SUPPLEMENT

Supplemental Materials and Methods

Genotyping of *FCGR3A* single nucleotide polymorphisms

FCGR3A SNPs were genotyped by Pyrosequencing assays. Two *FCGR3A* gene-specific fragments containing rs10127939 (SNP 230T>A>G) or rs396991 (SNP 559T>G) were generated with gene-specific PCR. The sense primer (5'-TTC AAG AAA AGG AAA TTG GTG-3') and the antisense primer (5'-CCC AAG ACC TAC TTA GAG CTA-3') were used to amplify a *FCGR3A*-specific genomic DNA fragment, which was used as the template for nested PCR with the sense primer (5'-TAC AGG GTG CTC GAG AAG GA-3') and the biotin-labeled antisense primer (5'- CGA GGC CTG GCT TGA GAT-3') to generate a DNA fragment (114 bps) containing the *FCGR3A* rs10127939 (SNP 230T>A>G) for Pyrosequencing assay. The genotype frequency of rs10127939 is shown in Supplemental Table 1. To determine the *FCGR3A* rs396991 genotypes, the sense primer (5'-CTG GTG TTT ACA TTG AGT TCT C-3') and the antisense primer (5'-CTG ATT CTG GAG GCT GGT TCT ACA-3') were used to amplify a *FCGR3A*-specific genomic DNA fragment, which was used as the template in a nest PCR with the biotin-labeled sense primer (5'-CAA AGG CAG GAA GTA TTT TCA T -3') and the antisense primer (5'-ACC TTG AGT GAT GGT GAT GTT CA -3') to generate a DNA fragment (145 bps) containing the *FCGR3A* SNP 559 for the Pyrosequencing assay. Pyrosequencing assays were performed on a PSQ-HS-96A Pyrosequencer (Qiagen) according to the vendor's instruction with the sense primer (5'-AGT GGT TTC ACA ATG AGA G-3') for rs10127939 and the antisense primer (5'-GAC ACA TTT TTA CTC CCA

A-3') rs396991. Pyrosequencing data were analyzed for SNP genotyping calls and relative quantities of SNP alleles.

Assessment of copy number variation

The copy number variation (CNV) status of the *FCGR3A* gene was also determined by Pyrosequencing methodology. Two Pyrosequencing assays were used to compare relative quantities of *FCGR2B* vs *FCGR2C* and *FCGR3A* vs *FCGR3B* (abbreviated as assays 2BC and 3AB, respectively). In each assay, a PCR primer pair was designed to amplify regions in two genes that are identical except for a paralogous sequence variant (PSV). The two "alleles" of the PSV serve as markers for the two genes. For each assay, PCR was carried out to amplify each site prior to Pyrosequencing using the following forward primers and biotinylated reverse primers: 2BC (F: 5'-AGT GAG TCA CTC CAC CTC TCT GTG-3'; R: 5'-TGT GTG CTG TTA CTG CCT ACC AG-3'), 3AB (F: 5'-TCC ACC TGG GTA CCA AGT CTC T-3'; R: 5'-TTG AGG GTC CTT TCT CCA TTT AA-3'). PCR products were purified and Pyrosequenced as described earlier for the rs396991 and rs10127939 genotyping reactions. The following sequencing primers were used for pyrosequencing: 2BC (5'-GGA AAA TGG GGA CAC TA-3'), 3AB (5'-TCT CTG TGA AGA CAA ACA TT-3'). Pyrosequencing data were analyzed using the absolute quantification method to determine the relative quantity of each PSV and, therefore, each gene. Each Pyrosequencing assay was performed in duplicate and the peak intensity was averaged. Donors for which one of the replicate reactions failed were excluded.

After Pyrosequencing, the relative gene quantities from 2BC and 3AB for each donor were first translated from percentages into their closest whole-number ratio (0% = 0: X, 25% = 1:3, 33% = 1:2, 40% = 2:3, 50% = 1:1, 60% = 3:2, 66% = 2:1, 75% = 3:1, 100% = X: 0). Donors with 1:1 ratios for both assays were classified as having no CNV. CNV in the *FCGR* gene cluster has been demonstrated to occur between two regions of paralogy [41]. Additionally, CNV of *FCGR2A* and *FCGR2B* has not been reported. Therefore, deletions and duplications of *FCGR3A* were expected to occur with deletions and duplications of *FCGR2C*, similar to the well described deletions and duplications of the *FCGR2C-FCGR3B* unit [41]. *FCGR3A* and *FCGR3B* CNVs were distinguished via the ratios for the 2BC and 3AB assays. Because the quantities of *FCGR2C* and *FCGR3B* are denominators in their respective assays, donors with deletions and duplications of the *FCGR2C-FCGR3B* unit have equivalent ratios for the 2BC and 3AB assays. However, since the quantity of *FCGR3A* is the numerator in the 3AB assay ratio, donors with deletion/duplication of the *FCGR3A-FCGR2C* unit have inversed ratios for 2BC and 3AB. Since *FCGR2B* does not vary in copy number, we were able to calculate *FCGR2C* gene copy number from the 2BC ratios. Knowing *FCGR2C* copy number, and which *FCGR3* gene was deleted or duplicated with *FCGR2C* allowed us to assign copy number to that *FCGR3* gene and the 3AB assay ratio allowed us to calculate the gene copy number of the other FCGR3 gene. The assay-ratio pairs observed for *FCGR3A* CNV were as follows: 2BC=2:1, 3AB=1:2 for single deletions of the 3A-2C; 2BC=2:3, 3AB=3:2 for single duplications of 3A-2C; and 2BC=1:2, 3AB=2:1 for double duplications of 3A-2C. We did not observe any 3A-null individuals, nor did we observe any individuals with 3A CNV in the absence of 2C CNV. CNV calls were used with

relative quantitation data of rs396991 and rs10127939 alleles to make genotyping calls for 3A CNV donors.

Supplemental Table 1. Genotype frequency of FcγRIIIa-66L/H/R allele in Controls, SLE participants, and SLE participants stratified by nephritis phenotype

		FcγRIIIa-66L/H/R genotype, no. (%) of subjects						
		66L/L	66L/R	66L/H	66R/R	66R/H	66H/H	66L/R/H
EA	ALL	1585(78.5%)	159(7.9%)	249(12.3%)	5(0.2%)	9(0.4%)	11(0.5%)	1(0.0%)
	CNTL	929(78.4%)	98(8.3%)	142(12.0%)	2(0.2%)	6(0.5%)	7(0.6%)	1(0.1%)
	SLE all	656(78.7%)	61(7.3%)	107(12.8%)	3(0.4%)	3(0.4%)	4(0.5%)	0(0.0%)
	SLE neph (-)	508(78.6%)	52(8.0%)	77(11.9%)	3(0.5%)	2(0.3%)	4(0.6%)	0(0.0%)
	SLE neph (+)	148(78.7%)	9(4.8%)	30(16.0%)	0(0.0%)	1(0.5%)	0(0.0%)	0(0.0%)
AA	ALL	1518(94.8%)	26(1.6%)	52(3.2%)	1(0.1%)	1(0.1%)	3(0.2%)	0(0.0%)
	CNTL	907(95.2%)	11(1.2%)	33(3.5%)	1(0.1%)	0(0.0%)	1(0.1%)	0(0.0%)
	SLE all	611(94.3%)	15(2.3%)	19(2.9%)	0(0.0%)	1(0.2%)	2(0.3%)	0(0.0%)
	SLE neph (-)	314(92.1%)	13(3.8%)	12(3.5%)	0(0.0%)	1(0.3%)	1(0.3%)	0(0.0%)
	SLE neph (+)	297(96.7%)	2(0.7%)	7(2.3%)	0(0.0%)	0(0.0%)	1(0.3%)	0(0.0%)

EA: European Americans; AA: African Americans; SLE neph (-): SLE patients negative for nephritis; SLE neph (+): SLE positive for nephritis.

Supplemental Table 2. The frequencies of *FCGR3A* gene copy number and the alleles of the 559 and 230 SNPs

		FCGR3A Copy Number			Alleles of 559		Alleles of 230		
		<2	2	>2	G	T	T	G	A
European Americans	All	1.09%	94.75%	4.16%	31.72%	68.28%	88.79%	4.31%	6.90%
	CNTL	1.35%	94.60%	4.05%	32.42%	67.58%	88.72%	4.49%	6.78%
	SLE	0.72%	94.96%	4.32%	30.72%	69.28%	88.88%	4.06%	7.06%
African Americans	All	1.25%	95.75%	3.00%	33.84%	66.16%	97.31%	0.90%	1.79%
	CNTL	1.26%	95.59%	3.15%	34.06%	65.94%	97.51%	0.67%	1.82%
	SLE	1.23%	95.99%	2.78%	33.51%	66.49%	97.02%	1.22%	1.76%