airway bronchiolar cell progenitor cells are a major sentinel cell responsible for Th2 polarizing and mucogenic cytokine production.

We and others have found that NFκB is activated in upper (nasal and tracheal) airway cells in response to TLR3 stimulation and intracellular viral replication,33–35 yet these cells produce less CCL20/TSLP/IL6 than lower airway cells. The explanation for the intriguing discrepancy how NFκB activation in bronchiolar epithelial cells produces a distinct expression pattern from that in upper airway cells lies in differences in the chromatin organization of these cells. Small airway epithelial cells are preprogrammed/primed to elaborate unique cytokine expression patterns.

Epigenetic control of gene expression programs through ‘innate training’

Epigenetics is a term that refers to heritable changes in gene expression that are controlled independently of primary DNA sequence. These changes are stable and inheritable, influencing environmental susceptibility to airway disease.36 The mechanisms for epigenetic control can be at several levels, including direct DNA modification (methylation and oxidation), histone post-translational modifications, and changes in microRNA (miRNA) expression. Epigenetic changes play important roles in cellular differentiation, cell fate decisions, and cell state transitions that underlie Th2 polarization, dendritic cell (DC) activation, and epithelial cellular adaptation.37,38

Although epigenetic regulation can be stable, recent work has shown that histone modifications, chromatin accessibility, and miRNA can be dynamically changed in response to innate signaling. This process has been best described in monocyte biology and referred to as ‘training innate immunity’ resulting in immunological memory.39 However, epigenetic reprogramming also occurs in the epithelium, affecting extracellular matrix remodeling and inducible type III interferon (IFN) response.40 In this review, we will focus on the innate training in epithelial cells relevant to coupling AEs with airway remodeling.

Innate-inducible DNA oxidative modifications function as an epigenetic regulator

Liganded TLRs induce reactive oxidative stress (ROS), a second messenger that stimulates the release of growth factors and cytokines linked to airway remodeling. In concert with the second messenger function, inducible ROS produce oxidative DNA damage, an event that alters gene expression programs in addition to its role as a potential mutagen.41 Among the DNA bases, guanine is the most highly sensitive base to ROS because of its low oxidation potential. Oxidation of guanine results in the formation of 7,8 dihydro-8-oxoguanine (8-oxoG) at guanine-rich promoter regions.42 8-oxoG has emerged as dynamic and reversible epigenetic signal in oxidative innate immune responses because this modification is selectively recognized by 8-oxoguanine DNA glycosylase (OGG1). OGG1 is a pleiotropic protein important in DNA damage repair and innate signaling. Of relevance here, OGG1 binding facilitates the recruitment of active transcription factor, NFκB, to promoters of a subset of highly inflammation-inducible genes (Figure 1). These genes control expression of neutrophilic chemokines, including chemokine (C-X-C motif) ligand 2 (CXCL2), a cytokine important in the rapid leukocytic inflammation in response to TNF43 and pollen allergens.44

OGG1-induced recruitment of NFκB to regulatory chromatin is associated with rapid and highly inducible gene expression. Mechanistically, activated NFκB is bound to the transcriptional elongation complex (PTEFb), a complex composed of cyclin-dependent kinase (CDK)9 and bromodomain-containing protein 4 (BRD4).45 BRD4 facilitates phosphorylation of RNA polymerase II,46 regulating its enzymatic processivity and RNA splicing functions, resulting in the rapid expression of inflammatory genes.47-48 In addition, we recently found that the association of RelA also induced the atypical histone acetyl transferase (HAT) activity of BRD4, acetylating histone H3 on Lys (K) 122, a modification that destabilizes nucleosomes, enhancing transcription through gene bodies.49,50 Consequently, the coactivator enzymatic properties of BRD4 mediates cytokine production, neutrophilia, leukocytic infiltration, and clinical manifestations of disease.26,32,51–53 In this manner, OGG1 nucleates chromatin remodeling complexes to innate genes (Figure 1).

Innate-inducible epigenetic marks affect genomic organization

The core component of chromatin is the nucleosome, a unit consisting of 142 bases of DNA wrapped around a histone octamer. The octamer is composed of two sets of histone H2A, H2B and H3 and H4 molecules with the internucleosome DNA stability H1 monomer. The nucleosomes protect DNA from damage and occlude transcription factors from binding; consequently, highly expressed genes are associated with nucleosome-free upstream control regions. There has been an explosion of the detailed biochemical understanding of histone modifications that affect nucleosomal structure and function.54 Genes whose regulatory elements are associated with acetylated histones (e.g., the H3K27 acetylation mark) are typically in a configuration accessible to transcription factors and can be constitutively or inducibly expressed. By contrast, genes with methylated histones, for example, H3K27(me)3, are in heterochromatin states and silenced.55

Increasingly, it has been recognized that genes involved in innate responses are associated with both activated and inactivated histone marks, so-called ‘metastable’ genes.40,56
Epigenetic mechanisms control cell-state transition

Chronic oxidative stress induced by innate signaling prompt adaptive cell-state transitions of the normal epithelium to a dedifferentiated mesenchymal-like state, called type II epithelial mesenchymal transition (EMT). Type II EMT involves extensive cytosolic restructuring, resulting in the loss of apical-basal polarity, dissolution of adherens junctions, enhanced motility, and expression of fibrotic genes (see Kalluri and Weinberg [2009] and Ijaz, Pazdrak, Kalita, and colleagues [2014] for in-depth reviews). As a consequence of this process, epithelial cells acquire stem-cell-like properties, permitting the transitioned mesenchymal cell to repopulate regions of denuded epithelium, promoting tissue repair and extracellular matrix remodeling.

The EMT program involves coordinating epigenetic reprogramming of ~3000 genes mediated by a core group of mesenchymal transcription factors, including SNAI1 and RelA. This program inhibits expression of differentiated epithelial cadherin (CDH1) and upregulates core EMT transcription factors, mesenchymal intermediate filaments, and extracellular matrix (ECM)-modifying genes (Figure 2). Transition to the mesenchymal state is the product of sequential cell-state changes beginning from the

Nucleosomes binding metastable genes can be dynamically shifted between active and inactivate states in response to cellular stimuli resulting in the processes of derepression and activation, working in parallel, the process of derepression and activation results in highly dynamic increase in gene expression.

BRD4 is a dynamically responsive chromatin modifying and organizing factor

Through its acetyl lysine-binding bromodomains, BRD4 is essential for the maintenance of higher order chromatin configuration. In particular, BRD4 is enriched in enhancer regions (aka ‘superenhancers’) with other chromatin modifying factors controlling the expression of tissue-specific genes. These superenhancers result in high-level, constitutive gene expression and coordinate with expression of distant gene through looping interactions. These interchromosomal contacts are thought to maintain gene expression programs controlling cell-type identity. In response to inflammatory/TLR signaling, BRD4 superenhancers are repositioned to inflammatory and fibrotic gene expression networks.
REVIEW – Inducible epigenetic reprogramming pathways

differentiated epithelial state transitioning into uncommitted ‘partial EMT (pEMT)’ state(s).\(^6^3\) Chromatin immunoprecipitation and histone-profiling studies have shown that initial responses in type II EMT are mediated predominately by coordinate reversible histone marks in the absence of changes in DNA methylation. Genome-wide ChiP-seq studies found a reduction in the heterochromatin mark H3 Lys9 dimethylation (H3K9Me2), an increase in the euchromatin mark H3 Lys4 trimethylation (H3K4Me3).\(^6^4\) Other histone profiling focusing on early changes in EMT have found accumulation of the repressive H3K27(me)3 mark,\(^6^5\) a post-translational modification maintained by the PRC repressor complex, associated with type III IFN silencing.\(^4^0\)

**Activated NFκB·BRD4 drives EMT programs**

Our recent unbiased RNA sequencing studies discovered that NFκB is upstream of the ‘core’ mesenchymal transcription factors SNAI1, zinc finger homeodomain enhancer-binding protein (ZEB), and V-Jun avian sarcoma virus 17 oncogene homolog (JUN) qualifying its status as a ‘master transcriptional regulator’ of EMT. Master regulators of the EMT are a subset of transcription factors engaged in the coordinate regulation of ‘cliques’ of downstream transcription factors by maintaining their expression by the formation of superenhancers.\(^5^8,6^2\) Systems-level studies have shown the essential role of BRD4 in mediating the coordinated gene expression changes underlying EMT.\(^6^2\) NFκB activation repositions BRD4-enriched superenhancers to inflammation-related genes in a cell type-dependent manner.\(^5^8\) In the case of type II EMT in airway remodeling, we have shown that NFκB repositions BRD4 to the promoters of mesenchymal regulatory factors, including SNAI, ZEB, and basic helix-loop-helix transcription factor (Twist),\(^6^6\) activating their expression by the transcriptional elongation.

**Targeting therapeutics to the OGG1-8-oxoG and NFκB-BRD4 complexes in airway inflammation/remodeling**

Collectively, the studies mentioned earlier provide a rich mechanistic understanding of the mucosal response to innate inflammation. ROS generated by TLR signaling induce site-specific 8-oxoG formation in regions of open chromatin. Binding of OGG1 to its substrate gene regulatory regions and consequential alterations in adjacent DNA sequences facilitates the recruitment of NFκB-BRD4 complex resulting in the rapid expression of innate inflammatory genes. Over time, repetitive NFκB activation of innate inflammation either by aeroallergens,\(^5^7\) oxidized DNA base products,\(^5^8\) or viral infections\(^6^9\) induces innate training as a result of epigenetic
reprogramming and elaboration of the epithelial mesenchymal transition (Figure 1).

These studies inform the development of small molecule inhibitors of OGG1 and BRD4 as first-in-class inhibitors of innate-induced epigenetic reprogramming important in airway remodeling. High throughput screening of inhibitors of OGG1 binding to genomic 8-oxoG resulted in the identification of TH5487, a cell-permeable active-site binding inhibitor. TH5487 prevents OGG1 chromatin binding at nontoxic concentrations suppressing inflammatory gene expression and TNF-induced lung inflammation in vivo. These data demonstrated that epigenetic inhibitors targeting oxidative DNA repair can reduce airway inflammation. The effect of the small molecule OGG1 inhibitors recapitulates the earlier discovery that genetic deficiency of OGG1 is associated with resistance to inflammation.

**BRD4 inhibitor development**

Advancement of small molecule inhibitors directed to BRD4 has been the subject of intense medicinal chemistry work. These approaches initially focused on fragment-based ligand design based on the structurally conserved bromodomain (BD) important in low-affinity acetylated-histone recognition, important in chromatin interaction. Consequently, a series of nonselective BD small molecule inhibitors were developed with interesting properties in antiproliferation, anti-inflammation, and antifibrotic activity. Because of the structural similarity of the BDs across the entire BET family, the majority of these first generation inhibitors were not BRD4-selective.

Our laboratory’s recent development and validation of a highly specific BRD4 inhibitor with nanomolar binding affinity and 30-fold specificity over the closely related BRD2 isoform has advanced the field by providing a useful probe for the testing of the role of BRD4 in pathophysiological conditions in vivo. These BRD4 inhibitors disrupt BRD4 activity at multiple levels, including disruption of the extensive BRD4 protein–protein interaction complex, dissolution BRD4-rich superenhancers, and inhibition of its atypical HAT activity. Consequently, BRD4 inhibitors show potential to interfere with mucosal inflammation and airway remodeling in response to viruses and allergen challenges. Reduction in airway remodeling has been through reversal of the mesenchymal

| Inhibitor       | Sponsor                  | Indication                                                                 | CT.gov identifier |
|-----------------|--------------------------|----------------------------------------------------------------------------|-------------------|
| Apabetalone     | Steeve Provencher        | Pulmonary artery hypertension                                               | NCT03655704      |
| SF1126          | SignalRX Pharmaceuticals, Inc. | Advanced hepatocellular cancer                                               | NCT03059147      |
| AZD9150         | AstraZeneca              | Relapsed/refractor non-Hodgkin’s lymphoma (PRISM)                           | NCT03527147      |
| AZDS153         | AstraZeneca              | Refractory solid tumors                                                     | NCT03205176      |
| AZDS153         | AstraZeneca              | Lymphoma                                                                   | NCT03205176      |
| PLX51107        | MDACC/National Cancer Institute | AML/myelodysplastic syndrome                                               | NCT04022785      |
| Olaparib        | National Cancer Institute | Metastatic CA with DNA repair defects                                       | NCT03375307      |
| BSM-986158      | Dana Farber Cancer Institute | Bromodomain and extraterminal domain (BET) inhibitor BMS-986158 in pediatric cancer | NCT03936465      |
| CPI-0610        | Constellation Pharmaceuticals | Peripheral nerve tumors                                                   | NCT02986919      |
| CPI-0610        | Constellation Pharmaceuticals | Lymphoma                                                               | NCT01949883      |
| GSK2820151      | GlaxoSmithKline          | Metastatic and unresectable solid tumors                                  | NCT02630251      |
| GSK25762        | GlaxoSmithKline          | Pharmacokinetics, pharmacodynamics, and clinical activity in NUT midline carcinoma and other cancers | NCT01587703, NCT01587703, NCT01943851 |
| INCB057643      | Incyte Corporation       | Advanced-stage cancer                                                      | NCT02711137      |
| ODM-207         | Orion                    | Solid tumors                                                               | NCT03035591      |
| CC-90010        | Celgene                  | Lymphoma, solid tumors                                                     | NCT03220347      |
| FT-1101         | Forma Therapeutics       | AML, myelodysplastic syndrome                                              | NCT02543879      |
| ABBV-744        | AbbVie                   | Prostate cancer                                                            | NCT03360006      |
| RVX-000222      | Resverlogix Corporation  | T2DM, CAD                                                                  | NCT02586155      |
| RVX-000222      | Resverlogix Corporation  | Chronic kidney failure                                                     | NCT03160430      |

AML, acute myeloid leukemia; CAD, coronary artery disease; DNA, deoxyribonucleic acid; NUT, nuclear carcinoma of the testis; T2DM, type 2 diabetes mellitus.
transition and reduction in the unfolded protein response. A recent demonstration of BRD4 inhibitors in TLR3-induced remodeling has been demonstrated by unbiased optical clearing secondary harmonic generation in a mouse model.

These first-generation inhibitors have been advanced to identify small molecule inhibitors for selective BDs as well as chemistries that degrade the target molecule using proteolysis targeting chimera (PROTAC), promoting ubiquitin-mediated proteolysis of the target molecule (reviewed in Cochran, Conery, and Sims [2019]). BRD4 inhibitors being tested in CT.gov-registered clinical trials in humans are shown in Table 1. The majority of indications are related to the treatment of solid and hematological malignancies.

Advancing BRD4 inhibitors into the clinic for treatment of airway remodeling will require the advancement of biomarkers of BRD4 effect. Biomarkers of BRD4 inhibition in airway disease have been identified recently using systems-level pharmacoproteomics approaches. This latter study discovered that BRD4 inhibitors interfered with vascular permeability and pericyte-myofibroblast transition, indicating that BRD4 inhibitors ameliorate multiple downstream homeostatic components of the coordinate mucosal injury-repair response.

### Summary

Inducible mucosal epigenetic responses to mucosal injury underlie a coordinate inflammatory and remodeling response. In this review, we present evidence for guanine residue oxidation as an inflammation-inducible epigenetic mark. This epigenetic change is dynamic and highly reversible. 8-oxoG is recognized by OGG1, recruiting the NFκB transcription factor–BRD4 complex. The evidence that BRD4 is a central nexus in inflammation-mediated remodeling is compelling. Efforts to disrupt the 8-oxoG–OGG1–NFκB–BRD4 epigenetic cascade has been successful with the development of two classes of OGG1 and BRD4 inhibitors. These compounds will find numerous clinical applications in treatment of acute inflammation and chronic remodeling.

### Contributions:
Both authors contributed equally to the preparation of this review. The named authors meet the International Committee of Medical Journal Editors (ICMJE) criteria for authorship for this article, take responsibility for the integrity of the work as a whole, and have given their approval for this version to be published.

### Disclosure and potential conflicts of interest:
Dr Brasier reports grants from the National Institutes of Health in relation to this work. He is the founder of Quadragenics, but receives no direct funding from this. In addition, he has a patent PCT/US2017/066107 issued. Dr Boldogh declares that he has no conflicts of interest. The International Committee of Medical Journal Editors (ICMJE) Potential Conflicts of Interests form for the authors is available for download at [http://www.drugsincontext.com/wp-content/uploads/2019/10/dic.2019-8-3-COI.pdf](http://www.drugsincontext.com/wp-content/uploads/2019/10/dic.2019-8-3-COI.pdf)

### Acknowledgments:
Funding acknowledgments: US National Institute of Allergy and Infectious Diseases NIAID 2P01AI062885 and the National Center for Advancing Translational Science NCATS UL1TR00237.

### Funding declaration:
There was no funding associated with the preparation of this article.

### Copyright:
Copyright © 2019 Brasier AR, Boldogh I. [https://doi.org/10.7573/dic.2019-8-3](https://doi.org/10.7573/dic.2019-8-3). Published by Drugs in Context under Creative Commons License Deed CC BY NC ND 4.0 which allows anyone to copy, distribute, and transmit the article provided it is properly attributed in the manner specified below. No commercial use without permission.

### Correct attribution:
Copyright © 2019 Brasier AR, Boldogh I. Published by Drugs in Context under Creative Commons License Deed CC BY NC ND 4.0.

### Article URL:
[https://www.drugsincontext.com/targeting-inducible-epigenetic-reprogramming-pathways-in-chronic-airway-remodeling/](https://www.drugsincontext.com/targeting-inducible-epigenetic-reprogramming-pathways-in-chronic-airway-remodeling/)

### Correspondence:
Allan R Brasier, MD, Institute for Clinical and Translational Research, University of Wisconsin-Madison School of Medicine and Public Health, 4246 Health Sciences Learning Center, 750 Highland Ave, Madison, WI 53705, USA. [abrasier@wisc.edu](mailto:abrasier@wisc.edu)

### Provenance:
invited; externally peer reviewed.

### Submitted:
26 August 2019; Peer review comments to author: 11 September 2019; Revised manuscript received: 16 September 2019; Accepted: 19 September 2019; Publication date: 23 October 2019.

### Drugs in Context
is published by BioExcel Publishing Ltd. Registered office: Plaza Building, Lee High Road, London, England, SE13 SPT.

BioExcel Publishing Limited is registered in England Number 10038393. VAT GB 252 7720 07.

For all manuscript and submissions enquiries, contact the Editor-in-Chief [gordon.mallarkey@bioexcelpublishing.com](mailto:gordon.mallarkey@bioexcelpublishing.com)

For all permissions, rights and reprints, contact David Hughes [david.hughes@bioexcelpublishing.com](mailto:david.hughes@bioexcelpublishing.com)

### References

1. The Global Asthma Report 2018. Auckland, New Zealand, 2018. [http://www.globalasthmareport.org/Global%20Asthma%20Report%202018.pdf](http://www.globalasthmareport.org/Global%20Asthma%20Report%202018.pdf) [Last accessed October 10, 2019].

2. National Asthma Education and Prevention Program. Expert panel report 3 (ewpr-3): guidelines for the diagnosis and management of asthma-summary report 2007. J Allergy Clin Immunol. 2007;120(Suppl):S94–138. [http://dx.doi.org/10.1016/j.jaci.2007.09.043](http://dx.doi.org/10.1016/j.jaci.2007.09.043)

3. Busse WW, Lemanske RF. Asthma. N Engl J Med. 2001;344:350–362. [http://dx.doi.org/10.1056/NEJM200102013440507](http://dx.doi.org/10.1056/NEJM200102013440507)
4. Dougherty RH, Fahy JV. Acute exacerbations of asthma: epidemiology, biology and the exacerbation-prone phenotype. *Clin Exp Allergy*. 2009;39(2):193–202. http://dx.doi.org/10.1111/j.1365-2222.2008.03157.x

5. Lane S, Molina J, Plusa T. An international observational prospective study to determine the cost of asthma exacerbations (coax). *Respir Med*. 2006;100(3):434–450. http://dx.doi.org/10.1016/j.rmed.2005.06.012

6. Denlinger LC, Phillips BR, Ramratnam S, et al. Inflammatory and comorbid features of patients with severe asthma and frequent exacerbations. *Am J Respir Crit Care Med*. 2017;195(3):302–313. http://dx.doi.org/10.1164/rccm.201602-0419OC

7. Calhoun WJ, Haselkorn T, Miller DP, Omachi TA. Asthma exacerbations and lung function in patients with severe or difficult-to-treat asthma. *J Allergy Clin Immunol*. 2015;136(4):1125–1127 e1124. http://dx.doi.org/10.1016/j.jaci.2015.05.014

8. Griswold SK, Nordstrom CR, Clark S, Gaeta TJ, Price ML, Camargo CA, Jr. Asthma exacerbations in North American adults: who are the “frequent fliers” in the emergency department? *Chest*. 2005;127(5):1579–1586. http://dx.doi.org/10.1378/chest.127.5.1579

9. Moore WC, Bleecker ER, Curran-Everett D, et al. Characterization of the severe asthma phenotype by the national heart, lung, and blood institute’s severe asthma research program. *J Allergy Clin Immunol*. 2007;119(2):405–413. http://dx.doi.org/10.1016/j.jaci.2006.11.639

10. Romagnoli M, Caramori G, Braccioni F, et al. Near-fatal asthma phenotype in the enfumosa cohort. *Clin Exp Allergy*. 2007;37(4):552–557. http://dx.doi.org/10.1111/j.1365-2222.2007.02683.x

11. Zomer-Kooijker K, van der Ent CK, Ermers MJJ, et al. Increased risk of wheeze and decreased lung function after respiratory syncytial virus infection. *PLoS ONE*. 2014;9(1):e87162. http://dx.doi.org/10.1371/journal.pone.0087162

12. Blanken MO, Rovers MM, Molenaar JM, et al. Respiratory syncytial virus and recurrent wheeze in healthy preterm infants. *N Engl J Med*. 2013;368(19):1791–1799. http://dx.doi.org/10.1056/NEJMoai2119177

13. Bont L, Steijn M, van Aalderen WM, Kimpen JL. Impact of wheezing after respiratory syncytial virus infection on health-related quality of life. *Pediatr Infect Dis J*. 2004;23(5):414–417. http://dx.doi.org/10.1091/inf.0000122604.32137.29

14. Bont L, van Aalderen WM, Versteegh J, et al. Airflow limitation during respiratory syncytial virus lower respiratory tract infection predicts recurrent wheezing. *Pediatr Infect Dis J*. 2001;20(3):277–282. http://dx.doi.org/10.1097/00006454-200103000-00012

15. Korppi M, Piippo-Savolainen E, Korhonen K, Remes S. Respiratory morbidity 20 years after RSV bronchiolitis in the first year of life. *Eur Respir J*. 2013;41(4):1032–1038. http://dx.doi.org/10.1183/09031936.00130312

16. Loukogeorgakis S, Kariou A, Christodoulopoulos N, et al. Recurrent wheezing in infancy: Is there a role for RSV bronchiolitis? *Pediatr Pulmonol*. 2004;40(5):455–461. http://dx.doi.org/10.1002/ppul.20058

17. Sigurs N, Aljasmin F, Kjellman B, et al. Asthma and allergy patterns over 2 years after severe RSV bronchiolitis in the first year of life. *Thorax*. 2010;65(12):1045–1052. http://dx.doi.org/10.1136/thx.2009.121582

18. Sigurs N, Gustafsson PM, Bjarnason R, et al. Severe respiratory syncytial virus bronchiolitis in infancy and asthma and allergy at age 13. *Am J Respir Crit Care Med*. 2005;171(2):137–141. http://dx.doi.org/10.1164/rccm.200406-730OC

19. Stein RT, Sherril D, Morgan WJ, et al. Respiratory syncytial virus in early life and risk of wheeze and allergy by age 13 years. *Lancet*. 1999;354(9189):541–545. http://dx.doi.org/10.1016/S0140-6736(98)10321-5

20. Bui DS, Burgess JA, Lowe AJ, et al. Childhood lung function predicts adult chronic obstructive pulmonary disease and asthma–chronic obstructive pulmonary disease overlap syndrome. *Am J Respir Crit Care Med*. 2017;196(1):39–46. http://dx.doi.org/10.1164/rccm.201606-1272OC

21. Grainge CL, Lau LC, Ward JA, et al. Effect of bronchoconstriction on airway remodeling in asthma. *N Engl J Med*. 2011;364(21):2006–2015. http://dx.doi.org/10.1056/NEJMoai1014350

22. Bergeron C, Tulic MK, Hamid Q. Airway remodeling in asthma: from benchside to clinical practice. *Can Respir J*. 2010;17(4):e85–93. http://dx.doi.org/10.1155/2010/318029

23. Al-Muhsen S, Johnson JR, Hamid Q. Remodeling in asthma. *J Allergy Clin Immunol*. 2011;128(3):451–462; quiz 463–454. http://dx.doi.org/10.1016/j.jaci.2011.04.047

24. Takizawa H. Remodeling in small airways of asthma. *Respir Med CME*. 2008;1(2):69–74. http://dx.doi.org/https://doi.org/10.1016/j.rmed.2008.05.001

25. Tian B, Hosoki K, Liu Z, et al. Mucosal bromodomain-containing protein 4 mediates aeroallergen-induced inflammation and remodeling. *J Allergy Clin Immunol*. 2018. http://dx.doi.org/10.1016/j.jaci.2018.09.029

26. Tian B, Hosoki K, Liu Z, et al. Mucosal bromodomain-containing protein 4 mediates aeroallergen-induced inflammation and remodeling. *J Allergy Clin Immunol*. 2018. http://dx.doi.org/10.1016/j.jaci.2018.09.029

27. Denney L, Byrne AJ, Shea TJ, et al. Pulmonary epithelial cell-derived cytokine TGF-β1 is a critical cofactor for enhanced innate lymphoid cell function. *Immunity*. 2015;43(5):945–958. http://dx.doi.org/10.1016/j.immuni.2015.10.012

28. Olszewska-Pazdrak B, Casola A, Saito T, et al. Cell-specific expression of RANTES, MCP-1, and MIP-1α by lower airway epithelial cells and eosinophils infected with respiratory syncytial viruses. *J Virol*. 1998;72(6):4756–4764.

29. Zhang Y, Luxon BA, Casola A, Garofalo RP, Jamaluddin M, Brasier AR. Expression of respiratory syncytial virus-induced chemokine gene networks in lower airway epithelial cells revealed by cDNA microarrays. *J Virol*. 2001;75(19):9044–9058. http://dx.doi.org/10.1128/JVI.75.19.9044-9058.2001
30. Zhao Y, Jamaluddin M, Zhang Y, et al. Systematic analysis of cell-type differences in the epithelial secretome reveals insights into the pathogenesis of respiratory syncytial virus-induced lower respiratory tract infections. *J Immunol* (Baltimore, MD: 1950). 2017;198(8):3345–3364. http://dx.doi.org/10.4049/jimmunol.1601291

31. Tian B, Liu Z, Yang J, et al. Selective antagonists of the bronchial epithelial NF-κB-bromodomain-containing protein 4 pathway in viral-induced airway inflammation. *Cell Rep.* 2018;23(4):1138–1151. http://dx.doi.org/10.1016/j.celrep.2018.03.106

32. Tian B, Yang J, Zhao Y, et al. Central role of the NF-κB pathway in the Scgb1a1-expressing epithelium in mediating respiratory syncytial virus-induced airway inflammation. *J Virol.* 2018;92(11). http://dx.doi.org/10.1128/JVI.00441-18

33. Saito T, Deskin RW, Casola A, et al. Respiratory syncytial virus induces selective production of the chemokine RANTES by upper airway epithelial cells. *J Infect Dis.* 1999;177:497–504. http://dx.doi.org/10.1086/348267

34. Tian B, Zhang Y, Luxon BA, et al. Identification of NF-κB-dependent gene networks in respiratory syncytial virus-infected cells. *J Virol.* 2002;76(13):6800–6814. http://dx.doi.org/10.1128/JVI.76.13.6800-6814.2002

35. Mastronarde JG, He B, Monick MM, Mukaida N, Matsushima K, Hunninghake GW. Induction of interleukin (IL)-8 gene expression by respiratory syncytial virus involves activation of nuclear factor (NF)-κB and NF-IL-6. *J Infect Dis.* 1996;174:262–267. http://dx.doi.org/10.1093/infdis/i6d.2.262

36. Yang J, Schwartz DA. Epigenetic control of gene expression in the lung. *Am J Respir Crit Care Med.* 2011;183(10):1295–1301. http://dx.doi.org/10.1164/rcrm.201010-1579PP

37. Mortaz E, Masjedi MR, Barnes PJ, Adcock IM. Epigenetics and chromatin remodeling play a role in lung disease. *Tanaffos.* 2011;10(4):7–16.

38. Kidd CD, Thompson PJ, Barrett L, Baltic S. Histone modifications and asthma. The interface of the epigenetic and genetic landscapes. *Am J Respir Cell Mol Biol.* 2016;54(1):3–12. http://dx.doi.org/10.1165/rcmb.2015-0050TR

39. Netaa MG. Training innate immunity: the changing concept of immunological memory in innate host defence. *Eur J Clin Invest.* 2013;43(8):881–884. http://dx.doi.org/10.1111/eci.12132

40. Yang J, Tian B, Sun H, Garofalo RP, Brasier AR. Epigenetic silencing of IRF1 dysregulates type III interferon responses to respiratory virus infection in epithelial to mesenchymal transition. *Nat Microbiol.* 2017;2:17086. http://dx.doi.org/10.1038/nmicrobiol.2017.86

41. Choudhary S, Boldogh I, Brasier AR. Inside-out signaling pathways from nuclear reactive oxygen species control pulmonary innate immunity. *J Innate Immun.* 2016. http://dx.doi.org/10.1159/000442254

42. Pan L, Zhu B, Hao W, et al. Oxidized guanine base lesions function in 8-oxoguanine DNA glycosylase1-mediated epigenetic regulation of nuclear factor κB-driven gene expression. *J Biol Chem.* 2016. http://dx.doi.org/10.1074/jbc.M116.751453

43. Hao W, Qi T, Pan L, et al. Effects of the stimuli-dependent enrichment of 8-oxoguanine DNA glycosylase1 on chromatinized DNA. *Redox Biol.* 2018;18:43–53. http://dx.doi.org/10.1016/j.redox.2018.06.002

44. Bacsi A, Aguilera-Aguirre L, Szczesny B, et al. Down-regulation of 8-oxoguanine DNA glycosylase1 expression in the airway epithelium ameliorates allergic lung inflammation. *DNA Repair (Amst).* 2013;12(1):18–26. http://dx.doi.org/10.1016/j.dnarep.2012.10.002

45. Nowak DE, Tian B, Jamaluddin M, et al. RelA Ser276 phosphorylation is required for activation of a subset of NF-kappaB-dependent genes by recruiting cyclin-dependent kinase 9/cyclin T1 complexes. *Mol Cell Biol.* 2008;28(11):3623–3638.

46. Devaiah BN, Lewis BA, Cherman N, et al. BRD4 is an atypical kinase that phosphorylates serine2 of the RNA polymerase ii carboxy-terminal domain. *Proc Natl Acad Sci U S A.* 2012;109(18):6927–6932. http://dx.doi.org/10.1073/pnas.1120422109

47. Brasier AR, Tian B, Jamaluddin M, Kalita MK, Garofalo RP, Lu M. Rela ser276 phosphorylation-coupled lys310 acetylation controls transcriptional elongation of inflammatory cytokines in respiratory syncytial virus infection. *J Virol.* 2011;85(22):11752–11769. http://dx.doi.org/10.1128/JVI.05360-11

48. Yang J, Zhao Y, Kalita M, et al. Systematic determination of human cyclin dependent kinase (cdk)-9 interactome identifies novel functions in RNA splicing mediated by the ddx5/17 rna helicases. *Mol Cell Proteomics.* 2015. http://dx.doi.org/10.1074/mcp.M115.049221

49. Tian B, Yang J, Zhao Y, et al. BRD4 Couples NF-κB/RelA with Airway Inflammation and the IRF-RIG-I Amplification Loop in Respiratory Syncytial Virus Infection. *J Virol.* 2017;91. http://dx.doi.org/10.1128/JVI.00007-17

50. Devaiah BN, Case-Borden C, Gegonne A, et al. BRD4 is a histone acetyltransferase that evicts nucleosomes from chromatin. *Nat Struct Mol Biol.* 2016;23(6):540–548. http://dx.doi.org/10.1038/nsmb.3228

51. Liu Z, Wang P, Chen H, et al. Drug discovery targeting bromodomain-containing protein 4. *J Med Chem.* 2017;60(11):4533–4558. http://dx.doi.org/10.1021/acs.jmedchem.6b01761

52. Liu Z, Tian B, Chen H, Wang P, Brasier AR, Zhou J. Discovery of potent and selective BRD4 inhibitors capable of blocking TLR3-induced acute airway inflammation. *Eur J Med Chem.* 2018;151:450–461. https://doi.org/10.1016/j.ejmech.2018.04.006

53. Tian B, Liu Z, Litvinov J, et al. Efficacy of novel highly specific bromodomain-containing protein 4 inhibitors in innate inflammation-driven airway remodeling. *Am J Respir Cell Mol Biol.* 2019;60(1):68–83. http://dx.doi.org/10.1165/rcmb.2017-0445OC
REVIEW – Inducible epigenetic reprogramming pathways

54. Tessarz P, Kouzarides T. Histone core modifications regulating nucleosome structure and dynamics. Nat Rev Mol Cell Biol. 2014;15(11):703–708. http://dx.doi.org/10.1038/nrm3890

55. Young MD, Willson TA, Wakefield MJ, et al. ChIP-seq analysis reveals distinct H3K27me3 profiles that correlate with transcriptional activity. Nucleic Acids Res. 2011;39(17):7415–7427. http://dx.doi.org/10.1093/nar/gkr416

56. Yang J, Tian B, Brasier AR. Targeting chromatin remodelling in inflammation and fibrosis. In: Donev R, ed. Advances in Protein Chemistry and Structural Biology. Elsevier; 2017.

57. Wang R, Li Q, Helfer CM, Jiao J, You J. Bromodomain protein BRD4 associated with acetylated chromatin is important for maintenance of higher-order chromatin structure. J Biol Chem. 2012;287(14):10738–10752. http://dx.doi.org/10.1074/jbc.M111.323493

58. Whyte WA, Orlando DA, Hnisz D, et al. Master transcription factors and mediator establish super-enhancers at key cell identity genes. Cell. 2013;153(2):307–319. http://dx.doi.org/10.1016/j.cell.2013.03.035

59. Kalluri R, Weinberg RA. The basics of epithelial-mesenchymal transition. J Clin Invest. 2009;119(6):1420–1428. http://dx.doi.org/10.1172/JCI39104

60. Ijaz T, Pazdrak K, Kalita M, et al. Systems biology approaches to understanding epithelial-mesenchymal transition (EMT) in mucosal remodelling and signalling in asthma. World Allergy Organ J. 2014;7(1):13. http://dx.doi.org/10.1186/1939-4551-7-13

61. Tian B, Li X, Kalita M, et al. Analysis of the TGFβ-induced program in primary airway epithelial cells shows essential role of NF-κB/RelA signaling network in type ii epithelial mesenchymal transition. BMC Genomics. 2015;16(1):529. http://dx.doi.org/10.1186/s12864-015-1707-x

62. Chang H, Liu Y, Xue M, et al. Synergistic action of master transcription factors controls epithelial-to-mesenchymal transition. Nucleic Acids Res. 2016;44(6):2514–2527. http://dx.doi.org/10.1093/nar/gkw126

63. Jordan NV, Johnson GL, Abell AN. Tracking the intermediate stages of epithelial-mesenchymal transition in epithelial stem cells and cancer. Cell Cycle. 2011;10(17):2865–2873. http://dx.doi.org/10.4161/cc.10.17.17188

64. McDonald OG, Wu H, Timp W, Doi A, Feinberg AP. Genome-scale epigenetic reprogramming during epithelial-to-mesenchymal transition. Nat Struct Mol Biol. 2011;18(8):867–874. http://dx.doi.org/10.1038/nsmb.2084

65. Lu C, Sidoli S, Kulej K, Ross K, Wu CH, Garcia BA. Coordination between TGF-β cellular signaling and epigenetic regulation during epithelial mesenchymal transition. Epigenetics Chromatin. 2019;12(1):11. http://dx.doi.org/10.1186/s13072-019-0256-y

66. Tian B, Zhao Y, Sun H, Zhang Y, Yang J, Brasier AR. BRD4 mediates NF-κB-dependent epithelial-mesenchymal transition and pulmonary fibrosis via transcriptional elongation. Am J Physiol Lung Cell Mol Physiol. 2016;311(6):L1183–L1201. http://dx.doi.org/10.1152/ajplung.00224.2016

67. Tian B, Hosoki K, Liu Z, et al. Mucosal bromodomain-containing protein 4 (BRD4) mediates aeroallergen-induced inflammation and remodeling. J Allergy Clin Immunol. 2019;143(4):1380–1394. http://dx.doi.org/10.1016/j.jaci.2018.09.029

68. Aguilera-Aguirre L, Hosoki K, Bacsi A, et al. Whole transcriptome analysis reveals a role for OGG1-initiated DNA repair signaling in airway remodeling. Free Radic Biol Med. 2015;89:20–33. http://dx.doi.org/10.1016/j.freeradbiomed.2015.07.007

69. Tian B, Patrikeev I, Ochoa L, et al. NF-kB mediates mesenchymal transition, remodeling, and pulmonary fibrosis in response to chronic inflammation by viral RNA patterns. Am J Respir Cell Mol Biol. 2017;56(4):506–520. http://dx.doi.org/10.1165/rcmb.2016-0259OC

70. Visnes T, Cazares-Korner A, Hao W, et al. Small-molecule inhibitor of OGG1 suppresses proinflammatory gene expression and inflammation. Science. 2018;362(6416):834–839. http://dx.doi.org/10.1126/science.aar8048

71. Touati E, Michel V, Thiberge JM, et al. Deficiency in OGG1 protects against inflammation and mutagenic effects associated with H. pylori infection in mouse. Helicobacter. 2006;11(5):494–505. http://dx.doi.org/10.1111/j.1525-3537.2006.00442.x

72. Duan Y, Guan Y, Qin W, Zhai X, Yu B, Liu H. Targeting BRD4 for cancer therapy: inhibitors and degraders. MedChemComm. 2018;9(11):1779–1802. http://dx.doi.org/10.1039/C8MD00198G

73. Liu Z, Wang P, Chen H, et al. Drug discovery targeting bromodomain-containing protein 4 (BRD4). J Med Chem. 2017;60:4533–4558. http://dx.doi.org/10.1021/acs.jmedchem.6b01761

74. Huang B, Yang KD, Zhou MM, Ozato K, Chen LF. BRD4 coactivates transcriptional activation of NF-kappaB via specific binding to acetylated rela. Mol Cell Biol. 2009;29(5):1375–1387. http://dx.doi.org/10.1128/MCB.01365-08

75. Zhang J, Jamaluddin M, Zhang Y, et al. Type II epithelial-mesenchymal transition upregulates protein N-glycosylation to maintain proteostasis and extracellular matrix production. J Proteome Res. 2019. http://dx.doi.org/10.1021/acs.jproteome.9b00342

76. Ochoa LF, Kholodnykh A, Villarreal P, et al. Imaging of murine whole lung fibrosis by large scale 3D microscopy aided by tissue optical clearing. Sci Rep. 2018;8(1):13348. http://dx.doi.org/10.1038/s41598-018-31182-2

77. Cochran AG, Conery AR, Sims RJ, 3rd. Bromodomains: a new target class for drug development. Nature Rev Drug Discov. 2019;18(8):609–628. http://dx.doi.org/10.1038/s41573-019-0030-7

78. Zhao Y, Tian B, Sun H, et al. Pharmacoproteomics reveal novel protective activity of bromodomain containing 4 inhibitors on vascular homeostasis in TLR3-mediated airway remodeling. J Proteomics. 2019;205:103415. http://dx.doi.org/10.1016/j.jprot.2019.103415