

Letter to the Editor

A genome-wide association study reveals 2 new susceptibility loci for atopic dermatitis

To the Editor:

Association studies have identified a total of 23 European and Asian genetic susceptibility loci for atopic dermatitis (AD), although these explain only a small fraction of the estimated total heritability.^{1,2}

To identify further risk loci for AD, we analyzed an imputed data set of more than 1.6 million genetic markers from 924 unrelated German tertiary care cases and 5506 population-based control subjects, followed by an additional replication study in a further 1383 AD cases and 1728 control subjects (see the [Methods](#) section and [Table E1](#) in this article's Online Repository at www.jacionline.org). Cases used for the screen had also been genotyped on Affymetrix 500K/5.0 platforms (Affymetrix, Santa Clara, Calif) and used for a previous genome-wide association study (GWAS).³

Genome-wide single nucleotide polymorphism (SNP) genotyping of patients was performed by an Affymetrix service facility (South San Francisco, Calif) using the Affymetrix Genome-Wide Human SNP Array 6.0 (1000k), according to the manufacturer's protocols. SNP genotype imputation was carried out with MACH version 1.10.16 and HapMap II CEU phased haplotypes release 22 (http://hapmap.ncbi.nlm.nih.gov/downloads/phasing/2007-08_rel22/phased/) as the reference data set to increase marker density. After stringent quality control filtering (see the [Methods](#) section in this article's Online Repository), the initial GWAS sample set consisted of 870 cases and 5293 control subjects with 1,623,390 markers. Genetic heterogeneity was found to be moderate, with an estimated genomic inflation factor λ_{1000} value of 1.076, and the quantile-quantile plot showed substantial deviation from the expected distribution of genome-wide P values in the tail of the distribution (see [Fig E1](#) in this article's Online Repository at www.jacionline.org).

After visual inspection of the regional plots of 812 SNPs showing a disease association with P values of 10^{-3} or less by means of clumping (clump command with default settings in PLINK: $P1 \leq 10^{-3}$, $P2 \leq .05$, $r^2 \geq 0.8$, kb = 250), 98 SNPs were carried forward to replication genotyping using ligation-based SNPlex or TaqMan (Applied Biosystems, Foster City, Calif) technology. After quality control (see the [Methods](#) section in this article's Online Repository), 1383 AD cases, 1728 control subjects, and 69 SNPs remained for association analysis (complete results are presented in [Table E2](#) in this article's Online Repository at www.jacionline.org). Ten of these SNPs showed a nominally significant association with P values of less than .05 in the replication cohort. In the combined analysis (GWAS and replication data sets) 5 loci achieved P values of less than 5×10^{-7} ([Table I](#) and see [Fig E2](#) in this article's Online Repository at www.jacionline.org for regional association plots). A marker within the filaggrin (*FLG*) locus achieved genome-wide significance in the combined analysis (rs12144049, $P_{\text{comb}} = 1.02 \times 10^{-16}$). After excluding carriers of common *FLG* mutations (R501X, 2282del4, R2447X, and S3247X), this loci no longer showed a significant association. Furthermore, an SNP located within the *RAD50/IL13/KIF3A* locus at 5q31.1 showed strong association (rs3091307, $P_{\text{comb}} = 4.18 \times 10^{-7}$), which tags previously reported susceptibility markers in the *RAD50/KIF3A* locus:

rs2040704 ($r^2 = 0.68$),⁴ rs3798135 ($r^2 = 0.76$), and rs2240032 ($r^2 = 0.76$).⁵ In contrast, it is in low linkage disequilibrium ($r^2 < 0.36$) with the *IL13* risk variants rs848¹ and rs20541,² supporting the notion of several independent association signals in this region.¹

Twelve further susceptibility loci reported previously in European population GWASs were replicated successfully ($P_{\text{GWAS}} \leq 5 \times 10^{-2}$). Only 1 reported locus, *ACTL9*, failed in the screen ($P_{\text{GWAS}} = 5.23 \times 10^{-2}$). Regarding susceptibility loci reported exclusively in GWASs of Asian populations, 5 loci (*GLB1*, *CCDC80*, *ZNF365*, *OR10A/3/NLRP10*, and *CYP24A1/PFDN4*) showed evidence for association in Europeans for the first time. Thus 21 of 23 reported AD risk loci could be confirmed. Detailed results of all AD susceptibility loci previously reported in GWASs from population with European and Asian ancestry are provided in [Table II](#).^{1-3,6-8}

Two novel loci not previously implicated in AD reached classical genome-wide significance ($P_{\text{comb}} < 5 \times 10^{-8}$) in the combined analysis: xin actin-binding repeat-containing protein 2 (*XIRP2*), also known as *CYMA3*, at chromosome 2q24.3, with the lead variant rs6720763 ($P_{\text{comb}} = 4.37 \times 10^{-8}$; odds ratio, 1.29) located in intron 2, and doublesex and mab-3-related transcription factor-like family A1 (*DMRTA1*) at chromosome 9p21.3, with the lead variant rs10738626 ($P_{\text{comb}} = 1.45 \times 10^{-8}$; odds ratio, 0.81) located 73 kb upstream.

Thus far, very little is known about the function of *XIRP2* and *DMRTA1*. For *XIRP2*, a cross-linking function between skeletal muscle and F-actin⁹ and an involvement as an interactor in the cohesion-*RAD21* interactome has been reported,¹⁰ and *DMRTA1* has primarily been implicated in sex differentiation.¹¹

In silico bioinformatic analysis of all SNPs in full linkage disequilibrium ($r^2 = 1$ based on 1000G data) with the lead variants (see the supplementary text section and [Tables E3a, E3b, E4a, and E4b](#) in this article's Online Repository at www.jacionline.org) indicated that the *XIRP2* lead variant rs6720763 is located within a potentially regulatory region because it is positioned within a DNaseI-hypersensitive site, transcription factors have been observed to bind to that region, alterations in transcription factor binding are predicted, and the DNA sequence is highly conserved. In an expression quantitative trait locus analysis with the MuTHER study data,¹² none of the investigated *DMRTA1* and *XIRP2* SNPs showed expression quantitative trait locus evidence on both loci.

PCR analysis in a panel of 13 different human tissues and cell types and immunohistochemistry in healthy epidermis and lesional AD epidermis showed RNA expression in skin and keratinocytes and immunoreactivity in lesional and healthy epidermis for both *XIRP2* and *DMRTA1*. Furthermore, both genes showed high mRNA expression in skeletal muscle, spleen, and testis tissue, and in addition, *DMRTA1* was expressed in the pancreas, lung, kidney, brain, small intestine, colon, and thymus (see the [Results](#) section and [Figs E3-E5](#) in this article's Online Repository at www.jacionline.org). Electrophoretic mobility shift assays with Cy5-labeled oligonucleotides using nuclear extracts derived from human HaCaT keratinocytes and competition shifts with different amounts of unlabeled probes (see the [Results](#) section in this article's Online Repository) showed allele-specific protein-DNA interactions for both identified risk SNPs ([Fig 1](#) and see [Fig E6](#) in this article's Online Repository at www.jacionline.org);

TABLE I. SNPs with a P value of 5×10^{-7} or less in the combined analysis

dbSNP ID	Chromosome	Position	A1, A2	Locus	GWAS				Replication				Combined	
					Cases: 870, control subjects: 5293				Cases: 1383, control subjects: 1728				Cases: 2253, control subjects: 7021	
					AF _{ca}	AF _{co}	P_{GWAS}	OR (95% CI)	AF _{ca}	AF _{co}	P_{Rep}	OR (95% OR)	P_{comb}	OR
rs12144049	1	150707534	C, T	<i>EDC</i>	0.369	0.279	1.87×10^{-8}	1.47 (1.31-1.64)	0.346	0.286	5.89×10^{-7}	1.33 (1.19-1.48)	1.02×10^{-16}	1.39
rs6720763	2	167700532	C, T	<i>XIRP2</i> (intron)	0.221	0.18	5.03×10^{-5}	1.31 (1.15-1.48)	0.211	0.174	2.65×10^{-4}	1.27 (1.12-1.44)	4.37×10^{-8}	1.29
rs3091307	5	132017035	A, G	<i>RAD50</i> <i>/IL13/</i> <i>KIF3A</i>	0.276	0.228	5.42×10^{-5}	1.3 (1.14-1.47)	0.283	0.247	1.69×10^{-3}	1.2 (1.07-1.35)	4.18×10^{-7}	1.24
rs10738626	9	22363457	C, T	<i>DMRTA1</i>	0.443	0.494	1.96×10^{-6}	0.77 (0.69-0.86)	0.474	0.517	7.95×10^{-4}	0.84 (0.76-0.93)	1.45×10^{-8}	0.81
rs1665050	15	57080897	A, G	<i>RNF111</i> (intron)	0.299	0.252	7.12×10^{-5}	1.27 (1.13-1.43)	0.296	0.255	3.48×10^{-4}	1.23 (1.1-1.38)	9.65×10^{-8}	1.25

SNPs are ranked by chromosomal position (National Center for Biotechnology Information build 36 [hg18]). ORs and 95% CIs for allele A1 are shown.

A1, Minor allele; A2, major allele; AF_{ca}/AF_{co}, allele frequencies in cases/control subjects; OR, odds ratio; P_{GWAS} / P_{Rep} / P_{comb} , P values in GWAS/replication/combined analysis.

TABLE II. Replication results of 23 established AD loci from GWASs

Locus	Reported gene(s)	Reported SNP(s)	References	Top SNP in region	Position	EA/RA	OR	P value
Loci identified through GWASs in European populations								
1q21.3	Tags <i>FLG</i> signal	rs3126085	1, 3, 6	rs12144049	152440176	C/T	1.47 (1.31-1.64)	1.87×10^{-8}
4q27	<i>IL2/IL21</i>	rs17389644	1	rs17454584	123497697	A/G	0.83 (0.77-0.89)	1.60×10^{-4}
5q31.1	<i>RAD50/IL13/IL4/KIF3A</i>	rs1295686, rs2897442, rs848	1, 7, 8	rs3091307	131995843	G/A	1.3 (1.14-1.47)	5.42×10^{-5}
6p21.33	<i>HLA-C/HLA-B/MICA</i>	rs9368677, rs2251396	2, 8	rs3021366	31324100	A/C	0.48 (0.33-0.78)	2.36×10^{-5}
6p21.33	<i>BAT1</i>	rs2844509	2	rs3130048	31510924	C/T	0.75 (0.68-0.82)	4.88×10^{-5}
6p21.33	<i>C6orf48</i>	rs9368699	2	rs9368699	31802541	C/T	0.51 (0.32-0.83)	2.55×10^{-4}
6p21.33	<i>TNXB/CREBL1</i>	rs12153855	2	rs12153855	32074804	C/T	0.77 (0.68-0.86)	5.55×10^{-3}
11p12	<i>PRR5L</i>	rs12295535	1	rs7945962	36344202	A/G	0.79 (0.74-0.84)	2.16×10^{-5}
11q13.1	<i>OVOL1</i>	rs479844	7, 8	rs479844	65551710	T/C	1.20 (1.25-1.14)	5.22×10^{-4}
11q13.5	<i>C11orf30/LRRC32</i>	rs7927894, rs11236809, rs7110818	1, 3, 8	rs2155219	76281593	T/G	0.81 (0.76-0.86)	1.74×10^{-4}
16p13.13	<i>CLEC16A/DEXI</i>	rs9923856, rs2041733	1, 8	rs2041733	11223454	C/G	1.23 (1.18-1.28)	1.31×10^{-4}
17q21.32-33	<i>ZNF652</i>	rs16948048	1	rs7209400	47440466	C/T	0.85 (0.80-0.90)	3.32×10^{-3}
19p13.2	<i>ACTL9</i>	rs2164983	7	rs12611036	8789381	A/G	1.12 (1.08-1.18)	5.23×10^{-2}
Loci identified through GWASs in Asian populations								
2q12.1	<i>IL18R1/IL18RAP/SLC9A4</i>	rs13015714, rs759382	1, 8	rs13015714	102945378	T/G	1.23 (1.17-1.29)	6.55×10^{-4}
3p22.3	<i>GLB1</i>	rs7613051	8	rs13091893	33065339	G/C	1.13 (1.07-1.19)	3.43×10^{-2}
3q13.2	<i>CCDC80</i>	rs12634229	8	rs2129844	112376308	T/C	1.52 (1.37-1.67)	2.93×10^{-3}
5q22.1	<i>TMEM232/SLC25A46</i>	rs7701890	6	rs11241070	109858821	G/A	1.17 (1.08-1.26)	6.47×10^{-2}
6p21.32	<i>GPSM3</i>	rs176095	8	rs6941112	32158319	A/G	1.31 (1.25-1.37)	1.66×10^{-5}
7p22.2	<i>CARD11</i>	rs4722404	8	rs11983024	3081727	C/G	0.82 (0.76-0.88)	1.75×10^{-3}
10q21.2	<i>ZNF365</i>	rs10995251	8	rs7919747	64380336	T/C	0.71 (0.62-0.8)	3.28×10^{-4}
11p15.4	<i>OR10A3/NLRP10</i>	rs878860	8	rs10769866	7968359	G/C	0.88 (0.82-0.94)	1.89×10^{-2}
20q13.2	<i>CYP24A1/PFDN4</i>	rs16999165	8	rs6013912	52807221	C/T	0.80 (0.74-0.86)	8.61×10^{-5}
20q13.33	<i>TNFRSF6B</i>	rs909341	1, 6	rs16984240	62328742	T/C	0.79 (0.71-0.87)	6.83×10^{-3}

Previously reported genes and SNPs (dbSNP ID) are sorted by chromosomal position (locus). The top associated SNP from imputed GWAS data for each locus (± 250 kb) is displayed. EA/RA, Effect/reference allele; OR, odds ratio.

however, further studies are needed to substantiate the effects on expression and causality.

In summary, through analysis of imputed SNP data in 870 German cases and 5293 German control subjects, followed by replication in 1383 additional German cases and 1728 control subjects, we could confirm 14 of the previously established 15 European AD susceptibility loci and, for the first time, replicate 5 of 8 loci thus far only reported in Asians and identified 2 new susceptibility loci with genome-wide significance in the combined analysis at chromosomes 2q24.3 (*XIRP2*) and 9p21.3

(*DMRTA1*). We provided preliminary functional evidence for a regulatory potential of the lead variants at these 2 novel loci, but further fine mapping and functional analyses will be needed to clarify whether the lead SNP is the true causal variant and to further elucidate its functional role. The 2 novel loci increase the explained heritability for AD in Europeans from 19.2% to 21.5% (see Table E5 in this article's Online Repository at www.jacionline.org).

We thank all the participating probands.

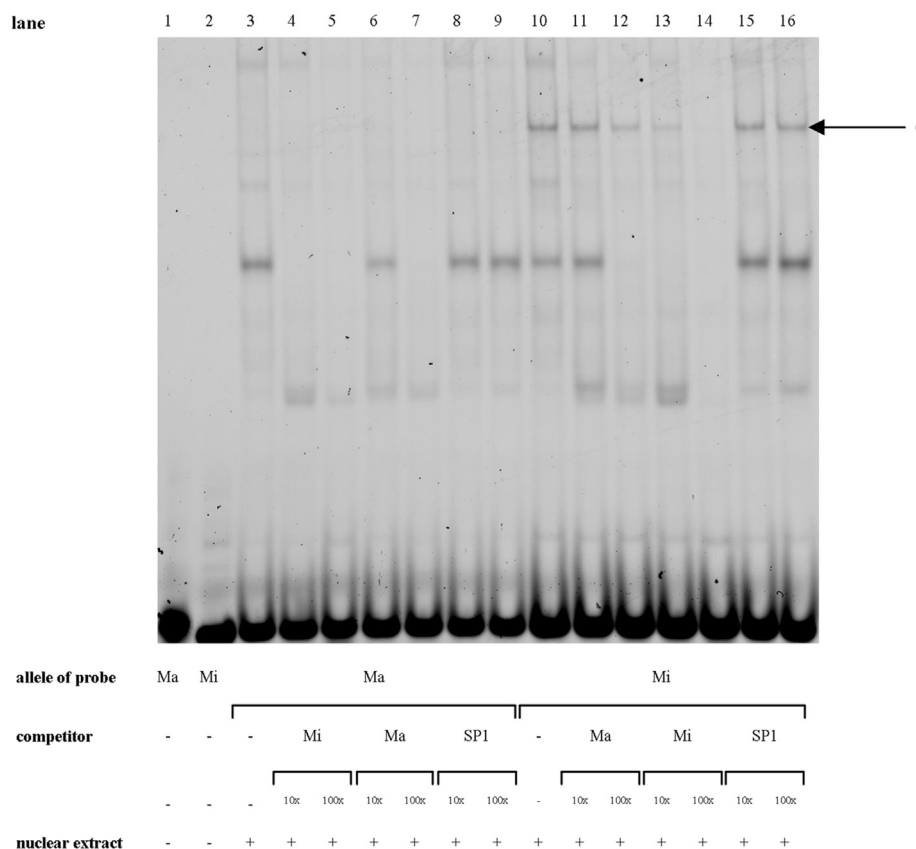


FIG 1. Allele-specific protein-DNA interactions of rs6720763 (*XIRP2*). Only the probe containing the minor (*Mi*; risk) allele shows allele-specific protein binding (lane 10, arrow c) compared with the major (*Ma*) allele (lane 3). Unlabeled minor allele probes efficiently competed with labeled minor allele probes (lanes 13 and 14) in contrast to the major allele (lanes 11 and 12) or SP1 (lanes 15 and 16).

Heidi Schaarschmidt, *ME^{a,*}*
 David Ellinghaus, *PhD^{a,*}*
 Elke Rodríguez, *PhD^b*
 Anja Kretschmer, *PhD^{b,c}*
 Hansjörg Baurecht, *MSc^b*
 Simone Lipinski, *PhD^d*
 Ulf Meyer-Hoffert, *MD, PhD^b*
 Jürgen Harder, *PhD^b*
 Wolfgang Lieb, *MD^d*
 Natalija Novak, *MD^e*
 Regina Fölster-Holst, *MD^b*
 Jorge Esparza-Gordillo, *PhD^{f,g}*
 Ingo Marenholz, *PhD^{f,g}*
 Franz Ruschendorf, *PhD^f*
 Norbert Hubner, *PhD^f*
 Eva Reischl, *PhD^c*
 Melanie Waldenberger, *PhD^c*
 Christian Gieger, *PhD^c*
 Thomas Illig, *PhD^h*
 Michael Kabesch, *MDⁱ*
 Xue-Jun Zhang, *MD^j*
 Feng-Li Xiao, *PhD^j*
 Young-Ae Lee, *MD^{k,g,*}*
 Andre Franke, *PhD^{a,*}*
 Stephan Weidinger, *MD^{b,*}*

Epidemiology and Institute of Epidemiology II, Helmholtz Zentrum München, German Research Center for Environmental Health, Neuherberg, Germany; ^athe Department of Dermatology and Allergy, University of Bonn, Bonn, Germany; ^bMax-Delbrück-Centrum (MDC) for Molecular Medicine, Berlin-Buch, Germany; ^cPediatric Allergy, Experimental and Clinical Research Center, Universitätsmedizin Berlin, Berlin, Germany; ^dHannover Unified Biobank, Hannover Medical School, Hannover, Germany; ^ethe Department of Pediatric Pneumology and Allergy, KUNO University Children's Hospital Regensburg, Regensburg, Germany; and ^fthe Institute of Dermatology at No. 1 Hospital, Anhui Medical University, Hefei, China. E-mail: sweidinger@dermatology.uni-kiel.de.

*These authors contributed equally to this work.

The project received infrastructure support through the DFG Cluster of Excellence "Inflammation at Interfaces" (grants EXC306 and EXC306/2) and was supported by the German Federal Ministry of Education and Research (BMBF) within the framework of the e:Med research and funding concept (sysINFLAME, grant no. 01ZX1306A) and the PopGen 2.0 network (01EY1103). The KORA research platform was initiated and financed by the Helmholtz Center Munich, German Research Center for Environmental Health, which is funded by the German Federal Ministry of Education and Research (BMBF) and by the State of Bavaria.

Disclosure of potential conflict of interest: N. Novak has received research support from the Germany Research Council (SFB704) and ALK-Abelló, has received consultancy fees from Leti Pharma and HAL Allergy, and has received lecture fees from Novartis, GlaxoSmithKline, MSD, ALK-Abelló, Bencard Allergy Therapeutics, HAL Allergy, and Stalergenes. M. Kabesch has received research support from the European Union (KBBE-2007-2-2-06, LSH-2004-1.2.5-1), the German Ministry of Education and Research (01ZX1306E, 01GS0810), and the German Research Foundation (SFB587B16) and has received lecture fees from European Respiratory Society, the European Academy of Allergy and Clinical Immunology, the American Thoracic Society, Novartis, and GlaxoSmithKline. S. Weidinger has received research support from NGFN (01GS0809, 01GS1818), Novartis, Pfizer, and Biogen Idec; is a board member for Novartis, Astellas, and Nutricia; has received consultancy fees from Novartis, Astellas, Pfizer, and Galderma; and has received lecture fees from Novartis, Pfizer, Pierre-Fabre, and Roche-Posay. The rest of the authors declare that they have no relevant conflicts of interest.

From ^athe Institute of Clinical Molecular Biology and ^dthe Institute of Epidemiology and Biobank PopGen, Christian-Albrechts-Universität of Kiel, Kiel, Germany; ^bthe Department of Dermatology, Venereology and Allergology, University Hospital Schleswig-Holstein, Campus Kiel, Kiel, Germany; ^cthe Research Unit of Molecular

REFERENCES

1. Ellinghaus D, Baurecht H, Esparza-Gordillo J, Rodriguez E, Matanovic A, Marenholz I, et al. High-density genotyping study identifies four new susceptibility loci for atopic dermatitis. *Nat Genet* 2013;45:808-12.
2. Weidinger S, Willis-Owen SA, Kamatani Y, Baurecht H, Morar N, Liang L, et al. A genome-wide association study of atopic dermatitis identifies loci with overlapping effects on asthma and psoriasis. *Hum Mol Genet* 2013;22:4841-56.
3. Esparza-Gordillo J, Weidinger S, Folster-Holst R, Bauerfeind A, Ruschendorf F, Patone G, et al. A common variant on chromosome 11q13 is associated with atopic dermatitis. *Nat Genet* 2009;41:596-601.
4. Esparza-Gordillo J, Schaarschmidt H, Liang L, Cookson W, Bauerfeind A, Lee-Kirsch MA, et al. A functional IL-6 receptor (IL6R) variant is a risk factor for persistent atopic dermatitis. *J Allergy Clin Immunol* 2013;132:371-7.
5. Weidinger S, Gieger C, Rodriguez E, Baurecht H, Mempel M, Klopp N, et al. Genome-wide scan on total serum IgE levels identifies FCER1A as novel susceptibility locus. *PLoS Genet* 2008;4:e1000166.
6. Sun LD, Xiao FL, Li Y, Zhou WM, Tang HY, Tang XF, et al. Genome-wide association study identifies two new susceptibility loci for atopic dermatitis in the Chinese Han population. *Nat Genet* 2011;43:690-4.
7. Paternoster L, Standl M, Chen CM, Ramasamy A, Bonnelykke K, Duijts L, et al. Meta-analysis of genome-wide association studies identifies three new risk loci for atopic dermatitis. *Nat Genet* 2012;44:187-92.
8. Hirota T, Takahashi A, Kubo M. Genome-wide association study identifies eight new susceptibility loci for atopic dermatitis in the Japanese population. *Nat Genet* 2012;44:1222-6.
9. Pacholsky D, Vakeel P, Himmel M, Lowe T, Stradal T, Rottner K, et al. Xin repeats define a novel actin-binding motif. *J Cell Sci* 2004;117:5257-68.
10. Panigrahi AK, Zhang N, Otta SK, Pati D. A cohesin-RAD21 interactome. *Biochem J* 2012;442:661-70.
11. Ottolenghi C, Fellous M, Barbieri M, McElreavey K. Novel paralogy relations among human chromosomes support a link between the phylogeny of doublesex-related genes and the evolution of sex determination. *Genomics* 2002;79:333-43.
12. Nica AC, Parts L, Glass D, Nisbet J, Barrett A, Sekowska M, et al. The architecture of gene regulatory variation across multiple human tissues: the MuTHER study. *PLoS Genet* 2011;7:e1002003.

<http://dx.doi.org/10.1016/j.jaci.2015.01.047>

A genome-wide association study reveals two new susceptibility loci for atopic dermatitis

Supplementary information

Heidi Schaarschmidt, ME^{1*}, David Ellinghaus, PhD^{1*}, Elke Rodríguez, PhD², Anja Kretschmer, PhD^{2,3}, Hansjörg Baurecht, MSc², Simone Lipinski, PhD¹, Ulf Meyer-Hoffert, MD PhD², Jürgen Harder, PhD², Wolfgang Lieb, MD⁴, Natalija Novak, MD⁵, Regina Fölster-Holst, MD², Jorge Esparza-Gordillo, PhD^{6,7}, Ingo Marenholz, PhD^{6,7}, Franz Ruschendorf, PhD⁶, Norbert Hubner, PhD⁶, Eva Reischl, PhD³, Melanie Waldenberger, PhD³, Christian Gieger, PhD³, Thomas Illig, PhD⁸, Michael Kabesch, MD⁹, Xue-Jun Zhang, MD¹⁰, Feng-Li Xiao, PhD¹⁰, Young-Ae Lee, MD^{6,7*}, Andre Franke, PhD^{1*}, Stephan Weidinger, MD^{2*}

1 Institute of Clinical Molecular Biology, Christian-Albrechts-University of Kiel, Kiel, Germany.

2 Department of Dermatology, Venereology and Allergology, University Hospital Schleswig-Holstein, Campus Kiel, Kiel, Germany.

3 Research Unit of Molecular Epidemiology and Institute of Epidemiology II, Helmholtz Zentrum München, German Research Center for Environmental Health, Neuherberg, Germany.

4 Institute of Epidemiology and Biobank PopGen, Christian-Albrechts-University of Kiel, Kiel, Germany.

2

5 Department of Dermatology and Allergy, University of Bonn, Bonn.

6 Max-Delbrück-Centrum (MDC) for Molecular Medicine, Berlin-Buch, Germany.

7 Pediatric Allergy, Experimental and Clinical Research Center, Universitätsmedizin
Berlin, Berlin, Germany.

8 Hannover Unified Biobank, Hannover Medical School, Hannover, Germany.

9 Department of Pediatric Pneumology and Allergy, KUNO University Children's
Hospital Regensburg, Regensburg, Germany.

10 Institute of Dermatology at No.1 Hospital, Anhui Medical University, Hefei, China.

*These authors contributed equally

Corresponding author:

Prof. Dr. Stephan Weidinger

Department of Dermatology, Venereology and Allergology

University Hospital Schleswig-Holstein, Campus Kiel

Schittenhelmstrasse 7

24105 Kiel

Tel.: +49-431-597-2732

Fax: +49-431-597-1815

Mail: sweidinger@dermatology.uni-kiel.de

Supplementary Methods

Study participants

All cases and controls of the discovery GWAS set and the replication set were of German origin. The institutional ethics review boards of the participating centers had approved the study protocol, and written informed consent had been obtained from all study participants or their legal guardians, respectively.

For the actual GWAS set a total of 924 unrelated AD cases recruited in German university hospitals (Charité Universitätsmedizin Berlin, University of Kiel, Technical University Munich) were used. The diagnosis of AD was made by experienced physicians according to the standard criteria of Hanifin and Rajka and the UK Working Party^{1, 2}. The 5,506 controls were obtained from the PopGen Biobank³, the KORA S3 and S4 survey, an independent population-based sample from the general population living in the region of Augsburg, southern Germany⁴, and from ISAAC Phase II study⁵. The replication set consisted of 1,415 independent AD cases from Germany (University of Kiel, Technical University Munich and Charité Universitätsmedizin Berlin) and further 1,748 independent controls from PopGen Biobank and Charité Universitätsmedizin Berlin. **Table E1** provides a summary of the study groups.

Genome-wide SNP genotyping, quality control (QC) and imputation

Initially, the GWAS marker dataset comprised 934,968 SNPs. SNPs on the X, Y and mitochondrial chromosomes as well as CNV-related SNPs were excluded. SNPs that had > 5% missing data, a minor allele frequency (MAF) <1% and an exact Hardy-Weinberg equilibrium in controls $P_{\text{HWE}} \leq 10^{-4}$ were further excluded. Samples with genotyping call rate (CR) $\leq 90\%$ were removed. We excluded samples from each pair of unexpected duplicates or relatives, as well as samples with outlier heterozygosity of ± 5 standard deviation (s.d.) away from the mean. The remaining GWAS samples were tested for population stratification using the principal components stratification method, as implemented in EIGENSTRAT, and population outliers were subsequently excluded. After this stringent quality control, the genotype data of 719,603 autosomal SNPs in 870 cases and 5,293 controls were available for imputation.

Out of the 2,543,887 imputed SNPs, we analyzed only those SNPs that could be imputed with high confidence (estimated r^2 between imputed and true genotypes > 0.8) and had a MAF $> 1\%$ in cases or in controls ($n=1,623,390$). To take imputation uncertainty into account, association analysis between the phenotype and the dosage data (expected allele counts) was performed using the logistic regression framework for dosage data, as implemented in MACH2DAT⁶. To control for potential confounding due to population stratification, we adjusted for the top ten eigenvectors from EIGENSTRAT as covariates in the regression analysis.

Replication SNP genotyping and QC using SNPLex and TaqMan

In order to validate the initial GWAS association results we genotyped 98 SNPs in the replication set using ligation-based SNPLex- or TaqMan technology from Applied Biosystems as previously described⁷. Before association analysis the following quality control criteria were applied using PLINK v.1.07⁸ to the genotype data: individual call rate (CR) had to be $\geq 50\%$ [32 cases and 20 controls were removed], exact Hardy-Weinberg equilibrium in controls $P_{HWE} \geq 10^{-4}$ [4 SNPs failed] and SNP call rate $\geq 90\%$ [25 SNPs failed]. Subsequently, 1,383 AD cases, 1,728 controls and 69 SNPs remained for replication analysis after quality control. Statistical analysis of genotype data was carried out using PLINK. Replication and joint P -values were calculated using PLINK's meta-analysis function with its standard error of odds ratio weighting option (inverse variance weighting).

Estimation of explained heritability for AD

Calculation of the heritability explained by all AD risk markers was performed using the method described by So et al.⁹. Under the assumption of a disease prevalence of 15%, we combined the explained heritability estimates for each single established locus including the two newly identified loci. Risk allele frequencies and OR for reported loci were extracted from the following references^{10, 11}, whereas for the new loci we used data from the GWAS screen.

In-silico data

To assess the potential functional relevance of candidate genes (*XIRP2* and *DMRTA1*) online databases were screened. In particular we used STRING (v9.1), a Search Tool for the Retrieval of Interacting Genes/Proteins (<http://string-db.org/>). This automated text mining approach searches primary databases that hold the experimental data as well as hand-curated databases serving expert annotations¹².

DNaseI hypersensitive sites, histone marks for regulatory regions and Chip-Seq data from the “ENCyclopedia of DNA Elements”¹³ (GRCh37/hg19, all “Integrated Regulation from ENCODE Tracks”) was used for predictions of the functional relevance of the associated SNPs. The Genomatix software was used for the prediction of transcription factor binding sites which were altered by SNPs¹⁴. Sequence conservation was analyzed by using DCODE on the ECR browser comparing the conservation between human, rhesus, dog, and mouse (high > 50% conservation, low < 50% conservation)¹⁵. Furthermore, we performed expression quantitative trait loci (eQTL) analyses using the Genevar database with the implemented MuTHER study data^{16,17}. For *in-silico* analyses all SNPs in full LD ($r^2=1$, based on 1000G data) with the *DMRTA1* and *XIRP2* lead SNPs were included.

Expression Analysis

A transcript expression study of *XIRP2* and *DMRTA1* was conducted using a human cDNA tissue panel (Clontech, Palo Alto, CA), cDNA from adult human skin (Alpha Diagnostics international, San Antonio, TX) as well as RNA extracted from primary keratinocytes isolated

from human foreskin obtained at the Department of Dermatology, University Hospital Schleswig-Holstein, Kiel and cultured under standard conditions¹⁸. We applied sequence specific intron spanning primers (*XIRP2* forward primer 5'->3': TCAAGAAGCAGCCAGGAAAT and reverse primer 5'->3': GGGCAGACTTTTCAAAGTGC; *DMRTA1* forward primer 5'->3': CTTGAGACAGGCCAGTGGTT and reverse primer 5'->3': TCTTCTTGTCCATTCTGGCA) and standard PCR conditions (Tanneal: 61°C and 60°C, respectively). Reactions were carried out on the Thermocycler 96well GeneAmp 9700 (Life Technologies GmbH, Darmstadt, Germany) and visualized on 2% agarose gels. For the negative amplification control sterilized water was used and *β-actin* / *GAPDH* (for keratinocytes) served as a loading control.

Immunohistochemistry

Histological sections were obtained from 4mm punch biopsies from lesional skin of AD patients and healthy volunteers recruited at the Department of Dermatology, University Hospital Schleswig-Holstein, Kiel. Immunohistochemical staining of paraffin-embedded tissue was done by using monoclonal rabbit anti-XIRP2 antibody (Sigma, Hamburg, Germany; dilution 1:50) and rabbit anti-DMRTA1 antibody (Novus Biologicals, Littleton, CO; dilution 1:200), followed by a biotinylated secondary goat anti-rabbit IgG antibody (Dako Cytomation, Hamburg, Germany; dilution 1:100) and subsequently incubated with Vector Universal DAB Kit (Vector, Burlingame, CA). Counterstaining was done with hematoxylin.

Nuclear protein extraction and Electrophoretic Mobility Shift Assay (EMSA)

In order to investigate allele-specific protein-DNA interactions for the identified risk variants, we performed EMSAs using the nontumorigenic human skin keratinocyte cell line HaCaT (CLS, Cell Lines Service GmbH, Eppelheim, Germany). Nuclear extracts of HaCaTs were produced by using the Nuclear Extract Kit (Active Motif). Protein concentration was determined by the BCA Protein Assay Reagent (Thermo Scientific). 5' Cy5-labeled and unlabeled oligonucleotides containing the major or minor allele of rs6720763/ rs10738626 or the consensus sequence for Specificity protein 1 (SP1) were purchased from Metabion (**Table E6**). Oligonucleotides were annealed to obtain double-stranded DNA probes. Binding reaction was carried out with or without different concentrations of unlabeled competitor oligonucleotides using 5 µg of nuclear extract in 1 x binding buffer (4% v/v Glycerol, 1 mM MgCl₂, 0.5 mM EDTA, 0.5 mM DTT, 50 mM NaCl, 10 mM TrisHCl pH7.5) with 0.5 µg poly dI-dC (Roche Diagnostics) and 1 ng of labeled probe in a total volume of 10 µl for 20 min at 4 °C. Protein-DNA complexes were separated on a 5.3% polyacrylamide gel by electrophoresis in 0.5 x tris-borate-EDTA (TBE) buffer. Band patterns were visualized by scanning the gel with the Typhoon Trio + (GE Healthcare). All EMSA experiments were repeated at least once. Technical validation always yielded identical results.

Supplementary Results

New AD susceptibility loci

Another potential candidate SNP, rs1665050 within intron 1 of *RNF111*, reached a *P*-value close to genome-wide significance in the combined analysis ($P_{\text{comb}} = 9.65 \times 10^{-8}$, OR = 1.25), see **Table 1** in the main manuscript. *RNF111* encodes a nuclear RING-domain containing E3 ubiquitin ligase Arcadia which enhances TGF- β signaling through ubiquitin-dependent degradation of different intracellular proteins¹⁹. TGF- β signaling is essential for the maintenance of immune tolerance²⁰, and impaired TGF- β signaling has been implicated in inflammatory and autoimmune processes, and promote Th2-mediated phenotypes^{21,22} through dysregulation of regulatory T-cell (T_{Reg}) activity²³.

In-silico data and in vitro experiments

Using STRING v9.1 (the Search Tool for the Retrieval of Interacting Genes/Proteins)¹², we identified *NSMAF* (Neutral Sphingomyelinase (N-SMase) Activation Associated Factor) as interaction partner of *XIRP2* based on experiments ($P = 5.65 \times 10^{-9}$). NSMAF interacts with TNF-p55 and leads to the activation of the N-SMase pathway, which mediates inflammation²⁴. N-SMase is an enzyme which is responsible for the breakdown of sphingomyelin into

phosphocholine and ceramide. A reduced SMase activity is correlated with decreased ceramide production²⁵, which is supposed to play a role in AD pathogenesis^{26,27}.

Only one common variant, rs7569147, tags the *XIRP2* lead SNP with complete LD. Here, no alterations in transcription factor binding are predicted and no binding of transcription factors has been observed (**Table E3a and E3b**). Variant rs10738626 upstream of *DMRTA1* is in complete LD with 37 other common variants ($r^2=1$). According to the ENCODE, DCODE and Genomatix data none of the *DMRTA1* SNPs is favorable over the other SNPs. Therefore we proceeded with the SNP with the lowest p-value (rs10738626) (**Table E4a and E4b**).

To characterize whether the SNPs cause differential transcription factor binding which might highlight a potential functional relevance of these variants, polymorphism-specific electrophoretic mobility shift assays (EMSAs) with Cy5-labeled oligonucleotides were performed using nuclear extracts derived from human HaCaT keratinocytes. An additional DNA-protein complex (c) was observed only in the presence of the risk-associated minor allele C of rs6720763 in *XIRP2* (**Figure 1**, lane 10), but not for the major allele (**Figure 1**, lane 3-9). Specificity of the observed protein-DNA-complex was demonstrated by competition shifts with different amounts of unlabeled probes (10%, 100%), which revealed an efficient competition for probes containing the minor allele (**Figure 1**, lanes 13+14) in contrast to oligonucleotides with the major allele and SP1

(**Figure 1**, lanes 11+12, 15+16, respectively). Free probes of both major and minor alleles without incubation with nuclear extract served as negative control (**Figure 1**, lanes 1+2). In the EMSA analysis of rs10738626 (*DMRTA1*) five additional complex formations (c1-c5) were detectable in nuclear extracts from HaCaT keratinocytes. Two additional complex formations (c1+c2) could be shown for the risk-associated major allele T (**Figure E6**, lane 3), and three additional complex formations (c3-c5) for the protective minor allele C (**Figure E6**, lane 10). Competition testing revealed clear specificity for c2 and c5 (**Figure E6**, lanes 4-9, 11-16, respectively), and a more efficient competition for c1, c3 and c4 (**Figure E6**, lanes 4-9 for c1, 11-16 for c3+c4). Free probes of both major and minor alleles without incubation with nuclear extract served as negative control (**Figure E6**, lanes 1+2).

Supplementary Tables

Table E1: Distribution of the sample sets before and after applying quality control (QC) criteria.

imputed GWAS			replication panel	
before QC	924	5,506	1,415	1,748
after QC	870	5,293	1,383	1,728
center	Cases	controls	cases	controls
Kiel (CAU/PopGen)	275	1,227	424	1,457
Munich (TU)	236	–	638	–
Berlin (Charité)	359	–	321	271
Munich(KORA F4)	–	1,775	–	–
Munich (KORA F3)	–	1,619	–	–
Hanover (ISAAC)	–	672	–	–

Table E2: Association results of 69 SNPs selected for replication. A1: minor allele; A2: major allele; AF_ca/AF_co: allele frequencies in cases/controls; 11/12/22_ca/co: genotype counts cases/controls; P_{GWAS} , P_{Repl} , P_{comb} : P -values in GWAS, replication, combined analysis. Odds ratios (OR) and 95% confidence intervals (CI) for allele A1 are shown. Top SNPs per locus with P -values $< 5 \times 10^{-7}$ in the combined analysis are highlighted in grey / bold italics.

dbSNP ID	chr	pos. (bp)	A1,A2	GWAS				AF_ca	replication				combined		
				AF_ca	AF_co	P_{GWAS}	OR (95% CI)		11/12/22_ca	AF_co	11/12/22_co	P_{Repl}	OR (95% OR)	P_{comb}	OR
rs10489149	1	14127254	G,C	0.137	0.103	2.89×10^{-4}	1.34 (1.15-1.56)	0.101	18/235/1085	0.091	20/263/1377	1.91×10^{-1}	1.12 (0.94-1.33)	6.38×10^{-2}	1.24
rs620478	1	38972814	G,A	0.15	0.11	6.50×10^{-6}	1.44 (1.23-1.67)	0.125	15/298/999	0.137	29/411/1273	1.75×10^{-1}	0.9 (0.77-1.05)	2.18×10^{-2}	0.79
rs1341341	1	56505067	A,G	0.376	0.424	8.74×10^{-5}	0.81 (0.73-0.9)	0.424	244/672/451	0.42	299/816/567	7.56×10^{-1}	1.02 (0.92-1.13)	1.27×10^{-2}	0.91
rs12731002	1	56547434	G,A	0.023	0.046	5.95×10^{-6}	0.49 (0.34-0.68)	0.05	0/132/1193	0.043	3/141/1558	2.23×10^{-1}	1.16 (0.91-1.48)	1.68×10^{-1}	1.4
rs12144049	1	150707534	C,T	0.369	0.279	1.87×10^{-8}	1.47 (1.31-1.64)	0.346	173/570/579	0.286	132/666/829	5.89×10^{-7}	1.33 (1.19-1.48)	1.02×10^{-16}	1.39
rs6704503	1	150707919	A,G	0.489	0.416	9.48×10^{-5}	1.33 (1.2-1.48)	0.483	321/628/365	0.422	269/815/518	3.21×10^{-6}	1.28 (1.15-1.42)	1.85×10^{-12}	1.31
rs11265282	1	158041032	C,T	0.12	0.158	2.94×10^{-4}	0.75 (0.64-0.88)	0.157	40/335/947	0.145	54/387/1262	2.10×10^{-1}	1.1 (0.95-1.26)	1.57×10^{-1}	0.93
rs7586380	2	13535344	T,C	0.259	0.219	1.01×10^{-4}	1.27 (1.14-1.43)	0.238	69/489/757	0.222	80/597/1030	1.26×10^{-1}	1.1 (0.97-1.24)	9.65×10^{-5}	0.93
rs1344915	2	60752167	T,G	0.338	0.394	1.54×10^{-5}	0.79 (0.71-0.88)	0.384	191/649/502	0.389	234/816/602	7.23×10^{-1}	0.98 (0.88-1.09)	1.34×10^{-3}	0.88
rs1362349	2	102318404	C,G	0.529	0.481	7.30×10^{-4}	1.19 (1.08-1.33)	0.497	314/677/322	0.5	426/846/427	8.32×10^{-1}	0.99 (0.89-1.1)	1.19×10^{-2}	1.09
rs10931651	2	149955546	A,G	0.199	0.162	4.15×10^{-5}	1.33 (1.17-1.53)	0.176	40/384/894	0.165	43/473/1180	2.50×10^{-1}	1.08 (0.95-1.24)	1.63×10^{-4}	1.2
rs6720763	2	167700532	C,T	0.221	0.18	5.03×10^{-5}	1.31 (1.15-1.48)	0.211	51/466/832	0.174	43/508/1159	2.65×10^{-4}	1.27 (1.12-1.44)	4.37×10^{-8}	1.29
rs6731283	2	167782076	T,G	0.281	0.235	2.33×10^{-6}	1.36 (1.19-1.54)	0.253	67/539/722	0.233	82/601/959	6.75×10^{-2}	1.12 (0.99-1.26)	4.23×10^{-6}	0.92
rs6746061	2	167782139	A,T	0.172	0.131	2.25×10^{-5}	1.38 (1.19-1.59)	0.165	34/377/937	0.15	37/439/1234	1.08×10^{-1}	1.12 (0.98-1.29)	3.49×10^{-5}	1.24
rs6759681	2	167782212	G,A	0.118	0.083	8.88×10^{-6}	1.48 (1.25-1.75)	0.09	10/224/1116	0.087	13/271/1427	6.24×10^{-1}	1.05 (0.88-1.25)	2.92×10^{-4}	0.83
rs4673964	2	215812151	C,T	0.308	0.355	3.54×10^{-5}	0.79 (0.7-0.88)	0.354	174/609/568	0.345	195/783/723	4.45×10^{-1}	1.04 (0.94-1.16)	2.59×10^{-2}	0.92
rs873838	3	25388042	C,A	0.516	0.473	6.56×10^{-5}	1.24 (1.11-1.38)	0.469	278/670/360	0.476	383/836/464	5.76×10^{-1}	0.97 (0.88-1.08)	1.90×10^{-2}	1.09
rs6797592	3	69601236	C,T	0.105	0.137	4.70×10^{-5}	0.71 (0.6-0.84)	0.138	28/309/989	0.13	35/377/1302	4.11×10^{-1}	1.06 (0.92-1.24)	5.22×10^{-2}	0.89
rs230515	4	103690463	C,T	0.303	0.35	1.81×10^{-4}	0.8 (0.71-0.9)	0.313	122/582/617	0.327	189/735/778	2.37×10^{-1}	0.94 (0.84-1.04)	6.45×10^{-4}	1.07
rs10428456	4	160405734	C,T	0.169	0.218	9.87×10^{-4}	0.71 (0.62-0.82)	0.205	56/436/846	0.216	83/576/1062	3.05×10^{-1}	0.94 (0.83-1.06)	7.80×10^{-5}	0.83

rs16872847	5	4187335	T,C	0.42	0.472	8.11x10 ⁻⁵	0.81 (0.72-0.9)	0.445	257/673/405	0.458	349/862/494	3.16x10 ⁻¹	0.95 (0.86-1.05)	6.21x10 ⁻⁴	1.08
rs12654436	5	6605278	G,A	0.278	0.323	1.56x10 ⁻⁴	0.8 (0.71-0.9)	0.289	101/570/666	0.301	147/703/805	2.92x10 ⁻¹	0.94 (0.84-1.05)	8.33x10 ⁻⁴	1.08
rs16899437	5	81592502	G,A	0.088	0.059	2.06x10 ⁻⁵	1.55 (1.28-1.89)	0.059	3/140/1103	0.059	6/189/1520	9.98x10 ⁻¹	1 (0.8-1.25)	9.43x10 ⁻⁴	0.78
rs37555	5	82259439	G,C	0.149	0.116	1.38x10 ⁻⁵	1.41 (1.22-1.64)	0.124	23/284/1023	0.131	27/391/1284	4.40x10 ⁻¹	0.94 (0.81-1.1)	9.62x10 ⁻³	0.82
rs13354690	5	130858516	G,A	0.012	0.032	3.48x10 ⁻⁵	0.43 (0.28-0.67)	0.025	1/66/1284	0.016	0/53/1608	1.13x10 ⁻²	1.59 (1.11-2.29)	7.07x10 ⁻¹	1.85
rs10463891	5	131625291	A,G	0.387	0.339	4.38x10 ⁻⁵	1.26 (1.13-1.4)	0.369	173/635/523	0.359	215/783/694	4.19x10 ⁻¹	1.04 (0.94-1.16)	5.39x10 ⁻⁴	1.14
rs3091307	5	132017035	G,A	0.276	0.228	5.42x10 ⁻⁵	1.3 (1.14-1.47)	0.283	110/545/697	0.247	110/608/957	1.69x10 ⁻³	1.2 (1.07-1.35)	4.18x10⁻⁷	1.24
rs17690965	5	132058566	C,G	0.308	0.264	4.88x10 ⁻⁴	1.23 (1.09-1.38)	0.327	148/570/608	0.286	140/691/868	6.18x10 ⁻⁴	1.21 (1.09-1.35)	1.02x10 ⁻⁶	1.22
rs1006329	5	168532429	G,A	0.238	0.297	6.14x10 ⁻⁴	0.74 (0.65-0.83)	0.328	141/594/602	0.322	171/765/784	6.31x10 ⁻¹	1.03 (0.92-1.14)	4.56x10 ⁻³	1.16
rs457759	6	5060595	C,A	0.438	0.492	9.29x10 ⁻⁵	0.81 (0.73-0.9)	0.461	280/681/384	0.472	382/798/475	4.15x10 ⁻¹	0.96 (0.87-1.06)	8.51x10 ⁻⁴	1.08
rs7765733	6	5097754	C,T	0.286	0.331	7.76x10 ⁻⁵	0.8 (0.71-0.89)	0.308	129/560/638	0.311	156/749/802	8.30x10 ⁻¹	0.99 (0.89-1.1)	3.90x10 ⁻³	0.89
rs9387633	6	98287263	G,A	0.292	0.248	9.82x10 ⁻⁵	1.26 (1.12-1.43)	0.268	96/519/712	0.263	105/641/871	6.81x10 ⁻¹	1.03 (0.91-1.15)	2.36x10 ⁻³	0.9
rs1636895	7	22301009	T,C	0.156	0.196	1.13x10 ⁻⁵	0.73 (0.63-0.84)	0.184	45/397/884	0.187	51/543/1127	7.09x10 ⁻¹	0.98 (0.86-1.11)	1.93x10 ⁻³	1.13
rs1636896	7	22301652	T,C	0.306	0.356	2.38x10 ⁻⁵	0.78 (0.69-0.88)	0.35	146/656/552	0.353	220/762/721	8.18x10 ⁻¹	0.99 (0.89-1.1)	2.86x10 ⁻³	1.11
rs6977628	7	28811671	T,C	0.325	0.269	5.77x10 ⁻⁶	1.3 (1.16-1.45)	0.299	133/527/665	0.273	126/681/900	2.63x10 ⁻²	1.14 (1.02-1.27)	1.41x10 ⁻⁶	0.93
rs2439279	8	32585951	G,A	0.42	0.481	3.45x10 ⁻⁵	0.8 (0.72-0.89)	0.448	286/637/425	0.447	353/807/531	9.33x10 ⁻¹	1 (0.91-1.11)	4.72x10 ⁻³	0.9
rs7829383	8	32660202	G,A	0.354	0.412	3.31x10 ⁻⁵	0.8 (0.71-0.88)	0.385	187/662/497	0.389	259/809/638	7.45x10 ⁻¹	0.98 (0.89-1.09)	1.87x10 ⁻³	1.11
rs13249751	8	109135758	A,T	0.262	0.218	3.63x10 ⁻⁴	1.25 (1.11-1.4)	0.27	93/522/697	0.266	107/685/895	7.70x10 ⁻¹	1.02 (0.91-1.14)	6.78x10 ⁻³	1.12
rs10738626	9	22363457	C,T	0.443	0.494	1.96x10 ⁻⁶	0.77 (0.69-0.86)	0.474	313/656/383	0.517	388/870/447	7.95x10 ⁻⁴	0.84 (0.76-0.93)	1.44x10⁻⁸	0.81
rs10813809	9	32391030	C,T	0.359	0.417	1.97x10 ⁻⁶	0.77 (0.69-0.86)	0.397	190/656/459	0.411	284/830/586	2.65x10 ⁻¹	0.94 (0.85-1.05)	6.28x10 ⁻⁵	0.86
rs16909765	9	122212339	A,C	0.187	0.151	1.17x10 ⁻⁵	1.37 (1.19-1.57)	0.145	22/342/967	0.155	36/454/1205	2.73x10 ⁻¹	0.92 (0.8-1.07)	1.38x10 ⁻²	1.13
rs11259268	10	14763910	A,G	0.136	0.17	6.62x10 ⁻⁵	0.74 (0.64-0.86)	0.184	54/395/917	0.168	43/487/1175	9.95x10 ⁻²	1.12 (0.98-1.28)	1.73x10 ⁻¹	0.93
rs7100925	10	56630929	C,T	0.344	0.29	7.78x10 ⁻⁶	1.3 (1.16-1.46)	0.314	139/553/631	0.302	141/745/816	3.01x10 ⁻¹	1.06 (0.95-1.18)	9.59x10 ⁻⁵	1.17
rs2093558	10	98130038	G,T	0.46	0.503	8.05x10 ⁻⁵	0.81 (0.73-0.90)	0.494	341/658/357	0.489	420/832/459	6.69x10 ⁻¹	1.02 (0.92-1.13)	2.24x10 ⁻³	1.12
rs1868997	11	14850796	T,C	0.325	0.37	2.61x10 ⁻⁵	0.79 (0.7-0.88)	0.368	172/663/535	0.36	232/744/701	5.53x10 ⁻¹	1.03 (0.93-1.15)	1.63x10 ⁻²	0.91
rs1945566	11	21559214	T,C	0.103	0.072	1.26x10 ⁻⁵	1.52 (1.27-1.82)	0.085	14/201/1129	0.073	12/229/1483	8.78x10 ⁻²	1.18 (0.98-1.42)	1.10x10 ⁻⁵	0.87
rs10430934	11	26606000	G,T	0.512	0.447	1.30x10 ⁻⁶	1.3 (1.17-1.45)	0.438	264/657/432	0.428	312/832/558	4.25x10 ⁻¹	1.04 (0.94-1.15)	1.04x10 ⁻⁴	1.16
rs3812724	11	78486119	G,A	0.099	0.074	8.63x10 ⁻⁵	1.46 (1.22-1.75)	0.084	7/213/1129	0.086	11/272/1432	8.26x10 ⁻¹	0.98 (0.82-1.17)	7.70x10 ⁻³	0.82
rs2555614	13	32686314	G,A	0.378	0.333	9.93x10 ⁻⁵	1.25 (1.11-1.39)	0.332	141/598/587	0.328	194/727/781	7.26x10 ⁻¹	1.02 (0.92-1.14)	3.08x10 ⁻³	0.91
rs842385	13	46155793	G,T	0.3	0.257	5.07x10 ⁻⁵	1.28 (1.14-1.43)	0.278	97/551/690	0.29	135/682/822	3.07x10 ⁻¹	0.94 (0.84-1.06)	3.66x10 ⁻²	0.86
rs7993551	13	88025453	A,G	0.106	0.137	1.76x10 ⁻⁵	0.7 (0.59-0.83)	0.143	23/326/950	0.139	37/396/1263	6.10x10 ⁻¹	1.04 (0.9-1.2)	1.94x10 ⁻²	0.88

rs1448564	14	40274788	C,A	0.204	0.165	3.56x10 ⁻⁵	1.33 (1.16-1.52)	0.185	44/406/889	0.178	48/493/1112	5.29x10 ⁻¹	1.04 (0.91-1.19)	6.09x10 ⁻⁴	0.89
rs7140785	14	56965594	T,G	0.137	0.094	6.36x10 ⁻⁶	1.46 (1.25-1.72)	0.13	24/303/1023	0.145	34/412/1214	1.03x10 ⁻¹	0.88 (0.76-1.03)	5.37x10 ⁻²	0.79
rs1665050	15	57080897	A,G	0.299	0.252	7.12x10 ⁻⁵	1.27 (1.13-1.43)	0.296	121/561/676	0.255	100/669/937	3.48x10 ⁻⁴	1.23 (1.1-1.38)	9.65x10⁻⁸	1.25
rs11630106	15	57244224	A,G	0.42	0.369	1.70x10 ⁻⁴	1.23 (1.1-1.36)	0.407	222/671/476	0.384	264/769/657	6.14x10 ⁻²	1.1 (1-1.22)	7.24x10 ⁻⁵	1.16
rs4965117	15	96293254	C,T	0.293	0.246	1.03x10 ⁻⁴	1.26 (1.12-1.41)	0.267	95/521/715	0.256	128/616/958	3.36x10 ⁻¹	1.06 (0.94-1.19)	5.93x10 ⁻⁴	1.15
rs7193563	16	17933165	C,G	0.245	0.292	5.82x10 ⁻⁵	0.78 (0.69-0.88)	0.309	134/577/658	0.28	134/691/890	1.28x10 ⁻²	1.15 (1.03-1.28)	4.49x10 ⁻¹	0.97
rs11859061	16	19562272	T,C	0.29	0.335	6.58x10 ⁻⁵	0.79 (0.7-0.88)	0.333	143/604/588	0.335	182/744/727	8.83x10 ⁻¹	0.99 (0.89-1.11)	5.0x10 ⁻³	1.11
rs1581054	16	54654634	C,T	0.421	0.475	9.28x10 ⁻⁴	0.84 (0.75-0.93)	0.451	239/702/368	0.44	334/822/538	3.98x10 ⁻¹	1.05 (0.94-1.16)	9.11x10 ⁻²	1.12
rs4796793	17	37795736	G,C	0.3	0.252	9.26x10 ⁻⁶	1.3 (1.16-1.47)	0.287	96/567/661	0.265	116/673/919	6.03x10 ⁻²	1.12 (1-1.25)	7.65x10 ⁻⁶	0.93
rs11664104	18	2984861	G,A	0.309	0.267	9.51x10 ⁻⁵	1.26 (1.12-1.41)	0.246	73/513/751	0.251	104/642/950	7.11x10 ⁻¹	0.98 (0.87-1.1)	1.11x10 ⁻²	0.88
rs17637670	18	2986922	G,A	0.197	0.152	1.66x10 ⁻⁶	1.42 (1.23-1.64)	0.149	29/344/973	0.159	51/441/1211	2.80x10 ⁻¹	0.93 (0.8-1.07)	6.62x10 ⁻³	0.81
rs12373294	18	39920535	C,T	0.343	0.294	3.23x10 ⁻⁵	1.27 (1.13-1.42)	0.334	142/595/581	0.312	164/725/799	7.58x10 ⁻²	1.1 (0.99-1.23)	3.0x10 ⁻⁵	1.18
rs17808190	18	54168503	T,G	0.253	0.297	1.96x10 ⁻⁴	0.8 (0.7-0.9)	0.294	112/565/663	0.287	153/668/874	5.46x10 ⁻¹	1.04 (0.93-1.16)	4.25x10 ⁻²	1.13
rs6110715	20	15592956	G,A	0.085	0.051	2.47x10 ⁻⁶	1.64 (1.35-2)	0.074	9/184/1172	0.066	11/204/1496	2.23x10 ⁻¹	1.13 (0.93-1.38)	1.89x10 ⁻⁵	0.83
rs2752914	20	51051356	C,T	0.469	0.409	2.92x10 ⁻⁵	1.26 (1.13-1.4)	0.428	228/657/414	0.431	319/820/553	8.50x10 ⁻¹	0.99 (0.89-1.1)	6.13x10 ⁻³	1.11
rs6013912	20	52235684	C,T	0.347	0.401	8.61x10 ⁻⁵	0.8 (0.71-0.89)	0.375	163/653/488	0.382	234/820/632	6.03x10 ⁻¹	0.97 (0.88-1.08)	2.07x10 ⁻³	0.89
rs2835541	21	37240765	G,T	0.4	0.446	6.20x10 ⁻⁴	0.83 (0.75-0.93)	0.446	261/638/403	0.444	322/794/505	8.84x10 ⁻¹	1.01 (0.91-1.12)	2.37x10 ⁻²	0.92
rs378376	21	43335510	C,A	0.334	0.282	3.65x10 ⁻⁵	1.27 (1.14-1.41)	0.301	109/569/630	0.321	181/728/789	9.50x10 ⁻²	0.91 (0.82-1.02)	8.38x10 ⁻²	0.85

Table E3a: Results of *in silico* analyses for *XIRP2*. Associated SNPs in LD = 1 with the lead SNP are listed. ENCODE: number of cell lines with positive DNaseI Hypersensitive Sites (DNaseI HS); chromatin state in keratinocytes (NHEC)/lymphocytes (NHLF); predicted number of transcription factor binding sites (TFBS) (details see Table E3b). Genomatix: loss/gain of TFBS. Genevar: expression quantitative trait loci (eQTLs) in lymphoblastoid cell lines (LCL) and skin. DCODE: conservation status.

		ENCODE					Genomatix		Genevar (Muther)		DCODE conservation
associated SNP (LD=1)	P_{GWAS}	gene	# of cell lines with DNaseI HS	chromatin state		# of TFBS	lost TFBS	new TFBS	LCL	skin	
				NHEC	NHLF				eQTL (<i>XIRP2</i>) evidence ($P \leq 0.05$)	eQTL (<i>XIRP2</i>) evidence ($P \leq 0.05$)	
rs6720763	5,03E-05	<i>XIRP2</i>	24	NA	strong enhancer	2	-	HBOX (Homeobox transcription factor) PAR/bZIP, ZFHX (Two-handed zinc finger homeodomain transcription factors)	NA	NA	high
rs7569147	7,59E-03	<i>XIRP2</i>	9	NA	weak enhancer	NA	-	-	NA	NA	high

Table E3b: Detailed ENCODE information of cell lines with DNaseI HS and predicted transcription factors listed in Table E3a.

associated SNP	cell lines with DNaseI HS	cell line (detail)	transcription factor (cell line)
rs6720763	AG04449	fetal buttock/thigh fibroblast	GATA2 (HUVEC)
	AG04450	fetal lung fibroblast	FOS (HUVEC)
	CD34+ mobilized	hematopoietic progenitor cells	
	CMK	acute megakaryocytic leukemia cells	
	HBMEC	brain microvascular endothelial cells	
	HCF	cardiac fibroblasts	
	HCFaa	cardiac fibroblasts- adult atrial	
	HCPEpiC	choroid plexus epithelial cells	
	HFF-Myc	foreskin fibroblast cells expressing canine cMyc	
	HIPEpiC	iris pigment epithelial cells	
	HMVEC-dAd	adult dermal microvascular endothelial cells	
	HMVEC-dBI-Neo	neonatal blood microvascular endothelial cells, dermal-derived	
	HMVEC-dLy-Ad	adult lymphatic microvascular endothelial cells, dermal-derived	
	HMVEC-LLy	lymphatic microvascular endothelial cells, lung-derived	
	HNPCEpiC	non-pigment ciliary epithelial cells	
	HPAF	pulmonary artery fibroblasts	
	HPF	pulmonary fibroblasts isolated from lung tissue	
	HRGEC	renal glomerular endothelial cells	
	HUVEC	umbilical vein endothelial cells	
	Jurkat	T lymphoblastoid derived from an acute T cell leukemia	
	NHDF-Ad	adult dermal fibroblasts	
	PANC-1	pancreatic carcinoma	
	SK-N-MC	neuroepithelioma cell line derived from a metastatic supra-orbital human brain tumor	
	WI-38	embryonic lung fibroblast cells	
rs7569147	HCPEpiC	choroid plexus epithelial cells	
	Jurkat	T lymphoblastoid derived from an acute T cell leukemia	
	NHDF-Ad	adult dermal fibroblasts	
	WI-38	embryonic lung fibroblast cells	
	HMF	mammary fibroblasts	
	HRPEpiC	retinal pigment epithelial cells	
	Medullo	Medulloblastoma	
	NT2-D1	malignant pluripotent embryonal carcinoma	
	WERI-Rb-1	Retinoblastoma	

Table E4a: Results of *in silico* analyses for *DMRTA1*. Associated SNPs in LD = 1 with the lead SNP are listed. ENCODE: number of cell lines with positive DNaseI Hypersensitive Sites (DNaseI HS); chromatin state in keratinocytes (NHEC)/lymphocytes (NHLF); predicted number of transcription factor binding sites (TFBS) (details see Table E4b). Genomatix: loss/gain of TFBS. Genevar: expression quantitative trait loci (eQTLs) in lymphoblastoid cell lines (LCL) and skin. DCODE: conservation status.

			ENCODE				Genomatix		Genevar (Muther)		DCODE conservation
associated SNP (LD=1)	P _{GWAS}	gene	chromatin state						LCL	skin	
			# of cell lines with DNaseI HS	NHEC	NHLF	# of TFBS	lost TFBS	new TFBS	eQTL (<i>DMRTA1</i>) evidence (P≤0.05)	eQTL (<i>DMRTA1</i>) evidence (P≤0.05)	
rs10738626	1,96E-06	<i>DMRTA1</i>	NA	NA	NA	NA	E2FF (E2F-myc activator/cell cycle regulator)	YTBP (Yeast TATA binding protein factor) ETSF (Human and murine ETS1 factors) AP2F (Activator protein 2)	NA	NA	low
rs10757319	NA		NA	NA	NA	NA	1	2	NA	NA	low
rs10757316	NA		NA	NA	NA	NA	-	2	NA	NA	low
rs10965335	NA		NA	NA	NA	NA	2	1	NA	NA	high
rs11536672	NA		NA	NA	NA	NA	3	1	NA	NA	high
rs11534173	NA		NA	NA	NA	NA	1	1	NA	NA	high
rs10811699	6,99E-05		NA	NA	NA	NA	1	1	NA	NA	high
rs10757315	NA		NA	NA	NA	NA	1	1	NA	NA	low
rs2383223	1,38E-04		NA	NA	NA	NA	4	-	NA	NA	high
rs7038483	NA		NA	NA	NA	NA	3	1	NA	NA	high
rs10811698	NA		NA	NA	NA	NA	-	-	NA	NA	low
rs1854491	NA		NA	NA	NA	NA	-	2	NA	NA	low
rs1854490	NA		NA	NA	NA	NA	-	1	NA	NA	low
rs10738625	3,00E-04		2	NA	NA	NA	1	4	NA	NA	low

rs1536462	2,92E-04	NA	NA	NA	NA	4	1	NA	NA	low
rs1536461	NA	NA	NA	NA	NA	1	1	NA	NA	low
rs1536460	3,02E-04	NA	NA	NA	NA	3	-	NA	NA	high
rs10811697	2,80E-04	NA	NA	NA	1	1	-	NA	NA	high
rs10811696	NA	NA	NA	NA	NA	1	1	NA	NA	low
rs10811695	2,77E-04	9	NA	NA	NA	2	4	NA	NA	low
rs10738624	NA	NA	NA	NA	NA	1	2	NA	NA	low
rs10757312	NA	NA	NA	NA	NA	2	2	NA	NA	low
rs10738623	NA	NA	NA	NA	NA	1	-	NA	NA	low
rs10757311	NA	NA	NA	NA	NA	1	4	NA	NA	low
rs1327063	2,78E-04	4	NA	NA	NA	1	-	NA	NA	high
rs1536459	2,67E-04	4	NA	NA	NA	3	2	NA	NA	high
rs943396	2,70E-04	NA	NA	NA	NA	3	2	NA	NA	low
rs10811694	2,47E-04	2	NA	NA	NA	1	5	NA	NA	low
rs7871660	NA	2	NA	NA	NA	1	4	NA	NA	low
rs1360136	3,32E-04	NA	NA	NA	NA	1	2	NA	NA	low
rs2025795	3,42E-04	NA	NA	NA	NA	4	1	NA	NA	low
rs7872794	3,76E-04	NA	NA	NA	NA	3	1	NA	NA	low
rs10757309	3,72E-04	NA	NA	NA	NA	1	-	NA	NA	high
rs1409775	NA	NA	NA	NA	NA	1	3	NA	NA	low
rs10811692	6,60E-04	NA	NA	NA	NA	1	1	NA	NA	low
rs1327061	3,57E-04	NA	NA	NA	5	1	3	NA	NA	high
rs1359741	4,03E-04	NA	NA	NA	NA	2	1	NA	NA	low
rs716579	NA	NA	NA	NA	NA	1	-	NA	NA	low

Table E4b: Detailed ENCODE information of cell lines with DNaseI HS and predicted transcription factors listed in Table E4a.

associated SNP	cell lines with DNaseI HS	cell line (detail)	transcription factor (cell line)
rs10738625	H1-hESC	embryonic stem cells	
	H7-hESC	embryonic stem cells	
rs10811697			MAFK (HepG2)
rs10811695	Caco-2	colorectal adenocarcinoma	
	Chorion	chorion cells	
	FibroP	fibroblasts taken from individuals with Parkinson's disease	
	GM12892	B-lymphocyte	
	GM19238	B-lymphocyte	
	H1-hESC	embryonic stem cells	
	HSMMtube	skeletal muscle myotubes differentiated from the HSMM cell line	
	iPS	induced pluripotent stem cell derived from skin fibroblast	
	Chorion	chorion cells	
	HSMMtube	skeletal muscle myotubes differentiated from the HSMM cell line	
rs1327063	MCF-7	mammary gland, adenocarcinoma	
	pHTE	primary tracheal epithelial cells	
	Chorion	chorion cells	
	HSMMtube	skeletal muscle myotubes differentiated from the HSMM cell line	
rs1536459	MCF-7	mammary gland, adenocarcinoma	
	pHTE	primary tracheal epithelial cells	
	Chorion	chorion cells	
	HSMMtube	skeletal muscle myotubes differentiated from the HSMM cell line	
rs10738623	Hepatocytes	primary hepatocytes	
	pHTE	primary tracheal epithelial cells	
	Melano	epidermal melanocytes	
rs10757311	Osteobl	osteoblasts (NHOst)	
rs1327061			EP300 (HeLa-S3)
			FOSL2 (A549)
			FOS (HUVEC)
			FOS (MCF10A-Er-Src)
			NR3C1 (A549)
			FOXA1 (A549)

Table E5: Explained AD heritability in Europeans including all previously reported loci and the 2 novel risk variants. Calculation was performed as previously described in ¹⁰ and ⁹. RA freq: risk allele frequency.

locus	variant	chromosome	RA freq	explained heritability
<i>FLG</i>	R501X, 222del4, R2447, S3247X	1q21.3	0.04	0.071
<i>IL18R1/IL18RAP/SLC9A4</i>	rs759382	2q12.1	0.24	0.006
<i>IL2/IL21</i>	rs17389644	4q27	0.23	0.004
<i>RAD50/IL13/IL4/KIF3A</i>	rs848	5q31.1	0.20	0.016
<i>HLA-C/HLA-B/MICA</i>	rs9368677, rs2251396	6p21.33	0.28	0.010
<i>BAT1</i>	rs2844509	6p21.33	0.74	0.010
<i>C6orf48</i>	rs9368699	6p21.33	0.04	0.010
<i>TNXB/CREBL1</i>	rs12153855	6p21.32	0.10	0.018
<i>PRR5L</i>	rs12295535	11p13	0.02	0.006
<i>OVOL1</i>	rs479844	11q13.1	0.56	0.003
<i>C11orf30/LRRC32</i>	rs7110818	11q13.5	0.44	0.012
<i>CLEC16A/DEXI</i>	rs2041733	16p13.13	0.45	0.009
<i>ZNF652</i>	rs16948048	17q21.32	0.38	0.005
<i>ACTL9</i>	rs2164983	19p13.2	0.15	0.002
<i>TNFRSF6B</i>	rs909341	20q13.33	0.77	0.010
<i>XIRP2</i>	rs6720763	2q24.3	0.18	0.009
<i>DMRTA1</i>	rs10738626	9p21.3	0.51	0.014
total of explained heritability				0.215

Table E6: EMSA oligonucleotide sequences. Cy5-labeled and unlabeled oligonucleotides containing the major (Ma) or minor (Mi) allele of rs6720763 and rs10738626 or the consensus sequence for SP1 (specificity protein 1). Forward (F) and reverse (R) oligonucleotides are listed. SNP alleles are highlighted in red.

oligonucleotide	5' -> 3' sequence
F_rs6720763_Ma_T	TATCAACATTTTACCTGAAGCATATTGAAAA
R_rs6720763_Ma_A	TTTCAATATGCTTCAGGTAAAATGTTGATA
F_rs6720763_Mi_C	TATCAACATTTTACCCGAAGCATATTGAAAA
R_rs6720763_Mi_G	TTTCAATATGCTTCGGTAAAATGTTGATA
F_rs10738626_Ma_C	CCATGTATATTTCTCGCCATAGGGAAACAT
R_rs10738626_Ma_G	ATGTTTCCCTATGGCGAGGAAATATACATGG
F_rs10738626_Ma_T	CCATGTATATTTCTTGCCATAGGGAAACAT
R_rs10738626_Ma_A	ATGTTTCCCTATGGCAAGGAAATATACATGG
F_SP1	ATTCGATCGGGGCGGGGCGAGC
R_SP1	GCTCGCCCGCCCGATCGAAT

Supplementary Figures

Figure E1: quantile-quantile (Q-Q) plot of the dataset after quality control including 6,163 samples (870 cases, 5,293 controls) and 1,623,390 SNPs. The Q-Q plot of the association statistics displays whether more significant results were generated than expected by chance. 95% confidence intervals are shown for the expected associations. The estimated genomic inflation factor is $\lambda_{1000}=1.076$, indicating minimal undetected population stratification.

Figure E2: Regional association plots of 2q24.3 (*XIRP2*), 9p21.3 (*DMRTA1*), 1q21.3 (*EDC*), 5q31.1 (*RAD50/IL13/KIF3A*) and 15q22.1 (*RNF111*). Presented are $-\log_{10} P$ -values as a function of chromosomal position (NCBI's build 36 (hg18)). Blue dot: lead SNP; differently colored dots/diamond: analyzed genotyped/imputed SNPs, the fill color represents the strength of LD (r^2) with the lead SNP (see color legend). Red dotted lines: $r^2 > 0.8$; blue line: recombination rates (cM/Mb).

Figure E3: *XIRP2* and *DMRTA1* RNA expression using a human cDNA tissue panel and keratinocytes, as determined by endpoint-PCR. Water served as a negative control, *GAPDH* as a positive control for keratinocytes and β -actin as a positive control for the cDNA tissue panel.

Figure E4: *XIRP2* immunohistochemistry analysis in healthy controls and AD patients. Immunostaining exhibited low expression of *XIRP2* in the epidermis of a) healthy control persons and b) lesions of AD patients (n=8, bars represent 50 μ m).

Figure E5: DMRTA1 immunohistochemistry analysis in healthy controls and AD patients.

DMRTA1 was detected in the epidermis of a) healthy control persons as well as in b) lesions of AD patients (n=8, bars represent 50 μ m).

Figure E6: Allele specific protein-DNA interactions of rs10738626 (*DMRTA1*). Two additional complex formations (arrows c1+c2) could be shown for the risk-associated major allele T (lane 3), and three additional complex formations (arrows c3-c5) for the protective minor allele C (lane 10). Competition testing revealed specificity for c2 and c5 (lanes 4-9, 11-16, respectively), and a more efficient competition for c1, c3 and c4 (lanes 4-9 for c1, 11-16 for c3+c4).

References

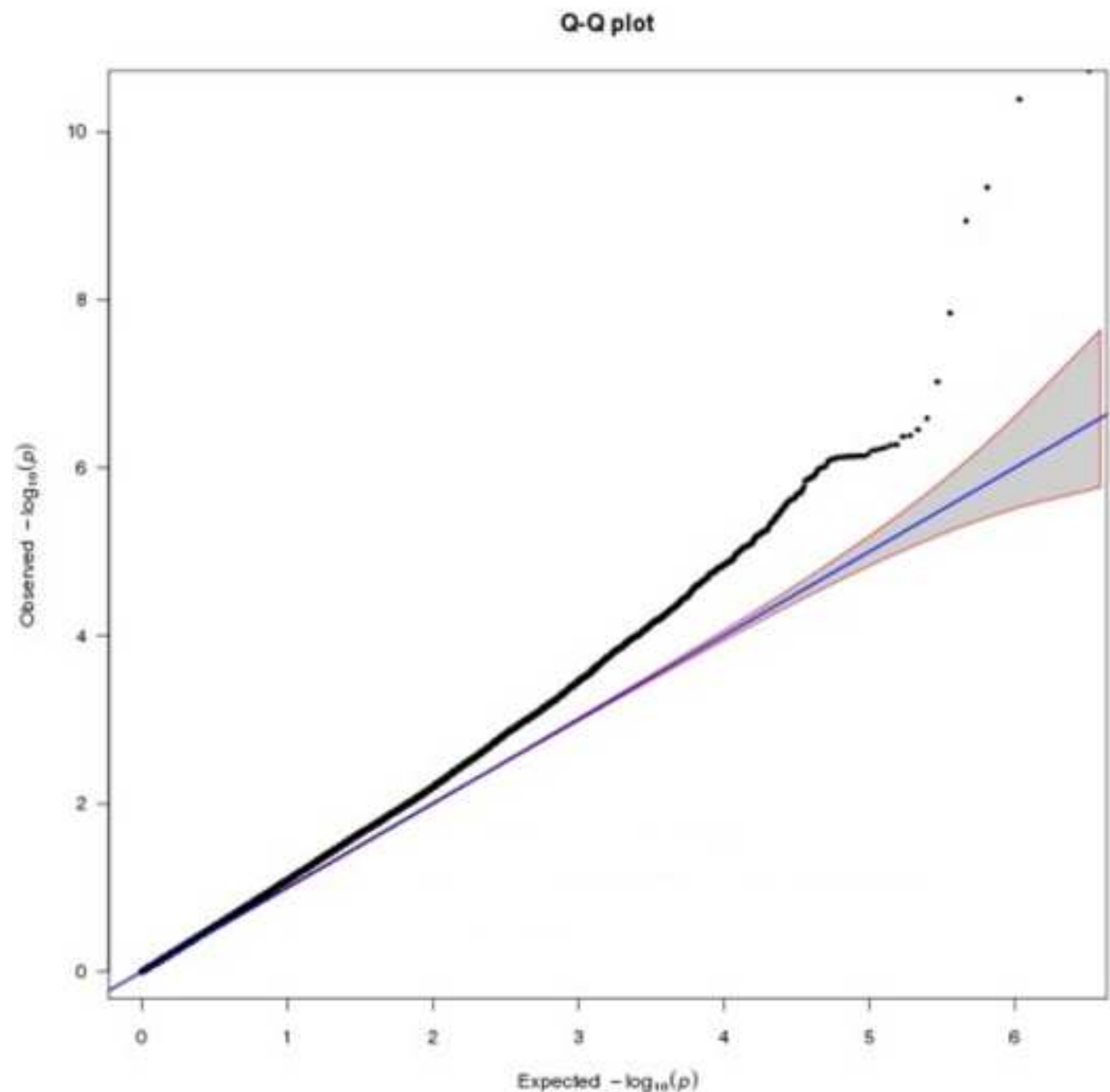
1. Williams HC, Burney PG, Hay RJ, Archer CB, Shipley MJ, Hunter JJ, et al. The U.K. Working Party's Diagnostic Criteria for Atopic Dermatitis. I. Derivation of a minimum set of discriminators for atopic dermatitis. *The British journal of dermatology* 1994; 131:383-96.
2. Hanifin JM, Rajka G. Diagnostic features of atopic eczema. *Acta Derm Venereol* 1980;44-7.
3. Krawczak M, Nikolaus S, von Eberstein H, Croucher PJ, El Mokhtari NE, Schreiber S. PopGen: population-based recruitment of patients and controls for the analysis of complex genotype-phenotype relationships. *Community Genet* 2006; 9:55-61.
4. Wichmann HE, Gieger C, Illig T. KORA-gen--resource for population genetics, controls and a broad spectrum of disease phenotypes. *Gesundheitswesen* 2005; 67 Suppl 1:S26-30.
5. Weiland SK, Bjorksten B, Brunekreef B, Cookson WO, von Mutius E, Strachan DP. Phase II of the International Study of Asthma and Allergies in Childhood (ISAAC II): rationale and methods. *Eur Respir J* 2004; 24:406-12.

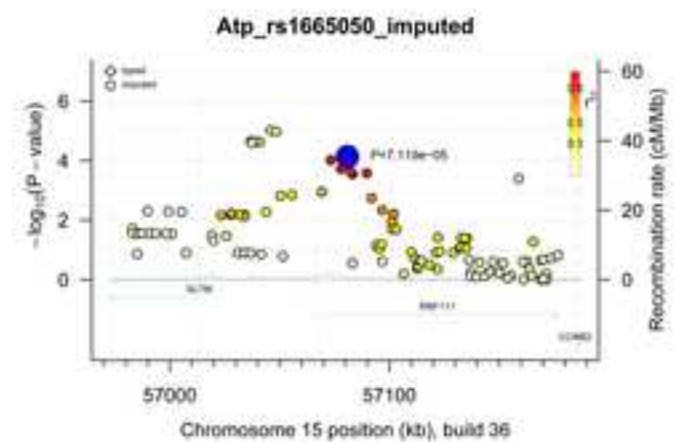
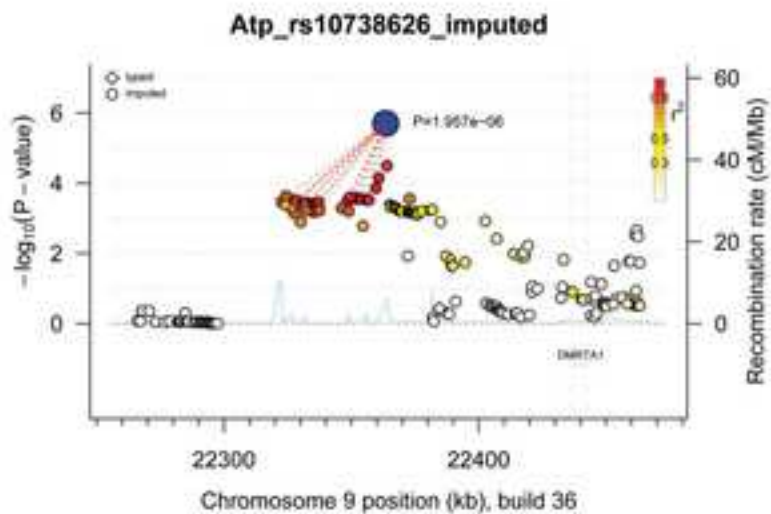
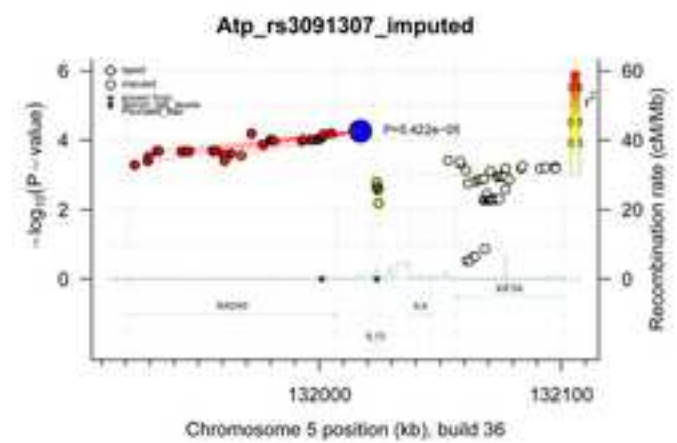
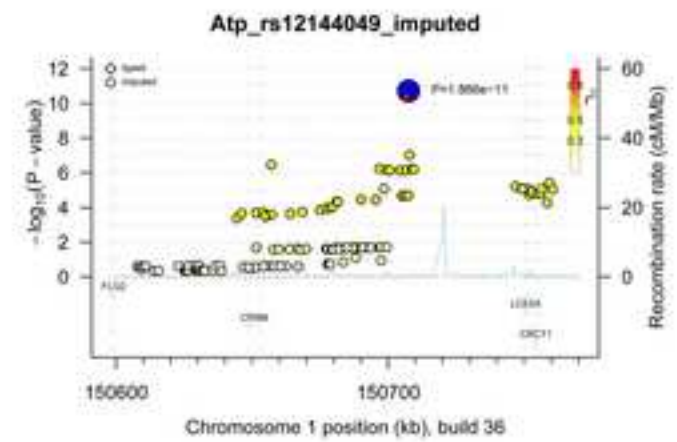
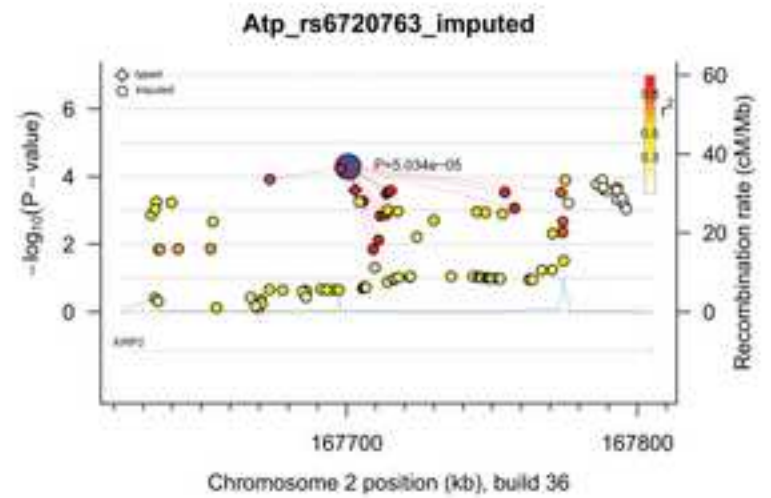
6. Li Y, Willer CJ, Ding J, Scheet P, Abecasis GR. MaCH: using sequence and genotype data to estimate haplotypes and unobserved genotypes. *Genet Epidemiol* 2010; 34:816-34.
7. Esparza-Gordillo J, Weidinger S, Folster-Holst R, Bauerfeind A, Ruschendorf F, Patone G, et al. A common variant on chromosome 11q13 is associated with atopic dermatitis. *Nat Genet* 2009; 41:596-601.
8. Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MA, Bender D, et al. PLINK: a tool set for whole-genome association and population-based linkage analyses. *Am J Hum Genet* 2007; 81:559-75.
9. So HC, Gui AH, Cherny SS, Sham PC. Evaluating the heritability explained by known susceptibility variants: a survey of ten complex diseases. *Genet Epidemiol* 2011; 35:310-7.
10. Ellinghaus D, Baurecht H, Esparza-Gordillo J, Rodriguez E, Matanovic A, Marenholz I, et al. High-density genotyping study identifies four new susceptibility loci for atopic dermatitis. *Nat Genet* 2013; 45:808-12.

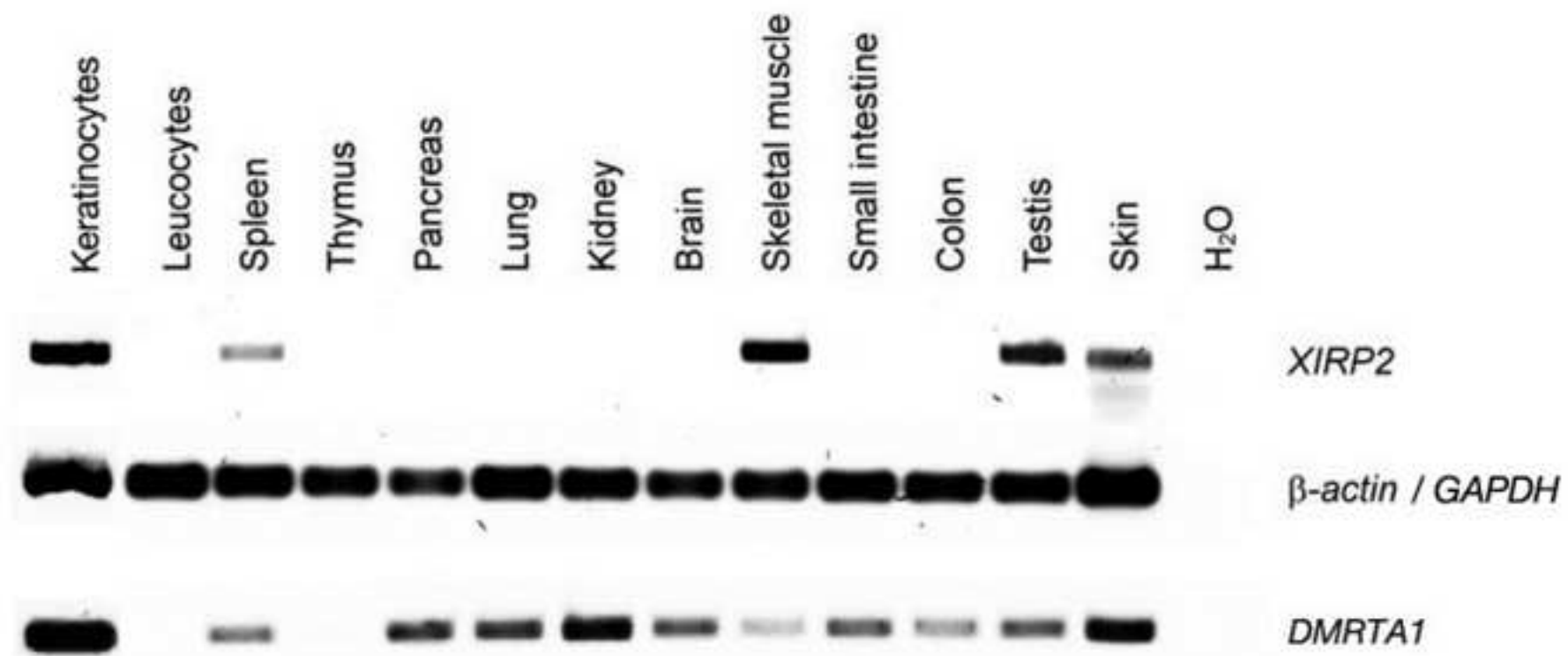
11. Weidinger S, Willis-Owen SA, Kamatani Y, Baurecht H, Morar N, Liang L, et al. A genome-wide association study of atopic dermatitis identifies loci with overlapping effects on asthma and psoriasis. *Hum Mol Genet* 2013; 22:4841-56.
12. Franceschini A, Szklarczyk D, Frankild S, Kuhn M, Simonovic M, Roth A, et al. STRING v9.1: protein-protein interaction networks, with increased coverage and integration. *Nucleic Acids Res* 2013; 41:D808-15.
13. Consortium EP. An integrated encyclopedia of DNA elements in the human genome. *Nature* 2012; 489:57-74.
14. Cartharius K, Frech K, Grote K, Klocke B, Haltmeier M, Klingenhoff A, et al. MatInspector and beyond: promoter analysis based on transcription factor binding sites. *Bioinformatics* 2005; 21:2933-42.
15. Loots GG, Ovcharenko I. Dcode.org anthology of comparative genomic tools. *Nucleic Acids Res* 2005; 33:W56-64.
16. Yang TP, Beazley C, Montgomery SB, Dimas AS, Gutierrez-Arcelus M, Stranger BE, et al. Genevar: a database and Java application for the analysis and visualization of SNP-gene associations in eQTL studies. *Bioinformatics* 2010; 26:2474-6.

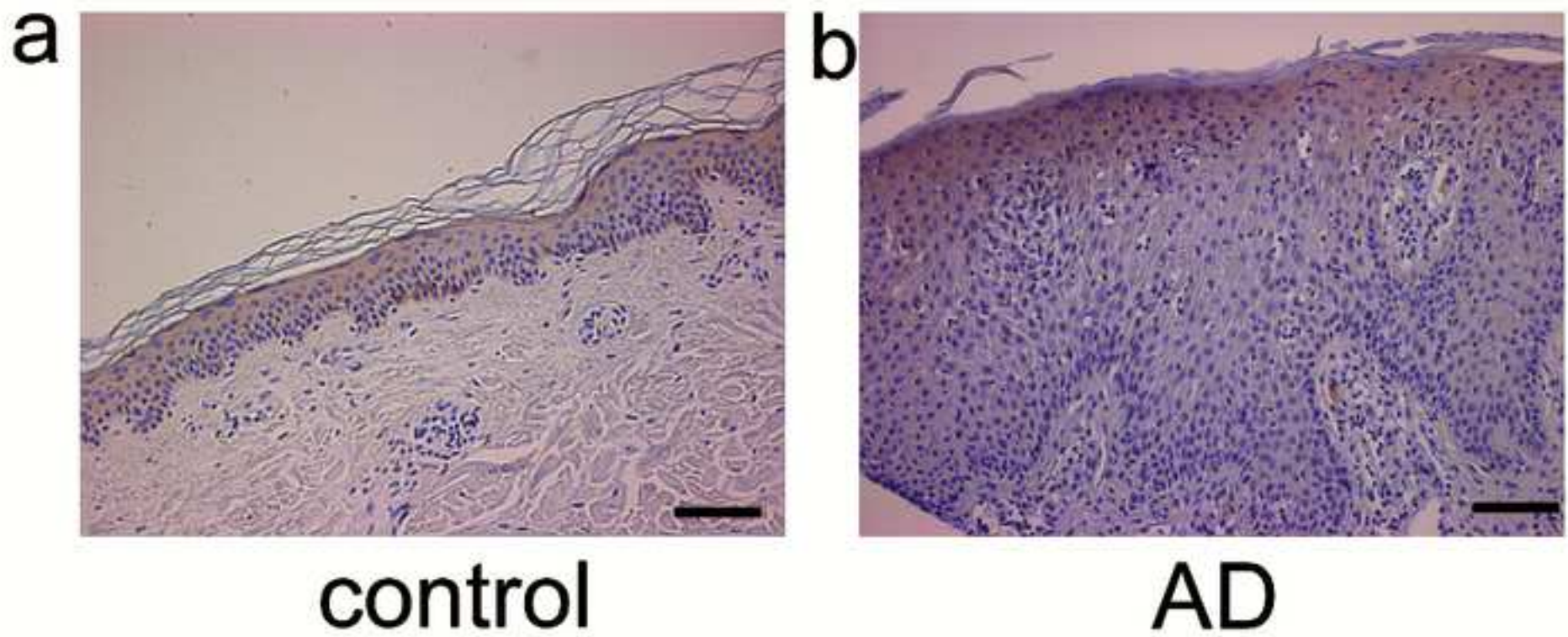
17. Nica AC, Parts L, Glass D, Nisbet J, Barrett A, Sekowska M, et al. The architecture of gene regulatory variation across multiple human tissues: the MuTHER study. *PLoS Genet* 2011; 7:e1002003.
18. Roth SA, Simanski M, Rademacher F, Schroder L, Harder J. The pattern recognition receptor NOD2 mediates *Staphylococcus aureus*-induced IL-17C expression in keratinocytes. *The Journal of investigative dermatology* 2014; 134:374-80.
19. Miyazono K, Kamiya Y, Morikawa M. Bone morphogenetic protein receptors and signal transduction. *J Biochem* 2010; 147:35-51.
20. Li MO, Wan YY, Sanjabi S, Robertson AK, Flavell RA. Transforming growth factor-beta regulation of immune responses. *Annu Rev Immunol* 2006; 24:99-146.
21. Shull MM, Ormsby I, Kier AB, Pawlowski S, Diebold RJ, Yin M, et al. Targeted disruption of the mouse transforming growth factor-beta 1 gene results in multifocal inflammatory disease. *Nature* 1992; 359:693-9.
22. Leveen P, Larsson J, Ehinger M, Cilio CM, Sundler M, Sjostrand LJ, et al. Induced disruption of the transforming growth factor beta type II receptor gene in mice causes a lethal inflammatory disorder that is transplantable. *Blood* 2002; 100:560-8.

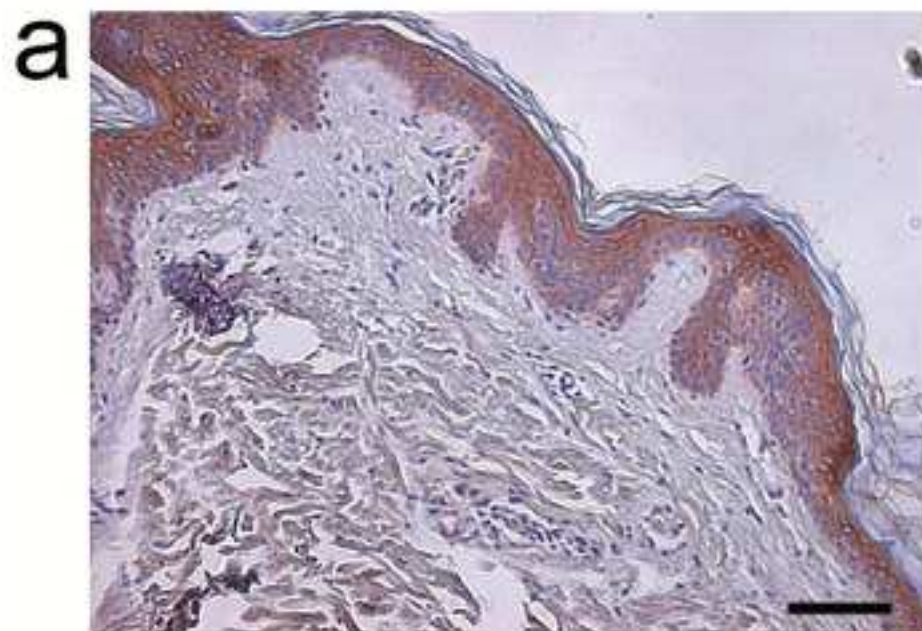
23. Frischmeyer-Guerrero PA, Guerrero AL, Oswald G, Chichester K, Myers L, Halushka MK, et al. TGFbeta receptor mutations impose a strong predisposition for human allergic disease. *Sci Transl Med* 2013; 5:195ra94.
24. Adam-Klages S, Schwandner R, Adam D, Kreder D, Bernardo K, Kronke M. Distinct adapter proteins mediate acid versus neutral sphingomyelinase activation through the p55 receptor for tumor necrosis factor. *J Leukoc Biol* 1998; 63:678-82.
25. Jensen JM, Folster-Holst R, Baranowsky A, Schunck M, Winoto-Morbach S, Neumann C, et al. Impaired sphingomyelinase activity and epidermal differentiation in atopic dermatitis. *J Invest Dermatol* 2004; 122:1423-31.
26. Hatano Y, Terashi H, Arakawa S, Katagiri K. Interleukin-4 suppresses the enhancement of ceramide synthesis and cutaneous permeability barrier functions induced by tumor necrosis factor-alpha and interferon-gamma in human epidermis. *J Invest Dermatol* 2005; 124:786-92.
27. Imokawa G, Abe A, Jin K, Higaki Y, Kawashima M, Hidano A. Decreased level of ceramides in stratum corneum of atopic dermatitis: an etiologic factor in atopic dry skin? *J Invest Dermatol* 1991; 96:523-6.



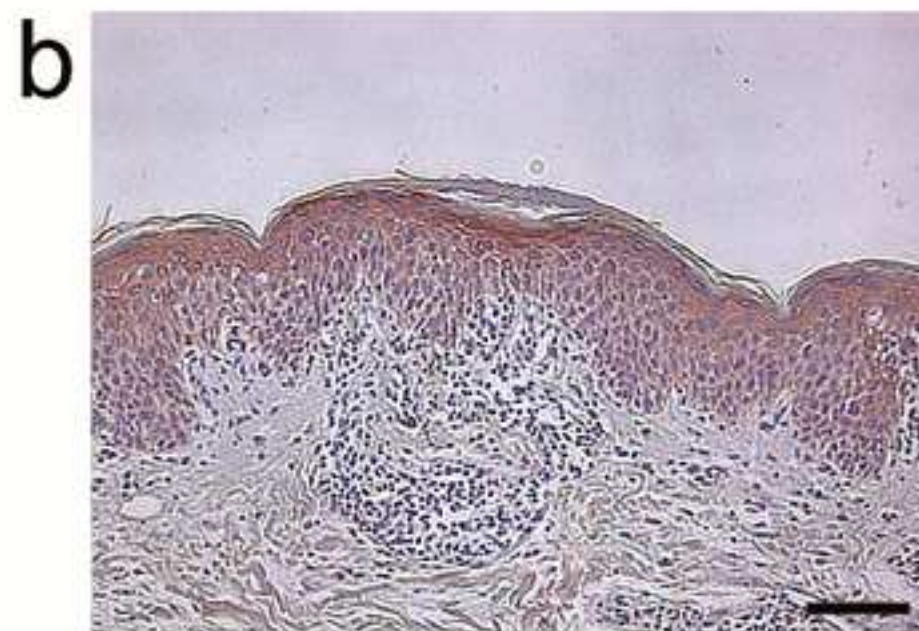








control



AD

Repository E Figure 6

[Click here to download high resolution image](#)

