Large genetic panel analysis of British and French childhood-onset lupus cohorts

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Objectives

Systemic lupus erythematosus (SLE) is a rare immunological disorder where genetic factors are considered important in causation. Monogenic lupus has been associated with around 30 genotypes in humans, and 60 in mice; whilst genome-wide association studies have identified more than 90 risk loci. We aimed to determine the genetic contribution of rare and predicted pathogenic gene variants in a population of unselected cases of childhood-onset SLE (cSLE).

Methods

We designed a next generation sequencing (NGS) panel comprising 147 genes, including all known lupus causing (KLC) genes in humans, and potentially lupus causing (PLC) genes identified through GWAS and animal models. We screened 117 probands fulfilling ACR criteria for SLE, ascertained through British and French cohorts of cSLE, and compared these data with those of 791 ethnically matched controls from the 1000 Genomes Project, 574 controls from the FREX consortium.

Results

Mendelian genotypes were confirmed in eight probands, involving mutations in *C1QA, C1QC C2, DNASE1L3* and *IKZF1*. Seven additional patients carried heterozygous mutations in complement or type I interferon associated autosomal recessive genes, with decreased levels of the encoded proteins recorded in two patients. Rare, predicted damaging variants were significantly enriched in the cSLE cohort compared to controls; 25% of SLE probands versus 4.6% of controls were identified to harbour at least one rare, predicted damaging variant ($p = 4.14 \times 10^{-15}$).

Conclusions

Inborn errors of immunity account for 7% of cSLE, with defects of innate immunity representing the main monogenic contribution. An accumulation of rare, predicted damaging variants in SLE-associated genes may contribute to disease expression and clinical heterogeneity.

Keywords: Monogenic lupus; paediatric rheumatology; genetics; innate immunity; complement deficiency

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Introduction

The term systemic lupus erythematosus (SLE) describes a rare, heterogeneous set of phenotypes characterized by the presence of autoantibodies targeting nuclear autoantigens and type I interferon upregulation. Familial aggregation and higher concordance rates between monozygotic compared to dizygotic twins suggest a major hereditary component to pathogenesis[1]. Genome-wide association studies (GWAS) have identified more than 90 SLE-associated loci[2]. GWAS defined variants are common, confer small effects on disease susceptibility and typically fall outside of coding regions. In contrast, monogenic forms of SLE, of which almost 30 have been described in humans, and greater than 60 in mice[3], involve highly penetrant rare variants in protein encoding DNA[4].

Mendelian forms of disease can help define the involvement of discrete pathways in pathogenesis. For example, both classical complement gene mutations and DNASE1L3 deficiency highlight the importance of efferocytosis in lupus pathology[5,6], whilst the relevance of type I interferon signalling to SLE is underlined by the association with the Mendelian type I interferonopathies[7,8]. Finally, B cells are a key player in lupus causation[9], with PKCδ deficiency the first described B cell related form of monogenic lupus[10], and heterozygous germline mutations in *IKZF1*, encoding IKAROS, a B cell transcription factor, a cause of autoimmunity including SLE[11].

Recent sequencing studies suggest a false dichotomy in categorizing diseases according to strict complex or Mendelian models, an alternative possibility highlighting the importance of combinations of small numbers of rare variants promoting disease in a single individual and at a population level[12]. Immune responses are variable in humans, with up to 40% of this diversity estimated to be explained by genetic variation[13]. In association with environmental factors, such genetic polymorphisms may promote tolerance breakdown[14], as exemplified by the lupus phenotype.

Here, we describe a large-scale molecular analysis of childhood-onset SLE (cSLE), indicating a mutational spectrum directly causing, or strongly influencing, pathogenesis.

Material and methods

Patients

We identified 117 unrelated probands from the UK Juvenile-onset SLE (JSLE)[15] and French SLE GENIAL cohorts fulfilling American College of Rheumatology (ACR) criteria for lupus, diagnosed before the age of 16 years. The study was approved by the relevant ethics committees (see the Appendix for further details). The combined cohort comprised the following ethnicities: European (50%), African (33%) and South Asian (16%), classified according to International Genome Sample resource criteria (figure 1A).

Gene selection

To identify genetic variations potentially responsible for SLE, we selected a set of genes based on: i) known Mendelian forms of lupus ('known lupus causing' – KLC - genes; n = 28), some also detected by GWAS and/or in murine studies (table S1); ii) defined causes of monogenic lupus in mouse models (n = 62) and/or genes demonstrating an association with SLE identified through GWAS (n = 67), with 11 genes common to both. The latter group are referred to as 'putative lupus causing' (PLC) genes (table S2). Defined as of 2013 when the study was initiated, this set comprised 147 genes in total.

DNA sequencing, variant discovery and filtering

Details of our sequencing protocols and variant analyses are provided in the Appendix. In brief, targeted enrichment and sequencing of the coding regions of 147 gene panel was undertaken using DNA extracted from peripheral blood. Single nucleotide variants (SNV) and indels were identified according to strict bioinformatic protocols, aligned with 1000 Genomes Project (1000g) sequence sets, and variant pathogenicity assessed using SIFT, Polyphen-2 and Combined Annotation Dependent Depletion (CADD) v1.3 scores. A rarity filter considered SNVs according to their population frequencies within the Exome Aggregation Consortium (ExAC) dataset, employing 1% and 0.1% in the case of putative biallelic mutations, or 0.01% and 0.001% for heterozygous variants in the case of indels or SNVs respectively, as our thresholds (threshold (heterozygous) = threshold (homozygous)^2 i.e. 10^{-4} for indels and 10^{-6} for SNVs). A pathogenicity filter considered selected indels with a CADD score higher than 15, and SNVs according to the formula: CADD score ≥ 15 AND sift score ≤ 0.05 AND Polyphen 2 score ≥ 0.9. A cartoon of the characterization of different sequence variants is given in figure S1 in the Appendix. For the FREX consortium and UK controls, only SNVs were analysed.

Control cohorts

Control data were derived from the 1000 Genomes Project (1000g). The healthy control cohort comprised 791 unrelated individuals demonstrating a similar ethnic distribution to the cSLE cohort (50% European, 33% African; 16% South Asian), built by random sampling among the 1000g sample set. Two additional control populations were ascertained through the FREX consortium, a French-specific database containing the exome data of a reference control panel of 574 individuals of French ancestry (http://lysine.univ-brest.fr/FrExAC/) and a UK control panel: 74 adults from the north west of England, sequenced and analysed using exactly the same targeted exon panel, sequencing platform and filtering strategy as employed in the lupus cohort. Additional information is given in Appendix.

***In vitro* assays, structural analyses**

Details of *in vitro* experiments, structural studies and protein interaction network construction are given in the Appendix.

Statistical analysis

Fisher's exact test was used to compare the frequencies of variants in the control and cSLE cohorts. Data normality was assessed using a Shapiro-Wilk test and quantile-quantile plots. The mean of SNPs per individual was compared between control and cSLE cohorts using a Mann-Whitney U test (Wilcoxon rank-sum test). P-values were adjusted using the Bonferroni correction where appropriate. Odd ratios were calculated using the standard formula, with S1/2 the number of variants that passed a filter and F1/2 those which failed for the two groups compared (OR = S1*F2 / F1*S2).

Role of the funding source

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Results

Population characteristics

Our patient cohort demonstrated a median age at disease onset of 9 (range 1.8 - 16) years and sex ratio of 25/92 (male:female = 1:3.7) (figure 1B, table S3). Similar to previous studies of cSLE, major organ involvement was frequent, with renal and cerebral disease noted in 59% and 23% of individuals respectively. A family history of lupus was reported in 20% of cases (first or second degree relatives).

Next generation sequencing identified a total of 30,955 variants in the 147 KLC and PLC genes across 117 patients (figures 1C, 1D) in the protein coding part of the gene (Supplemental Figure S1). Filtering according to the strategy described above was used to select for very rare variants, allowing us to exclude 99.95% of the initially identified polymorphisms. A similar strategy was applied to the control cohort of 791 ethnically matched individuals from the 1000g (SNVs and indels) and from the FREX consortium (SNVs) (figure 1E). An additional confirmatory cohort was generated from UK control panel: 74 adults from the north west of England sequenced and analysed using exactly the same targeted exon panel, sequencing platform and filtering strategy as employed in the lupus cohort.

Characterization of Mendelian genotypes

After filtering, 15 unrelated probands were identified to harbour a total of 14 either heterozygous or biallelic variants in 11 KLC genes (table 1), relating to components of the classical complement pathway (*C1QA, C1QC, C2, C3, C8B, C9*), DNASE1L3 deficiency (*DNASE1L3*), type I interferonopathies (*TREX1, RNASEH2C, ACP5)* or B-cell dysfunction (*IKZF1*). Ten of these 14 variants were novel (i.e. not seen in publicly available databases), with five having been reported previously as disease causing.

Segregation was compatible with autosomal recessive inheritance due to complement deficiency in six pedigrees. Biallelic mutations in either *C1QC* or *C1QA* were identified in three families (figure 2A). *In silico* structural analysis of C1qC predicted that the p.Gly164Ser substitution, already reported as pathogenic[16], destabilizes the globular head of the C1q complement protein (figure 2B). Two other variants in *C1QC* and *C1QA* were nonsense mutations. C1q was undetectable in patient 1 and 2 (Pt1, Pt2) (figure 2C). Patients 4 to 6 carried the same biallelic deletion in the *C2* gene (figure 2D), previously described as a cause of lupus with incomplete penetrance[17]. CH50 and C2 levels were decreased in Pt6, whist C3 and C4 were normal (figure 2E).

Two predicted damaging variants in *DNASE1L3* were identified in Pt7, and biallelic inheritance confirmed by parental testing (figure 3A). The nonsense mutation has been reported as disease causing[18], whilst the deletion of exon 5 was novel, located in the deoxyribonuclease domain (figure 3B) and confirmed by TAQman qPCR.

Sequencing of the family of Pt8 demonstrated segregation of a novel missense *IKZF1* variant with disease status in two additional relatives (figure 3C). A previous GWAS identified significant association with lupus at the *IKZF1* locus[19], whilst loss-of-function germline mutations have been recently reported to cause systemic autoimmunity and B cell deficiency[11]. Thus, *IKZF1*, initially considered as a PLC gene, was subsequently classified as a KLC gene. The p.Asp120Val IKZF1 variant is located in the first zinc finger of the protein (figure 3D). Confocal microscopy of NIH-3T3 cells transfected with wild type (WT) and mutant constructs revealed a punctate staining pattern characteristic of pericentromeric heterochromatin binding and localization of the WT protein, whereas the mutant protein exhibited diffuse nuclear staining, as previously observed for other pathogenic missense variants[20]. We also performed an electrophoretic mobility-shift assay (EMSA) using nuclear extract of transfected cells, and observed that the Asp120Val protein was unable to bind a consensus-binding sequence (similar to a previously reported mutation Arg164Gln) (figure 3D). Finally, we transduced Granta cells with inducible lenti-vector pInducer21 (ORF-EG)

(Addgene) expressing either WT-IKZF1 or Asp120Val-IKZF1, and recorded a decrease of transcriptional activity of the mutant construct by RNAseq analysis (figure 3E, F). Notably, all three affected patients carrying the p.Asp120Val mutation had normal immunoglobulin levels and did not present features of immunodeficiency.

Heterozygous variants in autosomal recessive KLC genes (Mendeliome) and PLC genes

Seven probands were identified to carry a monoallelic, rare, predicted damaging variant in a KLC gene previously described to cause SLE as an autosomal recessive trait (table 2). A *TREX1* p.Ser82Leufs*9 frameshift mutation had already been reported as disease-causing in Aicardi-Goutières syndrome (AGS)[21] . Other predicted damaging variants were recorded in *RNASEH2C*, *ACP5, C3*, *C8B* and *C9*. In this group of seven patients the sex ratio was 4:3, similar to the subset of probands described immediately above, and differing from the overall composition of our cohort (figure S2). Furthermore, the mean age at onset was earlier in this group than in the cohort overall. Pt15, carrying a p.Ile1093Ser *C3* substitution, presented with a phenotype characterized by early onset glomerulonephritis (lupus nephritis with C1q deposits) and thrombotic microangiopathy. He benefited from treatment with eculizumab and is still in remission after three years of therapy. C3 was undetectable on several assessments, representing the lowest levels recorded in the whole cSLE cohort (figure 3G). Heterozygous variants in *C3* have been reported as possibly causal of SLE[22] and evolution with eculizumab strongly support a causal impact of the mutation. A C9 variant was similarly associated with decreased protein levels in patient plasma (Pt14) (figure 3H). The *C8B* variant seen in Pt13 did not impact on plasmatic C8 concentration. The other patients were not available for additional investigations.

Focusing on variants in PLC genes, we detected 17 rare, predicted damaging PLC variants in 16 additional unrelated probands (Table 2).

To test in an unbiased manner if the accumulation of rare, predicted damaging heterozygous KLC and PLC variants was specifically associated with cSLE status, we constituted first a control cohort by analysing the genomic sequence data of 791 healthy individuals from the 1000g database, ensuring a similar distribution of ethnicity to our patient group, as shown by the PCA analysis of the frequent (MAF>1%) SNPs (figure S3). We applied the same algorithm to the controls as used in our cSLE data set. In so doing, we observed a statistically significant excess of rare, predicted damaging variants within the cSLE population compared to 1000g controls after adding filtering for rarity plus pathogenicity (figure 4A), with an odds ratio of 17.2 (CI 95% = 3.36-90,34) in KLC genes (figure 4C). For PLC genes, a significant difference was also identified with the same filtering strategy (rarity plus pathogenicity) (figure 4B), with an enrichment of 4.4 (CI95% = [2.02;8.18]) in the lupus population (figure 4C, D). These data comprised all variants in our gene panel in the SLE and 1000g cohort datasets, generated respectively by targeted sequencing and whole genome sequencing. Of note, sequence coverage was different between our in-house derived sequence and those of the 1000g (table S4). To further consider the comparability of our patient and control groups, we undertook a confirmatory analysis using data from a second control population (FREX Consortium)(figure 4A, 4B), demonstrating a higher coverage across our gene panel (table S4). Again, the overall mutational load was significantly increased for KLC and PLC genes in the cSLE population, with a greater odds ratio when considering rarity plus pathogenicity status, compared to rarity only, with an OR that reached 11.1 (CI95% = [2.151;57.202]) and 2.83 (CI95%=[1.398;5.735]) for KLC and PLC genes respectively under this latter filtering strategy (figure 4C, 4D). Coverage information of the variants selected from the panel are represented in supplemental table S5. A similar trend was also observed when we considered 74 controls from a United Kingdom cohort of healthy adults performed using the same panel sequencing sharing identical coverage and analysed with the same gene panel and filtering strategies, but the analysis did

not reach significance (table S6). In addition to the global evaluation of rare genetic variants, we tested if rare variants within the genes identified in the panel were differentially distributed in the lupus patients compared to controls. To do so, we performed rare variant association tests using the sequence Kernel association (SKAT-O) method, which combines burden and variance component tests and has been used to explore rare genetic variants association testing. We observed a higher burden of rare variants in the lupus population in three genes: *C1QC*, *RNASEH2C*, *MECP2* and *CD19* (table S7). Finally, we compared the combined frequency of KLC and PLC rare and predicted damaging variants per proband in the panel and 1000g, and identified a statistically significant increase of filtered variants in one, two and three genes per individual in the SLE population (figure 4E,F). Taken together, these mutational load and gene specific analyses suggest an enhanced contribution of rare and predicted damaging monoallelic variants in KLC or PLC genes to cSLE susceptibility.

Discussion

Immunoprofiling of cSLE patients has highlighted the heterogeneous nature of this phenotype[23]. Whilst the basis of such heterogeneity remains poorly defined, the identification of Mendelian forms of SLE related to different pathogenic mechanisms indicates that genetic factors are likely important. Low frequency variants are not captured by GWAS, nor do they confer sufficiently large effect sizes to be detected by classical linkage analysis in small family studies[4]. Meanwhile, the observation of variable penetrance and expression in Mendelian disorders indicates that other genes or environmental factors can impact phenotype. Here, we add to the understanding of germline predisposition to early-onset SLE.

A major challenge of our study was to set appropriate thresholds, in terms of population frequency and pathogenicity prediction, in order to capture likely causal variants. Given the identification of unequivocal Mendelian genotypes, the algorithms we employed appear valid. This bioinformatics strategy also demonstrated that the distribution of filtered variants was not equal in ethnically matched patient and control populations. Such stringent filtering, leading to an enrichment for likely pathogenic alleles, risks excluding possible disease-causing variants. For example, a p.Asp105Ala heterozygous variant in RNASEH2B, previously considered as causal for SLE[7], was excluded using this filtering strategy (data not shown). As such, our results likely represent a conservative estimate of the associated high-penetrant genetic load.

Our analysis identified eight probands with mutations in KLC genes consistent with Mendelian causation. Complement deficiencies accounted for 5% of probands in our cohort and a defect of efferocytosis is also relevant in DNASE1L3 deficiency[6]. Only one patient in our cohort was identified with a monogenic disease involving B cell dysfunction. That is, a novel mutation in *IKZF1*, encoding the transcription factor IKAROS, playing a role in B cell development. No mutations were identified in *PRKCD*, previously associated with monogenic lupus[10]. Thus, whilst SLE is often considered as a B cell driven disease, the majority of Mendelian forms observed in our cSLE relates to mutations in innate immunity related genes.

In cSLE patients compared to controls, we observed an increased frequency of monoallelic variants in KLC genes described to cause lupus as an autosomal recessive trait in both type I interferonopathy and complement related genes. In this group the sex ratio was 4:3, and the onset of disease was very early, possibly suggesting a significant impact of the observed heterozygous variants on disease induction. Among these variants, a *TREX1* mutation has already been reported in AGS, a recognized type I interferonopathy associated with an increased risk of SLE[8]. Günther et al. also described an increase of rare heterozygous variants in components of the RNase H2 complex in lupus[7]. Using our filtering strategy, one patient was detected with a heterozygous *RNASEH2C* predicted pathogenic substitution. Of note, the SKAT-O test also highlighted a higher rare-disease burden for this gene in the lupus population. We recorded an *ACP5* missense variant in one patient. Biallelic mutations

in *ACP5* are a cause of Mendelian lupus, and an increase of predicted pathogenic heterozygous variants in this gene has been reported in an SLE cohort[24]. The patient carrying the novel variant in C3 displayed features of lupus nephritis and haemolytic uremic syndrome (HUS), with C3 deficiency a known cause of HUS. We also noticed markedly decreased levels of C3 in this patient, strongly arguing for a genetic contribution of the variant to the phenotype. Overall, these observations support the possibility that genes known as directly causal of classical Mendelian lupus may play an important role in driving susceptibility to complex disease. Indeed, a recent report using whole genome sequencing identified an enrichment of heterozygous variants in genes related to monogenic lupus in an SLE cohort[25]. In addition, *de novo* monoallelic mutations have been described in KLC genes, including *PRKCD*, in cSLE patients, further supporting the contention of a genetic driven threshold for the occurrence of autoimmunity[26].

We identified 15 patients with heterozygous, putative mutations in PLC genes. The frequency of these rare, predicted damaging variants was also significantly increased in cSLE patients compared to the controls, suggesting that they might represent novel SLE predisposing alleles. Furthermore, some filtered variants co-segregated in a single individual. For example, Pt8 carried a *LYN* substitution associated with the novel *IKZF1* mutation discussed above, whilst Pt10 was identified with a novel, predicted damaging variant in each of *ACP5* and *RASGRP3*. It seems plausible that a lupus phenotype might be driven by pathogenic variants in two or more genes. Consistent with this hypothesis, we demonstrated an accumulation of rare and predicted damaging variants in individual cSLE patients compared to controls. It is important to emphasize that the number of rare variants *per se* did not vary between these populations; rather, it was the combination of variant frequency and pathogenicity score which distinguished the two groups in terms of monoallelic variant burden, with an odds ratio between patients and controls of from 11.1 to 17.1, and from 2.83 to 4.4 in the KLC and PLC gene sets respectively. Of note, a recent study in patients with C1R deficiency also suggested that common SLE-associated variants may influence the severity of an otherwise monogenic disease state[27].

Using our strategy, focusing on exons and polymorphisms with predicted functional consequences, we identified putative disease causing variants in the coding regions of genes already detected in GWAS studies. As examples, we recorded two unrelated patients carrying the same variant in *PDHX*, and a novel *PTPN22* variant was observed in one patient. Rare variants may contribute to the burden of the disease, with their accumulation influencing age at onset and clinical spectrum. Rare variants in two genes identified in GWAS studies, *BLK* and *BANK1*, were also recently identified in lupus patients with functional impact on B cell biology[28]. In our study, the bioinformatic filter we used was more stringent, and filtered out some of these variants in these genes. In addition, we may have missed functionally relevant regulatory variants located in non-coding regions of the genome that were not captured by the panel. Thus, given the stringency of the bioinformatic algorithm that we employed, our study probably underestimates the genetic contribution of the genes selected here. Furthermore, parental DNA was not systematically available in all patients, preventing us from detecting *de novo* mutations that might have otherwise been identified by trio analyses[25]. Finally, by limiting our analysis to a 147 gene panel we omit several other genes that may contribute to a Mendelian susceptibility to lupus. Functional studies were only performed when material was available, and to robustly address the contribution of interesting variants, their introduction into cellular and/or animal models may prove helpful.

Studying extreme phenotypes of complex disease represents a powerful strategy to simplify and understand human pathology. Our analysis highlights the heterogeneity of the genetic basis of cSLE, with 25% of the cSLE population carrying at least one rare and predicted damaging variant. Numerous clinical trials have failed to demonstrate a positive effect of a variety of medications in SLE, and current clinical and laboratory criteria are clearly not sufficient to detect specific factors determining treatment outcome. Explicitly considering

rare, high penetrant gene variants might identify patient subsets with distinct therapeutic responses, thereby enabling personalized treatments according to genetic background and molecular taxonomy[28].

Research in Context

Evidence before this study

Familial aggregation, and higher concordance rates between monozygotic (20 – 40%) relative to dizygotic twins and other full siblings (2 – 5%), suggest a major hereditary component to the pathogenesis of systemic lupus erythematosus (SLE). Whilst genome-wide association studies (GWAS) have identified more than 90 loci as robustly associated with the lupus phenotype, it is an overlooked fact that Mendelian forms of lupus have been described in the context of almost 30 discrete genotypes in humans, referred to here as the Mendeliome, and more than 60 single gene defects in mice. At present, the significance of these observations for the understanding of lupus biology remains unclear.

Added value of this study

To address the issue of the genetic contribution to lupus in children, we performed an analysis of 147 genes at depth, selected on the basis of their involvement in lupus pathogenesis, in 117 unrelated probands with SLE demonstrating onset before the age of 16 years. Within the Mendeliome, we observed mutant genotypes to be present in 7% of probands. Furthermore, heterozygous, predicted pathogenic variants in genes previously implicated in lupus causation were significantly enriched in the SLE cohort compared to controls. Of further interest, our study supports the concept of oligogenicity in SLE genetic susceptibility, with an accumulation of rare, predicted pathogenic variants in different genes in patients.

Implication of the available evidence

Numerous clinical trials in SLE have failed to demonstrate a positive effect of a variety of study drugs, and current clinical and laboratory criteria are clearly not sufficient to detect specific factors determining treatment outcome. Our results are consistent with an underlying heterogeneity of the lupus phenotype, with dysfunction of innate immunity playing an important role. Our data suggest that an exploration of genetic burden in single individuals may be informative in tailoring future personalized therapeutic interventions.

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medicine/research/ukjsle/jsle/) and GENIAL Study Group.

References

1. Criswell LA. The genetic contribution to systemic lupus erythematosus. Bull NYU Hosp Jt Dis 2008;66(3):176–83.
2. Bentham J, Morris DL, Graham DSC, et al. Genetic association analyses implicate aberrant regulation of innate and adaptive immunity genes in the pathogenesis of systemic lupus erythematosus. Nat Genet 2015;47(12):1457–64.
3. Kono DH, Theofilopoulos AN. Genetics of SLE in mice. Springer Semin Immunopathol 2006;28(2):83–96.
4. Manolio TA, Collins FS, Cox NJ, et al. Finding the missing heritability of complex diseases. Nature 2009;461(7265):747–53.
5. Al-Mayouf SM, Sunker A, Abdwani R, et al. Loss-of-function variant in DNASE1L3 causes a familial form of systemic lupus erythematosus. Nat Genet 2011;43(12):1186–8.
6. Sisirak V, Sally B, D'Agati V, et al. Digestion of Chromatin in Apoptotic Cell Microparticles Prevents Autoimmunity. Cell 2016;166(1):88–101.
7. Günther C, Kind B, Reijns MAM, et al. Defective removal of ribonucleotides from DNA promotes systemic autoimmunity. J Clin Invest 2015;125(1):413–24.
8. Lee-Kirsch MA, Gong M, Chowdhury D, et al. Mutations in the gene encoding the 3′-5′ DNA exonuclease TREX1 are associated with systemic lupus erythematosus. Nat Genet 2007;39(9):1065.
9. Lipsky PE. Systemic lupus erythematosus: an autoimmune disease of B cell hyperactivity. Nat Immunol 2001;2(9):764.
10. Belot A, Kasher PR, Trotter EW, et al. Protein kinase cδ deficiency causes mendelian systemic lupus erythematosus with B cell-defective apoptosis and hyperproliferation. Arthritis Rheum 2013;65(8):2161–71.
11. Hoshino A, Okada S, Yoshida K, et al. Abnormal hematopoiesis and autoimmunity in human subjects with germline IKZF1 mutations. J Allergy Clin Immunol 2017;140(1):223–31.
12. Jordan DM, Do R. Using Full Genomic Information to Predict Disease: Breaking Down the Barriers Between Complex and Mendelian Diseases. Annu Rev Genomics Hum Genet 2018;19:289–301.
13. Brodin P, Davis MM. Human immune system variation. Nat Rev Immunol 2017;17(1):21–9.
14. Watson L, Leone V, Pilkington C, et al. Disease activity, severity, and damage in the UK juvenile-onset systemic lupus erythematosus cohort. Arthritis Rheum 2012;64(7):2356–65.
15. López-Lera A, Torres-Canizales JM, Garrido S, Morales A, López-Trascasa M. Rothmund-Thomson syndrome and glomerulonephritis in a homozygous C1q-deficient patient due to a Gly164Ser C1qC mutation. J Invest Dermatol 2014;134(4):1152–4.
16. Johnson CA, Densen P, Wetsel RA, Cole FS, Goeken NE, Colten HR. Molecular heterogeneity of C2 deficiency. N Engl J Med 1992;326(13):871–4.
17. Ozçakar ZB, Foster J, Diaz-Horta O, et al. DNASE1L3 mutations in hypocomplementemic urticarial vasculitis syndrome. Arthritis Rheum 2013;65(8):2183–9.
18. Han J-W, Zheng H-F, Cui Y, et al. Genome-wide association study in a Chinese Han population identifies nine new susceptibility loci for systemic lupus erythematosus. Nat Genet 2009;41(11):1234–7.
19. Kuehn HS, Boisson B, Cunningham-Rundles C, et al. Loss of B Cells in Patients with Heterozygous Mutations in IKAROS. N Engl J Med 2016;374(11):1032–43.
20. Crow YJ, Chase DS, Lowenstein Schmidt J, et al. Characterization of human disease phenotypes associated with mutations in TREX1, RNASEH2A, RNASEH2B, RNASEH2C, SAMHD1, ADAR, and IFIH1. Am J Med Genet A 2015;167A(2):296–312.
21. Nozal P, Garrido S, Martínez-Ara J, et al. Case report: lupus nephritis with autoantibodies to complement alternative pathway proteins and C3 gene mutation. BMC Nephrol 2015;16:40.

22. Banchereau R, Hong S, Cantarel B, et al. Personalized Immunomonitoring Uncovers Molecular Networks that Stratify Lupus Patients. Cell 2016;165(6):1548–50.
23. An J, Briggs TA, Dumax-Vorzet A, et al. Tartrate-Resistant Acid Phosphatase Deficiency in the Predisposition to Systemic Lupus Erythematosus. Arthritis Rheumatol Hoboken NJ 2017;69(1):131–42.
24. Almlöf JC, Nystedt S, Leonard D, et al. Whole-genome sequencing identifies complex contributions to genetic risk by variants in genes causing monogenic systemic lupus erythematosus. Hum Genet 2019;138(2):141–50.
25. Pullabhatla V, Roberts AL, Lewis MJ, et al. De novo mutations implicate novel genes in systemic lupus erythematosus. Hum Mol Genet 2018;27(3):421–9.
26. Demirkaya E, Zhou Q, Smith CK, et al. Brief Report: Deficiency of Complement 1r Subcomponent in Early-Onset Systemic Lupus Erythematosus: The Role of Disease-Modifying Alleles in a Monogenic Disease. Arthritis Rheumatol Hoboken NJ 2017;69(9):1832–9.
27. Jiang SH, Athanasopoulos V, Ellyard JI, et al. Functional rare and low frequency variants in BLK and BANK1 contribute to human lupus. Nat Commun 2019;10(1):2201.
28. Barturen G, Beretta L, Cervera R, Van Vollenhoven R, Alarcón-Riquelme ME. Moving towards a molecular taxonomy of autoimmune rheumatic diseases. Nat Rev Rheumatol 2018;14(3):180.
29. Carbonella A, Mancano G, Gremese E, et al. An autosomal recessive DNASE1L3-related autoimmune disease with unusual clinical presentation mimicking systemic lupus erythematosus. Lupus 2017;26(7):768–72.

Contributions

AB and YJC conceived the study, planned, designed and interpreted experiments and wrote the initial draft. GIR performed PCR sequencing and participated in variant analysis with TL. EG, GM performed SKAT-O analyses. QR and RB performed the initial filtering of variants and the first analyses on the dataset. JOS, PARF, CB, TS, TL, TB, GIR analysed the panel data, 1000g and FREX data. Complement dosage was performed by MNS. Autoantibody screening was performed by NF. Structural 3D structures were provided by NT and CG. Biobanking and processing of samples were undertaken by EC and IR. SL and AH generated EMSA for Ikaros variant. Functional immunological and transcriptome studies were performed by SOO, MM, MT, SV, and KK. ES, CF, BR, RC, PR, BK, JCL, SD, PA, HR, BBM and MB provided clinical samples and critically reviewed patient data. GENIAL Investigators, UK JSLE Study Group provided additional samples. The FREX Consortium and samples from Professor G Evans, Manchester Centre for Genomic Medicine with sequencing funded by the BRC provided control data. All authors reviewed the manuscript and agreed to its submission.

Figure 1. **Demographic characteristics of the cSLE cohort and bioinformatic analysis strategy**. Ethnicity (A), age and sex (B) of the 117 probands in the cSLE cohort. (C) Schematic of the bioinformatic pipeline used to detect rare predicted damaging variants, leading to the definition of three categories of patient: 8 patients with monogenic SLE; 8 patients carrying monoallelic variants in autosomal recessive known lupus causing (KLC) genes; 16 additional patients with filtered variants in potentially lupus causing (PLC) genes. Total and individual variant numbers are presented before and after the application of filters of rarity and pathogenicity in the SLE (D) and 1000g (E) cohorts. Note that three patients in the KLC groups carried additional rare and predicted damaging variants in PLC genes, as indicated in brackets.

Figure 2. **Complement deficiencies in cSLE**. Pedigrees of families with mutations in *C1q* (A), *C2* (D),(black shading represents affected cSLE status), with identified mutations shown below each pedigree. (B) Structure of C1q globular domain with C1qA (red), C1qB (green) and C1qC (blue), calcium ion (yellow) and representation of the Gly164 residue (magenta). This amino acid is highly conserved across the C1q family. The glycine at position 164 is located near the junction between the collagen-like stem (red, green and blue circles) with the globular domain. The glycine is tightly packed into the structure, and any substitution is predicted to destabilize the assembly of the three chains. (C) ELISA quantification of complement components in sera from patients with C1Q and (E) C2 deficiency.

Figure 3: Other genetic defects in the Mendeliome. (A) Family pedigree of Pt 7. (B) Representation of the DNASE1L3 protein showing the T97Ifs*2 substitution and the deletion identified in Pt7 (red) and parents with linear representation of DNASE1L3 and the only other mutation so-far reported within the coding region (black). (C) Family pedigree of Pt 8 with (D) position of the D120V mutation (red) in the IKAROS zinc finger domain required for DNA-binding, and previously reported mutations (black). NIH3T3 cells were transfected with HA-tagged WT or D120V mutant expression vectors labelled with anti-HA antibody and an Alexa 488-conjugated (green) secondary antibody. Cells were visualized using confocal microscopy. Mutant D120V was analysed for DNA binding with the use of an electrophoretic mobilityshift assay (EMSA), and compared to the already reported defective variant R162Q (E) Heat map of regulated genes by RNA-Seq analyses in Granta cells with transfection of inducible WT-IKZF1 or D120V-IKZF1 (left panel without induction, right panel following induction). (F) Transcriptomic activity of WT and D120V variant.

Figure 4. **Rare and predicted damaging variants in cSLE and phenotypes**. (A) Heterozygous variant mean of SNPs in known lupus causing (KLC) genes previously associated with lupus causation as an autosomal recessive trait, in cSLE probands compared to controls in 1000g and FREX database. (B) Heterozygous variant mean of SNPs in putative lupus causing (PLC) genes in cSLE probands compared to controls in 1000g and FREX database. (C) Odds ratio for the mutational load in KLC (A) and PLC (B) genes of SNPs under the different filtering conditions of the lupus patients compared to 1000g or FREX cohorts (E) Number of KLC and PLC genes per individual in which a rare, predicted damaging variant was identified in cSLE probands and controls. (F) Representation of genes with either filtered variants (inner white circle) or without filtered variants (outer grey circle). Chromosome localization and number of filtered variants (innermost coloured dots) are indicated. Variants were divided into three categories: causal for monogenic disease (red), heterozygous in KLC genes previously associated with lupus causation as an autosomal recessive trait (orange), or PLC (blue) genes. Putative di- or tri-genic associations of variants in a single proband are signified by connecting lines.

Supplementary figure S1. Coding region filter. The boxes outline what we refer to as the coding region i.e. exons and canonical splice site nucleotides.

Supplementary figure S2. Population comparability
Principal component analysis of the distribution of frequent variants in the cSLE panel and 1000g populations showing three subgroups of populations within the two populations.

Supplementary figure S3. **Phenotype according to genotype**. Spider plots of the frequency of clinical features seen in: A) cSLE patients with Mendelian disease (n = 8); (B) probands carrying heterozygous variants in autosomal recessive KLC genes (n = 8); (C) probands with variants in PLC genes (n = 16); and (D) probands with no identified filtered variants (n = 85). Sex ratios and age at diagnosis are provided for each plot. Sex ratio, male:female; age, median age at onset in years; SD: standard deviation.

Table 1: Patients carrying filtered variant in known lupus causing (KLC) genes (bold) (with additional filtered variants, underlined), defining Mendelian SLE (Pt 1 to 8) or carrying heterozygous rare, predicted damaging variants in genes known to cause autosomal recessive lupus (Pt9 to 15)

Patient	Gene	Variant (HGVS)	Protein change	Zygoty	SIFT	Polyphen 2	CADD	ACMG classification	ExAc	Refe-rences
Pt1	***C1QC***	NM_001114101.3:c.490G>A	p.Gly164Ser	Homozygous	Deleterious	Probably damaging	33	Likely pathogenic	1/121376	[27]
Pt2	***C1QC***	NM_001114101.3:c.121delC	p.Leu41CysfsTer97	Compound heterozygous	-	-	26.2	Pathogenic	Novel	
		NM_001114101.3:c.490G>A	p.Gly164Ser	Compound heterozygous	Deleterious	Probably damaging	33	Likely pathogenic	1/121376	
	CD19	NM_001178098.2:c.1638delA	p.Gly547GlufsTer33	Heterozygous	-	-	25.4	-	Novel	
	CLEC16A	NM_001114101.3:c.490G>A	p.His129Asp	Heterozygous	-	-	28.9	Undetermined significance	Novel	
Pt3	***C1QA***	NM_001347465.2:c.44delT	p.Ile15AsnfsTer7	Homozygous	-	-	21.3	Likely pathogenic	Novel	
Pt4	***C2***	NM_000063.4:c.841_849+19del	p.Val281_Arg283del	Homozygous	-	-	32	Likely pathogenic	619/121204 1 hom	[28]
Pt5	***C2***	NM_000063.4:c.841_849+19del	p.Val281_Arg283del	Homozygous	-	-	32	Likely pathogenic	619/121204 1 hom	
Pt6	***C2***	NM_000063.4:c.841_849+19del	p.Val281_Arg283del	Homozygous	-	-	32	Likely pathogenic	619/121204 1 hom	
Pt7	***DNASE1L3***	NM_004944.3:c.290_291delCA	p.Thr97IlefsTer2	Compound heterozygous	-	-	25.1	Likely pathogenic	10/121384	[29]
		Exon 5 deletion	-	Compound heterozygous	-	-		-	Novel	
Pt8	***IKZF1***	NM_006060.6:c.199A>T	p.Asp120Val	Heterozygous	Deleterious	Probably damaging	26.3	Likely pathogenic	Novel	
	Lyn	NM_002350.3:c.773T>C	p.Phe258Ser	Heterozygous	Deleterious	Probably damaging	25.8	Likely pathogenic	Novel	

Pt9	***TREX1***	NM_033629.6:c.236_243dupCTGCAGCC	p.Ser82LeufsTer9	Heterozygous	-	-	24.6	Pathogenic	Novel	
Pt10	***ACP5***	NM_001111035.3:c.79C>A	p.Arg27Ser	Heterozygous	Deleterious	Probably damaging	28.1	Undetermined significance	Novel	[23]
	RASGRP3	NM_001349975.2:c.686C>G	p.Ala229Gly	Heterozygous	Deleterious	Probably damaging	31	Undetermined significance	Novel	
Pt11	***RNASEH2C***	NM_032193.3:c.472C>G	p.His158Asp	Heterozygous	Deleterious	Probably damaging	28.8	Undetermined significance	Novel	
Pt12	***C9***	NM_001737.5:c.1209_1222del	p.Asp403GlufsTer5	Heterozygous	-	-	34	Undetermined significance	8/121256	
Pt13	***C8B***	NM_000066.3:c.-99_-98delinsAAG	-	Heterozygous	-	-	16.31		Novel	
Pt14	***C9***	NM_001737.5:c.1490C>T	p.Ala497Val	Heterozygous	Deleterious	Probably damaging	29.2	Likely pathogenic	Novel	
Pt15	***C3***	NM_000064.4:c.3278T>G	p.Ile1093Ser	Heterozygous	Deleterious	Probably damaging	31	Likely pathogenic	Novel	

Table 2: Rare, predicted damaging variants per patient in putative lupus causing (PLC) genes (GWAS and mouse models)

Patient	Gene	Heterozygous variants	Protein change	SIFT	Polyphen2	CADD
Pt16	***IRF7***	NM_004031.2:c.926A>T	p.Lys309Met	Deleterious	Probably damaging	24.9
Pt17	***CLEC16A***	NM_015226.3:c.908_910delGAG	p.Gly303del	-	-	15.75
	NFE2L2	NM_006164.4:c.-11_9del	-	-	-	22.8
	TNXB	ENST00000375244.7:c:12727G>A	p.Gly4243Arg	Deleterious	Probably damaging	23.9
Pt18	***TRAF3IP2***	NM_147686.3:c.1366G>A	p.Val456Met	Deleterious	Probably damaging	32
Pt19	***PDHX***	NM_003477.2:c.171T>G	p.Ile57Met	Deleterious	Probably damaging	24.0
Pt20	***JAZF1***	NM_175061.4:c.-23delA	-	-	-	18.15
Pt21	***PHRF1***	NM_020901.3:c.1355C>T	p.Pro452Leu	Deleterious	Possibly damaging	23.9
Pt22	***CD19***	NM_001178098.1:c.1318G>A	p.Glu440Lys	Deleterious	Probably damaging	33
Pt23	***CBLB***	NM_170662.4:c.621T>G	p.Ile207Met	Deleterious	Probably damaging	23.9
	TMEM39A	NM_018266.2:c.689_696del	p.Thr230Argfs*21_	-	-	20.7
	TMEM39A	NM_018266.2:c681_687del	p.Ser227Argfs*20	-	-	15.14
Pt24	***TNXB***	NM_019105.6:c.4535_4552del	p.Asp1512_Val1517del	-	-	16.12
Pt25	***PTPN22***	NM_015967.5:c.314C>T	p.Pro105Leu	Deleterious	Probably damaging	29.6
Pt26	***PDHX***	NM_003477.2:c.171T>G	p.Ile57Met	Deleterious	Probably damaging	24.0
Pt27	***WDFY4***	NM_020945.1:c657G>T	p.Gln219His	Deleterious	Probably damaging	18.49
Pt28	***CYBB***	NM_000397.3:c.1102G>T	p.Ala368Ser	Deleterious	Probably damaging	25.2
Pt29	***MECP2***	NM_001110792.1:c851C>T	p.Pro284Leu	Deleterious	Probably damaging	27.9

Supplementary appendix

Here is provided supplemental information by the authors.

Materials and Methods

Patients

One hundred and seventeen unrelated probands were identified from the UK Juvenile-onset SLE (JSLE) Cohort Study (Watson et al., 2012) and French SLE GENIAL (Clinicaltrials.gov, NCT01992666) cohorts. All patients fulfilled American College of Rheumatology (ACR) criteria for lupus and were diagnosed before the age of 16 years. Linked clinical data were recorded in the two respective nationwide databases. The study was approved by the Sud-Est III ethics committee (#2013-011B), French Data Protection Authority (CNIL, #DR-2013-354), French Advisory Committee on Healthcare Research Data Processing (CCTIRS, #2013.223), the National Research Ethics Service North West, Liverpool East, UK (reference 06/Q1502/77) and the Leeds (East) Research Ethics Committee, UK (reference number 10/H1307/132). In agreement with the International Genome Sample resource (IGSR; http://www.internationalgenome.org) criteria for the classification of ethnicity, the combined cohort of pediatric onset SLE examined here was made up of the following ethnicities: European (50%), African (33%) and South Asian (16%) (Figure 1A).

Controls

Control data were derived from the 1000 Genomes Project (1000g)[16]. The healthy control cohort comprised 791 unrelated individuals demonstrating a similar ethnic distribution to the cSLE cohort (50% European, 33% African; 16% South Asian), built by random sampling among the 1000g sample set. Two additional control populations were ascertained through the FREX consortium, a French-specific database of 574 individuals of French ancestry and a UK control panel: 74 adults from the north west of England,
The FREX consortium exome database is a French-specific database containing the exome data of a reference control panel of individuals of French ancestry (n = 574). (Website: http://lysine.univ-brest.fr/FrExAC/). The FREX cohort is a federative project that aims at building such a database and giving access to the scientific community exome data that can serve as controls in association studies. A total of 574 healthy individuals sampled in 6 French regions have been sequenced using the Agilent V5+UTR exome capture kit and genotyped on Illumina Core Exome SNP-chip.
A cohort of 74 adults from the north west of England and were screened using an identical pipeline, arising from the CTD project (LEAP), led by Pr. Ian Bruce and funded by the BRC. The patients were sequenced and analysed using exactly the same targeted exon panel, sequencing platform and filtering strategy as employed in the lupus cohort.

Panel designation

We designed a next-generation sequencing panel selecting genes after a systematic search of Pubmed to January 2013, considering combinations of the following terms: “Mouse models of lupus; GWAS studies related to lupus; monogenic lupus”.

DNA sequencing

Targeted enrichment and sequencing of our panel of 146 genes was undertaken using 200 ng of DNA extracted from peripheral blood. Enrichment was performed with the Sure Select Custom Target Enrichment Kit (Agilent Technologies, Santa Clara, CA, USA) for the Illumina HiSeq system following the manufacturer's protocols. Samples were paired-end sequenced on on a Illumina HiSeq 2500 sequencer (Illumina Inc, San Diego, CA, USA). More than 96% of targeted bases were sequenced to a read depth of greater than 30. Rare variants, defined here as a frequency on the Exome Aggregation Consortium (ExAC, 0.2) database of <0.1% for SNPs and <1% for indels, were selected for further analysis. Studied variants were confirmed by Sanger sequencing (primers available on request).

Variant discovery and annotation

Variant discovery analysis was performed following Genome Analysis Toolkit (GATK) best practice guidelines(Van der Auwera et al., 2013). Raw data (fastq files) were preliminarily split according to the number of reads for optimal base quality score recalibration. Reads were then aligned to the reference genome assembly GRCh37 with BWA-MEM (version 0.7.12), using appropriate read group annotations. Duplicate reads were marked using Picard (version 1.140). We utilized GATK (version 3.4) to realign reads around indels with the 1000 Genomes Project (1000g)(The 1000 Genomes Project Consortium, 2010) phase 1 indel call reference and 1000g gold standard indel sets. Same procedure was performed for the Frex consortium database and UK control population for SNPs only. Base quality scores were recalibrated using the two aforementioned indel sets and the dbSNP138 release. Analysis-ready reads belonging to the same sample, but with different lane number, were then merged. To increase the quality of the results, an additional round of duplicate read marking and realignment was performed on merged files.

Single nucleotide variants (SNV) and indels were called simultaneously on each sample via local reassembly using the GATK HaplotypeCaller tool to produce an intermediate variant set. Joint genotyping was then performed on the files from all 117 individuals. Variants were filtered using variant quality score recalibration, and low quality (GQ < 20) reads removed. Copy number variants (CNVs) were identified according to a read-depth approach. ExomeDepth v1.1.6(Plagnol et al., 2012) was applied to non-duplicate sequencing reads aligned to GRCh37. CNV calling was performed utilizing other samples generated through the same sequencing run and bioinformatics strategy. Variants were annotated using Ensembl Variant Effect Predictor (VEP) 83 and Annovar(Wang et al., 2010). We considered SIFT(Ng and Henikoff, 2003), Polyphen-2(Adzhubei et al., 2013) and Combined Annotation Dependent Depletion (CADD) v1.3(Kircher et al., 2014) scores for computational predictions of variant pathogenicity. For the UK control population, CADD>15 only was considered for the analysis.

Variant filtering

Variants extracted from the raw data in VCF files were filtered using the following steps: segregation into two subsets, insertions/deletions (indels) and SNVs; filtering of indels and SNVs according to their population frequencies within the ExAC dataset, where we employed 1% and 0.1% respectively in the case of putative biallelic mutations, or 0.01% and 0.001% in the case of heterozygous variants, as our thresholds (threshold (heterozygous) = threshold (homozygous)^2 i.e. 10^{-4} for indels and 10^{-6} for SNVs). Finally, we selected indels with a CADD score higher than 15, and SNVs according to the formula: CADD score ≥ 15 AND sift score ≤ 0.05 AND Polyphen 2 score ≥ 0.9. A cartoon of the characterization of different sequence variants is given in Figure S1 in the Supplementary Appendix. For the UK control population, CADD>15 only was considered for the analysis.

cDNA cloning, expression and DNA binding studies

cDNA coding for IKZF1 was a kind gift from A. Liston (Leuven). cDNA was PCR amplified and cloned in a p3xFlagCMV vector (Sigma-Aldrich, France) that enables expression of IKAROS N-terminal Flag tagged protein. Directed mutagenesis was performed on this construct to obtain the IKZF1 c.A359T (p.D120V) variant. For IKAROS nuclear staining experiments, Flag-tagged human WT or D120V Ikaros plasmids were transfected into NIH3T3 cells using JetPEI transfection reagent (PolyPlus Transfection, France). Forty eight hours after transfection, cells were stained using primary mouse anti-Flag antibody (Sigma-Aldrich, France) and secondary anti-mouse AF488 antibody (Jackson ImmunoResearch, UK). Nuclei were stained using DAPI (Thermo Fisher Scientific, France). Images were

collected on a LSM 710 confocal microscope (Zeiss, Germany) with a 63x immersion objective.

C1q Structure analysis

Structural analysis was performed using MacPymol 1.3 software and C1q structure PDB 1PK6(Gaboriaud et al., 2003).

IZKF1 functional data

Granta cell sample preparation
Granta cells were transduced with inducible lenti-vector pInducer21 (ORF-EG) (Addgene) expressing either wt-IKZF1 or D120V-IKZF1. Four days following transduction, GFP positive cells were sorted using a FACSAria Cell Sorter (Becton-Dickinson, San Jose, USA). For the IKZF1 transcriptomic study, cells were plated at 1.106 cells/ml in 6 well plate and treated or not with doxycycline at 0.5µg/ml during 6 hours. Cells were then pelleted and total RNA extracted using Direct-Zol RNA MicroPrep w/ Zymp-Spin Columns and following manufacturer's instructions (Ozyme). Total RNA was quantified using Quantifluor RNA system (Promega).

Library preparation and RNASeq and analysis: Libraries were generated from 1µg of total RNA from doxycycline induced or non induced Granta cell lines. Libraries were generated with SENSE™ mRNA-Seq Library Prep Kit V2 (Lexogen) following the manufacturer's instructions. Tagged library quality was checked on D1000 screen tape and analysed on Tape station 4200 (Agilent). Libraries sequencing was performed by the GenomEast platform, a member of the "France Génomique" consortium (ANR-10-INBS- 0009). Cutadapt 1.10 (--adapter AGATCGGAAGAGCACACGTCTGAACTCCAGTCAC --quality-cutoff 20,20 –anywhere "Af100g" --minimum-length 40) was used for read preprocessing: adapter, poly-A tail and low-quality (Phred quality score below 20) base trimming, removal of reads shorter than 40 bp after trimming. Reads mapping to rRNA sequences were also discarded. Reads were then mapped onto the hg38 assembly of Homo sapiens genome using STAR (7) version 2.5.3a (--twopassMode Basic). Gene expression was quantified using htseq-count (8) release 0.6.1p1 (--mode union --minaqual 10) and gene annotations from Ensembl release 94. Statistical analysis was performed using R and DESeq2 1.16.1 Bioconductor library (9). Genes were selected as differentially expressed using the following thresholds: adjusted p-value lower than 0.05 and absolute value of log2 Fold-Change greater than 1.

Statistical analysis
Fisher's exact test was used to compare the frequencies of variants in the control and cSLE cohorts. Data normality was assessed using a Shapiro-Wilk test and quantile-quantile plots. The mean of SNPs per individual was compared between control and cSLE cohorts using a Mann-Whitney U test (Wilcoxon rank-sum test). P-values were adjusted using the Bonferroni correction where appropriate. Odd ratios were calculated using the standard formula, with S1/2 the number of variants that passed a filter and F1/2 those which failed for the two groups compared (OR = S1*F2 / F1*S2).
SKAT-O analysis was performed rare variants with a MAF of 1% or 0.1% within the selected genes to analyse the genes differentially distributed in the lupus patients compared to controls The SKAT-O method combines burden and variance component tests (Lee et al., 2012).

Supplemental table S1: Genotypes associated with putative monogenic SLE, known lupus-causing genes (KLC, also referred as Mendeliome) (n = 28)(defined as of 2013)

Gene	Pathogenesis	Kegg pathways	References
ACP5	Interferon signaling	ko00740 Riboflavin metabolism ko01100 Metabolic pathways ko04142 Lysosome ko04380 Osteoclast differentiation ko05323 Rheumatoid arthritis	(An et al., 2016; Briggs et al., 2011; Lausch et al., 2011)
C1Q(A,B,C)	Complement	ko04610 Complement and coagulation cascades ko05020 Prion diseases ko05133 Pertussis ko05142 Chagas disease (American trypanosomiasis) ko05150 Staphylococcus aureus infection ko05322 Systemic lupus erythematosus	(Botto et al., 1998; Martens et al., 2009)
C1R	Complement	ko04145 Phagosome ko04610 Complement and coagulation cascades ko05133 Pertussis ko05150 Staphylococcus aureus infection ko05322 Systemic lupus erythematosus	(Lee et al., 1978)
C1S	Complement	ko04610 Complement and coagulation cascades ko05133 Pertussis ko05150 Staphylococcus aureus infection ko05322 Systemic lupus erythematosus	(Amano et al., 2008; Dragon-Durey et al., 2001; Lee et al., 1978)
C2	Complement	map00140 Steroid hormone biosynthesis	(Stern et al., 1976)
C3	Complement	hsa04145 Phagosome hsa04610 Complement and coagulation cascades hsa05133 Pertussis hsa05134 Legionellosis hsa05140 Leishmaniasis	(Tsukamoto et al., 2005)

		hsa05142 Chagas disease (American trypanosomiasis) hsa05150 Staphylococcus aureus infection hsa05152 Tuberculosis hsa05167 Kaposi sarcoma-associated herpesvirus infection hsa05168 Herpes simplex infection hsa05203 Viral carcinogenesis hsa05322 Systemic lupus erythematosus	
C4A	Complement	hsa04610 Complement and coagulation cascades hsa05133 Pertussis hsa05150 Staphylococcus aureus infection hsa05322 Systemic lupus erythematosus	(Chen et al., 2000; Fischer et al., 1998; Paul et al., 2002)
C4B	Complement	hsa04610 Complement and coagulation cascades hsa05133 Pertussis	(Hartung et al., 1992; Yang et al., 2007)
C5	Complement	ko04610 Complement and coagulation cascades ko05020 Prion diseases ko05133 Pertussis ko05150 Staphylococcus aureus infection ko05168 Herpes simplex infection ko05322 Systemic lupus erythematosus	(Asghar et al., 1991)
C6	Complement	ko04610 Complement and coagulation cascades ko05020 Prion diseases ko05322 Systemic lupus erythematosus	(Trapp et al., 1987)
C7	Complement	ko04610 Complement and coagulation cascades ko05020 Prion diseases ko05322 Systemic lupus erythematosus	(Segurado et al., 1992)

C8A	Complement	ko04610 Complement and coagulation cascades ko05020 Prion diseases ko05146 Amoebiasis ko05322 Systemic lupus erythematosus	(Jasin, 1977)
C8B	Complement	ko04610 Complement and coagulation cascades ko05020 Prion diseases ko05146 Amoebiasis ko05322 Systemic lupus erythematosus	(Jasin, 1977)
C9	Complement	ko04610 Complement and coagulation cascades ko05020 Prion diseases ko05146 Amoebiasis ko05322 Systemic lupus erythematosus	(Kawai et al., 1989)
DNASE1	Interferon signaling		(Bodaño et al., 2006; Napirei et al., 2000; Yasutomo et al., 2001)
DNASE1L3	Interferon signaling		(Al-Mayouf et al., 2011; Ozçakar et al., 2013)
FAS	Central tolerance	ko01524 Platinum drug resistance ko04010 MAPK signaling pathway ko04060 Cytokine-cytokine receptor interaction ko04115 p53 signaling pathway ko04210 Apoptosis ko04217 Necroptosis ko04650 Natural killer cell mediated cytotoxicity ko04668 TNF signaling pathway ko04932 Non-alcoholic fatty liver disease (NAFLD) ko04940 Type I diabetes mellitus ko05010 Alzheimer disease ko05142 Chagas disease (American trypanosomiasis) ko05143 African	(Adachi et al., 1993; Watson et al., 1992)

		trypanosomiasis ko05161 Hepatitis B ko05162 Measles ko05163 Human cytomegalovirus infection ko05164 Influenza A ko05165 Human papillomavirus infection ko05167 Kaposi sarcoma-associated herpesvirus infection ko05168 Herpes simplex infection ko05200 Pathways in cancer ko05205 Proteoglycans in cancer ko05320 Autoimmune thyroid disease ko05330 Allograft rejection ko05332 Graft-versus-host disease	
FASLG	Central tolerance	ko01524 Platinum drug resistance ko04010 MAPK signaling pathway ko04014 Ras signaling pathway ko04060 Cytokine-cytokine receptor interaction ko04068 FoxO signaling pathway ko04151 PI3K-Akt signaling pathway ko04210 Apoptosis ko04217 Necroptosis ko04650 Natural killer cell mediated cytotoxicity ko04722 Neurotrophin signaling pathway ko04932 Non-alcoholic fatty liver disease (NAFLD) ko04940 Type I diabetes mellitus ko05142 Chagas disease (American trypanosomiasis) ko05143 African trypanosomiasis	(Lynch et al., 1994; Takahashi et al., 1994)

		ko05161 Hepatitis B ko05162 Measles ko05163 Human cytomegalovirus infection ko05164 Influenza A ko05165 Human papillomavirus infection ko05168 Herpes simplex infection ko05200 Pathways in cancer ko05205 Proteoglycans in cancer ko05320 Autoimmune thyroid disease ko05330 Allograft rejection ko05332 Graft-versus-host disease	
IFIH1	Interferon signaling	ko04622 RIG-I-like receptor signaling pathway ko05161 Hepatitis B ko05162 Measles ko05164 Influenza A ko05168 Herpes simplex infection	(Robinson et al., 2011)
IKZF1	Central tolerance		(Hoshino et al., 2017)
MFGE8	Efferocytosis		(Hanayama et al., 2004; Hu et al., 2009; Yamaguchi et al., 2008)
PRKCD	Central tolerance	ko04062 Chemokine signaling pathway ko04140 Autophagy - animal ko04270 Vascular smooth muscle contraction ko04621 NOD-like receptor signaling pathway ko04625 C-type lectin receptor signaling pathway ko04666 Fc gamma R-mediated phagocytosis ko04722 Neurotrophin signaling pathway ko04750 Inflammatory mediator regulation of TRP channels ko04912 GnRH signaling pathway	(Belot et al., 2013; Mecklenbräuker et al., 2002; Miyamoto et al., 2002)

		ko04915 Estrogen signaling pathway ko04930 Type II diabetes mellitus ko04931 Insulin resistance ko04933 AGE-RAGE signaling pathway in diabetic complications	
RNASEH2A, RNASEH2B, RNASEH2C	Interferon signaling	ko03030 DNA replication	(Ramantani et al., 2010)
TREX1	Interferon signaling	ko04623 Cytosolic DNA-sensing pathway	(Lee-Kirsch et al., 2007)

Supplemental table S2: Genes selected from GWAS or mouse models of SLE, putative lupus-causing genes (PLC) (n = 119) (defined as of 2013)

Gene	Kegg pathways	-	Ascertainment	References
AFF1	ko05202	Transcriptional misregulation in cancer	GWAS	(Okada et al., 2012)
APCS			Mouse model	(Ehrenstein et al., 1998, 2000)
ATG5	ko04136	Autophagy - other	GWAS	(Harley et al., 2008)
	ko04137	Mitophagy - animal		
	ko04138	Autophagy - yeast		
	ko04140	Autophagy - animal		
	ko04211	Longevity regulating pathway		
	ko04213	Longevity regulating pathway - multiple species		
	ko04216	Ferroptosis		
	ko04621	NOD-like receptor signaling pathway		
	ko04622	RIG-I-like receptor signaling pathway		
	ko05131	Shigellosis		
BANK1			GWAS	(Kozyrev et al., 2008)
BCL2	ko01521	EGFR tyrosine kinase inhibitor resistance	Mouse model	(Kozyrev et al., 2008; Liphaus et al., 2006)
	ko01522	Endocrine resistance		
	ko01524	Platinum drug resistance		
	ko04064	NF-kappa B signaling pathway		
	ko04066	HIF-1 signaling pathway		
	ko04071	Sphingolipid signaling pathway		
	ko04115	p53 signaling pathway		

	ko04140	Autophagy - animal		
	ko04141	Protein processing in endoplasmic reticulum		
	ko04151	PI3K-Akt signaling pathway		
	ko04210	Apoptosis		
	ko04215	Apoptosis - multiple species		
	ko04217	Necroptosis		
	ko04261	Adrenergic signaling in cardiomyocytes		
	ko04340	Hedgehog signaling pathway		
	ko04510	Focal adhesion		
	ko04621	NOD-like receptor signaling pathway		
	ko04630	JAK-STAT signaling pathway		
	ko04722	Neurotrophin signaling pathway		
	ko04725	Cholinergic synapse		
	ko04915	Estrogen signaling pathway		
	ko04928	Parathyroid hormone synthesis, secretion and action		
	ko04933	AGE-RAGE signaling pathway in diabetic complications		
	ko05014	Amyotrophic lateral sclerosis (ALS)		
	ko05145	Toxoplasmosis		
	ko05152	Tuberculosis		
	ko05161	Hepatitis B		
	ko05169	Epstein-Barr virus infection		
	ko05200	Pathways in cancer		
	ko05206	MicroRNAs in cancer		
	ko05210	Colorectal cancer		
	ko05215	Prostate cancer		
	ko05222	Small cell lung cancer		
	ko05226	Gastric cancer		
	ko05418	Fluid shear stress and atherosclerosis		

BLK			GWAS; mouse model	(Hom et al., 2008; Samuelson et al., 2012)
C8ORF12			GWAS	(Hom et al., 2008)
CBLB	ko04012 ko04120 ko04144 ko04625 ko04660 ko04910 ko05162	ErbB signaling pathway Ubiquitin mediated proteolysis Endocytosis C-type lectin receptor signaling pathway T cell receptor signaling pathway Insulin signaling pathway Measles	Mouse model	(Bachmaier et al., 2000; Yi et al., 2000)
CD19	hsa04151 hsa04640 hsa04662 hsa05169 hsa05340	PI3K-Akt signaling pathway Hematopoietic cell lineage B cell receptor signaling pathway Epstein-Barr virus infection Primary immunodeficiency	Mouse model	(Rickert et al., 1995; Zhou et al., 1994)
CD22	hsa04514 hsa04640 hsa04662	Cell adhesion molecules (CAMs) Hematopoietic cell lineage B cell receptor signaling pathway	Mouse model	(Cornall et al., 1998; O'Keefe et al., 1996)
CD40LG	hsa04060 hsa04064 hsa04514 hsa04660	Cytokine-cytokine receptor interaction NF-kappa B signaling pathway Cell adhesion molecules (CAMs) T cell receptor signaling pathway	GWAS; mouse model	(Higuchi et al., 2002; Manea et al., 2009)

	hsa04672 hsa05144 hsa05145 hsa05310 hsa05320 hsa05322 hsa05330 hsa05340 hsa05416	Intestinal immune network for IgA production Malaria Toxoplasmosis Asthma Autoimmune thyroid disease Systemic lupus erythematosus Allograft rejection Primary immunodeficiency Viral myocarditis		
CD44	ko04512 ko04640 ko05131 ko05169 ko05205 ko05206	ECM-receptor interaction Hematopoietic cell lineage Shigellosis Epstein-Barr virus infection Proteoglycans in cancer MicroRNAs in cancer	GWAS	(Lessard et al., 2011)
CD48	ko04650	Natural killer cell mediated cytotoxicity	Mouse model	(Kumar et al., 2006; Morel et al., 2001)
CDKN1A	ko01522 ko01524 ko04012 ko04066 ko04068 ko04110 ko04115 ko04151 ko04218 ko04630	Endocrine resistance Platinum drug resistance ErbB signaling pathway HIF-1 signaling pathway FoxO signaling pathway Cell cycle p53 signaling pathway PI3K-Akt signaling pathway Cellular senescence JAK-STAT signaling pathway	GWAS; Mouse model	(Balomenos et al., 2000; Kim et al., 2009)

	ko04921	Oxytocin signaling pathway			
	ko04928	Parathyroid hormone synthesis, secretion and action			
	ko04934	Cushing syndrome			
	ko05160	Hepatitis C			
	ko05161	Hepatitis B			
	ko05163	Human cytomegalovirus infection			
	ko05165	Human papillomavirus infection			
	ko05166	HTLV-I infection			
	ko05167	Kaposi sarcoma-associated herpesvirus infection			
	ko05169	Epstein-Barr virus infection			
	ko05200	Pathways in cancer			
	ko05202	Transcriptional misregulation in cancer			
	ko05203	Viral carcinogenesis			
	ko05205	Proteoglycans in cancer			
	ko05206	MicroRNAs in cancer			
	ko05210	Colorectal cancer			
	ko05211	Renal cell carcinoma			
	ko05212	Pancreatic cancer			
	ko05213	Endometrial cancer			
	ko05214	Glioma			
	ko05215	Prostate cancer			
	ko05216	Thyroid cancer			
	ko05217	Basal cell carcinoma			
	ko05218	Melanoma			
	ko05219	Bladder cancer			
	ko05220	Chronic myeloid leukemia			
	ko05222	Small cell lung cancer			
	ko05223	Non-small cell lung cancer			
	ko05224	Breast cancer			

	ko05225 ko05226	Hepatocellular carcinoma Gastric cancer		
CFLAR	ko04064 ko04140 ko04210 ko04217 ko04668 ko05142	NF-kappa B signaling pathway Autophagy - animal Apoptosis Necroptosis TNF signaling pathway Chagas disease (American trypanosomiasis)	Mouse model	(Qiao et al., 2010; Shenoy et al., 2001)
CLEC16A			GWAS	(Zhang et al., 2011)
CORO1A	ko04145 ko05152	Phagosome Tuberculosis	Mouse model	(Haraldsson et al., 2008)
CR2	ko04610 ko04640 ko04662 ko05169	Complement and coagulation cascades Hematopoietic cell lineage B cell receptor signaling pathway Epstein-Barr virus infection	Mouse model	(Boackle et al., 2001; Chen et al., 2000)
CRP			GWAS	(Edberg et al., 2008)
CSF2	ko04060 ko04630 ko04640 ko04650 ko04657 ko04660	Cytokine-cytokine receptor interaction JAK-STAT signaling pathway Hematopoietic cell lineage Natural killer cell mediated cytotoxicity IL-17 signaling pathway T cell receptor signaling pathway	Mouse model	(Dranoff et al., 1994; Enzler et al., 2003)

	ko04664	Fc epsilon RI signaling pathway		
	ko04668	TNF signaling pathway		
	ko05132	Salmonella infection		
	ko05146	Amoebiasis		
	ko05166	HTLV-I infection		
	ko05167	Kaposi sarcoma-associated herpesvirus infection		
	ko05202	Transcriptional misregulation in cancer		
	ko05221	Acute myeloid leukemia		
	ko05323	Rheumatoid arthritis		
CTLA4	ko04514	Cell adhesion molecules (CAMs)	GWAS; Mouse model	(Barreto et al., 2009; Tivol et al., 1995)
	ko04660	T cell receptor signaling pathway		
	ko05320	Autoimmune thyroid disease		
	ko05323	Rheumatoid arthritis		
CYBB	ko04066	HIF-1 signaling pathway	Mouse model	(Campbell et al., 2012)
	ko04145	Phagosome		
	ko04216	Ferroptosis		
	ko04217	Necroptosis		
	ko04621	NOD-like receptor signaling pathway		
	ko04670	Leukocyte transendothelial migration		
	ko04933	AGE-RAGE signaling pathway in diabetic complications		
	ko05140	Leishmaniasis		
DEF6			Mouse model	(Fanzo et al., 2006)
E2F2	ko01522	Endocrine resistance	Mouse model	(Murga et al., 2001)
	ko04110	Cell cycle		
	ko04218	Cellular senescence		

	ko04934	Cushing syndrome		
	ko05161	Hepatitis B		
	ko05163	Human cytomegalovirus infection		
	ko05166	HTLV-I infection		
	ko05167	Kaposi sarcoma-associated herpesvirus infection		
	ko05200	Pathways in cancer		
	ko05206	MicroRNAs in cancer		
	ko05212	Pancreatic cancer		
	ko05214	Glioma		
	ko05215	Prostate cancer		
	ko05218	Melanoma		
	ko05219	Bladder cancer		
	ko05220	Chronic myeloid leukemia		
	ko05222	Small cell lung cancer		
	ko05223	Non-small cell lung cancer		
	ko05224	Breast cancer		
	ko05225	Hepatocellular carcinoma		
	ko05226	Gastric cancer		
ELF1	ko04214	Apoptosis - fly	GWAS	(Yang et al., 2007)
EP300	ko04024	cAMP signaling pathway	Mouse model	(Forster et al., 2007; Shikama et al., 2003)
	ko04066	HIF-1 signaling pathway		
	ko04068	FoxO signaling pathway		
	ko04110	Cell cycle		
	ko04310	Wnt signaling pathway		
	ko04330	Notch signaling pathway		
	ko04350	TGF-beta signaling pathway		
	ko04520	Adherens junction		
	ko04630	JAK-STAT signaling pathway		

	ko04720	Long-term potentiation		
	ko04916	Melanogenesis		
	ko04919	Thyroid hormone signaling pathway		
	ko04922	Glucagon signaling pathway		
	ko05016	Huntington disease		
	ko05152	Tuberculosis		
	ko05161	Hepatitis B		
	ko05164	Influenza A		
	ko05165	Human papillomavirus infection		
	ko05166	HTLV-I infection		
	ko05167	Kaposi sarcoma-associated herpesvirus infection		
	ko05168	Herpes simplex infection		
	ko05169	Epstein-Barr virus infection		
	ko05200	Pathways in cancer		
	ko05203	Viral carcinogenesis		
	ko05206	MicroRNAs in cancer		
	ko05211	Renal cell carcinoma		
	ko05215	Prostate cancer		
ETS1	ko04013	MAPK signaling pathway - fly	GWAS	(Han et al., 2009; Yang et al., 2010)
	ko04014	Ras signaling pathway		
	ko04218	Cellular senescence		
	ko04320	Dorso-ventral axis formation		
	ko05166	HTLV-I infection		
	ko05200	Pathways in cancer		
	ko05211	Renal cell carcinoma		
FCGR2A	ko04145	Phagosome	GWAS	(Harley et al., 2008)
	ko04380	Osteoclast differentiation		
	ko04611	Platelet activation		

	ko04666 ko05140 ko05150 ko05152 ko05322	Fc gamma R-mediated phagocytosis Leishmaniasis Staphylococcus aureus infection Tuberculosis Systemic lupus erythematosus		
FCGR2B	ko04145 ko04380 ko04662 ko04666 ko05150 ko05152 ko05162	Phagosome Osteoclast differentiation B cell receptor signaling pathway Fc gamma R-mediated phagocytosis Staphylococcus aureus infection Tuberculosis Measles	GWAS	(Bolland and Ravetch, 2000; Takai et al., 1996)
FCGR3A	ko04145 ko04380 ko04650 ko05140 ko05150 ko05152 ko05322	Phagosome Osteoclast differentiation Natural killer cell mediated cytotoxicity Leishmaniasis Staphylococcus aureus infection Tuberculosis Systemic lupus erythematosus	GWAS	(Edberg et al., 2002)
FCGR3B	ko04145 ko04380 ko04650 ko05140 ko05150 ko05152 ko05322	Phagosome Osteoclast differentiation Natural killer cell mediated cytotoxicity Leishmaniasis Staphylococcus aureus infection Tuberculosis Systemic lupus erythematosus	GWAS	Fanciulli et al. 2007, Nat. Genet

FLI1	ko05202	Transcriptional misregulation in cancer	GWAS; mouse model	(Morris et al., 2010; Zhang et al., 1995)
FYB	ko04015	Rap1 signaling pathway	GWAS	(Addobbati et al., 2013)
GADD45A	ko04010 ko04068 ko04110 ko04115 ko04210 ko04218 ko05200 ko05202 ko05210 ko05212 ko05213 ko05214 ko05216 ko05217 ko05218 ko05220 ko05222 ko05223 ko05224 ko05225 ko05226	MAPK signaling pathway FoxO signaling pathway Cell cycle p53 signaling pathway Apoptosis Cellular senescence Pathways in cancer Transcriptional misregulation in cancer Colorectal cancer Pancreatic cancer Endometrial cancer Glioma Thyroid cancer Basal cell carcinoma Melanoma Chronic myeloid leukemia Small cell lung cancer Non-small cell lung cancer Breast cancer Hepatocellular carcinoma Gastric cancer	Mouse model	(Hollander et al., 1999; Li et al., 2010)
GPR132			Mouse model	(Le et al., 2001)
ICA1	ko04940	Type I diabetes mellitus	GWAS	(Harley et al., 2008)
ICMT	ko00900	Terpenoid backbone biosynthesis	Mouse model	(Doyle et al., 2003)

	ko01130	Biosynthesis of antibiotics		
IFNA17	hsa04060	Cytokine-cytokine receptor interaction	GWAS; mouse model	(Li et al., 2005)
	hsa04151	PI3K-Akt signaling pathway		
	hsa04217	Necroptosis		
	hsa04620	Toll-like receptor signaling pathway		
	hsa04621	NOD-like receptor signaling pathway		
	hsa04622	RIG-I-like receptor signaling pathway		
	hsa04623	Cytosolic DNA-sensing pathway		
	hsa04630	Jak-STAT signaling pathway		
	hsa04650	Natural killer cell mediated cytotoxicity		
	hsa05152	Tuberculosis		
	hsa05160	Hepatitis C		
	hsa05161	Hepatitis B		
	hsa05162	Measles		
	hsa05163	Human cytomegalovirus infection		
	hsa05164	Influenza A		
	hsa05165	Human papillomavirus infection		
	hsa05167	Kaposi sarcoma-associated herpesvirus infection		
	hsa05168	Herpes simplex infection		
	hsa05200	Pathways in cancer		
	hsa05320	Autoimmune thyroid disease		
IFNG	ko03050	Proteasome	Mouse model	(Hirankarn et al., 2009; Seery et al., 1997)
	ko04060	Cytokine-cytokine receptor interaction		
	ko04066	HIF-1 signaling pathway		
	ko04217	Necroptosis		
	ko04350	TGF-beta signaling pathway		
	ko04380	Osteoclast differentiation		
	ko04612	Antigen processing and presentation		

	ko04630	JAK-STAT signaling pathway		
	ko04650	Natural killer cell mediated cytotoxicity		
	ko04657	IL-17 signaling pathway		
	ko04658	Th1 and Th2 cell differentiation		
	ko04659	Th17 cell differentiation		
	ko04660	T cell receptor signaling pathway		
	ko04940	Type I diabetes mellitus		
	ko05132	Salmonella infection		
	ko05140	Leishmaniasis		
	ko05142	Chagas disease (American trypanosomiasis)		
	ko05143	African trypanosomiasis		
	ko05144	Malaria		
	ko05145	Toxoplasmosis		
	ko05146	Amoebiasis		
	ko05152	Tuberculosis		
	ko05162	Measles		
	ko05164	Influenza A		
	ko05168	Herpes simplex infection		
	ko05169	Epstein-Barr virus infection		
	ko05200	Pathways in cancer		
	ko05321	Inflammatory bowel disease (IBD)		
	ko05322	Systemic lupus erythematosus		
	ko05323	Rheumatoid arthritis		
	ko05330	Allograft rejection		
	ko05332	Graft-versus-host disease		
	ko05418	Fluid shear stress and atherosclerosis		
IKBKE	ko04620	Toll-like receptor signaling pathway	GWAS	(Wang et al., 2013)
	ko04621	NOD-like receptor signaling pathway		
	ko04622	RIG-I-like receptor signaling pathway		
	ko04623	Cytosolic DNA-sensing pathway		

	ko04625	C-type lectin receptor signaling pathway		
	ko04657	IL-17 signaling pathway		
	ko05160	Hepatitis C		
	ko05161	Hepatitis B		
	ko05162	Measles		
	ko05164	Influenza A		
	ko05165	Human papillomavirus infection		
	ko05167	Kaposi sarcoma-associated herpesvirus infection		
	ko05168	Herpes simplex infection		
IKZF3			GWAS	(Lessard et al., 2012; Wang et al., 1998)
IL10	ko04060	Cytokine-cytokine receptor interaction	GWAS	(Wang et al., 2013; Yin et al., 2002)
	ko04068	FoxO signaling pathway		
	ko04625	C-type lectin receptor signaling pathway		
	ko04630	JAK-STAT signaling pathway		
	ko04660	T cell receptor signaling pathway		
	ko04672	Intestinal immune network for IgA production		
	ko05133	Pertussis		
	ko05140	Leishmaniasis		
	ko05142	Chagas disease (American trypanosomiasis)		
	ko05143	African trypanosomiasis		
	ko05144	Malaria		
	ko05145	Toxoplasmosis		
	ko05146	Amoebiasis		
	ko05150	Staphylococcus aureus infection		
	ko05152	Tuberculosis		
	ko05169	Epstein-Barr virus infection		

	ko05310 ko05320 ko05321 ko05322 ko05330	Asthma Autoimmune thyroid disease Inflammatory bowel disease (IBD) Systemic lupus erythematosus Allograft rejection		
IL2	ko04060 ko04151 ko04625 ko04630 ko04658 ko04659 ko04660 ko04672 ko04940 ko05142 ko05162 ko05166 ko05200 ko05320 ko05321 ko05330 ko05332	Cytokine-cytokine receptor interaction PI3K-Akt signaling pathway C-type lectin receptor signaling pathway JAK-STAT signaling pathway Th1 and Th2 cell differentiation Th17 cell differentiation T cell receptor signaling pathway Intestinal immune network for IgA production Type I diabetes mellitus Chagas disease (American trypanosomiasis) Measles HTLV-I infection Pathways in cancer Autoimmune thyroid disease Inflammatory bowel disease (IBD) Allograft rejection Graft-versus-host disease	Mouse model	(Crispín and Tsokos, 2009; Schorle et al., 1991)
IL2RA	hsa04060 hsa04144 hsa04151	Cytokine-cytokine receptor interaction Endocytosis PI3K-Akt signaling pathway	Mouse model	(Carr et al., 2009; Willerford et al., 1995)

	hsa04630 hsa04640 hsa04658 hsa04659 hsa05162 hsa05166 hsa05200	Jak-STAT signaling pathway Hematopoietic cell lineage Th1 and Th2 cell differentiation Th17 cell differentiation Measles HTLV-I infection Pathways in cancer		
IL2RB	hsa04060 hsa04144 hsa04151 hsa04630 hsa04658 hsa04659 hsa05162 hsa05166 hsa05200 hsa05202	Cytokine-cytokine receptor interaction Endocytosis PI3K-Akt signaling pathway Jak-STAT signaling pathway Th1 and Th2 cell differentiation Th17 cell differentiation Measles HTLV-I infection Pathways in cancer Transcriptional misregulation in cancer	Mouse model	(Lieberman and Tsokos, 2010; Suzuki et al., 1995)
IL4	ko04060 ko04151 ko04630 ko04640 ko04657 ko04658 ko04659 ko04660 ko04664	Cytokine-cytokine receptor interaction PI3K-Akt signaling pathway JAK-STAT signaling pathway Hematopoietic cell lineage IL-17 signaling pathway Th1 and Th2 cell differentiation Th17 cell differentiation T cell receptor signaling pathway Fc epsilon RI signaling pathway	GWAS	(Erb et al., 1997; Wu et al., 2003; Yu et al., 2010)

	ko04672	Intestinal immune network for IgA production		
	ko05140	Leishmaniasis		
	ko05162	Measles		
	ko05200	Pathways in cancer		
	ko05310	Asthma		
	ko05320	Autoimmune thyroid disease		
	ko05321	Inflammatory bowel disease (IBD)		
	ko05330	Allograft rejection		
IRAK1	ko04010	MAPK signaling pathway	GWAS	(Jacob et al., 2007; Sawalha et al., 2008)
	ko04064	NF-kappa B signaling pathway		
	ko04620	Toll-like receptor signaling pathway		
	ko04624	Toll and Imd signaling pathway		
	ko04722	Neurotrophin signaling pathway		
	ko05133	Pertussis		
	ko05140	Leishmaniasis		
	ko05142	Chagas disease (American trypanosomiasis)		
	ko05145	Toxoplasmosis		
	ko05152	Tuberculosis		
	ko05162	Measles		
	ko05169	Epstein-Barr virus infection		
IRF5	ko04620	Toll-like receptor signaling pathway	GWAS	(Sigurdsson et al., 2005)
IRF7	hsa04620	Toll-like receptor signaling pathway	GWAS	(Fu et al., 2011; Harley et al., 2008)
	hsa04621	NOD-like receptor signaling pathway		
	hsa04622	RIG-I-like receptor signaling pathway		
	hsa04623	Cytosolic DNA-sensing pathway		
	hsa05160	Hepatitis C		

	hsa05161 hsa05162 hsa05164 hsa05167 hsa05168 hsa05203	Hepatitis B Measles Influenza A Kaposi sarcoma-associated herpesvirus infection Herpes simplex infection Viral carcinogenesis		
IRF8	hsa05133	Pertussis	GWAS	(Lessard et al., 2012)
ITGAM	hsa04015 hsa04145 hsa04514 hsa04610 hsa04640 hsa04670 hsa04810 hsa05133 hsa05134 hsa05140 hsa05146 hsa05150 hsa05152 hsa05202 hsa05221	Rap1 signaling pathway Phagosome Cell adhesion molecules (CAMs) Complement and coagulation cascades Hematopoietic cell lineage Leukocyte transendothelial migration Regulation of actin cytoskeleton Pertussis Legionellosis Leishmaniasis Amoebiasis Staphylococcus aureus infection Tuberculosis Transcriptional misregulation in cancer Acute myeloid leukemia	GWAS	(Harley et al., 2008; Nath et al., 2008)
ITPR3	hsa04020 hsa04022 hsa04070 hsa04114	Calcium signaling pathway cGMP-PKG signaling pathway Phosphatidylinositol signaling system Oocyte meiosis	GWAS	(Oishi et al., 2008)

	hsa04210	Apoptosis		
	hsa04218	Cellular senescence		
	hsa04270	Vascular smooth muscle contraction		
	hsa04371	Apelin signaling pathway		
	hsa04540	Gap junction		
	hsa04611	Platelet activation		
	hsa04621	NOD-like receptor signaling pathway		
	hsa04625	C-type lectin receptor signaling pathway		
	hsa04713	Circadian entrainment		
	hsa04720	Long-term potentiation		
	hsa04723	Retrograde endocannabinoid signaling		
	hsa04724	Glutamatergic synapse		
	hsa04725	Cholinergic synapse		
	hsa04726	Serotonergic synapse		
	hsa04728	Dopaminergic synapse		
	hsa04730	Long-term depression		

	hsa04742 hsa04750 hsa04911 hsa04912 hsa04915 hsa04918 hsa04921 hsa04922 hsa04924 hsa04925 hsa04927 hsa04928 hsa04934 hsa04970 hsa04971 hsa04972 hsa05010 hsa05163 hsa05167 hsa05205	Taste transduction Inflammatory mediator regulation of TRP channels Insulin secretion GnRH signaling pathway Estrogen signaling pathway Thyroid hormone synthesis Oxytocin signaling pathway Glucagon signaling pathway Renin secretion Aldosterone synthesis and secretion Cortisol synthesis and secretion Parathyroid hormone synthesis, secretion and action Cushing syndrome Salivary secretion Gastric acid secretion Pancreatic secretion Alzheimer disease Human cytomegalovirus infection Kaposi sarcoma-associated herpesvirus infection Proteoglycans in cancer		
JAZF1	H00409	Type II diabetes mellitus	GWAS	(Gateva et al., 2009)
JUNB	ko04380 ko04668	Osteoclast differentiation TNF signaling pathway	Mouse model	(Meixner et al., 2008; Pflegerl et al., 2009)
KLK1	hsa04614 hsa04961	Renin-angiotensin system Endocrine and other factor-regulated	GWAS	(Liu et al., 2009)

		calcium reabsorption		
KRAS	hsa01521	EGFR tyrosine kinase inhibitor resistance	Mouse model	(Quaio et al., 2012)
	hsa01522	Endocrine resistance		
	hsa04010	MAPK signaling pathway		
	hsa04012	ErbB signaling pathway		
	hsa04014	Ras signaling pathway		
	hsa04015	Rap1 signaling pathway		
	hsa04062	Chemokine signaling pathway		
	hsa04068	FoxO signaling pathway		
	hsa04071	Sphingolipid signaling pathway		
	hsa04072	Phospholipase D signaling pathway		
	hsa04137	Mitophagy - animal		
	hsa04140	Autophagy - animal		
	hsa04150	mTOR signaling pathway		
	hsa04151	PI3K-Akt signaling pathway		
	hsa04210	Apoptosis		
	hsa04211	Longevity regulating pathway		
	hsa04213	Longevity regulating pathway - multiple species		
	hsa04218	Cellular senescence		
	hsa04360	Axon guidance		
	hsa04370	VEGF signaling pathway		
	hsa04371	Apelin signaling pathway		
	hsa04540	Gap junction		
	hsa04550	Signaling pathways regulating pluripotency of stem cells		
	hsa04625	C-type lectin receptor signaling pathway		
	hsa04650	Natural killer cell mediated cytotoxicity		
	hsa04660	T cell receptor signaling pathway		
	hsa04662	B cell receptor signaling pathway		

	hsa04664	Fc epsilon RI signaling pathway		
	hsa04714	Thermogenesis		
	hsa04720	Long-term potentiation		
	hsa04722	Neurotrophin signaling pathway		
	hsa04725	Cholinergic synapse		
	hsa04726	Serotonergic synapse		
	hsa04730	Long-term depression		
	hsa04810	Regulation of actin cytoskeleton		
	hsa04910	Insulin signaling pathway		
	hsa04912	GnRH signaling pathway		
	hsa04914	Progesterone-mediated oocyte maturation		
	hsa04915	Estrogen signaling pathway		
	hsa04916	Melanogenesis		
	hsa04917	Prolactin signaling pathway		
	hsa04919	Thyroid hormone signaling pathway		
	hsa04921	Oxytocin signaling pathway		
	hsa04926	Relaxin signaling pathway		
	hsa04933	AGE-RAGE signaling pathway in diabetic complications		
	hsa04960	Aldosterone-regulated sodium reabsorption		
	hsa05034	Alcoholism		
	hsa05160	Hepatitis C		
	hsa05161	Hepatitis B		
	hsa05163	Human cytomegalovirus infection		
	hsa05165	Human papillomavirus infection		
	hsa05166	HTLV-I infection		
	hsa05167	Kaposi sarcoma-associated herpesvirus infection		
	hsa05200	Pathways in cancer		
	hsa05203	Viral carcinogenesis		
	hsa05205	Proteoglycans in cancer		

	hsa05206	MicroRNAs in cancer		
	hsa05210	Colorectal cancer		
	hsa05211	Renal cell carcinoma		
	hsa05212	Pancreatic cancer		
	hsa05213	Endometrial cancer		
	hsa05214	Glioma		
	hsa05215	Prostate cancer		
	hsa05216	Thyroid cancer		
	hsa05218	Melanoma		
	hsa05219	Bladder cancer		
	hsa05220	Chronic myeloid leukemia		
	hsa05221	Acute myeloid leukemia		
	hsa05223	Non-small cell lung cancer		
	hsa05224	Breast cancer		
	hsa05225	Hepatocellular carcinoma		
	hsa05226	Gastric cancer		
	hsa05230	Central carbon metabolism in cancer		
	hsa05231	Choline metabolism in cancer		
LAT	hsa04014	Ras signaling pathway	Mouse model	(Aguado et al., 2002; Sommers et al., 2002)
	hsa04015	Rap1 signaling pathway		
	hsa04064	NF-kappa B signaling pathway		
	hsa04650	Natural killer cell mediated cytotoxicity		
	hsa04658	Th1 and Th2 cell differentiation		
	hsa04659	Th17 cell differentiation		
	hsa04660	T cell receptor signaling pathway		
	hsa04664	Fc epsilon RI signaling pathway		
	hsa04666	Fc gamma R-mediated phagocytosis		
LBH			GWAS	(Yu et al., 2013)
LRRC18			GWAS	(Yang et al., 2010)

LY9			Mouse model	(Kumar et al., 2006; Morel et al., 2001)
LYN	ko04062	Chemokine signaling pathway	GWAS, Mouse model	(Harley et al., 2008; Hibbs et al., 1995)
	ko04064	NF-kappa B signaling pathway		
	ko04611	Platelet activation		
	ko04662	B cell receptor signaling pathway		
	ko04664	Fc epsilon RI signaling pathway		
	ko04666	Fc gamma R-mediated phagocytosis		
	ko04730	Long-term depression		
	ko05120	Epithelial cell signaling in Helicobacter pylori infection		
	ko05167	Kaposi sarcoma-associated herpesvirus infection		
	ko05169	Epstein-Barr virus infection		
	ko05203	Viral carcinogenesis		
MAN2A1	hsa00510	N-Glycan biosynthesis	Mouse model	(Chui et al., 1997)
	hsa01100	Metabolic pathways		
MAP3K1	ko04010	MAPK signaling pathway		(Cedeño et al., 2003; Sawalha et al., 2008)
	ko04120	Ubiquitin mediated proteolysis		
	ko04530	Tight junction		
	ko04622	RIG-I-like receptor signaling pathway		
	ko04722	Neurotrophin signaling pathway		
	ko04912	GnRH signaling pathway		
	ko05161	Hepatitis B		
	ko05166	Human T-cell leukemia virus 1 infection		
MARCO	ko04145	Phagosome	Mouse model	(Rogers et al., 2009)

MARK2			Mouse model	(Hurov et al., 2001)
MBL2	hsa04145 hsa04610 hsa05150	Phagosome Complement and coagulation cascades Staphylococcus aureus infection	GWAS	(Font et al., 2007)
MECP2			Mouse model	(Sawalha et al., 2008)
MERTK			Mouse model	(Scott et al., 2001; Wu et al., 2011)
MTA2			Mouse model	(Lu et al., 2008)
NFE2L2	ko04141 ko04212 ko05200 ko05225 ko05418	Protein processing in endoplasmic reticulum Longevity regulating pathway - worm Pathways in cancer Hepatocellular carcinoma Fluid shear stress and atherosclerosis	GWAS; mouse model	(Córdova et al., 2010; Li et al., 2004)
P2RX7	ko04020 ko04080 ko04621	Calcium signaling pathway Neuroactive ligand-receptor interaction NOD-like receptor signaling pathway	Mouse model	(Elliott et al., 2005; Portales-Cervantes et al., 2010)
PDCD1	hsa04514 hsa04660	Cell adhesion molecules (CAMs) T cell receptor signaling pathway	GWAS	(Nishimura et al., 1999; Prokunina et al., 2002)
PDHX	ko01100	Metabolic pathways	GWAS	(Lessard et al., 2011)
PECAM1	ko04514 ko04670 ko05144	Cell adhesion molecules (CAMs) Leukocyte transendothelial migration Malaria	Mouse model	(Wilkinson et al., 2002)

	ko05418	Fluid shear stress and atherosclerosis		
PHRF1			GWAS	(Harley et al., 2008)
PPARD	ko03320 ko04310 ko05200 ko05221	PPAR signaling pathway Wnt signaling pathway Pathways in cancer Acute myeloid leukemia	Mouse model	(Mukundan et al., 2009)
PPARG	ko03320 ko04152 ko04211 ko04380 ko04714 ko05016 ko05200 ko05202 ko05216	PPAR signaling pathway AMPK signaling pathway Longevity regulating pathway Osteoclast differentiation Thermogenesis Huntington disease Pathways in cancer Transcriptional misregulation in cancer Thyroid cancer	Mouse model	(Oxer et al., 2011; Rosner et al., 2011; Yeh et al., 2008)
PRDM1			GWAS	(Gateva et al., 2009)
PRKCB	ko01521 ko04010 ko04012 ko04014 ko04015 ko04020 ko04062 ko04064 ko04066	EGFR tyrosine kinase inhibitor resistance MAPK signaling pathway ErbB signaling pathway Ras signaling pathway Rap1 signaling pathway Calcium signaling pathway Chemokine signaling pathway NF-kappa B signaling pathway HIF-1 signaling pathway	GWAS	(Sheng et al., 2011)

	ko04070	Phosphatidylinositol signaling system			
	ko04071	Sphingolipid signaling pathway			
	ko04150	mTOR signaling pathway			
	ko04270	Vascular smooth muscle contraction			
	ko04310	Wnt signaling pathway			
	ko04370	VEGF signaling pathway			
	ko04510	Focal adhesion			
	ko04540	Gap junction			
	ko04650	Natural killer cell mediated cytotoxicity			
	ko04662	B cell receptor signaling pathway			
	ko04666	Fc gamma R-mediated phagocytosis			
	ko04670	Leukocyte transendothelial migration			
	ko04713	Circadian entrainment			
	ko04720	Long-term potentiation			
	ko04723	Retrograde endocannabinoid signaling			
	ko04724	Glutamatergic synapse			
	ko04725	Cholinergic synapse			
	ko04726	Serotonergic synapse			
	ko04727	GABAergic synapse			
	ko04728	Dopaminergic synapse			
	ko04730	Long-term depression			
	ko04750	Inflammatory mediator regulation of TRP channels			
	ko04911	Insulin secretion			
	ko04912	GnRH signaling pathway			
	ko04916	Melanogenesis			
	ko04918	Thyroid hormone synthesis			
	ko04919	Thyroid hormone signaling pathway			
	ko04921	Oxytocin signaling pathway			
	ko04925	Aldosterone synthesis and secretion			
	ko04928	Parathyroid hormone synthesis, secretion			

		and action		
	ko04931	Insulin resistance		
	ko04933	AGE-RAGE signaling pathway in diabetic complications		
	ko04960	Aldosterone-regulated sodium reabsorption		
	ko04961	Endocrine and other factor-regulated calcium reabsorption		
	ko04970	Salivary secretion		
	ko04971	Gastric acid secretion		
	ko04972	Pancreatic secretion		
	ko04973	Carbohydrate digestion and absorption		
	ko05031	Amphetamine addiction		
	ko05032	Morphine addiction		
	ko05140	Leishmaniasis		
	ko05143	African trypanosomiasis		
	ko05146	Amoebiasis		
	ko05161	Hepatitis B		
	ko05163	Human cytomegalovirus infection		
	ko05200	Pathways in cancer		
	ko05205	Proteoglycans in cancer		
	ko05206	MicroRNAs in cancer		
	ko05214	Glioma		
	ko05223	Non-small cell lung cancer		
	ko05225	Hepatocellular carcinoma		
	ko05231	Choline metabolism in cancer		
PTEN	ko00562	Inositol phosphate metabolism	Mouse model	(Di Cristofano et al., 1998, 1999)
	ko01521	EGFR tyrosine kinase inhibitor resistance		
	ko04068	FoxO signaling pathway		
	ko04070	Phosphatidylinositol signaling system		
	ko04071	Sphingolipid signaling pathway		

	ko04115	p53 signaling pathway		
	ko04140	Autophagy - animal		
	ko04150	mTOR signaling pathway		
	ko04151	PI3K-Akt signaling pathway		
	ko04212	Longevity regulating pathway - worm		
	ko04218	Cellular senescence		
	ko04510	Focal adhesion		
	ko04931	Insulin resistance		
	ko05161	Hepatitis B		
	ko05165	Human papillomavirus infection		
	ko05200	Pathways in cancer		
	ko05206	MicroRNAs in cancer		
	ko05213	Endometrial cancer		
	ko05214	Glioma		
	ko05215	Prostate cancer		
	ko05218	Melanoma		
	ko05222	Small cell lung cancer		
	ko05224	Breast cancer		
	ko05225	Hepatocellular carcinoma		
	ko05230	Central carbon metabolism in cancer		
PTPN22			GWAS; mouse model	(Kyogoku et al., 2004; Zikherman et al., 2009)
PTPN6	ko04520	Adherens junction	Mouse model	(Green and Shultz, 1975; Shultz et al., 1993; Tsui et al., 1993)
	ko04630	JAK-STAT signaling pathway		
	ko04650	Natural killer cell mediated cytotoxicity		
	ko04660	T cell receptor signaling pathway		
	ko04662	B cell receptor signaling pathway		

	ko05140 ko05205	Leishmaniasis Proteoglycans in cancer		
PTPRC	ko04514 ko04660 ko04666 ko05340	Cell adhesion molecules (CAMs) T cell receptor signaling pathway Fc gamma R-mediated phagocytosis Primary immunodeficiency	Mouse model	(Jury et al., 2007; Majeti et al., 2000)
PTTG1	hsa04110 hsa04114 hsa05166	Cell cycle Oocyte meiosis HTLV-I infection	GWAS	(Harley et al., 2008)
PXK			GWAS	(Harley et al., 2008)
RASGRP1	hsa04010 hsa04014 hsa04015 hsa04662 hsa05200	MAPK signaling pathway Ras signaling pathway Rap1 signaling pathway B cell receptor signaling pathway Pathways in cancer	Mouse model	(Priatel et al., 2007)
RASGRP3			GWAS	(Han et al., 2009)
RASSF5	hsa04014 hsa04015 hsa04218 hsa04670 hsa05200 hsa05223	Ras signaling pathway Rap1 signaling pathway Cellular senescence Leukocyte transendothelial migration Pathways in cancer Non-small cell lung cancer	Mouse model	(Katagiri et al., 2004, 2011)
RC3H1			Mouse model	(Vinuesa et al., 2005)
RXRA			Mouse model	(Núñez et al., 2010; Rosner et

				al., 2011)
SCUBE1			GWAS	(Harley et al., 2008)
SELP	hsa04514 hsa05144 hsa05150	Cell adhesion molecules (CAMs) Malaria Staphylococcus aureus infection	GWAS	(Jacob et al., 2007)
SH2D2A	hsa04370	VEGF signaling pathway	Mouse model	(Drappa et al., 2003)
SLAMF1			Mouse model	(Keszei et al., 2011)
SLAMF6			GWAS	(Kumar et al., 2006; Morel et al., 2001; Wandstrat et al., 2004)
SLC15A4			GWAS	(Han et al., 2009)
STAT1	hsa04062 hsa04217 hsa04380 hsa04620 hsa04621 hsa04625 hsa04630 hsa04658 hsa04659 hsa04917 hsa04919 hsa04933 hsa05140	Chemokine signaling pathway Necroptosis Osteoclast differentiation Toll-like receptor signaling pathway NOD-like receptor signaling pathway C-type lectin receptor signaling pathway Jak-STAT signaling pathway Th1 and Th2 cell differentiation Th17 cell differentiation Prolactin signaling pathway Thyroid hormone signaling pathway AGE-RAGE signaling pathway in diabetic complications Leishmaniasis	GWAS	(Remmers et al., 2007)

	hsa05145 hsa05152 hsa05160 hsa05161 hsa05162 hsa05164 hsa05165 hsa05167 hsa05168 hsa05200 hsa05212 hsa05321	Toxoplasmosis Tuberculosis Hepatitis C Hepatitis B Measles Influenza A Human papillomavirus infection Kaposi sarcoma-associated herpesvirus infection Herpes simplex infection Pathways in cancer Pancreatic cancer Inflammatory bowel disease (IBD)		
STAT4	ko04217 ko04630 ko04658 ko05161 ko05200 ko05321	Necroptosis JAK-STAT signaling pathway Th1 and Th2 cell differentiation Hepatitis B Pathways in cancer Inflammatory bowel disease (IBD)	GWAS	(Remmers et al., 2007)
STRA13	ko03460	Fanconi anemia pathway	Mouse model	(Sun et al., 2001)
TGFB1	ko04010 ko04060 ko04068 ko04110 ko04218 ko04350 ko04380 ko04390 ko04659	MAPK signaling pathway Cytokine-cytokine receptor interaction FoxO signaling pathway Cell cycle Cellular senescence TGF-beta signaling pathway Osteoclast differentiation Hippo signaling pathway Th17 cell differentiation	Mouse model	(Dang et al., 1995; Geiser et al., 1993)

	ko04672	Intestinal immune network for IgA production		
	ko04926	Relaxin signaling pathway		
	ko04932	Non-alcoholic fatty liver disease (NAFLD)		
	ko04933	AGE-RAGE signaling pathway in diabetic complications		
	ko05140	Leishmaniasis		
	ko05142	Chagas disease (American trypanosomiasis)		
	ko05144	Malaria		
	ko05145	Toxoplasmosis		
	ko05146	Amoebiasis		
	ko05152	Tuberculosis		
	ko05161	Hepatitis B		
	ko05166	HTLV-I infection		
	ko05200	Pathways in cancer		
	ko05205	Proteoglycans in cancer		
	ko05210	Colorectal cancer		
	ko05211	Renal cell carcinoma		
	ko05212	Pancreatic cancer		
	ko05220	Chronic myeloid leukemia		
	ko05225	Hepatocellular carcinoma		
	ko05226	Gastric cancer		
	ko05321	Inflammatory bowel disease (IBD)		
	ko05323	Rheumatoid arthritis		
	ko05410	Hypertrophic cardiomyopathy (HCM)		
	ko05414	Dilated cardiomyopathy (DCM)		
TLR7	ko04620	Toll-like receptor signaling pathway	GWAS; mouse model	(Fairhurst et al., 2008; Pisitkun et al., 2006)
	ko05162	Measles		
	ko05164	Influenza A		

TLR9	ko04620 ko05142 ko05143 ko05144 ko05152 ko05162 ko05168	Toll-like receptor signaling pathway Chagas disease (American trypanosomiasis) African trypanosomiasis Malaria Tuberculosis Measles Herpes simplex infection	GWAS; mouse model	(Christensen et al., 2005; Huang et al., 2012)
TMEM39A			GWAS	(Lessard et al., 2012)
TNFAIP3	ko04064 ko04217 ko04621 ko04657 ko04668 ko05162 ko05169	NF-kappa B signaling pathway Necroptosis NOD-like receptor signaling pathway IL-17 signaling pathway TNF signaling pathway Measles Epstein-Barr virus infection	GWAS	(Graham et al., 2008)
TNFRSF13B	hsa04060 hsa04672 hsa05340	Cytokine-cytokine receptor interaction Intestinal immune network for IgA production Primary immunodeficiency	Mouse model	(Seshasayee et al., 2003)
TNFRSF13C	hsa04060 hsa04064 hsa04672 hsa05166 hsa05340	Cytokine-cytokine receptor interaction NF-kappa B signaling pathway Intestinal immune network for IgA production HTLV-I infection Primary immunodeficiency	Mouse model	(Jury et al., 2007)
TNFSF13B	hsa04060	Cytokine-cytokine receptor interaction	GWAS; mouse model	(Mackay et al.,

	hsa04064 hsa04672 hsa05323	NF-kappa B signaling pathway Intestinal immune network for IgA production Rheumatoid arthritis		1999)
TNFSF14	hsa04060 hsa04064 hsa05168	Cytokine-cytokine receptor interaction NF-kappa B signaling pathway Herpes simplex infection	GWAS	(Wang et al., 2001)
TNFSF4	hsa04060	Cytokine-cytokine receptor interaction	GWAS	(Cunninghame Graham et al., 2008)
TNIP1			GWAS	(Gateva et al., 2009; Han et al., 2009)
TNXB	hsa04151 hsa04510 hsa04512 hsa05165 hsa05206	PI3K-Akt signaling pathway Focal adhesion ECM-receptor interaction Human papillomavirus infection MicroRNAs in cancer	GWAS	(Kamatani et al., 2008)
TRAF3IP2			Mouse model	(Perricone et al., 2013)
TRIM21	hsa05322	Systemic lupus erythematosus	Mouse model	(Espinosa et al., 2009; Frank et al., 1993)
TROVE2	hsa05322	Systemic lupus erythematosus	Mouse model	(Schulte-Pelkum et al., 2009; Xue et al., 2003)
TYK2	hsa04217 hsa04380 hsa04621	Necroptosis Osteoclast differentiation NOD-like receptor signaling pathway	GWAS	(Sigurdsson et al., 2005)

	hsa04630 hsa04658 hsa04659 hsa05145 hsa05160 hsa05162 hsa05164 hsa05165 hsa05167 hsa05168 hsa05169	Jak-STAT signaling pathway Th1 and Th2 cell differentiation Th17 cell differentiation Toxoplasmosis Hepatitis C Measles Influenza A Human papillomavirus infection Kaposi sarcoma-associated herpesvirus infection Herpes simplex infection Epstein-Barr virus infection		
UBE2L3	hsa04120 hsa05012	Ubiquitin mediated proteolysis Parkinson disease	GWAS	(Harley et al., 2008)
UHRF1BP1			GWAS	(Gateva et al., 2009)
WDFY4			GWAS	(Yang et al., 2010)
XKR6			GWAS	(Budarf et al., 2011)

Adachi, M., Watanabe-Fukunaga, R., and Nagata, S. (1993). Aberrant transcription caused by the insertion of an early transposable element in an intron of the Fas antigen gene of lpr mice. Proc. Natl. Acad. Sci. U.S.A. *90*, 1756–1760.

Aguado, E., Richelme, S., Nuñez-Cruz, S., Miazek, A., Mura, A.-M., Richelme, M., Guo, X.-J., Sainty, D., He, H.-T., Malissen, B., et al. (2002). Induction of T helper type 2 immunity by a point mutation in the LAT adaptor. Science *296*, 2036–2040.

Al-Mayouf, S.M., Sunker, A., Abdwani, R., Abrawi, S.A., Almurshedi, F., Alhashmi, N., Al Sonbul, A., Sewairi, W., Qari, A., Abdallah, E., et al. (2011). Loss-of-function variant in DNASE1L3 causes a familial form of systemic lupus erythematosus. Nat. Genet. *43*, 1186–1188.

Amano, M.T., Ferriani, V.P.L., Florido, M.P.C., Reis, E.S., Delcolli, M.I.M.V., Azzolini, A.E.C.S., Assis-Pandochi, A.I., Sjöholm, A.G., Farah, C.S., Jensenius, J.C., et al. (2008). Genetic analysis of complement C1s deficiency associated with systemic lupus erythematosus highlights alternative splicing of normal C1s gene. Molecular Immunology *45*, 1693–1702.

Asghar, S.S., Venneker, G.T., van Meegen, M., Meinardi, M.M.H.M., Hulsmans, R.-F.H.J., and de Waal, L.P. (1991). Hereditary deficiency of C5 in association with discoid lupus erythematosus. Journal of the American Academy of Dermatology *24*, 376–378.

Belot, A., Kasher, P.R., Trotter, E.W., Foray, A.-P., Debaud, A.-L., Rice, G.I., Szynkiewicz, M., Zabot, M.-T., Rouvet, I., Bhaskar, S.S., et al. (2013). Protein kinase cδ deficiency causes mendelian systemic lupus erythematosus with B cell-defective apoptosis and hyperproliferation. Arthritis Rheum. *65*, 2161–2171.
Bodaño, A., González, A., Ferreiros-Vidal, I., Balada, E., Ordi, J., Carreira, P., Gómez-Reino, J.J., and Conde, C. (2006). Association of a non-synonymous single-nucleotide polymorphism of DNASEI with SLE susceptibility. Rheumatology (Oxford) *45*, 819–823.
Botto, M., Agnola, C.D., Bygrave, A.E., Thompson, E.M., Cook, H.T., Petry, F., Loos, M., Pandolfi, P.P., and Walport, M.J. (1998). Homozygous C1q deficiency causes glomerulonephritis associated with multiple apoptotic bodies. Nature Genetics *19*, 56–59.
Briggs, T.A., Rice, G.I., Daly, S., Urquhart, J., Gornall, H., Bader-Meunier, B., Baskar, K., Baskar, S., Baudouin, V., Beresford, M.W., et al. (2011). Tartrate-resistant acid phosphatase deficiency causes a bone dysplasia with autoimmunity and a type I interferon expression signature. Nature Genetics *43*, 127–131.
Budarf, M.L., Goyette, P., Boucher, G., Lian, J., Graham, R.R., Claudio, J.O., Hudson, T., Gladman, D., Clarke, A.E., Pope, J.E., et al. (2011). A targeted association study in systemic lupus erythematosus identifies multiple susceptibility alleles. Genes Immun. *12*, 51–58.
Carr, E.J., Clatworthy, M.R., Lowe, C.E., Todd, J.A., Wong, A., Vyse, T.J., Kamesh, L., Watts, R.A., Lyons, P.A., and Smith, K.G.C. (2009). Contrasting genetic association of IL2RA with SLE and ANCA-associated vasculitis. BMC Med. Genet. *10*, 22.
Cedeño, S., Cifarelli, D.F., Blasini, A.M., Paris, M., Placeres, F., Alonso, G., and Rodriguez, M.A. (2003). Defective activity of ERK-1 and ERK-2 mitogen-activated protein kinases in peripheral blood T lymphocytes from patients with systemic lupus erythematosus: potential role of altered coupling of Ras guanine nucleotide exchange factor hSos to adapter protein Grb2 in lupus T cells. Clin. Immunol. *106*, 41–49.
Chen, Z., Koralov, S.B., and Kelsoe, G. (2000). Complement C4 inhibits systemic autoimmunity through a mechanism independent of complement receptors CR1 and CR2. J. Exp. Med. *192*, 1339–1352.
Christensen, S.R., Kashgarian, M., Alexopoulou, L., Flavell, R.A., Akira, S., and Shlomchik, M.J. (2005). Toll-like receptor 9 controls anti-DNA autoantibody production in murine lupus. J. Exp. Med. *202*, 321–331.
Chui, D., Oh-Eda, M., Liao, Y.F., Panneerselvam, K., Lal, A., Marek, K.W., Freeze, H.H., Moremen, K.W., Fukuda, M.N., and Marth, J.D. (1997). Alpha-mannosidase-II deficiency results in dyserythropoiesis and unveils an alternate pathway in oligosaccharide biosynthesis. Cell *90*, 157–167.
Córdova, E.J., Velázquez-Cruz, R., Centeno, F., Baca, V., and Orozco, L. (2010). The NRF2 gene variant, -653G/A, is associated with nephritis in childhood-onset systemic lupus erythematosus. Lupus *19*, 1237–1242.
Crispín, J.C., and Tsokos, G.C. (2009). Transcriptional regulation of IL-2 in health and autoimmunity. Autoimmun Rev *8*, 190–195.
Cunninghame Graham, D.S., Graham, R.R., Manku, H., Wong, A.K., Whittaker, J.C., Gaffney, P.M., Moser, K.L., Rioux, J.D., Altshuler, D., Behrens, T.W., et al. (2008). Polymorphism at the TNF superfamily gene TNFSF4 confers susceptibility to systemic lupus erythematosus. Nat. Genet. *40*, 83–89.
Dang, H., Geiser, A.G., Letterio, J.J., Nakabayashi, T., Kong, L., Fernandes, G., and Talal, N. (1995). SLE-like autoantibodies and Sjögren's syndrome-like lymphoproliferation in TGF-beta knockout mice. J. Immunol. *155*, 3205–3212.
Di Cristofano, A., Pesce, B., Cordon-Cardo, C., and Pandolfi, P.P. (1998). Pten is essential for embryonic development and tumour suppression. Nat. Genet. *19*, 348–355.
Di Cristofano, A., Kotsi, P., Peng, Y.F., Cordon-Cardo, C., Elkon, K.B., and Pandolfi, P.P. (1999). Impaired Fas response and autoimmunity in

Pten+/- mice. Science *285*, 2122–2125.
Dragon-Durey, M.-A., Quartier, P., Frémeaux-Bacchi, V., Blouin, J., Barace, C. de, Prieur, A.-M., Weiss, L., and Fridman, W.-H. (2001). Molecular Basis of a Selective C1s Deficiency Associated with Early Onset Multiple Autoimmune Diseases. The Journal of Immunology *166*, 7612–7616.
Drappa, J., Kamen, L.A., Chan, E., Georgiev, M., Ashany, D., Marti, F., and King, P.D. (2003). Impaired T cell death and lupus-like autoimmunity in T cell-specific adapter protein-deficient mice. J. Exp. Med. *198*, 809–821.
Ehrenstein, M.R., O'Keefe, T.L., Davies, S.L., and Neuberger, M.S. (1998). Targeted gene disruption reveals a role for natural secretory IgM in the maturation of the primary immune response. Proc. Natl. Acad. Sci. U.S.A. *95*, 10089–10093.
Ehrenstein, M.R., Cook, H.T., and Neuberger, M.S. (2000). Deficiency in serum immunoglobulin (Ig)M predisposes to development of IgG autoantibodies. J. Exp. Med. *191*, 1253–1258.
Elliott, J.I., McVey, J.H., and Higgins, C.F. (2005). The P2X7 receptor is a candidate product of murine and human lupus susceptibility loci: a hypothesis and comparison of murine allelic products. Arthritis Res. Ther. *7*, R468-475.
Erb, K.J., Rüger, B., von Brevern, M., Ryffel, B., Schimpl, A., and Rivett, K. (1997). Constitutive expression of interleukin (IL)-4 in vivo causes autoimmune-type disorders in mice. J. Exp. Med. *185*, 329–339.
Espinosa, A., Dardalhon, V., Brauner, S., Ambrosi, A., Higgs, R., Quintana, F.J., Sjöstrand, M., Eloranta, M.-L., Ní Gabhann, J., Winqvist, O., et al. (2009). Loss of the lupus autoantigen Ro52/Trim21 induces tissue inflammation and systemic autoimmunity by disregulating the IL-23-Th17 pathway. J. Exp. Med. *206*, 1661–1671.
Fairhurst, A.-M., Hwang, S., Wang, A., Tian, X.-H., Boudreaux, C., Zhou, X.J., Casco, J., Li, Q.-Z., Connolly, J.E., and Wakeland, E.K. (2008). Yaa autoimmune phenotypes are conferred by overexpression of TLR7. Eur. J. Immunol. *38*, 1971–1978.
Fischer, M.B., Ma, M., Hsu, N.C., and Carroll, M.C. (1998). Local Synthesis of C3 Within the Splenic Lymphoid Compartment Can Reconstitute the Impaired Immune Response in C3-Deficient Mice. The Journal of Immunology *160*, 2619–2625.
Font, J., Ramos-Casals, M., Brito-Zerón, P., Nardi, N., Ibañez, A., Suarez, B., Jiménez, S., Tàssies, D., García-Criado, A., Ros, E., et al. (2007). Association of mannose-binding lectin gene polymorphisms with antiphospholipid syndrome, cardiovascular disease and chronic damage in patients with systemic lupus erythematosus. Rheumatology (Oxford) *46*, 76–80.
Frank, M.B., Itoh, K., Fujisaku, A., Pontarotti, P., Mattei, M.G., and Neas, B.R. (1993). The mapping of the human 52-kD Ro/SSA autoantigen gene to human chromosome 11, and its polymorphisms. Am. J. Hum. Genet. *52*, 183–191.
Fu, Q., Zhao, J., Qian, X., Wong, J.L.H., Kaufman, K.M., Yu, C.Y., Hwee Siew Howe, Tan Tock Seng Hospital Lupus Study Group, Mok, M.Y., Harley, J.B., et al. (2011). Association of a functional IRF7 variant with systemic lupus erythematosus. Arthritis Rheum. *63*, 749–754.
Gateva, V., Sandling, J.K., Hom, G., Taylor, K.E., Chung, S.A., Sun, X., Ortmann, W., Kosoy, R., Ferreira, R.C., Nordmark, G., et al. (2009). A large-scale replication study identifies TNIP1, PRDM1, JAZF1, UHRF1BP1 and IL10 as risk loci for systemic lupus erythematosus. Nat. Genet. *41*, 1228–1233.
Geiser, A.G., Letterio, J.J., Kulkarni, A.B., Karlsson, S., Roberts, A.B., and Sporn, M.B. (1993). Transforming growth factor beta 1 (TGF-beta 1) controls expression of major histocompatibility genes in the postnatal mouse: aberrant histocompatibility antigen expression in the pathogenesis of the TGF-beta 1 null mouse phenotype. Proc. Natl. Acad. Sci. U.S.A. *90*, 9944–9948.
Graham, R.R., Cotsapas, C., Davies, L., Hackett, R., Lessard, C.J., Leon, J.M., Burtt, N.P., Guiducci, C., Parkin, M., Gates, C., et al. (2008).

Genetic variants near TNFAIP3 on 6q23 are associated with systemic lupus erythematosus. Nat. Genet. *40*, 1059–1061.
Green, M.C., and Shultz, L.D. (1975). Motheaten, an immunodeficient mutant of the mouse. I. Genetics and pathology. J. Hered. *66*, 250–258.
Han, J.-W., Zheng, H.-F., Cui, Y., Sun, L.-D., Ye, D.-Q., Hu, Z., Xu, J.-H., Cai, Z.-M., Huang, W., Zhao, G.-P., et al. (2009). Genome-wide association study in a Chinese Han population identifies nine new susceptibility loci for systemic lupus erythematosus. Nat. Genet. *41*, 1234–1237.
Hanayama, R., Tanaka, M., Miyasaka, K., Aozasa, K., Koike, M., Uchiyama, Y., and Nagata, S. (2004). Autoimmune disease and impaired uptake of apoptotic cells in MFG-E8-deficient mice. Science *304*, 1147–1150.
Harley, J.B., Genetics (SLEGEN), T.I.C. for S.L.E., Alarcón-Riquelme, M.E., Criswell, L.A., Jacob, C.O., Kimberly, R.P., Moser, K.L., Tsao, B.P., Vyse, T.J., and Langefeld, C.D. (2008). Genome-wide association scan in women with systemic lupus erythematosus identifies susceptibility variants in *ITGAM*, *PXK*, *KIAA1542* and other loci. Nature Genetics *40*, 204–210.
Hartung, K., Baur, M.P., Coldewey, R., Fricke, M., Kalden, J.R., Lakomek, H.J., Peter, H.H., Schendel, D., Schneider, P.M., and Seuchter, S.A. (1992). Major histocompatibility complex haplotypes and complement C4 alleles in systemic lupus erythematosus. Results of a multicenter study. J Clin Invest *90*, 1346–1351.
Hibbs, M.L., Tarlinton, D.M., Armes, J., Grail, D., Hodgson, G., Maglitto, R., Stacker, S.A., and Dunn, A.R. (1995). Multiple defects in the immune system of Lyn-deficient mice, culminating in autoimmune disease. Cell *83*, 301–311.
Hoshino, A., Okada, S., Yoshida, K., Nishida, N., Okuno, Y., Ueno, H., Yamashita, M., Okano, T., Tsumura, M., Nishimura, S., et al. (2017). Abnormal hematopoiesis and autoimmunity in human subjects with germline IKZF1 mutations. J. Allergy Clin. Immunol. *140*, 223–231.
Hu, C.Y., Wu, C.S., Tsai, H.F., Chang, S.K., Tsai, W.I., and Hsu, P.N. (2009). Genetic polymorphism in milk fat globule-EGF factor 8 (MFG-E8) is associated with systemic lupus erythematosus in human. Lupus *18*, 676–681.
Huang, C.-M., Huang, P.-H., Chen, C.-L., Lin, Y.-J., Tsai, C.-H., Huang, W.-L., and Tsai, F.-J. (2012). Association of toll-like receptor 9 gene polymorphism in Chinese patients with systemic lupus erythematosus in Taiwan. Rheumatol. Int. *32*, 2105–2109.
Hurov, J.B., Stappenbeck, T.S., Zmasek, C.M., White, L.S., Ranganath, S.H., Russell, J.H., Chan, A.C., Murphy, K.M., and Piwnica-Worms, H. (2001). Immune system dysfunction and autoimmune disease in mice lacking Emk (Par-1) protein kinase. Mol. Cell. Biol. *21*, 3206–3219.
Jacob, C.O., Reiff, A., Armstrong, D.L., Myones, B.L., Silverman, E., Klein-Gitelman, M., McCurdy, D., Wagner-Weiner, L., Nocton, J.J., Solomon, A., et al. (2007). Identification of novel susceptibility genes in childhood-onset systemic lupus erythematosus using a uniquely designed candidate gene pathway platform. Arthritis Rheum. *56*, 4164–4173.
Jasin, H.E. (1977). Absence of the eighth component of complement in association with systemic lupus erythematosus-like disease. J. Clin. Invest. *60*, 709–715.
Jury, E.C., Flores-Borja, F., and Kabouridis, P.S. (2007). Lipid rafts in T cell signalling and disease. Semin. Cell Dev. Biol. *18*, 608–615.
Kamatani, Y., Matsuda, K., Ohishi, T., Ohtsubo, S., Yamazaki, K., Iida, A., Hosono, N., Kubo, M., Yumura, W., Nitta, K., et al. (2008). Identification of a significant association of a single nucleotide polymorphism in TNXB with systemic lupus erythematosus in a Japanese population. J. Hum. Genet. *53*, 64–73.
Katagiri, K., Ohnishi, N., Kabashima, K., Iyoda, T., Takeda, N., Shinkai, Y., Inaba, K., and Kinashi, T. (2004). Crucial functions of the Rap1 effector molecule RAPL in lymphocyte and dendritic cell trafficking. Nat. Immunol. *5*, 1045–1051.
Katagiri, K., Ueda, Y., Tomiyama, T., Yasuda, K., Toda, Y., Ikehara, S., Nakayama, K.I., and Kinashi, T. (2011). Deficiency of Rap1-binding

protein RAPL causes lymphoproliferative disorders through mislocalization of p27kip1. Immunity *34*, 24–38.
Kawai, T., Katoh, K., Narita, M., Tani, K., and Okubo, T. (1989). Deficiency of the 9th component of complement (C9) in a patient with systemic lupus erythematosus. J. Rheumatol. *16*, 542–543.
Keszei, M., Latchman, Y.E., Vanguri, V.K., Brown, D.R., Detre, C., Morra, M., Arancibia-Carcamo, C.V., Arancibia, C.V., Paul, E., Calpe, S., et al. (2011). Auto-antibody production and glomerulonephritis in congenic Slamf1-/- and Slamf2-/- [B6.129] but not in Slamf1-/- and Slamf2-/- [BALB/c.129] mice. Int. Immunol. *23*, 149–158.
Kumar, K.R., Li, L., Yan, M., Bhaskarabhatla, M., Mobley, A.B., Nguyen, C., Mooney, J.M., Schatzle, J.D., Wakeland, E.K., and Mohan, C. (2006). Regulation of B cell tolerance by the lupus susceptibility gene Ly108. Science *312*, 1665–1669.
Kyogoku, C., Langefeld, C.D., Ortmann, W.A., Lee, A., Selby, S., Carlton, V.E.H., Chang, M., Ramos, P., Baechler, E.C., Batliwalla, F.M., et al. (2004). Genetic association of the R620W polymorphism of protein tyrosine phosphatase PTPN22 with human SLE. Am. J. Hum. Genet. *75*, 504–507.
Lausch, E., Janecke, A., Bros, M., Trojandt, S., Alanay, Y., De Laet, C., Hübner, C.A., Meinecke, P., Nishimura, G., Matsuo, M., et al. (2011). Genetic deficiency of tartrate-resistant acid phosphatase associated with skeletal dysplasia, cerebral calcifications and autoimmunity. Nat. Genet. *43*, 132–137.
Lee, S.L., Wallace, S.L., Barone, R., Blum, L., and Harvey Chase, P. (1978). Familial deficiency of two subunits of the first component of complement. c1r and c1s associated with a lupus erythematosus-like disease. Arthritis & Rheumatism *21*, 958–967.
Lee-Kirsch, M.A., Gong, M., Chowdhury, D., Senenko, L., Engel, K., Lee, Y.-A., de Silva, U., Bailey, S.L., Witte, T., Vyse, T.J., et al. (2007). Mutations in the gene encoding the 3'-5' DNA exonuclease TREX1 are associated with systemic lupus erythematosus. Nat. Genet. *39*, 1065–1067.
Lessard, C.J., Adrianto, I., Kelly, J.A., Kaufman, K.M., Grundahl, K.M., Adler, A., Williams, A.H., Gallant, C.J., Marta E. Alarcón-Riquelme on behalf of the BIOLUPUS and GENLES Networks, Anaya, J.-M., et al. (2011). Identification of a systemic lupus erythematosus susceptibility locus at 11p13 between PDHX and CD44 in a multiethnic study. Am. J. Hum. Genet. *88*, 83–91.
Lessard, C.J., Adrianto, I., Ice, J.A., Wiley, G.B., Kelly, J.A., Glenn, S.B., Adler, A.J., Li, H., Rasmussen, A., Williams, A.H., et al. (2012). Identification of IRF8, TMEM39A, and IKZF3-ZPBP2 as susceptibility loci for systemic lupus erythematosus in a large-scale multiracial replication study. Am. J. Hum. Genet. *90*, 648–660.
Li, J., Stein, T.D., and Johnson, J.A. (2004). Genetic dissection of systemic autoimmune disease in Nrf2-deficient mice. Physiol. Genomics *18*, 261–272.
Lieberman, L.A., and Tsokos, G.C. (2010). The IL-2 Defect in Systemic Lupus Erythematosus Disease Has an Expansive Effect on Host Immunity. J Biomed Biotechnol *2010*.
Liu, K., Li, Q.-Z., Delgado-Vega, A.M., Abelson, A.-K., Sánchez, E., Kelly, J.A., Li, L., Liu, Y., Zhou, J., Yan, M., et al. (2009). Kallikrein genes are associated with lupus and glomerular basement membrane-specific antibody-induced nephritis in mice and humans. J. Clin. Invest. *119*, 911–923.
Lu, X., Kovalev, G.I., Chang, H., Kallin, E., Knudsen, G., Xia, L., Mishra, N., Ruiz, P., Li, E., Su, L., et al. (2008). Inactivation of NuRD Component Mta2 Causes Abnormal T Cell Activation and Lupus-like Autoimmune Disease in Mice. J. Biol. Chem. *283*, 13825–13833.
Lynch, D.H., Watson, M.L., Alderson, M.R., Baum, P.R., Miller, R.E., Tough, T., Gibson, M., Davis-Smith, T., Smith, C.A., and Hunter, K. (1994).

The mouse Fas-ligand gene is mutated in gld mice and is part of a TNF family gene cluster. Immunity *1*, 131–136.
Mackay, F., Woodcock, S.A., Lawton, P., Ambrose, C., Baetscher, M., Schneider, P., Tschopp, J., and Browning, J.L. (1999). Mice transgenic for BAFF develop lymphocytic disorders along with autoimmune manifestations. J. Exp. Med. *190*, 1697–1710.
Majeti, R., Xu, Z., Parslow, T.G., Olson, J.L., Daikh, D.I., Killeen, N., and Weiss, A. (2000). An inactivating point mutation in the inhibitory wedge of CD45 causes lymphoproliferation and autoimmunity. Cell *103*, 1059–1070.
Martens, H.A., Zuurman, M.W., de Lange, A.H.M., Nolte, I.M., van der Steege, G., Navis, G.J., Kallenberg, C.G.M., Seelen, M.A., and Bijl, M. (2009). Analysis of C1q polymorphisms suggests association with systemic lupus erythematosus, serum C1q and CH50 levels and disease severity. Ann. Rheum. Dis. *68*, 715–720.
Mecklenbräuker, I., Saijo, K., Zheng, N.-Y., Leitges, M., and Tarakhovsky, A. (2002). Protein kinase Cdelta controls self-antigen-induced B-cell tolerance. Nature *416*, 860–865.
Meixner, A., Zenz, R., Schonthaler, H.B., Kenner, L., Scheuch, H., Penninger, J.M., and Wagner, E.F. (2008). Epidermal JunB represses G-CSF transcription and affects haematopoiesis and bone formation. Nat. Cell Biol. *10*, 1003–1011.
Miyamoto, A., Nakayama, K., Imaki, H., Hirose, S., Jiang, Y., Abe, M., Tsukiyama, T., Nagahama, H., Ohno, S., Hatakeyama, S., et al. (2002). Increased proliferation of B cells and auto-immunity in mice lacking protein kinase Cdelta. Nature *416*, 865–869.
Morel, L., Blenman, K.R., Croker, B.P., and Wakeland, E.K. (2001). The major murine systemic lupus erythematosus susceptibility locus, Sle1, is a cluster of functionally related genes. Proc. Natl. Acad. Sci. U.S.A. *98*, 1787–1792.
Mukundan, L., Odegaard, J.I., Morel, C.R., Heredia, J.E., Mwangi, J.W., Ricardo-Gonzalez, R.R., Goh, Y.P.S., Eagle, A.R., Dunn, S.E., Awakuni, J.U.H., et al. (2009). PPAR-delta senses and orchestrates clearance of apoptotic cells to promote tolerance. Nat. Med. *15*, 1266–1272.
Napirei, M., Karsunky, H., Zevnik, B., Stephan, H., Mannherz, H.G., and Möröy, T. (2000). Features of systemic lupus erythematosus in Dnase1-deficient mice. Nature Genetics *25*, 177–181.
Nath, S.K., Han, S., Kim-Howard, X., Kelly, J.A., Viswanathan, P., Gilkeson, G.S., Chen, W., Zhu, C., McEver, R.P., Kimberly, R.P., et al. (2008). A nonsynonymous functional variant in integrin-alpha(M) (encoded by ITGAM) is associated with systemic lupus erythematosus. Nat. Genet. *40*, 152–154.
Nishimura, H., Nose, M., Hiai, H., Minato, N., and Honjo, T. (1999). Development of lupus-like autoimmune diseases by disruption of the PD-1 gene encoding an ITIM motif-carrying immunoreceptor. Immunity *11*, 141–151.
Núñez, V., Alameda, D., Rico, D., Mota, R., Gonzalo, P., Cedenilla, M., Fischer, T., Boscá, L., Glass, C.K., Arroyo, A.G., et al. (2010). Retinoid X receptor alpha controls innate inflammatory responses through the up-regulation of chemokine expression. Proc. Natl. Acad. Sci. U.S.A. *107*, 10626–10631.
Oishi, T., Iida, A., Otsubo, S., Kamatani, Y., Usami, M., Takei, T., Uchida, K., Tsuchiya, K., Saito, S., Ohnisi, Y., et al. (2008). A functional SNP in the NKX2.5-binding site of ITPR3 promoter is associated with susceptibility to systemic lupus erythematosus in Japanese population. J. Hum. Genet. *53*, 151–162.
Oxer, D.S., Godoy, L.C., Borba, E., Lima-Salgado, T., Passos, L.A., Laurindo, I., Kubo, S., Barbeiro, D.F., Fernandes, D., Laurindo, F.R., et al. (2011). PPARγ expression is increased in systemic lupus erythematosus patients and represses CD40/CD40L signaling pathway. Lupus *20*, 575–587.

Ozçakar, Z.B., Foster, J., Diaz-Horta, O., Kasapcopur, O., Fan, Y.-S., Yalçınkaya, F., and Tekin, M. (2013). DNASE1L3 mutations in hypocomplementemic urticarial vasculitis syndrome. Arthritis Rheum. *65*, 2183–2189.
Paul, E., Pozdnyakova, O.O., Mitchell, E., and Carroll, M.C. (2002). Anti-DNA autoreactivity in C4-deficient mice. European Journal of Immunology *32*, 2672–2679.
Perricone, C., Ciccacci, C., Ceccarelli, F., Di Fusco, D., Spinelli, F.R., Cipriano, E., Novelli, G., Valesini, G., Conti, F., and Borgiani, P. (2013). TRAF3IP2 gene and systemic lupus erythematosus: association with disease susceptibility and pericarditis development. Immunogenetics *65*, 703–709.
Pflegerl, P., Vesely, P., Hantusch, B., Schlederer, M., Zenz, R., Janig, E., Steiner, G., Meixner, A., Petzelbauer, P., Wolf, P., et al. (2009). Epidermal loss of JunB leads to a SLE phenotype due to hyper IL-6 signaling. PNAS *106*, 20423–20428.
Pisitkun, P., Deane, J.A., Difilippantonio, M.J., Tarasenko, T., Satterthwaite, A.B., and Bolland, S. (2006). Autoreactive B cell responses to RNA-related antigens due to TLR7 gene duplication. Science *312*, 1669–1672.
Portales-Cervantes, L., Niño-Moreno, P., Doníz-Padilla, L., Baranda-Candido, L., García-Hernández, M., Salgado-Bustamante, M., González-Amaro, R., and Portales-Pérez, D. (2010). Expression and function of the P2X(7) purinergic receptor in patients with systemic lupus erythematosus and rheumatoid arthritis. Hum. Immunol. *71*, 818–825.
Priatel, J.J., Chen, X., Zenewicz, L.A., Shen, H., Harder, K.W., Horwitz, M.S., and Teh, H.-S. (2007). Chronic immunodeficiency in mice lacking RasGRP1 results in CD4 T cell immune activation and exhaustion. J. Immunol. *179*, 2143–2152.
Prokunina, L., Castillejo-López, C., Oberg, F., Gunnarsson, I., Berg, L., Magnusson, V., Brookes, A.J., Tentler, D., Kristjansdóttir, H., Gröndal, G., et al. (2002). A regulatory polymorphism in PDCD1 is associated with susceptibility to systemic lupus erythematosus in humans. Nat. Genet. *32*, 666–669.
Quaio, C.R.D.C., Carvalho, J.F., da Silva, C.A., Bueno, C., Brasil, A.S., Pereira, A.C., Jorge, A.A.L., Malaquias, A.C., Kim, C.A., and Bertola, D.R. (2012). Autoimmune disease and multiple autoantibodies in 42 patients with RASopathies. Am. J. Med. Genet. A *158A*, 1077–1082.
Ramantani, G., Kohlhase, J., Hertzberg, C., Innes, A.M., Engel, K., Hunger, S., Borozdin, W., Mah, J.K., Ungerath, K., Walkenhorst, H., et al. (2010). Expanding the phenotypic spectrum of lupus erythematosus in Aicardi-Goutières syndrome. Arthritis Rheum. *62*, 1469–1477.
Remmers, E.F., Plenge, R.M., Lee, A.T., Graham, R.R., Hom, G., Behrens, T.W., de Bakker, P.I.W., Le, J.M., Lee, H.-S., Batliwalla, F., et al. (2007). STAT4 and the risk of rheumatoid arthritis and systemic lupus erythematosus. N. Engl. J. Med. *357*, 977–986.
Robinson, T., Kariuki, S.N., Franek, B.S., Kumabe, M., Kumar, A.A., Badaracco, M., Mikolaitis, R.A., Guerrero, G., Utset, T.O., Drevlow, B.E., et al. (2011). Autoimmune disease risk variant of IFIH1 is associated with increased sensitivity to IFN-α and serologic autoimmunity in lupus patients. J. Immunol. *187*, 1298–1303.
Rogers, N.J., Lees, M.J., Gabriel, L., Maniati, E., Rose, S.J., Potter, P.K., and Morley, B.J. (2009). A defect in Marco expression contributes to systemic lupus erythematosus development via failure to clear apoptotic cells. J. Immunol. *182*, 1982–1990.
Rosner, C., Kruse, P.H., Hermes, M., Otto, N., and Walter, L. (2011). Rhesus macaque inhibitory and activating KIR3D interact with Mamu-A-encoded ligands. J. Immunol. *186*, 2156–2163.
Sawalha, A.H., Webb, R., Han, S., Kelly, J.A., Kaufman, K.M., Kimberly, R.P., Alarcón-Riquelme, M.E., James, J.A., Vyse, T.J., Gilkeson, G.S., et al. (2008a). Common variants within MECP2 confer risk of systemic lupus erythematosus. PLoS ONE *3*, e1727.
Sawalha, A.H., Jeffries, M., Webb, R., Lu, Q., Gorelik, G., Ray, D., Osban, J., Knowlton, N., Johnson, K., and Richardson, B. (2008b). Defective

T-cell ERK signaling induces interferon-regulated gene expression and overexpression of methylation-sensitive genes similar to lupus patients. Genes Immun. *9*, 368–378.
Schorle, H., Holtschke, T., Hünig, T., Schimpl, A., and Horak, I. (1991). Development and function of T cells in mice rendered interleukin-2 deficient by gene targeting. Nature *352*, 621–624.
Schulte-Pelkum, J., Fritzler, M., and Mahler, M. (2009). Latest update on the Ro/SS-A autoantibody system. Autoimmun Rev *8*, 632–637.
Scott, R.S., McMahon, E.J., Pop, S.M., Reap, E.A., Caricchio, R., Cohen, P.L., Earp, H.S., and Matsushima, G.K. (2001). Phagocytosis and clearance of apoptotic cells is mediated by MER. Nature *411*, 207–211.
Segurado, O.G., Arnaiz-Villena, A., Iglesias-Casarrubios, P., Martinez-Laso, J., Vicario, J.L., Fontan, G., and Lopez-Trascasa, M. (1992). Combined total deficiency of C7 and C4B with systemic lupus erythematosus (SLE). Clinical & Experimental Immunology *87*, 410–414.
Seshasayee, D., Valdez, P., Yan, M., Dixit, V.M., Tumas, D., and Grewal, I.S. (2003). Loss of TACI causes fatal lymphoproliferation and autoimmunity, establishing TACI as an inhibitory BLyS receptor. Immunity *18*, 279–288.
Sheng, Y.-J., Gao, J.-P., Li, J., Han, J.-W., Xu, Q., Hu, W.-L., Pan, T.-M., Cheng, Y.-L., Yu, Z.-Y., Ni, C., et al. (2011). Follow-up study identifies two novel susceptibility loci PRKCB and 8p11.21 for systemic lupus erythematosus. Rheumatology (Oxford) *50*, 682–688.
Shultz, L.D., Schweitzer, P.A., Rajan, T.V., Yi, T., Ihle, J.N., Matthews, R.J., Thomas, M.L., and Beier, D.R. (1993). Mutations at the murine motheaten locus are within the hematopoietic cell protein-tyrosine phosphatase (Hcph) gene. Cell *73*, 1445–1454.
Sigurdsson, S., Nordmark, G., Göring, H.H.H., Lindroos, K., Wiman, A.-C., Sturfelt, G., Jönsen, A., Rantapää-Dahlqvist, S., Möller, B., Kere, J., et al. (2005). Polymorphisms in the tyrosine kinase 2 and interferon regulatory factor 5 genes are associated with systemic lupus erythematosus. Am. J. Hum. Genet. *76*, 528–537.
Sommers, C.L., Park, C.-S., Lee, J., Feng, C., Fuller, C.L., Grinberg, A., Hildebrand, J.A., Lacaná, E., Menon, R.K., Shores, E.W., et al. (2002). A LAT mutation that inhibits T cell development yet induces lymphoproliferation. Science *296*, 2040–2043.
Stern, R., Fu, S.M., Fotino, M., Agnello, V., and Kunkel, H.G. (1976). Hereditary C2 deficiency. Association with skin lesions resembling the discoid lesion of systemic lupus erythematosus. Arthritis & Rheumatism *19*, 517–522.
Sun, H., Lu, B., Li, R.Q., Flavell, R.A., and Taneja, R. (2001). Defective T cell activation and autoimmune disorder in Stra13-deficient mice. Nat. Immunol. *2*, 1040–1047.
Suzuki, H., Kündig, T.M., Furlonger, C., Wakeham, A., Timms, E., Matsuyama, T., Schmits, R., Simard, J.J., Ohashi, P.S., and Griesser, H. (1995). Deregulated T cell activation and autoimmunity in mice lacking interleukin-2 receptor beta. Science *268*, 1472–1476.
Takahashi, T., Tanaka, M., Brannan, C.I., Jenkins, N.A., Copeland, N.G., Suda, T., and Nagata, S. (1994). Generalized lymphoproliferative disease in mice, caused by a point mutation in the Fas ligand. Cell *76*, 969–976.
Trapp, R.G., Fletcher, M., Forristal, J., and West, C.D. (1987). C4 binding protein deficiency in a patient with atypical Behçet's disease. J Rheumatol *14*, 135–138.
Tsui, H.W., Siminovitch, K.A., de Souza, L., and Tsui, F.W. (1993). Motheaten and viable motheaten mice have mutations in the haematopoietic cell phosphatase gene. Nat. Genet. *4*, 124–129.
Tsukamoto, H., Horiuchi, T., Kokuba, H., Nagae, S., Nishizaka, H., Sawabe, T., Harashima, S., Himeji, D., Koyama, T., Otsuka, J., et al. (2005). Molecular analysis of a novel hereditary C3 deficiency with systemic lupus erythematosus. Biochemical and Biophysical Research Communications *330*, 298–304.

Vinuesa, C.G., Cook, M.C., Angelucci, C., Athanasopoulos, V., Rui, L., Hill, K.M., Yu, D., Domaschenz, H., Whittle, B., Lambe, T., et al. (2005). A RING-type ubiquitin ligase family member required to repress follicular helper T cells and autoimmunity. Nature *435*, 452–458.
Wandstrat, A.E., Nguyen, C., Limaye, N., Chan, A.Y., Subramanian, S., Tian, X.-H., Yim, Y.-S., Pertsemlidis, A., Garner, H.R., Morel, L., et al. (2004). Association of extensive polymorphisms in the SLAM/CD2 gene cluster with murine lupus. Immunity *21*, 769–780.
Wang, C., Ahlford, A., Laxman, N., Nordmark, G., Eloranta, M.-L., Gunnarsson, I., Svenungsson, E., Padyukov, L., Sturfelt, G., Jönsen, A., et al. (2013). Contribution of IKBKE and IFIH1 gene variants to SLE susceptibility. Genes Immun. *14*, 217–222.
Wang, J., Lo, J.C., Foster, A., Yu, P., Chen, H.M., Wang, Y., Tamada, K., Chen, L., and Fu, Y.X. (2001). The regulation of T cell homeostasis and autoimmunity by T cell-derived LIGHT. J. Clin. Invest. *108*, 1771–1780.
Wang, J.H., Avitahl, N., Cariappa, A., Friedrich, C., Ikeda, T., Renold, A., Andrikopoulos, K., Liang, L., Pillai, S., Morgan, B.A., et al. (1998). Aiolos regulates B cell activation and maturation to effector state. Immunity *9*, 543–553.
Watson, M.L., Rao, J.K., Gilkeson, G.S., Ruiz, P., Eicher, E.M., Pisetsky, D.S., Matsuzawa, A., Rochelle, J.M., and Seldin, M.F. (1992). Genetic analysis of MRL-lpr mice: relationship of the Fas apoptosis gene to disease manifestations and renal disease-modifying loci. J. Exp. Med. *176*, 1645–1656.
Wilkinson, R., Lyons, A.B., Roberts, D., Wong, M.-X., Bartley, P.A., and Jackson, D.E. (2002). Platelet endothelial cell adhesion molecule-1 (PECAM-1/CD31) acts as a regulator of B-cell development, B-cell antigen receptor (BCR)-mediated activation, and autoimmune disease. Blood *100*, 184–193.
Willerford, D.M., Chen, J., Ferry, J.A., Davidson, L., Ma, A., and Alt, F.W. (1995). Interleukin-2 receptor alpha chain regulates the size and content of the peripheral lymphoid compartment. Immunity *3*, 521–530.
Wu, J., Ekman, C., Jönsen, A., Sturfelt, G., Bengtsson, A.A., Gottsäter, A., Lindblad, B., Lindqvist, E., Saxne, T., and Dahlbäck, B. (2011). Increased plasma levels of the soluble Mer tyrosine kinase receptor in systemic lupus erythematosus relate to disease activity and nephritis. Arthritis Res. Ther. *13*, R62.
Wu, M.C., Huang, C.M., Tsai, J.J.P., Chen, H.Y., and Tsai, F.J. (2003). Polymorphisms of the interleukin-4 gene in chinese patients with systemic lupus erythematosus in Taiwan. Lupus *12*, 21–25.
Xue, D., Shi, H., Smith, J.D., Chen, X., Noe, D.A., Cedervall, T., Yang, D.D., Eynon, E., Brash, D.E., Kashgarian, M., et al. (2003). A lupus-like syndrome develops in mice lacking the Ro 60-kDa protein, a major lupus autoantigen. PNAS *100*, 7503–7508.
Yamaguchi, H., Takagi, J., Miyamae, T., Yokota, S., Fujimoto, T., Nakamura, S., Ohshima, S., Naka, T., and Nagata, S. (2008). Milk fat globule EGF factor 8 in the serum of human patients of systemic lupus erythematosus. J. Leukoc. Biol. *83*, 1300–1307.
Yang, W., Shen, N., Ye, D.-Q., Liu, Q., Zhang, Y., Qian, X.-X., Hirankarn, N., Ying, D., Pan, H.-F., Mok, C.C., et al. (2010). Genome-wide association study in Asian populations identifies variants in ETS1 and WDFY4 associated with systemic lupus erythematosus. PLoS Genet. *6*, e1000841.
Yang, Y., Chung, E.K., Wu, Y.L., Savelli, S.L., Nagaraja, H.N., Zhou, B., Hebert, M., Jones, K.N., Shu, Y., Kitzmiller, K., et al. (2007). Gene Copy-Number Variation and Associated Polymorphisms of Complement Component C4 in Human Systemic Lupus Erythematosus (SLE): Low Copy Number Is a Risk Factor for and High Copy Number Is a Protective Factor against SLE Susceptibility in European Americans. The American Journal of Human Genetics *80*, 1037–1054.
Yasutomo, K., Horiuchi, T., Kagami, S., Tsukamoto, H., Hashimura, C., Urushihara, M., and Kuroda, Y. (2001). Mutation of *DNASE1* in people

with systemic lupus erythematosus. Nature Genetics *28*, 313–314.
Yeh, C.-S., Chung, F.-Y., Chen, C.-J., Tsai, W.-J., Liu, H.-W., Wang, G.-J., and Lin, S.-R. (2008). PPARgamma-2 and BMPR2 genes were differentially expressed in peripheral blood of SLE patients with osteonecrosis. DNA Cell Biol. *27*, 623–628.
Yin, Z., Bahtiyar, G., Zhang, N., Liu, L., Zhu, P., Robert, M.E., McNiff, J., Madaio, M.P., and Craft, J. (2002). IL-10 regulates murine lupus. J. Immunol. *169*, 2148–2155.
Yu, H.-H., Liu, P.-H., Lin, Y.-C., Chen, W.J., Lee, J.-H., Wang, L.-C., Yang, Y.-H., and Chiang, B.-L. (2010). Interleukin 4 and STAT6 gene polymorphisms are associated with systemic lupus erythematosus in Chinese patients. Lupus *19*, 1219–1228.
Yu, Z.-Y., Lu, W.-S., Zuo, X., Hu, J., Yao, S., Li, Y., Han, J.-W., Sun, L.-D., Cheng, Y.-L., Xu, Q., et al. (2013). One novel susceptibility locus associate with systemic lupus erythematosus in Chinese Han population. Rheumatol. Int. *33*, 2079–2083.
Zikherman, J., Hermiston, M., Steiner, D., Hasegawa, K., Chan, A., and Weiss, A. (2009). PTPN22 deficiency cooperates with the CD45 E613R allele to break tolerance on a non-autoimmune background. J. Immunol. *182*, 4093–4106

Supplementary table S3 : Clinical data of the 117 patients

	Gene filtered (variants details in table I and II)	Gender	Age at onset	SLE Past Familial history	Mucocutaneous	Musculoskeletal	Cardiorespiratory	Lupus Nephritis	Neurological	Haematological	anti-dsDNA	anti-Sm	anti-phospholipid	ANA
Pt1	**C1QC hom**	Male	6	1	1	1	1	1	0	1	0	1	0	1
Pt2	**C1QC Het compounds,** CD19, CLEC16A,	Male	3	0	1	1	0	1	1	1	0	0	0	0
Pt3	**C1QA hom**	Male	4	0	1	1	0	0	1	1	0	0	1	1
Pt4	**C2 hom**	Female	11	0	1	0	0	1	0	1	0	1	0	1
Pt5	**C2 hom**	Female	4	0	1	1	0	0	0	0	0	0	0	0
Pt6	**C2 hom**	Female	15	0	1	0	0	0	0	1	1	1	1	1
Pt7	**DNASE1L3 hom**	Male	4	0	1	1	0	1	0	1	1	0	0	1
Pt8	**IKZF1,** LYN	Female	14	1	1	0	1	1	0	1	1	1	0	1
Pt9	**TREX1**	Female	7	0	1	1	0	1	0	1	1	0	0	1
Pt10	**ACP5,** RASGRP3	Male	4	0	1	1	0	1	0	1	1	0	1	1
Pt11	**RNASEH2C**	Female	8	0	1	1	0	1	0	1	1	0	0	1
Pt12	**C9 het**	Female	11	0	1	1	1	1	1	1	1	0	1	1
Pt13	**C8B het**	Male	11	0	1	1	0	0	1	1	1	0	1	1
Pt14	**C9 het**	Male	7	0	0	0	0	1	0	1	1	0	0	1
Pt15	**C3 het**	Male	2	0	0	1	0	1	0	1	0	0	0	0
Pt16	IRF7	Female	8	0	1	1	0	1	1	1	0	0	0	1
Pt17	CLEC16A, NFE2L2, TBX	Female	10	0	1	1	1	1	0	0	1	0	0	1
Pt18	TRAF3IP2,	Female	10	0	1	1	1	1	1	1	1	0	0	1
Pt19	PDHX	Female	7	1	1	1	0	0	0	1	0	0	0	1
Pt20	JAZF1	Female	9	1	1	0	0	0	0	0	1	0	0	1
Pt21	PHRF1	Female	8	0	0	0	0	1	0	1	1	0	0	1
Pt22	CD19	Female	10	0	1	1	0	0	0	1	1	0	0	1
Pt23	CBLB, TMEM39A	Male	16	1	0	1	1	0	0	1	0	1	0	1

Pt24	TNXB	Female	2.5	0	0	1	0	1	0	1	0	0	1	1
Pt25	PTPN22	Female	9	1	1	0	0	1	0	1	1	0	0	1
Pt26	PDHX	Male	10	1	0	0	0	0	0	1	0	1	0	1
Pt27	WDFY4	Female	10	0	0	0	0	1	0	1	1		0	1
Pt28	CYBB	Male	10	0	1	0	0	0	0	0	0	1	0	1
Pt29	MECP2	Male	14	0	1	1	0	0	1	0	1	1	1	1
Pt30		Female	11	0	1	1	0	0	1	1	0	0	0	1
Pt31		Female	11	1	1	1	1	1	0	1	1	0	0	1
Pt32		Female	8	0	1	1	1	0	1	1	1	0	1	1
Pt33		Male	5	0	1	1	0	1	0	1	0	0	0	0
Pt34		Female	10	0	0	1	0	1	0	1	1	0	0	1
Pt35		Female	8	0	1	1	0	1	0	1	0	1	0	1
Pt36		Male	9	0	1	1	0	0	0	1	0	0	1	1
Pt37		Female	8	1	1	1	1	1	0	1	0	0	0	1
Pt38		Female	9	0	0	1	0	1	1	1	0	0	1	1
Pt39		Female	2	0	1	1	0	0	0	1	1	0	0	1
Pt40		Female	11	0	1	1	1	0	1	1	0	0	0	1
Pt41		Female	9	0	1	1	0	1	0	1	0	0	0	0
Pt42		Female	3	0	1	1	0	1	1	1	0	0	0	1
Pt43		Male	8	0	1	0	0	1	0	1	0	0	0	0
Pt44		Male	10	1	1	1	0	1	0	1	1	0	0	1
Pt45		Female	7	1	1	0	0	0	0	1	1	0	1	1
Pt46		Female	8	0	1	1	1	1	1	1	1	0	0	1
Pt47		Male	2	1	1	1	0	0	0	1	1	0	0	1
Pt48		Female	10	1	1	1	0	1	1	1	1	0	0	1
Pt49		Female	5	0	1	1	1	1	0	1	1	0	0	1
Pt50		Female	4	0	1	1	0	1	0	1	0	0	0	1
Pt51		Female	10	0	1	1	1	1	0	1	1	0	0	1

Pt52		Female	10	0	1	1	0	1	1	1	1	0	1	1
Pt53		Female	11	0	1	1	0	1	0	1	1	0	0	1
Pt54		Female	10	0	1	1	1	1	0	1	0	0	0	0
Pt55		Female	10	0	1	0	0	1	0	1	1	0	0	0
Pt56		Female	8	1	1	1	0	1	1	1	1	0	0	1
Pt57		Female	6	0	1	1	0	1	0	1	1	0	0	1
Pt58		Female	8	0	1	1	0	1	0	1	0	0	1	1
Pt59		Female	4	1	1	1	0	1	0	1	1	0	0	1
Pt60		Female	11	0	1	1	0	0	0	1	1	0	0	1
Pt61		Female	10	0	1	1	1	1	0	1	0	0	0	0
Pt62		Female	7	0	1	0	1	1	0	1	1	0	1	1
Pt63		Female	9	0	1	0	0	1	0	1	0	0	0	1
Pt64		Female	10	1	1	1	0	1	0	1	0	0	0	0
Pt65		Male	11	0	1	1	0	0	1	1	0	0	0	1
Pt66		Female	4	1	1	1	0	1	0	1	1	0	0	1
Pt67		Female	16	0	1	0	0	0	1	1	1	0	0	1
Pt68		Female	3	0	1	0	1	1	0	0	0	0	0	0
Pt69		Male	10	0	1	0	0	0	0	0	0	0	0	0
Pt70		Female	13	1	0	1	0	0	0	1	1	0	0	1
Pt71		Female	4	0	0	1	0	0	0	1	0	1	0	1
Pt72		Female	10	1	1	0	0	0	0	0	1	1	1	1
Pt73		Female	11	0	0	0	1	1	0	0	1	0	1	1
Pt74		Male	6	0	0	1	0	0	0	0	1	0	1	1
Pt75		Female	8	0	1	0	0	0	0	0	1	0	1	1
Pt76		Female	8	0	1	1	0	0	0	1	1	0	0	1
Pt77		Female	12	0	1	0	0	1	0	1	1	0	0	1
Pt78		Female	11	0	1	1	1	0	0	1	1	0	0	1
Pt79		Female	11	0	1	1	1	0	0	0	1	0	0	1

Pt80		Female	11	0	1	1	0	0	0	0	1	1	1	1
Pt81		Female	11	0	1	0	0	0	0	0	1	0	0	1
Pt82		Female	13	0	0	1	0	1	0	0	1	0	0	1
Pt83		Female	13	0	1	1	0	0	0	1	1	0	0	1
Pt84		Female	10	0	1	1	1	1	0	0	1	0	0	1
Pt85		Female	13	0	0	1	0	0	1	1	1	0	0	1
Pt86		Female	10	0	0	1	0	0	0	1	1	0	0	1
Pt87		Female	9	0	1	1	0	0	0	1	1	0	0	1
Pt88		Female	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Pt89		Male	16	1	0	1	0	1	0	0	1	0	1	1
Pt90		Female	11	0	1	0	0	0	0	1	1	1	1	1
Pt91		Female	9	0	1	1	0	1	0	1	0	0	0	1
Pt92		Female	12	0	1	0	1	1	0	1	1	0	1	1
Pt93		Male	11	0	0	0	0	0	0	0	1	1	0	1
Pt94		Female	5	0	0	0	0	1	0	0	0	0	1	1
Pt95		female	14	0	1	1	0	0	0	1	1	0	1	1
Pt96		Male	6	0	1	1	0	1	0	1	1	1	0	1
Pt97		Female	6	0	0	0	0	1	1	1	0	0	1	1
Pt98		Female	12	0	1	1	1	0	1	1	1	1	NA	1
Pt99		Female	16	0	0	1	1	0	0	1	1	0	1	1
Pt100		Female	14	0	0	0	0	0	0	1	0	0	1	1
Pt101		Female	16	0	1	1	1	1	0	1	1	0	0	1
Pt102		Female	4	1	1	0	0	1	0	1	1	0	1	1
Pt103		Male	8	0	1	0	0	1	0	1	1	0	0	1
Pt104		Female	6	1	1	0	0	0	0	0	0	0	0	1
Pt105		Female	14	0	1	0	0	1	0	1	1	0	0	1
Pt106		Female	13	0	0	0	0	1	1	1	1	0	0	0
Pt107		Female	16	0	1	1	0	1	0	1	1	0	1	1

Pt108		Female	10	0	1	1	0	0	0	0	1	0	0	1
Pt109		Female	13	0	1	0	0	1	0	1	1	0	0	1
Pt110		Female	13	1	0	0	0	0	1	1	0	0	1	0
Pt111		Female	8	0	1	1	0	0	0	1	1	0	0	1
Pt112		Female	10	0	1	1	0	1	1	0	0	0	1	1
Pt113		Male	8	0	1	1	0	1	1	1	0	0	0	1
Pt114		Female	9	0	1	1	0	1	1	1	0	0	0	1
Pt115		Female	11	0	1	1	0	1	0	1	1	0	1	0
Pt116		Female	13	0	1	1	0	1	0	0	1	1	0	1
Pt117		Female	14	0	0	1	0	1	0	1	1	0	1	1

Supplemental Table S4: Targeted versus Exome sequencing coverage and depth of the panel geneset

A/	Panel Lupus					1000g data-Bed				
	coverage	mean(%)	median(%)	min(%)	max(%)	coverage	mean	median	min	max
	1x	1	1	0,9995	1	1x	0,9339	0,9466	0,8353	0,9683
	5x	0,9999	0,9999	0,9991	1	5x	0,8924	0,9055	0,7498	0,9577
	10x	0,9998	0,9999	0,9985	1	10x	0,8592	0,8696	0,7251	0,9471
	20x	0,9996	0,9998	0,9959	1	20x	0,7905	0,7946	0,5403	0,9295

B/	FrEx-Bed				
	coverage	Mean	Median	Min	Max
	1x	0,9986	0,9989	0,9824	0,9999
	5x	0,9914	0,9933	0,9332	0,9991
	10x	0,9764	0,9797	0,8872	0,9970
	20x	0,9243	0,9265	0,8083	0,9920

Supplemental Table S5: Coverage and depth of rare SNP variants filtered in the panel in 1000g and Frex control database

				1000g covered individuals (n=791)		FREX covered individuals (n=574)	
Mutations				20X	30X	20X	30X
1	22974028	C1QC	c.490G>A	791	774	574	574
1	114401155	PTPN22	c.314C>T	606	407	228	31
2	33749115	RASGRP3	c.686C>G	669	658	574	574
3	105470408	CBLB	c.621T>G	768	752	574	573
5	39288980	C9	c.1490C>T	791	788	569	561
6	111887757	TRAF3IP2	c.1366G>A	791	791	574	574
7	50444429	IKZF1	c.199A>T	674	659	574	574
8	56866526	LYN	c.773T>C	790	786	564	406
10	49933991	WDFY4	c657G>T	328	299	574	567
11	605628	PHRF1	c.1355C>T	674	663	574	574
11	613556	IRF7	c.926A>T	487	291	569	479
11	34952961	PDHX	c.171T>G	732	605	574	571
11	65487277	RNASEH2C	c.472C>G	781	726	574	574
16	11063059	CLEC16A	c.385C>G	625	525	565	515
16	28948790	CD19	c.1318G>A	785	748	574	574
19	6693047	C3	c.3278T>G	779	708	574	574
19	11688054	ACP5	c.79C>A	773	686	574	574
X	37663334	CYBB	c.1102G>T	791	768	574	550
X	153296464	MECP2	c851C>T	782	715	574	574
			Mean of individuals	706.15	649.94	554.26	525.94
			% of individuals	0.89	0.82	0.96	0.91

Supplemental Table S6: Mean of filtered variants from the jSLE panel and UK healthy controls (adults, n=74). Processing of samples and bioinformatic filtering were identical within the two groups. Rare variants, defined here as a frequency on the Exome Aggregation Consortium (ExAC) database of <0.1% for SNPs and <1% for indels, were selected for further analysis. CADD>15 was considered for the pathogenic score.

		SNVs	
Filtering		**JSLE**	**Controls**
All	**KLC**	69.39316239	65.43835616
	PLC	322.2478632	314.1780822
ExAc	**KLC**	0.341880342	0.383561644
	PLC	2.564102564	7.835616438
CADD	**KLC**	0.057591623	0.041884817
	PLC	0.35078534	0.261780105

Supplemental Table S7: SKAT-O test for genes filtered in by the algorithm. Frequency of 0.1% and 1% in Exac were tested.

gene	chr	pos	ExAC_Fq	pSKAT-O
ACP5	19	11687884	0.10%	0.1166
ACP5	19	11687884	1%	0.1166
C1QC	1	22973915	0.10%	0.0057
C1QC	1	22973915	1%	0.0057
C2	6	31896638	0.10%	0.6901
C2	6	31896638	1%	0.6901
C3	19	6680256	0.10%	0.0667
C3	19	6680256	1%	0.0607
C8B	1	57409469	0.10%	1.0000
C8B	1	57409469	1%	1.0000
C9	5	39288980	0.10%	0.1171
C9	5	39288980	1%	0.2263
CBLB	3	105378034	0.10%	0.5063
CBLB	3	105378034	1%	0.5063
CD19	16	28948667	0.10%	0.0109
CD19	16	28948667	1%	0.0109
CLEC16A	16	11051758	0.10%	0.1743
CLEC16A	16	11051758	1%	0.4079
CYBB	X	37658219	0.10%	0.6885
CYBB	X	37658219	1%	0.6885
DNASE1L3	3	58179089	0.10%	0.6531
DNASE1L3	3	58179089	1%	0.6531
IKZF1	7	50444387	0.10%	0.1164
IKZF1	7	50444387	1%	0.1164
IRF7	11	613097	0.10%	0.4161
IRF7	11	613097	1%	0.4161
JAZF1	7	27872484	0.10%	0.5979
JAZF1	7	27872484	1%	0.5979
LYN	8	56863106	0.10%	0.2587
LYN	8	56863106	1%	0.2587
MECP2	X	153296464	0.10%	0.0256
MECP2	X	153296464	1%	0.0256
NFE2L2	2	178096309	0.10%	0.5976
NFE2L2	2	178096309	1%	0.0732
PDHX	11	34938246	0.10%	1.0000
PDHX	11	34938246	1%	1.0000
PHRF1	11	591436	0.10%	0.0527
PHRF1	11	587372	1%	0.1683
PTPN22	1	114372222	0.10%	0.2984
PTPN22	1	114372222	1%	0.2984
RASGRP3	2	33740227	0.10%	0.1167
RASGRP3	2	33740227	1%	0.1167
RNASEH2C	11	65487277	0.10%	0.0252
RNASEH2C	11	65487277	1%	0.0252
TMEM39A	3	119165965	0.10%	0.5948
TMEM39A	3	119165965	1%	0.5948
TNXB	6	32009134	0.10%	0.1259
TNXB	6	32009134	1%	0.1060
TRAF3IP2	6	111887757	0.10%	0.1163
TRAF3IP2	6	111887757	1%	0.2599
TREX1	3	48508607	0.10%	0.5967
TREX1	3	48508607	1%	0.5967

WDFY4	10	49933991	0.10%	0.2680
WDFY4			1%	0.6425

Figure 1

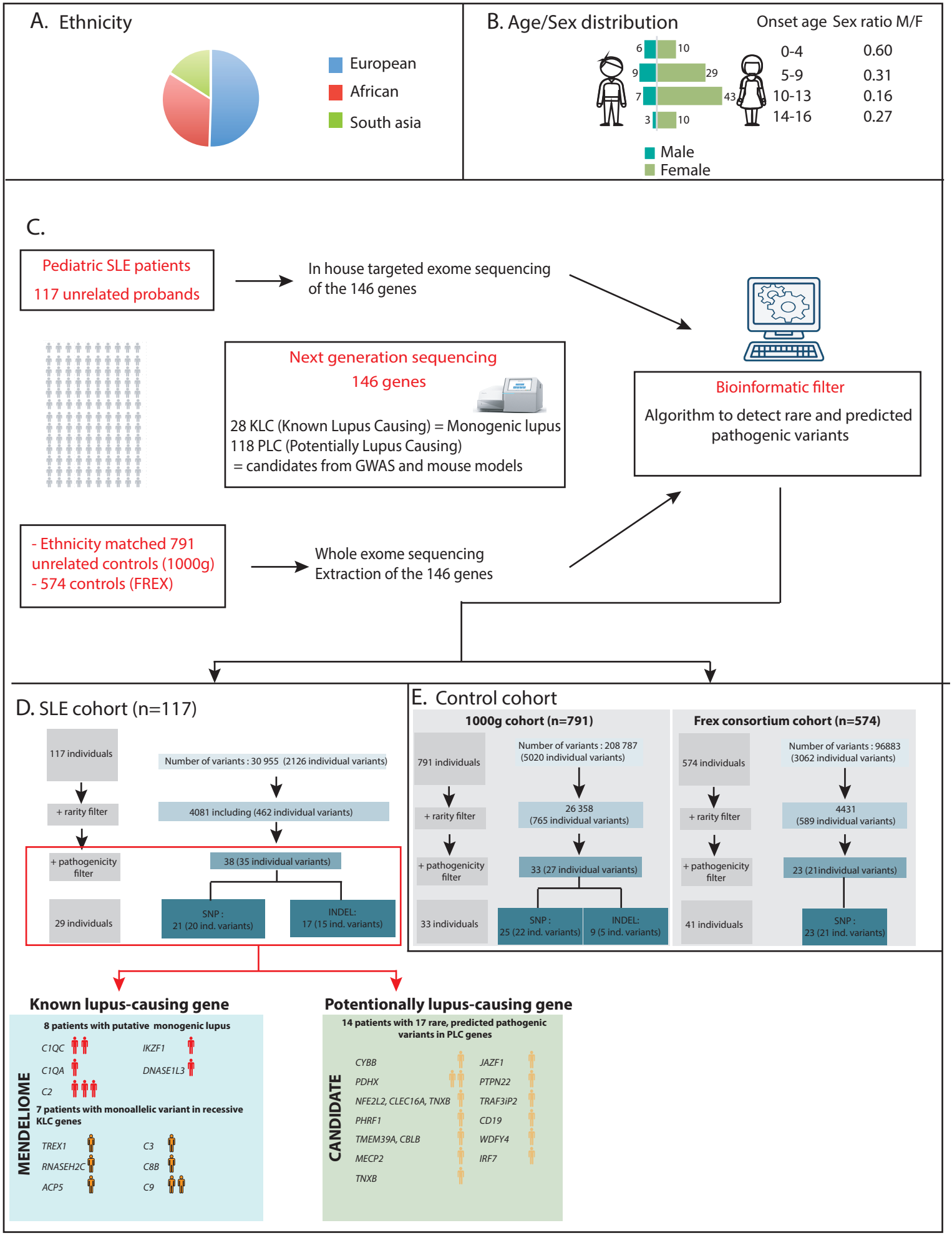


Figure 1

Figure 2

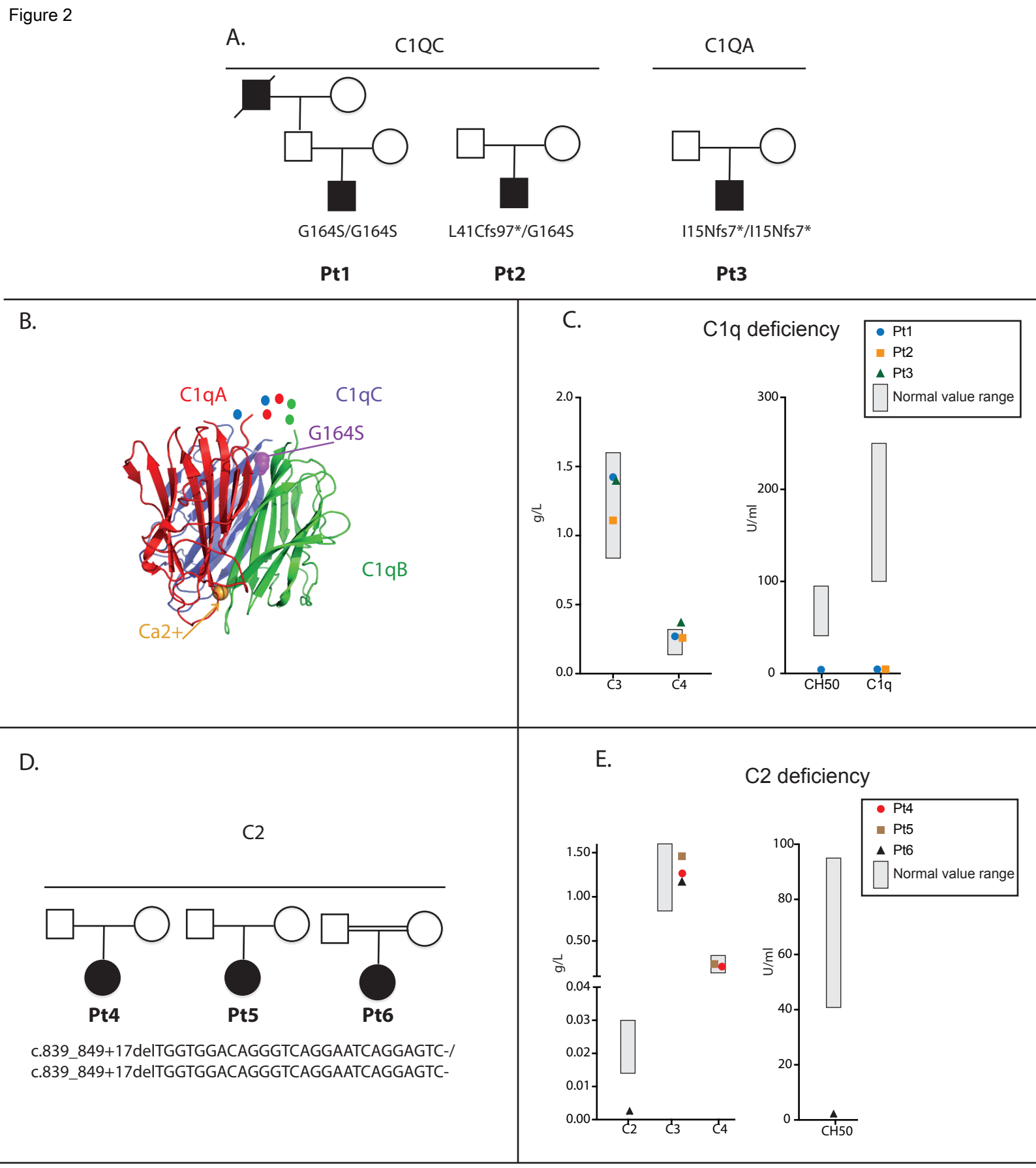


Figure 2

Figure 3

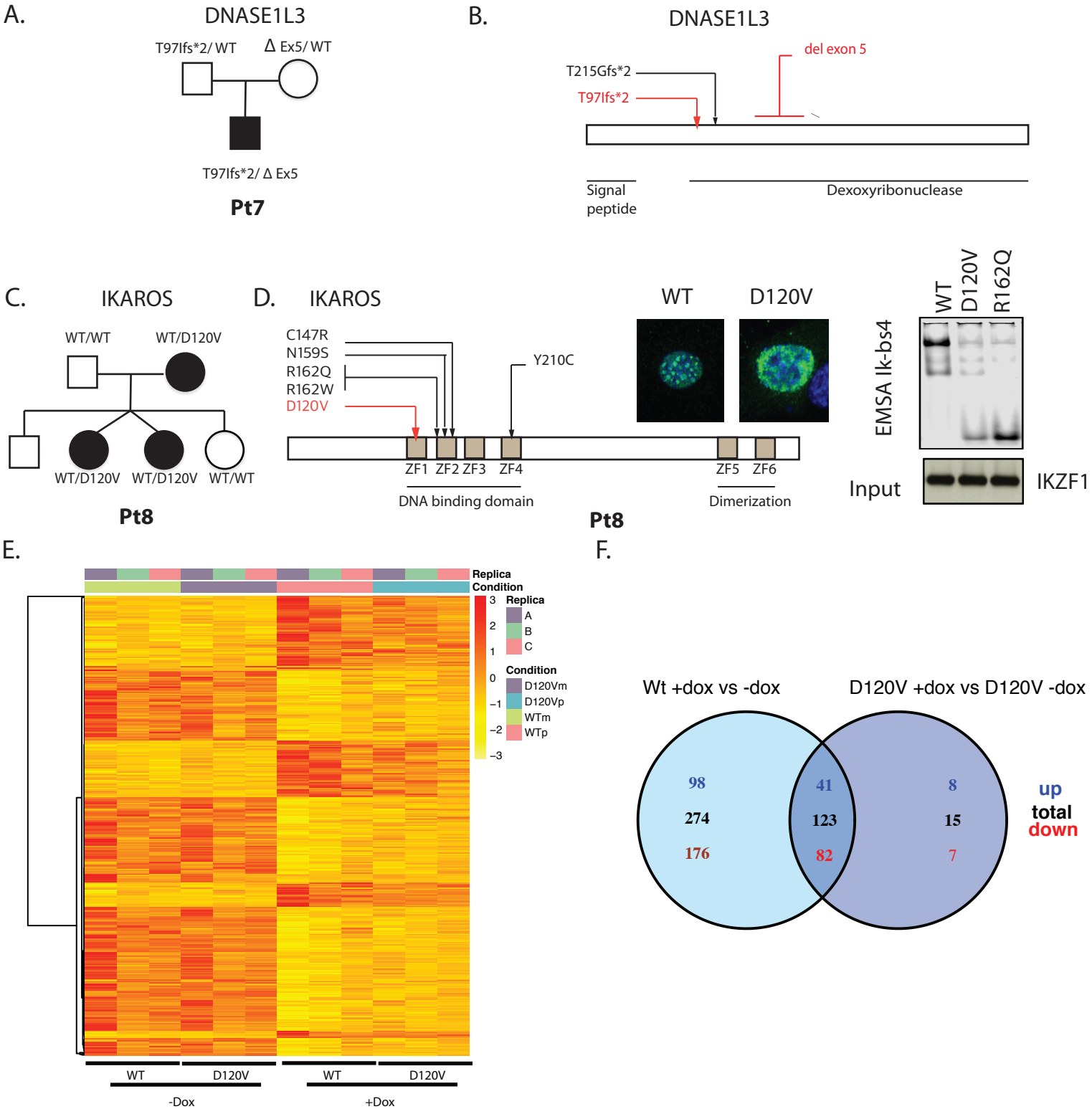
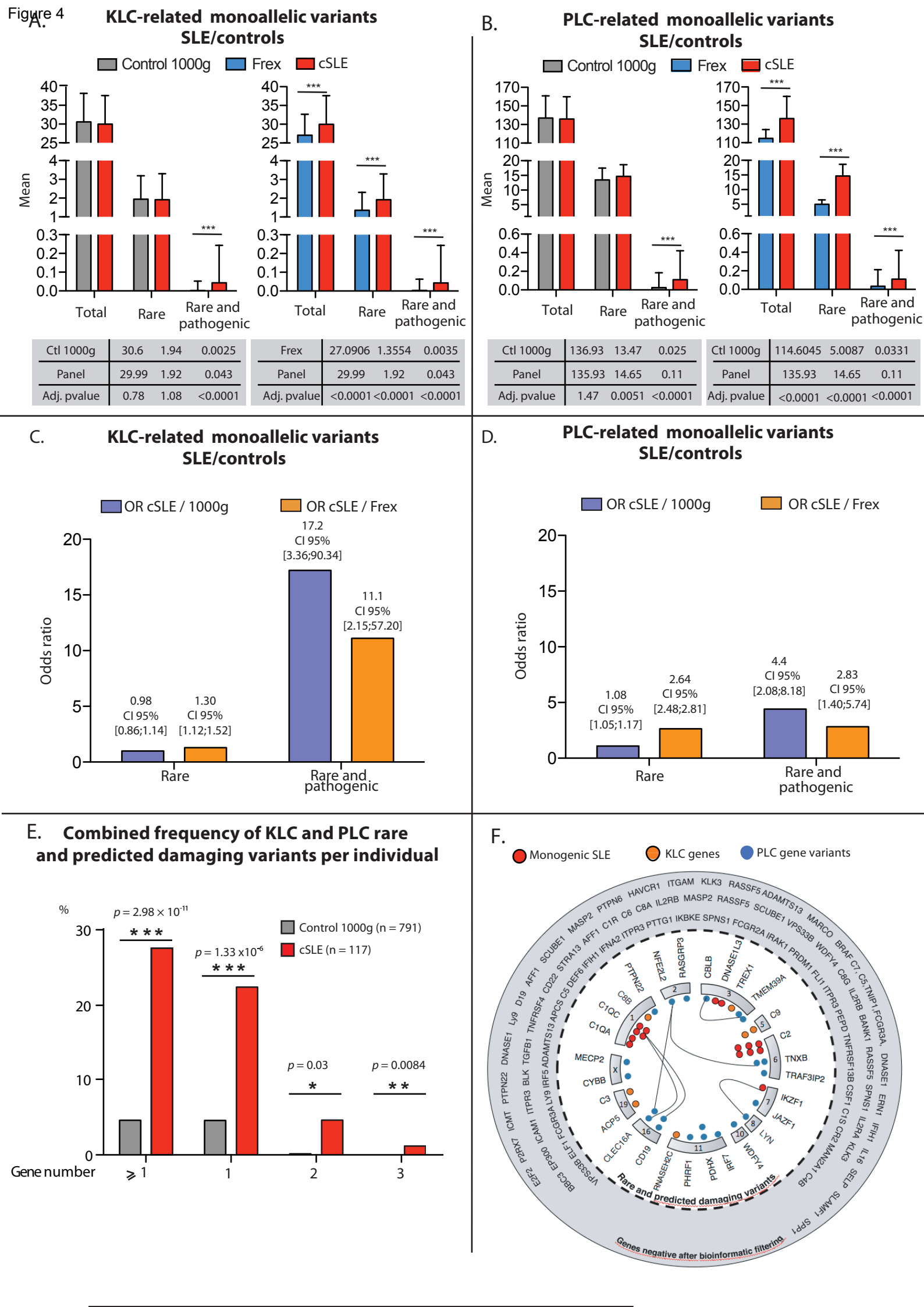


Figure 3

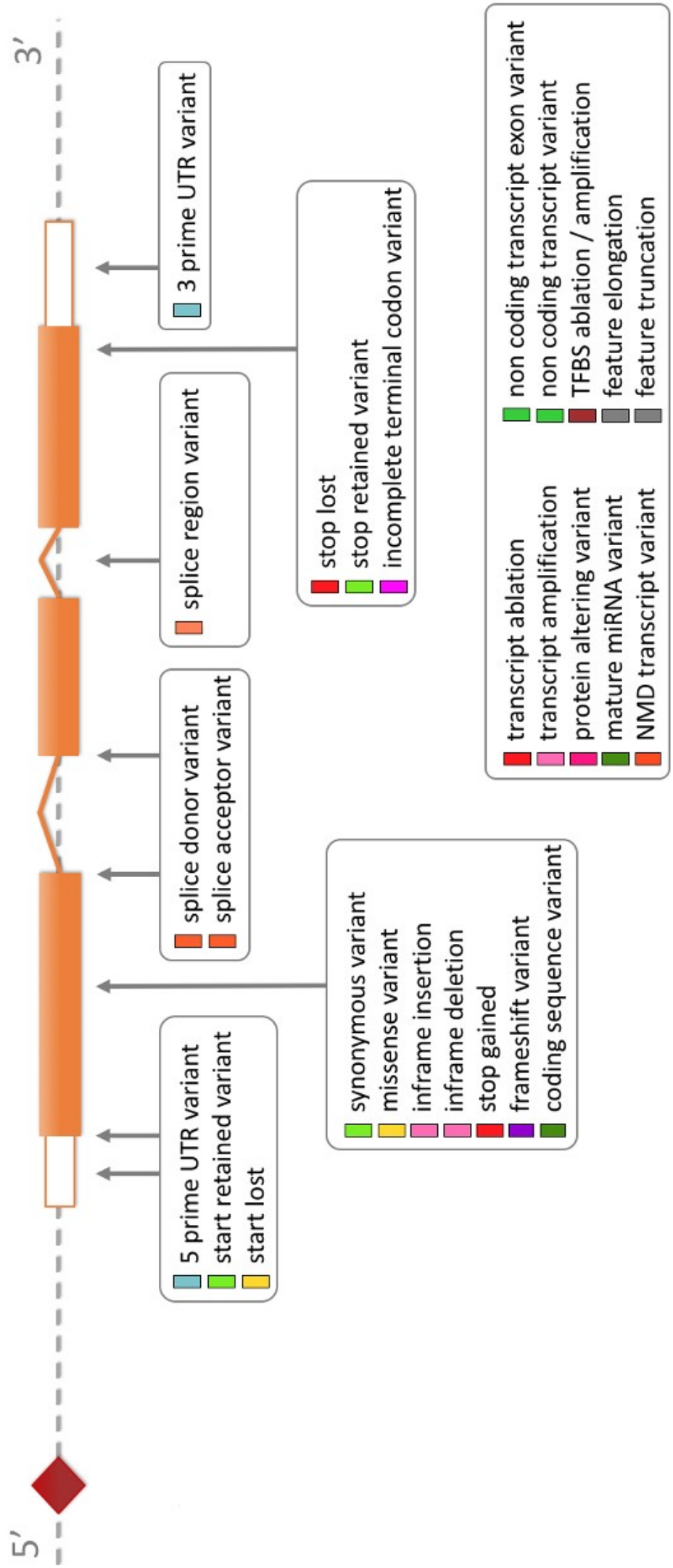
Figure 4



A.								
Ctl 1000g	30.6	1.94	0.0025	Frex	27.0906	1.3554	0.0035
Panel	29.99	1.92	0.043	Panel	29.99	1.92	0.043
Adj. pvalue	0.78	1.08	<0.0001	Adj. pvalue	<0.0001	<0.0001	<0.0001

B.								
Ctl 1000g	136.93	13.47	0.025	Ctl 1000g	114.6045	5.0087	0.0331
Panel	135.93	14.65	0.11	Panel	135.93	14.65	0.11
Adj. pvalue	1.47	0.0051	<0.0001	Adj. pvalue	<0.0001	<0.0001	<0.0001

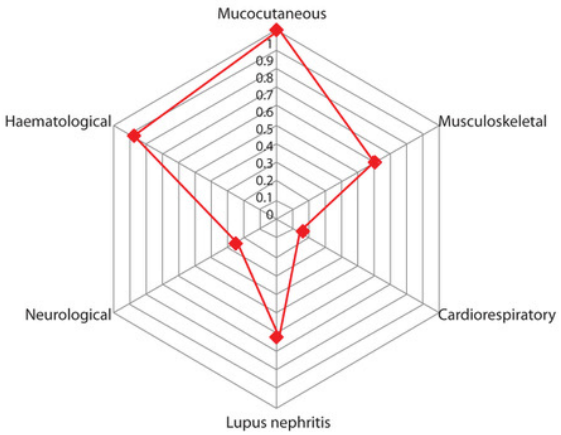
Supplemental Figure 1



Supplemental Figure 2

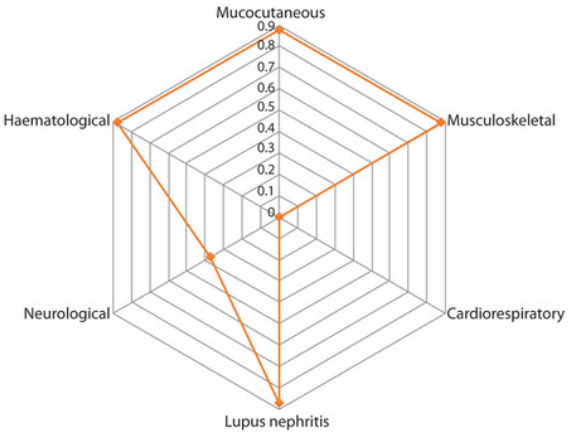
A. Mendelian Lupus (n=8)

sex ratio = 1
Age at onset = 7
(SD +/- 4.4)



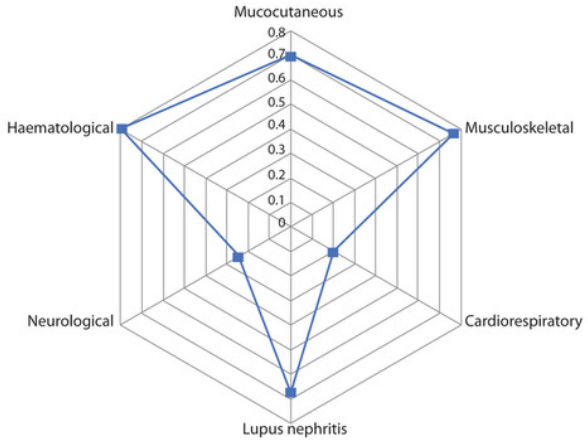
B. Patients carrying variants in KLC genes (n=7)

sex ratio = 1.33
Age at onset = 7
(SD +/- 3)



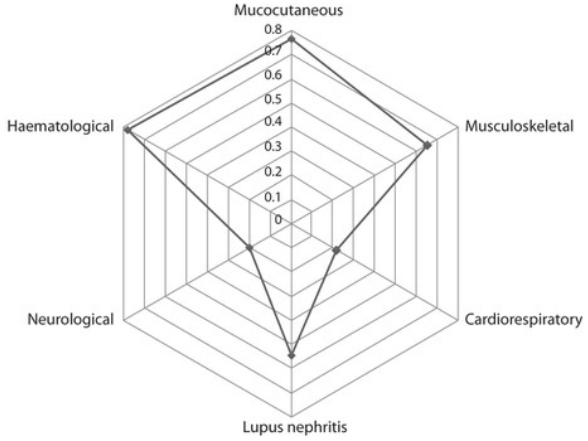
C. Patients carrying variants in PLC genes (n=14)

sex ratio = 0.4
Age at onset = 10
(SD +/- 3



D. Patients without filtered variants (n=88)

sex ratio = 0.17
Age at onset = 9
(SD +/- 3.32



Supplemental Figure 3