

The Role of Gene-Environment Interactions in the Development of Respiratory Disorders

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ABSTRACT Gene-environment interactions are thought to be critical for several respiratory diseases such as COPD and lung cancer. For some pulmonary diseases, the etiology is well understood as a function of single gene defects; for others, the etiology is mainly determined by exposure to some strong environmental risk factor. But for the overwhelming majority of diseases of the respiratory systems, it is clear that the greatest magnitude of disease burden relates to complex interactions between multiple determinants in one's environment and one's genetic blueprint. Understanding the fundamental gene-environmental interactions in the development of respiratory disorders such as asthma should lead to earlier identification of susceptible individuals and more effective approaches for disease prevention. Moreover, identifying the genes and the pathways that serve as genetic modifiers of adaptive responses to the changing environmental stimuli could lead to new insights into respiratory biology and the identification of new therapeutic targets.

INTRODUCTION

Great progress has been made in understanding the genetic and environmental basis of respiratory diseases. For some pulmonary diseases, the etiology is well understood as a function of single gene defects; for others, the etiology is mainly determined by exposure to some strong environmental risk factor. But for the overwhelming majority of diseases of the respiratory systems, it is clear that the greatest magnitude of disease burden relates to complex interactions between multiple determinants in one's environment and one's genetic blueprint (Au et al. 2001). Even conditions with a single gene defect have widely differing phenotypes within and between families, partly based on gene modifications and distinctive cellular milieus and partly on the basis of divergent environments. Thus, the inter-

action between genes and the environment plays a major role in the pathogenesis of the most common respiratory diseases. Environmental stimuli may activate conserved genetic responses in the lungs and blood inflammatory cells, and thereby initiate or modify the course of the disease process, acting to promote or suppress the disease phenotype. In many instances, discrete signaling pathways are triggered that allow people to adapt to new environmental stimuli or acquired disease states. Identifying the genes and the pathways that serve as genetic modifiers of adaptive responses to the changing environmental stimuli could lead to new insights into respiratory biology and the identification of new therapeutic targets. The primary focus of this presentation is to review the interrelationship between (1) certain environmental factors and genetic susceptibility to respiratory diseases; and (2) evaluate candidate genes that have been linked to most common respiratory diseases in ongoing molecular genetics studies (Xu et al. 2001).

GENETIC FACTORS

Recent attention is focused on understanding the genetic basis for individual susceptibility to the development of allergic as well as respiratory diseases (Feijen et al. 2000; Silverman, 2001). In more recent studies, an emphasis has been concentrated on establishing an association between inheritance of polymorphic chemical metabolizing genes and development of environmental cancer (e.g., lung cancer among cigarette smokers). The early reports of such associations have been very encouraging (Mucci et al. 2001). However, some reported positive associations were not substantiated in subsequent studies using larger sample sizes and different ethnic populations (Au et al. 2001). Some confounding factors that contribute to the discrepancies include ethnic-dependent distribution of variant gene alleles, differential expression of metabolizing genes, and inadequate study design.

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ENVIRONMENTAL FACTORS

The term „environment,, is used in the most general way to signify all chemical, physical, and biologic agents with which humans come in contact, whether that contact is via air, water or food, and whether the contact is occupational, residential, lifestyle, or whatever. In addition, it includes the social milieu which mediates those contacts and which provides a psychological dimension to the human environment. There are a wide variety of known and suspected environmental risk factors for respiratory diseases (Weiss 1999; Walter et al. 2000). Epidemiologic research has demonstrated the important roles played by such factors as smoking, diet, alcoholism, poverty, social support networks, urban air pollution, sedentary lifestyle, infectious agents, and a variety of occupational dusts and fumes (Barnes 1999). These factors have been linked to one or more classes of respiratory disease. Whether measured externally (e.g., industrial hygiene) or internally (e.g., biomarkers of exposure), it is becoming possible to gain greater knowledge of the complex environments in which we live. In regard to chronic diseases, the challenge is not only to measure environmental factors, but to measure those that were present at the etiologically relevant time (i.e., years or even decades before clinical manifestation of disease).

GENE-ENVIRONMENT INTERACTIONS

By taking into account both of these dimensions (gene and environment) simultaneously, it will be possible to dramatically increase the capacity to understand what is happening in the genetic dimension and in the environmental dimension (Walter et al. 2000). This will improve our capacity to: a) identify and characterize the genetic determinants of disease; b) identify and characterize the environmental determinants of disease so that preventive strategies can be planned; c) identify high-risk individuals in the population for targeted prevention; d) reveal novel pathways in these diseases as targets for new therapeutic strategies; and e) tailor drug therapy using genotype information to maximize the beneficial effects and minimize the adverse side effects of new pharmacological therapies.

Few diseases are the consequence of a single genetic or environmental event (Williams 2001). For example, only a small percentage of all cancer cases can be attributed to a single defective or mutant gene, typically identified from familial

clusters of disease (Almand and Carbone 2001). Instead, common allelic variants of susceptibility genes with relatively low penetrance account for a higher percentage of cancers and other chronic diseases overall. Thus, properly controlling environmental, occupational, or lifestyle exposures could prevent more diseases than just those caused by rare, highly penetrant alleles that lead to familial clusters. To get a more accurate picture of risk from exposures, researchers need to understand the nature, significance, and distribution of susceptible genotypes in the general population. Molecular and genetic techniques advanced by the Human Genome Project now make possible studies of individual differences in susceptibility to diseases and dysfunction linked to environmental exposure.

EXAMPLES OF GENE-ENVIRONMENT INTERACTIONS IN RESPIRATORY DISEASES

Chronic Obstructive Pulmonary Disease (COPD)

Chronic obstructive pulmonary disease, or COPD, is a disease that encompasses one or more of the following: (1) emphysema; (2) chronic bronchitis; and (3) chronic asthma. Current understanding of the pathogenesis of COPD, a source of substantial morbidity and mortality, suggests that chronic inflammation leads to the airways obstruction and parenchymal destruction that characterize this condition. Environmental factors, especially tobacco smoke exposure, are known to accelerate longitudinal decline of lung function, and there is substantial evidence that upregulation of inflammatory pathways plays a vital role in this process. Genetic regulation of both inflammatory responses and anti-inflammatory protective mechanisms likely underlies the heritability of COPD observed in family studies (Miki and Satoh 1999; Weiss 1999; Silverman, 2001).

A variety of studies have examined candidate gene loci with association studies, comparing the distribution of variants in genes hypothesized to be involved in the development of COPD in COPD patients and control subjects. For most genetic loci which have been tested, there have been inconsistent results (Silverman 2001). Genetic heterogeneity could contribute to difficulty in replicating associations between studies. In addition, case-control association studies are susceptible to supporting associations based

purely on population stratification, which can result from incomplete matching between cases and controls—including differences in ethnicity. No association studies in COPD have been reported which used family-based controls, a study design which is immune to such population stratification effects. More importantly, no linkage studies have been published in COPD to identify regions of the genome which are likely to contain COPD susceptibility genes—regions in which association studies are likely to be more productive (Walter et al. 2000).

Despite the high prevalence of and mortality from COPD, extensive research on the underlying pathophysiology and specific therapeutics for this disease is, relatively, in its infancy. Several novel molecular targets are being investigated as potential treatments for the disease (Hay 2001). The most exciting new class of compounds is the phosphodiesterase 4 inhibitors; Ariflo (SB 207499)—a member of this class, and the most advanced in development (Phase III)—was reported recently to have significant clinical efficacy in patients with chronic obstructive pulmonary disease (Barnes 2001; Huang et al. 2001).

Emphysema can best be characterized as the progressive destruction of the grape-like sacs that fulfill the lung's basic function: exchanging oxygen in the air for carbon dioxide in the cardiovascular system. Emphysema is the chief "culprit" in COPD. Emphysema begins with the destruction of alveoli, small sac-like structures (resembling bunches of grapes) in the lungs where oxygen from the air is exchanged for carbon dioxide in the blood. The walls of the alveoli are thin and fragile, and are easily damaged. The damage is irreversible and results in permanent "holes" in the tissues of the lower lungs. As alveoli are destroyed, the lungs are able to transfer less and less oxygen to the bloodstream, causing shortness of breath during exercise and eventually even at rest. The lungs also lose their elasticity, so the patient experiences great difficulty exhaling. The bronchial tubes leading to the air sacs may collapse, which traps air in the lungs. This is the condition known as emphysema.

Role of Alpha 1-Antitrypsin Deficiency (AATD) in Emphysema

In normal and healthy individuals, AAT protects the lungs from a natural enzyme (called neutrophil elastase) that helps fight bacteria and clean up dead lung tissue. However, this enzyme

can also eventually damage lung tissue if not neutralized by AAT. If allowed to progress, this form of emphysema becomes chronic and lung tissue continues to be destroyed; eventually it is fatal if the progress is not slowed down or halted. While there are different causes of emphysema (such as smoking and AAT deficiency), the physical signs and symptoms in each case are similar.

AATD related emphysema is caused by an inherited lack of a protective protein called alpha1-antitrypsin (AAT). Every person inherits two AAT genes, one from each parent. A person has AAT deficiency only if he or she inherits two abnormal genes. People who have only one abnormal gene and one normal AAT gene are "carriers" and do not have the disease. Their AAT levels may be lower than normal, but not as low as the deficiency state and are not generally thought to cause significant health problems. People who have AAT deficiency will pass on one abnormal gene to their children, but they will be "carriers" and will not have AAT deficiency. There is a test for determining if a person is a carrier or is AAT deficient.

The marked variability in the development of COPD in response to cigarette smoking has been known for decades, but severe AAT deficiency (PI Z allele) remains the only proven genetic risk factor for COPD (Sigsgaard et al. 2000). With cigarette smoking, PI Z subjects tend to develop more severe pulmonary impairment at an earlier age than non-smoking PI Z individuals. However, PI Z individuals exhibit wide variability in pulmonary function impairment, even among individuals with similar smoking histories. Therefore, other genes and environmental exposures are also likely involved.

Chronic bronchitis is a first step toward impending emphysema/COPD. It often results from „ordinary„ chest infections (colds, flu). Folks with chronic bronchitis know that every, or nearly every, cold they get will "go straight to the lungs". With proper care it need not lead to COPD; (2) in a COPD patient chronic bronchitis contributes to the „gunk„ that we spit up regularly.

Chronic asthma is the third constituent of COPD. Not all COPD patients have asthma, and most certainly most asthma patients do not have COPD. But many of us suffer from it greatly. Already sick, our airways are easily constricted by all manner of airborne nasties. It is why we carry our inhalers. Some authorities do not consider asthma a component of COPD because unlike emphysema and chronic bronchitis, asthma can

be reversed. Asthma is a chronic inflammatory disorder characterised by airflow obstruction. The inflammatory process involves mast cells, antigen presenting cells, eosinophils, neutrophils, airway epithelial cells and TH2 lymphocytes. These cells produce a broad array of pro inflammatory mediators and cytokines that lead to the pathophysiological changes seen in asthma. The improved understanding of this complex disease, the specific cells and the complex mediators has lead to newer insights into the efficacy of various novel and potential therapies (Babu and Holgate 2000).

Asthma has strong genetic and environmental components that interact both in the induction and subsequent expression of the disease phenotypes (Barnes 1999; Holgate 1999; Weiss 1999; Wiesh et al. 1999). Asthma is a complex disease with a phenotype that has been clinically difficult to define. Associated phenotypes including bronchial hyperresponsiveness and atopy have provided useful objective alternatives in genetic and epidemiologic studies. Although asthma genes have not yet been identified, much progress has been made toward this goal. Genetic studies indicate that multiple genes are involved in the pathogenesis of this disease, and chromosomal regions likely to harbor asthma susceptibility genes have been replicated in several studies. Environmental factors, including smoking, diet, and viral respiratory infections, have also been implicated in the etiology of asthma. Directly linking these exposures as causes of asthma, however, has also proved difficult.

Furthermore, interaction between susceptibility genes and environmental factors is probable and is a challenge currently being pursued by investigators worldwide. This phenomenon—denoted as effect modification of environmental exposure by genetic constitution, or gene by environment interaction—suggests that some genetic markers could indicate susceptibility to environmental factors. Thus, understanding the fundamental gene-environmental interactions in the development of asthma should lead to earlier identification of susceptible individuals and more effective approaches for disease prevention.

Lung Cancer

Lung cancer is a result of multiple gene-environment interactions occurring over several decades (Haugen et al. 2000; Almand and Carbone 2001). Lung cancer is a useful model for the study

of the interplay between genetic factors and environmental exposure since the primary etiology is well established. Several polymorphic enzymes that may be important determinants of susceptibility have been demonstrated (Reszka and Wasowicz 2001). Data also provide evidence for sex differences in lung cancer susceptibility. Furthermore, certain chemical carcinogens may contribute to the carcinogenic process in the lung epithelial cells by inducing genomic instability either directly or indirectly through inflammatory processes. Our understanding of lung cancer biology has rapidly expanded in recent years. Lung cancer, unlike most human cancers, can be traced to an environmental risk factor in the majority of cases, and this fact is reflected in the vast number of genetic alterations discovered in lung tumors whose pathogenesis is believed to be mediated by carcinogen exposure. The discovery of these alterations has led to a greater understanding of tumor development.

A vast number of studies are focused on investigating genetic polymorphism in order to estimate genetic contribution to the development of cancer. Possible cancer susceptibility genes have been sought among oncogenes, tumor suppressor genes, DNA repair genes and genes encoding phase I and phase II enzymes (Shields and Harris 2000).

Genetic Polymorphism of Glutathione S-Transferase and Lung Cancer Risk

Large individual differences in the biotransformation of xenobiotics have been explained on the basis of genetic polymorphisms in some detoxifying enzymes, regardless of environmental and occupational exposure (Mucci et al. 2001). Among these enzymes, glutathione S-transferases (GST) constitute a large multigene family of phase II enzymes involved in detoxification of potentially genotoxic chemicals (Reszka and Wasowicz 2001). Five genetic polymorphisms of GST have been well documented (Strange et al. 2001). Total or partial deletions and (or) single nucleotide polymorphisms in alleles encoding GSTM1, GSTM3, GSTP1, GSTT1, GSTZ1 are associated with reduction of enzymatic activity toward several substrates of different GST isoenzymes. In addition, molecular epidemiology studies indicate that a single genetic polymorphism of glutathione S-transferase appears to be a moderate lung cancer risk factor (Hou et al. 2001). However, the risk is higher when interactions with more GST polymorphisms and other risk factors

(e.g. cigarette smoking) occur (Reszka and Wasowicz 2001). Individuals with decreased rate of detoxification, with „high risk,, glutathione S-transferase genotypes have a slightly higher level of carcinogen-DNA adducts and more cytogenetic damages.

CONCLUDING REMARKS

Recent attention is focused on understanding the genetic and environmental basis for individual susceptibility to the development of chronic respiratory diseases. Indeed, gene-environment interactions are thought to be critical for several respiratory diseases such as COPD and lung cancer. These disorders are the result of multiple gene-environment interactions occurring over several decades. Further, more emphasis is being concentrated on establishing an association between inheritance of polymorphic chemical metabolizing genes and development of environmental cancer (e.g., lung cancer among cigarette smokers). AATD-related emphysema is caused by an inherited lack of a protective protein called alpha1-antitrypsin (AAT). Several of the GST genes are polymorphic in humans and are currently being investigated as possible cancer-risk modifiers. Moreover, understanding the fundamental gene-environmental interactions in the development of respiratory disorders such as asthma should lead to earlier identification of susceptible individuals and more effective approaches for disease prevention.

REFERENCES

- Almand B, Carbone DP 2001. Biological considerations in lung cancer. *Cancer Treat Res*, **105**: 1-30.
- Au WW, Oh HY, Grady J, Salama SA, Heo MY 2001. Usefulness of genetic susceptibility and biomarkers for evaluation of environmental health risk. *Environ Mol Mutagen*, **37**: 215-252.
- Babu KS, Holgate ST. Potential and novel therapies for asthma. *J Assoc Physicians India*, **48**: 1096-1102.
- Barnes KC 1999. Gene-environment and gene-gene interaction studies in the molecular genetic analysis of asthma and atopy. *Clin Exp Allergy*, **29 (Suppl 4)**: 47-51.
- Barnes PJ 2001. Potential novel therapies for chronic obstructive pulmonary disease. *Novartis Found Symp*, **234**: 255-672.
- Feijen M, Gerritsen J, Postma DS 2000. Genetics of allergic disease. *Br Med Bull*, **56**: 894-907.
- Haugen A, Ryberg D, Mollerup S, Zienoldiny S, Skaug V, Svendsrud DH 2000. Gene-environment interactions in human lung cancer. *Toxicol Lett*, **112-113**: 233-237.
- Hay DW 2000. Chronic obstructive pulmonary disease: emerging therapies. *Curr Opin Chem Biol*, **4**: 412-419.
- Holgate ST 1999. Genetic and environmental interaction in allergy and asthma. *J Allergy Clin Immunol*, **104**: 1139-1146.
- Hou SM, Falt S, Nyberg F 2001. Glutathione S-transferase T1-null genotype interacts synergistically with heavy smoking on lung cancer risk. *Environ Mol Mutagen*, **38**: 83-86.
- Huang Z, Ducharme Y, Macdonald D, Robichaud A 2001. The next generation of PDE4 inhibitors. *Curr Opin Chem Biol*, **5**: 432-438.
- Miki M, Satoh K 1999. Genetic risk factors for chronic obstructive pulmonary disease (COPD). *Nippon Rinsho*, **57**: 1954-1958.
- Mucci LA, Wedren S, Tamimi RM, Trichopoulos D, Adami HO 2001. The role of gene-environment interaction in the aetiology of human cancer: examples from cancers of the large bowel, lung and breast. *J Intern Med*, **249**: 477-493 (2001).
- Reszka E, Wasowicz W 2001. Significance of genetic polymorphisms in glutathione S-transferase multigene family and lung cancer risk. *Int J Occup Med Environ Health*, **14**: 99-113.
- Shields PG, Harris CC 2000. Cancer risk and low-penetrance susceptibility genes in gene-environment interactions. *J Clin Oncol*, **18**: 2309-2315.
- Sigsgaard T, Brandslund I, Omrand O, Hjort C, Lund ED, Pedersen OF, Miller MR 2000. S and Z alpha1-antitrypsin alleles are risk factors for bronchial hyperresponsiveness in young farmers: an example of gene/environment interaction. *Eur Respir J*, **16**: 50-55.
- Silverman EK 2001. Genetics of chronic obstructive pulmonary disease. *Novartis Found Symp*, **234**: 45-58.
- Strange RC, Spiteri MA, Ramachandran S, Fryer AA 2001. Glutathione-S-transferase family of enzymes. *Mutat Res*, **482**: 21-26.
- Walter R, Gottlieb DJ, O'Connor GT 2000. Environmental and genetic risk factors and gene-environment interactions in the pathogenesis of chronic obstructive lung disease. *Environ Health Perspect*, **108 (suppl 4)**: 733-742.
- Weiss ST 1999. Gene by environment interaction and asthma. *Clin Exp Allergy*, **29 (Suppl 2)**: 96-99.
- Wiesch DG, Meyers DA, Bleecker ER 1999. Genetics of asthma. *J Allergy Clin Immunol*, **104**: 895-901.
- Williams JA 2001. Single nucleotide polymorphisms, metabolic activation and environmental carcinogenesis: why molecular epidemiologists should think about enzyme expression. *Carcinogenesis*, **22**: 209-214.
- Xu J, Meyers DA, Ober C, Blumenthal MN, Mellen B, Barnes KC, King RA, Lester LA, Howard TD, Solway J, Langefeld CD, Beaty TH, Rich SS, Bleecker ER, Cox NJ 2001. Genomewide screen and identification of gene-gene interactions for asthma-susceptibility loci in three U.S. populations: collaborative study on the genetics of asthma. *Am J Hum Genet*, **68**: 1437-1446.