

Review

Interactions between genes and environmental factors in asthma and atopy: new developments

Claudia Sengler, Susanne Lau, Ulrich Wahn and Renate Nickel

Department of Pediatric Pneumology and Immunology, Charité, Humboldt University Berlin, Berlin, Germany

Correspondence: Renate Nickel, Department of Pediatric Pneumology and Immunology, Charité, Humboldt University, Augustenburger Platz 1, D-13353 Berlin, Germany. Tel: +49 30 4505 66131; fax: +49 30 4505 66931; e-mail: renate.nickel@charite.de

Received: 10 April 2001

Respir Res 2002, **3**:7

Revisions requested: 7 June 2001

Revisions received: 10 July 2001

Accepted: 27 July 2001

© 2002 BioMed Central Ltd

Published: 31 October 2001

(Print ISSN 1465-9921; Online ISSN 1465-993X)

Abstract

Asthma and associated phenotypes are complex traits most probably caused by an interaction of multiple disease susceptibility genes and environmental factors. Major achievements have occurred in identifying chromosomal regions and polymorphisms in candidate genes linked to or associated with asthma, atopic dermatitis, IgE levels and response to asthma therapy. The aims of this review are to explain the methodology of genetic studies of multifactorial diseases, to summarize chromosomal regions and polymorphisms in candidate genes linked to or associated with asthma and associated traits, to list genetic alterations that may alter response to asthma therapy, and to outline genetic factors that may render individuals more susceptible to asthma and atopy due to environmental changes.

Keywords: asthma, atopy, environment, genetics, infection

Introduction

The prevalence of asthma, allergic rhinitis and atopic dermatitis (AD) has dramatically increased over the past decades. These atopy-related diseases are the most common chronic disorders in childhood in Western societies. Several epidemiological studies have evaluated environmental risk factors that may explain the steady increase of allergic disease. (Note: in this article, allergic diseases/atopic disorders include asthma, allergic rhinitis and AD. Atopy-associated phenotypes/traits include, in addition, allergic sensitization, elevated total serum IgE and eosinophilia.) There is growing evidence that contact to bacterial antigens (such as endotoxin) and viral infections in early childhood is protective with regard to development of allergic disease in later life [1–6]. Although changes in lifestyle significantly contribute to disease expression, heritability has been shown to play a major role in the pathogenesis of allergic disease.

Multiple twin and family analyses strongly imply a genetic basis for atopy-related traits (for a review, see [7]). A recent study of 11,688 Danish twin pairs (comparing identical and non-identical twin pairs) suggested that 73% of asthma susceptibility is due to genetic factors [8]. However, atopy-associated phenotypes, including asthma, do not appear to follow any Mendelian inheritance pattern, which is characteristic for complex genetic (multifactorial) traits. The dissection of these traits is hampered by phenocopy, incomplete penetrance, and genetic heterogeneity [9]. The complexity of the genetics of asthma and other atopy-associated phenotypes is reflected by an increasingly large number of chromosomal regions showing (weak to moderate as defined in [9]) evidence for linkage, as well as various genetic variations in multiple candidate genes that are associated with asthma and associated phenotypes.

β_2 AR = beta-2 adrenergic receptor; AD = atopic dermatitis; IL = interleukin; 5-LO = 5-lipoxygenase; SNP = single nucleotide polymorphism; TDT = transmission disequilibrium test.

Methodology in genetic studies of complex traits

Microsatellite marker analysis

The majority of studies on genetics of complex traits to date have been based on microsatellite marker (synonym, short tandem repeat polymorphisms [STRP]) analyses. These genetic markers (whose biological function is as yet unknown) typically contain a variable number of tandem repeats of dinucleotide, trinucleotide or tetranucleotide DNA sequences (e.g. the tetranucleotide [TATA]_n). The high degree of polymorphism results in great variation between individuals. Short tandem repeat polymorphisms of known location are found densely spaced throughout the genome and are used for genome-wide searches as well as for the analysis of candidate gene regions.

Study designs in analyses of complex genetic traits

Most studies on the genetics of asthma are based on allele sharing methods, transmission disequilibrium test (TDT) analysis, or tests for associations. The allele sharing methods approach involves testing how often a genetic marker (or a chromosomal region) is shared by affected pedigree members. If allele sharing occurs significantly more often than expected by chance, linkage of the particular marker and disease can be assumed, indicating that the chromosomal region containing the genetic marker also contains a gene that contributes to disease expression.

The TDT approach is based on genotype analysis of affected subjects (no siblings are required) and their parents. The TDT tests whether genetic marker alleles from heterozygous parents are transmitted as frequently as expected by chance (by random, each parental allele is transmitted with a chance of 50%). Overtransmission of a particular marker allele indicates linkage of this allele with the respective 'disease' allele.

Association studies are usually applied once polymorphisms in candidate genes for asthma/atopy have been identified.

Linkage and candidate gene studies in asthma and atopy

Linkage studies

Results from linkage studies (genome-wide searches and candidate gene region analyses) are summarized in Supplementary Tables 1 and 2 (see also [10–13]). Multiple chromosomal regions were related to asthma and atopy. Few regions, however, have shown evidence for linkage in more than one population (Supplementary Table 2), which may be due to racial differences, to different definition of phenotypes, or (most probably) to insufficient numbers of affected sib-pairs in different study populations. The first successful attempts have been made to pool data from genetic studies of asthma and atopy in order to analyze major candidate gene regions [14,15].

Despite the high degree of inconsistent findings, it is intriguing that chromosomal regions linked to asthma have also shown evidence for linkage to other inflammatory and autoimmune diseases (such as psoriasis, inflammatory bowel disease, type I diabetes and multiple sclerosis). Becker *et al.* noted that susceptibility genes for various autoimmune and inflammatory diseases (including asthma) appear to cluster in 18 distinct chromosomal regions, implying common genetic elements in inflammatory disorders [16]. This speculation is further supported by two recent genome-wide searches for AD susceptibility genes. Regions linked to AD (Supplementary Table 1) have also been linked to psoriasis in other studies [17,18], suggesting that common genetic elements are involved in dermal inflammatory disorders.

Candidate gene studies

Multiple genes are involved in the allergic inflammation (shown in Supplementary Figure 1). Multiple single nucleotide polymorphisms (SNPs) in candidate genes for asthma and atopy have been identified, and they are presented in Supplementary Table 3. The (often conflicting) results have to be interpreted with caution, since multiple genotypes are commonly tested for a large number of atopy-related (sub)phenotypes (using various statistical approaches) in often considerably small sample sizes. Therefore, type 1 errors (false-positive results) are likely to occur. In addition, publication favors positive results and, therefore, many negative studies have not been published.

Functional studies

Many SNPs listed in Supplementary Table 3 have been tested for functional differences *in vitro*. However, when several (potentially) functional SNPs are expressed in one (regulatory region of a) gene (e.g. IL-4R α , IL-13, beta-2 adrenergic receptor [β_2 AR]), they are likely to interact. The individual analysis of SNPs may therefore not reflect *in vivo* gene function, as a recent study on β_2 AR haplotypes demonstrated (see ' β_2 -agonists' subsection). (Note: a haplotype is a particular combination of alleles in a defined region of a chromosome. It is used to describe combinations of polymorphisms in a gene/chromosomal area.) *In vitro* differences in gene expression or function due to SNPs therefore have to be interpreted with caution when several other polymorphisms within the same gene may also alter functionality *in vitro* and *in vivo*. This notion may explain some of the controversial findings with regard to associations of individual SNPs in polymorphic genes (e.g. IL-4R α).

The importance of haplotype analysis is further supported by the following studies. Seven SNPs in the IL-13 gene have been shown to be tightly linked [19], and therefore association studies of individual SNPs with the assumption of independent inheritance cannot easily be performed. Also, several SNPs within the IL-4R α gene have been iden-

tified (see Supplementary Table 3). An association of two individual SNPs (S503P and Q576R) with decreased serum IgE levels was observed in a Caucasian population; however, most significant results with regard to low IgE levels were seen when both SNPs occurred together [20].

Similarly, Ober *et al.* performed TDT analysis for IL-4R α SNPs and haplotypes in four populations. Again, the strongest evidence for linkage to asthma and/or atopy was observed with two-locus haplotype analysis [21].

Gene-gene interactions

Gene-gene interactions also have to be taken into consideration; for example, when both the genes encoding pro-inflammatory cytokines (i.e. IL-13) as well as their receptors (IL-4R α) and/or genes involved in signal transduction (STAT6) bear polymorphisms associated with disease. The first studies have demonstrated the feasibility and importance of gene-gene interaction in atopy [22].

Gene-environment interactions

Pharmacogenetics

A marked disparity in treatment response to pharmacotherapy is observed in asthma (as well as in many other diseases). Variations in genes encoding proteins that interact with specific drugs or drug metabolism are likely to contribute to variability in treatment response. The best examined gene with regard to pharmacogenetics is the gene encoding the enzyme cytochrome p450, for which various alterations that affect the metabolism of multiple drugs have been described. Gene chips for the determination of p450 alleles are commercially available in Sweden and North America (reviewed in [23]).

Pharmacotherapy of asthma and atopy comprises β -agonists, corticosteroids, anti-histamines (H1-receptor antagonists) and leukotriene (receptor) antagonists. The following pharmacogenetic studies have been performed.

β_2 -agonists

β_2 -agonists are the most widely used drugs in the treatment of asthma. They exert their primary effect on the β_2 AR of bronchial smooth muscle. The gene encoding the β_2 AR maps to chromosome 5q33 and is the best examined candidate gene with regard to pharmacogenetic studies in asthma. Several SNPs within the coding region and the promoter of the β_2 AR have been identified (summarized in [24]; see also Supplementary Table 3). Three mutations that result in amino acid changes of the mature protein (Gly16Arg, Gln27Glu, and Thr164Ile) lead to functional changes. The Gly16 variant was shown to undergo enhanced agonist-promoted downregulation *in vitro*. Interestingly, a strong association of the Gly16 polymorphism and nocturnal asthma could be demonstrated [25], which may explain the β_2 AR downregulation observed in nocturnal asthmatics but not in non-nocturnal asthmatics [26].

Conflicting results have been reported with regard to β_2 AR SNPs and response to β_2 -agonists (summarized in Supplementary Table 3). A large multicenter study demonstrated that patients homozygous for the Arg16 variant developed a decline in peak flow when using albuterol on a regular basis [27]. However, a recent haplotype analysis [24] indicated that β_2 AR SNPs are not independently transmitted and should presumably not be tested separately for associations with asthma or differences in drug response. Drysdale *et al.* demonstrated that only a limited number of β_2 AR haplotypes can be found in several ethnic groups, far less than theoretically possible. Furthermore, response to β_2 -agonists in asthmatic individuals significantly related to distinct haplotypes but not to individual SNPs. Consistent with the *in vivo* data, transfection of cells with the β_2 AR haplotype associated with a better response to β_2 -agonists resulted in significant greater mRNA levels and β_2 AR receptor density compared with the lower response haplotype [24].

Corticosteroids

Despite the large variability in the steroid response of asthmatic individuals, genetic variations in the glucocorticoid receptor gene have as yet to be identified in steroid-dependent asthmatics.

Anti-histamines

No functionally relevant polymorphism has been found in the H1-histamine receptor gene [23]. However, a functional mutation in the gene encoding the histamine-degrading enzyme *N*-methyltransferase has been related to asthma [28].

Leukotriene antagonists

Leukotrienes (LTC₄, LTD₄, LTE₄) contribute to airway inflammation and bronchoconstriction in asthmatic individuals. 5-lipoxygenase (5-LO) (gene symbol, ALOX5; chromosome, 10q11.2) is a crucial enzyme in the synthesis of leukotrienes. The activity of 5-LO determines, at least in part, the concentration of leukotrienes in the airways. Drazen *et al.* [29] described frequently occurring mutations in the 5-LO promoter (three to six tandem repeats of an Sp-1 binding site, with the wild-type allele having five copies) that result in a decreased transcriptional activity. In patients with asthma, only those individuals that expressed the wild-type 5-LO promoter responded well to therapy with a 5-LO inhibitor (ABT-761, the drug was never marketed), whereas individuals with both 5-LO promoter alleles mutated showed no significant improvement of lung function when treated. It has not yet been examined whether these mutations also affect the responses of asthma patients to leukotriene receptor antagonists (e.g. montelukast, zafirlukast).

Sanak *et al.* described a polymorphism within the leukotriene C4 synthase promoter that resulted in higher risk of

aspirin-induced asthma. This genetic variant may also alter response to treatment with drugs directed against leukotrienes [30].

In conclusion, the studies by Drazen *et al.* [29] and Drysdale *et al.* [24] in particular indicate that genetic testing may help to predict drug response and may eventually be used to optimize individual pharmacotherapy.

Genetics of host-defence

Apart from pharmacogenetic studies, the analysis of gene–environment interactions to date is hypothesis driven. The difficulties in quantifying and characterizing environmental risk factors for atopy (including onset and length of period of exposure) make these studies a challenge. Very high numbers of affected and unaffected subjects carefully characterized (longitudinally) for both the environmental setting and disease expression may be required to test for interactions between genetic variants and non-genetic influences.

However, recent genetic studies support the 'hygiene hypothesis', which postulates that atopy may be the result of a misdirected immune response in the absence of infection.

First, resistance to *Schistosoma mansoni* [31] and *Plasmodium falciparum* blood levels [32] (two independent studies) were linked to chromosome 5q31-33, a region (containing the IL-4 cytokine gene cluster) that has shown strong evidence for linkage to atopy-associated traits. Furthermore, a major locus closely linked to the interferon- γ receptor gene appears to control the switch from a T helper 2 to a T helper 1 cytokine profile during *S. mansoni* infection [33,34].

Second, SNPs within the Fc ϵ RI- β gene have been related to increased total serum IgE levels in heavily parasitized Australian aborigines, indicating a protective role in parasitic infection [35]. These SNPs have also been related to asthma, bronchial hyperresponsiveness, AD and atopy (see Supplementary Table 3). Fc ϵ RI- β maps to 11q13, a region that showed evidence for linkage to asthma and associated traits in multiple studies (Supplementary Table 3).

Finally, a polymorphism in the β_2 AR encoding gene (Arg16) that has been related to asthma (see Supplementary Table 3) was also associated with higher levels of parasitic infection [36].

Ethnic differences in 'host defense' and 'atopy' genes

Evolutionary pressure with regard to infectious agents has differed significantly between continents. Significant ethnic differences in genes involved in immune defense mechanisms have been reported and may be based on

differences in natural selection over the past centuries. Racial differences in genes involved in both host defense and allergic inflammation can best be demonstrated for CC chemokines and their receptors. CC chemokines have been shown to be crucial mediators of the allergic inflammation due to their potent chemoattractant properties for eosinophils, basophils and T cells [37]. Observations with regard to ethnic differences are as follows.

Evidence for linkage to chromosome 17q11.2 (a region that contains the CC chemokine gene cluster) has been reported in African Americans [38] but not in any Caucasian population (Supplementary Table 1).

A 32 base pair deletion in the CC chemokine receptor CCR5 renders individuals resistant to infection with macrophage-tropic HIV strains. This mutation is found in >10% of Caucasians, whereas it cannot be found in African populations [39]. A reduced risk of asthma was reported for carriers of the CCR5 deletion [40]. However, this could not be confirmed by Mitchell *et al.* [41].

In contrast to Caucasian individuals, the Duffy Antigen/Receptor for Chemokines is not expressed on red blood cells in the vast majority of African people (a point mutation in the Duffy promoter abolishes erythrocyte gene expression). This confers an evolutionary advantage since Duffy-negative erythrocytes are resistant to infection by *Plasmodium vivax*, which is endemic in most of Africa. Duffy has been shown to bind with high affinity to chemokines of both the CXC and CC classes, and is believed to function as a clearance receptor for chemokines (reviewed in [42]). It can therefore be hypothesized that higher or longer exposure to chemokines due to the absence of Duffy on erythrocytes might contribute to asthma pathogenesis in subjects of African descent.

A functional mutation in the proximal promoter of the CC chemokine RANTES was significantly more frequent among individuals of African descent compared with Caucasian subjects [43]. This mutation was associated with AD [43], asthma and atopy [44].

The 3' untranslated region of eotaxin has shown a much higher degree of polymorphism in African American and Afro-Caribbean individuals than in Caucasians. Polymorphic alleles were in linkage disequilibrium with asthma in both African American and Afro-Caribbean families, but not in Caucasian families [45].

Ethnic differences have also been described for various other genes involved in the allergic inflammation (e.g. IL-4R α [21], β_2 AR [24]). We can therefore speculate that inconsistent findings in the genetic studies of asthma and atopy summarized in this article may partly be explained by ethnic differences in nature and frequencies of genetic

variants in disease susceptibility genes. We can also speculate that ethnic differences in inflammatory genes (in addition to environmental factors) may also underlie the significant worldwide differences in the prevalence of asthma, allergic rhinitis, and AD [46].

Conclusion

Asthma and atopy are complex, multifactorial disorders. Major strides have been made in identifying chromosomal regions and candidate genes linked to asthma. However, the significant increase in the prevalence of atopy-related disorders over the past decades cannot be explained by changes in gene frequencies. It is rather probable that various pre-existing genetic factors interacting with a dramatically changing environment (decline of infectious diseases, change in diet, immunizations, and others) have rendered a large percentage of the population susceptible to asthma and atopy. Genetic variations that evolved to improve resistance to infections may very probably be misdirected to promote allergic inflammation in the absence of infection in Western societies. Redundancies in host defense mechanisms may explain the large number of chromosomal regions as well as a steadily growing number of genetic variants related to atopy. Inconsistent findings summarized in this article may be explained by ethnic differences in host defense genes, but also by limitations to taking gene–gene as well as gene–environment interactions into account. Large prospective, multicenter studies, in addition to retrospective collaborations as described previously [14], may help to better understand genetic and environmental risk factors for atopy.

Acknowledgement

This article was supported by BMBF grant 01GC0002.

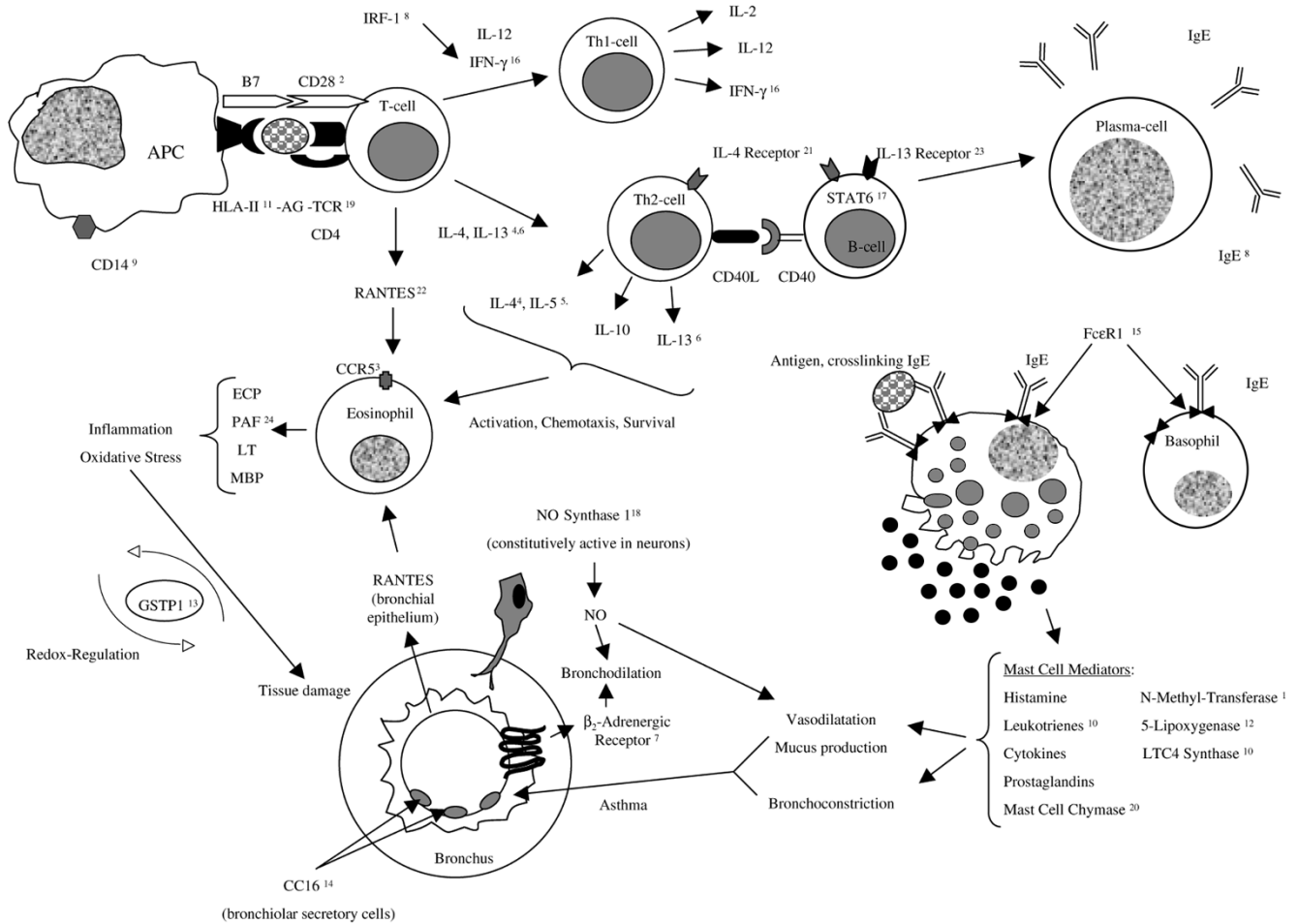
References

1. von Mutius E, Martinez FD, Fritzsche C, Nicolai T, Roell G, Thiemann HH: **Prevalence of asthma and atopy in two areas of West and East Germany.** *Am J Respir Crit Care Med* 1994, **149**: 358-364.
2. von Mutius E, Braun-Fahrländer C, Schierl R, Riedler J, Ehlermann S, Maisch S, Waser M, Nowak D: **Exposure to endotoxin or other bacterial components might protect against the development of atopy.** *Clin Exp Allergy* 2000, **30**:1230-1234.
3. von Mutius E, Martinez FD, Fritzsche C, Nicolai T, Reitmeier P, Thiemann HH: **Skin test reactivity and number of siblings.** *BMJ* 1994, **308**:692-695.
4. Von Ehrenstein OS, Von Mutius E, Illi S, Baumann L, Bohm O, von Kries R: **Reduced risk of hay fever and asthma among children of farmers.** *Clin Exp Allergy* 2000, **30**:187-193.
5. Illi S, von Mutius E, Lau S, Bergmann R, Niggemann B, Sommerfeld C, Wahn U: **Early childhood infectious diseases and the development of asthma up to school age: a birth cohort study.** *BMJ* 2001, **322**:390-395.
6. Gereda JE, Leung DY, Thatayatikom A, Streib JE, Price MR, Klinnert MD, Liu AH: **Relation between house-dust endotoxin exposure, type 1 T-cell development, and allergen sensitisation in infants at high risk of asthma.** *Lancet* 2000, **355**:1680-1683.
7. Sandford A, Weir T, Pare P: **The genetics of asthma.** *Am J Respir Crit Care Med* 1996, **153**:1749-1765.
8. Skadhauge LR, Christensen K, Kyvik KO, Sigsgaard T: **Genetic and environmental influence on asthma: a population-based study of 11,688 Danish twin pairs.** *Eur Respir J* 1999, **13**:8-14.
9. Lander ES, Schork NJ: **Genetic dissection of complex traits.** *Science* 1994, **265**:2037-2048.
10. Barnes KC: **Atopy and asthma genes – where do we stand?** *Allergy* 2000, **55**:803-817.
11. Cookson W: **The alliance of genes and environment in asthma and allergy.** *Nature* 1999, **402**:B5-B11.
12. Ober C, Moffatt MF: **Contributing factors to the pathobiology. The genetics of asthma.** *Clin Chest Med* 2000, **21**:245-261.
13. Palmer LJ, Cookson WO: **Genomic approaches to understanding asthma.** *Genome Res* 2000, **10**:1280-1287.
14. Lonjou C, Barnes K, Chen H, Cookson WO, Deichmann KA, Hall IP, Holloway JW, Laitinen T, Palmer LJ, Wjst M, Morton NE: **A first trial of retrospective collaboration for positional cloning in complex inheritance: assay of the cytokine region on chromosome 5 by the consortium on asthma genetics (COAG).** *Proc Natl Acad Sci USA* 2000, **97**:10942-10947.
15. Palmer LJ, Lonjou C, Barnes K, Chen H, Cookson WO, Deichmann KA, Holloway JW, Laitinen T, Wjst M, Morton NE: **Special Report: A retrospective collaboration on chromosome 5 by the International Consortium on Asthma Genetics (COAG).** *Clin Exp Allergy* 2001, **31**:152-154.
16. Becker KG, Simon RM, Bailey-Wilson JE, Freidlin B, Biddison WE, McFarland HF, Trent JM: **Clustering of non-major histocompatibility complex susceptibility candidate loci in human autoimmune diseases.** *Proc Natl Acad Sci USA* 1998, **95**: 9979-9984.
17. Cookson WO, Ubhi B, Lawrence R, Abecasis GR, Walley AJ, Cox HE, Coleman R, Leaves NI, Trembath RC, Moffatt MF, Harper JI: **Genetic linkage of childhood atopic dermatitis to psoriasis susceptibility loci.** *Nat Genet* 2001, **27**:372-373.
18. Lee YA, Wahn U, Kehrt R, Tarani L, Businco L, Gustafsson D, Andersson F, Oranje AP, Wolkertstorfer A, Berg A, Hoffmann U, Kuster W, Wienker T, Ruschendorf F, Reis A: **A major susceptibility locus for atopic dermatitis maps to chromosome 3q21.** *Nat Genet* 2000, **26**:470-473.
19. Graves PE, Kabesch M, Halonen M, Holberg CJ, Baldini M, Fritzsche C, Weiland SK, Erickson RP, von Mutius E, Martinez FD: **A cluster of seven tightly linked polymorphisms in the IL-13 gene is associated with total serum IgE levels in three populations of white children.** *J Allergy Clin Immunol* 2000, **105**: 506-513.
20. Kruse S, Japha T, Tedner M, Sparholt SH, Forster J, Kuehr J, Deichmann KA: **The polymorphisms S503P and Q576R in the interleukin-4 receptor alpha gene are associated with atopy and influence the signal transduction.** *Immunology* 1999, **96**: 365-371.
21. Ober C, Leavitt SA, Tsalenko A, Howard TD, Hoki DM, Daniel R, Newman DL, Wu X, Parry R, Lester LA, Solway J, Blumenthal M, King RA, Xu J, Meyers DA, Bleeker ER, Cox NJ: **Variation in the interleukin 4-receptor alpha gene confers susceptibility to asthma and atopy in ethnically diverse populations.** *Am J Hum Genet* 2000, **66**:517-526.
22. Barnes KC, Mathias RA, Nickel R, Freidhoff LR, Stockton ML, Xue X, Naidu RP, Levett PN, Casolaro V, Beaty TH: **Testing for gene-gene interaction controlling total IgE in families from Barbados: Evidence of sensitivity regarding linkage heterogeneity among families.** *Genomics* 2001, **71**:246-251.
23. Hall IP: **Pharmacogenetics of asthma.** *Eur Respir J* 2000, **15**: 449-451.
24. Drysdale CM, McGraw DW, Stack CB, Stephens JC, Judson RS, Nandabalan K, Arnold K, Ruano G, Liggett SB: **Complex promoter and coding region beta 2-adrenergic receptor haplotypes alter receptor expression and predict in vivo responsiveness.** *Proc Natl Acad Sci USA* 2000, **97**:10483-10488.
25. Turki J, Pak J, Green SA, Martin RJ, Liggett SB: **Genetic polymorphisms of the beta 2-adrenergic receptor in nocturnal and nonnocturnal asthma. Evidence that Gly16 correlates with the nocturnal phenotype.** *J Clin Invest* 1995, **95**:1635-1641.
26. Szeffler SJ, Ando R, Cicutto LC, Surs W, Hill MR, Martin RJ: **Plasma histamine, epinephrine, cortisol, and leukocyte beta-adrenergic receptors in nocturnal asthma.** *Clin Pharmacol Ther* 1991, **49**:59-68.
27. Israel E, Drazen JM, Liggett SB, Boushey HA, Cherniack RM, Chinchilli VM, Cooper DM, Fahy JV, Fish JE, Ford JG, Kraft M, Kunselman S, Lazarus SC, Lemanske RF, Martin RJ, McLean DE, Peters SP, Silverman EK, Sorkness CA, Szeffler SJ, Weiss ST, Yandava

- CN: **The effect of polymorphisms of the beta(2)-adrenergic receptor on the response to regular use of albuterol in asthma.** *Am J Respir Crit Care Med* 2000, **162**:75-80.
28. Yan L, Galinsky RE, Bernstein JA, Liggett SB, Weinshilboum RM: **Histamine N-methyltransferase pharmacogenetics: association of a common functional polymorphism with asthma.** *Pharmacogenetics* 2000, **10**:261-266.
 29. Drazen JM, Yandava CN, Dube L, Szczerback N, Hippensteel R, Pillari A, Israel E, Schork N, Silverman ES, Katz DA, Drajesk J: **Pharmacogenetic association between ALOX5 promoter genotype and the response to anti-asthma treatment.** *Nat Genet* 1999, **22**:168-170.
 30. Sanak M, Pierzchalska M, Bazan-Socha S, Szczeklik A: **Enhanced expression of the leukotriene C(4) synthase due to overactive transcription of an allelic variant associated with aspirin-intolerant asthma.** *Am J Respir Cell Mol Biol* 2000, **23**:290-296.
 31. Marquet S, Abel L, Hillaire D, Dessein H, Kalil J, Feingold J, Weissenbach J, Dessein AJ: **Genetic localization of a locus controlling the intensity of infection by Schistosoma mansoni on chromosome 5q31-q33.** *Nat Genet* 1996, **14**:181-184.
 32. Rihet P, Traore Y, Abel L, Aucan C, Traore-Leroux T, Fumoux F: **Malaria in humans: Plasmodium falciparum blood infection levels are linked to chromosome 5q31-q33.** *Am J Hum Genet* 1998, **63**:498-505.
 33. Dessein AJ, Hillaire D, Elwali NE, Marquet S, Mohamed-Ali Q, Mirghani A, Henri S, Abdelhameed AA, Saeed OK, Magzoub MM, Abel L: **Severe hepatic fibrosis in Schistosoma mansoni infection is controlled by a major locus that is closely linked to the interferon-gamma receptor gene.** *Am J Hum Genet* 1999, **65**:709-721.
 34. Roberts M, Butterworth AE, Kimani G, Kamau T, Fulford AJ, Dunne DW, Ouma JH, Sturrock RF: **Immunity after treatment of human schistosomiasis: association between cellular responses and resistance to reinfection.** *Infect Immun* 1993, **61**:4984-4993.
 35. Palmer LJ, Pare PD, Faux JA, Moffatt MF, Daniels SE, LeSouef PN, Bremner PR, Mockford E, Gracey M, Spargo R, Musk AW, Cookson WO: **Fc epsilon R1-beta polymorphism and total serum IgE levels in endemically parasitized Australian aborigines.** *Am J Hum Genet* 1997, **61**:182-188.
 36. Ramsay CE, Hayden CM, Tiller KJ, Burton PR, Hagel I, Palenque M, Lynch NR, Goldblatt J, LeSouef PN: **Association of polymorphisms in the beta2-adrenoreceptor gene with higher levels of parasitic infection.** *Hum Genet* 1999, **104**:269-274.
 37. Nickel R, Beck LA, Stellato C, Schleimer RP: **Chemokines and allergic disease.** *J Allergy Clin Immunol* 1999, **104**:723-742.
 38. **A genome-wide search for asthma susceptibility loci in ethnically diverse populations. The Collaborative Study on the Genetics of Asthma (CSGA).** *Nat Genet* 1997, **15**:389-392.
 39. Martinson JJ, Chapman NH, Rees DC, Liu YT, Clegg JB: **Global distribution of the CCR5 gene 32-basepair deletion.** *Nat Genet* 1997, **16**:100-103.
 40. Hall IP, Wheatley A, Christie G, McDougall C, Hubbard R, Helms PJ: **Association of CCR5 delta32 with reduced risk of asthma.** *Lancet* 1999, **354**:1264-1265.
 41. Mitchell TJ, Walley AJ, Pease JE, Venables PJ, Wiltshire S, Williams TJ, Cookson WO: **Delta 32 deletion of CCR5 gene and association with asthma or atopy.** *Lancet* 2000, **356**:1491-1492.
 42. Hadley TJ, Peiper SC: **From malaria to chemokine receptor: the emerging physiologic role of the Duffy blood group antigen.** *Blood* 1997, **89**:3077-3091.
 43. Nickel RG, Casolaro V, Wahn U, Beyer K, Barnes KC, Plunkett BS, Freidhoff LR, Sengler C, Plitt JR, Schleimer RP, Caraballo L, Naidu RP, Levett PN, Beaty TH, Huang SK: **Atopic dermatitis is associated with a functional mutation in the promoter of the C-C chemokine RANTES.** *J Immunol* 2000, **164**:1612-1616.
 44. Fryer AA, Spiteri MA, Bianco A, Hepple M, Jones PW, Strange RC, Makki R, Tavernier G, Smilie FI, Custovic A, Woodcock AA, Ollier WE, Hajeer AH: **The -403 G → A promoter polymorphism in the RANTES gene is associated with atopy and asthma.** *Genes Immun* 2000, **1**:509-514.
 45. Nickel R, Barnes KC, Sengler C, Casolaro V, Freidhoff LR, Weber P, Naidu R, Caraballo L, Ehrlich E, Plitt J, Schleimer RP, Huang SK, Beaty T: **Evidence for linkage of chemokine polymorphisms to asthma in populations of African descent [abstract].** *J Allergy Clin Immunol* 1999, **103**:S174.
 46. **Worldwide variation in prevalence of symptoms of asthma, allergic rhinoconjunctivitis, and atopic eczema: ISAAC. The International Study of Asthma and Allergies in Childhood (ISAAC) Steering Committee.** *Lancet* 1998, **351**:1225-1232.

Supplementary material

Supplementary Figure 1



Simplified cartoon of the allergic inflammation showing the mechanisms and molecules involved starting from antigen-presentation to organ-specific allergic reactions. Outlined are most candidate genes for atopy-associated phenotypes that have been analyzed to date (superscript numbers refer to Supplementary Table 3). APC, antigen presenting cell; AG, antigen; B7, costimulatory molecule B7; CC16, Clara cell protein 16; CCR5, CC chemokine receptor 5; CD, cluster of differentiation; CD40L, CD40 ligand; ECP, eosinophilic cationic protein; Fc ϵ R1- β , Fc epsilon receptor 1 beta (IgE receptor); GSTP1, glutathione-S-transferase 1; HLA, human leukocyte antigen; IFN, interferon; IL, interleukin; IRF-1, interferon regulatory factor 1; LT, leukotriene; LTC4, leukotriene C4 synthase; MBP, major basic protein; NO, nitric oxide; PAF, platelet activating factor; RANTES, regulated on activation, normal T cell expressed and secreted; STAT, signal transducer and activator of transcription; TCR, T-cell receptor; Th, T helper cell.

Supplementary Table 1

Results of genome-wide searches (asthma and associated traits)

Population	Study												
	CSGA 1 (1997)			Hizawa et al. (1998)		Daniels et al. (1996) C (Australian)	Ober et al. (1998) C (Hutterites)	Ober et al. (2000) [†] C (Hutterites)	Wjst et al. (1999) C (German/Swedish)	Yokouchi et al. (2000) C (Japanese)	EGEA (2000) C (French)	Lee et al. (2000) C (German)	Cookson et al. (2001) C (British)
Individuals (n)	117 [‡]	215 [‡]	48 [‡]	281	299	364	371/292	693	415	196	493	839	
Sib-pairs (n)	ns	ns	ns	ns	ns	172	ns	ns	156	65	ns	199	213
1q21													AD*
1p31											Asthma *		
2pter									Asthma*, IgE**				
									BHR*, RAST**				
2q22-33			Asthma*		IgE*								
3q21												AD***	
4q35						BHR**, atopy*				Asthma**			
5p13-15	Asthma**							BHR (D5S1470)					
5q23-33				IgE*			BHR**	BHR (D5S1462)		Asthma***			
6p21.3-23					IgE*	Eos***			Asthma*, IgE*				
									RAST*				
7p						IgE*, BHR** (D7S2250, D7S484)							

Continued overleaf

Supplementary Table 1 Continued

Population	Study											
	CSGA 1 (1997)			Hizawa <i>et al.</i> (1998)		Daniels <i>et al.</i> (1996) C (Australian)	Ober <i>et al.</i> (1998) C (Hutterites)	Ober <i>et al.</i> (2000) [†] C (Hutterites)	Wjst <i>et al.</i> (1999) C (German/Swedish)	Yokouchi <i>et al.</i> (2000) C (Japanese)	EGEA (2000) C (French)	Lee <i>et al.</i> (2000) C (German)
8p23.3				IgE*				Asthma (D8S1136)				
9 (DS1784)								Asthma*, IgE*				
								RAST*				
11p13-15		Asthma *								IgE*		
11q13						IgE**, ST***				IgE*		
12q13-24.2		Asthma*					BHR*	Asthma*		Eos**		
13q11-32.2		Asthma*			IgE*	Atopy**				Asthma**		Eos*
14q11.2-13		Asthma*										
14q32								BHR (gata193a07)				
16q-tel												IgE**
16q23-24						IgE**		ST (D16S539)				
17p11.1-q11.2	Asthma*											
17q12-21										Asthma/ST*		
17q25												AD**
19q13		Asthma*					Asthma*	BHR (D19S900)		BHR*		
20p												Asthma**
21q21		Asthma*										

AA, African American; AD, Atopic dermatitis; BHR, bronchial hyperresponsiveness; C, Caucasian; eos, eosinophil count; H, Hispanic; ns, not specified; RAST, radio allerge sorbent test; ST, positive skin test. * $P \leq 0.01$, ** $P \leq 0.001$, *** $P \leq 0.0001$. † Statistical significance ($P \leq 0.05$) in both tests, likelihood ratio χ^2 test and transmission disequilibrium test. ‡ Only affected individuals, neither number of all individuals nor number of sib-pairs specified [17,18,38,S1–S7].

Supplementary Table 2**Chromosomal regions showing evidence for linkage to asthma and related phenotypes**

Chromosomal region	Phenotype (linkage)	Population	Reference
5q23-33	Total IgE	Dutch, Amish, Caucasian (UK)	[S8–S10]
	Total IgE, specific IgE, eosinophil count	Australian	[S11]
	Specific IgE	African American	[S12]*
	Atopic dermatitis	Caucasian (UK), German/Swedish	[S13,S14]
	Asthma/atopy	Japanese	[S6,S15]*
	Eosinophil count	Caucasian (US)	[S16]
	Hypereosinophilia	Caucasian (US)	[S17]
	Bronchial hyperresponsiveness	Dutch, Hutterites	[S3,S18]*
6p21-23	Specific IgE	Caucasian (US)	[S1*,S19]
	Asthma, IgE	German/Swedish	[S5]*
	Allergic asthma	Colombian	[S20]
	Eosinophil count	Caucasian (Australian)	[S2]*
11q13	IgE	Caucasian (UK)	[S21,S22]
	IgE, positive skin test	Caucasian (Australian)	[S2]*
	Specific IgE	Caucasian (Australian), African American	[S11,S12]
	Bronchial hyperresponsiveness, log IgE	Caucasian (UK)	[S10]
	Bronchial hyperresponsiveness	Caucasian	[S23]
	Atopy	Japanese	[S24]
	Asthma, atopy	Caucasian	[S25]
12q14-24	Atopic dermatitis	German	[S26]
	Asthma	German/Swedish Caucasian (US)	[38,S27]*
		Caucasian (UK)	[S28]
	Asthma, atopy	Afro-Caribbean	[S29]
	Total IgE, asthma	Afro-Caribbean, Amish	[S30]
	Total IgE	German	[S31]
	Bronchial hyperresponsiveness	Hutterites	[S3]*
13q11-32	Eosinophil count	French	[S7]*
	Atopic dermatitis	German/Swedish	[S14]
	Specific IgE	Caucasian (UK)	[S1]*
	Atopy	Caucasian (Australian)	[S2]*
	Eosinophil count	French	[S7]*
	Asthma	Caucasian (US), Japanese	[38*,S6*]
	Atopic asthma	Japanese	[S32]

* Genome-wide searches.

Supplementary Table 3

Polymorphisms/mutations in candidate genes for asthma and related phenotypes

Chromosomal location	Number*	Molecule	Single nucleotide polymorphism/mutation†	Phenotype related	Population	Functional (in vitro)	Reference								
1p32	1	Histamine-N-methyl-transferase	C314T (Thr105Ile)	Asthma	Caucasian	Yes	[28]								
2q33-34	2	CD28 CTLA-4	T19int3C	None	Caucasian		[S33]								
			A49G	None	Caucasian		[S33]								
			C-318T	None	Caucasian										
3p21	3	CCR5	CC5-Δ32	Asthma	Caucasian	Yes	[40]								
				No Association	British/Australian		[41]								
5q31-34	4	IL-4	C-590T (= C-589T)	Total IgE	Japanese	Yes	[S34]								
				Total IgE	American		[S35]								
				Specific IgE, wheeze	Australian		[S36]								
				FEV1	Caucasian		[S37]								
	5	IL-5	identical {	C+33T None C-1055T C-1112T A-1512C C1923T G2525A C2580A C2749T	None	Japanese		[S38]							
					None	Caucasian		[S39]							
					Total IgE	Japanese		[S40]							
					Allergic asthma	Australian		[S41]							
					Total IgE	Dutch		[S42]							
					Total IgE	American/German		[19]							
7	β ₂ -AR	identical {	Arg130Gln G4257A Gln110Arg	High IgE, atopic dermatitis Atopic and intrinsic asthma	German British/Japanese		[S43] [S44]								
						G-1023A C-709A G-654A C-468G C-406T T-367C T-47C T-20C G46A (= Gly16Arg) C79G (= Gln27Glu) G252A C491T (= Thr164Ile) C523A Gly16Arg	Drug response (Haplotype studies)	Caucasian/African American Asian/Hispanic-Latino	Yes	[24]					
											Gly16Arg	Drug response	American	Yes	[27]
												FEV1 and FVC	Hutterites		[S45]
												Asthma severity	Not specified		[S46]
											Gly16Arg	Nocturnal asthma	American		[25]
												Steroid-dependent asthma	American		[S47]
												None	German		[S48]
											Gln27Glu	Drug response	Not specified		[S49]
												Drug response	American	Yes	[27]
Drug response	Not specified		[S49]												
None	None	German		[S48]											
	Total IgE	Caucasian		[S50]											
	FEV1	British		[S51]											
8	IRF-1	-300 G/T 4396 A/G 6355 G/A	None	Japanese		[S52]									
			None												
			None												
9	CD14	-159 C → T	Total IgE	Caucasian		[S53]									
			None	Hispanic		[S53]									
			Total IgE	British		[S54]									
5q35	10	LTC4 synthase	-444 C	None	Japanese		[S54]								
				Aspirin-induced asthma	Caucasian	Yes	[30]								
				None	American	No	[S55]								

Supplementary Table 3 Continued

Chromosomal location	Number*	Molecule	Single nucleotide polymorphism/mutation†	Phenotype related	Population	Functional (in vitro)	Reference
6p21	11	HLA-II		Specific IgE	Multiple		[S56]
		TNF	-308 TNF2	BHR	Caucasian		[S57]
		LT- α	LT alpha Ncol*1/ TNF-308*2 haplotype	Asthma	British		[S58]
6p21-12	24	PAF-acetyl-hydrolase	-308 TNF1 LT-alpha*2	Asthma	Australian		[S59]
			Ile198Thr	Atopic Asthma/ total IgE	German/British	Yes	[S60]
10	12	5-LO (promoter)	Ala379Val	Asthma/specific IgE		Yes	
11q12-13	14	CC16	Arg92His	None		Yes	
	15	Fc ϵ R1- β	(GGGCGG) _{n=3-6} A38G	Drug Response	Not specified	Yes	[29]
12q13	17	GSTP1	Ile105Val	Asthma	Australian		[S61]
			G2964A	None	British/Japanese		[S62]
			None	Atopy	British	No	[S63]
			Leu181Leu	Atopy	Japanese		[S64]
			Leu181/Leu183	Atopy, BHR	British/Australian	No	[S65]
			E237G (= Gly237Glu)	None	Australian		[S51]
			Rsal_in2*B	Atopy	Australian	No	[S66]
			Rsal_ex7 (+ Rsal_in2*B)	None	Japanese		[S67]
			-109C/T	Asthma	Australian		[S68]
			Ile105Val	Total IgE, atopic dermatitis, asthma	Not specified		[S69]
12q14	16	IFN- γ	None	Total IgE	Japanese		[S70]
			(AAT) _{n\geq12}	IgE/Skin Test/FEV1	Caucasian		[S71]
12q24	18	NO-synthase 1	G2964A	Asthma	Japanese		[S72]
14p11	19	TCR	None		Australian/Venezuelan		[S73]
			VA8.1(*)2	Exhaled NO	Mixed		[S74]
16p12-p11	21	IL-4R α	Q576R (= Arg551Gln)	Specific IgE	Caucasian		[S75]
				Eczema	Japanese		[S76]
				Total IgE	Not specified		[S77]
				None	Japanese		[S78]
				None	Italian		[S79]
				Atopic dermatitis	Japanese		[S80]
				With high IgE			
				Atopy, high IgE	American	Yes	[S81]
17q11-q12	22	RANTES		Asthma severity (FEV1)	American		[S82]
				None	Italian		[S83]
				Atopic dermatitis	Japanese		[S84]
				Low IgE	German	Yes	[20]
				Low IgE			[20]
				None			
				None			
				IgE/atopic asthma	Japanese	Yes	[S85]
				None	Japanese		[S86]
				Ile50Val			
Xq	23	IL-13R α 1	S503P (= 478Pro)				
			E400A				
Xq	23	IL-13R α 1	C431R				
			Ile50Val				
Xq	23	IL-13R α 1	Ile50Val				
			Ser727Ala				
Xq	23	IL-13R α 1	Glu375Ala				
			Cys406Arg				
Xq	23	IL-13R α 1	Ser411Leu				
			Ser761Pro				
Xq	23	IL-13R α 1	Ser478Pro				
			Gln551Arg				
Xq	23	IL-13R α 1	G-401A	Atopy/asthma	Hutterites/Caucasian		[21]
			G-403A	(Haplotype studies)	African American/Hispanic		
Xq	23	IL-13R α 1	A1398G	Atopic dermatitis	Caucasian	Yes	[43]
				Asthma, atopy	Caucasian		[44]
Xq	23	IL-13R α 1	A1398G	Total IgE	British		[S44]

Divergent positions of identical mutations are due to different database accession numbers used for reference. * Numbers refer to molecules in Supplementary Figure 1. † Nomenclature as in references. β_2 -AR, β_2 adrenergic receptor; BHR, bronchial hyperresponsiveness; CC16, Clara cell protein 16; CCR5, CC chemokine receptor 5; CD, cluster of differentiation; Fc ϵ R1- β , Fc epsilon receptor 1 beta (IgE receptor); FVC, forced vital capacity; FEV1, forced expiratory volume in 1 s; GSTP1, glutathione-S-transferase 1; HLA, human leukocyte antigen; IFN, interferon; IL, interleukin; IL-4/13 R, interleukin 4/13 receptor; IRF-1, interferon regulatory factor 1; 5-LO, 5-lipoxygenase; LT, leukotriene; LT- α , lymphotoxin α ; LTC4, leukotriene C4 synthase; NO, nitric oxide; PAF, platelet activating factor; RANTES, regulated on activation, normal T cell expressed and secreted; STAT6, signal transducer and activator of transcription 6; TCR, T-cell receptor; TNF, tumor necrosis factor.

References

- S1. Hizawa N, Freidhoff LR, Chiu YF, Ehrlich E, Luehr CA, Anderson JL, Duffy DL, Dunston GM, Weber JL, Huang SK, Barnes KC, Marsh DG, Beaty TH: **Genetic regulation of Dermatophagoides pteronyssinus-specific IgE responsiveness: a genome-wide multipoint linkage analysis in families recruited through 2 asthmatic sibs. Collaborative Study on the Genetics of Asthma (CSGA).** *J Allergy Clin Immunol* 1998, **102**:436-442.
- S2. Daniels SE, Bhattacharya S, James A, Leaves NI, Young A, Hill MR, Faux JA, Ryan GF, le Souef PN, Lathrop GM, Musk AW, Cookson WO: **A genome-wide search for quantitative trait loci underlying asthma.** *Nature* 1996, **383**:247-250.
- S3. Ober C, Cox NJ, Abney M, Di Rienzo A, Lander ES, Changyaleket B, Gidley H, Kurtz B, Lee J, Nance M, Pettersson A, Prescott J, Richardson A, Schlenker E, Summerhill E, Willadsen S, Parry R: **Genome-wide search for asthma susceptibility loci in a founder population. The Collaborative Study on the Genetics of Asthma.** *Hum Mol Genet* 1998, **7**:1393-1398.
- S4. Ober C, Tsalenko A, Parry R, Cox NJ: **A second-generation genome-wide screen for asthma-susceptibility alleles in a founder population.** *Am J Hum Genet* 2000, **67**:1154-1162.
- S5. Wjst M, Fischer G, Immervoll T, Jung M, Saar K, Rueschendorf F, Reis A, Ulbrecht M, Gomolka M, Weiss EH, Jaeger L, Nickel R, Richter K, Kjellman NI, Griese M, von Berg A, Gappa M, Riedel R, Boehle M, van Koningsbruggen S, Schoberth P, Szczepanski R, Dorsch W, Silbermann M, Wichmann HE: **A genome-wide search for linkage to asthma. German Asthma Genetics Group.** *Genomics* 1999, **58**:1-8.
- S6. Yokouchi Y, Nukaga Y, Shibasaki M, Noguchi E, Kimura K, Ito S, Nishihara M, Yamakawa-Kobayashi K, Takeda K, Imoto N, Ichikawa K, Matsui A, Hamaguchi H, Arinami T: **Significant evidence for linkage of mite-sensitive childhood asthma to chromosome 5q31-q33 near the interleukin 12 B locus by a genome-wide search in Japanese families.** *Genomics* 2000, **66**:152-160.
- S7. Dizier MH, Besse-Schmittler C, Guilloud-Bataille M, Annesi-Maesano I, Boussaha M, Bousquet J, Charpin D, Degioanni A, Gormand F, Grimfeld A, Hochez J, Hyne G, Lockhart A, Luillier-Lacombe M, Matran R, Meunier F, Neukirch F, Pacheco Y, Parent V, Paty E, Pin I, Pison C, Scheinmann P, Thobie N, Vervloet D, Kauffmann F: **Genome screen for asthma and related phenotypes in the french EGEA study.** *Am J Respir Crit Care Med* 2000, **162**:1812-1818.
- S8. Meyers DA, Postma DS, Panhuysen CI, Xu J, Amelung PJ, Levitt RC, Bleeker ER: **Evidence for a locus regulating total serum IgE levels mapping to chromosome 5.** *Genomics* 1994, **23**:464-470.
- S9. Marsh DG, Neely JD, Breazeale DR, Ghosh B, Freidhoff LR, Ehrlich-Kautzky E, Schou C, Krishnaswamy G, Beaty TH: **Linkage analysis of IL4 and other chromosome 5q31.1 markers and total serum immunoglobulin E concentrations.** *Science* 1994, **264**:1152-1156.
- S10. Doull IJ, Lawrence S, Watson M, Begishvili T, Beasley RW, Lampe F, Holgate T, Morton NE: **Allelic association of gene markers on chromosomes 5q and 11q with atopy and bronchial hyper-responsiveness.** *Am J Respir Crit Care Med* 1996, **153**:1280-1284.
- S11. Palmer LJ, Daniels SE, Rye PJ, Gibson NA, Tay GK, Cookson WO, Goldblatt J, Burton PR, LeSouef PN: **Linkage of chromosome 5q and 11q gene markers to asthma-associated quantitative traits in Australian children.** *Am J Respir Crit Care Med* 1998, **158**:1825-1830.
- S12. Hizawa N, Freidhoff LR, Ehrlich E, Chiu YF, Duffy DL, Schou C, Dunston GM, Beaty TH, Marsh DG, Barnes KC, Huang SK: **Genetic influences of chromosomes 5q31-q33 and 11q13 on specific IgE responsiveness to common inhaled allergens among African American families. Collaborative Study on the Genetics of Asthma (CSGA).** *J Allergy Clin Immunol* 1998, **102**:449-453.
- S13. Forrest S, Dunn K, Elliott K, Fitzpatrick E, Fullerton J, McCarthy M, Brown J, Hill D, Williamson R: **Identifying genes predisposing to atopic eczema.** *J Allergy Clin Immunol* 1999, **104**:1066-1070.
- S14. Beyer K, Nickel R, Freidhoff L, Bjorksten B, Huang SK, Barnes KC, MacDonald S, Forster J, Zepp F, Wahn V, Beaty TH, Marsh DG, Wahn U: **Association and linkage of atopic dermatitis with chromosome 13q12-14 and 5q31-33 markers.** *J Invest Dermatol* 2000, **115**:906-908.
- S15. Noguchi E, Shibasaki M, Arinami T, Takeda K, Maki T, Miyamoto T, Kawashima T, Kobayashi K, Hamaguchi H: **Evidence for linkage between asthma/atopy in childhood and chromosome 5q31-q33 in a Japanese population.** *Am J Respir Crit Care Med* 1997, **156**:1390-1393.
- S16. Martinez FD, Solomon S, Holberg CJ, Graves PE, Baldini M, Erickson RP: **Linkage of circulating eosinophils to markers on chromosome 5q.** *Am J Respir Crit Care Med* 1998, **158**:1739-1744.
- S17. Rioux JD, Stone VA, Daly MJ, Cargill M, Green T, Nguyen H, Nutman T, Zimmerman PA, Tucker MA, Hudson T, Goldstein AM, Lander E, Lin AY: **Familial eosinophilia maps to the cytokine gene cluster on human chromosomal region 5q31-q33.** *Am J Hum Genet* 1998, **63**:1086-1094.
- S18. Postma DS, Bleeker ER, Amelung PJ, Holroyd KJ, Xu J, Panhuysen CI, Meyers DA, Levitt RC: **Genetic susceptibility to asthma - bronchial hyperresponsiveness coinherit with a major gene for atopy.** *N Engl J Med* 1995, **333**:894-900.
- S19. Hizawa N, Collins G, Rafnar T, Huang SK, Duffy DL, Weber JL, Freidhoff LR, Ehrlich E, Marsh DG, Beaty TH, Barnes KC: **Linkage analysis of Dermatophagoides pteronyssinus-specific IgE responsiveness with polymorphic markers on chromosome 6p21 (HLA-D region) in Caucasian families by the transmission/disequilibrium test. Collaborative Study on the Genetics of Asthma (CSGA).** *J Allergy Clin Immunol* 1998, **102**:443-448.
- S20. Caraballo LR, Hernandez M: **HLA haplotype segregation in families with allergic asthma.** *Tissue Antigens* 1990, **35**:182-186.
- S21. Cookson WO, Sharp PA, Faux JA, Hopkin JM: **Linkage between immunoglobulin E responses underlying asthma and rhinitis and chromosome 11q.** *Lancet* 1989, **1**:1292-1295.
- S22. Young RP, Sharp PA, Lynch JR, Faux JA, Lathrop GM, Cookson WO, Hopkin JM: **Confirmation of genetic linkage between atopic IgE responses and chromosome 11q13.** *J Med Genet* 1992, **29**:236-238.
- S23. van Herwerden L, Harrap SB, Wong ZY, Abramson MJ, Kutin JJ, Forbes AB, Raven J, Lanigan A, Walters EH: **Linkage of high-affinity IgE receptor gene with bronchial hyperreactivity, even in absence of atopy.** *Lancet* 1995, **346**:1262-1265.
- S24. Shirakawa T, Hashimoto T, Furuyama J, Takeshita T, Morimoto K: **Linkage between severe atopy and chromosome 11q13 in Japanese families.** *Clin Genet* 1994, **46**:228-232.
- S25. Collee JM, ten Kate LP, de Vries HG, Kliphuis JW, Bouman K, Scheffer H, Gerritsen J: **Allele sharing on chromosome 11q13 in sibs with asthma and atopy [letter].** *Lancet* 1993, **342**:936.
- S26. Folster-Holst R, Moises HW, Yang L, Fritsch W, Weissenbach J, Christophers E: **Linkage between atopy and the IgE high-affinity receptor gene at 11q13 in atopic dermatitis families.** *Hum Genet* 1998, **102**:236-239.
- S27. Wjst M, Wichmann HE: **Collaborative study on the genetics of asthma in Germany.** *Clin Exp Allergy* 1995, **25**(suppl 2):23-25.
- S28. Wilkinson J, Grimley S, Collins A, Thomas NS, Holgate ST, Morton N: **Linkage of asthma to markers on chromosome 12 in a sample of 240 families using quantitative phenotype scores.** *Genomics* 1998, **53**:251-259.
- S29. Barnes KC, Freidhoff LR, Nickel R, Chiu YF, Juo SH, Hizawa N, Naidu RP, Ehrlich E, Duffy DL, Schou C, Levett PN, Marsh DG, Beaty TH: **Dense mapping of chromosome 12q13.12-q23.3 and linkage to asthma and atopy.** *J Allergy Clin Immunol* 1999, **104**:485-491.
- S30. Barnes KC, Neely JD, Duffy DL, Freidhoff LR, Breazeale DR, Schou C, Naidu RP, Levett PN, Renault B, Kucherlapati R, Iozzino S, Ehrlich E, Beaty TH, Marsh DG: **Linkage of asthma and total serum IgE concentration to markers on chromosome 12q: evidence from Afro-Caribbean and Caucasian populations.** *Genomics* 1996, **37**:41-50.
- S31. Nickel R, Wahn U, Hizawa N, Maestri N, Duffy DL, Barnes KC, Beyer K, Forster J, Bergmann R, Zepp F, Wahn V, Marsh DG: **Evidence for linkage of chromosome 12q15-q24.1 markers to high total serum IgE concentrations in children of the German Multicenter Allergy Study.** *Genomics* 1997, **46**:159-162.
- S32. Kimura K, Noguchi E, Shibasaki M, Arinami T, Yokouchi Y, Takeda K, Yamakawa-Kobayashi K, Matsui A, Hamaguchi H: **Linkage and association of atopic asthma to markers on chromosome 13 in the Japanese population.** *Hum Mol Genet* 1999, **8**:1487-1490.
- S33. Heinzmann A, Plesnar C, Kuehr J, Forster J, Deichmann KA: **Common polymorphisms in the CTLA-4 and CD28 genes at**

- 2q33 are not associated with asthma or atopy.** *Eur J Immunogenet* 2000, **27**:57-61.
- S34. Noguchi E, Shibasaki M, Arinami T, Takeda K, Yokouchi Y, Kawashima T, Yanagi H, Matsui A, Hamaguchi H: **Association of asthma and the interleukin-4 promoter gene in Japanese.** *Clin Exp Allergy* 1998, **28**:449-453.
- S35. Rosenwasser LJ, Klemm DJ, Dresback JK, Inamura H, Mascali JJ, Klinnert M, Borish L: **Promoter polymorphisms in the chromosome 5 gene cluster in asthma and atopy.** *Clin Exp Allergy* 1995, **25**(suppl 2):74-78, [discussion] 95-76.
- S36. Walley AJ, Cookson WO: **Investigation of an interleukin-4 promoter polymorphism for associations with asthma and atopy.** *J Med Genet* 1996, **33**:689-692.
- S37. Burchard EG, Silverman EK, Rosenwasser LJ, Borish L, Yandava C, Pillari A, Weiss ST, Hasday J, Lilly CM, Ford JG, Drazen JM: **Association between a sequence variant in the IL-4 gene promoter and FEV(1) in asthma.** *Am J Respir Crit Care Med* 1999, **160**:919-922.
- S38. Kawashima T, Noguchi E, Arinami T, Yamakawa-Kobayashi K, Nakagawa H, Otsuka F, Hamaguchi H: **Linkage and association of an interleukin 4 gene polymorphism with atopic dermatitis in Japanese families.** *J Med Genet* 1998, **35**:502-504.
- S39. Dizier MH, Sandford A, Walley A, Philippi A, Cookson W, Demenais F: **Indication of linkage of serum IgE levels to the interleukin-4 gene and exclusion of the contribution of the (-590 C to T) interleukin-4 promoter polymorphism to IgE variation.** *Genet Epidemiol* 1999, **16**:84-94.
- S40. Suzuki I, Yamaguchi E, Hizawa N, Itoh A, Kawakami Y: **A new polymorphism in the 5' flanking region of the human interleukin (IL)-4 gene.** *Immunogenetics* 1999, **49**:738-739.
- S41. Pereira E, Goldblatt J, Rye P, Sanderson C, Le Souef P: **Mutation analysis of interleukin-5 in an asthmatic cohort.** *Hum Mutat* 1998, **11**:51-54.
- S42. van der Pouw Kraan TC, van Veen A, Boeijs LC, van Tuyl SA, de Groot ER, Stapel SO, Bakker A, Verweij CL, Aarden LA, van der Zee JS: **An IL-13 promoter polymorphism associated with increased risk of allergic asthma.** *Genes Immun* 1999, **1**:61-65.
- S43. Liu X, Nickel R, Beyer K, Wahn U, Ehrlich E, Freidhoff LR, Bjorksten B, Beaty TH, Huang SK: **An IL13 coding region variant is associated with a high total serum IgE level and atopic dermatitis in the German multicenter atopy study (MAS-90).** *J Allergy Clin Immunol* 2000, **106**:167-170.
- S44. Heinzmann A, Mao XQ, Akaiwa M, Kreomer RT, Gao PS, Ohshima K, Umeshita R, Abe Y, Braun S, Yamashita T, Roberts MH, Sugimoto R, Arima K, Arinobu Y, Yu B, Kruse S, Enomoto T, Dake Y, Kawai M, Shimazu S, Sasaki S, Adra CN, Kitaichi M, Inoue H, Yamauchi K, Tomichi N, Kurimoto F, Hamasaki N, Hopkin JM, Izuhara K, Shirakawa T, Deichmann KA: **Genetic variants of IL-13 signalling and human asthma and atopy.** *Hum Mol Genet* 2000, **9**:549-559.
- S45. Summerhill E, Leavitt SA, Gidley H, Parry R, Solway J, Ober C: **Beta(2)-adrenergic receptor Arg16/Arg16 genotype is associated with reduced lung function, but not with asthma, in the Hutterites.** *Am J Respir Crit Care Med* 2000, **162**:599-602.
- S46. Holloway JW, Dunbar PR, Riley GA, Sawyer GM, Fitzharris PF, Pearce N, Le Gros GS, Beasley R: **Association of beta2-adrenergic receptor polymorphisms with severe asthma.** *Clin Exp Allergy* 2000, **30**:1097-1103.
- S47. Reihnsaus E, Innis M, MacIntyre N, Liggett SB: **Mutations in the gene encoding for the beta 2-adrenergic receptor in normal and asthmatic subjects.** *Am J Respir Cell Mol Biol* 1993, **8**:334-339.
- S48. Deichmann KA, Schmidt A, Heinzmann A, Kruse S, Forster J, Kuehr J: **Association studies on beta2-adrenoceptor polymorphisms and enhanced IgE responsiveness in an atopic population.** *Clin Exp Allergy* 1999, **29**:794-799.
- S49. Tan S, Hall IP, Dewar J, Dow E, Lipworth B: **Association between beta 2-adrenoceptor polymorphism and susceptibility to bronchodilator desensitisation in moderately severe stable asthmatics.** *Lancet* 1997, **350**:995-999.
- S50. Dewar JC, Wilkinson J, Wheatley A, Thomas NS, Doull I, Morton N, Lio P, Harvey JF, Liggett SB, Holgate ST, Hall IP: **The glutamine 27 beta2-adrenoceptor polymorphism is associated with elevated IgE levels in asthmatic families.** *J Allergy Clin Immunol* 1997, **100**:261-265.
- S51. Hall IP, Wheatley A, Wilding P, Liggett SB: **Association of Glu 27 beta 2-adrenoceptor polymorphism with lower airway reactivity in asthmatic subjects.** *Lancet* 1995, **345**:1213-1214.
- S52. Noguchi E, Shibasaki M, Arinami T, Yamakawa-Kobayashi K, Yokouchi Y, Takeda K, Matsui A, Hamaguchi H: **Mutation screening of interferon regulatory factor 1 gene (IRF-1) as a candidate gene for atopy/asthma.** *Clin Exp Allergy* 2000, **30**:1563-1567.
- S53. Baldini M, Lohman IC, Halonen M, Erickson RP, Holt PG, Martinez FD: **A Polymorphism* in the 5' flanking region of the CD14 gene is associated with circulating soluble CD14 levels and with total serum immunoglobulin E.** *Am J Respir Cell Mol Biol* 1999, **20**:976-983.
- S54. Gao PS, Mao XQ, Baldini M, Roberts MH, Adra CN, Shirakawa T, Holt PG, Martinez FD, Hopkin JM: **Serum total IgE levels and CD14 on chromosome 5q31.** *Clin Genet* 1999, **56**:164-165.
- S55. Van Sambek R, Stevenson DD, Baldasaro M, Lam BK, Zhao J, Yoshida S, Yandora C, Drazen JM, Penrose JF: **5' flanking region polymorphism of the gene encoding leukotriene C4 synthase does not correlate with the aspirin-intolerant asthma phenotype in the United States.** *J Allergy Clin Immunol* 2000, **106**:72-76.
- S56. Moffatt MF, Cookson WO: **The genetics of specific allergy.** *Monogr Allergy* 1996, **33**:71-96.
- S57. Li Kam, Wa TC, Mansur AH, Britton J, Williams G, Pavord I, Richards K, Campbell DA, Morton N, Holgate ST, Morrison JF: **Association between -308 tumour necrosis factor promoter polymorphism and bronchial hyperreactivity in asthma.** *Clin Exp Allergy* 1999, **29**:1204-1208.
- S58. Moffatt MF, Cookson WO: **Tumour necrosis factor haplotypes and asthma.** *Hum Mol Genet* 1997, **6**:551-554.
- S59. Albuquerque RV, Hayden CM, Palmer LJ, Laing IA, Rye PJ, Gibson NA, Burton PR, Goldblatt J, Lesouef PN: **Association of polymorphisms within the tumour necrosis factor (TNF) genes and childhood asthma.** *Clin Exp Allergy* 1998, **28**:578-584.
- S60. Kruse S, Mao XQ, Heinzmann A, Blattmann S, Roberts MH, Braun S, Gao PS, Forster J, Kuehr J, Hopkin JM, Shirakawa T, Deichmann KA: **The Ile198Thr and Ala379Val variants of plasmatic PAF-acetylhydrolase impair catalytic activities and are associated with atopy and asthma.** *Am J Hum Genet* 2000, **66**:1522-1530.
- S61. Laing IA, Goldblatt J, Eber E, Hayden CM, Rye PJ, Gibson NA, Palmer LJ, Burton PR, Le Souef PN: **A polymorphism of the CC16 gene is associated with an increased risk of asthma.** *J Med Genet* 1998, **35**:463-467.
- S62. Gao PS, Mao XQ, Kawai M, Enomoto T, Sasaki S, Tanabe O, Yoshimura K, Shaldon SR, Dake Y, Kitano H, Coull P, Shirakawa T, Hopkin JM: **Negative association between asthma and variants of CC16(CC10) on chromosome 11q13 in British and Japanese populations.** *Hum Genet* 1998, **103**:57-59.
- S63. Shirakawa T, Li A, Dubowitz M, Dekker JW, Shaw AE, Faux JA, Ra C, Cookson WO, Hopkin JM: **Association between atopy and variants of the beta subunit of the high-affinity immunoglobulin E receptor.** *Nat Genet* 1994, **7**:125-129.
- S64. Shirakawa T, Mao XQ, Sasaki S, Enomoto T, Kawai M, Morimoto K, Hopkin J: **Association between atopic asthma and a coding variant of Fc epsilon RI beta in a Japanese population [letter].** *Hum Mol Genet* 1996, **5**:2068.
- S65. Hill MR, James AL, Faux JA, Ryan G, Hopkin JM, le Souef P, Musk AW, Cookson WO: **Fc epsilon RI-beta polymorphism and risk of atopy in a general population sample.** *BMJ* 1995, **311**:776-779.
- S66. Hill MR, Cookson WO: **A new variant of the beta subunit of the high-affinity receptor for immunoglobulin E (Fc epsilon RI-beta E237G): associations with measures of atopy and bronchial hyper-responsiveness.** *Hum Mol Genet* 1996, **5**:959-962.
- S67. Ishizawa M, Shibasaki M, Yokouchi Y, Noguchi E, Arinami T, Yamakawa-Kobayashi K, Matsui A, Hamaguchi H: **No association between atopic asthma and a coding variant of Fc epsilon R1 beta in a Japanese population.** *J Hum Genet* 1999, **44**:308-311.
- S68. Palmer LJ, Rye PJ, Gibson NA, Moffatt MF, Goldblatt J, Burton PR, Cookson WO, Lesouef PN: **Association of Fc epsilon R1-beta polymorphisms with asthma and associated traits in Australian asthmatic families.** *Clin Exp Allergy* 1999, **29**:1555-1562.
- S69. Cox HE, Moffatt MF, Faux JA, Walley AJ, Coleman R, Trembath RC, Cookson WO, Harper JI: **Association of atopic dermatitis to the beta subunit of the high affinity immunoglobulin E receptor.** *Br J Dermatol* 1998, **138**:182-187.

- S70. Hizawa N, Yamaguchi E, Jinushi E, Kawakami Y: **A common FCER1B gene promoter polymorphism influences total serum IgE levels in a Japanese population.** *Am J Respir Crit Care Med* 2000, **161**:906-909.
- S71. Fryer AA, Bianco A, Hepple M, Jones PW, Strange RC, Spiteri MA: **Polymorphism at the glutathione S-transferase GSTP1 locus. A new marker for bronchial hyperresponsiveness and asthma.** *Am J Respir Crit Care Med* 2000, **161**:1437-1442.
- S72. Gao PS, Mao XQ, Roberts MH, Arinobu Y, Akaiwa M, Enomoto T, Dake Y, Kawai M, Sasaki S, Hamasaki N, Izuhara K, Shirakawa T, Hopkin JM: **Variants of STAT6 (signal transducer and activator of transcription 6) in atopic asthma.** *J Med Genet* 2000, **37**:380-382.
- S73. Hayden C, Pereira E, Rye P, Palmer L, Gibson N, Palenque M, Hagel I, Lynch N, Goldblatt J, Lesouef P: **Mutation screening of interferon-gamma (IFN γ) as a candidate gene for asthma.** *Clin Exp Allergy* 1997, **27**:1412-1416.
- S74. Wechsler ME, Grasmann H, Deykin A, Silverman EK, Yandava CN, Israel E, Wand M, Drazen JM: **Exhaled nitric oxide in patients with asthma. Association with nos1 genotype.** *Am J Respir Crit Care Med* 2000, **162**:2043-2047.
- S75. Moffatt MF, Schou C, Faux JA, Cookson WO: **Germline TCR-A restriction of immunoglobulin E responses to allergen.** *Immunogenetics* 1997, **46**:226-230.
- S76. Mao XQ, Shirakawa T, Yoshikawa T, Yoshikawa K, Kawai M, Sasaki S, Enomoto T, Hashimoto T, Furuyama J, Hopkin JM, Morimoto K: **Association between genetic variants of mast-cell chymase and eczema.** *Lancet* 1996, **348**:581-583.
- S77. Mao XQ, Shirakawa T, Enomoto T, Shimazu S, Dake Y, Kitano H, Hagihara A, Hopkin JM: **Association between variants of mast cell chymase gene and serum IgE levels in eczema.** *Hum Hered* 1998, **48**:38-41.
- S78. Kawashima T, Noguchi E, Arinami T, Kobayashi K, Otsuka F, Hamaguchi H: **No evidence for an association between a variant of the mast cell chymase gene and atopic dermatitis based on case-control and haplotype-relative-risk analyses.** *Hum Hered* 1998, **48**:271-274.
- S79. Pascale E, Tarani L, Meglio P, Businco L, Battiloro E, Cimino-Reale G, Verna R, D'Ambrosio E: **Absence of association between a variant of the mast cell chymase gene and atopic dermatitis in an Italian population.** *Hum Hered* 2001, **51**:177-179.
- S80. Tanaka K, Sugiura H, Uehara M, Sato H, Hashimoto-Tamaoki T, Furuyama J: **Association between mast cell chymase genotype and atopic eczema: comparison between patients with atopic eczema alone and those with atopic eczema and atopic respiratory disease.** *Clin Exp Allergy* 1999, **29**:800-803.
- S81. Hershey GK, Friedrich MF, Esswein LA, Thomas ML, Chatila TA: **The association of atopy with a gain-of-function mutation in the alpha subunit of the interleukin-4 receptor.** *N Engl J Med* 1997, **337**:1720-1725.
- S82. Rosa-Rosa L, Zimmermann N, Bernstein JA, Rothenberg ME, Khurana Hershey GK: **The R576 IL-4 receptor alpha allele correlates with asthma severity.** *J Allergy Clin Immunol* 1999, **104**:1008-1014.
- S83. Patuzzo C, Trabetti E, Malerba G, Martinati LC, Boner AL, Pescolliderung L, Zanoni G, Pignatti PF: **No linkage or association of the IL-4Ralpha gene Q576R mutation with atopic asthma in Italian families.** *J Med Genet* 2000, **37**:382-384.
- S84. Oiso N, Fukai K, Ishii M: **Interleukin 4 receptor alpha chain polymorphism Gln551Arg is associated with adult atopic dermatitis in Japan.** *Br J Dermatol* 2000, **142**:1003-1006.
- S85. Mitsuyasu H, Izuhara K, Mao XQ, Gao PS, Arinobu Y, Enomoto T, Kawai M, Sasaki S, Dake Y, Hamasaki N, Shirakawa T, Hopkin JM: **Ile50Val variant of IL4R alpha upregulates IgE synthesis and associates with atopic asthma.** *Nat Genet* 1998, **19**:119-120.
- S86. Noguchi E, Shibasaki M, Arinami T, Takeda K, Yokouchi Y, Kobayashi K, Imoto N, Nakahara S, Matsui A, Hamaguchi H: **No association between atopy/asthma and the Ile50Val polymorphism of IL-4 receptor.** *Am J Respir Crit Care Med* 1999, **160**:342-345.