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Fine-mapping of IgE-associated loci 1q23, 5q31, and 12q13 using 1000 Genomes Project data

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asthma; atopy; fine-mapping; genetic polymorphisms; immunoglobulin E.

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Abstract

Background: Genome-wide association studies (GWAS) repeatedly identified 1q23 (*FCERIA*), 5q31 (*RAD50-IL13* and *IL4*), and 12q13 (*STAT6*) as major susceptibility loci influencing the regulation of total serum IgE levels. As GWAS may be insufficient to capture causal variants, we performed fine-mapping and re-genotyping of the three loci using 1000 Genomes Project datasets.

Methods: Linkage disequilibrium tagging polymorphisms and polymorphisms of putative functional relevance were genotyped by chip technology (24 polymorphisms) or MALDI-TOF-MS (40 polymorphisms) in at least 1303 German children (651 asthmatics). The effect of polymorphisms on total serum IgE, IgE percentiles, and atopic diseases was assessed, and a risk score model was applied for gene-by-gene interaction analyses. Functional effects of putative causal variants from these three loci were studied *in silico*.

Results: Associations from GWAS were confirmed and extended. For 1q23 and 5q31, the majority of associations were found with mild to moderately elevated IgE levels, while in the 12q13 locus, single-nucleotide polymorphisms (SNPs) were associated with strongly elevated IgE levels. Gene-by-gene interaction analyses suggested that the presence of mutations in all three loci increases the risk for elevated IgE up to fourfold.

Conclusion: This fine-mapping study confirmed previous associations and identified novel associations of SNPs in 1q23, 5q31, and 12q13 with different levels of serum IgE and their concomitant contribution to IgE regulation.

Immunoglobulin E (IgE) mediates allergy and elevated serum IgE levels are associated with asthma, allergic rhinitis, and atopic dermatitis (eczema) (1). Genetic factors are important in determining IgE levels in blood (2), and genome-wide association studies (GWAS) have identified major susceptibility loci on chromosomes 1q23, 5q31, and 12q13 (3–5). The

source of the association signal from 1q23 seems to be *FCERIA*, a gene encoding the α -chain of the high-affinity receptor for IgE (Fc ϵ RI α) (2) expressed on human mast cells and basophils (6). Chromosome 5q31 harbors genes coding for IL-4, IL-13, and RAD50 (3). IL-4 and IL-13 are cytokines expressed mostly by type 2 helper (Th2) cells that bind

to their B-cell receptors to promote IgE synthesis through class switching (7, 8). A function of the DNA repair protein RAD50 in the context of IgE-mediated allergy has not yet been established (2). The association signal on chromosome 12q13 derives from *STAT6*, a gene encoding the STAT6 transcription factor, which migrates to the B-cell nucleus upon activation where it binds to DNA regions stimulating IgE production (9–11).

Genotyping arrays used in GWAS can capture major association loci but, at the same time, may be insufficient to identify causal variants from these loci (12). Thus, we performed a systematic fine-mapping and re-genotyping of total serum IgE association signals from 1q23, 5q31, and 12q13 based on comprehensive data from the 1000 Genomes Project (13). Furthermore, we investigated whether top-associated single-nucleotide polymorphisms (SNPs) from 1q23, 5q31, and 12q13 interact to affect IgE levels. Finally, the SNPs in these fine-mapped loci were further studied for associations with allergic disorders, such as asthma, atopy, atopic dermatitis, and allergic rhinitis.

Methods

Subjects and genotyping

In our previous GWAS on childhood asthma, 1422 children of German/Austrian ethnicity (728 asthmatics) were chip-genotyped by Illumina Sentrix HumanHap300 BeadChip and had complete information on disease status for asthma available (14). This dataset was a part of the GABRIEL Consortium meta-analyses (4). The majority of asthmatic cases (655) derived from the Multicenter Asthma Genetic in Childhood Study (MAGICS) (14, 15). Further 73 asthmatic children and all age- and sex-matched controls (694) were randomly selected from the large ($n = 5629$) cross-sectional International Study of Asthma and Allergy in Childhood, phase II (ISAAC II) (15, 16) of the same ethnic background. The very similar study protocols of MAGICS and ISAAC II were approved by the respective ethics committees, and written informed consent was obtained from the parents of every child from both studies. Table S1 describes the MAGICS and ISAAC II populations.

Continuous quality control and an update of consent in a follow-up study led to a decrease in the number of subjects to 1303 (651 asthmatics) for whom chip-genotyping results were available for the purposes of the present study. In addition, 151 children for whom not enough DNA for GWAS genotyping had been available or who had been recruited later in MAGICS were also included for genotyping of novel SNPs by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS) (17). As a result, 1454 subjects (763 asthmatics) were included in the fine-mapping analyses of the 1q23, 5q31, and 12q13 loci.

SNPs selection, genotyping, and quality control

A total of 464 SNPs/mutations of 1q23, 5q31, and 12q13 loci were identified in the 1000 Genomes CEU dataset ([ftp://ftp.](ftp://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/)

[1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/](ftp://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/)) (13). To systematically select SNPs of interest for our fine-mapping analyses, we applied a linkage disequilibrium (LD)-based tagging SNP approach for polymorphisms with minor allele frequency (MAF) $\geq 10\%$ ($n = 202$), leading to the identification of 55 tagging SNPs, from which putative false-positive SNP calls from the 1000 Genomes Project pilot dataset were excluded. We also incorporated 1000 Genomes variants not covered by tagging SNPs such as nonsynonymous coding mutations and SNPs with previously identified functional effects. Single-nucleotide polymorphisms not yet covered by the previous genotyping (14, 17) were genotyped by MALDI-TOF (successfully in 21 of 29 SNPs with a call rate of $>90\%$ and no deviation from Hardy-Weinberg Equilibrium). Additionally, variants in the investigated loci genotyped by us previously for different purposes were also incorporated. Thus, 64 SNPs were analyzed in the three loci. Details of SNPs selection are given in Figs S1–S3 and Data S1.

Total serum IgE and atopic sensitization testing

Total serum IgE was measured by the IMMULITE 2000 system (Siemens Healthcare Diagnostics, Eschborn, Germany). Allergic sensitization to common aeroallergens was tested in MAGICS by determining specific serum IgE using CLA-1 (Hitachi Chemical Diagnostics, Berkshire, UK) or Allergy Screen (Mediawiss Analytic, Moers, Germany) panels. In ISAAC II, atopic sensitization to typical inhalant allergens was assessed by skin prick testing and/or by Sx1 assay (ImmunoCAP; Phadia, Freiburg, Germany) (16). For details, please refer to Data S1.

Qualitative phenotypes under investigation

Asthma, atopic asthma, and nonatopic asthma were analyzed in a case-control setting based on subjects from MAGICS and ISAAC II (*see above*). The presence of both asthma and atopy was considered as atopic asthma. Nonatopic asthma was defined as the presence of asthma in the absence of atopy. Both asthma subphenotypes were compared to the control group of ISAAC II subjects without asthma and atopy. Atopic dermatitis, allergic rhinitis, and atopy were studied in subjects randomly selected from the cross-sectional ISAAC II cohort and compared to individuals without the respective condition (14, 15). Definitions of phenotypes are described in detail in Data S1.

Statistical analyses

All LD calculations including tagging procedure were conducted using Haploview (version 4.2) software (18). All LD values presented here correspond to the square of the correlation coefficient (r^2). In case of binary traits, additive genetic effects were modeled by logistic regression with odds ratios (ORs) showing the effect size as an output. The genotype effects on total serum IgE levels as a quantitative trait were analyzed in an age- and sex-adjusted additive model using

linear regression with the magnitude of the effect expressed by estimates (standard error). Before entering the model, IgE values were log-transformed for their approximation to normality. To investigate combined effects of top IgE-associated SNPs from 1q23, 5q31, and 12q13 on IgE levels, gene-by-gene interaction analyses were conducted using a risk score model (19) including the most strongly associated SNP from each region. The determination of risk alleles in dominant or recessive models was first applied to assign a risk value of either one or zero, corresponding to the presence or absence of genetic risk, respectively. Risk values were then summed up to compare samples with the same risk result to the reference population with a risk score of zero. Risk score effects were assessed by logistic regression analysis with adjustment for age and sex. Statistical analyses for single SNPs were carried out by Plink (version 1.07; <http://pngu.mgh.harvard.edu/~purcell/plink/>) (20) and by R (version 3.0.1; <http://cran.r-project.org/>) software for risk score model.

In silico prediction of function

Allele-specific effects of associated SNPs on gene function were studied *in silico* by Alibaba software (version 2.1; <http://www.gene-regulation.com/pub/programs/alibaba2/index.html>) for variants located upstream of the transcription start sites, Human Splicing Finder software (version 2.4.1; <http://www.umd.be/HSF/>) for intronic polymorphisms, SIFT software (<http://sift.bii.a-star.edu.sg/>) for mutations in coding regions, and RegRNA software (release 1.0, January 2006; <http://regrna.mbc.nctu.edu.tw/html/about.html>) for SNPs located in the 3'-untranslated regions. The results were further investigated with F-SNP software that combines SNP and gene function information from 16 databases (<http://compbio.cs.queensu.ca/F-SNP/>).

Results

LD structure

Twenty-six SNPs genotyped in 1q23 locus formed thirteen tagging bins (called further 'BINs') comprising 1–4 SNPs (Fig. S4A). In chromosome 5q31, 23 genotyped polymorphisms grouped in seventeen BINs of 1–5 SNPs (Fig. S4B). Finally, fifteen SNPs whose genotypes were available for 12q13 locus created thirteen BINs of 1–3 polymorphisms (Fig. S4C).

Association of 1q23, 5q31, and 12q13 SNPs with total serum IgE levels

To expand and dissect the association signal from previous GWAS and candidate studies on IgE levels, we performed fine-mapping of 1q23, 5q13, and 12q13 loci using the 1000 Genomes dataset (information based on deep sequencing) (13). In the 1q23 locus, top associations with IgE levels were identified for rs2427837 and rs2251746 (both BIN-4) (Tables 1 and S2). This locus has been explored by us and

others in detail (3, 5, 17, 21). However, the SNPs not analyzed previously, rs2427830 (BIN-13, *OR10J3*/downstream of *FCER1A*) and rs56366265 (BIN-8, intron of *FCER1A*), were also linked to elevated IgE levels (Table 1 and Fig. S4). In our IgE percentile analyses, the majority of associations in 1q23 locus were found with mildly-to-moderately elevated IgE levels (50th and 66th percentiles), as previously reported (17). Unlike rs56366265, which follows the pattern of the majority of 1q23 SNPs, rs2427830 showed an association with extreme IgE levels (90th percentile). Applying functional *in silico* prediction, rs2427830 'T' allele showed binding site for C/EBP beta transcription factor, while rs56366265 'A' allele demonstrated a potential to modify splicing events (Table S3). Beyond GWAS, here we identified two additional putatively functional SNPs, rs2427830 and rs56366265, which may also modify IgE levels.

In the 5q31 locus, moderate association signals with IgE levels were observed from many different SNPs residing in different tagging BINs. Thus, no peak association signal could be defined. The three IgE association top signal SNPs, rs3798135, rs2706347, and rs7737470 from 1st GWAS on IgE levels (3), were tagged by IgE-associated SNP rs2244012 (BIN-4, intron of *RAD50*), whereas the fourth top-associated SNP from the previous GWAS was covered in our present study by rs1881457. Some additional, significant association signals were observed for rs2243300 (BIN-14) and rs4426908 (BIN-17). Furthermore, rs739719 (BIN-2, downstream of *IL5*) and rs4426908 (BIN-17, downstream of *IL4*) were linked to IgE levels in this post-GWAS analysis for the first time (Table 1). *In silico* function prediction suggested that rs2244012 may have the potential to modify transcriptional regulation and its 'G' allele may alter splicing events (Table S3).

The peak IgE association signal from the 12q13 locus was confirmed for rs1059513 (BIN-6), which is in line with previous results from GWAS and candidate gene studies (5, 11). Intriguingly, we found association signals in 12q13 predominantly with extreme IgE levels, which is different from signals in 1q23 and 5q31. Only rs1059513 showed an association with mildly-to-moderately elevated IgE levels (50th and 66th percentiles) in the complete cohort (Table 1).

To further dissect the IgE association signals, IgE levels were analyzed in asthmatics and nonasthmatics separately (Tables 2 and S4). A strong IgE association signal in asthmatics was observed for rs2427830 (BIN-13 of the 1q23 locus). In the 5q31 locus, association with IgE levels was almost exclusively seen in nonasthmatics. The association of rs1059513 (BIN-6 of the 12q13 locus) with IgE levels has already been reported (11), but here we additionally found that IgE association signal is mainly driven by asthmatics.

Gene-by-gene interaction effects on total serum IgE levels

Next, combined effects of the three loci on total serum IgE levels were analyzed. For this purpose, one SNP showing the most significant association with serum IgE levels per locus (rs2427837, rs2243300, and rs1059513) was selected for the

Table 1 Polymorphisms in 1q23, 5q31, and 12q13 loci associated with total serum IgE levels

Chr	SNP	BIN	Minor allele	Total IgE in complete cohort*		50th percentile† (IgE cutoff 60.4 IU/ml)		66th percentile‡ (IgE cutoff 115 IU/ml)		90th percentile§ (IgE cutoff 457 IU/ml)	
				Estimate (SE)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
1q23	rs10489854	1	T	0.27 (0.14)	0.058	1.51 (1.00–2.28)	0.048	1.73 (1.18–2.54)	0.005	1.08 (0.68–1.71)	0.748
1q23	rs2511214	2	G	0.14 (0.06)	0.027	1.16 (0.97–1.38)	0.098	1.08 (0.92–1.27)	0.360	1.1 (0.90–1.35)	0.347
1q23	rs2427836	3	C	–0.15 (0.06)	0.016	0.85 (0.71–1.01)	0.064	0.87 (0.73–1.02)	0.094	0.91 (0.74–1.12)	0.380
1q23	rs2494264	3	A	–0.18 (0.06)	0.005	0.82 (0.69–0.98)	0.029	0.85 (0.72–1.00)	0.056	0.9 (0.74–1.10)	0.308
1q23	rs2494251	3	A	–0.16 (0.06)	0.014	0.85 (0.71–1.01)	0.066	0.88 (0.74–1.04)	0.133	0.9 (0.73–1.11)	0.318
1q23	rs2427837	4	A	–0.32 (0.07)	4.37 × 10^{–6}	0.65 (0.53–0.78)	7.02 × 10^{–6}	0.67 (0.56–0.81)	2.73 × 10^{–5}	0.74 (0.59–0.94)	0.014
1q23	rs2251746	4	C	–0.30 (0.07)	3.54 × 10^{–5}	0.66 (0.54–0.80)	3.56 × 10^{–5}	0.69 (0.57–0.83)	1.43 × 10^{–4}	0.77 (0.60–0.98)	0.037
1q23	rs7522607	5	C	0.19 (0.09)	0.028	1.16 (0.91–1.47)	0.237	1.10 (0.88–1.38)	0.390	1.23 (0.94–1.62)	0.126
1q23	rs12565775	6	C	0.21 (0.11)	0.048	1.38 (1.01–1.88)	0.043	1.49 (1.12–1.99)	0.006	1.12 (0.80–1.57)	0.517
1q23	rs7548864	6	A	0.23 (0.11)	0.037	1.62 (1.16–2.25)	0.004	1.56 (1.16–2.10)	0.004	1.08 (0.76–1.55)	0.660
1q23	rs12119226	6	T	0.18 (0.11)	0.093	1.40 (1.02–1.92)	0.039	1.48 (1.11–1.99)	0.008	1.03 (0.73–1.47)	0.862
1q23	rs56366265	8	A	0.12 (0.08)	0.152	1.27 (1.00–1.62)	0.045	1.30 (1.04–1.63)	0.019	1.22 (0.94–1.59)	0.139
1q23	rs16842010	10	C	0.15 (0.12)	0.231	1.30 (0.90–1.87)	0.158	1.44 (1.03–2.01)	0.034	1.09 (0.73–1.62)	0.686
1q23	rs2427829	11	A	0.15 (0.07)	0.030	1.09 (0.90–1.31)	0.387	1.10 (0.92–1.31)	0.315	1.16 (0.94–1.45)	0.168
1q23	rs2427830	13	T	0.22 (0.07)	0.004	1.18 (0.96–1.47)	0.122	1.17 (0.96–1.43)	0.125	1.43 (1.14–1.81)	0.002
5q31	rs739719	2	A	–0.27 (0.13)	0.040	0.84 (0.59–1.19)	0.330	0.77 (0.55–1.10)	0.150	0.68 (0.42–1.10)	0.118
5q31	rs2244012	4	G	0.15 (0.07)	0.036	1.19 (0.97–1.46)	0.093	0.99 (0.82–1.19)	0.895	1.13 (0.90–1.43)	0.297
5q31	rs6871536	4	C	0.15 (0.07)	0.045	1.17 (0.96–1.44)	0.125	0.99 (0.81–1.19)	0.880	1.15 (0.91–1.45)	0.253
5q31	rs1295686	12	T	0.15 (0.08)	0.048	1.23 (0.99–1.52)	0.066	1.05 (0.86–1.29)	0.635	0.99 (0.77–1.28)	0.969
5q31	rs20541	12	A	0.16 (0.08)	0.047	1.25 (1.00–1.55)	0.049	1.05 (0.85–1.29)	0.665	1.01 (0.78–1.31)	0.920
5q31	rs2243204	13	T	0.23 (0.11)	0.041	1.41 (1.02–1.95)	0.035	1.06 (0.79–1.42)	0.688	1.25 (0.88–1.77)	0.215
5q31	rs2243300	14	T	0.26 (0.11)	0.025	1.48 (1.06–2.06)	0.020	1.04 (0.77–1.40)	0.794	1.4 (0.99–1.99)	0.057
5q31	rs4426908	17	C	0.14 (0.07)	0.033	1.25 (1.04–1.51)	0.019	1.18 (0.99–1.41)	0.069	1.03 (0.83–1.28)	0.773
12q13	rs1059513	6	C	–0.34 (0.10)	0.001	0.63 (0.48–0.83)	8.38 × 10^{–4}	0.58 (0.44–0.77)	1.26 × 10^{–4}	0.77 (0.54–1.10)	0.153
12q13	rs703817	7	T	0.07 (0.06)	0.288	1.02 (0.86–1.22)	0.810	1.09 (0.92–1.29)	0.306	1.24 (1.00–1.52)	0.048
12q13	rs324011	11	T	0.06 (0.07)	0.410	1.03 (0.86–1.24)	0.749	1.05 (0.88–1.25)	0.577	1.29 (1.04–1.60)	0.022
12q13	rs167769	11	T	0.08 (0.07)	0.200	1.07 (0.89–1.28)	0.475	1.08 (0.91–1.29)	0.363	1.31 (1.05–1.62)	0.015
12q13	rs12368672	11	G	0.11 (0.06)	0.076	1.11 (0.93–1.32)	0.244	1.15 (0.98–1.36)	0.094	1.25 (1.02–1.53)	0.030

Chr, chromosome; SNP, single-nucleotide polymorphism; BIN, tagging bin; MAF, minor allele frequency; SE, standard error; CI, confidence interval.

Only SNPs showing at least one significant association in any of the four analyses are given here. For the complete results, see Table S2.

P-values are adjusted for age and gender; if < 0.05, they are considered to be significant (bold).

Number of individuals with nonmissing phenotype involved in the respective calculations for the different rounds of genotyping:

*total IgE complete cohort: MALDI-TOF MS (1261), CHIP (1145);

†50th: MALDI-TOF MS (827 cases/434 controls), CHIP (740 cases/405 controls);

‡66th: MALDI-TOF MS (633 cases/628 controls), CHIP (561 cases/584 controls); §90th: MALDI-TOF MS (253 cases/1008 controls), CHIP (221 cases/924 controls).

Table 2 Associations between polymorphisms and IgE levels as a quantitative variable in the complete cohort, asthmatics, or nonasthmatics

Chr	SNP	BIN	MAF	Total IgE in complete cohort*		Asthmatics†		Nonasthmatics‡	
				Estimate (SE)	P-value	Estimate (SE)	P-value	Estimate (SE)	P-value
1q23	rs2511214	2	0.48	0.14 (0.06)	0.027	0.16 (0.09)	0.066	0.14 (0.08)	0.072
1q23	rs2427836	3	0.47	-0.15 (0.06)	0.016	-0.16 (0.09)	0.082	-0.16 (0.08)	0.042
1q23	rs2494264	3	0.46	-0.18 (0.06)	0.005	-0.21 (0.09)	0.015	-0.19 (0.08)	0.019
1q23	rs2494251	3	0.45	-0.16 (0.06)	0.014	-0.20 (0.09)	0.025	-0.14 (0.08)	0.086
1q23	rs2427837	4	0.27	-0.32 (0.07)	4.37 × 10⁻⁶	-0.26 (0.10)	0.006	-0.32 (0.09)	2.17 × 10⁻⁴
1q23	rs2251746	4	0.27	-0.30 (0.07)	3.54 × 10⁻⁵	-0.22 (0.10)	0.037	-0.32 (0.09)	3.17 × 10⁻⁴
1q23	rs61828219	5	0.17	0.15 (0.08)	0.069	0.28 (0.11)	0.011	0.01 (0.10)	0.904
1q23	rs3845625	5	0.15	0.15 (0.08)	0.087	0.25 (0.11)	0.031	-0.03 (0.11)	0.809
1q23	rs7549785	5	0.16	0.15 (0.09)	0.085	0.40 (0.12)	0.002	0.00 (0.11)	0.968
1q23	rs7522607	5	0.17	0.19 (0.09)	0.028	0.30 (0.12)	0.013	0.07 (0.11)	0.540
1q23	rs12565775	6	0.09	0.21 (0.11)	0.048	0.16 (0.15)	0.278	0.26 (0.14)	0.055
1q23	rs7548864	6	0.09	0.23 (0.11)	0.037	0.08 (0.16)	0.621	0.33 (0.14)	0.019
1q23	rs2427829	11	0.28	0.15 (0.07)	0.030	0.24 (0.09)	0.010	0.06 (0.09)	0.482
1q23	rs2427830	13	0.21	0.22 (0.07)	0.004	0.35 (0.10)	6.14 × 10⁻⁴	0.05 (0.10)	0.619
5q31	rs739719	2	0.07	-0.27 (0.13)	0.040	-0.16 (0.20)	0.415	-0.32 (0.16)	0.042
5q31	rs2244012	4	0.25	0.15 (0.07)	0.036	0.10 (0.11)	0.354	0.17 (0.09)	0.055
5q31	rs6871536	4	0.25	0.15 (0.07)	0.045	0.08 (0.11)	0.427	0.16 (0.09)	0.074
5q31	rs1881457	4	0.24	0.15 (0.08)	0.056	0.04 (0.11)	0.718	0.25 (0.09)	0.009
5q31	rs1800925	4	0.24	0.15 (0.08)	0.050	0.08 (0.11)	0.460	0.21 (0.09)	0.028
5q31	rs72797364	6	0.08	-0.19 (0.12)	0.103	-0.08 (0.17)	0.633	-0.32 (0.15)	0.029
5q31	rs1295686	12	0.23	0.15 (0.08)	0.048	0.11 (0.11)	0.317	0.13 (0.10)	0.188
5q31	rs20541	12	0.22	0.16 (0.08)	0.047	0.10 (0.11)	0.370	0.13 (0.10)	0.200
5q31	rs2243204	13	0.09	0.23 (0.11)	0.041	0.29 (0.16)	0.067	0.15 (0.14)	0.283
5q31	rs2243300	14	0.08	0.26 (0.11)	0.025	0.36 (0.16)	0.028	0.12 (0.14)	0.386
5q31	rs4426908	17	0.29	0.14 (0.07)	0.033	0.14 (0.09)	0.127	0.11 (0.09)	0.207
12q13	rs1059513	6	0.10	-0.34 (0.10)	0.001	-0.38 (0.14)	0.008	-0.29 (0.13)	0.023
12q13	rs12368672	11	0.40	0.11 (0.06)	0.076	0.20 (0.09)	0.023	0.00 (0.08)	0.955

Only SNPs showing at least one significant association in any of the three analyses are given here. For the complete results, see Table S4. P-values are adjusted for age and gender; if < 0.05, they are considered to be significant (bold).

Chr, chromosome; SNP, single-nucleotide polymorphism; BIN, tagging bin; MAF, minor allele frequency; SE, standard error.

Number of individuals with nonmissing phenotype involved in the respective calculations for the different rounds of genotyping:

*total IgE complete cohort: MALDI-TOF MS (1261), CHIP (1145);

†total IgE asthmatics: MALDI-TOF MS (602), CHIP (517);

‡total IgE nonasthmatics: MALDI-TOF MS (651), CHIP (620).

Table 3 Chance of having elevated IgE levels with regard to risk score based on the number of risk genotypes

Risk score	N	Total IgE in complete cohort		50th percentile (IgE cutoff 60.4 IU/ml)		66th percentile (IgE cutoff 115 IU/ml)		90th percentile (IgE cutoff 457 IU/ml)	
		Estimate (SE)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
0	94	0	–	1	–	1	–	1	–
1 vs 0	517	0.43 (0.18)	0.017	1.82 (1.13–2.94)	0.015	1.63 (0.98–2.69)	0.059	1.27 (0.65–2.49)	0.484
2 vs 0	562	0.74 (0.17)	2.47 × 10⁻⁵	2.89 (1.78–4.70)	1.76 × 10⁻⁵	2.54 (1.55–4.17)	2.32 × 10⁻⁴	1.58 (0.82–3.03)	0.172
3 vs 0	89	0.94 (0.26)	5.23 × 10⁻⁴	4.30 (2.15–8.59)	3.79 × 10⁻⁵	2.63 (1.39–4.99)	0.003	2.37 (1.07–5.25)	0.033

The statistical analysis was conducted by linear or logistic regression analysis adjusting for age and sex.

P-values < 0.05 are considered to be significant (bold).

N, number of subjects; SE, standard error; CI, confidence interval.

analyses in a previously developed risk score model (19). While the presence of one risk genotype from any of these SNPs may lead to elevated serum IgE levels, carriers of all

three risk genotypes have an up to fourfold risk of elevated IgE levels when compared to subjects without any risk genotype (Tables 3 and S5).

Association of 1q23, 5q31, and 12q13 SNPs with asthma, atopic asthma, and nonatopic asthma

Only rs1295686 and rs20541 (BIN-12 of the 5q31 locus) were weakly associated with asthma. Different SNPs were found to correlate specifically with atopic asthma in 1q23, 5q31, and 12q13 loci (Table S6). Notably, rs2427830 in this list has never been reported to be linked with atopic asthma. Furthermore, none of the SNPs under investigation were associated with nonatopic asthma.

Association of 1q23, 5q31, and 12q13 SNPs with allergic rhinitis, atopic dermatitis, or atopy

Association between SNPs and atopic dermatitis, allergic rhinitis and atopy was explored in the cross-sectional ISAAC II study only (Table S7). In the 1q23 locus, associations with atopy and allergic rhinitis were found for several SNPs. Association signals with atopy were identified for rs3024979 (intron of *NAB2*) and rs2598483 (upstream of *STAT6*) of 12q13 locus, of which the first may modify splicing events (Table S3). None of the SNPs were associated with atopic dermatitis.

Discussion

GWAS is a powerful tool to identify novel loci associated with certain traits (22). However, even if followed by a classical HapMap-based fine-mapping and imputation, it cannot always identify real causal variants but rather their LD-based proxies. Therefore, we performed a very detailed, sequencing-based fine-mapping of three loci consistently associated with total serum IgE levels in GWAS, that is, chromosomes 1q23, 5q31, and 12q13 (3–5). For this purpose, we extracted the information on all mutations in the targeted regions from the 1000 Genomes Project dataset and applied a tagging approach for those with a considerable frequency ($MAF \geq 10\%$) (23, 24). In order to trap rare but putative functional variants irrespective of MAF, we also incorporated all 1000 Genomes amino acid-changing variants and SNPs previously shown to be functional (11) but not covered by our tagging. Additionally, in case of the 5q31 loci, we included also variants in the 5q31 locus control region (LCR) situated in the *RAD50* gene region (25), which could potentially affect the expression of Th2 cytokine genes *IL5*, *IL13* and *IL4* located nearby as well as *RAD50* itself (2). By identifying novel, potentially regulatory SNPs associated with total serum IgE and by applying more detailed analyses of association (percentile analyses and gene-by-gene interaction), this study not only reconfirms but also extends findings of previous GWAS (3–5).

As only already known association signals were investigated in more detail, no correction for multiple testing was applied. We acknowledge that this reasoning is only valid for total serum IgE and asthma, and therefore, results with other phenotypes have to be interpreted with caution. In our fine-mapping of the 1q23 locus, the strongest IgE association signal overall in the whole cohort was observed for rs2427837 and rs2251746 (both SNPs from BIN-4), which is in line with previous GWAS findings (3, 5). In the percentile-based analy-

ses, strong associations between SNPs rs56366265 and rs2427830 (from two independent BINs) and IgE were reported for the first time. Single-nucleotide polymorphism rs2427830 was associated with IgE at the 90th IgE percentile, which may correspond to its selective association with IgE observed in asthmatics (having higher IgE levels than nonasthmatics). The asthma-specific IgE signal of this SNPs seems especially interesting in view of its associations with atopic asthma, which may hint to mechanisms of atopic asthma development in childhood as suggested by Holt and Sly (26, 27).

Considering the location of a SNP rs2427830 downstream of *FCER1A* and also its close neighbor, *OR10J3*, the association between this polymorphism and serum IgE might be difficult to explain from the functional point of view, even though similar effects have already been reported for other genetic variants in the region (5, 17). One possible explanation might be that the observed association results from a complex interplay between several identified and yet not identified functional *FCER1A* SNPs. In addition, a SNP rs2427830 by, for example, its effect on C/EBP binding might itself affect *OR10J3* transcriptional activity, which could in turn influence serum IgE by yet unknown mechanism. It cannot be excluded that increased (general) inflammatory responses related to genetic effects also exerted by the locus (28) could partly contribute to the elevation of IgE levels, too.

A functional *IL13* variant (rs20541), reported to be a major source of the 5q31-derived IgE association signal by GWAS (4, 5), was also associated with IgE levels in our analyses. Further variants from *IL13* have been previously reported to also influence IgE levels (5, 29–32). However, in our fine-mapping study, it would be difficult to determine a peak association signal within this locus because several SNPs from different BINs showed associations of comparable effect size and significance. In a previous GWAS (3), the top association signal in chromosome 5q31 was observed for SNPs in *RAD50*. Here, we also observed *RAD50* SNPs, rs6871536 and rs2244012, to be associated with serum IgE levels. Single-nucleotide polymorphism rs6871536 is located in the distal region of *RAD50*, which at the same time is a crucial element of the 5q31 LCR, regulating and coordinating transcription of the 5q31 locus Th2 cytokine-expressing genes, such as *IL4* and *IL13* (2). Another IgE-associated *RAD50* SNP, rs2244012, localizes outside of the LCR, suggesting that *RAD50* SNPs might affect IgE levels also directly by yet unknown mechanisms. Taken together, fine-mapping of the 5q31 suggests that different SNPs from the locus influence serum IgE levels, potentially affecting more than one pathomechanism in IgE regulation.

The strongest IgE signal from 12q13 locus was observed for SNP rs1059513. Interestingly, several other SNPs showed only effects when extreme IgE values (90th percentile) were analyzed, suggesting a role in disease-related mechanisms rather than effect on basal IgE levels. This information was missed so far in nonpercentile analyses. The most interesting group of SNPs associated with highly elevated IgE consisted mostly of one BIN, comprising rs12368672 and rs167769 (associated with IgE in previous GWAS (3–5)) and SNP rs324011 which we had studied functionally for its effect on *STAT6* expression (33).

In addition to applying a percentile-based analysis of IgE in combination with classical continuous data analysis to 1000 Genomes data for fine-mapping, we performed a gene-by-gene interaction analysis combining one strongly associated SNP from each locus. An accumulation of risk genotypes from the three loci increases the risk for elevated IgE levels in a stepwise manner, especially for mild to moderate IgE percentiles (50th or 66th). As a statistical risk score model was applied, biological causality or biological interaction between SNPs cannot be deduced directly. Also, these interaction data need to be interpreted with caution because the population is rather small for this type of analyses.

In the present study, only SNPs rs1295686 and rs20541 from BIN-12 of the 5q31 locus showed an association with asthma, which is in line with the findings of some previous GWAS (4, 34). None of the SNPs in any of the three loci demonstrated an association with atopic dermatitis, which could suggest the involvement of other than IgE-affecting genetic factors in its pathogenesis (35). While chromosome 5q31 was associated with neither atopy nor allergic rhinitis, several SNPs in 1q23 and 12q13 loci demonstrated an association with the former or the latter. In most of the cases, SNPs associated with atopy showed at least a tendency for an association with allergic rhinitis and *vice versa*, suggesting a partial overlap of the genetic background in those traits.

In summary, this study confirms and extends findings of previous GWAS. Several novel associations with total IgE levels were found in the three loci (rs2427830, rs56366265, rs739719, and rs4426908). The use of percentile analysis suggests that genetic variants in these loci determine not only basal IgE levels but may also influence IgE responses related to the development of atopic diseases. Gene-by-gene interaction analyses suggested that the presence of mutations in all three loci increases the risk for elevated IgE. This knowledge may help to dissect the complexity of IgE effects in asthma and allergy and to study SNP effects in the regions more specifically in the future.

Author contributions

Vishwas Sharma contributed to the study design, data interpretation, bioinformatics analysis and wrote the first draft of manuscript; Sven Michel contributed to genotyping and writing of the manuscript, performed statistical analyses and data interpretation; Vincent Gaertner contributed to statistical analysis; Andre Franke, Sonja Zeilinger, Thomas Illig, and Michaela Schedel carried out MALDI-TOF MS genotyping; Christian Vogelberg, Andrea von Berg, Albrecht Bufer, Andrea Heinzmann, Otto Laub, Ernst Rietschel, Burkhard Simma, Thomas Frischer, and Jon Genuneit contributed to data collection; Daniel P. Potaczek and Michael Kabesch developed the study design, participated in data analysis and interpretation, and wrote the final version of the manuscript.

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Conflicts of interest

The authors declare that they have no conflicts of interest.

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1. Selection process of polymorphisms from chromosome 1q23.

Figure S2. Selection process of polymorphisms from chromosome 5q31.

Figure S3. Selection process of polymorphisms from chromosome 12q13.

Figure S4. Depiction of IgE-associated loci 1q23 (A), 5q31 (B), and 12q13 (C) with exon–intron structure of the genes, rs numbers of genotyped polymorphisms, tagging bins (in brackets), and pairwise linkage disequilibrium (r^2).

Table S1. Descriptive statistics of the populations used for the analysis.

Table S2. Associations between polymorphisms and total serum IgE levels as a quantitative variable (A) or the risk of having total serum IgE concentration above the certain percentiles (B–D) in the complete cohort.

Table S3. *In silico* functional analysis of SNPs associated with total serum IgE levels.

Table S4. Associations between polymorphisms and IgE levels as a quantitative variable in the complete cohort (A), asthmatics (B), or nonasthmatics (C).

Table S5. Creation of risk scores for gene-by-gene interaction analysis as previously established (14); top-associated SNPs of 1q23, 5q31, and 12q13 loci and the risk of elevated total serum IgE levels in the complete cohort were investigated.

Table S6. Associations between polymorphisms and asthma or its subphenotypes.

Table S7. Associations with atopic sensitization, allergic rhinitis, or atopic dermatitis.

Data S1. Detailed description of the study methodology, including study populations, atopy testing, characteristics of clinical phenotypes investigated, and selection of polymorphisms for genotyping.

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Fine mapping of IgE associated loci 1q23, 5q31 and 12q13 using 1000 Genomes Project data

[Supplement Data S1. Detailed description s-of the study methodology, including study populations, atopy testing IgE-testing, characteristics of clinical phenotypes investigated, and SNPs selection of polymorphisms for genotyping](#)

Description of the **source of study** populations

Commento [DPP1]: Or "study source"

Subjects investigated in this work derived from the Multicenter Asthma Genetic in Childhood Study (MAGICS) and the German arm of the International Study of Asthma and Allergies in Childhood, phase II (ISAAC II) (1, 2). In MAGICS, 1255 children were recruited between 2001 and 2007 in seven different clinical asthma centres from Germany or Austria. In ISAAC II, a large cross-sectional study conducted in 1995 and 1996, 5629 school children aged between 9 and 11 years were recruited in Munich and Dresden (Germany), as described in more details elsewhere (3, 4). Only children of German descent were included in subsequent genetic analyses. Both studies were approved by the respective ethics committees. Written informed consent was obtained from the parents of all the participating children.

Testing for atopic sensitization

Allergic sensitization against typical aero-allergens, such as *D. pteronyssinus*, *D. farinae*, cat epithelium, grass pollen and birch pollen, was tested in MAGICS by determination of specific serum IgE using CLA-1 (Hitachi Chemical Diagnostics, [Berkshire, Eschbort](#), UK) or AllergyScreen (Mediawiss Analytic, [Moers](#), Germany) panels. In ISAAC II, atopic sensitization was assessed by skin prick testing against common inhalant allergens, including *D. pteronyssinus*, *D. farinae*, *Alternaria tenuis*, cat dander, mixed grass and mixed tree pollen, and/ or using Sx1 assay (ImmunoCAP, [Phadia](#), [Freiburg](#), Germany) containing a cocktail of antibodies against typical

aero-allergens, such as local grass pollen, rye pollen, birch pollen, mugwort pollen, *D. pteronyssinus*, cat dander, dog dander and *Cladosporium herbarum*.

Definitions of the clinical phenotypes

In MAGICS, asthma was diagnosed in 865 children (mean age 11 years) by pediatric pulmonologists based on physical examination, clinical history, lung function tests, and allergy status. In ISAAC II, children were considered asthmatic if their parents reported a physician's diagnosis of asthma once or recurrent spastic or asthmatic bronchitis more than once. Both MAGICS and ISAAC II subjects were included in a case-control analysis of asthma, atopic asthma and non-atopic asthma conducted in the present work. Atopic asthma was equal to the co-existence of asthma and atopy, the latter defined by the positive results for specific IgE against at least one of allergens tested (RAST/ CAP class equivalent of >2) in MAGICS and the positive specific IgE results based on Sx1 testing (RAST/ CAP class of >2) in ISAAC II. The presence of asthma without an accompanying atopy was considered to define non-atopic asthma. "Non-atopic" status definition was based on negative results for all specific IgE tested (RAST/ CAP class equivalent of 0) in MAGICS, and negative skin prick tests (a wheal reaction <3 mm after subtraction of the negative control for all tested allergens) and Sx1 (RAST/CAP class of 0) in ISAAC II.

Atopic dermatitis, allergic rhinitis and atopy in general were studied in the cross-sectional set of subjects randomly selected from ISAAC II, as described in the main document. Atopic dermatitis and allergic rhinitis were defined by the doctor's diagnosis report in the parental questionnaire. Atopy was defined as a positive skin prick test result to any of the allergens tested (a wheal reaction ≥ 3 mm after subtraction of the negative control). Individuals having any of those three phenotypes were analyzed in comparison to those negative to atopic dermatitis, allergic rhinitis or atopy, respectively.

Details on the selection of polymorphisms for genotyping

To fine-map 1q23, 5q31 and 12q13 loci in detail, we obtained the data of all single-nucleotide polymorphisms (SNPs)/ mutations in the regions of interest (chromosome 1: 157509000-157555000 for 1q23 locus, chromosome 5: 131900000-132051400 for 5q31 locus and chromosome 12: 55765600-55800146 for 12q13 locus) from the 1000 Genomes [Project](http://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/) dataset for the CEU population ([ftp://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/](http://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/)) (5). In total, 464 SNPs (1q23: 134 SNPs; 5q31: 264 SNPs; and 12q13: 66 SNPs) were identified in the regions of interest, to which we applied: (I) a linkage disequilibrium (LD) -based tagging approach for SNPs with minor allele frequency $\geq 10\%$ (1q23: 37 SNPs; 5q31: 138 SNPs; and 12q13: 27 SNPs), and also incorporated variants not covered by tagging, including (II) non-synonymous coding mutations (1q23: 1 SNP; and 5q31: 1 SNP), (III) SNPs in the IInd fragment of *RAD50*-located 5q31 locus control region (LCR) hypersensitivity site (RHS) 5 of (5q31: 1 SNP) due to its functional relevance for the regulation of *IL13* and *IL4* gene expression (6-8), and (IV) previously identified functional SNPs (12q13: 1 SNP) (9). For pairwise tagging, criteria of Hardy-Weinberg Equilibrium (HWE) > 0.05 and $r^2 > 0.8$ were applied. A set of 55 tagging SNPs (1q23: 11 SNPs; 5q31: 28 SNPs; and 12q13: 16 SNPs) was identified by Haploview software (version 4.2; <http://www.broadinstitute.org/scientific-community/science/programs/medical-and-population-genetics/haploview/haploview>). Tagging (I) or other (II-IV) SNPs already covered by the previous rounds of genotyping, i.e. GWAS Illumina Sentrix HumanHap300 BeadChip chip-genotyping (1q23: 5 SNPs; 5q31: 9 SNPs; and 12q13: 6 SNPs) (1), subsequent (HapMap-based) matrix-assisted-laser-desorption/ionization-time-of-flight-mass-spectrometry (MALDI-TOF-MS) re-genotyping of 1q23 (9 SNPs) (10) or 5q31 (4 SNPs) (2), and additional, ~~also those, yet unpublished;~~ MALDI-TOF-MS genotyping data available to us (1q23: 2 SNPs; ~~5q31: 4 SNPs;~~ and 12q13: 1 SNPs), were removed from the genotyping list. Additionally, SNPs suspected to be false positive calls of 1000 [Genomes Project](http://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/) dataset-, such as X > Y or Z > X (Y) mutations on polyX/polyY borders, directly neighboring variants, SNPs presumed to derive from paralogous region, and

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otherwise suspected SNPs according to our own estimation and validation information from dbSNP NCBI database of short genomic variations (<http://www.ncbi.nlm.nih.gov/projects/SNP/>), were excluded from ~~re~~-genotyping list (5q31: 9 SNPs; and 12q13: 2 SNPs). Finally, 29 SNPs remained in a MALDI-TOF-MS re-genotyping list (1q23: 6 SNPs; 5q31: 13 SNPs; and 12q13: 10 SNPs), 21 of which (1q23: 4 SNPs; 5q31: 9 SNPs; and 12q13: 8 SNPs) were successfully genotyped. In addition to those 21 polymorphisms, 43 SNPs from the previous rounds of genotyping, including GWAS chip-genotyping (1q23: 8 SNPs; 5q31: 10 SNPs; and 12q13: 6 SNPs) or MALDI-TOF genotyping (1q23: 14 SNPs; 5q31: 4 SNPs; and 12q13: 1 SNP) were available for the statistical testing of the three loci making a total number of analyzed SNPs of 64. Selection process of the investigated SNPs is presented in ~~Supplementary Figures IS1, I and IIS3~~. Details on the methodology of genotyping, including primers and all the additional procedures of handling the samples could be obtained from the authors on request.

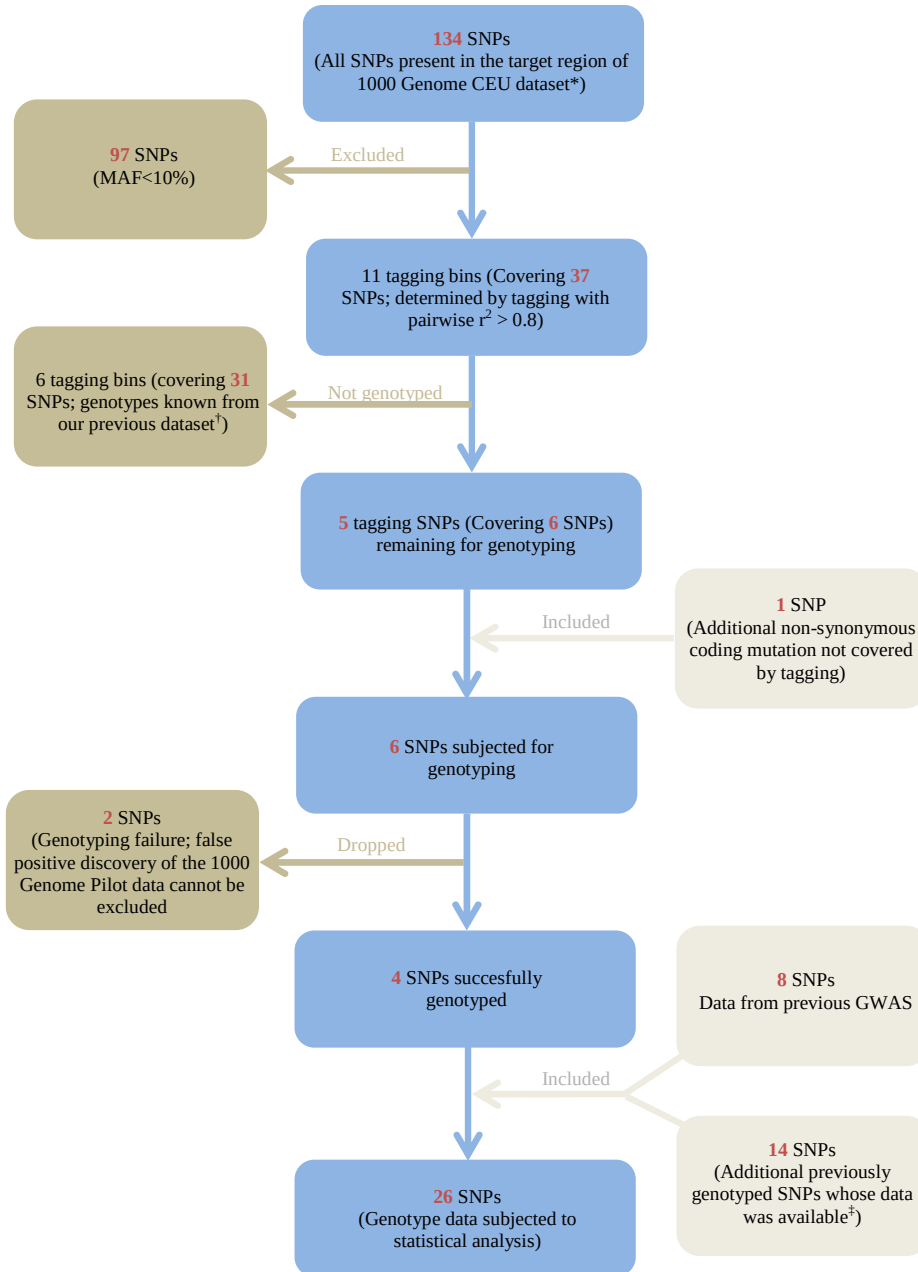
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Supplementary Figure S11. Selection process of polymorphisms from chromosome 1q23

1q23 region covered in the study (Chromosome 1: 157509000-157555000)

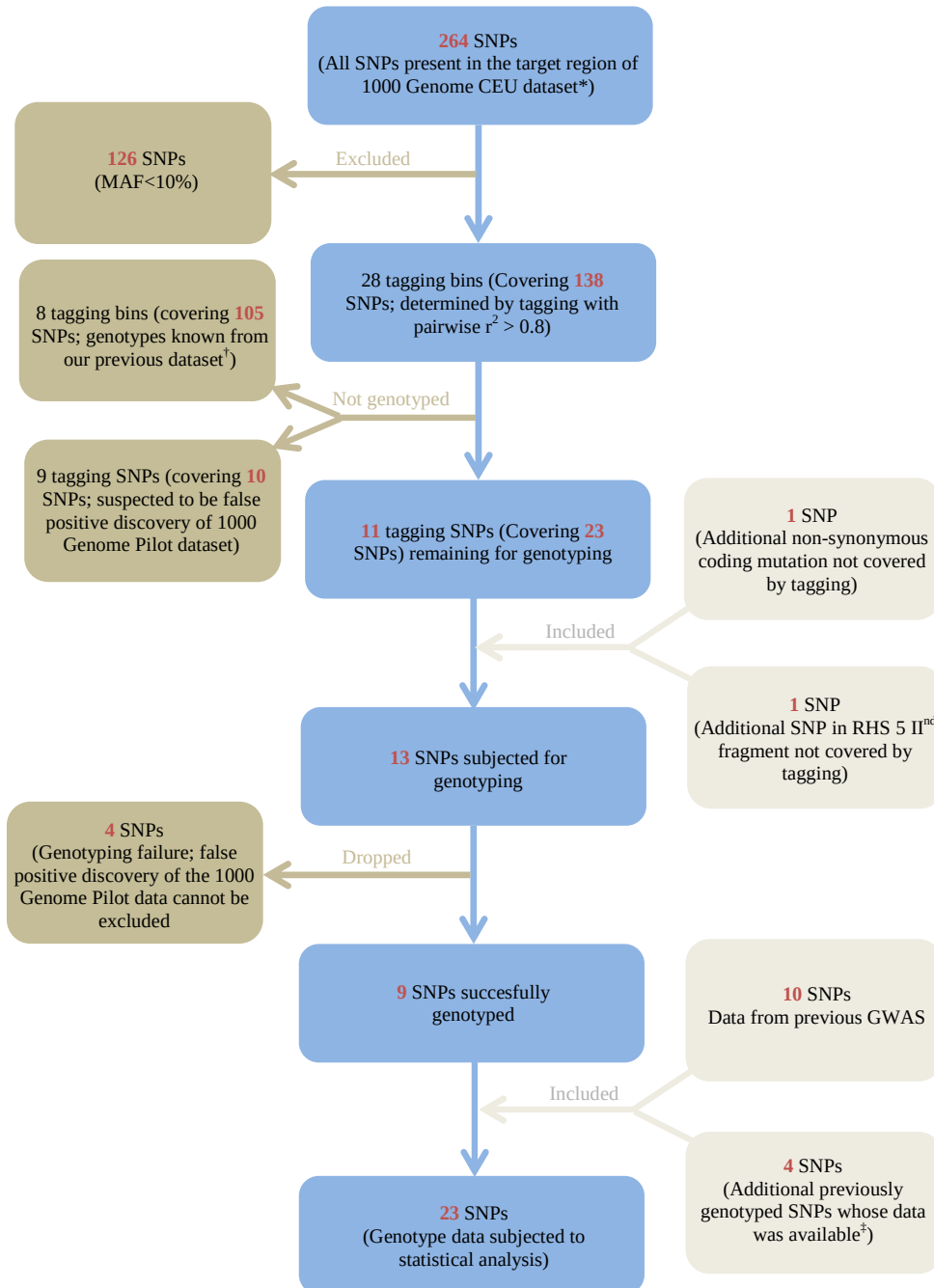


* ftp://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/ (5)

† Previous genotyping in our cohort includes here (a) the genome-wide association study on childhood asthma (1), (b) the study by Potaczek *et al.* (10) and (c) yet unpublished data

‡ From (a) the study by Potaczek *et al.* (10) and (b) yet unpublished data

Abbreviations: SNP, single-nucleotide polymorphism; MAF, minor allele frequency

Supplementary Figure HS2. Selection process of polymorphisms from chromosome 5q315q31 region covered in the study (Chromosome 5: 131900000-132051400)

* ftp://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/ (5)

† Previous genotyping in our cohort includes here (a) the genome-wide association study on childhood asthma (1) and (b) the study by Michel *et al.* 2010 (2)

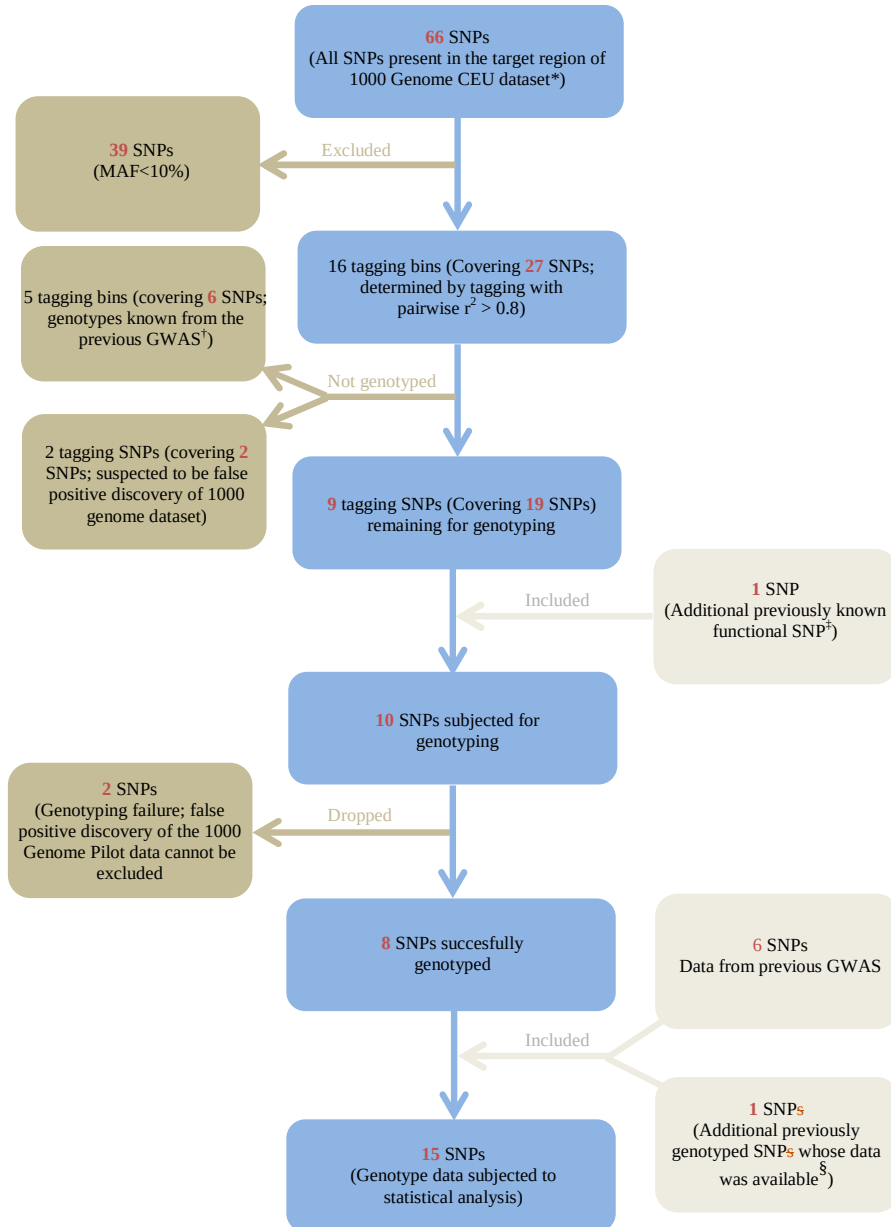
‡ From the study by Michel *et al.* 2010 (2)

Abbreviations: RHS; *RAD50* hypersensitive site; otherwise, see ~~Supplementary~~ Figure ~~4S1~~

Supplementary Figure HHS3. Selection process of polymorphisms from chromosome 12q13

Commento [u2]: Changes are also made in this Figure itself: in the right box on the bottom (two changes).

12q13 region covered in the study (Chromosome 12: 55765600-55800146)



* ftp://ftp.1000genomes.ebi.ac.uk/vol1/ftp/pilot_data/release/2010_07/ (5)

† Previous genotyping in our cohort includes here the genome-wide association study on childhood asthma (1)

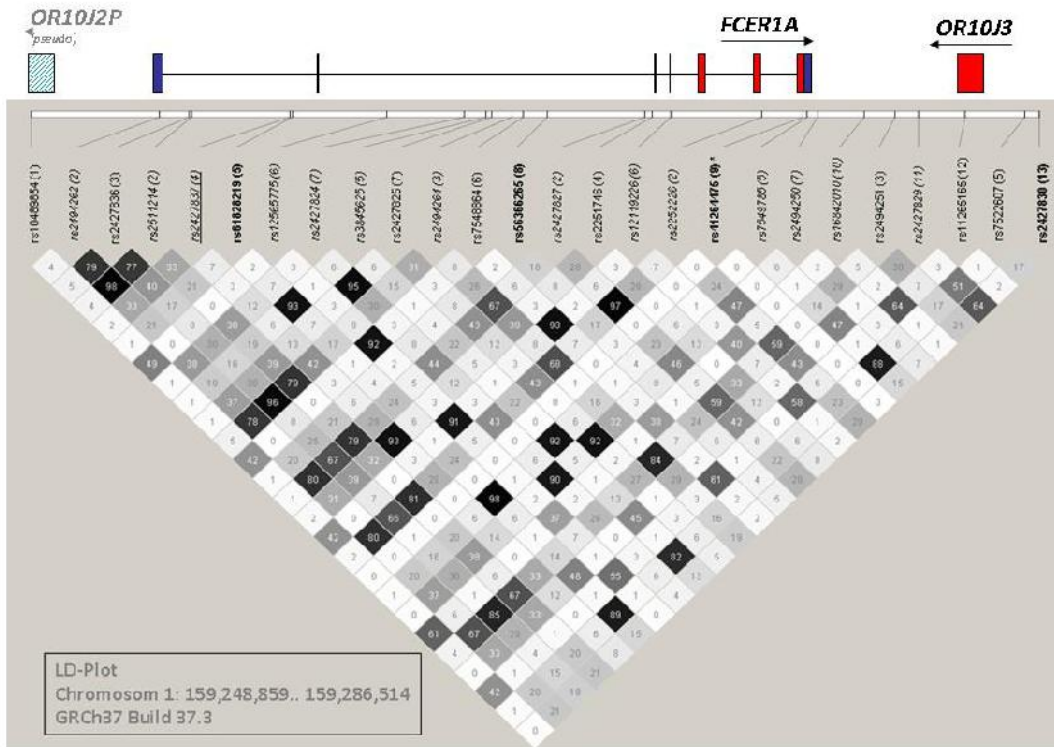
‡ Schedel *et al.* (11)

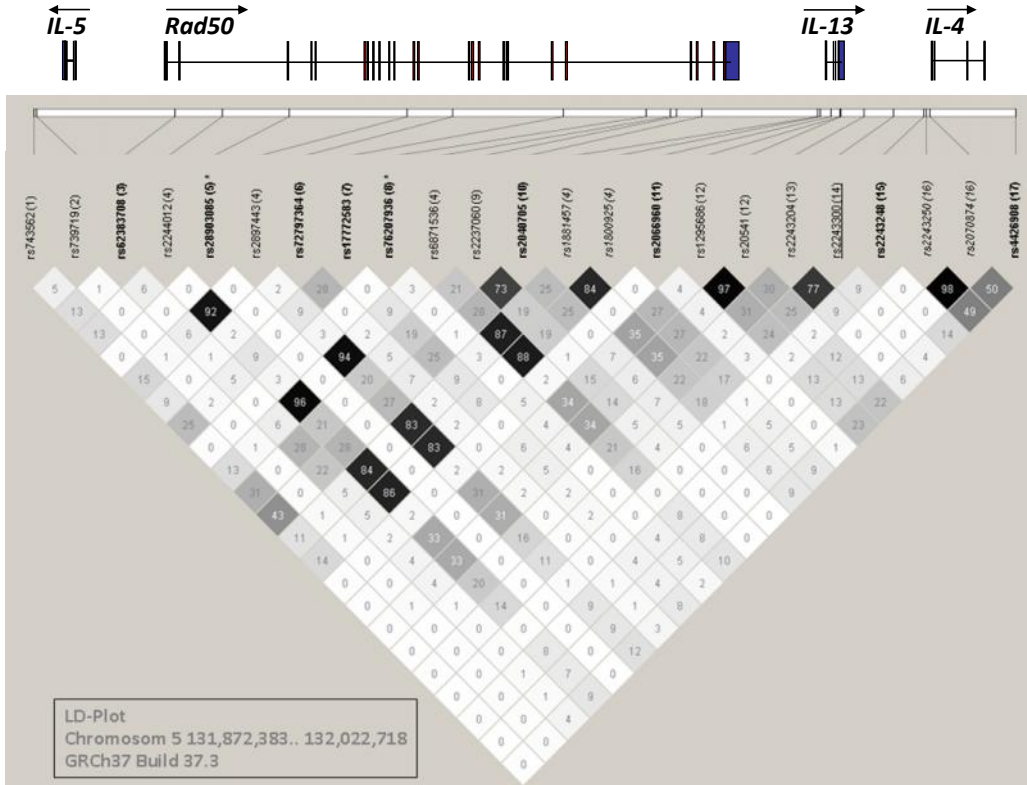
§ From yet unpublished data

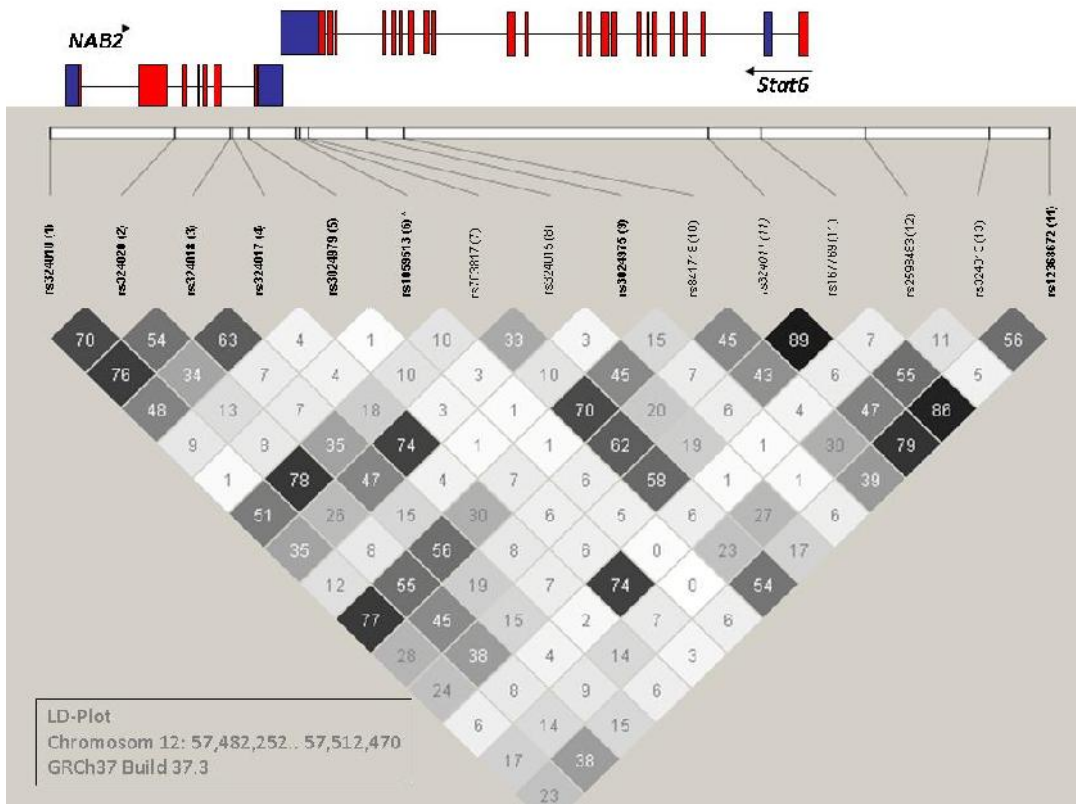
Abbreviations: see ~~Supplementary~~ Figure ~~IS1~~

Supplementary Figure IVS4. Depiction of IgE associated loci 1q23 ([aA](#)), 5q31 ([bB](#)) and 12q13 ([cC](#)) with exon-intron structure of the genes, rs numbers of genotyped polymorphisms, tagging bins (in brackets) and pairwise linkage disequilibrium (r^2). In r^2 plot, numbers represent r^2 values expressed as a percentage of its maximal value (1.0); shading of the squares represents r^2 magnitude with a black-to-white gradient showing highest (1.0) to lowest (0.0) r^2 value. In the gene scheme, red boxes correspond to coding exons, blue boxes to untranslated regions, and green oblique-stripped box to pseudogene.

Newly genotyped SNPs by MALDI-TOF-MS are given in bold. The SNPs genotyped by Illumina Sentrix HumanHap300 BeadChip in our previous GWAS (1) whose data are utilized in the present study are in normal text. The SNPs previously genotyped by MALDI-TOF-MS ((2, 10) [and including from](#) our unpublished data) are in italic. Top associated IgE signal among all the studied SNPs in the gene locus is underlined. Variants selected based on their (putative) functionality, i.e. non-synonymous coding mutations and SNP in *RAD50* hypersensitive site 5 not covered by tagging approach are marked with a star. Tagging bins are given in brackets after rs number.







Supplementary Table IS1. Descriptive statistics of the populations used for the analysis

	MAGICS	ISAAC II
Male sex, n* (%)	457/691 (66.1)	386/763 (50.6)
Age, mean (SD)	10.99 (2.90)	9.63 (0.59)
Asthmatic, n* (%)	691/691 (100)	72/755 (9.54)
Atopic sensitization, n* (%)	505/598 (84.4)	194/651 (29.8)
Total serum IgE levels, median (Q1-Q3)	260 (91-611)	69 (23-188)

* Number affected/ number with data available

Abbreviations: SD, standard deviation; for acronyms of the studies, please, see plain text.

Supplementary Table HS2. Associations between polymorphisms and total serum IgE levels as a quantitative variable (A) or the risk of having total serum IgE concentration above the certain percentiles (B-D) in the complete cohort

Chr	SNP	BIN	Minor allele	MAF	Source of genotyping	Total IgE in complete cohort (A)		50 th percentile (B)		66 th percentile (C)		90 th percentile (D)	
						Estimate (SE)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
1q23	rs10489854	1	T	0.06	CHIP	0.27 (0.14)	0.058	1.51 (1.00-2.28)	0.048	1.73 (1.18-2.54)	0.005	1.08 (0.68-1.71)	0.748
1q23	rs2494262	2	C	0.48	MALDI-TOF MS	0.12 (0.06)	0.057	1.14 (0.96-1.36)	0.148	1.06 (0.89-1.25)	0.521	1.07 (0.87-1.31)	0.540
1q23	rs2511214	2	G	0.48	MALDI-TOF MS	0.14 (0.06)	0.027	1.16 (0.97-1.38)	0.098	1.08 (0.92-1.27)	0.360	1.1 (0.90-1.35)	0.347
1q23	rs2427827	2	T	0.44	MALDI-TOF MS	0.11 (0.06)	0.069	1.12 (0.94-1.33)	0.206	1.04 (0.88-1.23)	0.627	1.07 (0.87-1.31)	0.540
1q23	rs2252226	2	C	0.45	MALDI-TOF MS	0.09 (0.06)	0.136	1.09 (0.91-1.29)	0.341	1.00 (0.85-1.18)	0.964	1.04 (0.85-1.27)	0.704
1q23	rs2427836	3	C	0.47	CHIP	-0.15 (0.06)	0.016	0.85 (0.71-1.01)	0.064	0.87 (0.73-1.02)	0.094	0.91 (0.74-1.12)	0.380
1q23	rs2494264	3	A	0.46	MALDI-TOF MS	-0.18 (0.06)	0.005	0.82 (0.69-0.98)	0.029	0.85 (0.72-1.00)	0.056	0.9 (0.74-1.10)	0.308
1q23	rs2494251	3	A	0.45	CHIP	-0.16 (0.06)	0.014	0.85 (0.71-1.01)	0.066	0.88 (0.74-1.04)	0.133	0.9 (0.73-1.11)	0.318
1q23	rs2427837	4	A	0.27	MALDI-TOF MS	-0.32 (0.07)	4.37*10⁻⁶	0.65 (0.53-0.78)	7.02*10⁻⁶	0.67 (0.56-0.81)	2.73*10⁻⁵	0.74 (0.59-0.94)	0.014
1q23	rs2251746	4	C	0.27	CHIP	-0.30 (0.07)	3.54*10⁻⁵	0.66 (0.54-0.80)	3.56*10⁻⁵	0.69 (0.57-0.83)	1.43*10⁻⁴	0.77 (0.60-0.98)	0.037
1q23	rs61828219	5	A	0.17	MALDI-TOF MS	0.15 (0.08)	0.069	1.10 (0.88-1.38)	0.406	1.08 (0.87-1.33)	0.487	1.14(0.88-1.47)	0.318
1q23	rs3845625	5	T	0.15	MALDI-TOF MS	0.15 (0.08)	0.087	1.15 (0.91-1.47)	0.248	1.11 (0.89-1.39)	0.343	1.11(0.85-1.46)	0.429
1q23	rs7549785	5	A	0.16	MALDI-TOF MS	0.15 (0.09)	0.085	1.10 (0.86-1.40)	0.442	1.07 (0.85-1.34)	0.572	1.2(0.92-1.57)	0.187
1q23	rs7522607	5	C	0.17	CHIP	0.19 (0.09)	0.028	1.16 (0.91-1.47)	0.237	1.10 (0.88-1.38)	0.390	1.23 (0.94-1.62)	0.126
1q23	rs12565775	6	C	0.09	MALDI-TOF MS	0.21 (0.11)	0.048	1.38 (1.01-1.88)	0.043	1.49 (1.12-1.99)	0.006	1.12 (0.80-1.57)	0.517
1q23	rs7548864	6	A	0.09	CHIP	0.23 (0.11)	0.037	1.62 (1.16-2.25)	0.004	1.56 (1.16-2.10)	0.004	1.08 (0.76-1.55)	0.660
1q23	rs12119226	6	T	0.08	MALDI-TOF MS	0.18 (0.11)	0.093	1.40 (1.02-1.92)	0.039	1.48 (1.11-1.99)	0.008	1.03(0.73-1.47)	0.862
1q23	rs2427824	7	T	0.26	MALDI-TOF MS	-0.03 (0.07)	0.622	0.96 (0.79-1.16)	0.649	0.91 (0.75-1.09)	0.300	0.97 (0.77-1.21)	0.765
1q23	rs2427825	7	T	0.27	CHIP	-0.03 (0.07)	0.712	0.94 (0.78-1.14)	0.547	0.92 (0.76-1.11)	0.367	0.95 (0.75-1.20)	0.640
1q23	rs2494250	7	G	0.28	MALDI-TOF MS	0.02 (0.07)	0.719	1.05 (0.87-1.27)	0.635	0.99 (0.83-1.19)	0.928	1 (0.80-1.25)	0.993
1q23	rs56366265	8	A	0.18	MALDI-TOF MS	0.12 (0.08)	0.152	1.27 (1.00-1.62)	0.045	1.30 (1.04-1.63)	0.019	1.22 (0.94-1.59)	0.139
1q23	rs41264475	9	A	0.01	MALDI-TOF MS	0.30 (0.32)	0.356	1.39 (0.53-3.64)	0.501	1.78 (0.73-4.31)	0.202	1.71 (0.69-4.27)	0.248
1q23	rs16842010	10	C	0.06	MALDI-TOF MS	0.15 (0.12)	0.231	1.30 (0.90-1.87)	0.158	1.44 (1.03-2.01)	0.034	1.09 (0.73-1.62)	0.686
1q23	rs2427829	11	A	0.28	MALDI-TOF MS	0.15 (0.07)	0.030	1.09 (0.90-1.31)	0.387	1.10 (0.92-1.31)	0.315	1.16 (0.94-1.45)	0.168

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Chr	SNP	BIN	Minor allele	MAF	Source of genotyping	Total IgE in complete cohort (A)		50 th percentile (B)		66 th percentile (C)		90 th percentile (D)	
						Estimate (SE)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
1q23	rs11265165	12	C	0.09	CHIP	0.09 (0.11)	0.444	1.26 (0.91-1.75)	0.166	1.31 (0.96-1.77)	0.084	1.06 (0.73-1.54)	0.763
1q23	rs2427830	13	T	0.21	MALDI-TOF MS	0.22 (0.07)	0.004	1.18 (0.96-1.47)	0.122	1.17 (0.96-1.43)	0.125	1.43 (1.14-1.81)	0.002
5q31	rs743562	1	T	0.45	CHIP	0.05 (0.06)	0.407	1.07 (0.89-1.27)	0.476	1.02 (0.86-1.21)	0.821	1.04 (0.84-1.28)	0.740
5q31	rs739719	2	A	0.07	CHIP	-0.27 (0.13)	0.040	0.84 (0.59-1.19)	0.330	0.77 (0.55-1.10)	0.150	0.68 (0.42-1.10)	0.118
5q31	rs62383708	3	T	0.16	MALDI-TOF MS	-0.10 (0.09)	0.241	0.86 (0.68-1.09)	0.207	0.88 (0.70-1.10)	0.270	0.84 (0.63-1.12)	0.234
5q31	rs2244012	4	G	0.25	CHIP	0.15 (0.07)	0.036	1.19 (0.97-1.46)	0.093	0.99 (0.82-1.19)	0.895	1.13 (0.90-1.43)	0.297
5q31	rs2897443	4	T	0.24	CHIP	0.11 (0.07)	0.125	1.14 (0.93-1.40)	0.204	0.95 (0.79-1.16)	0.627	1.08(0.85-1.37)	0.546
5q31	rs6871536	4	C	0.25	CHIP	0.15 (0.07)	0.045	1.17 (0.96-1.44)	0.125	0.99 (0.81-1.19)	0.880	1.15(0.91-1.45)	0.253
5q31	rs1881457	4	C	0.24	MALDI-TOF MS	0.15 (0.08)	0.056	1.24 (1.00-1.54)	0.051	0.97 (0.79-1.19)	0.760	1.07 (0.83-1.37)	0.594
5q31	rs1800925	4	T	0.24	MALDI-TOF MS	0.15 (0.08)	0.050	1.16 (0.94-1.44)	0.170	0.97 (0.79-1.19)	0.780	1.19 (0.93-1.52)	0.163
5q31	rs28903085	5	C	0.004	MALDI-TOF MS	-0.16 (0.44)	0.715	0.57 (0.18-1.83)	0.345	0.78 (0.24-2.53)	0.681	0.92 (0.20-4.29)	0.917
5q31	rs72797364	6	C	0.08	MALDI-TOF MS	-0.19 (0.12)	0.103	0.77 (0.56-1.05)	0.101	0.88 (0.64-1.21)	0.433	0.81 (0.54-1.22)	0.314
5q31	rs17772583	7	G	0.23	MALDI-TOF MS	-0.03 (0.07)	0.662	0.93 (0.76-1.14)	0.475	0.99 (0.81-1.20)	0.904	1.02 (0.80-1.30)	0.865
5q31	rs76207936	8	A	0.01	MALDI-TOF MS	0.36 (0.31)	0.245	1.70 (0.66-4.40)	0.272	1.23 (0.54-2.83)	0.618	0.83 (0.28-2.46)	0.731
5q31	rs2237060	9	G	0.39	CHIP	-0.04 (0.06)	0.528	0.97 (0.81-1.15)	0.710	1.06 (0.90-1.26)	0.479	0.89 (0.72-1.10)	0.281
5q31	rs2040705	10	A	0.46	MALDI-TOF MS	-0.01 (0.06)	0.853	0.99 (0.84-1.18)	0.929	1.12 (0.95-1.32)	0.167	0.93 (0.77-1.14)	0.508
5q31	rs2066960	11	A	0.11	MALDI-TOF MS	0.08 (0.10)	0.429	1.03 (0.79-1.35)	0.826	1.07 (0.83-1.39)	0.587	1.15 (0.84-1.57)	0.375
5q31	rs1295686	12	T	0.23	CHIP	0.15 (0.08)	0.048	1.23 (0.99-1.52)	0.066	1.05 (0.86-1.29)	0.635	0.99 (0.77-1.28)	0.969
5q31	rs20541	12	A	0.22	CHIP	0.16 (0.08)	0.047	1.25 (1.00-1.55)	0.049	1.05 (0.85-1.29)	0.665	1.01 (0.78-1.31)	0.920
5q31	rs2243204	13	T	0.09	CHIP	0.23 (0.11)	0.041	1.41 (1.02-1.95)	0.035	1.06 (0.79-1.42)	0.688	1.25 (0.88-1.77)	0.215
5q31	rs2243300	14	T	0.08	CHIP	0.26 (0.11)	0.025	1.48 (1.06-2.06)	0.020	1.04 (0.77-1.40)	0.794	1.4 (0.99-1.99)	0.057
5q31	rs2243248	15	G	0.07	MALDI-TOF MS	0.06 (0.11)	0.624	1.31 (0.94-1.81)	0.108	1.11 (0.83-1.50)	0.473	0.89 (0.61-1.30)	0.555
5q31	rs2243250	16	T	0.17	MALDI-TOF MS	0.14 (0.09)	0.113	1.15 (0.90-1.47)	0.276	1.10 (0.87-1.38)	0.433	1.06 (0.80-1.41)	0.677

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Chr	SNP	BIN	Minor allele	MAF	Source of genotyping	Total IgE in complete cohort (A)		50 th percentile (B)		66 th percentile (C)		90 th percentile (D)	
						Estimate (SE)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
5q31	rs2070874	16	T	0.18	MALDI-TOF MS	0.13 (0.09)	0.131	1.20 (0.94-1.52)	0.148	1.09 (0.87-1.37)	0.446	1 (0.76-1.33)	0.983
5q31	rs4426908	17	C	0.29	MALDI-TOF MS	0.14 (0.07)	0.033	1.25 (1.04-1.51)	0.019	1.18 (0.99-1.41)	0.069	1.03 (0.83-1.28)	0.773
12q13	rs324010	1	C	0.47	MALDI-TOF MS	0.03 (0.06)	0.631	1.05 (0.88-1.25)	0.567	1.05 (0.89-1.24)	0.568	0.93 (0.76-1.14)	0.482
12q13	rs324020	2	C	0.44	MALDI-TOF MS	0.07 (0.06)	0.278	1.07 (0.90-1.28)	0.426	1.12 (0.95-1.32)	0.181	1.19 (0.97-1.45)	0.098
12q13	rs324018	3	A	0.41	MALDI-TOF MS	0.06 (0.06)	0.357	1.13 (0.95-1.35)	0.166	1.06 (0.90-1.25)	0.509	0.94 (0.77-1.16)	0.585
12q13	rs324017	4	T	0.31	MALDI-TOF MS	0.05 (0.07)	0.463	1.16 (0.96-1.39)	0.130	1.07 (0.89-1.27)	0.474	0.91 (0.73-1.14)	0.409
12q13	rs3024979	5	A	0.09	MALDI-TOF MS	0.03 (0.10)	0.752	0.99 (0.74-1.32)	0.948	1.10 (0.84-1.45)	0.484	0.96 (0.68-1.34)	0.809
12q13	rs1059513	6	C	0.10	MALDI-TOF MS	-0.34 (0.10)	0.001	0.63 (0.48-0.83)	8.4*10⁻⁴	0.58 (0.44-0.77)	1.26*10⁻⁴	0.77 (0.54-1.10)	0.153
12q13	rs703817	7	T	0.50	CHIP	0.07 (0.06)	0.288	1.02 (0.86-1.22)	0.810	1.09 (0.92-1.29)	0.306	1.24 (1.00-1.52)	0.048
12q13	rs324015	8	T	0.25	CHIP	0.00 (0.07)	0.963	1.14 (0.93-1.40)	0.194	0.99 (0.82-1.20)	0.948	0.8 (0.62-1.02)	0.073
12q13	rs3024975	9	T	0.10	MALDI-TOF MS	0.05 (0.10)	0.639	1.04 (0.78-1.39)	0.785	1.02 (0.77-1.34)	0.903	0.99 (0.71-1.39)	0.975
12q13	rs841718	10	G	0.42	CHIP	0.02 (0.07)	0.792	1.09 (0.91-1.30)	0.374	1.05 (0.88-1.24)	0.612	0.89 (0.72-1.11)	0.295
12q13	rs324011	11	T	0.40	MALDI-TOF MS	0.06 (0.07)	0.410	1.03 (0.86-1.24)	0.749	1.05 (0.88-1.25)	0.577	1.29 (1.04-1.60)	0.022
12q13	rs167769	11	T	0.39	CHIP	0.08 (0.07)	0.200	1.07 (0.89-1.28)	0.475	1.08 (0.91-1.29)	0.363	1.31(1.05-1.62)	0.015
12q13	rs12368672	11	G	0.40	MALDI-TOF MS	0.11 (0.06)	0.076	1.11 (0.93-1.32)	0.244	1.15 (0.98-1.36)	0.094	1.25 (1.02-1.53)	0.030
12q13	rs2598483	12	T	0.10	CHIP	0.09 (0.10)	0.403	0.97 (0.73-1.28)	0.810	1.16 (0.89-1.52)	0.272	1.06 (0.77-1.47)	0.711
12q13	rs324013	13	G	0.49	CHIP	-0.04 (0.06)	0.513	0.97 (0.82-1.16)	0.766	1.00 (0.84-1.18)	0.986	0.85 (0.69-1.05)	0.138

Number of individuals with non-missing phenotype involved in the respective calculations for the different rounds of genotyping:

A) Total IgE complete cohort: MALDI-TOF MS (1261), CHIP (1145)

B) 50th percentile: MALDI-TOF MS (827 cases / 434 controls), CHIP (740 cases / 405 controls);

C) 66th percentile: MALDI-TOF MS (633 cases / 628 controls), CHIP (561 cases / 584 controls);

D) 90th percentile: MALDI-TOF MS (253 cases / 1008 controls), CHIP (221 cases / 924 controls);

All statistical analyses include adjustment for age and sex.

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Abbreviations: Chr, chromosome; SNP, single-nucleotide polymorphism; BIN, tagging bin; MAF, minor allele frequency; SE, standard error; CI, confidence interval; for abbreviations of genotyping methods, please, see plain text.

Supplementary Table H3S3. In-silico functional analysis of SNPs associated with total serum IgE levels

Chr	SNP	Minor allele	Location of SNPs*	Putative functional impact	Functional relevance by F-SNP software
1q23	rs10489854	T	Upstream of <i>FCER1A/OR10J2P</i> pseudogene (non-synonymous coding mutation by F-SNP software)	No difference detected ^a	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
1q23	rs2494262	C	Upstream of <i>FCER1A</i>	Oct-1 binds to 'A' but not to 'C' allele ^a	Transcriptional regulation changed by prediction tool TFSearch (FS score 0.5)
1q23	rs2511214	G	Intron of <i>FCER1A</i> (upstream by F-SNP software)	Potential splice site. EIEs new site is found in 'G' but not in 'T' allele ^b	Transcriptional regulation changed by prediction tool TFSearch (FS score 0.5)
1q23	rs2427827	T	Intron of <i>FCER1A</i>	EIEs site broken in 'T' but not in 'C' allele also IIEs new site is found in 'T' allele but not in 'C' allele ^b	Transcriptional regulation changed by prediction tool TFSearch and Consite (FS score 0.208)
1q23	rs2252226	C	Intron of <i>FCER1A</i>	Altered potential branch point and EIEs new site is found in 'C' but not in 'T' allele ^b	Transcriptional regulation changed by prediction tool TFSearch (FS score 0.176)
1q23	rs2427836	C	Intron of <i>FCER1A</i> (upstream by F-SNP software)	EIEs site broken in 'T' but not in 'C' allele and IIEs new site is found in 'T' but not in 'C' allele ^b	No change
1q23	rs2494264	A	Intron of <i>FCER1A</i>	Altered potential branch point. EIEs site broken and new site is found in 'A' but not in 'T' allele ^b	Transcriptional regulation changed/ exist by prediction tool GoldenPath and Consite (FS score 0.109)
1q23	rs2494251	A	Downstream of <i>FCER1A</i>	No suitable prediction program available	No suitable prediction program available
1q23	rs2427837	A	Intron of <i>FCER1A</i> (5' near <i>FCER1A</i> by F-SNP software)	EIEs site broken and new site is found in 'A' but not in 'G' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch, Consite and GoldenPath (FS score 0.268)
1q23	rs2251746	C	Intron of <i>FCER1A</i>	Altered potential branch point. EIEs site broken and new site is found in 'C' but not in 'T' allele and IIEs site broken in 'C' but not in 'T' allele ^b	Transcriptional regulation changed by prediction tool TFSearch and Consite (FS score 0.208)

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Chr	SNP	Minor allele	Location of SNPs*	Putative functional impact	Functional relevance by F-SNP software
1q23	rs61828219	A	Intron of <i>FCER1A</i>	Altered potential branch point and EIEs new site is found in 'A' but not in 'C' allele ^b	No result found
1q23	rs3845625	T	Intron of <i>FCER1A</i>	Altered potential branch point. EIEs site broken in 'T' but not in 'C' allele also IIEs new site is found in 'T' but not in 'C' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch and GoldenPath (FS score 0.242)
1q23	rs7549785	A	3'UTR of <i>FCER1A</i>	Allele specific miRNA binding ^d	Transcriptional regulation changed by prediction tool TFSearch (FS score 0.176)
1q23	rs7522607	C	Upstream of <i>OR10J3</i>	No difference detected ^a	No functional information exists
1q23	rs12565775	C	Intron of <i>FCER1A</i>	EIEs new site is found in 'C' but not in 'A' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch, Consite and GoldenPath (FS score 0.268)
1q23	rs7548864	A	Intron of <i>FCER1A</i>	Altered potential splice site and IIEs site broken in 'A' but not in 'G' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch, Consite and GoldenPath (FS score 0.268)
1q23	rs12119226	T	Intron of <i>FCER1A</i>	Altered potential splice site and EIEs new site is found in 'T' but not in 'C' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch and GoldenPath (FS score 0.315)
1q23	rs2427824	T	Intron of <i>FCER1A</i>	IIEs new site is found in 'T' but not in 'C' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch, Consite and GoldenPath (FS score 0.268)
1q23	rs2427825	T	Intron of <i>FCER1A</i>	No difference detected ^b	Transcriptional regulation changed/ exist by prediction tool Consite and GoldenPath (FS score 0.109)
1q23	rs2494250	G	Downstream of <i>FCER1A</i> (3' near <i>FCER1A</i> by F-SNP software)	No suitable prediction program available	Transcriptional regulation changed by prediction tool TFSearch and Consite (FS score 0.208)
1q23	rs56366265	A	Intron of <i>FCER1A</i>	Altered potential splice site, EIEs and IIEs site broken in 'A' but not in 'G' allele ^b	No result found

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Chr	SNP	Minor allele	Location of SNPs*	Putative functional impact	Functional relevance by F-SNP software
1q23	rs41264475	A	Exon of <i>FCER1A</i>	Altered amino acid from N247K ^c	Splicing regulation changed by prediction tool ESEfinder, ESRSearch and RESCUE ESE (FS score 0.365)
1q23	rs16842010	C	Downstream of <i>FCER1A</i>	No suitable prediction program available	No suitable prediction program available
1q23	rs2427829	A	Downstream of <i>FCER1A</i>	No suitable prediction program available	No suitable prediction program available
1q23	rs11265165	C	Exon of <i>OR10J3</i>	No difference detected ^c	Splicing regulation changed by prediction tool ESEfinder, ESRSearch and PESX (FS score 0.330)
1q23	rs2427830	T	Upstream of <i>OR10J3</i> / downstream of <i>FCER1A</i>	C/EBPbeta binds to 'T' but not to 'G' allele ^a	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
5q31	rs743562	T	Downstream of <i>IL5</i>	No suitable prediction program available	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
5q31	rs739719	A	Downstream of <i>IL5</i>	No suitable prediction program available	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
5q31	rs62383708	T	Intron of <i>RAD50</i>	EIEs site broken in 'T' but not in 'G' allele also IIEs new site is found in 'T' but not in 'G' allele ^b	No result found
5q31	rs2244012	G	Intron of <i>RAD50</i>	EIEs site broken in 'G' but not in 'A' allele also new site is found in 'G' but not in 'A' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch, Consite and GoldenPath (FS score 0.268)
5q31	rs2897443	T	Intron of <i>RAD50</i>	EIEs new site is found in 'A' but not in 'T' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch, Consite and GoldenPath (FS score 0.268)
5q31	rs6871536	C	Intron of <i>RAD50</i>	EIEs new site is found in 'C' but not in 'T' allele ^b	Transcriptional regulation changed/ exist by prediction tool TFSearch and GoldenPath (FS score 0.242)
5q31	rs1881457	C	Upstream of <i>IL13</i>	No difference ^a	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.065)

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Chr	SNP	Minor allele	Location of SNPs*	Putative functional impact	Functional relevance by F-SNP software
5q31	rs1800925	T	Upstream of <i>IL13</i>	ICSBP and C/EBPα binds to 'T' but not to 'C' allele ^a	Transcriptional regulation changed/ exist by prediction tool TFSearch and GoldenPath (FS score 0.242)
5q31	rs28903085	C	Exon of <i>RAD50</i>	Altered amino acid from I94L ^c	Splicing regulation changed by prediction tool ESEfinder, ESRSearch and RescueESE (FS score 0.652)
5q31	rs72797364	C	Intron of <i>RAD50</i>	Altered potential branch point and IIEs new site is found in 'C' but not in 'T' allele ^b	No result found
5q31	rs17772583	G	Intron of <i>RAD50</i>	Altered potential branch point. EIEs site broken in 'G' but not in 'A' allele also new site is found in 'G' but not in 'A' allele ^b	Transcriptional regulation changed by prediction tool TFSearch (FS score 0.176)
5q31	rs76207936	A	Intron of <i>RAD50</i>	Altered potential splice site and altered potential branch point ^b	No result found
5q31	rs2237060	G	Intron of <i>RAD50</i>	EIEs site broken in 'C' but not in 'A' Allele ^b	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.065)
5q31	rs2040705	A	Intron of <i>RAD50</i>	IIEs site broken in A but not in T allele ^b	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.065)
5q31	rs2066960	A	Intron of <i>IL13</i>	Altered potential branch point and EIEs site broken in 'A' but not in 'C' allele ^b	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
5q31	rs1295686	T	Intron of <i>IL13</i>	Altered potential branch point and altered potential splice sites. IIEs site broken in 'G' but not in 'A' allele ^b	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
5q31	rs20541	A	Exon of <i>IL13</i>	Altered amino acid from R144Q ^c	Splicing regulation changed by prediction tool ESRSearch and RESCUE ESE. Transcriptional regulation exist by GoldenPath (FS score 0.273)
5q31	rs2243204	T	Downstream of <i>IL13</i>	No suitable prediction program available	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.065)
5q31	rs2243300	T	Upstream of <i>IL4</i>	No difference ^a	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.065)
5q31	rs2243248	G	Upstream of <i>IL4</i>	No difference ^a	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)

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Chr	SNP	Minor allele	Location of SNPs*	Putative functional impact	Functional relevance by F-SNP software
5q31	rs2243250	T	Upstream of <i>IL4</i>	GR binds to 'A' and MIG1 binds to 'T' allele ^a	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
5q31	rs2070874	T	5'UTR of <i>IL4</i>	Ftz and C/EBP alpha binds to 'C' and C/EBP beta binds to 'T' allele ^a	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
5q31	rs4426908	C	Downstream of <i>IL4</i>	No suitable prediction program available	Transcriptional regulation exist by prediction tool Golden Path (FS score 0.101)
12q13	rs324010	C	Upstream of <i>NAB2</i>	Sp1 binds to 'C' but not to 'T' allele ^a	Transcriptional regulation changed/ exist by prediction tool TFSearch, and GoldenPath (FS score 0.242)
12q13	rs324020	C	Intron of <i>NAB2</i>	EIEs site broken in 'C' but not in 'A' allele ^b	No functional information exists
12q13	rs324018	A	Intron of <i>NAB2</i>	Altered potential branch point and EIEs new site is found in 'C' but not in 'A' allele ^b	No functional information exists
12q13	rs324017	T	Intron of <i>NAB2</i>	EIEs new site is found in 'T' but not in 'G' allele ^b	No difference
12q13	rs3024979	A	Intron of <i>NAB2</i>	EIEs and IIEs site broken in 'A' but not in 'T' ^b	No functional information exists
12q13	rs1059513	C	3'UTRs of <i>STAT6</i>	Allele specific miRNA binding ^d	No functional information exists
12q13	rs703817	T	3' UTR of <i>STAT6</i>	Allele specific miRNA binding ^d	No functional information exists
12q13	rs324015	T	3' UTR of <i>STAT6</i>	Allele specific miRNA binding ^d	No functional information exists
12q13	rs3024975	T	Intron of <i>STAT6</i>	EIEs site broken in 'T' but not in 'C' allele ^b	No functional information exists
12q13	rs841718	G	Intron of <i>STAT6</i>	Altered potential splice site. EIEs and IIEs site broken in 'C' but not in 'T' allele ^b	No functional information exists
12q13	rs324011	T	Intron of <i>STAT6</i>	EIEs site broken in 'T' but not in 'C' allele also IIEs new site is found in 'T' but not in 'C' allele ^b	No functional information exists
12q13	rs167769	T	Intron of <i>STAT6</i>	EIEs site broken in 'T' but not in 'C' allele and IIEs new site is found in 'T' but not in 'C' allele ^b	No functional information exists

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Chr	SNP	Minor allele	Location of SNPs*	Putative functional impact	Functional relevance by F-SNP software
12q13	rs12368672	G	Upstream of <i>STAT6</i> (intergenic by F-SNP software)	Sp1 binds to 'C' but not to 'G' allele ^a	No functional information exists
12q13	rs2598483	T	Upstream of <i>STAT6</i> (regulatory region/upstream by F-SNP software)	No difference ^a	Transcriptional regulation frameshift coding/regulatory region by prediction tool Ensembl NS, and Ensembl TR (FS score 0.398)
12q13	rs324013	G	Upstream of <i>STAT6</i> (intergenic by F-SNP software)	CREB and CPE binds to 'C' but not to 'T' allele ^a	No functional information exists

* The location of SNPs is based on dbSNP database

^a = Alibaba software (version 2.1)

b = Human Splicing Finder software (version 2.4.1)

c = SIFT software (<http://sift.bii.a-star.edu.sg/>)

^d = RegRNA software (release 1.0, Jan 2006)

Abbreviations: EIEs are exon-identity elements and IIEs are intron-identity elements, whose prediction is based on methods of Zhang et al., 2008 (12);

FS (functional significance) score value in 55% of disease related SNP falls ≥ 0.5 (13); otherwise, see [Supplementary Table HS2](#)

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Supplementary Table S4IV. Associations between polymorphisms and IgE levels as a quantitative variable in the complete cohort (A), asthmatics (B) or non-asthmatics (C)

Chr	SNP	BIN	MAF	Total IgE in complete cohort (A)		Asthmatics (B)		Non-asthmatics (C)	
				Estimate (SE)	P-value	Estimate (SE)	P-value	Estimate (SE)	P-value
1q23	rs10489854	1	0.06	0.27 (0.14)	0.058	0.20 (0.21)	0.334	0.26 (0.17)	0.132
1q23	rs2494262	2	0.48	0.12 (0.06)	0.057	0.15 (0.09)	0.086	0.12 (0.08)	0.137
1q23	rs2511214	2	0.48	0.14 (0.06)	0.027	0.16 (0.09)	0.066	0.14 (0.08)	0.072
1q23	rs2427827	2	0.44	0.11 (0.06)	0.069	0.15 (0.09)	0.093	0.14 (0.08)	0.084
1q23	rs2252226	2	0.45	0.09 (0.06)	0.136	0.14 (0.09)	0.104	0.09 (0.08)	0.238
1q23	rs2427836	3	0.47	-0.15 (0.06)	0.016	-0.16 (0.09)	0.082	-0.16 (0.08)	0.042
1q23	rs2494264	3	0.46	-0.18 (0.06)	0.005	-0.21 (0.09)	0.015	-0.19 (0.08)	0.019
1q23	rs2494251	3	0.45	-0.16 (0.06)	0.014	-0.20 (0.09)	0.025	-0.14 (0.08)	0.086
1q23	rs2427837	4	0.27	-0.32 (0.07)	4.37*10⁻⁶	-0.26 (0.10)	0.006	-0.32 (0.09)	2.17*10⁻⁴
1q23	rs2251746	4	0.27	-0.30 (0.07)	3.54*10⁻⁵	-0.22 (0.10)	0.037	-0.32 (0.09)	3.17*10⁻⁴
1q23	rs61828219	5	0.17	0.15 (0.08)	0.069	0.28 (0.11)	0.011	0.01 (0.10)	0.904
1q23	rs3845625	5	0.15	0.15 (0.08)	0.087	0.25 (0.11)	0.031	-0.03 (0.11)	0.809
1q23	rs7549785	5	0.16	0.15 (0.09)	0.085	0.40 (0.12)	0.002	0.00 (0.11)	0.968
1q23	rs7522607	5	0.17	0.19 (0.09)	0.028	0.30 (0.12)	0.013	0.07 (0.11)	0.540
1q23	rs12565775	6	0.09	0.21 (0.11)	0.048	0.16 (0.15)	0.278	0.26 (0.14)	0.055
1q23	rs7548864	6	0.09	0.23 (0.11)	0.037	0.08 (0.16)	0.621	0.33 (0.14)	0.019
1q23	rs12119226	6	0.08	0.18 (0.11)	0.093	0.10 (0.15)	0.516	0.24 (0.14)	0.096
1q23	rs2427824	7	0.26	-0.03 (0.07)	0.622	-0.07 (0.10)	0.496	0.05 (0.09)	0.588
1q23	rs2427825	7	0.27	-0.03 (0.07)	0.712	-0.08 (0.10)	0.447	0.07 (0.09)	0.419
1q23	rs2494250	7	0.28	0.02 (0.07)	0.719	-0.03 (0.10)	0.792	0.12 (0.09)	0.155
1q23	rs56366265	8	0.18	0.12 (0.08)	0.152	0.04 (0.12)	0.746	0.11 (0.11)	0.291
1q23	rs41264475	9	0.01	0.30 (0.32)	0.356	0.20 (0.38)	0.609	0.05 (0.49)	0.914
1q23	rs16842010	10	0.06	0.15 (0.12)	0.231	0.29 (0.18)	0.105	0.07 (0.16)	0.675
1q23	rs2427829	11	0.28	0.15 (0.07)	0.030	0.24 (0.09)	0.010	0.06 (0.09)	0.482

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Chr	SNP	BIN	MAF	Total IgE in complete cohort (A)		Asthmatics (B)		Non-asthmatics (C)	
				Estimate (SE)	P-value	Estimate (SE)	P-value	Estimate (SE)	P-value
1q23	rs11265165	12	0.09	0.09 (0.11)	0.444	0.14 (0.16)	0.389	0.04 (0.14)	0.804
1q23	rs2427830	13	0.21	0.22 (0.07)	0.004	0.35 (0.10)	6.14*10⁻⁴	0.05 (0.10)	0.619
5q31	rs743562	1	0.45	0.05 (0.06)	0.407	0.02 (0.10)	0.835	0.10 (0.08)	0.205
5q31	rs739719	2	0.07	-0.27 (0.13)	0.040	-0.16 (0.20)	0.415	-0.32 (0.16)	0.042
5q31	rs62383708	3	0.16	-0.10 (0.09)	0.241	0.03 (0.12)	0.835	-0.17 (0.11)	0.119
5q31	rs2244012	4	0.25	0.15 (0.07)	0.036	0.10 (0.11)	0.354	0.17 (0.09)	0.055
5q31	rs2897443	4	0.24	0.11 (0.07)	0.125	0.02 (0.11)	0.827	0.16 (0.09)	0.079
5q31	rs6871536	4	0.25	0.15 (0.07)	0.045	0.08 (0.11)	0.427	0.16 (0.09)	0.074
5q31	rs1881457	4	0.24	0.15 (0.08)	0.056	0.04 (0.11)	0.718	0.25 (0.09)	0.009
5q31	rs1800925	4	0.24	0.15 (0.08)	0.050	0.08 (0.11)	0.460	0.21 (0.09)	0.028
5q31	rs28903085	5	0.004	-0.16 (0.44)	0.715	-0.21 (0.66)	0.744	-0.07 (0.53)	0.900
5q31	rs72797364	6	0.08	-0.19 (0.12)	0.103	-0.08 (0.17)	0.633	-0.32 (0.15)	0.029
5q31	rs17772583	7	0.23	-0.03 (0.07)	0.662	0.01 (0.10)	0.889	-0.05 (0.09)	0.619
5q31	rs76207936	8	0.01	0.36 (0.31)	0.245	-0.13 (0.41)	0.750	0.75 (0.42)	0.074
5q31	rs2237060	9	0.39	-0.04 (0.06)	0.528	-0.05 (0.09)	0.558	-0.03 (0.08)	0.736
5q31	rs2040705	10	0.46	-0.01 (0.06)	0.853	-0.01 (0.09)	0.889	-0.03 (0.08)	0.687
5q31	rs2066960	11	0.11	0.08 (0.10)	0.429	0.06 (0.14)	0.664	0.13 (0.12)	0.308
5q31	rs1295686	12	0.23	0.15 (0.08)	0.048	0.11 (0.11)	0.317	0.13 (0.10)	0.188
5q31	rs20541	12	0.22	0.16 (0.08)	0.047	0.10 (0.11)	0.370	0.13 (0.10)	0.200
5q31	rs2243204	13	0.09	0.23 (0.11)	0.041	0.29 (0.16)	0.067	0.15 (0.14)	0.283
5q31	rs2243300	14	0.08	0.26 (0.11)	0.025	0.36 (0.16)	0.028	0.12 (0.14)	0.386
5q31	rs2243248	15	0.07	0.06 (0.11)	0.624	0.04 (0.16)	0.827	0.14 (0.14)	0.305
5q31	rs2243250	16	0.17	0.14 (0.09)	0.113	0.13 (0.12)	0.278	0.07 (0.12)	0.533

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Chr	SNP	BIN	MAF	Total IgE in complete cohort (A)		Asthmatics (B)		Non-asthmatics (C)	
				Estimate (SE)	P-value	Estimate (SE)	P-value	Estimate (SE)	P-value
5q31	rs2070874	16	0.18	0.13 (0.09)	0.131	0.08 (0.12)	0.482	0.10 (0.11)	0.370
5q31	rs4426908	17	0.29	0.14 (0.07)	0.033	0.14 (0.09)	0.127	0.11 (0.09)	0.207
12q13	rs324010	1	0.47	0.03 (0.06)	0.631	0.04 (0.08)	0.636	-0.02 (0.08)	0.825
12q13	rs324020	2	0.44	0.07 (0.06)	0.278	0.09 (0.09)	0.291	0.10 (0.08)	0.205
12q13	rs324018	3	0.41	0.06 (0.06)	0.357	0.06 (0.09)	0.497	0.00 (0.08)	0.971
12q13	rs324017	4	0.31	0.05 (0.07)	0.463	0.04 (0.09)	0.682	0.03 (0.09)	0.735
12q13	rs3024979	5	0.09	0.03 (0.10)	0.752	-0.05 (0.14)	0.718	0.14 (0.13)	0.276
12q13	rs1059513	6	0.10	-0.34 (0.10)	0.001	-0.38 (0.14)	0.008	-0.29 (0.13)	0.023
12q13	rs703817	7	0.50	0.07 (0.06)	0.288	-0.10 (0.09)	0.294	0.07 (0.08)	0.409
12q13	rs324015	8	0.25	0.00 (0.07)	0.963	-0.08 (0.10)	0.414	0.06 (0.09)	0.518
12q13	rs3024975	9	0.10	0.05 (0.10)	0.639	0.03 (0.14)	0.828	0.00 (0.13)	0.994
12q13	rs841718	10	0.42	0.02 (0.07)	0.792	0.01 (0.09)	0.882	0.01 (0.08)	0.871
12q13	rs324011	11	0.40	0.06 (0.07)	0.410	0.19 (0.10)	0.054	-0.06 (0.08)	0.485
12q13	rs167769	11	0.39	0.08 (0.07)	0.200	0.15 (0.10)	0.117	0.00 (0.08)	0.984
12q13	rs12368672	11	0.40	0.11 (0.06)	0.076	0.20 (0.09)	0.023	0.00 (0.08)	0.955
12q13	rs2598483	12	0.10	0.09 (0.10)	0.403	0.01 (0.14)	0.956	0.10 (0.13)	0.443
12q13	rs324013	13	0.49	-0.04 (0.06)	0.513	-0.11 (0.09)	0.225	-0.05 (0.08)	0.557*

Number of individuals with non-missing phenotype involved in the respective calculations for the different rounds of genotyping:

A) Total IgE complete cohort: MALDI-TOF MS (1261), CHIP (1145)

B) Total IgE asthmatics: MALDI-TOF MS (602), CHIP (517)

C) Total IgE non-asthmatics: MALDI-TOF MS (651), CHIP (620)

* A is the minor allele

Abbreviations: see [Supplementary Table HS2](#)

Supplementary Table VS5. Creation of risk scores for gene by gene interaction analysis as previously established (14);

top associated SNPs of 1q23, 5q31 and 12q13 loci and the risk of elevated total serum IgE levels in the complete cohort were investigated

Chr	SNP	Allele combinations			Risks	No risks
		increase IgE levels				
1q23	rs2427837	GG		GA AA	GG=1	non GG=0
5q31	rs2243300	TT	TG	GG	TT or TG =1	GG =0
12q13	rs1059513	TT		TC CC	TT=1	non TT=0
					max score = 3	min score = 0

Abbreviations: see **Supplementary Table HS2**

Supplementary Table VIS6. Associations between polymorphisms and asthma or its sub-phenotypes

Chr	SNP	BIN	MAF	Asthma (A)		Atopic asthma (B)		Non-atopic asthma (C)	
				Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
1q23	rs10489854	1	0.06	1.06 (0.76-1.48)	0.728	1.12 (0.73-1.72)	0.606	1.14 (0.59-2.22)	0.694
1q23	rs2494262	2	0.48	1.04 (0.89-1.21)	0.614	1.16 (0.97-1.40)	0.110	1.02 (0.75-1.39)	0.906
1q23	rs2511214	2	0.48	1.05 (0.90-1.21)	0.566	1.18 (0.98-1.42)	0.084	0.99 (0.73-1.35)	0.961
1q23	rs2427827	2	0.44	0.99 (0.85-1.15)	0.867	1.11 (0.92-1.33)	0.293	0.98 (0.72-1.34)	0.904
1q23	rs2252226	2	0.45	1.00 (0.86-1.16)	0.978	1.11 (0.92-1.34)	0.260	0.96 (0.70-1.31)	0.796
1q23	rs2427836	3	0.47	0.99 (0.85-1.16)	0.937	0.89 (0.74-1.07)	0.209	1.06 (0.77-1.45)	0.723
1q23	rs2494264	3	0.46	1.00 (0.86-1.16)	0.989	0.87 (0.73-1.05)	0.148	1.04 (0.77-1.41)	0.809
1q23	rs2494251	3	0.45	0.97 (0.83-1.13)	0.694	0.86 (0.71-1.03)	0.108	1.15 (0.84-1.58)	0.376
1q23	rs2427837	4	0.27	0.92 (0.78-1.08)	0.302	0.78 (0.64-0.96)	0.021	0.87 (0.62-1.22)	0.404
1q23	rs2251746	4	0.27	0.93 (0.78-1.10)	0.404	0.81 (0.65-1.00)	0.050	0.91 (0.64-1.29)	0.581
1q23	rs61828219	5	0.17	1.11 (0.91-1.36)	0.293	1.29 (1.01-1.64)	0.045	0.79 (0.51-1.23)	0.299
1q23	rs3845625	5	0.15	1.21 (0.98-1.49)	0.076	1.33 (1.03-1.71)	0.030	0.84 (0.54-1.31)	0.435
1q23	rs7549785	5	0.16	0.98 (0.79-1.20)	0.822	1.17 (0.90-1.52)	0.230	0.67 (0.42-1.09)	0.105
1q23	rs7522607	5	0.17	1.13 (0.92-1.40)	0.242	1.25 (0.97-1.61)	0.082	0.78 (0.49-1.23)	0.284
1q23	rs12565775	6	0.09	0.95 (0.74-1.23)	0.695	1.06 (0.77-1.45)	0.735	0.93 (0.54-1.59)	0.790
1q23	rs7548864	6	0.09	1.03 (0.79-1.34)	0.832	1.15 (0.83-1.59)	0.413	1.17 (0.69-1.97)	0.557
1q23	rs12119226	6	0.08	1.00 (0.77-1.30)	0.976	1.13 (0.81-1.56)	0.475	1.10 (0.65-1.85)	0.737
1q23	rs2427824	7	0.26	0.92 (0.78-1.09)	0.348	0.89 (0.72-1.10)	0.277	1.10 (0.79-1.55)	0.567
1q23	rs2427825	7	0.27	0.92 (0.78-1.09)	0.348	0.94 (0.77-1.16)	0.585	0.99 (0.69-1.41)	0.958
1q23	rs2494250	7	0.28	0.96 (0.81-1.13)	0.626	0.99 (0.81-1.22)	0.934	1.15 (0.83-1.60)	0.409
1q23	rs56366265	8	0.18	1.02 (0.83-1.25)	0.864	0.98 (0.76-1.26)	0.870	1.17 (0.78-1.75)	0.454
1q23	rs41264475	9	0.01	1.74 (0.77-3.94)	0.181	1.93 (0.67-5.52)	0.221	2.29 (0.54-9.75)	0.261
1q23	rs16842010	10	0.06	0.95 (0.71-1.29)	0.759	1.05 (0.72-1.52)	0.806	0.79 (0.41-1.52)	0.478
1q23	rs2427829	11	0.28	1.09 (0.93-1.29)	0.292	1.24 (1.01-1.51)	0.043	1.03 (0.74-1.45)	0.856

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Chr	SNP	BIN	MAF	Asthma (A)		Atopic asthma (B)		Non-atopic asthma (C)	
				Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
1q23	rs11265165	12	0.09	1.05 (0.80-1.38)	0.744	1.15 (0.82-1.61)	0.428	0.91 (0.51-1.63)	0.749
1q23	rs2427830	13	0.21	1.19 (0.99-1.43)	0.060	1.44 (1.15-1.82)	0.002	0.96 (0.64-1.42)	0.830
5q31	rs743562	1	0.45	0.94 (0.81-1.10)	0.432	0.95 (0.79-1.15)	0.621	1.10 (0.81-1.49)	0.537
5q31	rs739719	2	0.07	0.86 (0.63-1.18)	0.364	0.80 (0.55-1.17)	0.246	0.57 (0.28-1.17)	0.124
5q31	rs62383708	3	0.16	0.96 (0.78-1.18)	0.728	0.92 (0.72-1.18)	0.505	0.86 (0.57-1.30)	0.461
5q31	rs2244012	4	0.25	1.11 (0.93-1.33)	0.229	1.14 (0.91-1.41)	0.251	1.09 (0.77-1.53)	0.628
5q31	rs2897443	4	0.24	1.12 (0.94-1.34)	0.214	1.09 (0.88-1.36)	0.427	1.13 (0.80-1.60)	0.483
5q31	rs6871536	4	0.25	1.14 (0.95-1.36)	0.157	1.13 (0.91-1.41)	0.270	1.09 (0.77-1.54)	0.623
5q31	rs1881457	4	0.24	1.09 (0.91-1.31)	0.364	1.13 (0.90-1.42)	0.297	1.04 (0.72-1.50)	0.824
5q31	rs1800925	4	0.24	1.10 (0.91-1.32)	0.342	1.11 (0.88-1.41)	0.361	1.06 (0.74-1.52)	0.749
5q31	rs28903085	5	0.004	0.56 (0.18-1.73)	0.314	0.33 (0.09-1.29)	0.112	1.07 (0.22-5.22)	0.934
5q31	rs72797364	6	0.08	0.95 (0.72-1.25)	0.704	0.86 (0.61-1.21)	0.394	1.24 (0.74-2.07)	0.412
5q31	rs17772583	7	0.23	0.85 (0.71-1.01)	0.065	0.83 (0.67-1.03)	0.096	0.87 (0.60-1.25)	0.440
5q31	rs76207936	8	0.01	1.66 (0.79-3.49)	0.182	1.39 (0.58-3.34)	0.466	1.42 (0.37-5.43)	0.611
5q31	rs2237060	9	0.39	1.05 (0.90-1.22)	0.563	1.07 (0.89-1.29)	0.479	1.04 (0.76-1.40)	0.820
5q31	rs2040705	10	0.46	1.07 (0.92-1.24)	0.383	1.11 (0.93-1.33)	0.249	1.09 (0.82-1.45)	0.558
5q31	rs2066960	11	0.11	0.91 (0.72-1.16)	0.459	0.99 (0.74-1.32)	0.952	0.95 (0.59-1.53)	0.827
5q31	rs1295686	12	0.23	1.22 (1.01-1.47)	0.038	1.19 (0.95-1.48)	0.131	0.95 (0.65-1.39)	0.784
5q31	rs20541	12	0.22	1.23 (1.02-1.49)	0.031	1.19 (0.95-1.49)	0.134	0.98 (0.66-1.43)	0.901
5q31	rs2243204	13	0.09	1.13 (0.87-1.48)	0.371	1.18 (0.85-1.64)	0.315	0.86 (0.48-1.56)	0.625
5q31	rs2243300	14	0.08	1.09 (0.83-1.44)	0.522	1.14 (0.81-1.59)	0.451	0.84 (0.46-1.54)	0.578
5q31	rs2243248	15	0.07	0.84 (0.63-1.11)	0.215	0.87 (0.61-1.23)	0.434	0.91 (0.51-1.63)	0.762
5q31	rs2243250	16	0.17	1.23 (1.00-1.52)	0.053	1.23 (0.96-1.59)	0.103	0.68 (0.42-1.10)	0.115

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Chr	SNP	BIN	MAF	Asthma (A)		Atopic asthma (B)		Non-atopic asthma (C)	
				Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
5q31	rs2070874	16	0.18	1.22 (0.99-1.50)	0.057	1.22 (0.95-1.56)	0.124	0.67 (0.42-1.07)	0.090
5q31	rs4426908	17	0.29	1.12 (0.96-1.32)	0.156	1.25 (1.02-1.53)	0.031	0.78 (0.55-1.11)	0.165
12q13	rs324010	1	0.47	1.02 (0.88-1.19)	0.760	1.00 (0.83-1.20)	0.988	1.10 (0.81-1.50)	0.527
12q13	rs324020	2	0.44	0.95 (0.81-1.10)	0.474	1.03 (0.86-1.24)	0.752	0.84 (0.62-1.14)	0.269
12q13	rs324018	3	0.41	1.12 (0.96-1.31)	0.141	1.06 (0.88-1.28)	0.525	1.13 (0.83-1.52)	0.449
12q13	rs324017	4	0.31	1.04 (0.88-1.22)	0.669	0.98 (0.81-1.20)	0.854	1.10 (0.80-1.51)	0.579
12q13	rs3024979	5	0.09	1.03 (0.80-1.33)	0.797	1.34 (0.98-1.84)	0.070	1.07 (0.63-1.81)	0.807
12q13	rs1059513	6	0.10	0.89 (0.69-1.13)	0.337	0.76 (0.56-1.02)	0.071	1.23 (0.80-1.89)	0.350
12q13	rs703817	7	0.50	1.02 (0.87-1.19)	0.816	1.11 (0.92-1.34)	0.267*	0.93 (0.68-1.26)	0.625
12q13	rs324015	8	0.25	0.97 (0.82-1.16)	0.757	0.89 (0.71-1.10)	0.276	1.12 (0.80-1.58)	0.509
12q13	rs3024975	9	0.10	1.22 (0.95-1.57)	0.116	1.20 (0.89-1.62)	0.241	0.97 (0.59-1.60)	0.917
12q13	rs841718	10	0.42	0.97 (0.83-1.14)	0.712	0.91 (0.75-1.11)	0.370	1.05 (0.76-1.45)	0.772
12q13	rs324011	11	0.40	1.02 (0.86-1.19)	0.849	1.02 (0.83-1.24)	0.870	0.84 (0.60-1.17)	0.294
12q13	rs167769	11	0.39	1.06 (0.90-1.24)	0.491	1.05 (0.86-1.27)	0.633	0.92 (0.66-1.27)	0.605
12q13	rs12368672	11	0.40	1.05 (0.90-1.22)	0.528	1.08 (0.90-1.30)	0.417	0.91 (0.67-1.23)	0.533
12q13	rs2598483	12	0.10	1.18 (0.92-1.52)	0.184	1.56 (1.14-2.13)	0.006	1.19 (0.69-2.03)	0.535
12q13	rs324013	13	0.49	0.92 (0.79-1.07)	0.292	0.97 (0.81-1.18)	0.789	1.08 (0.79-1.49)	0.629

Number of individuals with non-missing phenotype involved in the respective calculations for the different rounds of genotyping:

A) Asthma: MALDI-TOF MS (763 cases / 683 controls), CHIP (651 cases / 652 controls)

B) Atopic asthma: MALDI-TOF MS (548 cases / 426 controls), CHIP (468 cases / 408 controls)

C) Non-atopic asthma: MALDI-TOF MS (113 cases / 426 controls), CHIP (98 cases / 408 controls)

* C is the minor allele

Abbreviations: see [Supplementary Table HS2](#)

Supplementary Table VHS7. Associations with atopic sensitization, allergic rhinitis or atopic dermatitis

Chr	SNP	BIN	MAF	Atopy (A)		Rhinitis (B)		Eczema (C)	
				Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
1q23	rs10489854	1	0.06	1.54 (0.95-2.49)	0.077	1.83 (1.01-3.33)	0.048	0.59 (0.29-1.19)	0.142
1q23	rs2494262	2	0.48	1.13 (0.89-1.44)	0.321	0.98 (0.70-1.37)	0.896	0.95 (0.72-1.26)	0.731
1q23	rs2511214	2	0.48	1.16 (0.92-1.48)	0.213	1.08 (0.78-1.52)	0.636	0.96 (0.73-1.26)	0.779
1q23	rs2427827	2	0.44	1.09 (0.86-1.39)	0.467	1.04 (0.74-1.46)	0.822	1.02 (0.77-1.34)	0.900
1q23	rs2252226	2	0.45	1.06 (0.83-1.34)	0.650	0.96 (0.69-1.35)	0.822	0.96 (0.73-1.26)	0.755
1q23	rs2427836	3	0.47	0.82 (0.65-1.05)	0.115	0.85 (0.61-1.19)	0.354	1.08 (0.82-1.42)	0.610
1q23	rs2494264	3	0.46	0.80 (0.63-1.02)	0.076	0.85 (0.61-1.19)	0.353	1.12 (0.85-1.47)	0.424
1q23	rs2494251	3	0.45	0.84 (0.66-1.07)	0.160	0.79 (0.56-1.11)	0.173	1.05 (0.79-1.38)	0.747
1q23	rs2427837	4	0.27	0.75 (0.57-0.99)	0.043	0.79 (0.54-1.17)	0.243	0.86 (0.63-1.17)	0.326
1q23	rs2251746	4	0.27	0.77 (0.58-1.02)	0.070	0.86 (0.58-1.26)	0.428	0.82 (0.60-1.13)	0.221
1q23	rs61828219	5	0.17	1.13 (0.83-1.55)	0.436	1.25 (0.82-1.91)	0.295	1.06 (0.74-1.51)	0.771
1q23	rs3845625	5	0.15	1.08 (0.77-1.51)	0.664	1.23 (0.78-1.92)	0.372	1.05 (0.72-1.54)	0.803
1q23	rs7549785	5	0.16	1.13 (0.82-1.55)	0.447	1.27 (0.83-1.95)	0.264	1.07 (0.75-1.53)	0.713
1q23	rs7522607	5	0.17	1.06 (0.76-1.47)	0.742	1.27 (0.82-1.96)	0.287	1.01 (0.69-1.48)	0.961
1q23	rs12565775	6	0.09	1.43 (0.97-2.09)	0.068	1.68 (1.04-2.71)	0.033	0.86 (0.53-1.37)	0.521
1q23	rs7548864	6	0.09	1.52 (1.03-2.24)	0.034	1.82 (1.13-2.94)	0.015	0.79 (0.48-1.31)	0.363
1q23	rs12119226	6	0.08	1.47 (0.99-2.18)	0.054	1.74 (1.07-2.84)	0.026	0.90 (0.55-1.46)	0.670
1q23	rs2427824	7	0.26	0.94 (0.72-1.24)	0.672	0.71 (0.48-1.07)	0.103	0.98 (0.72-1.33)	0.882
1q23	rs2427825	7	0.27	1.00 (0.77-1.30)	0.979	0.76 (0.52-1.13)	0.175	0.96 (0.71-1.31)	0.815
1q23	rs2494250	7	0.28	1.03 (0.79-1.33)	0.851	0.85 (0.59-1.24)	0.410	0.95 (0.71-1.29)	0.758
1q23	rs56366265	8	0.18	0.99 (0.72-1.37)	0.972	0.89 (0.57-1.40)	0.611	1.38 (0.98-1.96)	0.070
1q23	rs41264475	9	0.01	0.93 (0.19-4.63)	0.927	0.00 (0.00-∞)	0.999	0.66 (0.08-5.37)	0.694
1q23	rs16842010	10	0.06	1.26 (0.80-2.00)	0.323	1.59 (0.88-2.87)	0.124	0.82 (0.46-1.46)	0.500
1q23	rs2427829	11	0.28	1.11 (0.86-1.44)	0.427	1.18 (0.83-1.68)	0.364	0.94 (0.69-1.27)	0.692

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Chr	SNP	BIN	MAF	Atopy (A)		Rhinitis (B)		Eczema (C)	
				Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
1q23	rs11265165	12	0.09	1.25 (0.82-1.89)	0.301	1.96 (1.19-3.25)	0.009	1.11 (0.69-1.79)	0.664
1q23	rs2427830	13	0.21	1.37 (1.03-1.83)	0.033	1.40 (0.94-2.07)	0.096	1.09 (0.77-1.53)	0.642
5q31	rs743562	1	0.45	0.94 (0.74-1.19)	0.582	0.97 (0.70-1.35)	0.857	1.08 (0.82-1.42)	0.579
5q31	rs739719	2	0.07	0.74 (0.44-1.24)	0.252	0.84 (0.41-1.71)	0.624	0.89 (0.50-1.59)	0.689
5q31	rs62383708	3	0.16	0.91 (0.66-1.27)	0.586	0.90 (0.56-1.43)	0.644	1.00 (0.69-1.44)	0.993
5q31	rs2244012	4	0.25	1.09 (0.83-1.41)	0.549	1.02 (0.70-1.49)	0.903	1.03 (0.75-1.40)	0.876
5q31	rs2897443	4	0.24	1.15 (0.88-1.50)	0.315	1.12 (0.77-1.63)	0.553	1.00 (0.73-1.37)	0.988
5q31	rs6871536	4	0.25	1.11 (0.85-1.45)	0.457	1.07 (0.73-1.56)	0.734	1.00 (0.73-1.37)	0.999
5q31	rs1881457	4	0.24	1.28 (0.97-1.69)	0.084	1.16 (0.79-1.72)	0.449	0.95 (0.68-1.32)	0.744
5q31	rs1800925	4	0.24	1.15 (0.86-1.52)	0.342	1.05 (0.70-1.57)	0.821	1.09 (0.78-1.51)	0.617
5q31	rs28903085	5	0.004	0.00 (0.00-∞)	0.999	0.00 (0.00-∞)	0.999	0.66 (0.08-5.39)	0.696
5q31	rs72797364	6	0.08	0.67 (0.42-1.09)	0.106	0.67 (0.33-1.35)	0.261	1.06 (0.64-1.74)	0.829
5q31	rs17772583	7	0.23	0.83 (0.63-1.11)	0.215	0.88 (0.59-1.32)	0.542	1.02 (0.74-1.40)	0.929
5q31	rs76207936	8	0.01	1.13 (0.35-3.64)	0.843	1.39 (0.31-6.35)	0.668	0.35 (0.05-2.70)	0.314
5q31	rs2237060	9	0.39	1.06 (0.84-1.34)	0.648	1.20 (0.86-1.66)	0.287	1.00 (0.76-1.31)	0.997
5q31	rs2040705	10	0.46	1.19 (0.94-1.50)	0.143	1.22 (0.88-1.69)	0.239	0.96 (0.74-1.25)	0.774
5q31	rs2066960	11	0.11	0.90 (0.61-1.32)	0.582	1.20 (0.72-1.98)	0.482	0.72 (0.45-1.16)	0.178
5q31	rs1295686	12	0.23	0.92 (0.68-1.25)	0.607	0.86 (0.56-1.32)	0.479	0.99 (0.70-1.39)	0.950
5q31	rs20541	12	0.22	0.92 (0.68-1.24)	0.570	0.89 (0.57-1.37)	0.586	1.00 (0.71-1.41)	0.992
5q31	rs2243204	13	0.09	0.90 (0.58-1.40)	0.636	0.80 (0.42-1.53)	0.504	0.93 (0.56-1.55)	0.789
5q31	rs2243300	14	0.08	0.83 (0.53-1.31)	0.431	0.70 (0.35-1.40)	0.307	1.00 (0.61-1.64)	0.100
5q31	rs2243248	15	0.07	1.30 (0.86-1.95)	0.215	1.19 (0.68-2.10)	0.539	0.93 (0.55-1.56)	0.780
5q31	rs2243250	16	0.17	0.92 (0.65-1.30)	0.635	1.29 (0.83-2.00)	0.259	0.96 (0.65-1.43)	0.859

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Chr	SNP	BIN	MAF	Atopy (A)		Rhinitis (B)		Eczema (C)	
				Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
5q31	rs2070874	16	0.18	0.97 (0.69-1.36)	0.855	1.27 (0.82-1.97)	0.285	0.94 (0.64-1.39)	0.773
5q31	rs4426908	17	0.29	0.96 (0.74-1.25)	0.759	1.15 (0.80-1.64)	0.459	0.94 (0.70-1.26)	0.675
12q13	rs324010	1	0.47	0.91 (0.71-1.16)	0.436	0.76 (0.54-1.08)	0.123	1.17 (0.88-1.54)	0.276
12q13	rs324020	2	0.44	1.13 (0.89-1.43)	0.328	1.31 (0.94-1.83)	0.117	0.95 (0.72-1.24)	0.690
12q13	rs324018	3	0.41	0.87 (0.68-1.12)	0.288	0.82 (0.57-1.17)	0.266	1.09 (0.82-1.44)	0.551
12q13	rs324017	4	0.31	0.85 (0.65-1.12)	0.247	0.81 (0.55-1.19)	0.291	0.96 (0.71-1.30)	0.787
12q13	rs3024979	5	0.09	1.60 (1.10-2.32)	0.014	1.61 (0.99-2.61)	0.054	0.88 (0.54-1.41)	0.584
12q13	rs1059513	6	0.10	0.80 (0.53-1.20)	0.279	0.96 (0.56-1.66)	0.892	0.85 (0.54-1.35)	0.494
12q13	rs703817	7	0.50	1.13 (0.89-1.44)	0.302	1.40 (1.00-1.95)	0.051	0.97 (0.74-1.27)	0.807
12q13	rs324015	8	0.25	0.87 (0.65-1.15)	0.317	0.83 (0.56-1.24)	0.359	0.88 (0.64-1.22)	0.446
12q13	rs3024975	9	0.10	1.02 (0.68-1.51)	0.940	0.94 (0.53-1.67)	0.834	1.33 (0.87-2.05)	0.190
12q13	rs841718	10	0.42	0.89 (0.69-1.14)	0.359	0.70 (0.49-1.01)	0.055	1.12 (0.84-1.50)	0.423
12q13	rs324011	11	0.40	0.89 (0.69-1.15)	0.381	0.98 (0.69-1.40)	0.919	0.99 (0.74-1.32)	0.921
12q13	rs167769	11	0.39	0.99 (0.77-1.27)	0.938	1.12 (0.80-1.58)	0.504	1.04 (0.79-1.38)	0.770
12q13	rs12368672	11	0.40	0.97 (0.76-1.23)	0.778	1.08 (0.77-1.50)	0.671	1.06 (0.81-1.39)	0.688
12q13	rs2598483	12	0.10	1.86 (1.29-2.69)	8.32*10⁻⁴	1.59 (0.99-2.58)	0.057	0.83 (0.51-1.34)	0.442
12q13	rs324013	13	0.49	1.08 (0.85-1.38)	0.525	0.94 (0.67-1.32)	0.717	0.92 (0.69-1.21)	0.539

Number of individuals with non-missing phenotype involved in the respective calculations for the different rounds of genotyping:

A) Atopic sensitization: MALDI-TOF MS (192 cases / 536 controls), CHIP (183 cases / 515 controls)

B) Allergic rhinitis: MALDI-TOF MS (80 cases / 665 controls), CHIP (77 cases / 636 controls)

C) Atopic dermatitis: MALDI-TOF MS (131 cases / 603 controls), CHIP (123 cases / 578 controls)

Abbreviations: see [Supplementary-Table HS2](#)