Supplemental Material

Monogenic and Polygenic Contributions to Atrial Fibrillation Risk: Results from a National Biobank

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Tweet
TTN is the gene most commonly implicated in atrial fibrillation. Loss-of-function variants in *TTN* are highly penetrant, but polygenic risk explains a larger proportion of genetic susceptibility to atrial fibrillation.

Detailed Methods

Extended quality control in exome sequencing cohort

Whole exome sequencing variant calling

A 'Functionally Equivalent' dataset was created according to the primary analysis protocol[1] and was subject to GATK 3.0 variant calling. Variants with inbreeding coefficient<-0.03 or without at least one variant genotype of read depth ≥10, genotype quality ≥20 and, if heterozygous, allelic balance ≥0.20 were filtered out (https://biobank.ctsu.ox.ac.uk/showcase/label.cgi?id=170). Variants were annotated to genome build GRch38.

Sample quality control

In addition to sample quality control performed based on exome sequencing data, extended sample quality control was performed based on genotyping array data. Details on genotyping procedures and how quality metrics were derived are described in detail elsewhere[2]. In brief, genotyping was performed using Affymetrix UK biobank Axiom (450,000 samples) and Affymetrix UK BiLEVE axiom (50,000 samples) arrays[2]. Subsequently, the genetic data were imputed to the Haplotype Reference Consortium panel[3] and UK10K[4] + 1000 Genomes[5] panel. In the present analysis, samples that were outliers for heterozygosity or missingness were removed. In addition, individuals with putative sex chromosome aneuploidy or with a mismatch between self-reported and genetically inferred sex were excluded. Samples were further excluded if they were not used in the central kinship inference. We additionally restricted our analysis to individuals who were of white-European descent, as determined by Principal Component Analysis described previously[6]. Individuals who decided to revoke their consent were also excluded from the cohort, leaving 43,139 subjects. An unrelated subset of 41,335 individuals was also defined where no first, second or third degree relationships were present, as determined by KING coefficients of <0.0442[2, 7]. To identify the maximum number of unrelated individuals, we first calculated which individuals had excess relatives (2 or more) and iteratively removed these individuals until none remained. Then, for each pair of remaining related individuals, a single sample was removed at random.

Variant quality control for genetic relatedness matrix

A subset of high-quality variants from the genotyping array was used to estimate the genetic relatedness matrix used in SAIGE. Variants with MAF <1%, missingness >1% or that failed a stringent Hardy-Weinberg equilibrium test (p<0.001) were removed. Variants present in the MHC or the chromosome 8 inversion regions of the genome were additionally excluded. Finally, two rounds of linkage-disequilibrium-based pruning were performed (*--indep-pairwise 200 100 0.1* and *--indep-pairwise 200 100 0.05* in PLINK2.0[8]). After quality control, 93,491 high-quality independent variants remained.

Sample quality control in PRS validation cohort

The best performing AF PRS was identified in a validation cohort independent of the exome sequencing cohort. This group was formed using all individuals with genotyping array data for whom exome sequencing data was not available. Individuals were removed if they were related to any samples in the exome sequencing cohort by 3rd degree or closer (KING coefficient <0.0442) or if they were related to any other samples within the validation cohort. The remaining quality control steps were identical to those described in the extended sample quality control for the exome sequencing cohort, leaving 322,161 unrelated individuals of white-European descent with high-quality genotyping array data.

Extended procedure for PRS

LDpred algorithm

Derivation of multiple candidate PRSs for AF has been described in detail in a previous manuscript[9]. In short, LDpred employs a Bayesian approach that calculates posterior mean effect sizes for each variant, based on a

prior from GWAS summary statistics[10]. A subsequent shrinkage is applied based on the degree of linkage disequilibrium with other variants (the extent to which a given variant is correlated with other nearby variants in a reference population). A reference of European genome sequences from the 1000 Genomes panel (N=503) was used. The algorithm additionally takes into account a parameter ρ, which represent the assumed fraction of variants in the genome with nonzero effect sizes. A range of ρ values were used in the validation cohort: 1.0, 0.3, 0.1, 0.03, 0.01, 0.003, and 0.001. The PRS based on the value of ρ with the best predictive performance in the validation cohort was chosen and subsequently applied to the exome-sequencing cohort.

Individual scoring

Variant phasing and version 3 imputation in the UK Biobank have been described in detail previously[2]. Individuals were scored using genetic dosages of imputed variants in PLINK2.0[8], using function *--score cols=scoresums*. During this process, we restricted ourselves to variants with high imputation quality (INFO>0.3). Almost all LDpred-adjusted variants were present in the UK Biobank at high quality, as shown in a previous manuscript[11].

Online Table II. Exclusion criteria for electrocardiogram traits

ECG traits	RR interval	PR interval	QRS complex	Corrected QT interval	P wave duration
Exclusions	<600ms or >1500ms	<80ms or >320 ms	<60ms or >150ms	QRS complex >120ms, HR <40, or HR >120	<40ms or >180ms

Note, QT was corrected using Bazett's formula; HR: heart rate; ms: milliseconds

Online Table III. Area under the receiver-operator-curve (AUC) values and P-values for AF PRS based on different values of ρ using LDpred in the validation cohort

	With covariates (age, sex, PC1-4)			No covariates		
ρ	AUC	95%CI	P-value	AUC	95%CI	P-value
1	0.748	0.744-0.752	5.03E-167	0.57	0.565-0.575	1.97E-162
3.00E-01	0.749	0.744-0.753	7.40E-181	0.573	0.568-0.578	9.59E-176
1.00E-01	0.75	0.746-0.754	7.15E-216	0.579	0.574-0.584	1.83E-209
3.00E-02	0.753	0.749-0.757	3.42E-276	0.588	0.583-0.593	1.25E-267
1.00E-02	0.757	0.753-0.761	< 2.00E-280	0.6	0.594-0.605	< 2.00E-280
3.00E-03	***0.762***	***0.758-0.766***	***< 2.00E-280***	***0.613***	***0.608-0.618***	***< 2.00E-280***
1.00E-03	0.739	0.735-0.744	0.000803	0.509	0.504-0.514	0.000415

Abbreviations: AUC: area under the receiver operating characteristic curve, CI: confidence interval, PC: principle component of ancestry

Online Table IV. Risk of atrial fibrillation conferred by increasingly extreme tails of the polygenic risk score compared to the remainder of the population

Before removing cases of HF before AF					
Tail of PRS	**Sample size, N**	**No. AF cases in tail, N/ N total (%)**	**No. AF cases in remainder, N/ N total (%)**	**OR compared to remainder [95% CI]**	**P-value**
Top 33.3%	41335	758/13778 (5.5)	758/26080 (2.9)	2.2 [1.98-2.44]	<10E-15
Top 10%	41335	305/4135 (7.4)	1172/35723 (3.3)	2.53 [2.21-2.89]	<10E-15
Top 2.5%	41335	102/1033 (9.9)	1375/38825 (3.5)	3.21 [2.57-3.97]	<10E-15
Top 1%	41335	38/413 (9.2)	1439/39445 (3.6)	2.83 [1.97-3.95]	1.69E-07
Top 0.45%	41335	17/188 (9.0)	1460/39670 (3.7)	2.76 [1.60-4.47]	5.45E-04
Top 0.15%	41335	8/62 (12.9)	1469/39796 (3.7)	5.81 [2.53-11.89]	1.60E-04
Top 0.1%	41335	6/41 (14.6)	1471/39817 (3.7)	7.3 [2.76-16.63]	2.91E-04
After removing cases of HF before AF					
Tail of PRS	**Sample size, N**	**No. AF cases in tail, N/ N total (%)**	**No. AF cases in remainder, N/ N total (%)**	**OR compared to remainder [95% CI]**	**P-value**
Top 33.3%	41212	699/13737 (5.1)	655/26121 (2.5)	2.2 [1.99-2.48]	<10E-15
Top 10%	41212	284/4121 (6.9)	1070/35737 (3.0)	2.57 [2.24-2.95]	<10E-15
Top 2.5%	41212	95/1030 (9.2)	1259/38828 (3.2)	3.26 [2.59-4.06]	<10E-15
Top 1%	41212	36/412 (8.7)	1318/39446 (3.3)	2.93 [2.02-4.11]	1.48E-07
Top 0.44%	41212	17/181 (9.4)	1337/39677 (3.4)	3.24 [1.88-5.25]	8.48E-05
Top 0.2%	41212	10/82 (12.2)	1344/39776 (3.4)	5.3 [2.54-10.03]	5.43E-05
Top 0.1%	41212	6/41 (14.6)	1348/39817 (3.4)	7.86 [2.97-17.88]	1.88E-04

Abbreviations: AF: atrial fibrillation, OR: odds ratio, CI: confidence interval, PRS: polygenic risk score, HF: heart failure

Online Table V. Genome-wide significant gene from exome-wide gene-based burden analysis

					Unadjusted for PRS			Adjusted for PRS		
Gene	**Sample size, N**	**LOF carriers, N**	**LOF carriers in AF cases, N/Total N (%)**	**LOF carriers in controls, N/Total N (%)**	**OR**	**95% CI**	**P-value**	**OR**	**95% CI**	**P-value**
TTN	43139	554	49/1546 (3.17)	505/41593 (1.21)	2.71	1.97-3.66	2.50E-08	2.70	1.95-3.66	3.12E-08

Abbreviations: LOF: high confidence loss of function variants, AF: atrial fibrillation, OR: odds ratio, CI: confidence interval, PRS: polygenic risk score, SE: standard error

Online Table VI. Loss of function burden analysis results for previously reported monogenic candidate genes for AF

Gene	Sample size, N	LOF carriers, N	LOF carriers in AF cases, N/Total N (%)	LOF carriers in controls, N/Total N (%)	OR	95% CI	P-value	Ref
SYNE2	43139	154	1/1546(0.06)	153/41593(0.37)	0.26	[0.03-0.94]	0.04	[12]
RYR2	43139	832	21/1546(1.36)	811/41593(1.95)	0.60	[0.36-0.94]	0.09	[13]
SCN10A	43139	92	5/1546(0.32)	87/41593(0.21)	1.76	[0.57-4.16]	0.16	[14, 15]
KCNE5	43139	45	0/1546(0)	45/41593(0.11)	0.29	[0-2.07]	0.19	[16]
ZFHX3	43139	21	0/1546(0)	21/41593(0.05)	0.58	[0-4.39]	0.36	[12]
HCN4	43139	16	0/1546(0)	16/41593(0.04)	0.65	[0-5.17]	0.38	[17]
JPH2	43139	13	1/1546(0.06)	12/41593(0.03)	3.43	[0.35-16.11]	0.40	[18]
KCNE2	43139	12	0/1546(0)	12/41593(0.03)	0.79	[0.01-6.55]	0.41	[19-21]
KCNQ1	43139	20	0/1546(0)	20/41593(0.05)	0.87	[0.01-6.55]	0.43	[22-24]
NUP155	43139	14	0/1546(0)	14/41593(0.03)	0.83	[0.01-6.59]	0.45	[25]
KCNA5	43139	10	0/1546(0)	10/41593(0.02)	1.20	[0.01-10.17]	0.52	[26, 27]
SCN5A	43139	28	1/1546(0.06)	27/41593(0.06)	1.88	[0.21-7.5]	0.89	[28-30]
KCNN3	43139	151	5/1546(0.32)	146/41593(0.35)	0.89	[0.29-2.05]	0.90	[12]
Genes not testable for an association with AF due to an insufficient number of LOF carrier: *SCN1B, SCN2B, SCN3B, SCN4B, ABCC9, KCND3, KCNE1, KCNE3, KCNE4, KCNH2, KCNJ2, KCNJ5, KCNJ8, GATA4, GATA5, GATA6, GJA1, GJA5, GREM2, LMNA, NKX2-5, NKX2-6, NPPA*, and *PITX2*								

Abbreviations: LOF: high confidence loss of function variants, AF: atrial fibrillation, OR: odds ratio, CI: confidence interval, PRS: polygenic risk score, SE: standard error, Ref: reference.

Online Table VII. Loss of function burden analysis results for genes at AF GWAS loci

Gene	Sample size, N	LOF carriers, N	LOF carriers in AF cases, N/Total N (%)	LOF carriers in controls, N/Total N (%)	OR	95% CI	P-value
TTN	43139	554	49/1546(3.17)	505/41593(1.21)	2.71	1.97-3.66	2.50E-08
PKP2	43139	48	7/1546(0.45)	41/41593(0.1)	4.64	1.92-9.84	1.18E-03
DDX20	43139	99	10/1546(0.65)	89/41593(0.21)	3.24	1.57-6.04	1.98E-03
TMEM102	43139	826	15/1546(0.97)	811/41593(1.95)	0.44	0.25-0.73	3.73E-03
DENND3	43139	34	4/1546(0.26)	30/41593(0.07)	5.3	1.65-13.5	6.80E-03
KLHL38	43139	41	5/1546(0.32)	36/41593(0.09)	4.38	1.53-10.48	7.32E-03
TNK1	43139	64	6/1546(0.39)	58/41593(0.14)	3.27	1.27-7.13	1.35E-02
ADAM15	43139	34	4/1546(0.26)	30/41593(0.07)	4.3	1.33-11.11	1.45E-02
ATAD2	43139	2026	54/1546(3.49)	1972/41593(4.74)	0.72	0.53-0.94	1.83E-02
BID	43139	13	2/1546(0.13)	11/41593(0.03)	7.28	1.35-27	2.36E-02
ETV1	43139	11	2/1546(0.13)	9/41593(0.02)	7.27	1.26-29.86	2.60E-02
KCNJ1	43139	35	4/1546(0.26)	31/41593(0.07)	3.51	1.09-9.03	2.81E-02
HAUS4	43139	12	2/1546(0.13)	10/41593(0.02)	6.6	1.2-24.9	2.94E-02
USP18	43139	12	2/1546(0.13)	10/41593(0.02)	3.63	0.38-16.77	3.05E-02
PBXIP1	43139	58	5/1546(0.32)	53/41593(0.13)	2.89	1.04-6.56	3.16E-02
GFER	43139	54	5/1546(0.32)	49/41593(0.12)	2.94	1.05-6.78	3.20E-02
GIMAP8	43139	27	3/1546(0.19)	24/41593(0.06)	4.18	1.08-12.05	3.20E-02
POLR3D	43139	47	4/1546(0.26)	43/41593(0.1)	2.58	0.69-7.05	3.24E-02
KRTCAP2	43139	21	3/1546(0.19)	18/41593(0.04)	3.02	0.58-10.42	3.77E-02
SYNE2	43139	154	1/1546(0.06)	153/41593(0.37)	0.26	0.03-0.94	3.79E-02
ABCA3	43139	14	2/1546(0.13)	12/41593(0.03)	6.15	1.09-24.52	4.14E-02
TLE3	43139	84	5/1546(0.32)	79/41593(0.19)	1.86	0.51-4.82	4.92E-02

Note, among the 1181 potential genes/reading frames within +/- 500kb from the top 94 GWAS loci, 760 genes were not tested due to insufficient number of LOF carriers or because they were not protein-coding, GWAS: genome wide association studies, LOF: high confidence loss of function variants, AF: atrial fibrillation, OR: odds ratio, CI: confidence interval, PRS: polygenic risk score, SE: standard error

Online Table VIII. Frequency of LOF variants in genes implicated in monogenic forms of cardiovascular disease among the UK Biobank exomes

Gene	LOF carriers, N (%)	Relative frequency compared to LOF in canonical *TTN*	Relative frequency compared to LOF in cardiac *TTN*
***TTN* (canonical)**	554 (1.3)	1	2.8
***TTN* (cardiac)**	198 (0.44)	0.36	1
SCN5A	28 (0.065)	0.051	0.14
KCNQ1	20 (0.046)	0.036	0.10
MYBPC3	19 (0.044)	0.034	0.096
LDLR	12 (0.028)	0.022	0.061

Abbreviations: LOF: high-confidence loss-of-function

Online Table IX. *TTN* LOF variants among European ancestry in UK Biobank

Locus	Ref	Alt	Consequence	hgvsc	hgvsp	Control	AF	CMP	HF
chr2:178528273	C	T	splice donor	c.107377+1G>A		2	0	0	0
chr2:178528307	G	A	stop gained	c.107344C>T	p.Gln35782Ter	1	0	0	0
chr2:178528797	G	A	stop gained	c.106954C>T	p.Arg35652Ter	1	0	0	0
chr2:178529121	CT	C	frameshift	c.106629del	p.Ala35544ProfsTer2	1	0	0	0
chr2:178529958	A	T	splice donor	c.106531+2T>A		1	0	0	0
chr2:178530467	TG	T	frameshift	c.106147del	p.His35383MetfsTer22	1	0	0	0
chr2:178530809	GT	G	frameshift	c.105805del	p.Thr35269GlnfsTer24	1	0	0	0
chr2:178531423	AAC	A	frameshift	c.105190_105191del	p.Val35064PhefsTer4	1	0	0	0
chr2:178531668	G	A	stop gained	c.104947C>T	p.Gln34983Ter	2	0	0	0
chr2:178531788	G	A	stop gained	c.104827C>T	p.Arg34943Ter	2	0	0	0
chr2:178531887	TG	T	frameshift	c.104727del	p.Arg34910GlyfsTer5	7	1	0	0
chr2:178531890	AAAAGATATCAAATCTTGCAGACCTCT	A	frameshift	c.104699_104724del	p.Glu34900ValfsTer4	7	1	0	0
chr2:178531925	GA	G	frameshift	c.104689del	p.Ser34897ArgfsTer18	0	1	0	0
chr2:178532844	G	A	stop gained	c.103771C>T	p.Arg34591Ter	0	1	1	1
chr2:178532971	A	C	stop gained	c.103644T>G	p.Tyr34548Ter	0	1	0	0
chr2:178533652	G	GTTGA	frameshift	c.102959_102962dup	p.Ser34322GlnfsTer7	1	0	0	0
chr2:178534089	G	A	stop gained	c.102526C>T	p.Gln34176Ter	1	0	0	0
chr2:178534344	G	GGT	frameshift	c.102269_102270dup	p.Arg34091ThrfsTer8	2	0	0	0
chr2:178534619	C	T	stop gained	c.101996G>A	p.Trp33999Ter	0	1	0	0
chr2:178535516	T	TA	frameshift	c.101098_101099insT	p.Asp33700ValfsTer13	1	0	0	0
chr2:178536180	GA	G	frameshift	c.100566del	p.Pro33523GlnfsTer19	1	0	0	0
chr2:178536937	C	T	splice donor	c.100171+1G>A		1	0	0	0
chr2:178539764	CCT	C	frameshift	c.98299_98300del	p.Arg32767GlyfsTer2	1	0	0	0
chr2:178540303	C	T	stop gained	c.97863G>A	p.Trp32621Ter	1	0	0	0
chr2:178542492	C	A	stop gained	c.97264G>T	p.Glu32422Ter	0	1	1	1
chr2:178543508	T	TA	frameshift	c.96464dup	p.Thr32156AsnfsTer37	1	0	0	0
chr2:178544116	C	T	splice acceptor	c.96029-1G>A		1	0	0	0
chr2:178544241	G	C	stop gained	c.95988C>G	p.Tyr31996Ter	0	1	0	1
chr2:178545817	TACTG	T	splice donor	c.95415_95416+2del		2	0	0	0
chr2:178547518	CTTTAA	C	frameshift	c.94103_94107del	p.Ile31368SerfsTer34	1	0	0	0
chr2:178547890	TG	T	frameshift	c.93735del	p.Val31247Ter	0	1	0	0
chr2:178547907	CTGAT	C	frameshift	c.93715_93718del	p.Ile31239ValfsTer8	1	0	0	0
chr2:178547979	TC	T	frameshift	c.93646del	p.Asp31216ThrfsTer10	1	0	0	0
chr2:178548147	C	T	stop gained	c.93479G>A	p.Trp31160Ter	1	0	0	0
chr2:178548966	TGTAGATCA	T	frameshift	c.92652_92659del	p.Asp30885SerfsTer3	1	0	0	0
chr2:178549337	G	GCTTTT	frameshift	c.92284_92288dup	p.Ser30763ArgfsTer7	1	1	0	0
chr2:178551807	CTTTCT	C	frameshift	c.91088_91092del	p.Lys30363ArgfsTer4	1	0	0	0
chr2:178552221	T	A	stop gained	c.90679A>T	p.Lys30227Ter	0	1	1	1
chr2:178552236	G	GAA	frameshift	c.90663_90664insTT	p.Leu30222PhefsTer69	1	0	0	0
chr2:178552237	GGT	G	frameshift	c.90661_90662del	p.Thr30221ProfsTer14	1	0	0	0
chr2:178557324	TA	T	frameshift	c.87937del	p.Tyr29313MetfsTer88	1	0	0	0

Locus	Ref	Alt	Consequence	hgvsc	hgvsp	Control	AF	CMP	HF
chr2:178557545	TC	T	frameshift	c.87716del	p.Gly29239AspfsTer32	1	0	0	0
chr2:178559309	A	T	splice donor	c.86821+2T>A		1	0	0	0
chr2:178559385	TAATA	T	frameshift	c.86743_86746del	p.Tyr28915ThrfsTer22	1	0	0	0
chr2:178560364	G	A	stop gained	c.85768C>T	p.Arg28590Ter	1	0	0	0
chr2:178560865	G	A	stop gained	c.85267C>T	p.Arg28423Ter	2	0	0	0
chr2:178561762	CACTT	C	frameshift	c.84366_84369del	p.Glu28124IlefsTer49	1	0	1	1
chr2:178562528	TC	T	frameshift	c.83603del	p.Gly27868GlufsTer3	1	0	0	0
chr2:178562697	GC	G	frameshift	c.83434del	p.Ala27812ProfsTer59	1	0	0	0
chr2:178565497	G	A	stop gained	c.80635C>T	p.Gln26879Ter	1	0	0	0
chr2:178565651	C	T	stop gained	c.80481G>A	p.Trp26827Ter	0	1	0	0
chr2:178566149	GT	G	frameshift	c.79982del	p.Tyr26661SerfsTer16	1	0	0	0
chr2:178566162	T	TA	frameshift	c.79969_79970insT	p.Asp26657ValfsTer8	1	0	0	0
chr2:178566237	TC	T	frameshift	c.79894del	p.Glu26632AsnfsTer12	1	0	0	0
chr2:178566634	TG	T	frameshift	c.79497del	p.Thr26500LeufsTer24	1	0	0	0
chr2:178566838	G	A	stop gained	c.79294C>T	p.Arg26432Ter	0	1	0	1
chr2:178568033	GTTTC	G	frameshift	c.78095_78098del	p.Arg26032ThrfsTer41	1	0	0	0
chr2:178570804	G	A	stop gained	c.75328C>T	p.Arg25110Ter	1	0	0	0
chr2:178570882	G	A	stop gained	c.75250C>T	p.Arg25084Ter	0	1	0	0
chr2:178571261	CT	C	frameshift	c.74870del	p.Lys24957SerfsTer2	1	0	0	0
chr2:178572193	G	A	stop gained	c.73939C>T	p.Arg24647Ter	1	0	0	0
chr2:178573085	A	C	stop gained	c.73047T>G	p.Tyr24349Ter	1	0	0	0
chr2:178573323	AT	A	frameshift	c.72808del	p.Ile24270LeufsTer11	1	0	0	0
chr2:178573742	C	CAA	frameshift	c.72388_72389dup	p.Leu24130PhefsTer4	1	0	0	0
chr2:178573908	T	TC	frameshift	c.72223_72224insG	p.Lys24075ArgfsTer12	0	1	0	0
chr2:178575970	G	A	stop gained	c.70162C>T	p.Arg23388Ter	2	0	0	0
chr2:178576285	C	A	stop gained	c.69847G>T	p.Gly23283Ter	1	0	0	0
chr2:178579702	G	A	stop gained	c.67495C>T	p.Arg22499Ter	3	0	0	0
chr2:178579850	T	G	splice acceptor	c.67349-2A>C		3	0	0	0
chr2:178579938	C	T	splice donor	c.67348+1G>A		2	0	0	0
chr2:178580481	CAT	C	frameshift	c.66896_66897del	p.Asn22299SerfsTer15	1	0	0	0
chr2:178584726	G	A	stop gained	c.64915C>T	p.Arg21639Ter	1	0	0	0
chr2:178585291	G	A	stop gained	c.64453C>T	p.Arg21485Ter	1	0	0	0
chr2:178586802	GC	G	frameshift	c.64098del	p.Glu21366AspfsTer8	1	0	0	0
chr2:178587116	A	G	splice donor	c.64093+2T>C		1	0	0	0
chr2:178588700	G	A	stop gained	c.63025C>T	p.Arg21009Ter	1	0	0	0
chr2:178589076	AACAGTAC	A	frameshift	c.62642_62648del	p.Cys20881PhefsTer19	1	0	0	0
chr2:178589370	GTC	G	frameshift	c.62353_62354del	p.Asp20785HisfsTer3	1	0	0	0
chr2:178590170	G	A	stop gained	c.61555C>T	p.Arg20519Ter	2	0	0	0
chr2:178590260	T	A	stop gained	c.61465A>T	p.Arg20489Ter	2	0	0	0
chr2:178590974	T	A	stop gained	c.60751A>T	p.Lys20251Ter	1	0	0	0
chr2:178592916	CAG	C	frameshift	c.59201_59202del	p.Pro19734ArgfsTer5	0	1	1	1
chr2:178593172	C	A	splice donor	c.59035+1G>T		1	0	0	0
chr2:178593959	AC	A	splice donor	c.58432+1del		0	1	1	1
chr2:178598977	A	AT	frameshift	c.56732dup	p.Asp18911GlufsTer25	2	0	0	0
chr2:178599145	C	T	splice donor	c.56647+1G>A		1	0	0	0
chr2:178601739	G	A	stop gained	c.55351C>T	p.Arg18451Ter	1	0	0	0

Locus	Ref	Alt	Consequence	hgvsc	hgvsp	Control	AF	CMP	HF
chr2:178604215	C	CT	frameshift	c.54471dup	p.Val18158SerfsTer60	2	0	0	0
chr2:178605064	TC	T	frameshift	c.54112del	p.Glu18038ArgfsTer47	1	0	0	0
chr2:178605263	G	A	stop gained	c.53914C>T	p.Arg17972Ter	1	0	0	0
chr2:178607810	TTG	T	frameshift	c.52975_52976del	p.Gln17659ThrfsTer6	1	0	0	0
chr2:178608700	T	TTCAA	stop gained	c.52307_52310dup	p.Glu17437AspfsTer2	0	1	0	0
chr2:178609289	G	A	stop gained	c.52021C>T	p.Arg17341Ter	1	0	1	1
chr2:178609756	G	A	stop gained	c.51667C>T	p.Arg17223Ter	1	0	0	0
chr2:178610090	G	A	stop gained	c.51436C>T	p.Gln17146Ter	1	0	0	0
chr2:178610180	C	A	stop gained	c.51346G>T	p.Glu17116Ter	1	0	0	0
chr2:178611446	ACT	A	frameshift	c.50781_50782del	p.Arg16927SerfsTer14	1	0	0	0
chr2:178611606	GC	G	frameshift	c.50622del	p.Lys16874AsnfsTer12	1	0	0	0
chr2:178613158	TA	T	splice donor	c.49648+2del		2	1	0	0
chr2:178613938	C	T	splice acceptor	c.49346-1G>A		2	0	0	0
chr2:178614465	C	G	splice donor	c.49048+1G>C		0	1	0	0
chr2:178614871	CCT	C	frameshift	c.48734_48735del	p.Glu16245GlyfsTer32	1	0	0	0
chr2:178614930	G	GCATAC	frameshift	c.48672_48676dup	p.Ala16226GlyfsTer6	0	1	0	0
chr2:178616728	C	G	splice donor	c.48160+1G>C		0	1	0	0
chr2:178617839	G	A	stop gained	c.47512C>T	p.Arg15838Ter	1	0	0	0
chr2:178617857	G	A	stop gained	c.47494C>T	p.Arg15832Ter	1	0	0	0
chr2:178618468	A	AGTTAC	frameshift	c.46985_46989dup	p.Leu15664ValfsTer3	0	1	0	1
chr2:178620285	G	T	stop gained	c.46236C>A	p.Cys15412Ter	1	1	0	0
chr2:178621101	C	T	splice donor	c.45616+1G>A		1	0	0	0
chr2:178621150	C	CGTAA	frameshift	c.45564_45567dup	p.Val15190LeufsTer18	1	1	0	0
chr2:178621473	A	C	splice donor	c.45349+2T>G		1	0	0	0
chr2:178621474	C	A	splice donor	c.45349+1G>T		1	1	0	1
chr2:178621709	TG	T	frameshift	c.45114del	p.Asn15039ThrfsTer10	1	0	0	0
chr2:178622669	C	A	splice donor	c.44913+1G>T		1	0	0	0
chr2:178622684	G	A	stop gained	c.44899C>T	p.Arg14967Ter	0	1	0	1
chr2:178624544	A	AT	frameshift	c.44735dup	p.His14912GlnfsTer2	1	0	0	0
chr2:178624625	C	T	stop gained	c.44655G>A	p.Trp14885Ter	1	0	0	0
chr2:178629360	AG	A	frameshift	c.44364del	p.Tyr14789ThrfsTer15	1	0	0	0
chr2:178630250	G	A	stop gained	c.44272C>T	p.Arg14758Ter	1	0	0	0
chr2:178632700	T	A	stop gained	c.43306A>T	p.Lys14436Ter	0	1	0	1
chr2:178633279	CAA	C	frameshift	c.42992_42993del	p.Phe14331CysfsTer3	1	0	0	0
chr2:178633491	T	A	stop gained	c.42868A>T	p.Lys14290Ter	1	0	0	0
chr2:178633667	GGA	G	frameshift	c.42690_42691del	p.Pro14231LeufsTer8	1	0	0	0
chr2:178633670	TC	T	frameshift	c.42688del	p.Asp14230IlefsTer6	1	0	0	0
chr2:178635441	TC	T	frameshift	c.41882del	p.Gly13961GlufsTer11	1	0	0	0
chr2:178636098	G	A	stop gained	c.41473C>T	p.Arg13825Ter	0	1	0	1
chr2:178642236	C	T	splice donor	c.40558+1G>A		4	0	0	0
chr2:178646484	C	T	splice donor	c.40297+1G>A		2	0	0	0
chr2:178647063	C	A	splice donor	c.40222+1G>T		19	1	0	0
chr2:178647064	C	A	stop gained	c.40222G>T	p.Glu13408Ter	8	1	0	0
chr2:178649269	G	GT	frameshift	c.40035dup	p.Pro13346ThrfsTer13	1	0	0	0
chr2:178649816	C	A	splice donor	c.39895+1G>T		1	0	0	1
chr2:178650163	C	G	splice donor	c.39817+1G>C		1	0	0	0

Locus	Ref	Alt	Consequence	hgvsc	hgvsp	Control	AF	CMP	HF
chr2:178650755	TG	T	frameshift	c.39704del	p.Pro13235GlnfsTer75	1	0	0	0
chr2:178652542	C	T	splice acceptor	c.39044-1G>A		1	0	0	0
chr2:178652902	C	A	stop gained	c.38905G>T	p.Glu12969Ter	1	0	0	0
chr2:178653304	C	A	stop gained	c.38725G>T	p.Glu12909Ter	1	0	0	0
chr2:178653473	CT	C	frameshift	c.38660del	p.Lys12887ArgfsTer60	24	1	0	0
chr2:178654010	A	T	splice donor	c.38464+2T>A		1	0	0	0
chr2:178654536	T	G	splice acceptor	c.38207-2A>C		1	0	0	0
chr2:178657518	G	GGCTTTTTAGGA	frameshift	c.38007_38017dup	p.Pro12673LeufsTer278	1	0	0	0
chr2:178657735	T	A	stop gained	c.37906A>T	p.Lys12636Ter	2	0	0	0
chr2:178658104	T	A	stop gained	c.37765A>T	p.Lys12589Ter	2	0	0	0
chr2:178658356	C	T	splice acceptor	c.37628-1G>A		2	0	0	0
chr2:178661970	C	CT	frameshift	c.37178dup	p.Pro12394AlafsTer18	2	0	0	0
chr2:178662420	T	A	splice acceptor	c.36959-2A>T		4	0	0	0
chr2:178663270	A	AG	frameshift	c.36695dup	p.Glu12233Ter	1	0	0	0
chr2:178663903	C	T	splice acceptor	c.36365-1G>A		5	0	0	0
chr2:178664099	C	G	splice acceptor	c.36281-1G>C		1	0	0	0
chr2:178664443	GACAGTTAAGAATGTACCTTTGACAGGTACA	G	splice donor	c.36267_36280+16del		27	1	0	1
chr2:178664867	GT	G	frameshift	c.36102del	p.Glu12034AspfsTer124	0	1	0	0
chr2:178664879	TGGAA	T	frameshift	c.36087_36090del	p.Ser12030LysfsTer127	1	0	0	0
chr2:178665446	T	A	stop gained	c.35974A>T	p.Lys11992Ter	2	0	0	0
chr2:178665777	G	A	stop gained	c.35890C>T	p.Arg11964Ter	6	0	0	0
chr2:178666870	C	CT	frameshift	c.35828dup	p.Glu11945ArgfsTer6	1	0	0	0
chr2:178666896	C	A	stop gained	c.35803G>T	p.Glu11935Ter	1	0	0	0
chr2:178667239	C	A	stop gained	c.35794G>T	p.Glu11932Ter	2	0	0	0
chr2:178667276	AG	A	frameshift	c.35756del	p.Pro11919LeufsTer51	2	0	0	0
chr2:178667440	AC	A	splice donor	c.35713+1del		3	0	0	0
chr2:178667469	TAGGAA	T	frameshift	c.35681_35685del	p.Ile11894LysfsTer9	1	0	0	0
chr2:178667475	TAG	T	frameshift	c.35678_35679del	p.Thr11893AsnfsTer3	1	0	0	0
chr2:178672405	A	G	splice donor	c.34930+2T>C		4	0	0	0
chr2:178672634	C	T	splice donor	c.34855+1G>A		2	0	0	0
chr2:178675038	C	T	splice donor	c.34612+1G>A		2	0	0	0
chr2:178675722	T	A	stop gained	c.34486A>T	p.Lys11496Ter	1	1	0	0
chr2:178677634	TG	T	frameshift	c.34277del	p.Pro11426GlnfsTer42	3	1	0	0
chr2:178677873	GAGGT	G	frameshift	c.34035_34038del	p.Pro11346ArgfsTer121	1	0	0	0
chr2:178680253	C	T	splice donor	c.33418+1G>A		0	1	1	1
chr2:178681374	A	T	splice donor	c.33247+2T>A		1	0	0	0
chr2:178682904	C	A	splice acceptor	c.32888-1G>T		3	0	0	0
chr2:178684083	C	T	splice acceptor	c.32723-1G>A		1	0	0	0
chr2:178684741	C	A	stop gained	c.32563G>T	p.Glu10855Ter	1	1	0	0
chr2:178685252	C	T	splice donor	c.32470+1G>A		1	0	0	0
chr2:178685517	C	T	splice donor	c.32392+1G>A		2	0	0	0
chr2:178689289	C	A	splice donor	c.32011+1G>T		8	1	0	0
chr2:178689589	TC	T	frameshift	c.31852del	p.Glu10618LysfsTer7	1	0	1	1

Locus	Ref	Alt	Consequence	hgvsc	hgvsp	Control	AF	CMP	HF
chr2:178689897	C	T	splice acceptor	c.31763-1G>A		59	3	0	0
chr2:178692100	CT	C	splice acceptor	c.31679-2del		1	0	0	0
chr2:178692503	G	GT	frameshift	c.31671dup	p.Pro10558ThrfsTer5	2	0	0	0
chr2:178694598	C	G	splice donor	c.31426+1G>C		2	0	0	0
chr2:178696007	TTC	T	frameshift	c.31063_31064del	p.Glu10355SerfsTer10	1	0	0	0
chr2:178696271	T	C	splice acceptor	c.30803-2A>G		2	0	0	0
chr2:178698915	C	A	splice acceptor	c.30683-1G>T		4	0	0	0
chr2:178701120	CT	C	frameshift	c.30681del	p.Val10228LeufsTer6	1	0	0	0
chr2:178702527	GCTCT	G	frameshift	c.30356_30359del	p.Glu10119AlafsTer15	1	0	0	0
chr2:178704946	CCTCT	C	frameshift	c.29621_29624del	p.Glu9874GlyfsTer28	4	0	0	0
chr2:178706561	G	T	stop gained	c.29313C>A	p.Cys9771Ter	1	0	0	0
chr2:178706925	T	A	stop gained	c.29071A>T	p.Lys9691Ter	1	0	0	0
chr2:178706933	GC	G	frameshift	c.29062del	p.Ala9688GlnfsTer7	1	0	0	0
chr2:178706955	CT	C	splice acceptor	c.29042-2del		2	0	0	0
chr2:178714291	C	G	splice donor	c.26482+1G>C		3	1	0	0
chr2:178714985	C	T	splice donor	c.26200+1G>A		1	0	0	0
chr2:178715235	GA	G	frameshift	c.25950del	p.Pro8651LeufsTer2	1	0	0	0
chr2:178715707	A	T	stop gained	c.25707T>A	p.Tyr8569Ter	1	0	0	0
chr2:178717522	C	A	splice donor	c.25351+1G>T		2	0	0	0
chr2:178718149	TTACACTG	T	frameshift	c.24850_24856del	p.Gln8284LysfsTer36	1	0	0	0
chr2:178719452	C	A	splice acceptor	c.23939-1G>T		1	0	0	0
chr2:178719833	C	G	splice acceptor	c.23660-1G>C		1	0	0	0
chr2:178721872	CAT	C	frameshift	c.22789_22790del	p.Met7597ValfsTer15	1	0	0	0
chr2:178723045	C	A	splice donor	c.21961+1G>T		1	0	0	0
chr2:178723218	C	T	stop gained	c.21789G>A	p.Trp7263Ter	1	0	0	0
chr2:178723464	AG	A	frameshift	c.21635del	p.Ser7212LeufsTer15	1	0	0	0
chr2:178724539	C	T	splice acceptor	c.20837-1G>A		1	0	0	0
chr2:178727258	G	A	stop gained	c.20107C>T	p.Arg6703Ter	1	0	0	0
chr2:178727309	G	A	stop gained	c.20056C>T	p.Arg6686Ter	1	0	0	0
chr2:178727679	GAGATT	G	frameshift	c.19894_19898del	p.Asn6632LeufsTer7	1	0	0	0
chr2:178728221	ATC	A	frameshift	c.19601_19602del	p.Arg6534IlefsTer7	1	0	0	0
chr2:178729558	T	TG	frameshift	c.18597dup	p.Thr6200HisfsTer15	1	0	0	0
chr2:178729712	G	A	stop gained	c.18541C>T	p.Arg6181Ter	1	0	0	0
chr2:178730664	G	A	stop gained	c.17869C>T	p.Gln5957Ter	1	0	0	0
chr2:178730761	C	CT	frameshift	c.17771dup	p.Leu5925AlafsTer8	1	0	0	0
chr2:178731843	G	A	stop gained	c.17032C>T	p.Arg5678Ter	2	0	0	0
chr2:178732927	T	TA	frameshift	c.16248_16249insT	p.Ile5417TyrfsTer7	1	0	0	0
chr2:178733098	TA	T	frameshift	c.16077del	p.Phe5359LeufsTer28	3	0	0	0
chr2:178733371	G	A	stop gained	c.15922C>T	p.Arg5308Ter	2	0	0	0
chr2:178733497	G	A	stop gained	c.15796C>T	p.Arg5266Ter	1	0	0	0
chr2:178733518	C	A	splice acceptor	c.15776-1G>T		1	1	0	1
chr2:178734478	G	A	stop gained	c.15346C>T	p.Arg5116Ter	2	0	0	0
chr2:178734500	C	T	stop gained	c.15324G>A	p.Trp5108Ter	3	0	0	0
chr2:178734535	CA	C	frameshift	c.15288del	p.Cys5096TrpfsTer76	1	0	0	0
chr2:178734839	G	A	stop gained	c.15085C>T	p.Arg5029Ter	1	0	0	0
chr2:178735826	CAAACTTG	C	frameshift	c.14613_14619del	p.Asn4871LysfsTer10	1	0	0	0

Locus	Ref	Alt	Consequence	hgvsc	hgvsp	Control	AF	CMP	HF
chr2:178740151	CT	C	frameshift	c.13081del	p.Arg4361GlufsTer6	1	0	0	0
chr2:178740754	G	GT	frameshift	c.12478dup	p.Thr4160AsnfsTer11	1	0	0	0
chr2:178740827	CA	C	frameshift	c.12405del	p.Asn4135LysfsTer33	1	0	0	0
chr2:178741104	CTT	C	frameshift	c.12127_12128del	p.Lys4043ValfsTer7	1	0	0	0
chr2:178741304	G	A	stop gained	c.11929C>T	p.Gln3977Ter	1	0	0	0
chr2:178741575	AT	A	frameshift	c.11657del	p.Asp3886ValfsTer22	1	0	0	0
chr2:178741797	AT	A	frameshift	c.11435del	p.Asp3812ValfsTer20	0	1	0	0
chr2:178745884	C	A	stop gained	c.16516G>T	p.Glu5506Ter	6	0	0	0
chr2:178745884	C	CA	frameshift	c.16515dup	p.Glu5506Ter	1	0	0	0
chr2:178745946	A	ACTTTG	frameshift	c.16449_16453dup	p.Val5485AlafsTer3	1	0	0	0
chr2:178746042	C	CA	frameshift	c.16357dup	p.Cys5453LeufsTer22	0	1	0	0
chr2:178746047	G	C	stop gained	c.16353C>G	p.Tyr5451Ter	5	1	0	0
chr2:178746295	GTTCT	G	frameshift	c.16101_16104del	p.Lys5367AsnfsTer2	1	0	0	0
chr2:178746347	TG	T	frameshift	c.16052del	p.Pro5351HisfsTer5	2	0	0	0
chr2:178746457	CA	C	frameshift	c.15942del	p.Glu5315AsnfsTer2	1	0	0	0
chr2:178746748	G	A	stop gained	c.15652C>T	p.Arg5218Ter	1	0	0	0
chr2:178747087	G	A	stop gained	c.15313C>T	p.Arg5105Ter	1	0	0	0
chr2:178747087	G	GCTCTAGAGTCTCTCCTGGGGGTGTGGAGTATCA	stop gained	c.15312_15313insTGATACTCCACACCCCCAGGAGAGACTCTAGAG	p.Glu5104_Arg5105insTer	2	0	0	0
chr2:178747094	AG	A	frameshift	c.15305del	p.Thr5102IlefsTer2	1	0	0	0
chr2:178747120	T	A	stop gained	c.15280A>T	p.Arg5094Ter	2	0	0	0
chr2:178747139	G	GT	frameshift	c.15260_15261insA	p.Gly5089ArgfsTer43	1	0	0	0
chr2:178747224	AG	A	frameshift	c.15175del	p.Leu5059Ter	1	0	0	0
chr2:178747558	CA	C	frameshift	c.14841del	p.Val4948CysfsTer12	1	0	0	0
chr2:178747680	TG	T	frameshift	c.14719del	p.Gln4907AsnfsTer37	1	0	0	0
chr2:178747860	AT	A	frameshift	c.14539del	p.Ile4847Ter	5	0	0	0
chr2:178748025	G	GCCAA	frameshift	c.14371_14374dup	p.Ala4792ValfsTer25	1	0	0	0
chr2:178748109	G	C	stop gained	c.14291C>G	p.Ser4764Ter	1	0	0	0
chr2:178756292	T	TC	frameshift	c.11183dup	p.Leu3729ThrfsTer9	54	0	0	3
chr2:178756681	T	TA	frameshift	c.10794dup	p.Lys3599Ter	1	0	0	0
chr2:178757630	G	T	stop gained	c.10590C>A	p.Tyr3530Ter	2	0	0	0
chr2:178757721	T	TGAAACCATTGGATTTCTGGCTTGGGAATGCCA	frameshift	c.10467_10498dup	p.His3500LeufsTer16	2	0	0	0
chr2:178758982	A	G	splice donor	c.10303+2T>C		6	2	0	0
chr2:178764788	G	A	stop gained	c.9727C>T	p.Gln3243Ter	1	0	0	0
chr2:178767782	G	A	stop gained	c.9448C>T	p.Arg3150Ter	1	0	0	0
chr2:178769746	TG	T	frameshift	c.8834del	p.Thr2945LysfsTer20	1	0	0	0
chr2:178773207	A	AT	frameshift	c.7756dup	p.Ile2586AsnfsTer3	1	0	0	0
chr2:178773558	G	A	stop gained	c.7498C>T	p.Gln2500Ter	1	0	0	0
chr2:178774206	C	T	splice donor	c.7057+1G>A		1	0	0	0
chr2:178774375	CTTCTATATTTTCT	C	frameshift	c.6876_6888del	p.Ile2295GlyfsTer30	1	0	0	0
chr2:178774920	C	A	splice donor	c.6790+1G>T		1	0	0	0

Locus	Ref	Alt	Consequence	hgvsc	hgvsp	Control	AF	CMP	HF
chr2:178776073	G	A	stop gained	c.5791C>T	p.Gln1931Ter	1	0	0	0
chr2:178776378	GTA	G	frameshift	c.5484_5485del	p.Thr1829ArgfsTer22	1	0	0	0
chr2:178776745	T	A	stop gained	c.5119A>T	p.Lys1707Ter	1	0	0	0
chr2:178777004	AG	A	frameshift	c.4859del	p.Thr1620MetfsTer26	2	0	0	0
chr2:178777148	C	A	splice donor	c.4814+1G>T		1	0	0	0
chr2:178777234	CTTTCA	C	frameshift	c.4724_4728del	p.Met1575SerfsTer6	5	0	0	0
chr2:178779019	GA	G	frameshift	c.4062del	p.Leu1355PhefsTer42	1	0	0	0
chr2:178782240	A	AT	frameshift	c.3351dup	p.Ser1118IlefsTer21	1	0	0	0
chr2:178782872	G	A	stop gained	c.3034C>T	p.Arg1012Ter	1	0	0	0
chr2:178784175	G	C	stop gained	c.2670C>G	p.Tyr890Ter	1	0	0	0
chr2:178786030	G	A	stop gained	c.2188C>T	p.Gln730Ter	1	0	0	0
chr2:178786129	T	A	stop gained	c.2089A>T	p.Lys697Ter	0	1	0	0
chr2:178789977	CCTTCTCCTGAGTTATTTGCA	C	frameshift	c.1919_1938del	p.Val640AspfsTer19	2	0	0	0
chr2:178794436	G	GA	frameshift	c.1360_1361insT	p.Thr454IlefsTer25	2	0	0	0
chr2:178794437	T	TG	frameshift	c.1359_1360insC	p.Thr454HisfsTer25	2	0	0	0
chr2:178800574	A	ACCACAGGTGTAGGGATT	frameshift	c.387_403dup	p.Val135GlufsTer7	0	1	0	0
chr2:178800653	G	A	stop gained	c.325C>T	p.Arg109Ter	0	1	0	0

Abbreviations: Ref: reference allele, Alt: alternative allele, hgvcs: allelic changes in coding region, hgvsp: allelic changes in protein level, AF: atrial fibrillation, CMP: nonischemic cardiomyopathy, HF: heart failure.

Online Table X. *TTN* LOF association results with atrial fibrillation

Location of *TTN* LOF variants	Sample size, N	LOF carriers, N	*TTN* LOF carriers in AF cases, N /Total N (%)	*TTN* LOF carriers in controls, N /Total N (%)	OR [95%CI]	P-value
Canonical transcript	43139	554	49/1546 (3.17)	505/41593 (1.21)	2.71 [1.97-3.66]	2.50E-08
All transcripts	43139	591	51/1546 (3.30)	540/41593 (1.30)	2.66 [1.94-3.56]	2.80E-08
Cardiac exons (PSI≥90%)	43139	198	34/1546 (2.2)	164/41593 (0.4)	6.15 [4.07-9.06]	3.26E-14
Non-cardiac exons (PSI<90%)	43139	395	17/1546 (1.1)	378/41593 (0.91)	1.26 [0.74-2.00]	5.79E-01

Note, OR and 95% CI were estimated from Firth's logistic regression. OR: Odds ratio, CI: Confidence interval, PSI: percentage splicing index

Online Table XI. Sensitivity analyses for *TTN* LOF association with AF

Sensitivity analyses	All LOF variants in *TTN* (all transcripts)					LOF variants in *TTN* (cardiac exons)				
	Sample size, N	*TTN* LOF carriers in AF cases, N /Total N (%)	*TTN* LOF carriers in controls, N /Total N (%)	OR [95%CI]	P-value	Sample size, N	*TTN* LOF carriers in AF cases, N /Total N (%)	*TTN* LOF carriers in controls, N /Total N (%)	OR [95%CI]	P-value
Original result	43139	51/1546 (3.3)	540/41593 (1.3)	2.66 [1.94-3.56]	2.80E-08	43139	34/1546 (2.2)	164/41593 (0.4)	6.15 [4.07-9.06]	3.26E-14
Adjusting for PRS	43139	51/1546 (3.3)	540/41593 (1.3)	2.66 [1.94-3.58]	3.89E-08	43139	34/1546 (2.2)	164/41593 (0.4)	6.65 [4.38-9.84]	9.19E-15
Remove HF prior to AF	43007	43/1414 (3.0)	540/41593 (1.3)	2.44 [1.74-3.35]	1.70E-06	43007	27/1414 (1.9)	164/41593 (0.4)	5.35 [3.39-8.13]	1.12E-10
Remove HF prior to AF + Adjusting for PRS	43007	43/1414 (3.0)	540/41593 (1.3)	2.41 [1.71-3.32]	2.82E-06	43007	27/1414 (1.9)	164/41593 (0.4)	5.65 [3.57-8.65]	6.45E-11

Abbreviations: LOF: high confidence loss of function variants, AF: atrial fibrillation, OR: odds ratio, CI: confidence interval, PRS: polygenic risk score, HF: heart failure

Online Table XII. Association between RR interval and variants in *TTN*

	All LOF variants in *TTN* (all transcripts)			LOF variants in *TTN* (cardiac exons)		
Trait	***TTN* LOF carriers, N /Total N (%)**	**Effect size [95% CI]**	**P-value**	***TTN* LOF carriers, N /Total N (%)**	**Effect size [95% CI]**	**P-value**
RR interval	171/11522(0)	-0.22 [-0.37,-0.07]	4.06E-03	53/11522(0)	-0.52 [-0.79, -0.25]	1.40E-04

Abbreviations: LOF: high confidence loss of function variants, AF: atrial fibrillation, CI: confidence interval

Online Table XIII. Variability explained by models

Models	R^2	Improvement
Null model	0.211	-
Null model + *TTN*$_{LOF}$	0.213	0.002
Null model + *TTN*$_{LOF}$ + 13 AF genes	0.215	0.004
Null Model + PRS	0.258	0.047
Null model + *TTN*$_{LOF}$ + 13 AF genes + PRS	0.261	0.051

Note, Null model: covariates only model including age, sex, PCs 1-4, *TTN*$_{LOF}$: LOF variants in *TTN* exons highly expressed in cardiac tissue, 13 AF genes: *SYNE2, RYR2, SCN10A, KCNE5, ZFHX3, HCN4, JPH2, KCNE2, KCNQ1, NUP155, KCNA5, SCN5A, KCNN3* (with cumulative minor allele count ≥ 10), PRS: polygenic risk score

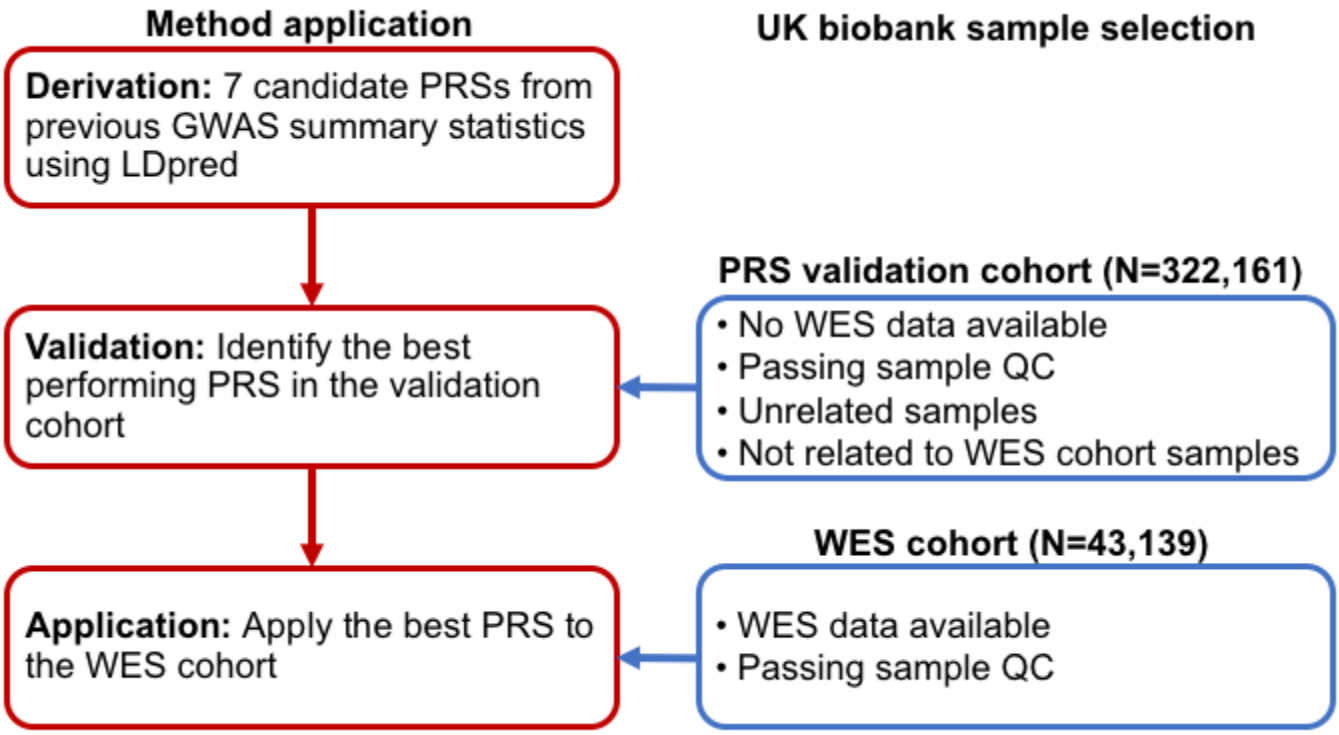
Online Table XIV. Results from LOF burden analysis for aggregation of LOFs in functional categories

Gene set	Sample size, N	LOF carriers, N	LOF carriers in AF cases, N/Total N (%)	LOF carriers in controls, N/Total N (%)	OR	95% CI	P-value
Potassium Channels	41212	251	4/1354(0.3)	247/39858(0.6)	0.52	[0.17-1.19]	1.32E-01
Potassium Channels IKs	41212	82	0/1354(0)	82/39858(0.2)	0.18	[0-1.27]	1.02E-01
Sodium Channels	41212	122	5/1354(0.4)	117/39858(0.3)	1.66	[0.61-3.62]	2.85E-01
Calcium Handling	41212	806	16/1354(1.2)	790/39858(2)	0.61	[0.36-0.97]	3.39E-02
Transcription Factors	41212	33	2/1354(0.1)	31/39858(0.1)	2.26	[0.45-7.07]	2.75E-01
Gap Junctions	41212	9	1/1354(0.1)	8/39858(0)	3.91	[0.4-19.17]	1.99E-01

Note, LOF variants in different candidate AF genes are aggregated into functional categories[31] and used for association tests; P-values, ORs and CIs are from firth's logistic regression performed in unrelated participants after excluding AF cases with heart failure prior to AF. LOF: loss-of-function variants, AF: atrial fibrillation, OR: odds ratio, CI: confidence interval, IKs: slowly activating potassium current.

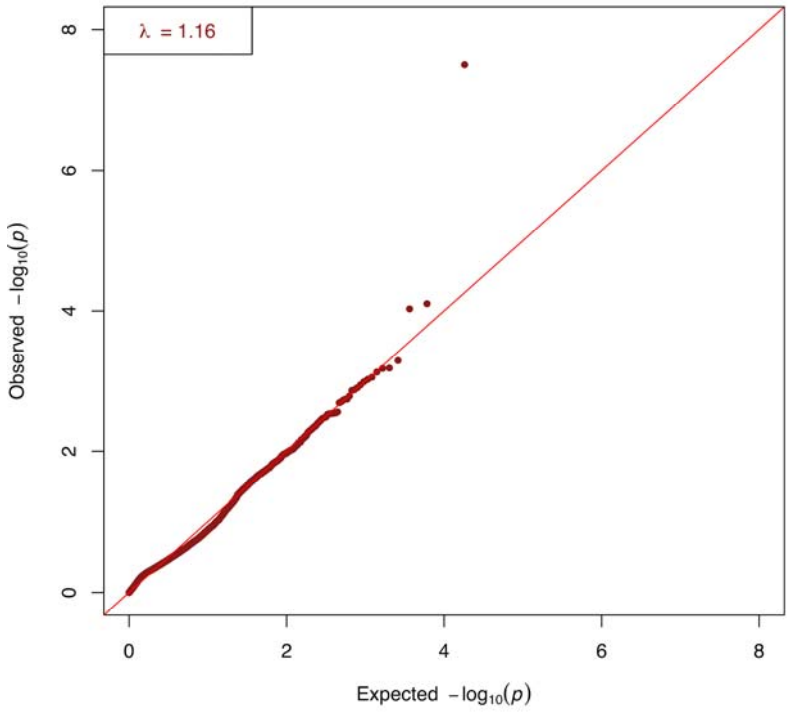
Potassium Channels include *ABCC9*, *HCN4, KCNA5, KCND3, KCNE1, KCNE2, KCNE3, KCNE4, KCNE5, KCNH2, KCNJ2, KCNJ5, KCNJ8, KCNN3, KCNQ1*; Potassium Channels IKs include *KCND3, KCNE1, KCNE2, KCNE3, KCNE4, KCNE5, KCNQ1*; Sodium Channels include *SCN1B, SCN2B, SCN3B, SCN4B, SCN5A, SCN10A*; Calcium Handling include *JPH2, RYR2*; Transcription Factors include *GATA4, GATA5, GATA6, NKX2-5, NKX2-6, PITX2, ZFHX3*; Gap Junctions include *GJA1, GJA5*

Supplemental Online Figure I



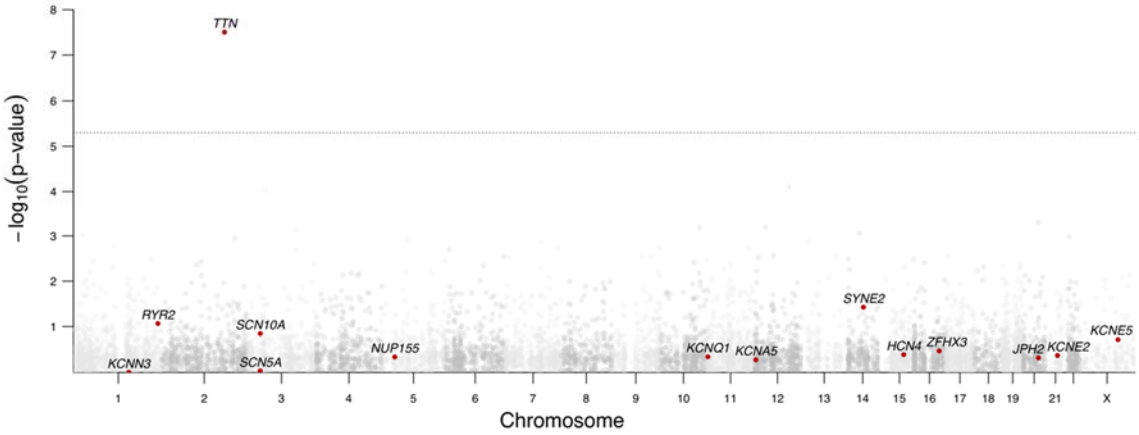
Online Figure I. Flowchart for AF polygenetic risk score validation and application in UK Biobank. Based on previous summary GWAS statistics[32], polygenic risk score (PRS) for atrial fibrillation (AF) was estimated using LDpred[9]. The estimated PRSs were validated in the validation cohort that did not include whole-exome sequenced participants in UK Biobank and tested in whole-exome sequencing (WES) cohort.

Supplemental Online Figure II



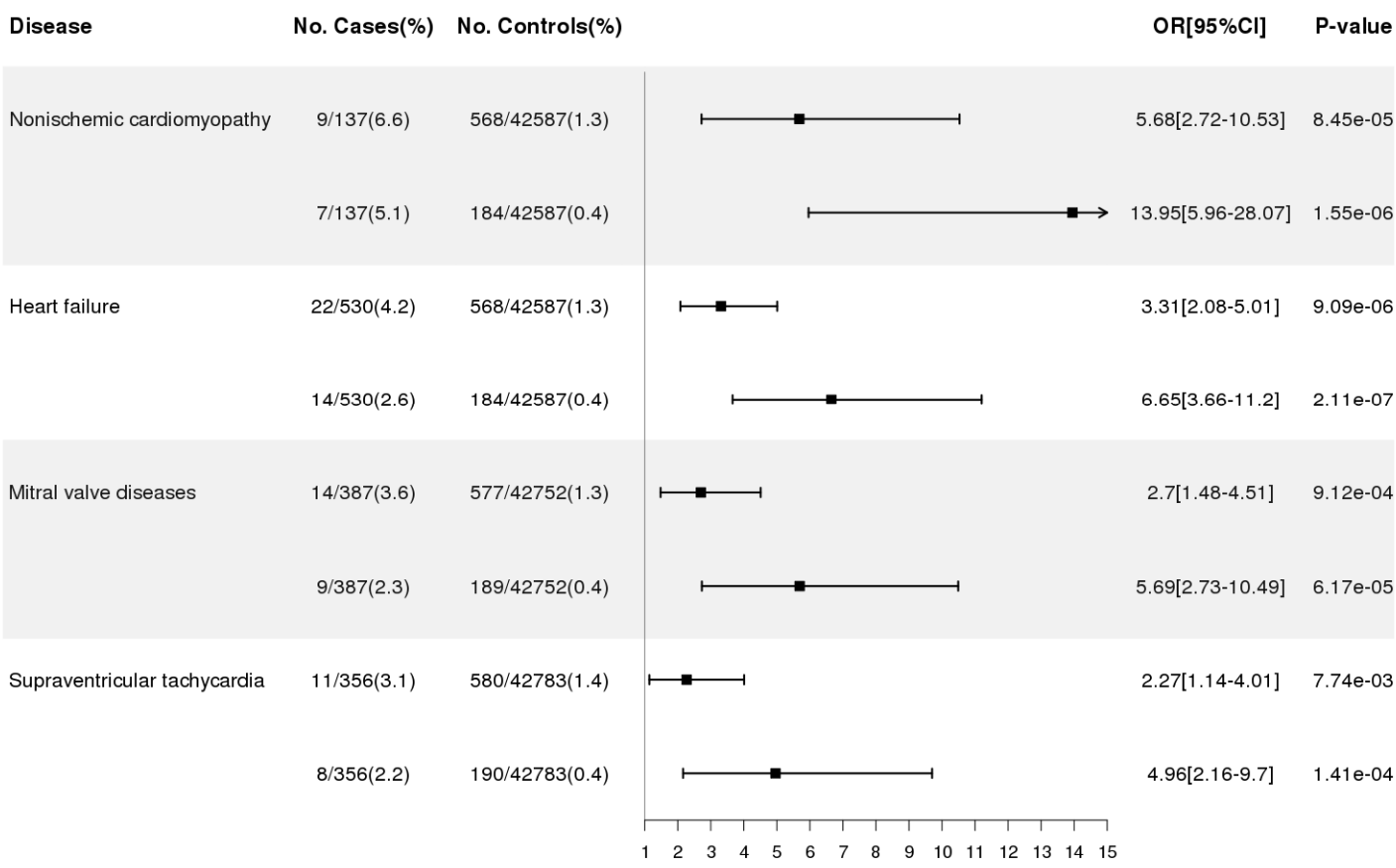
Online Figure II. Quantile-quantile plot for gene-based burden analysis for loss-of-function variants. The x-axis represents the expected significance for genes assuming no association, while the y-axis represents the observed significance in the analysis. The red line shows where the expected value and observed value for genes would fall assuming no association. In the present analysis, one gene strongly deviates from the null assumption. Although λ is considerably larger than 1, no significant genomic inflation is visible upon inspection, indicating limited systematic bias.

Supplemental Online Figure III



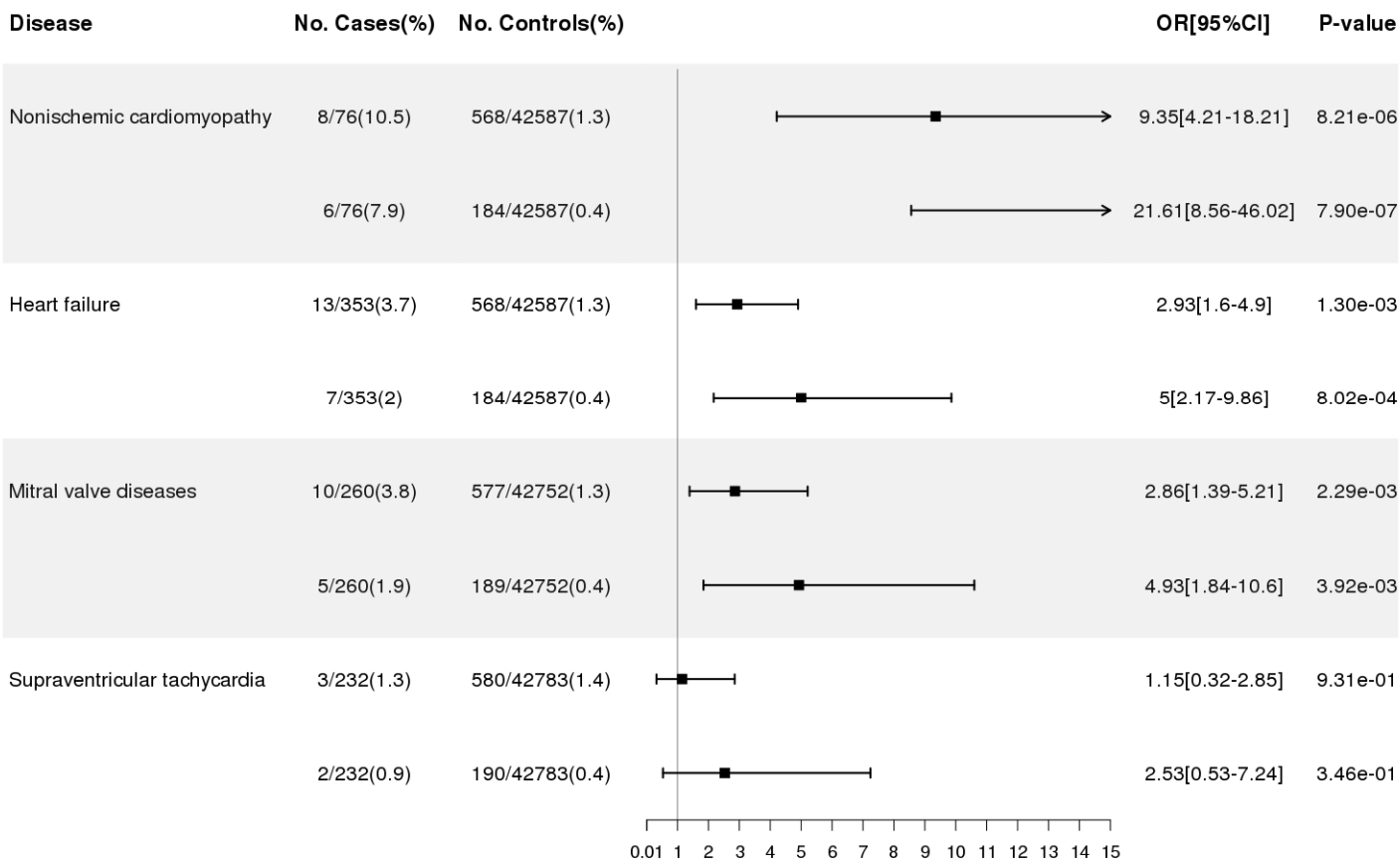
Online Figure III. Manhattan plot of the association between previously reported monogenic genes and atrial fibrillation. Red dots are the genes previously reported for association with atrial fibrillation. Among 37 previously reported monogenic candidate genes, 13 genes had enough carriers of loss-of-function variants to perform association tests.

Supplemental Online Figure IV



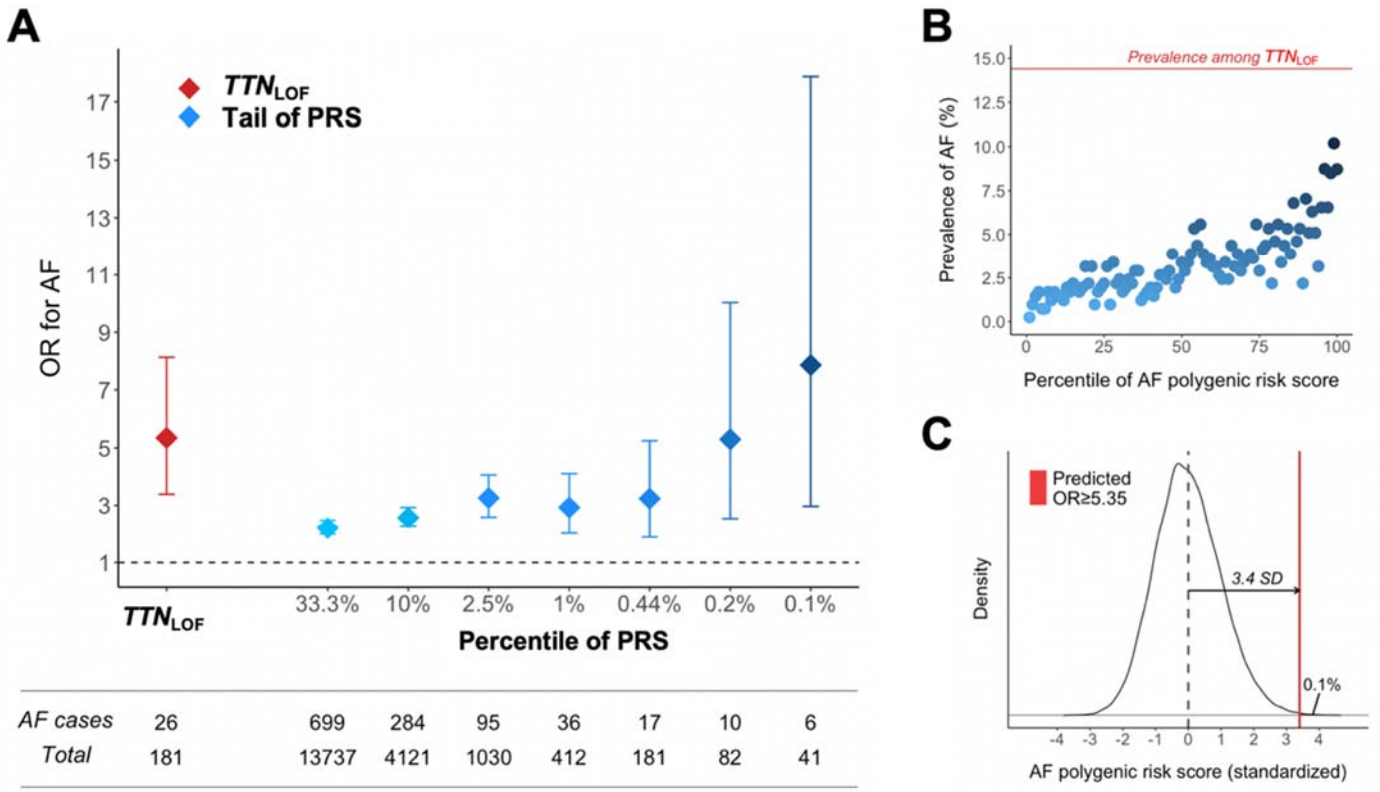
Online Figure IV. Forest plot for associated diseases and loss-of-function (LOF) variants in *TTN*. Online Figure IV illustrates the diseases significantly associated with LOF variants in *TTN.* The first and second rows per disease represent LOF variants in all transcripts of *TTN* and LOF variants in cardiac exons of *TTN*, respectively. Odds ratio and 95% confidence interval were estimated from Firth's logistic regression. OR: odds ratio.

Supplemental Online Figure V



Online Figure V. Sensitivity analyses between associated diseases and loss-of-function (LOF) variants in *TTN*. Online Figure V exhibits an association between diseases and LOF variants in *TTN*. Participants who had atrial fibrillation prior to the diagnosis of disease were removed in this analysis. The first and second rows per disease represent LOF variants in all transcripts of *TTN* and LOF variants in cardiac exons of *TTN*, respectively. Odds ratio (OR) and 95% confidence interval (CI) were estimated from Firth's logistic regression.

Supplemental Online Figure VI



Online Figure VI. Risk of atrial fibrillation conferred by loss-of-function variants in cardiac *TTN* compared to polygenic risk in the UK Biobank. Online Figure VIA shows the risk of atrial fibrillation (AF) conferred by loss-of-function variants in cardiac exons of *TTN* (TTN_{LOF}) and the risk conferred by high AF polygenic risk scores (PRS) in an unrelated subset of the exome sequencing cohort (N = 41,212). Increasingly extreme tails of the PRS distribution are compared to the remainder of the population and are shown in blue. TTN_{LOF} carriers, shown in red, are compared to noncarriers. Odds ratios (OR) are from Firth's logistic regression adjusted for sex, age and the first 4 principal components of ancestry. Individuals in the top 0.2% of PRS have a risk of AF comparable to the of TTN_{LOF} carriers. Online Figure VIB shows the prevalence of AF among percentiles of PRS in blue. The observed AF prevalence among all TTN_{LOF} carriers is indicated with the red line. Individuals in the highest percentiles of PRS do not attain an AF prevalence equivalent to that observed in TTN_{LOF} carriers. Online Figure VIC displays the distribution of the AF PRS in the population. Based on Firth's logistic regression, individuals at 3.4 standard deviations (SD) from the mean are predicted to be at equivalent AF risk to TTN_{LOF} carriers. Only 0.1% of the population is at equivalent or higher risk by PRS.

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