Clinical approaches toward asthma and COPD based on the heterogeneity of disease pathogenesis

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Abstract

Asthma and chronic obstructive pulmonary disease (COPD) are each heterogeneous disease classifications that include several clinical and pathophysiological phenotypes. This heterogeneity complicates characterization of each disease and, in some cases, hinders the selection of appropriate treatment. Therefore, in recent years, emphasis has been placed on improving our understanding of the various phenotypes of asthma and of COPD and identifying biomarkers for each phenotype. Likewise, the concept of the endotype has been gaining acceptance; an endotype is a disease subtype that is defined by unique or distinctive functional or pathophysiological mechanisms. Endotypes of asthma or COPD may be primarily characterised by increased susceptibility to type-2 inflammation, increased susceptibility to viral infections, bacterial colonization or impaired lung development. The "Dutch hypothesis" is as follows: gene variants underlying particular endotypes interact with detrimental environmental stimuli (e.g., smoking, viral infection, and air pollution) and contribute to the ultimate development of asthma, COPD, or both. Novel approaches that involve multidimensional assessment should facilitate identification and management of the components that generate this heterogeneity. Ultimately, patients with chronic inflammatory lung diseases may be treated based on these endotypes as determined by the respective biomarkers that correspond to individual endotypes instead of on disease labels such as asthma, COPD, or even asthma-COPD overlap syndrome (ACOS).

Introduction

Asthma and chronic obstructive pulmonary disease (COPD) are each heterogeneous disease classifications that involve chronic inflammation of the lower respiratory tract. Generally, the main features and pathophysiology differ substantially between the two, which allows clinicians to differentiate each entity and provide appropriate treatment. However, each disease is increasingly recognized as a complex of multiple phenotypes, and each phenotype has a somewhat unique natural history, severity, and treatment response [1]. This heterogeneity makes optimizing treatment a challenge, especially for patients that respond poorly to current therapies. The overlap between asthma and COPD is also increasingly evident; asthma-COPD overlap syndrome (ACOS) increasingly makes up a larger percentage of obstructive lung disease diagnoses and is associated with a higher overall health-care burden [2]. In 1961, Orie et al. [3] proposed that asthma and COPD are different expressions of one basic pulmonary disease whose expression in individuals presents differently depending on a combination of endogenous and exogenous factors (the Dutch hypothesis). They suggested that clinicians carefully phenotype patients with obstructive airway diseases so as to determine the heterogeneity of both asthma and COPD to better define these diseases and optimize treatments. Although there remains an ongoing debate regarding the relative merits of the "Dutch hypothesis", which again proposes that asthma and COPD are different expressions of one basic pulmonary disease" and the "British hypothesis", which argues they are distinct entities generated by different mechanisms [4], treatment strategies that target the particular causes underlying asthma or COPD will be more successful than the current strategies that merely decrease airway inflammation and airway obstruction without eradicating the underlying drivers of specific disease processes.

Recent genetic studies including genome-wide association studies (GWASs) have successfully identified many genetic loci responsible for susceptibility to asthma, COPD, or both. However, these loci explain only a small proportion of the heritability of these diseases because the phenotypic heterogeneity of asthma and COPD greatly complicates genetic analysis. A specific phenotype is likely to be more closely related to a specific pathogenetic mechanism, and focusing on a particular phenotype may increase the power of genetic studies and consequently lead to a better understanding of an endotype defined by a particular combination of phenotypes. In fact, larger GWASs are identifying individual genes that are more strongly linked to some asthma phenotypes than to others [5]. For example, we recently found strong evidence that hyaluronan synthase 2 (HAS2) is associated with asthma, specifically among non-smoking asthmatic patients or those with a limited smoking history of < 10 pack years [6]. Therefore, the genetic predisposition to the dysregulation of particular pathways may further help to define subgroups of asthma and COPD. In the end, this approach may lead to diagnosis for patients based on, in part, their genetic make-up, and to new therapeutic prospects.

Common pathways underlying asthma and COPD

One approach to exploring the Dutch hypothesis is to identify genes and mechanisms common to both asthma and COPD. Genetic studies indicate that common molecular pathways seem to underlie the pathogenesis of both asthma and COPD [7]. We previously searched PubMed for all asthma susceptibility genes and all COPD susceptibility genes to identify overlaps between these gene sets. Particular genes and molecular pathways show up at intersections of asthma and COPD gene sets [8]. We again briefly searched the PubMed database up to September 2012 to identify genes found to be associated with asthma, COPD, tuberculosis, or essential hypertension in at least two independent reports of candidate-gene associations or in GWASs. To learn how the identified genes interact with each other and other cellular proteins, we conducted pathway-based analysis using Ingenuity Pathway Analysis software. We identified 108 genes associated with asthma and 58 associated with COPD. We then grouped these susceptibility genes into networks based on functional annotation and identified 12 networks for asthma and 11 for COPD. Analysis of the networks for overlap between the two diseases revealed that a single complex network comprising 229 overlapping molecules could arise from these 23 networks. These overlapping molecules are significantly involved in canonical pathways including the "aryl hydrocarbon receptor signaling," "role of cytokines in mediating communication between immune cells," "glucocorticoid receptor signaling," and "IL-12 signaling and production in macrophages" pathways. The Jaccard similarity index for a comparison between asthma and COPD was 0.81 at the network-level comparison, and the odds ratio (OR) was 3.62 ($P < 0.0001$) for the asthma/COPD pair in comparison with the tuberculosis/essential hypertension pair. The Jaccard similarity index is a measure of the degree of association between two diseases; this index is determined based on the number of molecules in common between the diseases divided by the number of molecules unique to one or the other of the diseases. Accordingly, to calculate this index for asthma and COPD, of the 509 molecules in the 23 networks, the 229 molecules common to both was divided by the 280 molecules unique to one or the other, 190 and 90 molecules were unique to asthma and to COPD, respectively. Therefore, a particular network of shared genetic factors may lead to asthma when combined with specific environmental factors that are met at early in life, or conversely, this same network may lead to COPD when combined with different environmental factors early in life or with similar environmental factors later in life.

In recent years, the concept of the endotype has become increasingly important in the study of asthma and COPD. A phenotype is defined as a group of similar clinically observable characteristics that lack a documented and direct etiologic relationship with a distinct pathophysiologic mechanism. In contrast, the term endotype is used to describe “a subtype of a disease defined by a unique or distinctive functional or pathophysiological mechanism” [9]. Hypothetically, variation in environment influences on individual genotypes can lead to heterogeneous endotypes of asthma and COPD. Several clinical phenotypes can overlap in one patient and the same clinical phenotype could result from different endotypes. Therefore,

future treatment options are likely to target individual endotypes that are each defined by a distinct pathophysiological mechanism such as type-2 inflammation, viral infection, bacterial colonization or dysregulated lung growth (Table 1).

An endotype characterised by type-2 inflammation

Based on *in vitro* studies, Woodruff et al found three differentially expressed genes (*CLCA1*, *SERPINB2,* and *POSTN*) that are regulated by the type-2 cytokine IL-13. When patients are clustered on the basis of this signature type-2 expression, only 50% of the asthma patients cluster with the high type-2 phenotype, and healthy, non-atopic controls cluster with the low-type-2 asthma phenotype. Importantly, high type-2 asthma was associated with consistent clinical and inflammatory characteristics including increased blood and airway eosinophilia, airway hyper-responsiveness, a sub-epithelial basement membrane (SBM) thickening, high IgE levels, and expression of IL-5 and IL-13 in tissues [10]. Importantly, a clear association was found between type-2 phenotype and response to inhaled corticosteroids (ICS); the high type-2 phenotype was associated with robust FEV_1 improvement with moderate dose of ICS, while the low-type-2 phenotype was associated with no improvement at this ICS dose.

Another study by Christenson et al identified a genomic signature for type-2 inflammation in COPD [11]. They looked at airway-derived gene expression in an asthma cohort and two COPD cohorts, and found significant overlap between asthma and COPD with regard to gene expression specific for type-2 inflammation. For patients with COPD, a higher score of the genomic signature for type-2 inflammation score was associated with decreased lung function, increased airway wall eosinophil counts, higher blood eosinophil percentages, and responsiveness to both short-acting bronchodilator and ICSs. These findings are compatible with the Dutch hypothesis because the phenotypic feature of type-2 driven eosinophilic inflammation predicts favorable ICS treatment responsiveness both in patients with asthma and those with COPD.

Although a specific age cut-off for early-onset asthma has not been established, for most adults with asthma that originates in early childhood, the condition has an allergic component, and most people with asthma are likely to have this type-2 high phenotype. Indeed, the diversity of endotypes increases as age of onset increases, and late-onset of the disease is actually an important factor that blurs the border between asthma and COPD. I have recently proposed one plausible approach to using ICSs and LABAs/LAMAs in clinical practice for COPD patients that is based on both the extent of airflow obstruction and the presence of an asthma component or airway eosinophilic inflammation in the COPD presentation; this approach is a tentative move toward personalized treatment [12]. Sputum- or blood-eosinophil counts [13-16], FeNO [17-19], periostin [20-22] or IgE responsiveness [23] might identify two subpopulations, one in which ICSs could have potentially deleterious effects and another in which ICSs could be beneficial. Although

further clinical studies are needed for all these candidate biomarkers, it is likely that several of these, alone or in combination, will be useful in identifying patients with asthma or COPD who will benefit from type-2-targeted therapies. Given the high predictive value and ease of measurement of blood eosinophils, it is likely that these measurements will serve as an initial biomarker to predict response to ICSs and responses to biologic agents that target IL-4, 5, and 13. More clinical evidence and research will eventually identify panels of biomarkers that best predict responses to these biologic therapies.

An endotype characterised by increased susceptibility to viral infection

Respiratory viral infection is a common feature of some major human airway diseases, such as asthma and COPD. Exposure to respiratory viruses in early life is very common, and such exposure constitutes an independent risk factor for lung function abnormalities in adulthood, especially when the consequence of the childhood infection is severe acute lower respiratory tract infection (LRTI) [24]. Human rhinovirus (HRV) prevalence and load increase during COPD exacerbation, and patients with frequent exacerbations are more likely to experience HRV infection [25]. The first GWAS for asthma identified a novel asthma-associated locus on chromosome 17q12-q21 that encompassed two genes, *ORMDL3* and *GSDMB* [5]. Variations at this locus are associated with an approximately 2-fold increased risk of recurrent wheezing, asthma, asthma exacerbations, and bronchial hyperresponsiveness from early infancy to school age, but not with increased risk of eczema, rhinitis, or allergic sensitization [26]. Furthermore, the 17q21 locus is associated with asthma in children who had had HRV wheezing illnesses and with expression of two genes at this locus, suggesting a role of 17q21 variants in the development of HRV wheezing illnesses during early childhood as an underlying mechanism conferring susceptibility to early-onset asthma [27].

Another GWAS of a specific asthma phenotype characterized by recurrent, severe exacerbations occurring between 2 and 6 years of age provided strong evidence for a newly recognized susceptibility gene, *CDHR3*, which encodes cadherin-related family member 3 and is highly expressed in airway epithelium [28]. Compared with wild-type CDHR3, the CDHR3 (Cys529→Tyr, rs6967330) variant, which was linked to wheezing illnesses and hospitalizations for childhood asthma based on genetic analysis, exhibited approximately 10-fold increases in HRV-C binding and HRV progeny yields when transfected into cells *in vitro*; these findings indicate that the CDHR3 variant that is associated with asthma susceptibility facilitates HRV-C entry into host cells, and that the variant could be a risk factor for HRV-C wheezing illnesses [29].

We published results of a sub-analysis of a large Japanese GWAS [30] that had identified five asthma-associated loci at a genome-wide significant threshold; the entire data set was from 7171 adult individuals with asthma and 27,912 control subjects. In our sub-analysis [6], to identify more asthma-associated loci, only 240 asthma patients and 734 healthy controls, all recruited from a single geographical region were

included; additionally, only non-smoking asthmatic patients or those with a limited smoking history of less than 10 pack years were included. These inclusion criteria may have reduced the number of participants who had asthma-like symptoms due to environmental factors such as smoking, and thereby increased our ability to identify asthma-specific genetic effects. Using this approach, evidence that *hyaluronan synthase 2* (*HAS2*) is associated with asthma emerged. This gene encodes a glucosaminoglycan that is present in the extracellular matrix and is strongly expressed in the lungs. Furthermore, an asthma-associated SNP was shown to affect regulation of *HAS2* mRNA expression. Hyaluronan plays an essential role in many physiological and pathological processes, including cell migration, morphogenesis, tissue regeneration, wound repair, and tumor cell growth and invasion. The airway inflammation associated with asthma involves the accumulation of a hyaluronan-rich matrix following viral infection; the degraded hyaluronan matrix is removed by the leukocytes/macrophages that enter the tissue. Notably, the degraded hyaluronan matrix acts as a "danger signal" responsible for initiating host responses to the inflammatory process and likely participates in determining the extent of the response to viral infections [31].

Children who have disorders of the immune system often experience severe infections. The two most common immune disorders to consider among these young children are deficiencies in IgA or mannose binding lectin (MBL). In fact, asthmatic patients are more likely to have a diagnosis of selective IgA deficiency (sIgAD)/common variable immunodeficiency (CVID) than nonasthmatic individuals [32]. In addition, the T allele at *MBL2* rs1800450 is associated with lower serum MBL levels, and is a risk factor of developing wheezing [33].

Given that early-life factors, including viral infections, increase the risk of COPD [34], these genetic findings may indicate the existence of an endotype characterised by increased susceptibility to viral infections that underlies some phenotypes of asthma and COPD.

An endotype characterised by bacterial colonization in airways

The lower airways are all colonized with airway microbiota and the airway microbiomes in patients with asthma or COPD differ from those in healthy control subjects [35]. Although it is unknown whether the differences are the cause or consequence of the respective diseases or treatments [36], it is likely that disease-related bacterial colonization impacts negatively on clinical course by increasing the symptoms (chronic cough and expectoration), accelerating the rate of FEV_1 decline, promoting the occurrence of exacerbations, or some combination thereof [37, 38].

Ghebre et al. [39] performed cluster and factor analysis on sputum inflammatory mediators that generated clinically meaningful groupings within their sample of patients with asthma or COPD. An asthma-predominant group with eosinophilic inflammation and increased type-2 inflammatory mediators was

distinct from a COPD-predominant group with mixed eosinophilic and neutrophilic cells and increased proinflammatory cytokine levels. A third mixed group comprised asthma and COPD diagnoses and was associated with clinically chronic bronchitis, neutrophilic predominance, bacterial colonization, and increased levels of IL-1β and TNF-α. Furthermore, one-month-old neonates with bacterial colonization in the hypopharyngeal region (*Streptococcus pneumoniae*, *Haemophilus influenzae*, *Moraxella catarrhalis*, or a combination) have an increased risk for recurrent wheezing and asthma early in life, and this increased risk is independent of atopy [40]. These findings indicate that bacterial colonization has a role in some disease phenotypes, especially those phenotypes with neutrophilic inflammation. Importantly, these patients are consistently found to be largely refractory to ICS treatment, and thus have fewer treatment options than other groups [41].

Previously, we reported that the genes encoding CCL5/RANTES, tissue factor (TF), Clara cell secretory protein (CC16), or catalase (CAT) are associated with adult/late-onset asthma in a Japanese population [42-45]. Assuming that differentiating asthma on the basis of age at disease onset increases the power of genetic studies and will enhance our understanding of asthma pathogenesis, we performed a GWAS focusing on late-onset asthma (age of onset ≥ 40 years) in a 2-stage genetic association study of 4933 Japanese adults [46]. A meta-analysis combining data from three studies, two discovery and one replication, showed increased frequencies of the C alleles at both rs2523870 and rs2517548 in the late-onset asthma group (meta-P = 3.77×10^{-7} and 3.98×10^{-7}, respectively). These SNPs are close to each other; both are located between *HLA complex group 22* (*HCG22*) and *mucin 22* (*MUC22*) and are in tight linkage disequilibrium. SNP-gene associations for these two SNPs obtained by expression quantitative trait loci (eQTL) analysis indicated that *HCG22* mRNA expression in lymphoblastoid cell lines is strongly correlated with the number of C alleles, the asthma susceptibility alleles at either rs2523870 or rs2517548 ($P = 8.54 \times 10^{-87}$ or 3.23×10^{-84}, respectively). When we plotted the OR for association between rs2523870 and age-of-onset specific subsets of asthma, the OR gradually increased as the age-of-onset cut-off increased up to the 50 or older category (Fig 1); this finding clearly indicates that the genetic contribution of rs2523870 increases as the age at onset of asthma increased.

Interestingly, *HCG22* was originally identified as a candidate gene for diffuse panbronchiolitis (DPB) in the MHC class I region on chromosome 6p21.3 [47]. DPB is an idiopathic inflammatory disease characterized by chronic neutrophilic bronchiolitis and rhinosinusitis that cause mucus hypersecretion and airflow obstruction. The average age of onset of DPB is around 40 years, and two-thirds of those affected are non-smokers. HCG22 has characteristics similar to those of mucin, with tandem repeats consisting of serine and threonine-rich peptides. *HCG22*, a novel mucin-like gene, forms a mucin-like gene cluster together with *MUC22*, *MUC21*, and *DPCR1*. Expression of *HCG22* is increased in lung tissues.

When we investigated the genetic impact of rs2523870 on the development of DPB (N = 108) or COPD (N = 307) using 4044 healthy adults as controls, the C allele at rs2523870 (the risk allele for late-onset asthma) was also associated with DPB ($P = 3.06 \times 10^{-4}$; OR = 1.65) and with COPD ($P = 6.14 \times 10^{-3}$; OR = 1.28). The genetic associations found in patients with DPB or COPD and those with late-onset asthma along with the finding that both rs2523870 and rs2517548 are highly correlated with increased expression of *HCG22* strongly indicate that these SNPs confer increased risk of late-onset asthma and suggest that a common pathogenetic mechanism or a specific endotype underlies late-onset asthma, DPB, and COPD.

Mucus hypersecretion is a common feature of chronic airway diseases such as asthma, COPD, and DPB. Accumulation and activation of airway neutrophils are also important in these diseases, and the activated neutrophils recruited to the airways play several key roles in mucus hypersecretion. Impaired mucosal immunity associated with increased susceptibility to infection could underlie some of the distinct asthma and COPD phenotypes. Long-term macrolide therapy is highly effective in the treatment of DPB [48]. In addition, macrolide antibiotics have been demonstrated to be efficacious in the treatment of exacerbation-prone COPD and of severe neutrophilic asthma [49-51]. Macrolides such as erythromycin, clarithromycin, and azithromycin not only have antimicrobial properties, but also broad anti-inflammatory and immunomodulatory effects [52]. Nevertheless, chronic use of macrolides is associated with the occurrence of macrolide-resistant bacteria in the commensal flora of the pharynx of individual patients and also induces the risk of an increase in antibiotic resistance at the population level [53]. An electronic nose, which is a new non-invasive technology capable of distinguishing volatile organic compound (VOC) breath-prints in exhaled breath, can identify the presence of airway bacterial colonization in clinically stable patients with COPD [54]. Further research will determine the usefulness of breath-prints for diagnosing an endotype that may be particularly responsive to macrolide therapy.

An endotype characterised by reduced pulmonary function

David Barker proposed an original hypothesis stating that fetal development is an important influence on the development of adult disease [55], and this hypothesis is relevant to asthma and COPD [56]. Adverse factors affecting lung development during fetal life and early childhood reduce the attainment of maximum lung function and accelerate declines in lung function during adulthood; therefore, these adverse factors predispose individuals to reduced lung function and increased respiratory morbidity, particularly asthma and COPD, throughout life [49, 57, 58]. *TSLP* is a likely asthma-susceptibility locus [59], and we found that *TSLP* variants are associated with lower lung function in healthy individuals [60], which is consistent with the contention that genetic determinants of lung function influence susceptibility to asthma.

To date, GWASs on pulmonary function (e.g., predicted percentage of forced expiratory volume in 1

second (%FEV_1) and ratio of forced expiratory volume in 1 second to forced vital capacity (FEV_1/FVC)) have identified a number of risk loci in multiethnic populations [61-66]. Although our previous GWAS did not identify SNPs associated with pulmonary function at the level of genome-wide significance in a Japanese population, it does demonstrate that the heritability of pulmonary function can be explained by additive effects of multiple common SNPs, providing compelling evidence for a strong genetic influence on FEV_1/FVC [67]. Furthermore, our findings indicate that lung-function genes identified in previous GWASs in non-Japanese populations account for 4.3% to 12.0% of the entire estimated heritability of FEV_1/FVC in a Japanese population. Therefore, we constructed a multi-SNP genetic risk score (GRS) for reduced FEV_1/FVC using genotype information for 16 genes associated with lower FEV_1/FVC in a GWAS of Japanese populations as well as in previous GWASs of non-Japanese populations [68]. The GRS, which combines the modest effects of multiple SNPs into a single variable, is calculated as the weighted sum of the number of high-risk alleles. Both reduced growth and accelerated lung function decline lead to lower lung function levels in adults, but the GRS calculated using the 16 SNPs is not associated with annual lung function decline in the healthy participants; therefore, we believe that these 16 SNPs (or gene pathways involving these 16 SNPs) may be involved in deregulated lung growth or development rather than in the accelerated decline of lung function.

This GRS for lower FEV_1/FVC was consistently associated with the onset of asthma ($P = 9.6 \times 10^{-4}$) in two independent Japanese populations as well as with the onset of COPD ($P = 0.042$). The Cochran-Armitage trend test shows that the prevalence of asthma increases as GRS values increase ($P = 0.0059$) (Fig. 2). In contrast, the prevalence of atopic status does not change according to GRS values ($P = 0.46$), indicating that the association between the GRS and the presence of asthma is independent of atopic status. Clustering of asthma patients based on GRS values indicates that an increased GRS may be responsible for the development of a particular phenotype of asthma, a phenotype that is characterized by early onset, atopy, and more severe airflow obstruction. In a multinomial logistic regression analysis, the strongest association between GRS values and asthma is found in this particular cluster ($P = 3.6\times10^{-5}$). In fact, a similar cluster that is characterized by early-onset, atopic asthma with advanced airflow limitation has also been identified [69, 70].

Overall, these data further highlight the potential application of genomics in developing novel strategies to precision medicine that may improve long-term respiratory outcomes for children with lower lung function. Such strategies may enable earlier identification of at-risk infants and of the specific pathways involved with disease pathogenesis in individual cases, which will allow for earlier and more specific interventions to achieve greater respiratory health after altered lung development in early childhood.

Relationships of endotypes to gender, environmental factors and epigenetics

Accumulating epidemiologic data demonstrate sex differences with respect to prevalence and progression of airway diseases, including asthma and COPD. A wide range of factors have been examined to explain the sex differences noted in asthma and COPD, including hormonal influences, genetic predisposition, anatomic differences, differences in immune response, environmental factors, and comorbidities [71]. *TSLP* variants that play an important role in type-2 inflammation endotype are associated with asthma in a sex-specific fashion [72]. In addition, a significant difference in DNA methylation between males and females is evident in the 17q12-q21-gene promoter region, which underlies the endotype of increased susceptibility to virus infection. This finding suggests that sex-dependent DNA methylation can serve as a modifier of predisposition for asthma [73]. Accordingly, the endotypes described here are probably significantly influenced by sex difference.

The progression from early life insults to pediatric disease and finally chronic obstructive airway disease in adulthood involves complex genetic, epigenetic, and environmental interactions. In terms of the endotype characterised by impaired lung function, genetic variants that were identified in adults to be associated with lung function were not associated with neonatal lung function, but they were associated with the development of lung function measures during early childhood [74]. These findings suggest that a window of opportunity exists for interventions that target these genetic mechanisms. In fact, long-term improvements in air quality are associated with statistically and clinically significant positive effects on lung-function growth in children [75]. Measurement of lung function in an early life, therefore, may help us in preventing reduced lung function and in maintaining respiratory health among children.

With regard to the endotype characterised by bacterial colonisation, recent insights into the airway microbiome suggest potential effects of aberrant airway microbiomes in patients with asthma or COPD on the pathobiology of their disease. Children living on farms are exposed to a wider range of microbes than children not living on farms, and this exposure explains a substantial fraction of the inverse relation between asthma and growing up on a farm [76]. These findings indicate that the diversity of an environmental microbiome can influence lung health and disease. Detection of bacterial colonization may allow an early intervention by exposing susceptible individuals to a wide range of microbes, which may help ameliorate or prevent chronic inflammatory lung diseases in later life.

Immunoglobulin E (IgE) is a central mediator of type-2 inflammation. A genome-wide survey of epigenetic associations between serum IgE concentrations and methylation at loci concentrated in CpG islands in 95 nuclear pedigrees found that the top three loci accounted for 13% of IgE variation [77], explaining the 10-fold higher variance found compared with that derived from a large SNP GWAS [5]. This study, by identifying novel therapeutic targets and biomarkers for patient stratification in allergic diseases, highlights the potential importance of epigenetic responses to environmental factors that underlie

the type-2 inflammation endotype. Another study examined the genome-wide epigenetic response of airway epithelial cells (AECs) to IL-13 and showed that a single exposure of IL-13 selectively induce long-lasting DNA methylation changes in asthmatic airways that alter specific AEC pathways and contribute to asthma phenotypes/endotypes [78].

An endotype-oriented approach to asthma and COPD

Patients with asthma, COPD, or ACOS represent a heterogeneous group in terms of pathogenesis, clinical presentation, disease course, and prognosis; this heterogeneity indicates a wide range of disease mechanisms. Careful phenotypic characterisation of patient subpopulations is therefore required to make improvements in the treatment of these heterogeneous diseases. Clusters of phenotypes are likely to encompass specific endotypes, subgroups of these diseases each with distinct molecular mechanisms. Here, I have discussed what appear to be four important endotypes that seem to underlie susceptibility to asthma and COPD; each endotype is currently defined by one characteristic: susceptibilities to type-2 inflammation, susceptibility to viral infections, bacterial colonization, or impaired lung development.

So far, type-2 eosinophilic inflammation is the most studied endotype because most novel asthma treatments target the type-2 pathway. A concerted effort is now needed to sub-stratify asthma and COPD beyond the stratifications based on type-2 pathways and to use appropriate biomarkers that can identify patients likely to respond to specific and respective biologics. The application of modern technologies to the study of genomic alterations associated with viral infection, bacterial colonization, or lung growth may facilitate targeted development of new treatment options for patients with specific molecular abnormalities. In reality, asthma and COPD are not only heterogeneous diseases but also associated with complex medical conditions; therefore, different molecular characteristics associated with different endotypes may occur in varying proportions in any given patient.

Even at the present time, multidimensional information (such as sputum and/or peripheral blood eosinophil numbers, FeNO, serum IgE, allergen-specific IgE antibodies, airflow obstruction, previous exacerbation, age at onset, emphysema, pulmonary hypertension, and bacterial colonization) can help to provide clinicians with the actionable information necessary for true personalized treatment of a given patient; nevertheless, the specificity and efficacy of treatments for individuals can improve with a better understanding of endotypes and endotype-specific disease mechanisms.

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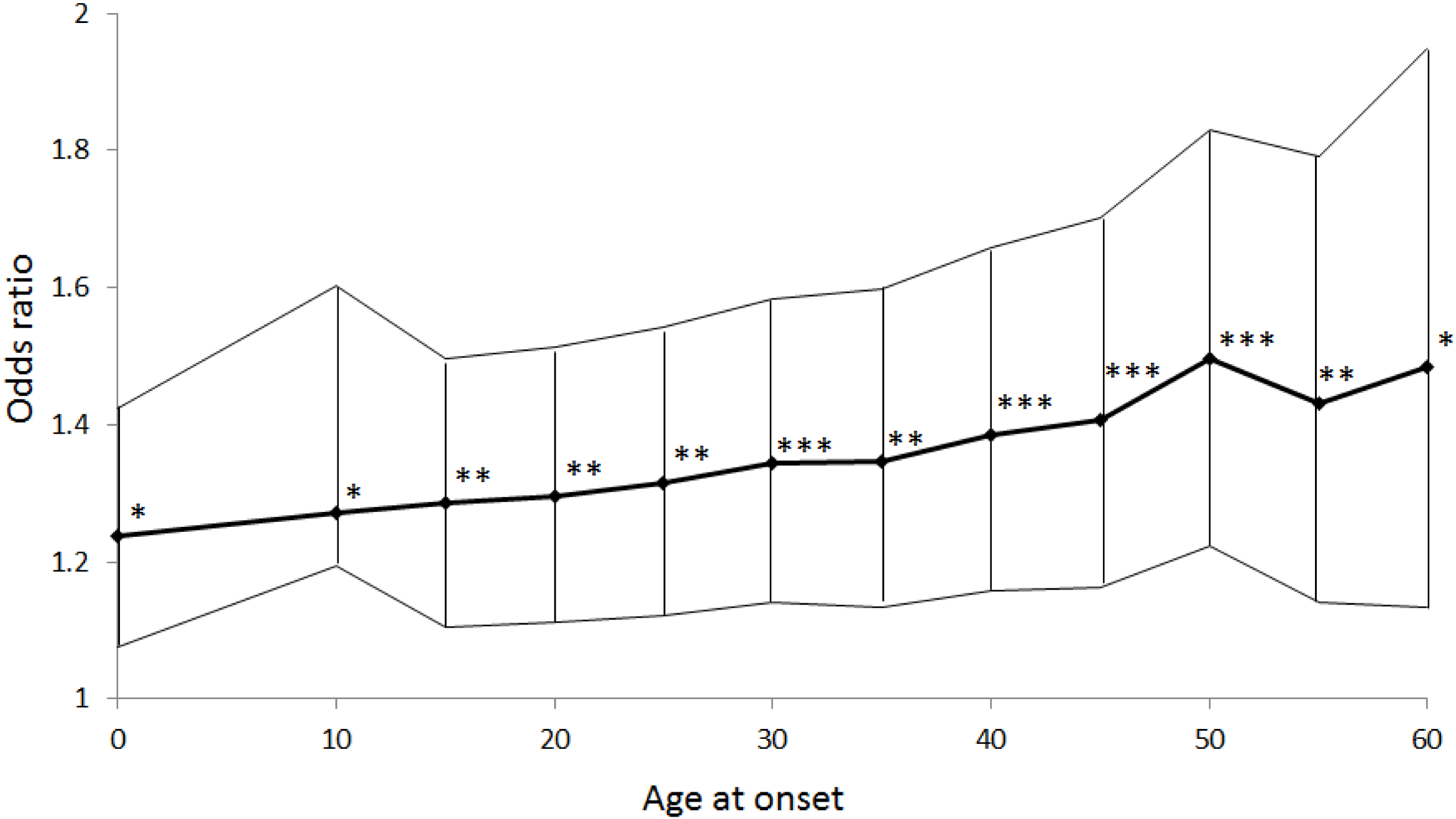
Table 1 Approaches to asthma and COPD based on endotypes

Endotype	Genotype	Biomarker or clinical features
Type-2 inflammation	Type-2 related genes (IL13, TSLP, IL33)	Eosinophils (blood, sputum), FeNO, IgE, periostin
Increased susceptibility to viral infections	ORMDL3/GSDMB(17q21), CDHR3, HAS2 MBL2	Repeated respiratory infections at an early stages of life, Serum IgA
Impaired lung development	Lung function genes (FAM13A, NCR3, TGFB2)	Impaired pulmonary function at an early age
Bacterial colonization	HCG22	Sputum culture, VOC

VOC; volatile organic compound


Odds ratio
2
1.8
1.6
1.4
1.2
1
0
10
20
30
40
50
60
Age at onset
*
*
**
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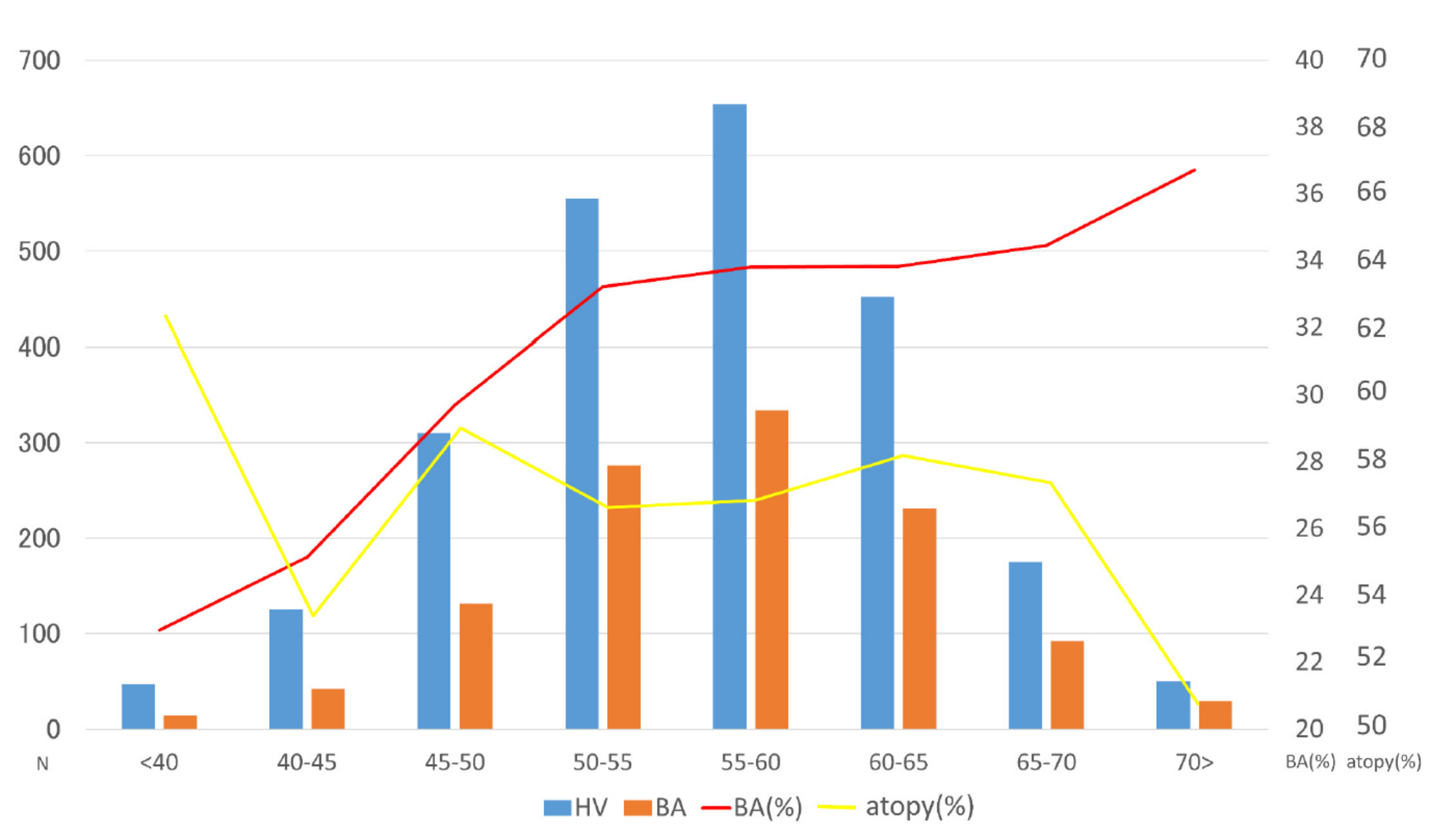
700
600
500
400
300
200
100
0
40
38
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34
32
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28
26
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68
66
64
62
60
58
56
54
52
50
N
<40
40-45
45-50
50-55
55-60
60-65
65-70
70>
BA(%)
atopy(%)
HV
BA
BA(%)
atopy(%)

Figure legends

Figure 1

Odds ratios for associations between *HCG22* and onset age-specific subsets of asthma

The OR gradually increases as the cut-off for the age-of-onset increases until the onset age reached 50 years or later. ORs significantly greater than 1 are highlighted (*P < 0.005, **P < 0.001, and ***P < 0.0005). Ref. 46

Figure 2

Prevalence of asthma according to genetic risk score values for lower lung function

The horizontal-axis shows the GRS ranges. The left vertical-axis shows the number of healthy individuals and asthmatic patients for each GRS range. The right vertical-axis shows the percentages of asthmatic patients and atopic individuals for each GRS range. The upper line shows the percentage of atopic individuals for each given GRS range. The lower line shows the percentage of asthmatic patients for each given GRS range. Atopy was defined as the presence of specific IgE antibody toward at least 1 common inhaled allergen. HV, healthy volunteer, BA, bronchial asthma. Ref. 68