Whole exome sequencing identifies variants in families with macular degeneration

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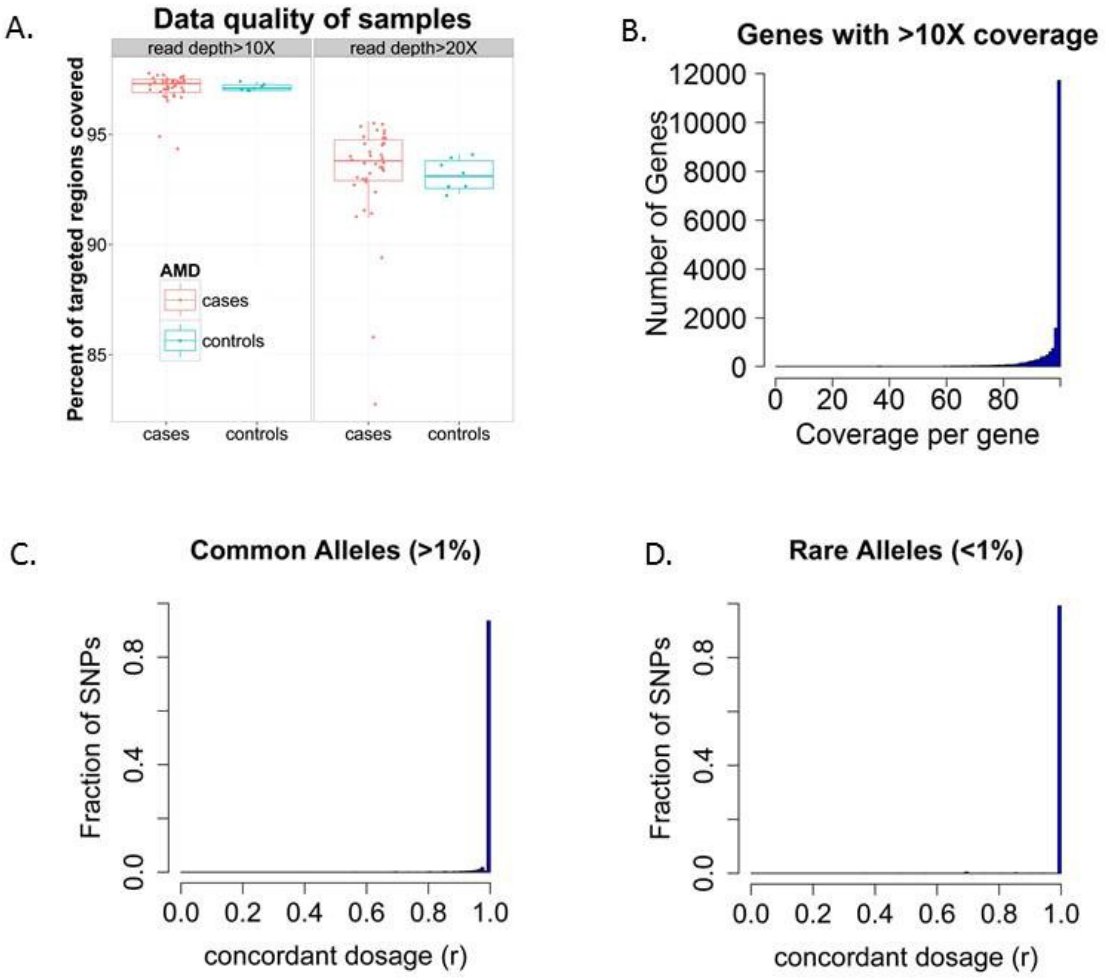
Supplementary Table S1-Other potential pathogenic variants suggested by XBrowse.

FAM	Genes	Location	REF	ALT	RSID	Amino Acid	Allele Frequencies in ESP	LOD score	Polyphen2	SIFT
Variants predicted to be both probably damaging by PolyPhen2 and deleterious by SIFT										
I	MUC4	chr3:195490456	T	G	.	N4573H	0	-0.90	prob.dam.	deleterious
IV	CCT4	chr2:62106083	A	C	.	L148W	0.0002	-5.40	prob.dam.	deleterious
IV	CNTN3	chr3:74316451	G	A	.	A928V	0	-7.89	prob.dam.	deleterious
IV	ALDH1L1	chr3:125865723	G	A	rs144099397	S344F	0.0005	-1.80	prob.dam.	deleterious
VI	MFN2	chr1:12067297	A	G	.	Q687R	0	0.65	prob.dam.	deleterious
VI	TAB2	chr6:149700430	C	T	.	T460M	0	0.65	prob.dam.	deleterious
VI	NOL11	chr17:65716047	A	T	.	D94V	0	0.65	prob.dam.	deleterious
VIII	AGT	chr1:230841679	C	T	rs74315283	R375Q	0	-1.80	prob.dam.	deleterious
VIII	EPSTI1	chr13:43543310	G	A	.	A84V	0.0001	-1.80	prob.dam.	deleterious
IX	GRM7	chr3:6903211	G	C	.	E46Q	0	-0.30	prob.dam.	deleterious
IX	MTMR14	chr3:9730694	C	T	.	P454L	0	-0.30	prob.dam.	deleterious
IX	CDAN1	chr15:43020878	C	T	rs200401359	G926R	0	-0.30	prob.dam.	deleterious
IX	ZNF836	chr19:52659758	T	A	.	K393M	0	-0.30	prob.dam.	deleterious
Variants predicted to be either probably damaging by PolyPhen2 or deleterious by SIFT										
I	KIF20B	chr10:91477454	G	A	rs149456198	V416I	0.0005	1.20	benign	deleterious
II	PDLIM7	chr5:176917023	C	T	.	R217H	0	1.22	NA	deleterious
III	CLASRP	chr19:45561058	T	C	.	V172A	0	-0.80	prob.dam.	tolerated
III	TULP4	chr6:158923961	C	T	rs140116628	T1089M	0.0003	-4.40	benign	deleterious
VI	ARHGEF10L	chr1:17961406	G	C	.	V608L	0	0.90	poss.dam.	deleterious
VI	TPO	chr2:1546296	C	T	.	R908C	0.0001	0.90	poss.dam.	deleterious
VII	OR1C1	chr1:247920937	C	T	.	V258I	0.0001	-3.06	poss.dam.	deleterious
VII	DDHD2	chr8:38097798	C	A	.	P210T	0.0002	-2.65	poss.dam.	deleterious

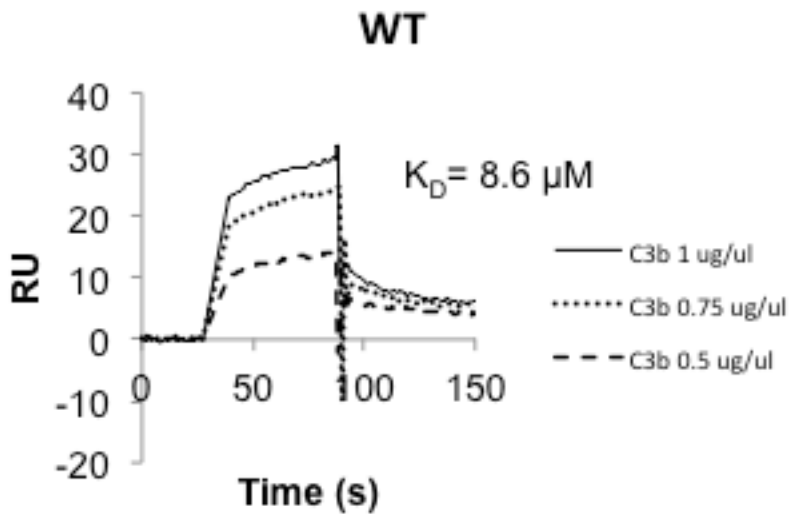
VII	SYT8	chr11:1857324	C	A	.	A156D	0	-8.40	poss.dam.	deleterious
VIII	ABCA9	chr17:67045529	G	C	rs150105567	R67G	0.0006	0.65	poss.dam.	deleterious
VIII	DNAH17	chr17:76482076	G	A	rs181353842	P2414L	0.0008	0.65	benign	deleterious
VIII	IGLC7	chr22:23265006	C	T	.	S81F	0	0.65	poss.dam.	deleterious
IX	GRM7	chr3:6903211	G	C	.	E46Q	0	0.65	prob.dam.	tolerated
IX	HLX	chr1:221053633	G	A	.	G1935D	0.0001	0.65	benign	deleterious
IX	SLC25A47	chr14:100795151	C	T	rs201454370	S139L	0.0004	0.65	benign	deleterious
IX	LRBA	chr4:151357949	T	A	rs147096866	D2294V	0.0002	0.65	poss.dam.	deleterious

FAM, family; Ref, reference allele; Alt, alternative allele; rsID, rs number; ESP, exome sequencing project; prob.dam., probably damaging; poss.dam., possibly damaging.

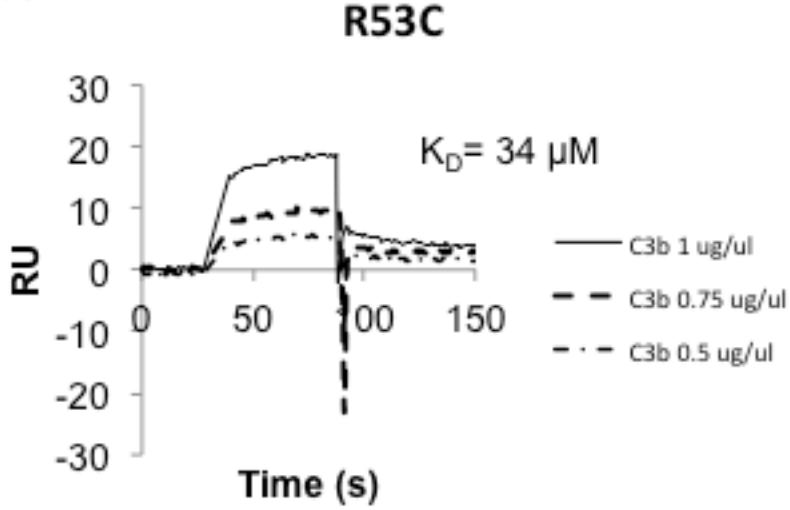


Supplemental Figure 1. Exome sequencing coverage and genotypes concordance.

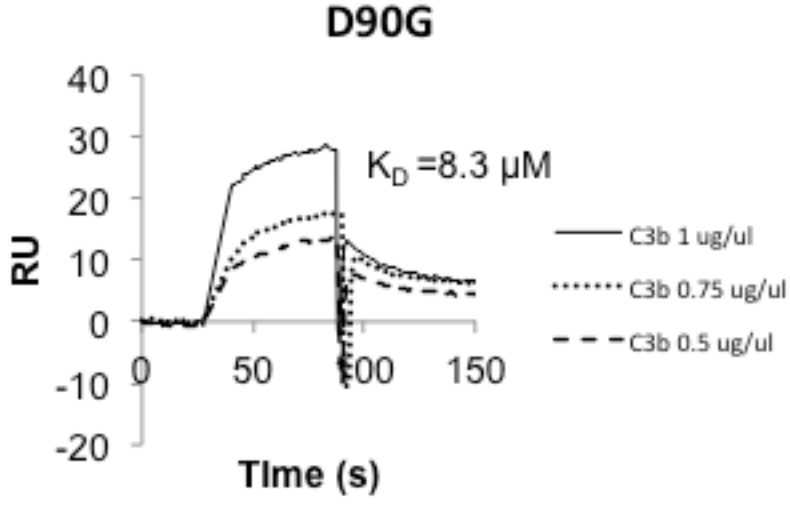
(A) All sequenced samples were required to have over 10X coverage at greater than 90% of the targeted regions(median = 97.25%) and over 20X coverage at greater than 80% of the targeted regions(median = 93.7%); (B) Distribution of the percent regions covered at >10X depth for each gene; (C,D) Histogram of correlations between minor allele dosages at 2,426 SNPs as determined by sequence-based and exome-array-based genotyping for common alleles (>1% frequency) and rare alleles (<1% frequency).


WT
K_D= 8.6 µM
RU
Time (s)
C3b 1 ug/ul
C3b 0.75 ug/ul
C3b 0.5 ug/ul

A


R53C
K_D= 34 µM
RU
Time (s)
C3b 1 ug/ul
C3b 0.75 ug/ul
C3b 0.5 ug/ul

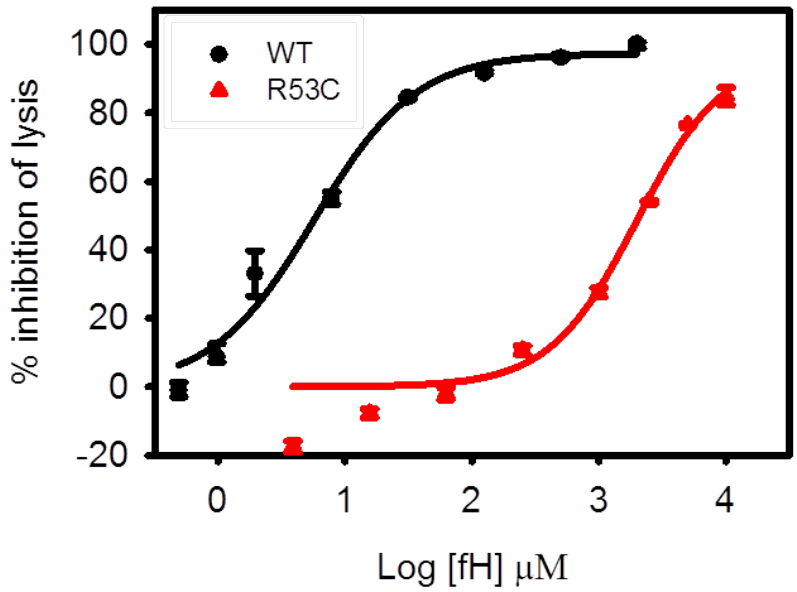
B


D90G
K_D =8.3 µM
RU
TIme (s)
C3b 1 ug/ul
C3b 0.75 ug/ul
C3b 0.5 ug/ul

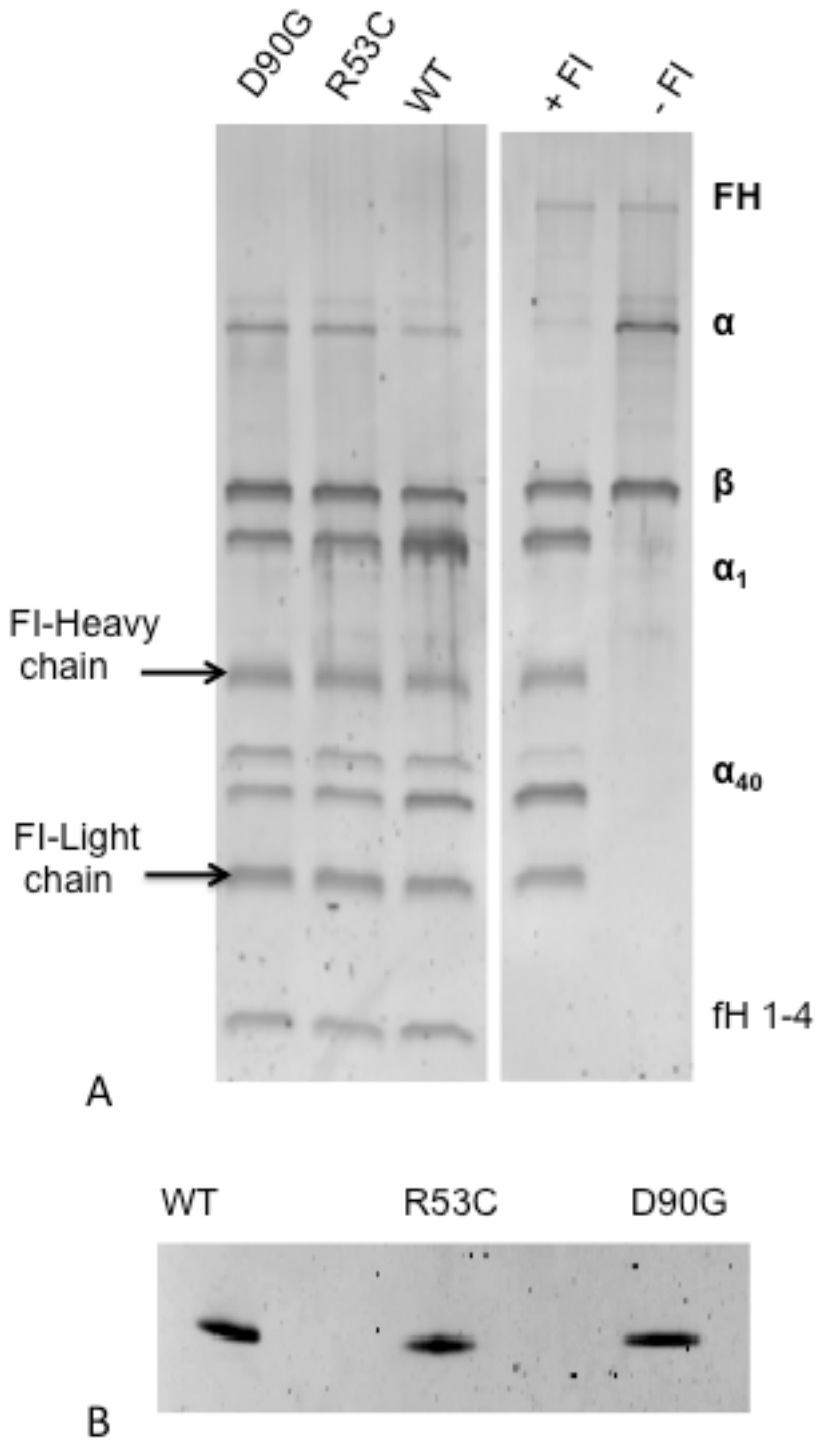
C

Supplemental Figure 2. CFH_{R53C}, but not CFH_{D90G}, demonstrates a weaker affinity for C3b compared to CFH_{WT}.

Overlaying sensograms show the steady state response for the binding of C3b (1-5 μM) to CFH1-4 proteins immobilized on a CM5 sensor chip. Response was plotted against concentration and the K_D was calculated using the 1:1 binding model in the BIAeval software.



Supplemental Figure 3. Hemolytic experiments confirm R53C's decay accelerating activity. In assays using sheep erythrocyte lysis, the decay defect of R53C is clear. 50% inhibition of lysis was achieved using 6.0 nM of WT. In contrast, 2000 nM (>300-fold more) was required using R53C.



Supplemental Figure 4. Representative gel of fluid phase cofactor assay.

(A) Representative gel shows that both mutants clearly fail to cleave α' at the same rate as WT and that as a result more α'remains and less α_1 and less α_{40} are generated (10% Tris-Gly SDS). (B) Equal amounts of each cofactor protein was present in each reaction (separate 12% Tris-Gly SDS).