PCSK6 and Survival in Idiopathic Pulmonary Fibrosis

Justin M. Oldham MD*[1], Richard J. Allen PhD*[2], Jose M. Lorenzo-Salazar MSc*[3], Philip L. Molyneaux MBBS*[4,5], Shwu-Fan Ma PhD[6], Chitra Joseph PhD[7], John S. Kim MD[6], Beatriz Guillen-Guio PhD[2,8], Tamara Hernández-Beeftink MSc[8,9,], Jonathan A. Kropski MD[10], Yong Huang MD[6], Cathryn T. Lee MD[11], Ayodeji Adegunsoye MD[11], Janelle Vu Pugashetti MD[12], Angela L. Linderholm PhD[12], Vivian Vo BS[12], Mary E. Strek MD[11], Jonathan Jou MD[13], Adrian Muñoz-Barrera[3]; Luis A. Rubio-Rodriguez[3]; Richard Hubbard MBBS[14,15], Nik Hirani MBBS[16], Moira K.B. Whyte MBBS[16], Simon Hart MBBS[17], Andrew G. Nicholson MBBS[4,5], Lisa Lancaster MD[10], Helen Parfrey MBBS[18], Doris Rassl MBBS[18], William Wallace MBBS[16], Eleanor Valenzi MD[19], Yingze Zhang PhD[19], Josyf Mychaleckyj, DPhil;[20] Amy Stockwell MS[21], Naftali Kaminski MD[22], Paul J. Wolters MD[23], Maria Molina-Molina MD[24,25], Nicholas E. Banovich[26], William A. Fahy[27], Fernando J. Martinez MD[28], Ian P. Hall BMBCh[7,15], Martin D. Tobin MBChB[2,29], Toby M. Maher BM[4, 30], Timothy S. Blackwell MD[10], Brian L. Yaspan PhD[21], R Gisli Jenkins MBBS#[4,5], Carlos Flores PhD#[3,8,25,31], Louise V Wain PhD#[2,29], Imre Noth MD#[6]

*These authors contributed equally

#These authors contributed equally

Affiliations

1. Division of Pulmonary and Critical Care Medicine, University of Michigan, Ann Arbor, MI USA

AJRCCM Articles in Press. Published February 13, 2023 as 10.1164/rccm.202205-0845OC

2. Department of Health Sciences, University of Leicester, Leicester, UK
3. Genomics Division, Instituto Tecnológico y de Energías Renovables (ITER), Santa Cruz de Tenerife, Spain
4. National Heart and Lung Institute, Imperial College London, London, UK
5. Royal Brompton and Harefield Hospitals, Guy's and St. Thomas' NHS Foundation Trust, London, UK
6. Division of Pulmonary and Critical Care Medicine, University of Virginia, Charlottesville, VA USA
7. University of Nottingham, Division of Respiratory Medicine, Nottingham, UK
8. Research Unit, Hospital Universitario Nuestra Señora de Candelaria, Santa Cruz de Tenerife, Spain
9. Research Unit, Hospital Universitario de Gran Canaria Dr. Negrin, Las Palmas de Gran Canaria, Spain
10. Division of Pulmonary and Critical Care Medicine, Vanderbilt University, Nashville, TN USA
11. Section of Pulmonary and Critical Care Medicine, University of Chicago, Chicago, IL USA
12. Division of Pulmonary, Critical Care and Sleep Medicine, University of California Davis, Davis, CA USA
13. Department of Surgery, University of Illinois College of Medicine, Peoria, IL
14. Division of Epidemiology and Public Health, University of Nottingham, Nottingham, United Kingdom

AJRCCM Articles in Press. Published February 13, 2023 as 10.1164/rccm.202205-0845OC

15. National Institute for Health Research, Nottingham Biomedical Research Centre, Nottingham University Hospitals NHS Trust, Nottingham, United Kingdom
16. Medical Research Council Centre for Inflammation Research, The University of Edinburgh, Edinburgh, United Kingdom
17. Respiratory Research Group, Hull York Medical School, Castle Hill Hospital, Cottingham, United Kingdom
18. Cambridge Interstitial Lung Disease Service, Royal Papworth Hospital, Cambridge, United Kingdom
19. Division of Pulmonary, Allergy and Critical Care Medicine, The University of Pittsburgh, Pittsburgh, PA USA
20. Center for Public Health Genomics; University of Virginia, Charlottesville, VA, USA
21. Genentech Inc., San Francisco, CA USA
22. Section of Pulmonary, Critical Care and Sleep Medicine, Yale School of Medicine, New Haven, CT USA
23. Division of Pulmonary, Critical Care, Allergy and Sleep Medicine, University of California at San Francisco, San Francisco, CA USA
24. Servei de Pneumologia, Laboratori de Pneumologia Experimental, IDIBELL, Campus de Bellvitge, Universitat de Barcelona, Barcelona, Spain,
25. CIBER de Enfermedades Respiratorias, Instituto de Salud Carlos III, Madrid, Spain
26. Translational Genomics Research Institute, Phoenix, AZ USA
27. Discovery Medicine, GlaxoSmithKline, Stevenage, UK
28. Weill Cornell Medical Center, New York, NY USA

AJRCCM Articles in Press. Published February 13, 2023 as 10.1164/rccm.202205-0845OC

29. National Institute for Health Research, Leicester Respiratory Biomedical Research Centre, Glenfield Hospital, Leicester, UK
30. Division of Pulmonary and Critical Care Medicine, University of Southern California, Los Angeles, CA USA
31. Facultad de Ciencias de la Salud, Universidad Fernando Pessoa Canarias, Las Palmas de Gran Canaria, Spain

Corresponding Author

Justin M. Oldham

6301 MSRB3

1150 W. Medical Center Dr.

Ann Arbor, MI 48109-5642

oldhamj@med.umich.edu

Author Contributions

JMO, RJA, JMLS, SFM, RGJ, CF, LVW and IN designed and supervised the study. JMO, PLM, JAK, AA, JVP, MES, RH, NH, MW, SH, HP, NK, PW, MMM, FJM, TMM, TSB, RGJ and IN recruited patients for the study. JMO, PLM, SFM, JAK, AL, VV, YZ, AS, NK, PW, IH, MDT, WAF, TMM, TSB, BLY, RGJ, CF and IN contributed to DNA extraction and genotyping. CJ, AGN, DR, WW and RGJ secured lung tissue and performed the IHC analysis. NEB, JAK, TSB contributed single cell sequencing data. JMO, RJA, JMLS, CJ, JSK, BGG, THB, JAK, YH, AS, NEB, TSB, BLY, RGJ, CF, LVW performed the analyses. JMO, PLM, JK, CTL, AA, JVP, MES, EV, YZ, KG, DK, NK, PW, MMM, FJM, TMM, RGJ and IN collected paired clinical data. JMO, JMLS, RJA, RGJ, CF, LW

AJRCCM Articles in Press. Published February 13, 2023 as 10.1164/rccm.202205-0845OC

and IN wrote the manuscript with the input of all authors. All authors approved the final version of this manuscript.

Funding

NHLBI - K23HL138190 (JMO), R56HL158935 (JMO), K23HL150301 (JSK), K23HL146942 (AA), T32HL007605 and Pulmonary Fibrosis Foundation (CTL), R01HL145372(JAK/NEB), RO1HL130796 (IN), UG3HL145266 (IN). Ministry of Science and Innovation RTC-2017-6471-1; AEI/FEDER UE (CF). Instituto de Salud Carlos III co-financed by the ERDF "A way of making Europe" from the EU PI20/00876 (CF), Instituto Tecnológico y de Energías Renovables OA17/008 (CF). Consejería de Educación-Gobierno de Canarias and Cabildo Insular de Tenerife BOC n.º 173, 24/08/2017 (JMLS). UK National Institute for Health Research (NIHR) - RGJ holds an NIHR Research Professorship (RP-2017-08-ST2-014) and has MRC grants (G0901226 and MR/V00235X/1). TMM is supported by an NIHR Clinician Scientist Fellowship (NIHR reference CS-2013–13–017). MDT is supported by an NIHR Senior Investigator Award (NIHR201371). The views expressed are our own and not necessarily those of the NHS, the NIHR, or the UK Department of Health. British Lung Foundation Chair in Respiratory Research (C17-3) (TMM), GlaxoSmithKline/British Lung Foundation Chair in Respiratory Research (C17-1) (LWW). Wellcome Trust - WT202849/Z/16/Z (MDT), 221680/Z/20/Z (BGG). For the purpose of open access, the author has applied a CC BY public copyright license to any Author Accepted Manuscript version arising from this submission.

BREATHE — The Health Data Research Hub for Respiratory Health (MC_PC_19004) (LWW)

AJRCCM Articles in Press. Published February 13, 2023 as 10.1164/rccm.202205-0845OC

Action for Pulmonary Fibrosis Research Fellows (RJA and PLM). This research used the ALICE and SPECTRE High Performance Computing Facilities at the University of Leicester and TeideHPC (https://teidehpc.iter.es/en/home) at Instituto Tecnológico y de Energías Renovables.

Role of the Funding Source

This study was funded by the agencies listed above, which had no role in the design or conduct of the study. All authors had full access to all data, and the corresponding author had final responsibility for the decision to submit for publication.

Running Head: PCSK6 and Survival in IPF

Descriptor number: 9.23 interstitial lung disease

Manuscript word count: 3304

Abstract word count: 250/250

At a Glance Commentary

Scientific Knowledge on the Subject: A host of gene variants that increase one's risk of developing idiopathic pulmonary fibrosis have been identified through genomic analyses. Few of these variants have been associated with differential outcomes, however, suggesting that genomic determinants of IPF susceptibility and progression may have limited overlap. To identify genomic determinants of IPF outcomes, a genome-wide association study of IPF survival was conducted.

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What This Study Adds to the Field: We identified four novel gene variants associated with differential IPF survival, including one in *PCSK*6 that reached genome-wide significance. Downstream analysis showed that PCSK6 was highly expressed in IPF lung parenchyma and that PCSK6 lung staining intensity, peripheral blood gene expression and plasma concentration were each associated with reduced transplant-free survival. These findings suggest that PCSK6 may play a potentially important role in IPF progression.

Some of the results of these studies have been previously reported in the form of a preprint (medRxiv, 7 May 2022 www.medrxiv.org/content/10.1101/2022.05.06.22274705v1).

This article has an online data supplement, which is accessible from this issue's table of content online at www.atsjournals.org

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Abstract

Rationale: Idiopathic pulmonary fibrosis (IPF) is a devastating disease characterized by limited treatment options and high mortality. A better understanding of the molecular drivers of IPF progression is needed.

Objective: To identify and validate molecular determinants of IPF survival.

Methods: A staged genome-wide association study (GWAS) was performed using paired genomic and survival data. Stage I cases were drawn from centers across the US and Europe and stage II cases from Vanderbilt University. Cox proportional hazards regression was used to identify gene variants associated with differential transplant-free survival (TFS). Stage I variants with nominal significance ($p<5x10^{-5}$) were advanced for stage II testing and meta-analyzed to identify those reaching genome-wide significance ($p<5x10^{-8}$). Downstream analyses were performed for genes and proteins associated with variants reaching genome-wide significance.

Main Results: After quality controls, 1481 stage I cases and 397 stage II cases were included in the analysis. After filtering, 9,075,629 variants were tested in stage I, with 158 meeting advancement criteria. Four variants associated with TFS with consistent effect direction were identified in stage II, including one in an intron of proprotein convertase subtilisin/kexin type 6 (*PCSK6*) reaching genome-wide significance (HR 4.11; 95%CI 2.54-6.67; $p=9.45x10^{-9}$). PCSK6 protein was highly expressed in IPF lung parenchyma. PCSK6 lung staining intensity, peripheral blood gene expression and plasma concentration were associated with reduced transplant-free survival.

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Conclusions: We identified four novel variants associated with IPF survival, including one in *PCSK6* that reached genome-wide significance. Downstream analyses suggested that PCSK6 protein plays a potentially important role in IPF progression.

Keywords

Idiopathic Pulmonary Fibrosis

Genome-wide association study

Genomics

Survival

PCSK6

protein

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Introduction

Idiopathic Pulmonary Fibrosis (IPF) is a devastating disease characterized by progressive lung scarring and poor survival.(1, 2) Two anti-fibrotic therapies have been approved for the treatment of IPF after randomized controlled trials demonstrated efficacy in slowing lung function decline.(3, 4) Despite this advance, outcomes remain poor and anti-fibrotic therapy appears to provide only modest survival benefit.(5) To improve IPF outcomes, novel therapeutic targets are needed.

We and others have identified molecular IPF risk factors through unbiased investigation of the genome, transcriptome, and proteome.(6-15) Among the strongest molecular determinants of IPF is a common variant in the promoter region of *MUC5B,* which increases the odds of developing IPF by nearly 5-fold per risk allele.(6-9) Despite this strong association with IPF onset, the MUC5B promoter was paradoxically associated with improved survival,(16) though this association was potentially confounded by index event bias.(17) Few other susceptibility-associated gene variants have been shown to reliably predict differential IPF survival, suggesting that molecular determinants of IPF susceptibility and progression may have limited overlap.

To better understand molecular drivers of IPF progression and identify new therapeutic targets, we conducted a two-stage, multi-center, international genome-wide association study (GWAS) of IPF survival, followed by downstream analysis of genes and proteins associated with top survival-associated variant to determine whether these circulating proteins also predicted differential survival. Some results of this study have been

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previously reported in the form of abstracts at the American Thoracic Society(18) and British Thoracic Society(19) national meetings.

Methods

Cohorts and case selection

All patients provided informed consent for research blood draw in accordance with protocols approved by the institutional review board at each participating institution. GWAS stage I cases consisted of unrelated IPF patients of European ancestry from three previously described case-control GWAS datasets from the United States (US),(9) United Kingdom (UK),(7) and a combined cohort from the US, UK and Spain (UUS)(6). Available outcome data was gathered for all cases meeting international consensus criteria for IPF(20) and survival plotted for individual cohorts within each dataset. Patients without available outcome data were excluded, as were clinical trial cohorts due to short follow-up (**Supplemental Methods**). Stage II cases consisted of previously described, unrelated IPF patients of European ancestry from Vanderbilt University.(21) Vital status was assessed in US and Spain-based cohorts by chart review and contact with family members and in UK-based cohorts by review of national vital status databases.

Genotyping and quality control

Genotypes were generated for stage I cases using SNP (single nucleotide polymorphism) genotyping arrays according to previously described methods.(6, 7, 9) A summary of arrays used for each cohort is provided in the **Supplemental Methods**. Imputation for stage I cases was performed using the Michigan Imputation Server using the Haplotype Reference

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Consortium panel (v1.1 2016). Genotypes for the stage II cases were determined by whole genome sequencing, as previously described.(21) Stringent quality control measures were applied with a two-tier variant filtering scheme, with variants having minor allele frequency (MAF) 0.5-1% retained when imputation $R^2 \geq 0.8$ and those with MAF≥1% retained when imputation $R^2 \geq 0.5$. Variants deviating from Hardy-Weinberg equilibrium ($p<1.0 \times 10^{-6}$) were removed.

Genome-wide survival analysis

The primary endpoint assessed was transplant-free survival (TFS), defined as the time in months from site-determined date of IPF diagnosis to event (death or lung transplant) or censoring date. Variants associated with differential TFS were identified using a multivariable Cox proportional hazards regression model adjusted for age, sex, center, and first ten genetic principal components, with principal components calculated separately for each cohort. Variant genotypes were treated as a continuous variable with each patient having an imputed genotype dosage between zero and two risk alleles. To avoid considering results obtained for just one of the three individual studies (US, UK and UUS), only those variants with association results available for at least two datasets (US, UK and UUS) were meta-analyzed using a fixed-effect inverse variance weighted meta-analysis METAL (v2011-03-25) to generate stage I results.

Variants nominally associated with TFS in stage I were defined as those with Wald $p<0.05$ in at least two datasets with the same direction of effect and $p<5.0 \times 10^{-5}$ in stage I meta-analysis. Conditional analysis of these SNPs to deduce their independence was performed

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with GCTA-COJO v1.26. The proportional hazards assumption was then assessed for each independent variant meeting advancement criteria by testing whether Schoenfeld residual rank varied by genotype strata. Variants that satisfied the proportional hazards assumption were advanced for stage II testing. Stage I and II cases were then meta-analyzed using METAL with the genome-wide significance threshold set at $p<5.0x10^{-8}$. *In silico* assessments were used to infer the biological effect of variants associated with TFS after stage II testing.

PCSK6 tissue expression

Formalin-fixed paraffin-embedded human lung tissue sections obtained from patients with IPF undergoing surgical lung biopsy were compared to control subjects undergoing lung resection for malignancy, with sections distal to areas of malignancy utilized. Immunohistochemistry was performed using standard methods (**Supplemental Methods**) and mean staining intensity of PCSK6 protein was compared between IPF cases and non-IPF controls using a Mann-Whitney U-test. Single-cell RNA-sequencing data from prior published datasets(11, 22, 23) were reanalyzed and jointly annotated using label transfer from an updated annotated version of GSE135893. Cell-type annotation was performed using the TransferAnchors function in Seurat v4(24). Data visualization was performed using Scanpy v1.7. 2(25). The code used for analysis and presentation is available at http://github.com/KropskiLab/ipf_survival_gwas.

PCSK6 clinical outcome association

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The association between circulating *PCSK6* gene expression and TFS was assessed using three previously published microarray datasets from the COMET trial, Imperial College and the University of Chicago (**Supplemental Methods**),(26) which were analyzed separately with results meta-analyzed and presented as a forest plot. Circulating plasma PCSK6 protein concentration was then determined in patients with IPF from UC-Davis and UChicago (**Supplemental Methods**), $\log_2$ transformed, and tested for TFS association using Cox proportional hazards regression.(27) The proportional hazards assumption was satisfied for all downstream survival analyses.

Results

Case selection for stage I

Patients comprising the US cohort included those from the University of Chicago (n=118) and University of Pittsburgh (n=200) **(Figure E1, Figure E2)**. Those comprising the UK cohort included patients from the University of Edinburgh (n=119), Trent Lung Fibrosis Study (n=210), a subset of those participating in the prospective, multi-center PROFILE study (NCT 01134822) (n=175), and aggregated patients from smaller UK centers (Hull and Papworth) (n=61) **(Figure E1, Figure E3)**. Patients comprising the UUS cohort included those from the University of Chicago (independent of those from the US cohort, n=187), PROFILE study (independent of those in the UK dataset, n=299), University of California (Davis and San Francisco) (n=84) and aggregated patients from centers in Spain (n=28) **(Figure E1, Figure E4).**

Baseline characteristics and outcomes

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Following phenotypic exclusions, 1481 patients comprising stage I were included in the analysis. These included 318 patients from the US dataset, 565 from the UK dataset, and 598 from the UUS dataset. Baseline characteristics for each dataset are shown in **Table 1**. The mean age ranged from 67 to 72 years and males comprised 71-75% of each dataset. The mean percent predicted FVC and diffusion capacity of the lung for carbon monoxide (DLCO) was lowest in patients comprising the US dataset and highest in the UK dataset. A majority of patients in each dataset were classified as gender, age, physiology(28) (GAP) stage I or II. Median survival was highest in the UK cohort (53.2 months), followed by the UUS cohort (40.6 months) and US cohort (39.3 months) (p=0.001) (**Figure E2**). Survival also varied substantially across centers comprising each cohort (**Figures E3-5**). Median survival was 48 months in the Vanderbilt University validation cohort (**Figure E6**). Survival was similar among stage I and II cases through 24 months of follow-up but could not be compared thereafter due to violation of the proportion hazards assumption (**Figure E6**).

Genome-wide survival analysis

After filtering, 7,873,835 variants in the US dataset, 8,591,398 variants in the UK dataset, and 8,620,496 variants in the UUS dataset were tested for TFS association. Quantile-quantile plots for each stage I cohort suggested acceptable inflation(29) **(Figure E7).** After stratifying stage I cohorts by minor allele frequency (MAF), inflation was higher for rare variants compared to low and high frequency variants, but within an acceptable range for each group (λ<1.1) (**Figure E8**). For meta-analysis, 9,075,629 variants were tested for TFS association in the aggregated stage I cohorts. One hundred and sixty-one independent SNPs

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had Wald $p<0.05$ in at least two datasets with the same direction of effect and $p<5.0x10^{-5}$ in stage I meta-analysis (**Figure 1**). Of those, 158 satisfied the proportional hazards assumption and advanced for stage II testing (**Table E1**).

Genotype data was available in the Vanderbilt University cohort for 154 of the 158 variants advanced from stage I. Six variants were associated with TFS in the Vanderbilt University cohort at $p<0.05$, including four with consistent effect direction that strengthened in TFS association after meta-analysis (**Table 2**; **Table E2**)**.** These four were rs184498750 near Succinate-CoA Ligase GDP/ADP-Forming Subunit Alpha (*SUCLG1*), rs60514164 near ubiquitin-conjugating enzyme E2Q family member 2 (*UBE2Q2*), rs35647788 in an intron of Proprotein Convertase Subtilisin/Kexin Type 6 (*PCSK6*), and rs3893252 in an intron of Deleted In Azoospermia-Associated Protein 1 (*DAZAP1*) (**Table 2**). Of these, rs35647788 (*PCSK6*) showed the strongest TFS association across stage I (HR 4.76; 95% CI 2.62-8.64; $p=2.96x10^{-7}$) and stage II (HR 3.12; 95% CI 1.37-7.11; $p=6.70x10^{-3}$) cohorts and crossed the genome-wide significance threshold in meta-analysis (HR 4.11; 95% CI 2.54-6.67; $p=9.45x10^{-9}$) (**Table 2**). With the exception of rs60514164 (*UBE2Q2*) (MAF=8%), these SNPs were low frequency, with MAF of ~1% in the study population. Regional association plots for each of the four variants are shown in **Figure E9**.

In multivariable analysis, each variant except rs3893252 (*DAZAP1*) maintained survival association after adjustment for relevant confounders of IPF survival (**Table E3**). Among patients with the rs35647788 (*PCSK6*) variant, all were heterozygotes (**Table E4**) and were evenly distributed across centers comprising the UK and UUS cohorts. No

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rs35647788 (*PCSK6*) variants were observed in the US cohort despite good imputation quality (r^2=0.74). In sensitivity analysis of the *PCSK6* variant, results were consistent when censoring transplants (**Table E5**). *In silico* testing revealed functional effects for each of the four variants associated with TFS after stage II meta-analysis **(Table E6)**, none of which had known association with fibrotic lung disease. Using GTEx, we found multiple common sentinel PCSK6 expression quantitative trait loci (eQTL) in high LD (D'=1) with rs35647788 (**Table E7**).

PCSK6 tissue expression

Morphologic assessment of histological sections from lung tissue in patients with IPF were compared with control subjects without fibrotic lung disease. In IPF lung, cytoplasmic PCSK6 expression localized to ciliated epithelial cells and alveolar epithelial cells and was markedly higher than PCSK6 expression in non-IPF control sections (**Figure 2**). Western Blot confirmed the presence of only a single PCSK6 band (**Figure E10**). Relative staining intensity was two-fold higher in IPF lung samples (n=86) compared with non-IPF controls (n=9) (p<0.001) (**Figure 3a**). Increased PCSK6 protein staining score was associated with reduced TFS in those with available survival data (n=71), with staining scores above the median associated with greater than 2-fold increased risk of death or lung transplant (HR 2.41; 95% CI 1.12-5.16; p=0.024) (**Figure 3b**). Interrogating previously published scRNA-seq data from IPF and control lungs, PCSK6 expression was highest in lymphatic endothelial cells and adventitial fibroblasts, while broad expression in the airway epithelium was observed (**Figure E11**).

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PCSK6 clinical outcome association

When assessing *PCSK6* gene expression in the COMET (n=75), Imperial College (n=55) and University of Chicago (n=45) cohorts, increasing PCSK6 expression was associated with increased mortality risk in each cohort, with each one-unit increased associated with greater than three-fold increased risk of death or lung transplant in meta-analysis (HR 3.43; 95% CI 1.62-7.25; p=0.0012) (**Figure 4a**). When assessing PCSK6 plasma concentration in patients with IPF from UC-Davis (n=138) and UChicago (n=181), increasing plasma PCSK6 concentration was associated with reduced TFS, with each one-unit change in log-transformed plasma concentration associated with a nearly 50% increase in outcome risk (HR 1.47; 95% CI 1.14, 1.89; p=0.0031). These results were consistent across UC-Davis (HR 1.47, 95% CI 1.14-1.89; p=0.08) and UChicago (HR 1.34, 95% CI 0.92-1.94; p=0.12) cohorts. After stratification of the combined cohort by tertiles, those with PCSK6 concentration in the highest tertile displayed significantly worse survival than those in the second and third tertiles (p=0.0018) (**Figure 4b**).

Overlap between IPF risk and transplant free survival

Variants previously associated with IPF risk(6-10) were investigated for outcome association (**Supplementary Methods**). None of the 15 genetic variants with previously associated with IPF risk(6-10) were associated with TFS after Bonferroni correction (p=0.0033) (**Table E8**). As previously reported,(16, 17) individuals with the *MUC5B* promoter polymorphism (rs3570590) displayed better overall survival, though this did not reach significance after adjustment for multiple testing. None of the four validated survival variants were associated with differential IPF risk (**Table E9**). When combing the effect of

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thousands of IPF risk variants in a polygenic risk score, this risk score was not significantly associated with TFS for any significance threshold used (**Figure E12**), again suggesting that variants that affect disease risk may have little impact on survival times after diagnosis.

Discussion

In this investigation, we conducted the first GWAS of IPF survival, identifying a variant intronic to *PCSK6* that associated with differential TFS at genome-wide significance in two independent IPF cohorts totaling nearly two thousand patients. We subsequently found that PCSK6 protein was highly expressed in IPF lung tissue, localizing to the airway epithelium, which plays a key role in IPF onset and progression.(30) Finally, we found that PCSK6 lung staining, peripheral blood gene expression and circulating plasma concentration negatively correlated with TFS across independent IPF cohorts. To our knowledge, this study is the first to systematically identify gene variants associated with IPF survival and the first to identify PCSK6 as a potentially relevant mediator of IPF progression.

PCSK6, also called *PACE4*, encodes a widely expressed calcium-dependent serine endoprotease, which we demonstrate is expressed most highly in the airway epithelium, adventitial fibroblasts, and lymphatic endothelial cells in the lung. PCSK6 is a critical mediator of TGF-β processing and is crucial for reproduction, embryological development and blood pressure regulation.(31-35) A *PCSK6* gene variant has been implicated in the development of hypertension(35) and dysregulated *PCSK6* gene expression has been linked to vascular disease,(36-38) and cardiac remodeling following myocardial ischemia.(39)

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These cardiovascular remodeling effects make PCSK6 of particular relevance to IPF, as *PCSK6* overexpression can lead to increased collagen I and III deposition, TGF-β activation and extracellular matrix formation,(39, 40) which are cardinal features of IPF pathogenesis.(2) Additionally, PCSK6 may bind tissue inhibitors of metalloproteinases (TIMPs),(41) potentially counteracting the anti-metalloproteinase activity of TIMPs.(42)

PCSK6 dysregulation has also been implicated in the development of cancer of the lung,(43) breast,(44) ovary(45) thyroid(46) and prostate.(47) PCSK6 has been shown to regulate apoptosis in prostate cancer(48) and pancreatic cancer(49) and also linked to increased cancer cell invasiveness by enhancing bioactivity of matrix metalloproteinases and cytokines.(50) Accordingly, PCSK6 has been proposed as an anti-tumor therapeutic target(51, 52) and a bioavailable formulation of an anti-PCSK6 molecule is currently under investigation.(53) *In vitro* PCSK6 inhibition has already been shown to reduce fibroblast proliferation, migration and invasion in rheumatoid arthritis-associated synovitis.(54) Given its relatively high expression in adventitial, including airway-associated, fibroblasts and the airway epithelium, PCSK6 could serve as a potential therapeutic in patients with pulmonary fibrosis. However, additional research is needed to better characterize the role PCSK6 may play in IPF progression before therapeutic blockade is considered.

Despite being the largest genomic analysis of IPF survival performed to date, the modest size of this cohort limited our ability to identify higher frequency SNPs with modest effect sizes. Additionally, the rare nature of the *PCSK6* variant identified in this study suggests that it is unlikely to singularly explain subsequent gene expression and protein findings. *In silico*

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analyses identified several nearby regulatory elements which may include common functional variants, with smaller effects, in linkage disequilibrium with this *PCSK6* variant. Further research is needed to determine whether rs35647788 and other rare variants in *PCSK6* have additive effects on gene expression, along with investigation of regulatory elements, expression quantitative trait loci and more complex structural variants that may be contributing to our findings.

These results, and others recently published by our group,(55) demonstrate the value of case-only GWAS to identify genes associated with a relevant trait or outcome within a disease state. This approach is subject to temporal selection bias, however; as the timing of blood draw is likely associated with outcome when using date of blood draw as the starting point for determining survival time.(56) This approach was necessary given difficulties defining the date of diagnosis, along with variable periods of pre-clinical disease in patients with IPF, but variability in survival across cohorts suggests temporal selection bias was likely present in our study. This study also suggests potentially important differences between genomic determinants of IPF susceptibility and survival, whereby those involved in disease onset may be independent of those driving disease progression. No survival-associated variant identified in this analysis was associated with IPF risk. While replicating prior IPF risk association for the *MUC5B* promoter polymorphism, we found only weak association with favorable survival, an observation that may be influenced by index event bias.(17) As none of the survival-associated variants showed an association with disease risk, it is unlikely these survival results are affected by index event bias. These findings have potential

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implications for drug development, as genes associated with IPF survival may represent more effective therapeutic targets than those associated with IPF onset.

This study has several limitations. Given sample size constraints for this rare disease, we pursued a two-stage approach with meta-analysis of candidate variants rather than a discovery/replication approach, which would have required substantially higher sample sizes in each cohort. Furthermore, there was heterogeneity in the distribution of the *PCSK6* variant, as this variant was not detected in any individuals from the US cohort. This was likely driven by the rare nature of this variant, but consistent effect association in the stage II cohort and genome-wide significance for the *PCSK6* variant after meta-analysis increases confidence that this represents a true association, as does the downstream clinical outcome analysis showing PCSK6 gene expression and protein concentration to be associated with differential TFS. Next, we relied on all-cause mortality when modeling these data, leaving it unclear what proportion of cases died due to IPF. This could have biased results to some extent if variants were associated with death events unrelated to IPF. There were also substantial differences in transplantation across cohorts, which could have biased results when modeling transplant-free survival. This did not appear to be an issue for *PCSK6* variant, which maintained strong association with death when censoring transplant events (**Table E5**). Additionally, a large proportion of patients comprising the UUS dataset were recruited after the US approval of pirfenidone and nintedanib, which may impact survival,(57) and patients recruited before 2012 may have been exposed to potentially harmful immunosuppression.(58) Finally, to facilitate accurate imputation, we focused individuals of European ancestry, leaving it unclear whether these findings would extend to

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patients of non-European ancestry. The *PCSK6* variant was not present in any of the 123 patients excluded due to non-European ancestry, but dedicated analysis in this population is warranted. Additional variants of interest that failed to reach genome-wide significance were also identified, including one in DAZAP1, which lies close to PCSK4 on chromosome 19. Additional research is needed to ascertain the role these genes nearby these variants may play in IPF progression.

Conclusion

Here we present results from the first GWAS of IPF survival conducted to date. This study sheds important light on the genetics of IPF progression and identified novel variants which may contribute to this process, including rs35647788 in an intron of *PCSK6*. Downstream analysis demonstrated PCSK6 protein lung staining, peripheral blood gene expression and circulating plasma concentration to be associated with reduced IPF survival, suggesting PCSK6 may serve as a potential therapeutic target in patients with IPF.

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Figure Legend

Figure 1. Figure 1. Manhattan plot of Stage I gene variants associated with IPF survival. Each dot represents a gene variant, arranged on the x-axis by chromosome. Variants falling above the blue line, which corresponds to $p<5x10^{-5}$, are considered to reach nominal significance, while those falling above the red line, which corresponds to $p<5x10^{-8}$, are considered to reach genome-wide significance. All variants crossing the nominal significance threshold were advanced for stage II testing.

Figure 2. PCSK6 immunohistochemistry showed increased cytoplasmic PCSK6 expression in ciliated epithelial cells (A) and alveolar epithelial cells (B-C) compared with normal lung control sections (D-F). Parallel IPF section sections (G-H) confirm increased PCSK6 staining (G) when compared to control section (H). Human kidney positive control (I) is provided for reference. Magnification is 20um magnification for panels A-F and 50um for panels G-I.

Figure 3. Comparison of relative PCSK6 staining intensity between IPF cases and non-IPF controls demonstrated significantly higher median intensity in IPF lungs ($p<0.001$) (Figure 3a). Survival was lower among IPF cases with PCSK6 staining intensity above the median (Figure 3b).

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Figure 4. Relationship between PCSK6 and clinically relevant IPF endpoints. Higher peripheral blood gene expression (a) and circulating plasma protein concentration (b) are associated with reduced transplant-free survival.

Acknowledgements

We thank all the patients that contributed biologic samples and clinical data for this research.

Declaration of interests

JMO reports grants from the National Heart, Lung, and Blood Institute (NHLBI), American Lung Association and American Thoracic Society related to the submitted work and personal fees from Genentech, Boehringer Ingelheim, United Therapeutics, Lupin pharmaceuticals and AmMax Bio unrelated to the submitted work. PLM has received industry-academic funding from AstraZeneca and GSK and has received speaker and consultancy fees from Boehringer Ingelheim and Hoffman-La Roche outside the submitted work. JSK reports grants from the NHLBI and Pulmonary Fibrosis Foundation. JAK reports grants/contracts from NIH/NHLBI, Department of Defense, Doris Duke Charitable Foundation, Three Lakes Foundation, Boehringer-Ingelheim, Bristol-Myers-Squibb, has served as a consultant for Boehringer-Ingelheim, Janssen and APIE, and reports nonfinancial study support from Genentech. AGN reports consultancy fees for Galapagos, Medical Quantitative Image Analysis and Boehringer Ingelheim, and fees for educational activities from Boehringer Ingelheim and UptoDate. AA reports grants from the National Heart, Lung, and Blood Institute (NHLBI), American College of Chest Physicians, and the

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Pulmonary Fibrosis Foundation, and speaking and advisory board fees from Genentech and Boehringer Ingelheim. MES has received grants from Boehringer Ingelheim and personal fees from Boehringer Ingelheim and Fibrogen. HP reports grants from Roche; personal fees from Boehringer Ingelheim, Roche and Pliant Therapeutics and is a Trustee for the charity Action for Pulmonary Fibrosis. IPH holds an NIHR Senior Investigator Award. AS and BLY are employees of Roche/Genentech with stock and stock options in Roche. TMM has received industry-academic funding from GlaxoSmithKline (GSK) R&D and UCB; and has received consultancy or speakers fees from Apellis, AstraZeneca, Bayer, Biogen Idec, Boehringer Ingelheim, Cipla, GSK R&D, InterMune, ProMetic, Roche, Sanofi-Aventis, Sanumed, and UCB. NK served as a consultant to Biogen Idec, Boehringer Ingelheim, Third Rock, Pliant, Samumed, NuMedii, Theravance, LifeMax, Three Lake Partners, Optikira, Astra Zeneca, Veracyte, Augmanity and CSL Behring, over the last 3 years, reports Equity in Pliant and a grant from Veracyte, Boehringer Ingelheim, BMS and non-financial support from MiRagen and Astra Zeneca. NK has IP on novel biomarkers and therapeutics in IPF licensed to Biotech. RGJ reports grants from GSK, UK MRC, Biogen, AstraZeneca, Galecto and NIHR; personal fees from Boehringer Ingleheim, Bristol Myers Squibb, Chiesi, Galapagos, GSK, Heptares, MedImmune, Pharmakea, Roche/Promedior, Pliant Therapeutics, and Veracyte; and is a Trustee for the charity Action for Pulmonary Fibrosis. LVW has received funding from GlaxoSmithKline. IN reports personal fees and consultancy, advisory board, speaker, and study contracts for Boehringer Ingelheim and Genentech; personal fees from Immuneworks, Sanofi Aventis, Global blood therapeutics; and has a patent for Toll-interacting protein (TOLLIP) and pharmacogenetics pending. All other authors declare no competing interests.

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Data Sharing Statement

GWAS Summary statistics for this study are available at https://github.com/genomicsITER/PFgenetics..

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Table 1. Baseline characteristics and outcomes of stage I and II datasets

		Stage I (n=1481)		Stage II (n=397)
Characteristic	**US* (n=318)**	**UK** (n=565)**	**UUS*** (n=598)**	**Vanderbilt****
Age	67.3 (8.9)	72.1 (8.4)	69.8 (8.3)	65.7 (9.0)
Male sex	234 (73.6)	403 (71.3)	449 (75.1)	295 (74.3)
FVC, % predicted	62.7 (17.9)	77.7 (18.5)	70.2 (18.6)	65.0 (16.1)
DLCO, % predicted	36.7 (14.6)	43.3 (14.3)	39.9 (14.6)	39.1 (13.5)
GAP Stage				
I	72 (23.1)	106 (35.2)	135 (24.2)	115 (29.8)
II	149 (47.8)	144 (47.8)	286 (51.4)	201 (52.1)
III	91 (29.2)	51 (16.9)	136 (24.4)	70 (18.1)
Death	189 (59.4)	366 (64.8)	257 (43.0)	202 (50.9)
Transplant	52 (16.4)	2 (0.4)	26 (4.4)	32 (8.1)
Death or transplant	241 (75.8)	366 (64.8)	283 (47.3)	234 (58.9)
Median survival months (IQR)	39.3 (13.4-70.6)	53.2 (24.8-92.5)	40.6 (19.9-75.5)	48 (15-105)

* n for missing data: FVC (n=6); DLCO (n=31): GAP Stage (n=6)
** n for missing data: FVC (n=241); DLCO (n=264): GAP Stage (n=241)
*** n for missing data: FVC (n=30); DLCO (n=41); GAP Stage (n=30)
**** n for missing data: FVC (n=4); DLCO (n=9); GAP stage (n=4)

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Table 2. Results for the SNPs nominally associated across stages and with same direction of effects on IPF survival

Variant location (hg19) and nearest gene						Stage I results				Stage II results			Meta-analysis	
Chr	**Position**	**SNP rsID**	**Gene**	**REF**	**EA**	**R^2**	**EAF**	**HR [95% CI]**	***P-value***	**EAF**	**HR [95 % CI]**	***P-value***	**HR [95 %CI]**	***P-value***
2	84291167	rs184498750	*SUCLG1*	G	T	0.86	1.1%	3.11 [1.88-5.15]	$9.83x10^{-6}$	1.2%	2.05 [1.79-4.18]	0.049	2.71 [1.79-4.08]	$2.07x10^{-6}$
15	76081200	rs60514164	*UBE2Q2*	C	T	0.77	7.0%	1.60 [1.32-1.95]	$2.35x10^{-6}$	7.7%	1.42 [1.04-1.93]	0.026	1.55 [1.31-1.83]	$2.23x10^{-7}$
15	101914234	rs35647788	*PCSK6*	C	T	0.93	0.8%	4.76 [2.62-8.64]	$2.96x10^{-7}$	0.8%	3.12 [1.37-7.11]	$6.72x10^{-3}$	4.11 [2.54-6.67]	$9.45x10^{-9}$
19	1412985	rs3893252	*DAZAP1*	C	T	0.82	0.6%	3.57 [1.97-6.49]	$2.91x10^{-5}$	1.5%	2.09 [1.05-4.15]	0.036	2.84 [1.81-4.45]	$5.81x10^{-6}$

Abbreviations: Ref = reference allele; EA = effect allele; R^2= lowest imputation quality value across studies; EAF = effect allele frequency; HR = hazard ratio; CI = confidence interval

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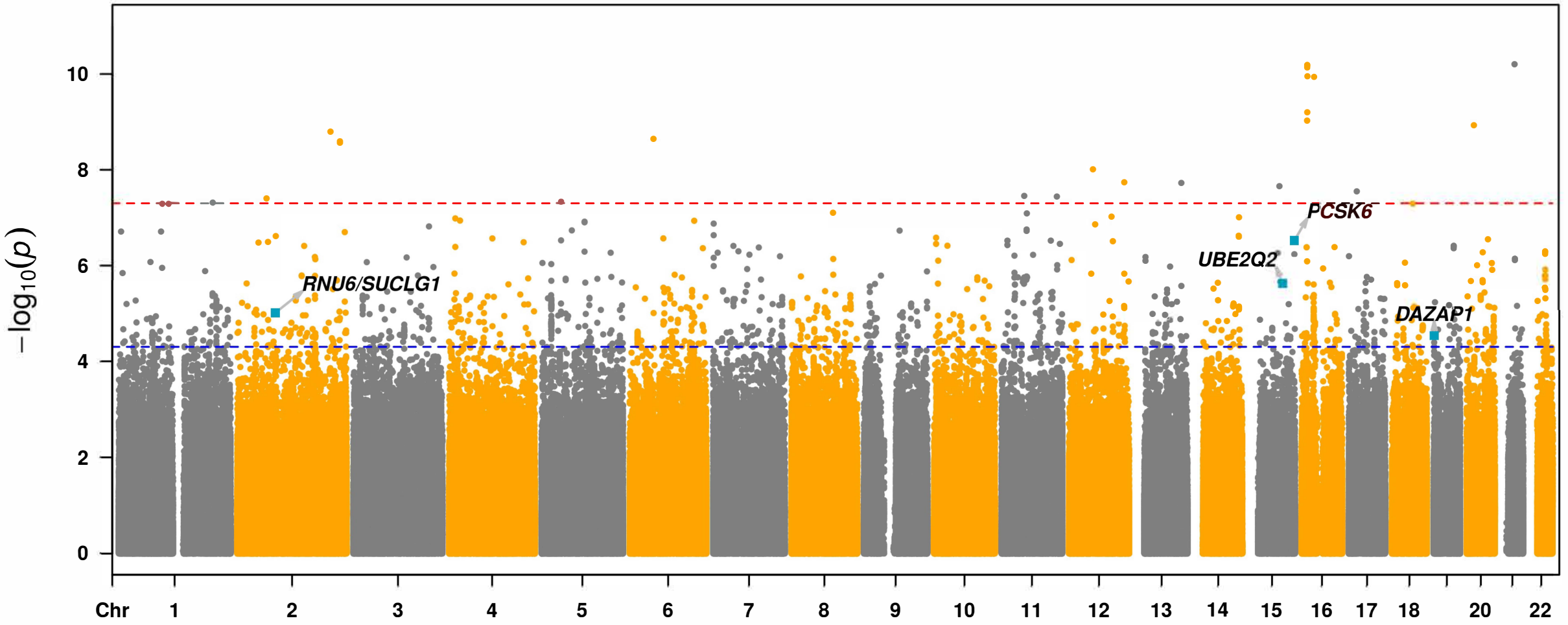
Figure 1



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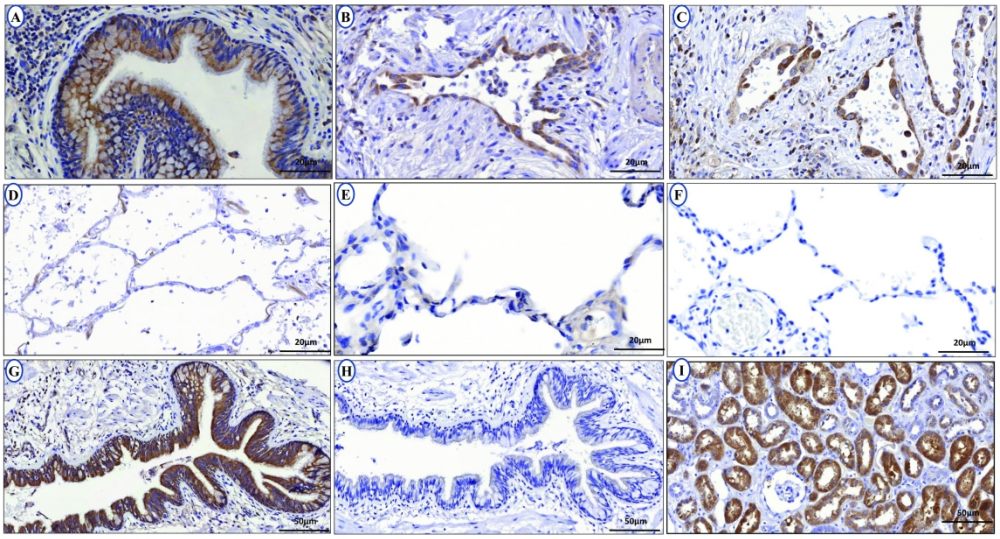

Figure 2. PCSK6 immunohistochemistry showed increased cytoplasmic PCSK6 expression in ciliated epithelial cells (A) and alveolar epithelial cells (B-C) compared with normal lung control sections (D-F). Parallel IPF section sections (G-H) confirm increased PCSK6 staining (G) when compared to control section (H). Human kidney positive control (I) is provided for reference. Magnification is 20um magnification for panels A-F and 50um for panels G-I.

338x190mm (300 x 300 DPI)

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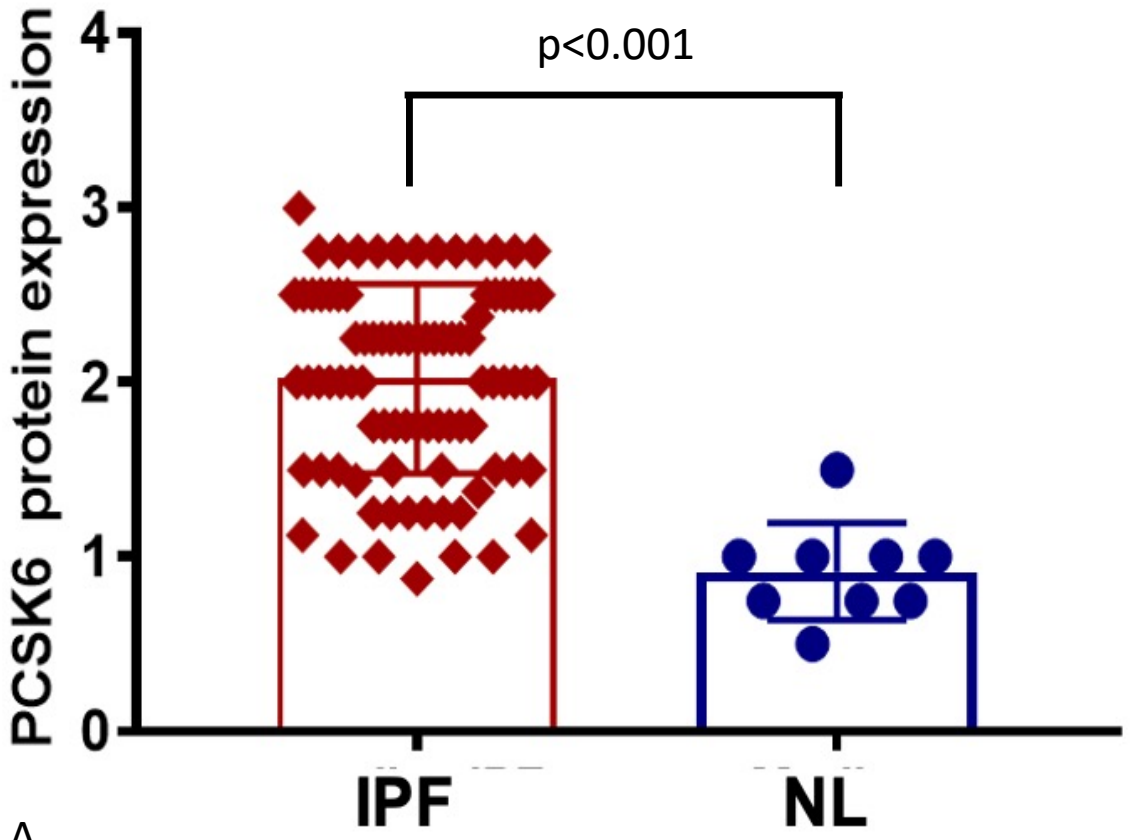
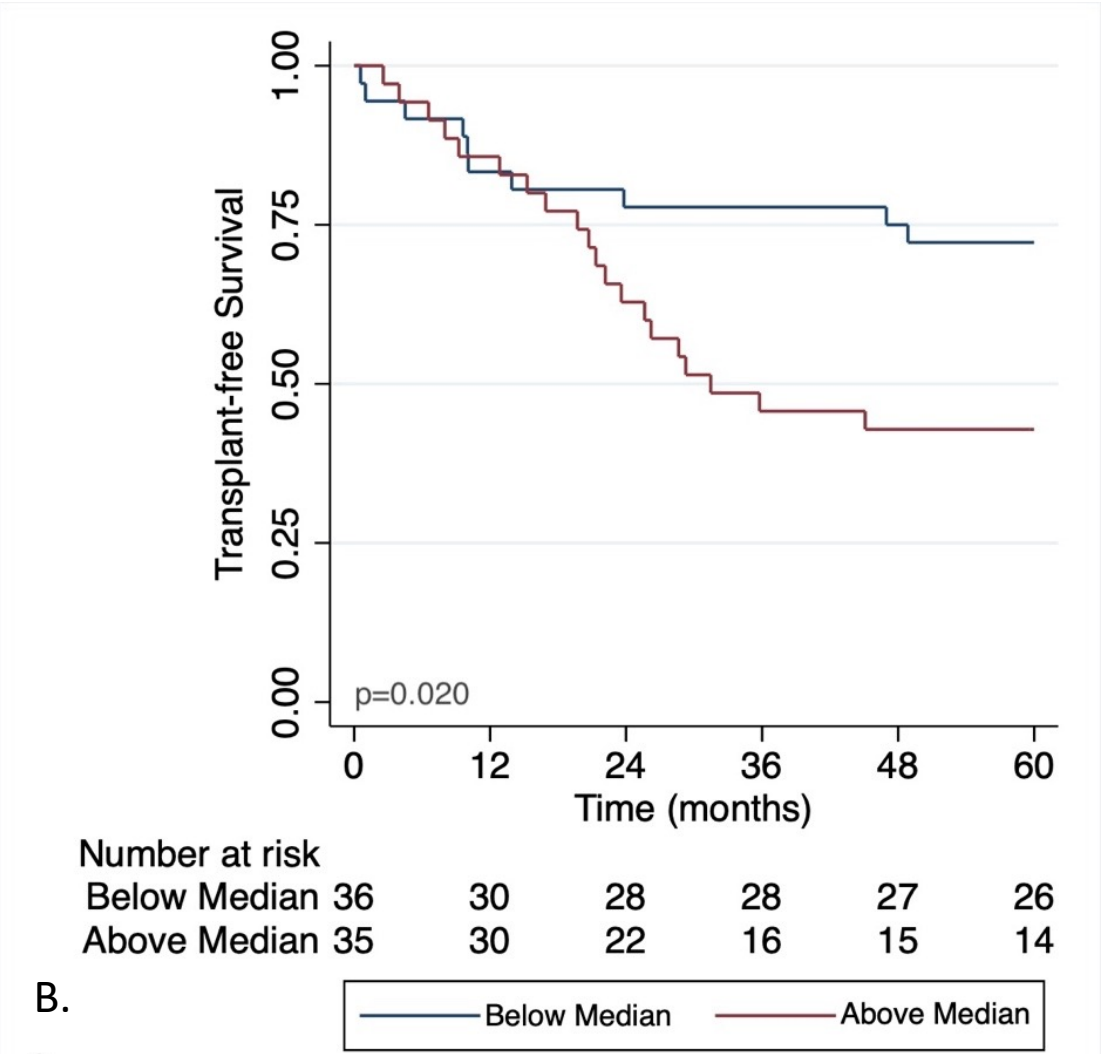
Figure 3



A.



B.

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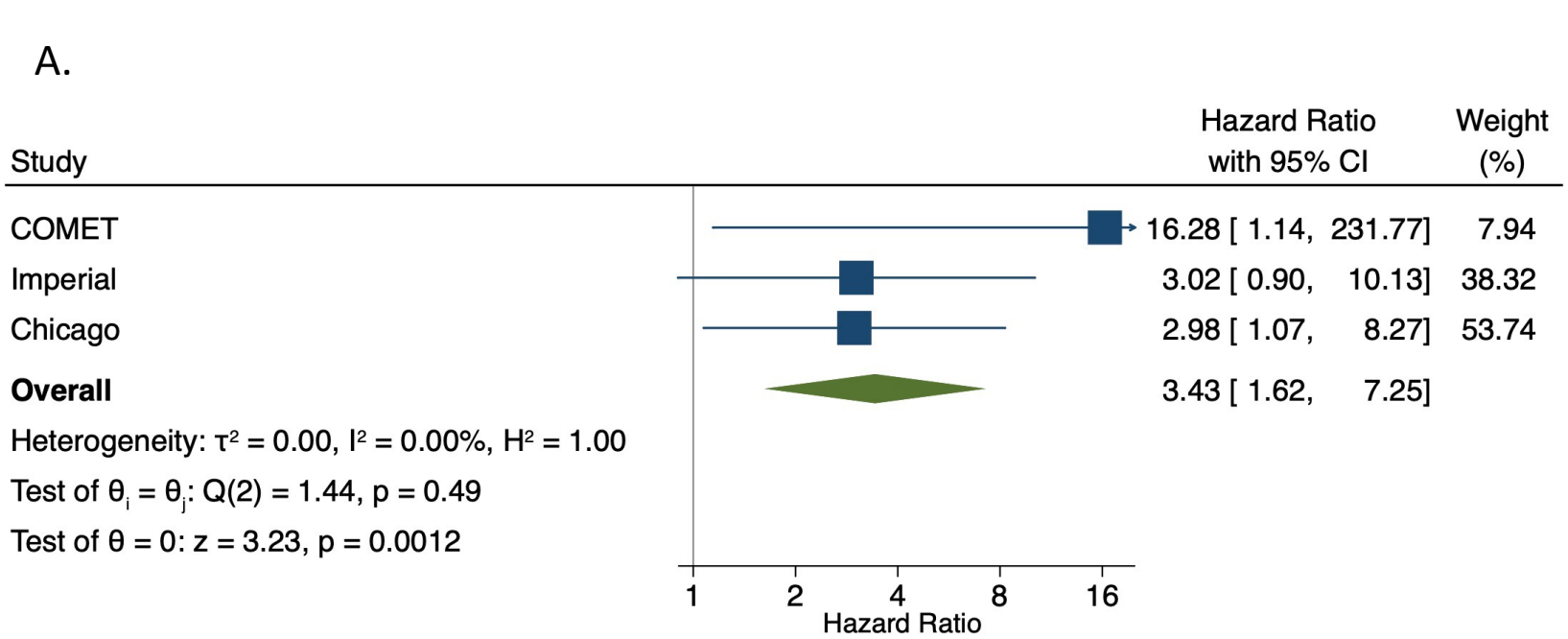
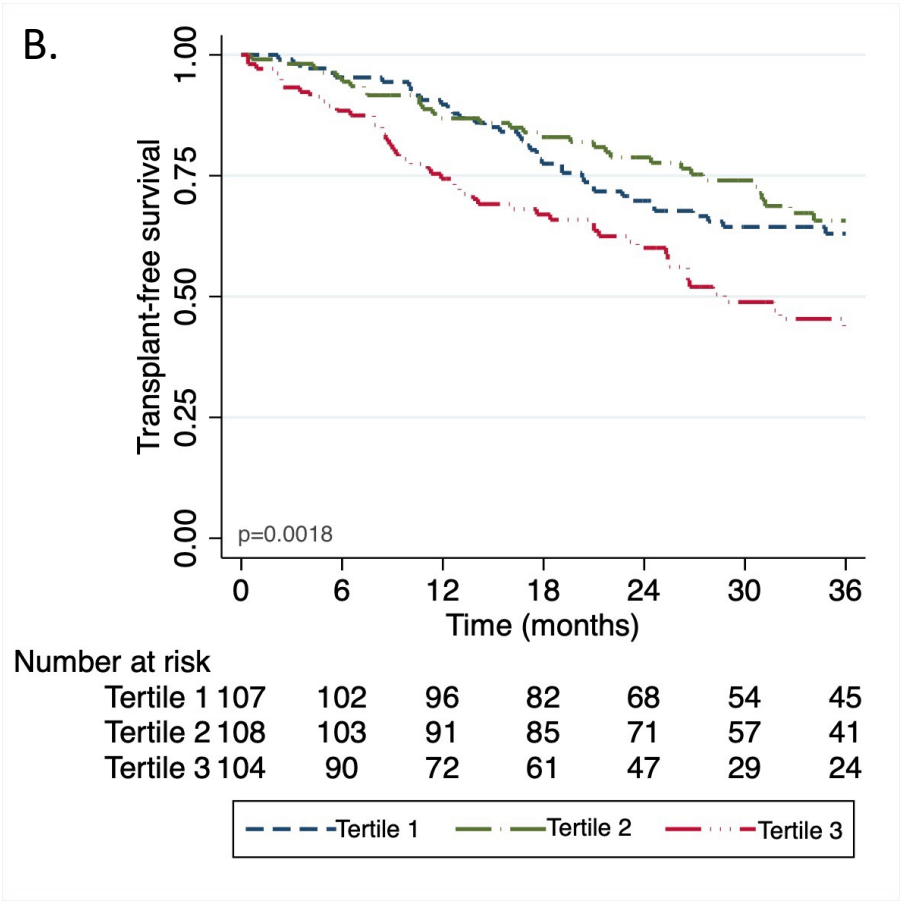
Figure 4





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PCSK6 and Genomic Determinants of Idiopathic Pulmonary Fibrosis Survival

Justin M. Oldham MD, Richard J. Allen PhD, Jose M. Lorenzo-Salazar MSc, Philip L. Molyneaux MBBS, Shwu-Fan Ma PhD, Chitra Joseph PhD, John S. Kim MD,Beatriz Guillen-Guio PhD, Tamara Hernández-Beeftink MSc, Jonathan Kropski MD, Yong Huang MD, Cathryn T. Lee MD, Ayodeji Adegunsoye MD, Janelle Vu Pugashetti MD, Angela L. Linderholm PhD, Vivian Vo BS, Mary E. Strek MD, Jonathan Jou, Adrian Muñoz-Barrera; Luis A. Rubio-Rodriguez; Richard Hubbard MBBS, Nik Hirani MBBS, Moira K.B. Whyte MBBS, Simon Hart MBBS, Andrew G. Nicholson MBBS, Lisa Lancaster MD, Helen Parfrey MBBS, Doris Rassl MBBS, William Wallace MBBS, Eleanor Valenzi MD, Yingze Zhang PhD, Josyf Mychaleckyj DPhil,Amy Stockwell MS, Naftali Kaminski MD, Paul J. Wolters MD, Maria Molina-Molina MD, William A. Fahy, Fernando J. Martinez MD, Ian P. Hall BMBCh, Martin D. Tobin MBChB, Toby M. Maher BM, Timothy S. Blackwell MD, Brian L. Yaspan PhD, R Gisli Jenkins MBBS, Carlos Flores PhD, Louise V Wain PhD, Imre Noth MD

ONLINE DATA SUPPLEMENT

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Supplemental Methods

Genotyping Platforms

Genotyping of stage I US cohort cases was performed using the Affymetrix (Santa Clara, CA) Genome-Wide Human SNP 6.0 Array.
Genotyping of stage I UK cohort cases was performed using the Affymetrix (Santa Clara, CA) Genome-Wide Human BiLEVE Array.
Genotyping of stage I UUS cohort cases was performed using the Affymetrix (Santa Clara, CA) Axiom UK Biobank array, except for Spanish cases, which used the Affymetrix Axiom Spain Biobank array.
Genotyping of stage II Vanderbilt university cases was performed with whole genome sequencing.

Functional effects of survival-associated variants

In silico assessments were used to infer the biological effect of variants associated with TFS after stage II testing. Gene prioritization was based on Open Targets Genetics scores v22.02. Potential regulatory effects were then assessed for epigenetic mechanisms [HaploReg v4.1, RegulomeDB v2.0.3], long-distance genomic interactions [Hi-C Unifying Genomic Interrogator (HUGIn) v1, considering tissue-specific $P \leq$

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Bonferroni corrected thresholds to select interactions], tissue-specific cis eQTLs and DNase I sensitivity QTLs (dsQTLs) [GTEx v8, considering a tissue-specific $P \leq 0.05$ as threshold, and SNPDelScore]. Proxies (D'=1) of variants associated with TFS were investigated for their association with putative causal genes across all tissues using GTEx v8. Effect of survival associated variants on other traits was assessed using PhenoScanner v2 ($P<0.001$). All web-based tools were accessed on the 09/09/2022.

Survival association for IPF susceptibility-associated variants

To assess the association between IPF survival and variants previously linked to IPF-risk,[1-4] polygenic risk score (PRS) analysis was performed using PRSice[5] (v1.25) to determine genetic overlap between IPF risk and survival. An IPF susceptibility PRS was calculated using weights from a previous IPF risk GWAS[1] and tested for its association with TFS in individuals in the UUS study who were not included in the IPF risk GWAS. Independent variants (selected through LD clumping with $r^2 \leq 0.1$) associated with IPF risk at $P \leq 0.001$[5] comprised the initial PRS with threshold adjustments made to identify the PRS that explained the highest proportion of IPF risk. The final PRS was then tested for TFS association with the aforementioned Cox proportional hazards regression model using the survival package in R v3.5.3. Given its large effect on IPF risk, analyses were then repeated excluding variants near the *MUC5B* promoter polymorphism (within 500 kb of rs35705950).

PCSK6 Immunohistochemistry

The tissue samples were obtained after informed consent and local ethics approval (South East Scotland SAHSC Bioresource-reference number 06/S1101/41; Brompton Node samples- reference number 15/SC/0101; Papworth Node Samples reference number 08/H0304/56+5; non-diseased controls-reference number (Q)GM030404 and Nottingham BRC samples- reference number 08/H0407/1). Formalin-Fixed Paraffin Embedded (FFPE) tissue samples were cut, 5µm thick, on positively charged Leica Surgipath X-tra slides. IHC staining was performed using the Novocastra Novolink™ Polymer Detection Systems kit (RE7280-K, Leica, Biosystems, Newcastle, UK) as previously described.[6] In brief, tissue sections were deparaffinised with xylene and rehydrated through 100% ethanol. Heat-induced (pH=6) citrate antigen retrieval was performed and rabbit PCSK6 antibody (HPA004774, Atlas antibodies, Sweden; 1:200 dilution of stock antibody) was incubated overnight at 4°C. 3-3′ Diaminobenzidine tetrahydrochloride (Novolink DAB substrate buffer plus) was used as the chromogen. Slides were counterstained with Novolink haematoxylin for 6 min, dehydrated and cover slipped. Normal kidney tissue was used as a positive tissue control, whereas no primary antibody was used as a negative control.

PCSK6 Western blotting

Cell Lysis Buffer (Cell Signalling, USA) supplemented with protease inhibitor cocktail (Sigma, USA) was used to collect the cell protein and the western blotting protein concentrations were determined by BCA assay using a commercially available kit (Thermo Fisher Scientific, Waltham, MA), according to the manufacturer's instructions. 20 µg protein were loaded per lane of a 4-12%, pre-cast Bis-Tris gradient gels (Thermo Fisher Scientific, Waltham, MA), subject to electrophoresis, and transferred onto a nitrocellulose membrane (Merck, GE10600002). Membranes were blocked for 1 h in 5% non-fat milk in tris-buffered saline containing 0.1% Tween, pH=7.4 (TBST). Membranes were incubated with rabbit anti-PCSK6 (HPA004774, Atlas antibodies, Sweden–1:1000 dilution of stock antibody) diluted in the blocking buffer for overnight at 4°C. A loading control of mouse Anti-β-Actin was also used to demonstrate protein loading (Merck; A5441; Mouse monoclonal- Anti-β-Actin antibody at 1:100000 dilution of stock antibody). Following day membranes was washed in TBST, incubated with an anti-mouse-HRP and anti-rabbit-HRP conjugated

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secondary antibodies (Dako, USA) at 1:2500 for 1hr at room temperature. Visualization was performed with Clarity Max™ ECL Substrate (Biorad, UK) on a Licor C-DiGit.

PCSK6 Gene Expression Testing

Messenger RNA was extracted from peripheral blood mononuclear cells in the COMET nad Chicago cohorts and from whole blood in the Imperial College cohort. Transcriptomic data were generated for the COMET cohort using the Affymetrix PrimeView Array, for th Imperial College cohort using the Affymetrix Human Gene 1.1 ST Array and for the UChicago cohort using bulk peripheral blood mononuclear cells RNA sequencing.

PCSK6 Plasma Testing

Stored frozen plasma in ethylenediaminetetraacetic acid aliquots from UC-Davis (n=187) and UChicago (n=139) were thawed and processed at UC-Davis in institutional batches. PCSK6 concentration was determined by enzyme-linked immunosorbent assay (ELISA) using a commercially available kit purchased from MyBioSource (San Diego, CA). The kit was run according to the manufacturer's instructions. Briefly, standards, undiluted plasma samples and a horseradish peroxidase-conjugated detection antibody were added to 96-well plates pre-coated with capture antibody before incubation for 1 h at 37°C. The wells were then washed and developed with chromogen solution included with the kit and immediately read at 450 nM using an ELISA plate reader. Data are reported as ng/mL. Intra- and inter-assay variability was controlled for using control standards on each plate. Protein concentration was log transformed for outcome modeling.

PCSK6 Protein Staining Intensity

The immunohistochemically stained slides were scanned using a ScanScope XT Slide Scanner (LeicaAperio Technologies, Vista, CA, USA) under 20x objective magnification (0.5 µm resolution) using Pannoramic Viewer (3DHISTECH Ltd Budapest, Hungary) slide viewing software. Both the percentage of staining and staining intensity of PCSK6 expression in lung sections were individually assessed. For PCSK6 quantification, the following scoring system of seven high-power fields at X40 per tissue section were used:

- **Score 0:** No cells stained
- **Score 0.5**: 1-10 cells stained at low intensity
- **Score 1.0:** 1-10 cells stained at high intensity
- **Score 1.5:** 11-25 cells stained at low intensity
- **Score 2.0:** 11-25 cells stained at high intensity
- **Score 2.5:** ≥26 cells stained at low intensity
- **Score 3.0:** ≥26 cells stained at high intensity

To normalize for varying numbers of regions of interest per slide, the mean score per slide was calculated. A Mann-Whitney U-test was used to compare between IPF and control lung samples. Statistical analysis was performed using GraphPad Prism 7.0 (GraphPad Software, San Diego, CA, USA).

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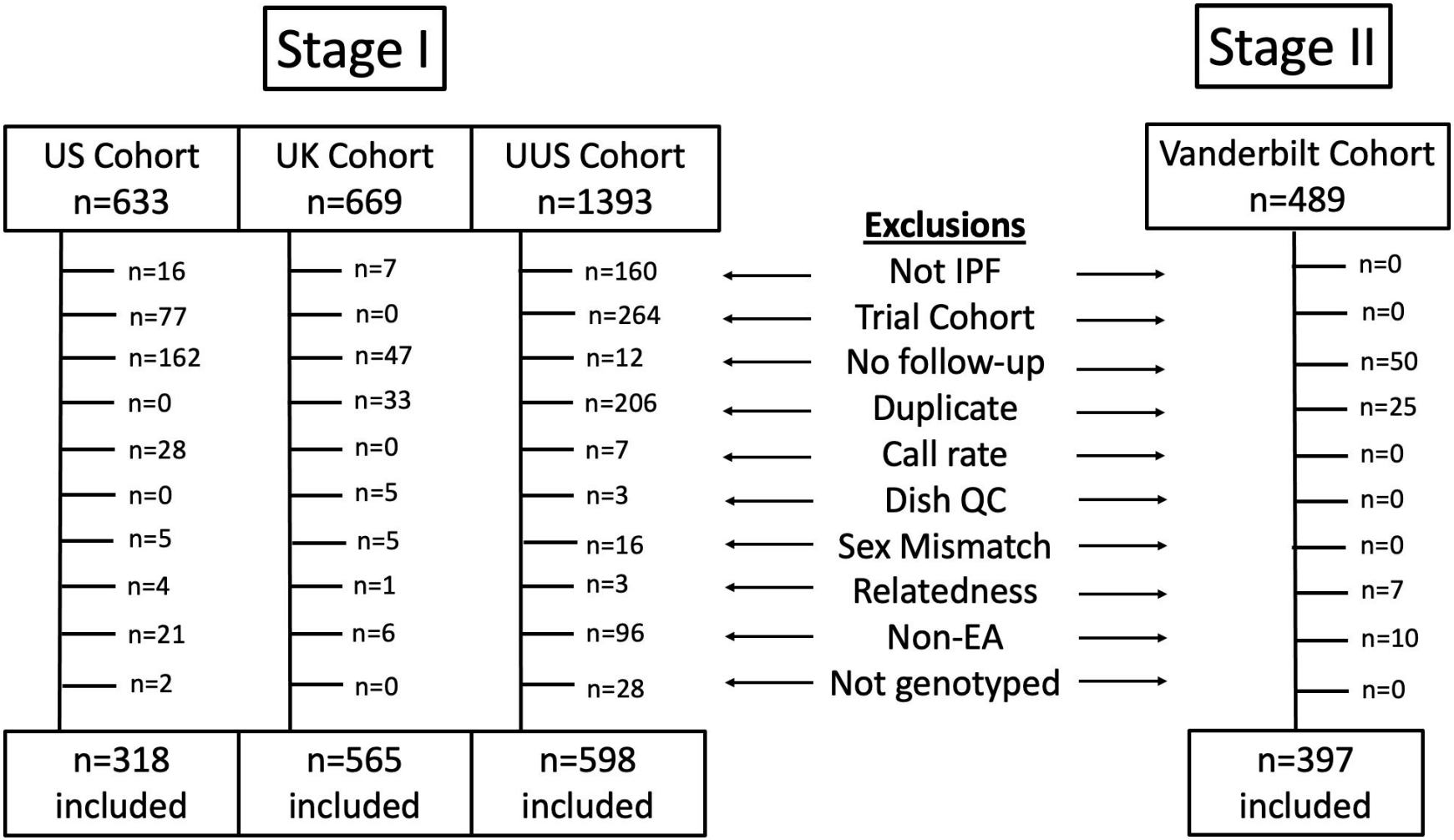

Figure E1. Quality control and filtering results

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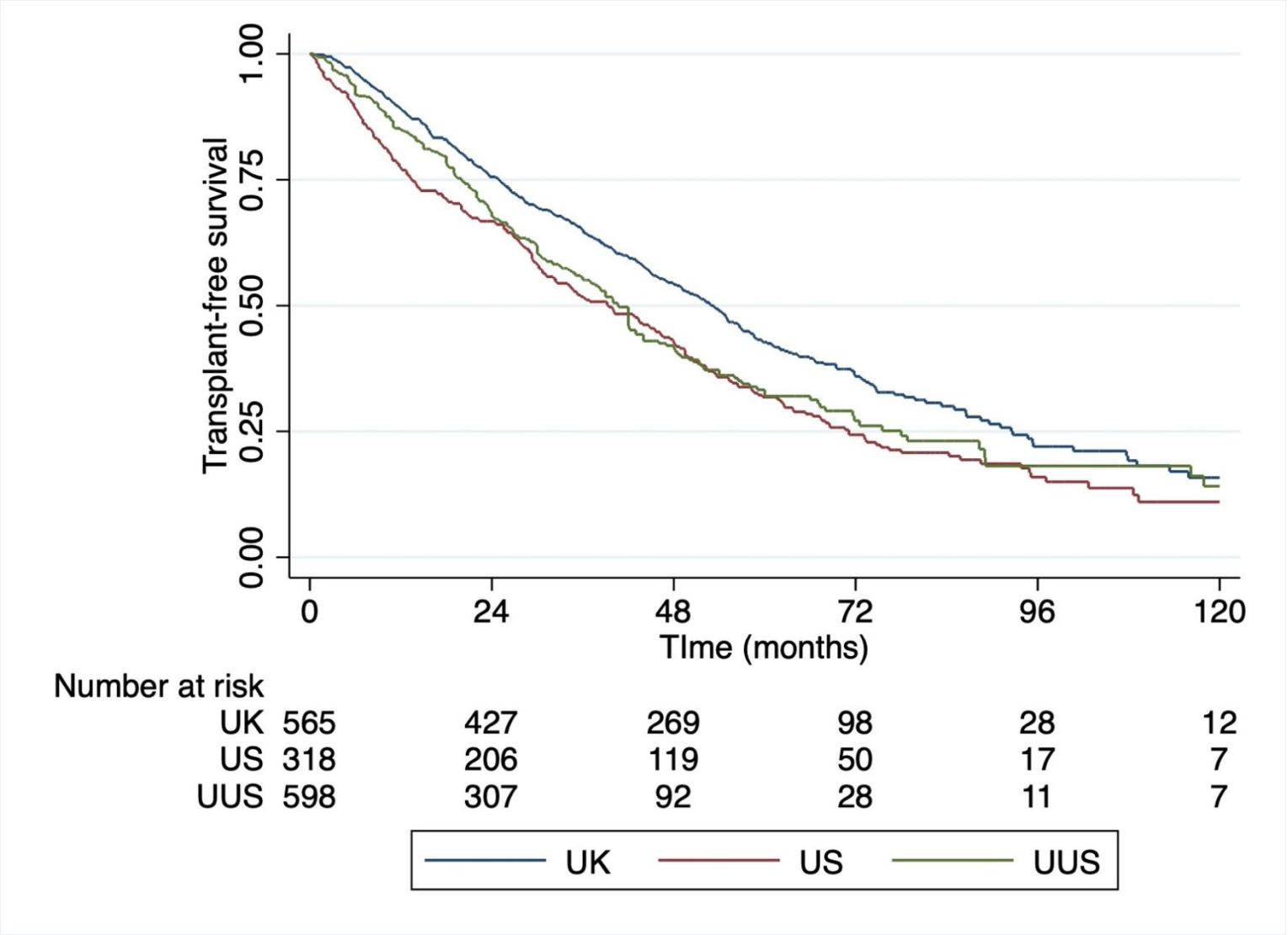

Figure E2. Kaplan-Meier survival plot for US, UK and UUS patients included in GWAS stage I. Median survival was highest in the UK cohort followed by UUS and US cohorts ($p_{logrank}$=0.001).

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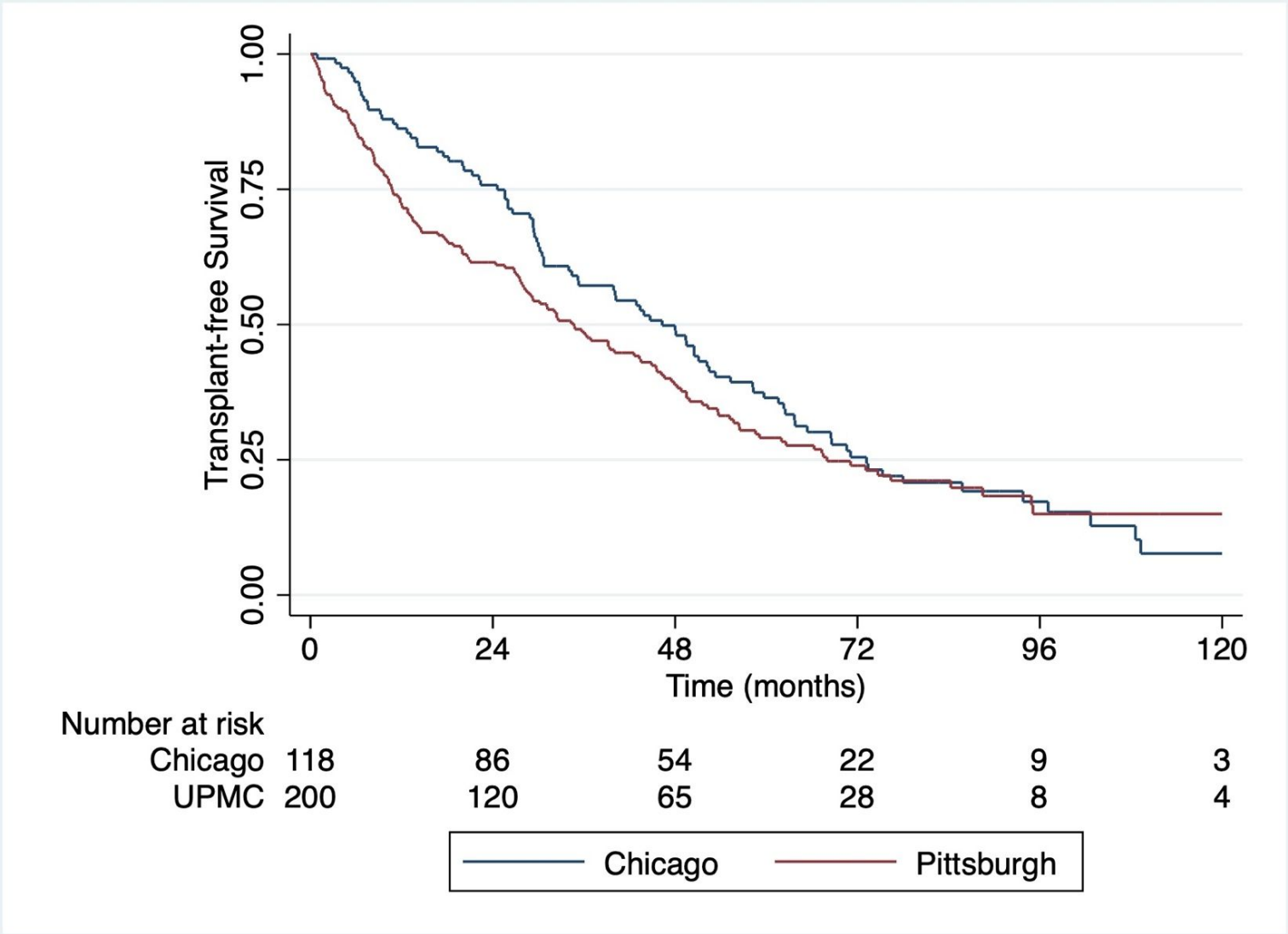

Figure E3. Kaplan-Meier survival plot for IPF cohorts within US dataset. Survival was similar between Chicago and UPMC cohorts ($p_{logrank}$=0.22).

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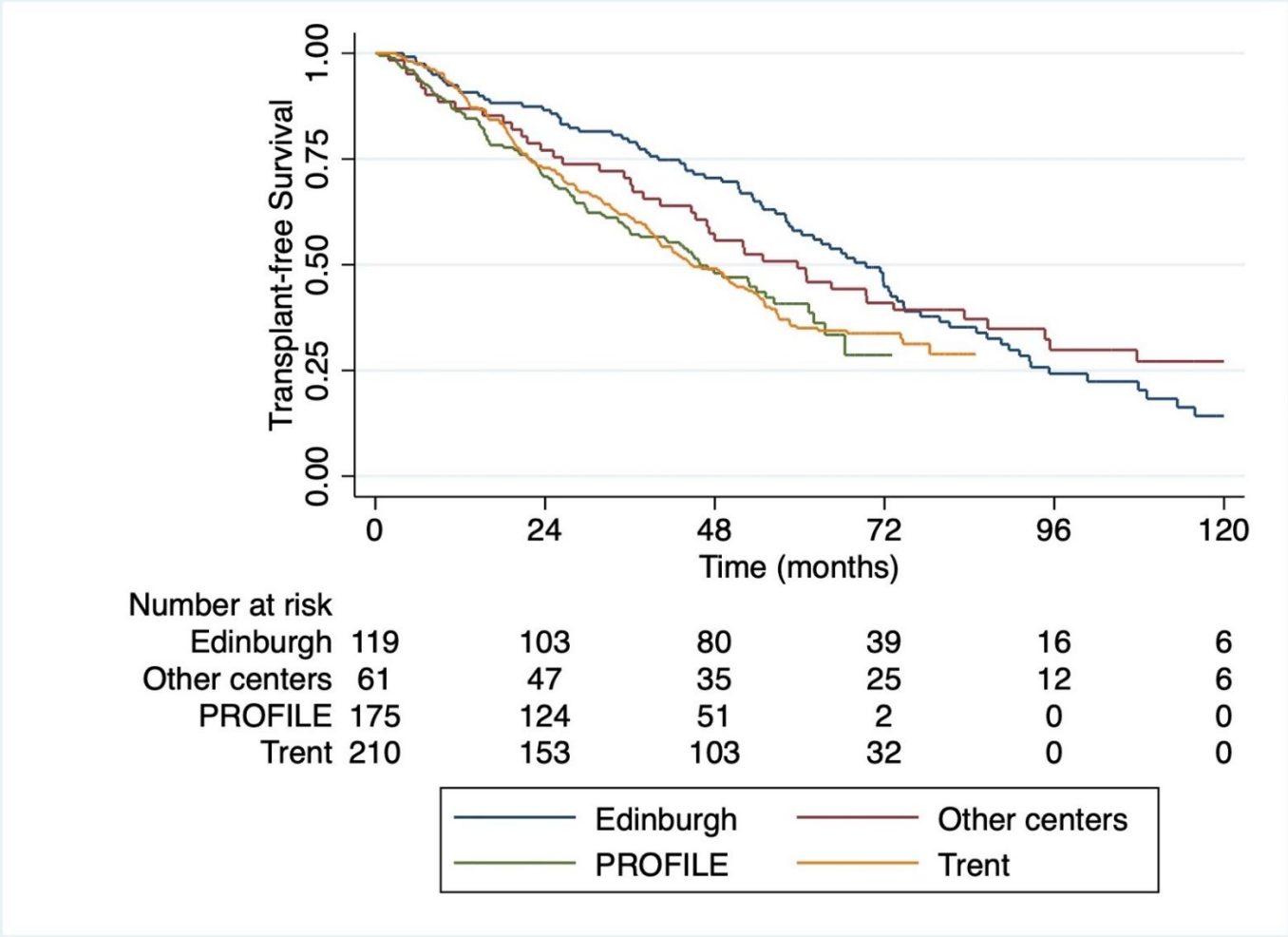

Figure E4. Kaplan-Meier survival plot for IPF cohorts within UK dataset. Median survival was highest in the Edinburgh cohort followed by smaller UK centers and PROFILE and Trent cohorts ($p_{logrank}$=0.0084).

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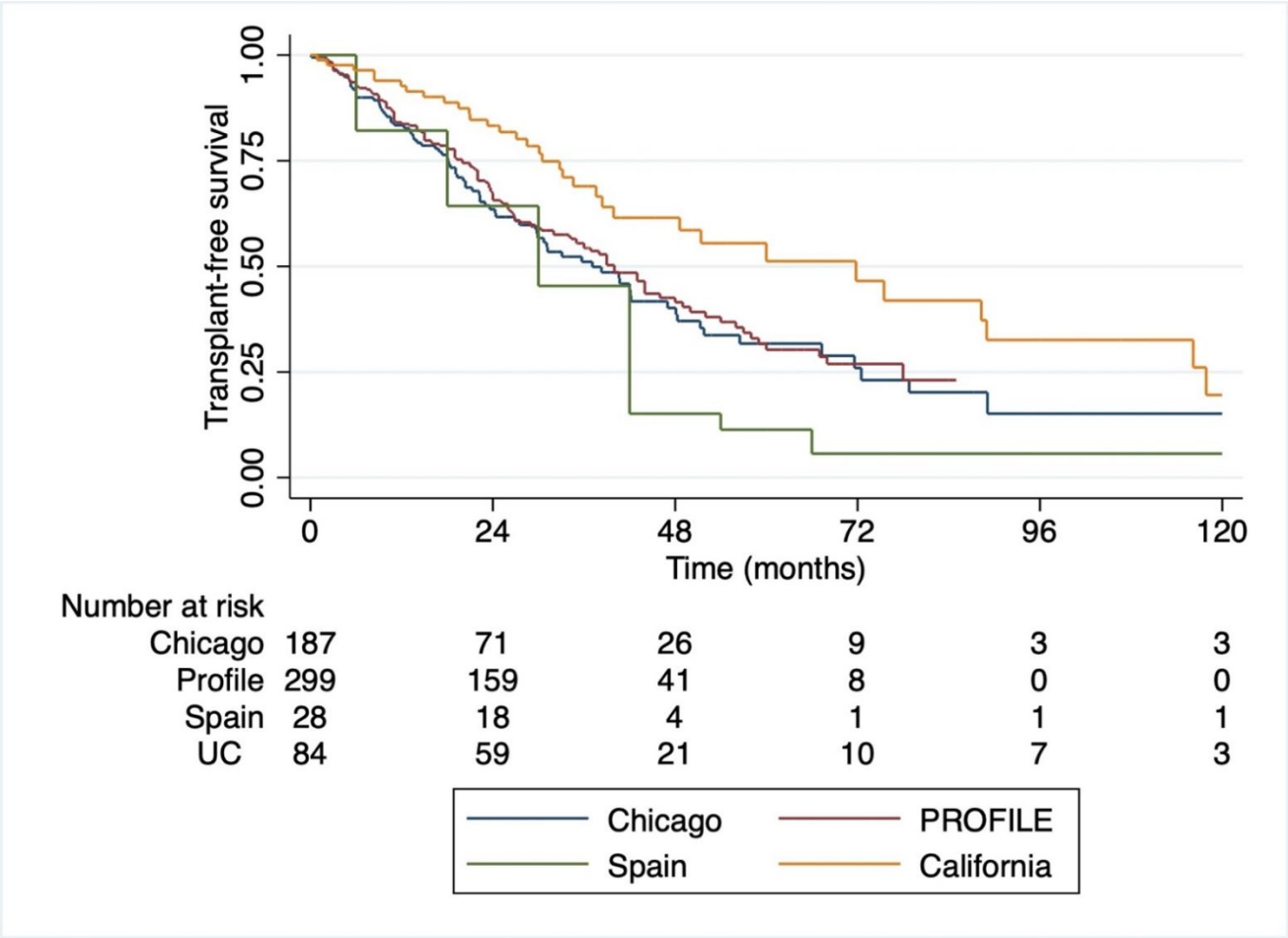

Figure E5. Kaplan-Meier survival plot for IPF cohorts within UUS dataset. Median survival was highest in the California cohort followed by Chicago, PROFILE and Spain cohorts ($p_{logrank}$=0.0025). PROFILE and Chicago cases are independent of those included in the UK and US cohorts, respectively.

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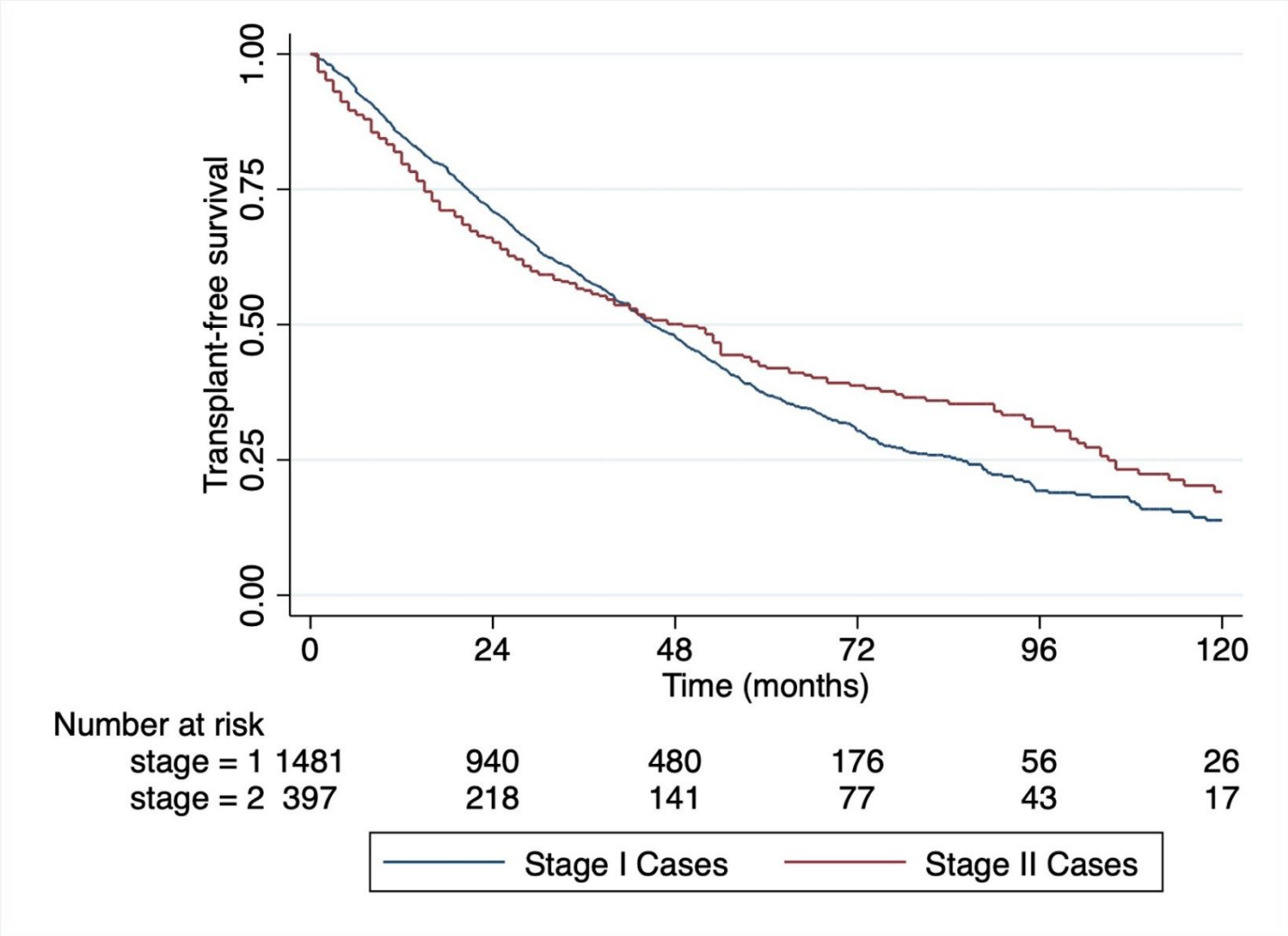

Figure E6. Kaplan-Meier survival plot for patients included in GWAS stage I and II cohorts. Survival was similar between stage I and II cases ($p_{logrank} = 0.27$)

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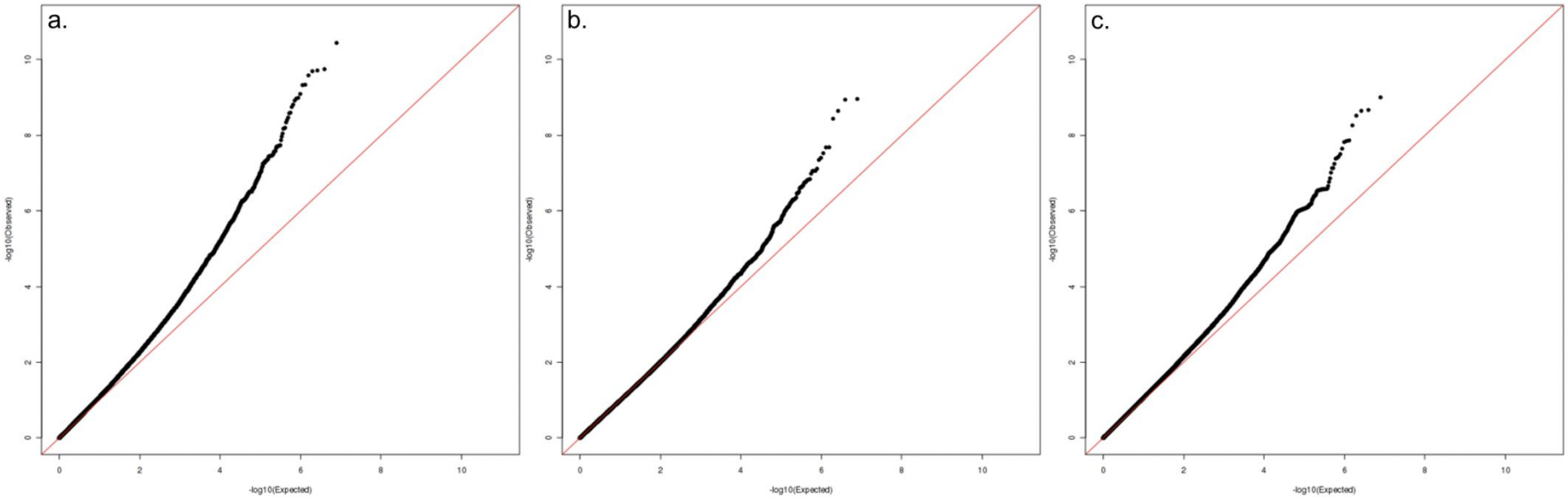

Figure E7. Quantile-quantile plots for US (λ=1.014) (a), UK (λ=0.996) (b) and UUS (λ=1.082) (c) cohorts.

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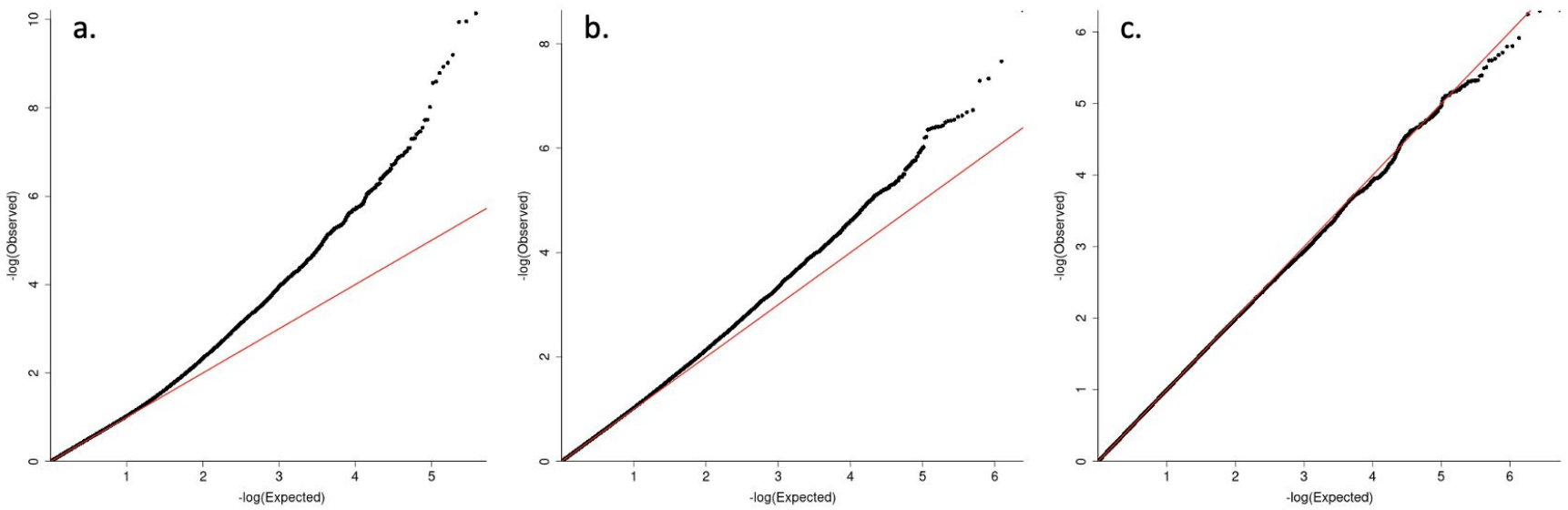

Figure E8. Quantile-quantile plots for minor allele frequency (MAF) bins, including MAF<1% (a) (λ=1.05), MAF 1-5% (b) (λ=1.02) and MAF >5% (c) (λ=1.00).

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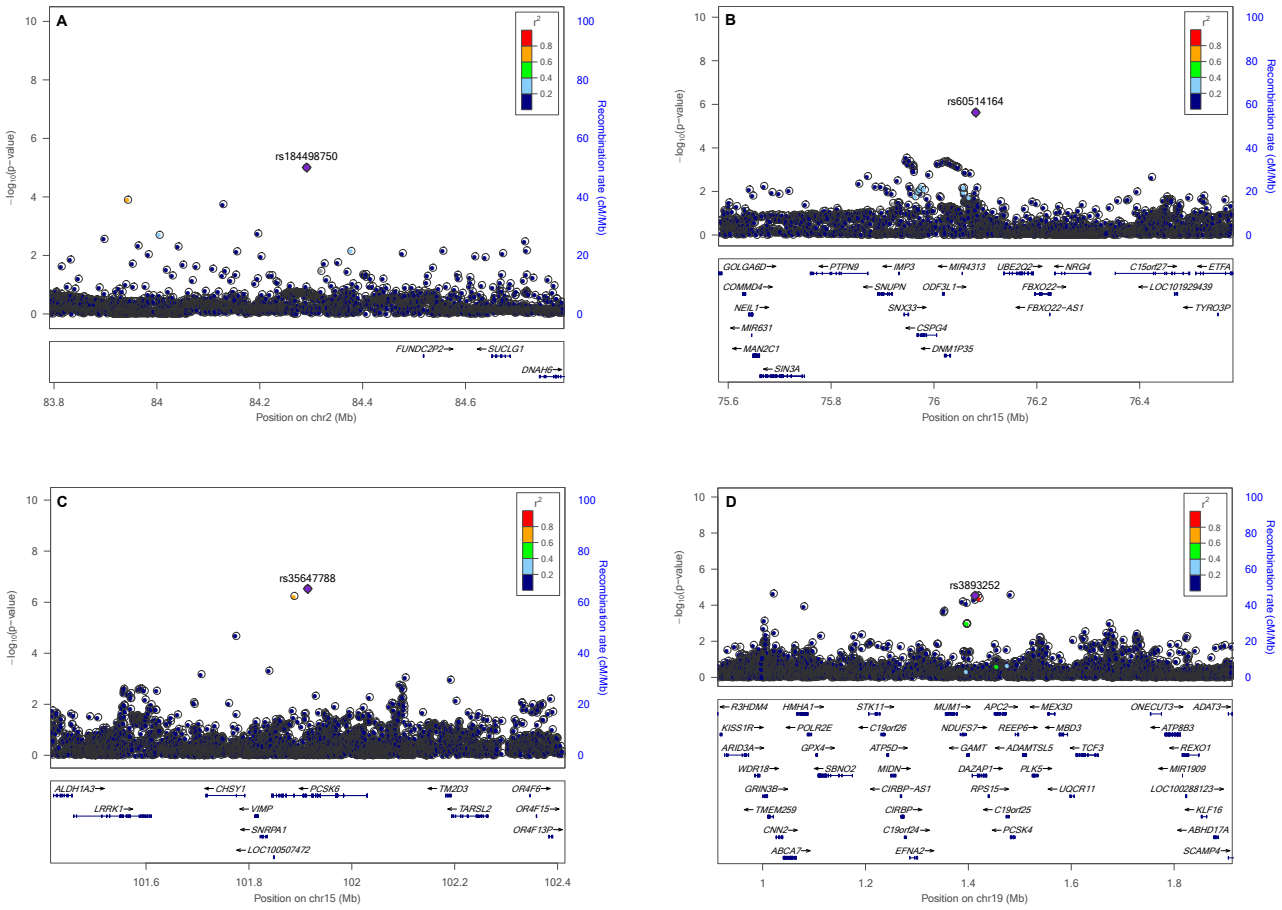

Figure E9. Regional plots for top variants after stage II meta-analysis. Y-axis displays -log10 transformed p-value and x-axis shows the hg19 genomic position. Estimated recombination rates (light blue line) are plotted on the right y-axis. The results for the remaining SNPs are color coded to reflect their degree of linkage disequilibrium with the leading SNP (indicated) based on pairwise r^2 values in Europeans.

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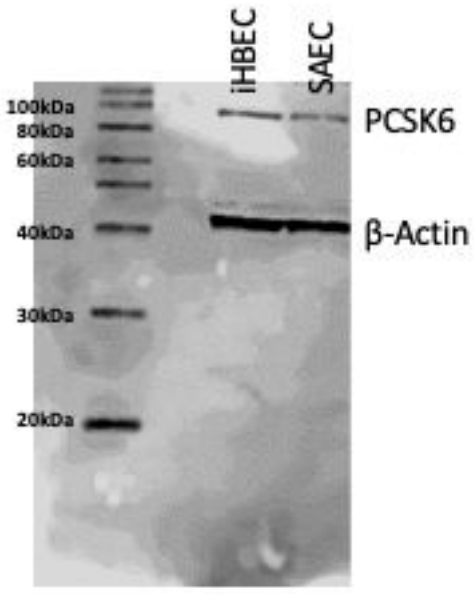

Figure E10. PCSK6 Western Blot. Single bands for PCSK6 (HPA004774) in the immortalized human bronchial epithelial cells (iHBECs) and small airway epithelial cells (SAEC) cells, at molecular weight of 100kDa, β-Actin (A544) was used as a loading control around 42kDa.

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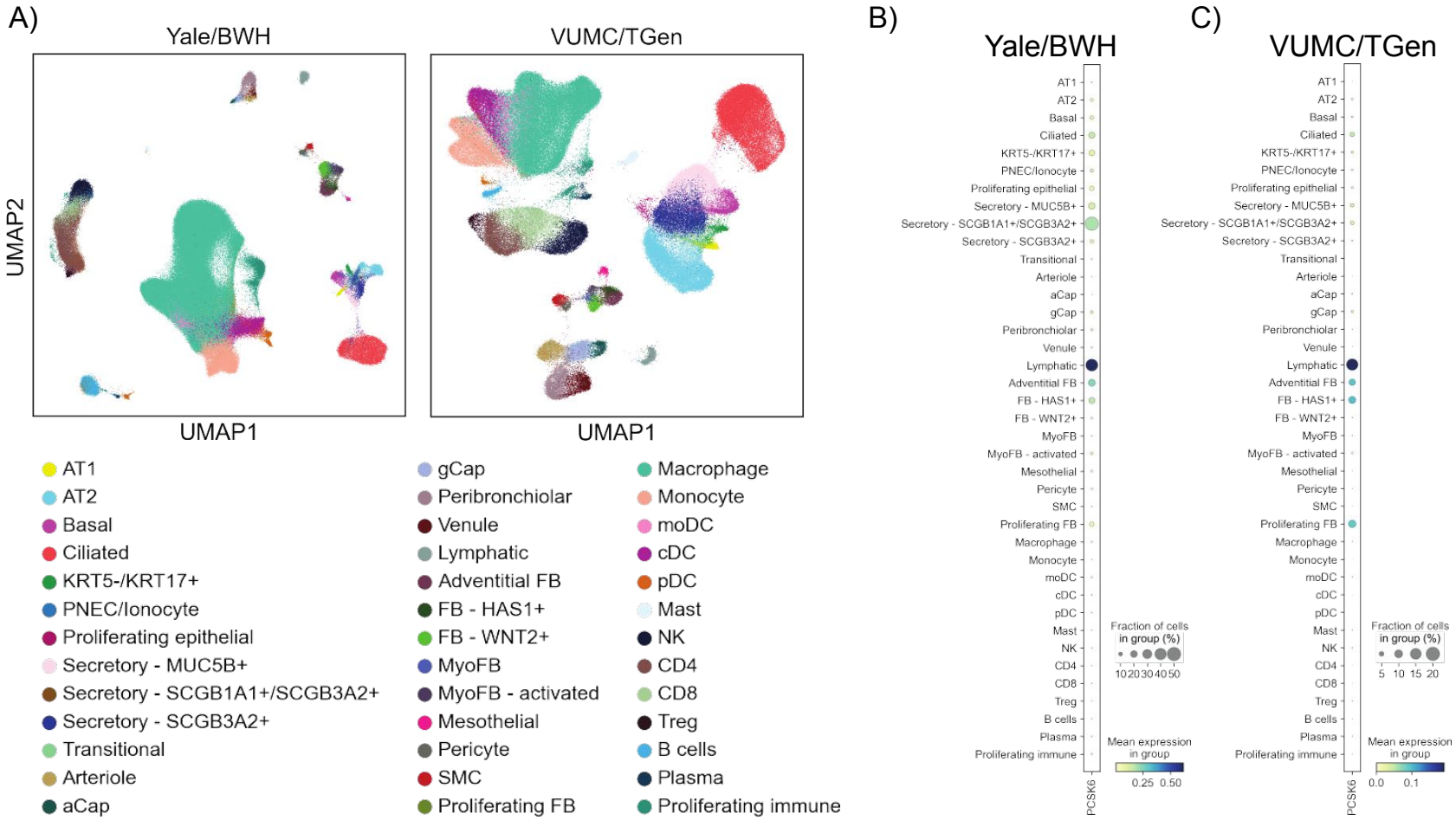

Figure E11. Cell-type specific expression of *PCSK6* in control and IPF lungs. IPF and control single-cell RNA-sequencing (scRNA-seq) data from two published datasets (Adams et al, *Science Advances 2020* (Yale/BWH) and Bui et al, *Nature Communications 2021* (VUMC/TGen) were reprocessed and cell-type annotation was performed using label transfer from a common reference dataset. A) Uniform manifold approximation and projection (UMAP) embedding of 215,093 cells from 32 control and 32 IPF lungs from Yale/BWH and 157,629 cells from 22 control and 27 IPF lungs from VUMC/TGen annotated by cell-type. Dotplots depicting cell-type specific expression of *PCSK6* from the Yale/BWH dataset (B) and the VUMC/TGen dataset (C).

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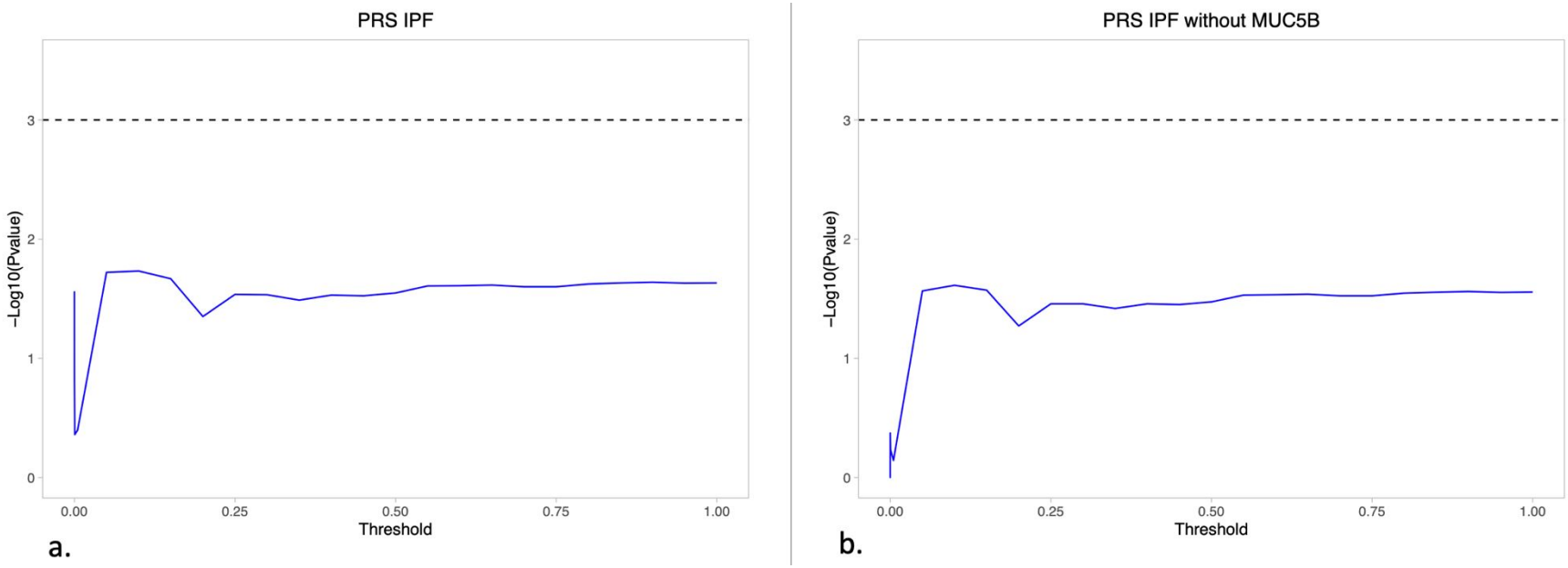

Figure E12. Strength of association between IPF risk polygenic risk score and transplant-free survival. The x axis shows the *p*-threshold used for determining which variants to include in the risk PRS calculation (i.e. as the value increases then more variants are included in the calculation) and the y axis shows the association of the PRS with TFS. The dotted line shows the significance threshold of p=0.001. Figure a) shows the association of the PRS when calculated using variants from across the genome, b) shows the strength of association when excluding the *MUC5B* region from the PRS calculation.

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Table E1. Stage I variants of nominal significance identified in the 3-way meta-analysis

Variant Location and Alleles					US cohort				UK cohort				UUS cohort				Stage-I meta-analysis	
Chr	Pos	SNP rsID	REF	EA	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	HR [95%CI]	P
1	10762349	rs7514663	C	T	NA	NA	NA	NA	0.91	0.0141	2.11 [1.16-3.84]	1.39E-2	0.92	0.0107	4.10 [2.01-8.37]	1.07E-4	2.74 [1.72-4.36]	2.15E-5
1	94606086	rs185926659	G	T	0.92	0.0050	6.68 [1.87-23.9]	3.44E-3	0.93	0.0072	5.24 [2.51-11.0]	1.05E-5	NA	NA	NA	NA	5.55 [2.91-10.6]	1.95E-7
1	100493151	rs148192333	T	C	NA	NA	NA	NA	0.93	0.0111	2.61 [1.36-5.02]	4.07E-3	0.89	0.0075	3.81 [1.63-8.92]	2.08E-3	2.98 [1.76-5.05]	4.75E-5
1	170154993	rs111749519	A	G	0.81	0.0124	2.7 [1.15-6.36]	2.30E-2	0.92	0.0094	2.91 [1.43-5.93]	3.31E-3	0.89	0.0080	2.68 [1.14-6.3]	2.32E-2	2.79 [1.74-4.47]	2.17E-5
1	188069617	rs190285902	C	G	NA	NA	NA	NA	0.87	0.0137	1.91 [1.03-3.56]	4.05E-2	0.85	0.0084	6.93 [2.95-16.3]	8.72E-6	2.92 [1.76-4.86]	3.69E-5
1	188151083	rs145551873	G	A	0.62	0.0117	4.72 [1.79-12.4]	1.66E-3	0.95	0.0226	1.88 [1.15-3.06]	1.15E-2	0.91	0.0125	2.75 [1.33-5.69]	6.43E-3	2.35 [1.61-3.45]	1.05E-5
1	200768521	rs11589092	G	A	0.97	0.0975	1.50 [1.08-2.07]	1.43E-2	0.98	0.0851	1.38 [1.07-1.77]	1.19E-2	0.97	0.0736	1.45 [1.06-1.99]	2.03E-2	1.43 [1.20-1.70]	4.68E-5
1	200861595	rs968549	C	T	0.95	0.9014	0.65 [0.47-0.89]	8.45E-3	1.00	0.9138	0.72 [0.56-0.92]	1.01E-2	1.00	0.9284	0.71 [0.52-0.98]	3.52E-2	0.7 [0.59-0.83]	4.29E-5
1	203327373	rs115684501	C	T	NA	NA	NA	NA	0.92	0.0082	3.08 [1.4-6.77]	5.01E-3	0.89	0.0059	6.94 [2.22-21.7]	8.55E-4	3.95 [2.05-7.61]	3.97E-5
1	214247966	rs115702239	C	A	NA	NA	NA	NA	0.86	0.0082	4.07 [1.71-9.68]	1.49E-3	0.83	0.0092	3.97 [1.82-8.67]	5.51E-4	4.02 [2.22-7.27]	4.36E-6
1	222319103	rs112891824	C	T	0.64	0.0100	4.97 [1.44-17.2]	1.11E-2	0.99	0.0159	2.26 [1.26-4.06]	6.53E-3	1.00	0.0159	2.19 [1.20-4.01]	1.04E-2	2.41 [1.60-3.62]	2.25E-5
1	242551121	rs34473966	A	C	NA	NA	NA	NA	0.89	0.0116	3.48 [1.85-6.55]	1.11E-4	0.88	0.0115	2.42 [1.12-5.22]	2.49E-2	3.03 [1.84-4.97]	1.24E-5
2	38114198	rs114698213	C	T	NA	NA	NA	NA	0.91	0.0126	2.97 [1.61-5.48]	4.79E-4	0.78	0.0146	2.36 [1.21-4.61]	1.17E-2	2.69 [1.70-4.26]	2.43E-5
2	38249347	rs150622385	G	A	NA	NA	NA	NA	0.85	0.0146	2.30 [1.25-4.25]	7.71E-3	0.80	0.0116	3.88 [1.94-7.73]	1.21E-4	2.87 [1.80-4.57]	9.72E-6
2	43602198	rs185306441	C	A	NA	NA	NA	NA	0.93	0.0137	2.01 [1.05-3.84]	3.51E-2	0.90	0.0120	4.41 [2.26-8.58]	1.28E-5	2.90 [1.80-4.65]	1.07E-5
2	47162883	rs115376562	T	C	NA	NA	NA	NA	0.88	0.0063	2.81 [1.24-6.38]	1.32E-2	0.85	0.0053	12.41 [4.81-32.0]	1.88E-7	5.15 [2.75-9.67]	3.34E-7
2	54643373	rs150765591	T	G	NA	NA	NA	NA	0.78	0.0106	2.87 [1.41-5.86]	3.73E-3	0.79	0.0105	4.21 [1.78-9.98]	1.09E-3	3.33 [1.9-5.82]	2.43E-5
2	84291167	rs184498750	G	T	NA	NA	NA	NA	0.86	0.0131	3.23 [1.76-5.92]	1.49E-4	0.86	0.0079	2.86 [1.20-6.81]	1.75E-2	3.11 [1.88-5.15]	9.83E-6
2	138337791	rs149029025	G	A	NA	NA	NA	NA	0.93	0.0069	3.82 [1.68-8.70]	1.41E-3	0.91	0.0055	4.04 [1.54-10.6]	4.45E-3	3.91 [2.07-7.38]	2.65E-5
2	142907569	rs142306180	C	A	0.92	0.0102	10.55 [4.16-26.8]	7.11E-7	NA	NA	NA	NA	0.95	0.0053	2.89 [1.03-8.16]	4.43E-2	5.88 [2.85-12.2]	1.63E-6
2	172941078	rs2357323	T	C	0.86	0.0053	4.51 [1.1-18.6]	3.68E-2	NA	NA	NA	NA	0.84	0.0089	4.57 [2.09-10.0]	1.44E-4	4.56 [2.23-9.32]	3.23E-5
2	172942149	rs56923699	G	A	0.86	0.0053	4.84 [1.17-20.0]	2.93E-2	NA	NA	NA	NA	0.85	0.0092	4.55 [2.09-9.93]	1.38E-4	4.62 [2.26-9.42]	2.59E-5
2	173111445	rs182271851	A	G	0.81	0.0085	3.08 [1.19-7.98]	2.09E-2	NA	NA	NA	NA	0.94	0.0051	8.84 [3.69-21.2]	1.05E-6	5.48 [2.79-10.8]	7.49E-7
2	226721440	rs377036225	G	A	NA	NA	NA	NA	0.89	0.0105	5.25 [2.45-11.3]	2.04E-5	0.87	0.0157	2.00 [1.06-3.78]	3.23E-2	3.03 [1.84-4.99]	1.39E-5

AJRCCM Articles in Press. Published February 13, 2023 as 10.1164/rccm.202205-0845OC

Table E1. Stage I variants of nominal significance identified in the 3-way meta-analysis

Variant Location and Alleles					US cohort				UK cohort				UUS cohort				Stage-I meta-analysis	
Chr	Pos	SNP rsID	REF	EA	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	HR [95%CI]	P
2	226758606	rs142266733	A	G	NA	NA	NA	NA	0.85	0.0118	4.16 [2.06-8.41]	6.95E-5	0.81	0.0135	2.35 [1.15-4.81]	1.95E-2	3.18 [1.91-5.31]	9.25E-6
2	240384770	rs188643417	G	A	NA	NA	NA	NA	0.83	0.0085	2.42 [1.07-5.49]	3.43E-2	0.88	0.0111	3.81 [2.02-7.18]	3.49E-5	3.19 [1.91-5.33]	9.73E-6
2	241253363	rs116377902	G	C	NA	NA	NA	NA	0.95	0.0070	3.82 [1.76-8.29]	6.75E-4	0.94	0.0071	2.74 [1.19-6.29]	1.73E-2	3.30 [1.85-5.87]	4.96E-5
2	241397273	rs75406448	A	G	0.67	0.0301	1.99 [1.04-3.81]	3.79E-2	0.97	0.0379	1.79 [1.22-2.62]	2.72E-3	0.97	0.0300	1.72 [1.10-2.69]	1.80E-2	1.80 [1.37-2.35]	2.29E-5
3	20681518	rs73177699	T	G	0.89	0.0174	2.16 [1.11-4.2]	2.30E-2	0.92	0.0122	2.46 [1.29-4.69]	6.07E-3	0.92	0.0068	4.56 [1.90-10.9]	6.75E-4	2.68 [1.76-4.07]	4.27E-6
3	59911730	rs77895452	C	A	0.82	0.0059	6.68 [2.13-21.0]	1.13E-3	NA	NA	NA	NA	0.91	0.0050	4.49 [1.55-13.0]	5.52E-3	5.39 [2.39-12.10]	4.91E-5
3	69741636	rs139644937	T	A	0.96	0.0062	7.63 [2.33-25.0]	7.84E-4	1.00	0.0071	2.74 [1.31-5.74]	7.68E-3	0.99	0.0071	2.59 [1.04-6.43]	4.08E-2	3.23 [1.91-5.48]	1.34E-5
3	75034442	rs142255892	C	T	NA	NA	NA	NA	0.78	0.0107	4.23 [2.07-8.64]	7.39E-5	0.71	0.0108	2.88 [1.30-6.36]	9.13E-3	3.59 [2.09-6.16]	3.54E-6
3	79693632	rs36022026	A	C	0.95	0.0616	1.83 [1.25-2.66]	1.76E-3	0.97	0.0675	1.47 [1.09-1.98]	1.19E-2	0.97	0.0722	1.4 [1.01-1.94]	4.30E-2	1.52 [1.25-1.85]	2.36E-5
3	112828302	rs2178398	C	G	0.99	0.1511	0.73 [0.56-0.95]	2.13E-2	0.96	0.1884	0.74 [0.60-0.91]	4.97E-3	0.97	0.1836	0.75 [0.59-0.94]	1.48E-2	0.74 [0.64-0.85]	2.02E-5
3	145344805	rs368666349	C	A	NA	NA	NA	NA	0.91	0.0069	2.25 [1.22-4.17]	9.74E-3	0.90	0.0096	4.04 [2.02-8.09]	7.91E-5	2.88 [1.80-4.60]	9.57E-6
4	11752169	rs191600377	T	C	NA	NA	NA	NA	0.81	0.0050	6.99 [2.34-20.9]	5.02E-4	0.85	0.0078	3.59 [1.65-7.79]	1.26E-3	4.54 [2.37-8.69]	5.15E-6
4	11982650	rs17260001	G	A	NA	NA	NA	NA	0.90	0.0056	8.38 [3.24-21.70]	1.18E-5	0.90	0.0073	3.22 [1.30-7.95]	1.13E-2	5.17 [2.65-10.10]	1.49E-6
4	12091599	rs192085162	G	A	NA	NA	NA	NA	0.90	0.0056	5.38 [2.21-13.05]	2.02E-4	0.92	0.0065	3.29 [1.26-8.59]	1.53E-2	4.33 [2.23-8.41]	1.51E-5
4	66572997	rs142199841	T	C	0.86	0.0084	3.62 [1.36-9.62]	9.86E-3	0.92	0.0054	4.87 [2.05-11.60]	3.35E-4	NA	NA	NA	NA	4.31 [2.22-8.34]	1.52E-5
4	74459567	rs79914686	T	C	NA	NA	NA	NA	0.82	0.0054	6.24 [2.24-17.40]	4.70E-4	0.81	0.0055	3.58 [1.24-10.3]	1.86E-2	4.82 [2.27-10.20]	4.12E-5
4	76495474	rs28641522	A	G	0.83	0.0067	11.56 [3.45-38.7]	7.14E-5	NA	NA	NA	NA	0.86	0.0061	3.17 [1.26-7.98]	1.44E-2	5.08 [2.36-10.9]	3.22E-5
4	122351715	rs6850444	G	C	0.90	0.3205	0.79 [0.64-0.97]	2.61E-2	0.99	0.3401	0.81 [0.69-0.96]	1.65E-2	0.98	0.3709	0.74 [0.62-0.89]	1.26E-3	0.78 [0.7-0.87]	1.14E-5
4	166645872	rs71618464	T	C	NA	NA	NA	NA	0.93	0.0100	3.19 [1.56-6.50]	1.46E-3	0.91	0.0074	4.62 [1.90-11.2]	7.11E-4	3.66 [2.08-6.44]	6.56E-6
4	175369612	rs139698405	C	T	NA	NA	NA	NA	0.81	0.0079	4.03 [1.57-10.40]	3.69E-3	0.81	0.0059	4.40 [1.76-11.00]	1.58E-3	4.21 [2.15-8.24]	2.69E-5
4	175401693	rs190563090	A	G	NA	NA	NA	NA	0.84	0.0093	2.77 [1.16-6.63]	2.23E-2	0.83	0.0053	11.41 [4.27-30.50]	1.20E-6	5.03 [2.59-9.78]	1.88E-6
5	36581565	rs138830243	C	T	0.82	0.0109	2.93 [1.1-7.81]	3.20E-2	0.86	0.0116	3.07 [1.57-5.98]	9.88E-4	0.84	0.0120	2.41 [1.10-5.27]	2.73E-2	2.81 [1.77-4.46]	1.11E-5
5	96796936	rs138577491	T	C	NA	NA	NA	NA	0.86	0.0063	5.85 [2.44-14.0]	7.56E-5	0.88	0.0061	3.95 [1.49-10.5]	5.92E-3	4.95 [2.55-9.62]	2.30E-6
5	106109149	rs571176768	G	C	0.92	0.0079	2.63 [1.04-6.63]	4.07E-2	0.97	0.0098	3.17 [1.62-6.22]	7.63E-4	0.95	0.0081	2.86 [1.27-6.44]	1.14E-2	2.95 [1.85-4.68]	4.77E-6
5	151623819	rs191657347	A	G	0.92	0.0069	3.74 [1.37-10.2]	1.02E-2	0.93	0.0054	4.51 [1.91-10.70]	6.06E-4	NA	NA	NA	NA	4.19 [2.15-8.16]	2.63E-5
5	154587673	rs536809954	G	A	NA	NA	NA	NA	0.89	0.0055	5.62 [2.38-13.30]	8.48E-5	0.86	0.0075	3.73 [1.54-9.06]	3.62E-3	4.65 [2.48-8.72]	1.75E-6

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Variant Location and Alleles					US cohort				UK cohort				UUS cohort				Stage-I meta-analysis	
Chr	Pos	SNP rsID	REF	EA	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	HR [95%CI]	P
5	154820302	rs13172001	G	A	NA	NA	NA	NA	0.87	0.0120	3.35 [1.77-6.36]	2.16E-4	0.87	0.0094	2.37 [1.12-5.02]	2.38E-2	2.92 [1.78-4.79]	2.26E-5
6	26118992	rs116594669	C	G	0.76	0.0107	3.52 [1.36-9.09]	9.33E-3	0.89	0.0080	3.57 [1.53-8.37]	3.35E-3	0.89	0.0143	2.02 [1.00-4.06]	4.90E-2	2.77 [1.71-4.49]	3.71E-5
6	75389935	rs145442342	G	A	0.74	0.0135	2.69 [1.18-6.11]	1.84E-2	0.93	0.0102	2.43 [1.20-4.93]	1.38E-2	0.92	0.0062	4.27 [1.66-11.0]	2.67E-3	2.86 [1.77-4.62]	1.65E-5
6	77044422	rs140590158	C	T	NA	NA	NA	NA	0.92	0.0206	2.61 [1.59-4.29]	1.53E-4	0.90	0.0190	2.2 [1.27-3.81]	4.87E-3	2.43 [1.67-3.53]	3.62E-6
6	84945776	rs116863455	G	A	NA	NA	NA	NA	0.90	0.0086	4.38 [2.17-8.85]	3.73E-5	0.86	0.0075	3.13 [1.20-8.15]	1.98E-2	3.92 [2.21-6.96]	3.16E-6
6	94466663	rs149057325	A	T	0.93	0.0288	1.81 [1.08-3.02]	2.35E-2	0.98	0.0262	2.00 [1.25-3.21]	3.86E-3	0.98	0.0289	1.62 [1.03-2.55]	3.58E-2	1.80 [1.36-2.39]	4.42E-5
6	94498243	rs143246710	C	T	0.94	0.0177	2.23 [1.18-4.24]	1.39E-2	0.97	0.0161	2.09 [1.19-3.65]	9.84E-3	0.97	0.0153	2.11 [1.17-3.81]	1.27E-2	2.14 [1.50-3.04]	2.38E-5
6	94531687	rs144768626	A	G	0.93	0.0176	2.3 [1.2-4.39]	1.17E-2	0.97	0.0168	2.08 [1.21-3.59]	8.30E-3	0.96	0.0159	1.96 [1.08-3.55]	2.63E-2	2.10 [1.48-2.97]	3.43E-5
6	102917669	rs139568075	C	T	NA	NA	NA	NA	0.80	0.0063	4.39 [1.81-10.6]	1.07E-3	0.80	0.0084	3.25 [1.44-7.35]	4.59E-3	3.75 [2.03-6.93]	2.35E-5
6	116041139	rs35483630	G	A	NA	NA	NA	NA	0.89	0.0060	4.47 [1.76-11.40]	1.64E-3	0.86	0.0098	3.72 [1.87-7.40]	1.76E-4	3.98 [2.26-7.03]	1.80E-6
6	116377677	rs181590625	G	A	NA	NA	NA	NA	0.90	0.0052	3.20 [1.11-9.24]	3.12E-2	0.88	0.0088	3.71 [1.81-7.59]	3.29E-4	3.54 [1.92-6.50]	4.89E-5
6	139248572	rs535860622	T	C	0.83	0.0068	3 [1.03-8.72]	4.35E-2	NA	NA	NA	NA	0.89	0.0058	8.84 [3.35-23.40]	1.10E-5	5.44 [2.57-11.50]	9.73E-6
6	143298800	rs149730644	C	T	0.85	0.0066	4.49 [1.33-15.2]	1.58E-2	NA	NA	NA	NA	0.95	0.0054	11.47 [4.50-29.30]	3.26E-7	8.13 [3.74-17.70]	1.19E-7
6	152786447	rs62427038	T	C	NA	NA	NA	NA	0.82	0.0086	3.44 [1.57-7.54]	1.97E-3	0.83	0.0117	2.95 [1.38-6.27]	5.08E-3	3.19 [1.83-5.55]	4.28E-5
6	164521835	rs188589004	G	A	NA	NA	NA	NA	0.82	0.0071	2.96 [1.35-6.48]	6.72E-3	0.83	0.0066	5.47 [2.13-14.10]	4.25E-4	3.76 [2.03-6.94]	2.34E-5
7	1065220	rs183625998	A	G	NA	NA	NA	NA	0.95	0.0054	6.22 [2.60-14.90]	3.95E-5	0.92	0.0055	4.54 [1.93-10.66]	5.13E-4	5.33 [2.86-9.93]	1.36E-7
7	1120020	rs192803195	C	T	NA	NA	NA	NA	0.91	0.0051	6.83 [2.68-17.4]	5.68E-5	0.90	0.0051	3.79 [1.6-9.01]	2.50E-3	5.03 [2.63-9.63]	1.04E-6
7	74953934	rs868964452	A	T	NA	NA	NA	NA	0.62	0.0180	3.21 [1.73-5.97]	2.19E-4	0.64	0.0125	2.21 [1.01-4.84]	4.68E-2	2.8 [1.71-4.59]	4.17E-5
7	75344084	rs117388086	C	A	NA	NA	NA	NA	0.91	0.0110	2.37 [1.19-4.71]	1.42E-2	0.91	0.0158	3.02 [1.67-5.46]	2.70E-4	2.71 [1.71-4.29]	2.23E-5
7	75358307	rs111489307	T	G	0.53	0.0137	3.19 [1.23-8.27]	1.68E-2	0.98	0.0104	3.33 [1.69-6.59]	5.34E-4	0.97	0.0092	2.83 [1.31-6.11]	7.96E-3	3.13 [1.98-4.96]	1.20E-6
7	101163592	rs149406562	C	A	NA	NA	NA	NA	0.93	0.0129	3.8 [2.10-6.87]	9.88E-6	0.94	0.0098	2.76 [1.30-5.85]	8.21E-3	3.38 [2.11-5.43]	4.26E-7
7	103029848	rs142639517	A	G	NA	NA	NA	NA	0.89	0.0082	4.02 [1.96-8.25]	1.47E-4	0.91	0.0050	2.95 [1.25-6.95]	1.35E-2	3.56 [2.03-6.23]	8.79E-6
7	125546344	rs74580469	T	C	NA	NA	NA	NA	0.93	0.0085	3.39 [1.61-7.13]	1.27E-3	0.88	0.0125	3.11 [1.50-6.45]	2.30E-3	3.25 [1.91-5.53]	1.36E-5
7	148106376	rs556475473	T	C	NA	NA	NA	NA	0.85	0.0062	5.33 [2.22-12.8]	1.82E-4	0.86	0.0052	5.17 [2.01-13.3]	6.39E-4	5.26 [2.73-10.10]	6.51E-7
7	152982958	rs73728959	G	A	NA	NA	NA	NA	0.99	0.0093	3.59 [1.70-7.57]	8.10E-4	0.96	0.0104	3.51 [1.60-7.66]	1.67E-3	3.55 [2.05-6.15]	6.53E-6
8	9496015	rs148904289	T	A	NA	NA	NA	NA	0.91	0.0053	3.54 [1.44-8.75]	6.06E-3	0.93	0.0078	3.58 [1.60-8.01]	1.87E-3	3.57 [1.93-6.59]	4.97E-5

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Table E1. Stage I variants of nominal significance identified in the 3-way meta-analysis

Variant Location and Alleles					US cohort				UK cohort				UUS cohort				Stage-I meta-analysis	
Chr	Pos	SNP rsID	REF	EA	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	HR [95%CI]	P
8	22381858	rs113465261	T	C	NA	NA	NA	NA	0.90	0.0060	2.90 [1.14-7.41]	2.58E-2	0.90	0.0051	5.20 [2.29-11.80]	8.49E-5	3.99 [2.12-7.50]	1.78E-5
8	23454598	rs193064386	G	T	NA	NA	NA	NA	0.87	0.0067	3.80 [1.69-8.55]	1.28E-3	0.88	0.0062	3.70 [1.53-8.92]	3.63E-3	3.75 [2.04-6.89]	2.01E-5
8	23626607	rs79562205	C	T	NA	NA	NA	NA	0.93	0.0075	3.64 [1.69-7.83]	9.58E-4	0.91	0.0097	2.91 [1.38-6.14]	4.93E-3	3.26 [1.89-5.63]	2.16E-5
8	80493124	rs116969973	G	A	NA	NA	NA	NA	0.94	0.0122	2.20 [1.22-3.96]	9.08E-3	0.88	0.0126	4.12 [2.02-8.38]	9.57E-5	2.80 [1.77-4.45]	1.21E-5
8	92675281	rs183828688	A	G	NA	NA	NA	NA	0.90	0.0072	7.55 [3.38-16.9]	8.31E-7	0.89	0.0065	3.35 [1.34-8.39]	9.84E-3	5.39 [2.91-9.98]	8.00E-8
8	120287198	rs117931584	A	G	0.72	0.0116	6.09 [2.43-15.3]	1.18E-4	NA	NA	NA	NA	0.84	0.0053	2.95 [1.07-8.11]	3.66E-2	4.37 [2.14-8.92]	4.93E-5
9	4064850	rs147994675	C	T	0.83	0.0113	2.47 [1.02-5.97]	4.43E-2	0.89	0.0107	2.12 [1.07-4.19]	3.15E-2	0.85	0.0130	3.11 [1.62-5.99]	6.68E-4	2.55 [1.66-3.91]	1.84E-5
9	7042171	rs190922348	C	T	0.82	0.0161	2.68 [1.31-5.47]	6.81E-3	0.92	0.0100	2.48 [1.26-4.88]	8.41E-3	0.93	0.0128	2.26 [1.05-4.86]	3.65E-2	2.48 [1.62-3.79]	2.84E-5
9	14343426	rs118165660	G	A	NA	NA	NA	NA	0.79	0.0257	2.53 [1.56-4.10]	1.74E-4	0.76	0.0184	2.08 [1.11-3.90]	2.26E-2	2.36 [1.60-3.48]	1.55E-5
9	24585027	rs113029495	G	A	0.87	0.0060	8.74 [2.78-27.5]	2.04E-4	0.94	0.0062	3.90 [1.74-8.73]	9.60E-4	NA	NA	NA	NA	5.01 [2.56-9.79]	2.45E-6
9	86260066	rs139230397	G	C	NA	NA	NA	NA	0.88	0.0056	4.67 [1.92-11.40]	6.63E-4	0.89	0.0089	3.63 [1.55-8.47]	2.92E-3	4.11 [2.20-7.70]	9.53E-6
9	92489004	rs187088797	C	G	NA	NA	NA	NA	0.88	0.0093	3.16 [1.59-6.29]	1.04E-3	0.91	0.0077	2.89 [1.27-6.55]	1.12E-2	3.05 [1.79-5.21]	4.47E-5
10	4620992	rs142562867	G	A	0.91	0.0112	3.48 [1.48-8.19]	4.34E-3	0.97	0.0160	1.9 [1.13-3.20]	1.55E-2	0.96	0.0142	2.40 [1.19-4.84]	1.42E-2	2.26 [1.54-3.32]	2.96E-5
10	11875431	rs111589468	G	A	0.84	0.0058	7 [2.02-24.3]	2.14E-3	NA	NA	NA	NA	0.89	0.0103	3.33 [1.63-6.78]	9.42E-4	3.99 [2.09-7.60]	2.60E-5
10	43302339	rs139494937	A	T	NA	NA	NA	NA	0.93	0.0170	2.41 [1.37-4.23]	2.19E-3	0.96	0.0086	3.88 [1.79-8.41]	5.70E-4	2.82 [1.78-4.47]	1.05E-5
10	73718436	rs186597433	C	T	0.72	0.0110	3.73 [1.48-9.37]	5.16E-3	NA	NA	NA	NA	0.95	0.0065	5.69 [2.45-13.20]	5.38E-5	4.70 [2.45-9.02]	3.14E-6
10	87849922	rs191887805	C	T	NA	NA	NA	NA	0.87	0.0063	5.87 [2.44-14.10]	7.84E-5	0.85	0.0056	2.66 [1.07-6.58]	3.47E-2	4.06 [2.14-7.73]	1.93E-5
10	120582311	rs117054238	T	C	NA	NA	NA	NA	0.84	0.0081	2.33 [1.10-4.94]	2.67E-2	0.81	0.0058	9.16 [3.39-24.8]	1.25E-5	3.74 [2.04-6.86]	2.08E-5
11	14115243	rs78904863	T	A	0.87	0.0226	3.46 [1.88-6.37]	6.56E-5	0.95	0.0197	1.69 [1.02-2.79]	4.04E-2	0.95	0.0257	1.70 [1.07-2.69]	2.45E-2	1.99 [1.47-2.70]	9.54E-6
11	28663945	rs111945608	T	C	0.70	0.0142	5.14 [2.34-11.3]	4.66E-5	0.91	0.0210	1.64 [1.00-2.70]	4.96E-2	0.93	0.0112	3.19 [1.62-6.26]	7.56E-4	2.45 [1.70-3.52]	1.43E-6
11	30638253	rs189350185	G	T	0.95	0.0075	4.81 [1.82-12.8]	1.58E-3	0.94	0.0073	3.01 [1.42-6.40]	4.04E-3	NA	NA	NA	NA	3.55 [1.94-6.51]	4.17E-5
11	33998481	rs76061348	T	C	NA	NA	NA	NA	0.89	0.0124	2.51 [1.33-4.73]	4.33E-3	0.91	0.0208	2.35 [1.37-4.04]	1.92E-3	2.42 [1.59-3.69]	3.75E-5
11	36108905	rs118080683	C	T	0.54	0.0146	2.9 [1.1-7.64]	3.14E-2	0.92	0.0153	3.01 [1.70-5.33]	1.64E-4	0.89	0.0165	2.20 [1.13-4.30]	2.11E-2	2.69 [1.79-4.04]	1.78E-6
11	44113701	rs186991973	T	C	0.87	0.0121	4.33 [1.99-9.43]	2.24E-4	NA	NA	NA	NA	0.92	0.0064	2.73 [1.16-6.42]	2.17E-2	3.51 [1.92-6.41]	4.52E-5
11	75214357	rs186259260	C	T	NA	NA	NA	NA	0.80	0.0053	3.64 [1.35-9.82]	1.07E-2	0.85	0.0076	5.00 [2.24-11.20]	8.46E-5	4.38 [2.31-8.31]	5.86E-6
11	81937642	rs72940008	C	G	NA	NA	NA	NA	0.85	0.0096	2.58 [1.31-5.11]	6.41E-3	0.83	0.0092	3.64 [1.71-7.74]	7.83E-4	2.99 [1.79-5.01]	3.00E-5

AJRCCM Articles in Press. Published February 13, 2023 as 10.1164/rccm.202205-0845OC

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Variant Location and Alleles					US cohort				UK cohort				UUS cohort				Stage-I meta-analysis	
Chr	Pos	SNP rsID	REF	EA	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	HR [95%CI]	P
11	123294624	rs118091577	G	T	NA	NA	NA	NA	0.86	0.0073	3.24 [1.75-6.03]	1.95E-4	0.86	0.0062	4.05 [1.75-9.37]	1.10E-3	3.49 [2.11-5.79]	1.22E-6
11	123312059	rs559811602	A	G	NA	NA	NA	NA	0.92	0.0057	8.99 [3.85-21.00]	4.04E-7	0.87	0.0051	3.57 [1.41-9.05]	7.35E-3	6.02 [3.18-11.40]	3.65E-8
11	126441296	rs189162333	A	G	0.57	0.0155	6.04 [2.37-15.4]	1.69E-4	0.67	0.0147	2.67 [1.33-5.35]	5.61E-3	NA	NA	NA	NA	3.50 [1.98-6.18]	1.51E-5
11	132893929	rs192803572	T	C	NA	NA	NA	NA	0.86	0.0091	4.23 [2.02-8.84]	1.29E-4	0.80	0.0061	3.4 [1.03-11.3]	4.50E-2	4.00 [2.12-7.54]	1.91E-5
11	134086356	rs140676527	G	A	0.83	0.0062	4.63 [1.48-14.5]	8.64E-3	0.92	0.0063	4.79 [2.06-11.20]	2.83E-4	NA	NA	NA	NA	4.74 [2.37-9.46]	1.05E-5
12	5522297	rs150598029	T	G	NA	NA	NA	NA	0.99	0.0053	5.98 [2.49-14.40]	6.51E-5	0.97	0.0078	2.36 [1.14-4.90]	2.13E-2	3.52 [1.98-6.25]	1.81E-5
12	6067283	rs185814268	A	G	NA	NA	NA	NA	0.86	0.0092	4.66 [2.23-9.74]	4.21E-5	0.85	0.0092	3.53 [1.51-8.22]	3.51E-3	4.16 [2.36-7.31]	7.79E-7
12	59333262	rs4760248	G	C	0.99	0.1086	1.63 [1.23-2.16]	6.93E-4	1.00	0.1090	1.28 [1.00-1.62]	4.56E-2	0.99	0.0997	1.32 [1.01-1.72]	3.88E-2	1.38 [1.18-1.61]	4.52E-5
12	77326010	rs117273906	C	T	0.95	0.0091	4.41 [1.8-10.8]	1.18E-3	0.99	0.0129	2.21 [1.20-4.06]	1.09E-2	0.99	0.0120	2.31 [1.14-4.67]	2.06E-2	2.57 [1.69-3.91]	1.07E-5
12	91345195	rs61926456	C	T	NA	NA	NA	NA	0.89	0.0129	3.21 [1.73-5.99]	2.35E-4	0.89	0.0103	2.11 [1.01-4.38]	4.60E-2	2.71 [1.68-4.39]	4.89E-5
12	124110116	rs188979614	C	T	0.85	0.0100	3.13 [1.29-7.59]	1.15E-2	0.93	0.0054	4.17 [1.77-9.8]	1.07E-3	0.92	0.0051	4.77 [1.57-14.5]	5.83E-3	3.88 [2.24-6.75]	1.50E-6
12	124161354	rs190737725	C	T	0.80	0.0090	4.16 [1.48-11.7]	6.90E-3	0.94	0.0053	4.20 [1.77-9.96]	1.12E-3	NA	NA	NA	NA	4.18 [2.13-8.22]	3.32E-5
13	43516465	rs192910570	A	G	NA	NA	NA	NA	0.82	0.0110	4.03 [2.11-7.71]	2.57E-5	0.84	0.0106	2.78 [1.03-7.55]	4.42E-2	3.63 [2.09-6.3]	4.37E-6
13	43523271	rs143441960	T	C	NA	NA	NA	NA	0.85	0.0144	2.99 [1.68-5.32]	2.04E-4	0.83	0.0118	2.52 [1.06-6.01]	3.70E-2	2.84 [1.75-4.63]	2.57E-5
13	50380270	rs35389309	A	G	0.90	0.0060	7.31 [2.39-22.4]	4.91E-4	NA	NA	NA	NA	0.89	0.0073	3.69 [1.58-8.59]	2.50E-3	4.72 [2.33-9.54]	1.61E-5
13	77117893	rs149658103	C	T	0.88	0.0159	2.23 [1.04-4.78]	3.90E-2	0.96	0.0226	1.85 [1.12-3.05]	1.58E-2	0.97	0.0144	2.36 [1.29-4.30]	5.22E-3	2.07 [1.46-2.94]	4.91E-5
13	92556834	rs9556131	T	A	0.92	0.1107	1.39 [1.01-1.93]	4.56E-2	0.98	0.0977	1.57 [1.24-2.00]	2.32E-4	0.97	0.1021	1.33 [1.03-1.73]	3.16E-2	1.45 [1.23-1.70]	5.68E-6
14	53451666	rs182533924	A	T	NA	NA	NA	NA	0.83	0.0072	5.29 [2.32-12.0]	7.24E-5	0.81	0.0067	4.61 [1.51-14.10]	7.21E-3	5.05 [2.58-9.88]	2.25E-6
14	54089652	rs187000606	C	T	NA	NA	NA	NA	0.88	0.0062	4.92 [2.13-11.34]	1.84E-4	0.90	0.0062	3.29 [1.42-7.61]	5.32E-3	4.06 [2.22-7.42]	5.29E-6
14	82049712	rs138142698	A	G	0.81	0.0087	3.78 [1.48-9.68]	5.56E-3	NA	NA	NA	NA	0.92	0.0081	3.78 [1.74-8.22]	7.86E-4	3.78 [2.02-7.07]	3.08E-5
14	92700530	rs117705014	G	A	NA	NA	NA	NA	0.89	0.0118	2.30 [1.26-4.22]	6.99E-3	0.86	0.0093	3.98 [1.96-8.08]	1.35E-4	2.87 [1.80-4.59]	1.03E-5
15	76081200	rs60514164	C	T	0.77	0.0578	2.11 [1.37-3.25]	6.94E-4	0.92	0.0676	1.54 [1.15-2.07]	4.22E-3	0.92	0.0848	1.47 [1.08-1.99]	1.51E-2	1.6 [1.32-1.95]	2.35E-6
15	82159556	rs116962753	T	C	0.66	0.0102	3.13 [1.22-8.01]	1.73E-2	0.80	0.0082	4.33 [1.99-9.40]	2.11E-4	NA	NA	NA	NA	3.83 [2.08-7.04]	1.61E-5
15	101914234	rs35647788	C	T	NA	NA	NA	NA	0.93	0.0069	5.18 [2.35-11.42]	4.60E-5	0.95	0.0098	4.25 [1.78-10.17]	1.14E-3	4.76 [2.62-8.64]	2.96E-7
16	13757971	rs183941002	G	T	NA	NA	NA	NA	0.95	0.0062	5.21 [2.28-11.90]	8.79E-5	0.92	0.0064	11.14 [4.78-26.00]	2.34E-8	7.44 [4.07-13.60]	6.60E-11
16	24509583	rs28510778	C	T	0.75	0.0388	1.92 [1.13-3.25]	1.57E-2	1.00	0.0517	1.48 [1.08-2.02]	1.48E-2	1.00	0.0609	1.48 [1.10-2.00]	9.50E-3	1.53 [1.25-1.88]	4.35E-5

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Variant Location and Alleles					US cohort				UK cohort				UUS cohort				Stage-I meta-analysis	
Chr	Pos	SNP rsID	REF	EA	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	HR [95%CI]	P
16	29437123	rs71387661	T	C	NA	NA	NA	NA	0.89	0.0246	2.02 [1.30-3.16]	1.95E-3	0.90	0.0151	2.63 [1.36-5.07]	3.90E-3	2.19 [1.51-3.18]	3.97E-5
16	29599816	rs147189264	G	A	NA	NA	NA	NA	0.88	0.0284	1.92 [1.25-2.95]	2.77E-3	0.88	0.0205	2.64 [1.50-4.68]	8.27E-4	2.14 [1.51-3.03]	1.67E-5
16	29603553	rs34944462	C	T	NA	NA	NA	NA	0.89	0.0284	1.93 [1.26-2.95]	2.64E-3	0.88	0.0204	2.65 [1.50-4.70]	7.97E-4	2.15 [1.52-3.04]	1.55E-5
16	57317482	rs148521985	C	T	NA	NA	NA	NA	0.90	0.0095	2.75 [1.34-5.67]	6.01E-3	0.89	0.0074	3.86 [1.72-8.7]	1.10E-3	3.18 [1.83-5.50]	3.70E-5
16	65985243	rs144433549	G	A	NA	NA	NA	NA	0.88	0.0055	8.02 [3.08-20.9]	2.08E-5	0.89	0.0064	3.11 [1.28-7.56]	1.25E-2	4.91 [2.52-9.56]	2.84E-6
16	83235650	rs11642140	T	C	NA	NA	NA	NA	0.99	0.0584	1.62 [1.19-2.20]	2.06E-3	0.99	0.0559	1.62 [1.17-2.26]	4.13E-3	1.62 [1.29-2.04]	3.58E-5
17	18860566	rs187631060	G	A	NA	NA	NA	NA	0.84	0.0061	4.40 [1.78-10.9]	1.36E-3	0.81	0.0050	10.27 [4.04-26.1]	9.66E-7	6.53 [3.37-12.68]	2.82E-8
17	32965617	rs11650798	G	C	0.61	0.5741	0.69 [0.55-0.88]	2.33E-3	0.98	0.5712	0.82 [0.70-0.96]	1.40E-2	0.98	0.5763	0.81 [0.68-0.96]	1.71E-2	0.79 [0.71-0.88]	1.88E-5
17	78670545	rs187199661	G	A	NA	NA	NA	NA	0.87	0.0061	3.48 [1.45-8.39]	5.34E-3	0.88	0.0071	4.04 [1.75-9.33]	1.06E-3	3.76 [2.02-6.97]	2.73E-5
17	78806880	rs139401580	C	T	NA	NA	NA	NA	0.93	0.0053	4.38 [1.82-10.6]	9.92E-4	0.92	0.0084	3.37 [1.65-6.89]	8.73E-4	3.76 [2.13-6.64]	4.93E-6
18	12796936	rs187303689	G	A	NA	NA	NA	NA	0.95	0.0062	2.63 [1.24-5.57]	1.19E-2	0.93	0.0065	8.12 [3.32-19.9]	4.56E-6	4.1 [2.28-7.36]	2.32E-6
18	12954976	rs191153148	C	T	NA	NA	NA	NA	0.88	0.0053	2.95 [1.12-7.73]	2.79E-2	0.85	0.0055	9.14 [3.26-25.6]	2.53E-5	4.89 [2.39-10.00]	1.43E-5
18	25516518	rs10469051	G	C	1.00	0.0739	1.66 [1.2-2.3]	2.41E-3	1.00	0.0655	1.44 [1.09-1.90]	9.58E-3	1.00	0.0653	1.38 [1.01-1.90]	4.58E-2	1.48 [1.24-1.77]	2.08E-5
18	30461003	rs141194630	T	C	0.81	0.0050	3.41 [1.01-11.6]	4.90E-2	0.87	0.0055	7.26 [3.01-17.5]	1.00E-5	NA	NA	NA	NA	5.70 [2.76-11.80]	2.58E-6
19	1021639	rs150244663	C	T	NA	NA	NA	NA	0.88	0.0278	2.13 [1.33-3.40]	1.54E-3	0.89	0.0210	2.41 [1.34-4.33]	3.13E-3	2.23 [1.54-3.23]	2.24E-5
19	1412576	rs147560834	G	A	NA	NA	NA	NA	0.91	0.0103	2.71 [1.35-5.44]	4.91E-3	0.93	0.0097	2.82 [1.45-5.50]	2.33E-3	2.77 [1.69-4.52]	4.98E-5
19	1412985	rs3893252	C	T	NA	NA	NA	NA	0.82	0.0051	10.40 [3.60-30.0]	1.50E-5	0.99	0.0067	2.18 [1.09-4.35]	2.81E-2	3.57 [1.97-6.49]	2.91E-5
19	1482080	rs78359732	C	A	NA	NA	NA	NA	1.00	0.0106	2.52 [1.34-4.72]	3.92E-3	0.99	0.0100	2.88 [1.51-5.51]	1.37E-3	2.68 [1.69-4.25]	2.60E-5
19	46090283	rs141771937	T	C	NA	NA	NA	NA	1.00	0.0133	2.33 [1.30-4.20]	4.71E-3	0.97	0.0125	4.31 [2.32-8.01]	3.87E-6	3.08 [1.99-4.76]	3.86E-7
19	49909139	rs559050154	G	A	NA	NA	NA	NA	0.91	0.0115	3.64 [1.86-7.14]	1.63E-4	0.88	0.0067	3.25 [1.09-9.68]	3.44E-2	3.54 [1.98-6.31]	1.88E-5
20	10720969	rs78188241	T	C	0.82	0.0211	2.97 [1.58-5.59]	7.15E-4	1.00	0.0301	1.53 [1.00-2.34]	4.81E-2	0.95	0.0212	2.55 [1.53-4.25]	3.35E-4	2.05 [1.52-2.76]	2.11E-6
20	11731982	rs117414348	A	G	NA	NA	NA	NA	0.84	0.0111	3.30 [1.77-6.17]	1.81E-4	0.80	0.0060	3.23 [1.30-8.00]	1.13E-2	3.28 [1.95-5.52]	8.01E-6
20	24359932	rs77942623	T	C	0.82	0.0051	7.75 [2.07-29.00]	2.34E-3	NA	NA	NA	NA	0.92	0.0052	6.97 [2.76-17.58]	3.96E-5	7.21 [3.27-15.90]	9.84E-7
20	36479360	rs76505427	G	T	NA	NA	NA	NA	0.86	0.0061	3.05 [1.31-7.10]	9.52E-3	0.86	0.0074	5.58 [2.54-12.3]	1.92E-5	4.16 [2.31-7.50]	2.03E-6
20	39496129	rs118012081	G	A	NA	NA	NA	NA	0.82	0.0071	2.80 [1.12-7.01]	2.77E-2	0.82	0.0082	6.78 [2.93-15.70]	7.77E-6	4.45 [2.37-8.39]	3.69E-6
20	40341113	rs142276787	T	C	0.88	0.0078	3.34 [1.15-9.65]	2.61E-2	0.95	0.0071	2.76 [1.21-6.28]	1.58E-2	0.94	0.0059	3.15 [1.37-7.25]	7.03E-3	3.02 [1.78-5.12]	3.89E-5

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Table E1. Stage I variants of nominal significance identified in the 3-way meta-analysis

Variant Location and Alleles					US cohort				UK cohort				UUS cohort				Stage-I meta-analysis	
Chr	Pos	SNP rsID	REF	EA	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	R2	EAF	HR [95%CI]	P	HR [95%CI]	P
20	42789152	rs12480648	C	T	0.95	0.0063	3.99 [1.21-13.10]	2.28E-2	0.94	0.0060	6.56 [2.41-17.90]	2.35E-4	NA	NA	NA	NA	5.40 [2.47-11.80]	2.38E-5
20	48957748	rs76672992	G	A	0.83	0.0067	4.50 [1.59-12.80]	4.72E-3	0.96	0.0106	2.98 [1.63-5.46]	3.86E-4	0.97	0.0127	2.78 [1.35-5.73]	5.62E-3	3.11 [2.02-4.79]	2.82E-7
20	61469020	rs71325411	A	G	NA	NA	NA	NA	0.75	0.0335	2.32 [1.49-3.61]	1.96E-4	0.74	0.0287	2.07 [1.20-3.59]	9.33E-3	2.22 [1.57-3.15]	7.70E-6
22	19257205	rs362240	A	G	NA	NA	NA	NA	0.90	0.0093	4.04 [1.94-8.41]	1.86E-4	0.89	0.0101	2.62 [1.35-5.11]	4.63E-3	3.22 [1.94-5.33]	5.48E-6
22	34904345	rs187511566	A	G	NA	NA	NA	NA	0.90	0.0053	6.70 [2.71-16.60]	3.73E-5	0.84	0.0056	3.69 [1.43-9.49]	6.79E-3	5.10 [2.62-9.92]	1.62E-6
22	36599507	rs9306307	A	G	0.89	0.2510	0.71 [0.55-0.91]	7.17E-3	0.96	0.2905	0.78 [0.65-0.93]	5.93E-3	0.97	0.2888	0.79 [0.65-0.97]	2.24E-2	0.77 [0.68-0.87]	1.70E-5
22	38095241	rs79385984	G	A	NA	NA	NA	NA	0.82	0.0104	2.86 [1.47-5.60]	2.09E-3	0.84	0.0077	3.62 [1.50-8.74]	4.21E-3	3.11 [1.81-5.34]	4.03E-5

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Table E2. Stage II validation of survival-associated variants identified in stage I.

Chr	SNP rs ID	Position	EAF	HR [95%CI]	*p*
1	rs7514663	10762349	0.008	0.91 [0.23-3.68]	0.90
1	rs185926659	94606086	NA	NA	NA
1	rs148192333	100493151	0.004	1.09 [0.27-4.44]	0.90
1	rs111749519	170154993	0.008	0.97 [0.40-2.35]	0.94
1	rs190285902	188069617	0.013	1.07 [0.48-2.42]	0.86
1	rs145551873	188151083	0.016	1.08 [0.53-2.19]	0.84
1	rs11589092	200768521	0.100	0.94 [0.70-1.25]	0.67
1	rs968549	200861595	0.903	1.07 [0.80-1.43]	0.66
1	rs115684501	203327373	0.004	1.91 [0.47-7.74]	0.36
1	rs115702239	214247966	0.008	2.17 [0.8-5.84]	0.13
1	rs112891824	222319103	0.016	1.12 [0.57-2.18]	0.74
1	rs34473966	242551121	NA	NA	NA
2	rs114698213	38114198	0.008	0.47 [0.12-1.88]	0.28
2	rs150622385	38249347	0.008	0.88 [0.32-2.39]	0.81
2	rs185306441	43602198	0.012	1.78 [0.91-3.48]	0.09
2	rs115376562	47162883	0.010	1.24 [0.55-2.80]	0.61
2	rs150765591	54643373	0.008	2.17 [0.96-4.92]	0.06
2	rs184498750	84291167	0.012	2.05 [1.00-4.18]	0.0493
2	rs149029025	138337791	NA	NA	NA
2	rs142306180	142907569	0.008	1.36 [0.56-3.32]	0.50
2	rs2357323	172941078	0.004	0.71 [0.18-2.86]	0.63
2	rs56923699	172942149	0.004	0.71 [0.18-2.86]	0.63
2	rs182271851	173111445	0.004	0.48 [0.12-1.94]	0.30
2	rs377036225	226721440	0.009	0.41 [0.13-1.29]	0.13
2	rs142266733	226758606	0.009	0.41 [0.13-1.29]	0.13
2	rs188643417	240384770	0.006	0.60 [0.15-2.43]	0.48

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Table E2. Stage II validation of survival-associated variants identified in stage I.

Chr	SNP rs ID	Position	EAF	HR [95%CI]	*p*
2	rs116377902	241253363	0.009	0.39 [0.10-1.56]	0.18
2	rs75406448	241397273	0.045	1.00 [0.65-1.53]	0.99
3	rs73177699	20681518	0.004	2.85 [0.90-9.04]	0.08
3	rs77895452	59911730	0.004	0.54 [0.13-2.27]	0.40
3	rs139644937	69741636	0.011	0.88 [0.41-1.87]	0.74
3	rs142255892	75034442	0.012	1.12 [0.51-2.42]	0.78
3	rs36022026	79693632	0.065	0.94 [0.66-1.33]	0.72
3	rs2178398	112828302	0.176	0.92 [0.72-1.16]	0.47
3	rs368666349	145344805	0.007	1.22 [0.30-4.94]	0.78
4	rs191600377	11752169	0.011	1.81 [0.80-4.11]	0.15
4	rs17260001	11982650	0.008	1.67 [0.61-4.58]	0.32
4	rs192085162	12091599	NA	NA	NA
4	rs142199841	66572997	0.007	0.72 [0.22-2.30]	0.58
4	rs79914686	74459567	0.002	3.95 [0.98-16.0]	0.05
4	rs28641522	76495474	0.007	3.1 [0.98-9.82]	0.05
4	rs6850444	122351715	0.352	0.89 [0.74-1.07]	0.22
4	rs71618464	166645872	0.008	1.56 [0.72-3.38]	0.26
4	rs139698405	175369612	0.009	0.57 [0.18-1.80]	0.34
4	rs190563090	175401693	0.009	0.57 [0.18-1.80]	0.34
5	rs138830243	36581565	0.006	1.13 [0.28-4.55]	0.87
5	rs138577491	96796936	0.010	0.48 [0.15-1.58]	0.23
5	rs571176768	106109149	NA	NA	NA
5	rs191657347	151623819	0.006	1.61 [0.51-5.14]	0.42
5	rs536809954	154587673	0.009	1.00 [0.37-2.70]	1.00
5	rs13172001	154820302	0.011	2.07 [0.97-4.42]	0.06
6	rs116594669	26118992	0.007	0.37 [0.05-2.66]	0.32

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Table E2. Stage II validation of survival-associated variants identified in stage I.

Chr	SNP rs ID	Position	EAF	HR [95%CI]	*p*
6	rs145442342	75389935	0.011	0.72 [0.29-1.76]	0.47
6	rs140590158	77044422	0.020	0.75 [0.38-1.47]	0.40
6	rs116863455	84945776	0.011	0.87 [0.32-2.35]	0.79
6	rs149057325	94466663	0.027	1.01 [0.60-1.69]	0.97
6	rs143246710	94498243	0.015	0.98 [0.52-1.87]	0.96
6	rs144768626	94531687	0.015	0.99 [0.52-1.88]	0.97
6	rs139568075*	102917669	0.009	0.10 [0.01-0.73]	0.02
6	rs35483630	116041139	0.010	0.67 [0.30-1.52]	0.34
6	rs181590625	116377677	0.008	0.63 [0.25-1.57]	0.32
6	rs535860622	139248572	0.006	1.30 [0.32-5.26]	0.71
6	rs149730644	143298800	NA	NA	NA
6	rs62427038	152786447	0.003	0.86 [0.21-3.48]	0.83
6	rs188589004	164521835	0.007	0.94 [0.30-2.97]	0.91
7	rs183625998	1065220	0.006	0.83 [0.20-3.36]	0.79
7	rs192803195	1120020	0.007	0.81 [0.20-3.28]	0.77
7	rs868964452	74953934	NA	NA	NA
7	rs117388086	75344084	0.012	0.39 [0.12-1.23]	0.11
7	rs111489307	75358307	0.016	0.64 [0.24-1.73]	0.38
7	rs149406562	101163592	0.012	2.00 [0.82-4.91]	0.13
7	rs142639517	103029848	0.010	1.20 [0.49-2.93]	0.68
7	rs74580469	125546344	0.010	0.33 [0.08-1.33]	0.12
7	rs556475473	148106376	NA	NA	NA
7	rs73728959	152982958	0.008	0.55 [0.14-2.23]	0.40
8	rs148904289	9496015	0.010	0.86 [0.27-2.70]	0.80
8	rs113465261	22381858	0.009	1.31 [0.58-2.97]	0.52
8	rs193064386	23454598	0.003	2.77 [0.67-11.5]	0.16

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Table E2. Stage II validation of survival-associated variants identified in stage I.

Chr	SNP rs ID	Position	EAF	HR [95%CI]	*p*
8	rs79562205	23626607	0.012	1.09 [0.48-2.46]	0.84
8	rs116969973	80493124	0.008	1.83 [0.74-4.53]	0.19
8	rs183828688*	92675281	0.007	0.38 [1.08-6.5]	0.03
8	rs117931584	120287198	0.011	0.98 [0.4-2.41]	0.97
9	rs147994675	4064850	0.013	1.13 [0.53-2.44]	0.75
9	rs190922348	7042171	0.010	1.54 [0.67-3.50]	0.31
9	rs118165660	14343426	0.017	1.03 [0.53-2.01]	0.93
9	rs113029495	24585027	0.007	0.79 [0.29-2.14]	0.65
9	rs139230397	86260066	0.003	1.77 [0.25-12.8]	0.57
9	rs187088797	92489004	0.008	1.72 [0.64-4.65]	0.29
10	rs142562867	4620992	0.017	0.82 [0.41-1.67]	0.59
10	rs111589468	11875431	0.006	0.70 [0.17-2.84]	0.62
10	rs139494937	43302339	0.020	1.36 [0.74-2.53]	0.32
10	rs186597433	73718436	0.006	2.31 [0.85-6.32]	0.10
10	rs191887805	87849922	0.007	0.75 [0.24-2.34]	0.62
10	rs117054238	120582311	0.006	2.14 [0.79-5.80]	0.13
11	rs78904863	14115243	0.022	0.55 [0.26-1.18]	0.13
11	rs111945608	28663945	0.019	0.56 [0.24-1.3]	0.18
11	rs189350185	30638253	0.007	0.51 [0.16-1.61]	0.25
11	rs76061348	33998481	0.015	0.68 [0.28-1.66]	0.39
11	rs118080683	36108905	0.029	0.57 [0.31-1.06]	0.07
11	rs186991973	44113701	0.007	2.16 [0.85-5.46]	0.10
11	rs186259260	75214357	0.010	1.17 [0.51-2.66]	0.71
11	rs72940008	81937642	0.013	0.63 [0.23-1.72]	0.37
11	rs118091577	123294624	0.003	NA	0.99
11	rs559811602	123312059	0.001	NA	0.99

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Table E2. Stage II validation of survival-associated variants identified in stage I.

Chr	SNP rs ID	Position	EAF	HR [95%CI]	*p*
11	rs189162333	126441296	0.023	1.11 [0.62-1.99]	0.73
11	rs192803572	132893929	0.007	0.40 [0.10-1.62]	0.20
11	rs140676527	134086356	0.004	0.40 [0.06-2.85]	0.36
12	rs150598029	5522297	0.010	0.57 [0.21-1.54]	0.27
12	rs185814268	6067283	0.007	1.41 [0.35-5.78]	0.63
12	rs4760248	59333262	NA	NA	NA
12	rs117273906	77326010	0.017	0.70 [0.29-1.71]	0.43
12	rs61926456	91345195	0.016	1.05 [0.51-2.17]	0.89
12	rs188979614	124110116	0.001	NA	0.99
12	rs190737725	124161354	0.002	0.67 [0.09-4.80]	0.69
13	rs192910570	43516465	0.017	0.63 [0.26-1.53]	0.30
13	rs143441960	43523271	0.020	0.72 [0.34-1.54]	0.40
13	rs35389309	50380270	0.001	NA	0.99
13	rs149658103	77117893	0.016	1.33 [0.68-2.60]	0.40
13	rs9556131	92556834	0.097	1.20 [0.88-1.64]	0.25
14	rs182533924	53451666	0.008	1.21 [0.45-3.27]	0.71
14	rs187000606	54089652	0.009	0.96 [0.39-2.34]	0.92
14	rs138142698	82049712	0.002	1.13 [0.28-4.58]	0.87
14	rs117705014	92700530	0.007	1.27 [0.47-3.43]	0.64
15	rs60514164	76081200	0.076	1.42 [1.04-1.93]	0.0256
15	rs116962753	82159556	0.013	0.73 [0.30-1.78]	0.49
15	rs35647788	101914234	0.008	3.12 [1.37-7.11]	0.0067
16	rs183941002	13757971	0.004	1.57 [0.58-4.29]	0.38
16	rs28510778	24509583	0.063	0.98 [0.67-1.43]	0.91
16	rs71387661	29437123	NA	NA	NA
16	rs147189264	29599816	0.025	1.19 [0.66-2.14]	0.57

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Table E2. Stage II validation of survival-associated variants identified in stage I.

Chr	SNP rs ID	Position	EAF	HR [95%CI]	*p*
16	rs34944462	29603553	0.025	1.19 [0.66-2.14]	0.56
16	rs148521985	57317482	0.002	NA	0.99
16	rs144433549	65985243	0.007	0.59 [0.19-1.87]	0.37
16	rs11642140	83235650	0.057	0.85 [0.57-1.27]	0.43
17	rs187631060	18860566	0.001	1.32 [0.18-9.67]	0.78
17	rs11650798	32965617	0.563	1.08 [0.9-1.29]	0.40
17	rs187199661	78670545	0.007	0.70 [0.22-2.21]	0.55
17	rs139401580	78806880	0.003	2.20 [0.54-8.94]	0.27
18	rs187303689	12796936	0.007	0.47 [0.11-1.92]	0.29
18	rs191153148	12954976	0.007	0.38 [0.09-1.57]	0.18
18	rs10469051	25516518	0.071	1.10 [0.76-1.59]	0.61
18	rs141194630	30461003	0.004	1.07 [0.27-4.32]	0.92
19	rs150244663	1021639	0.021	1.04 [0.61-1.8]	0.88
19	rs147560834	1412576	0.011	0.70 [0.29-1.72]	0.44
19	rs3893252	1412985	0.015	2.09 [1.05-4.15]	0.0357
19	rs78359732	1482080	0.010	0.84 [0.34-2.07]	0.70
19	rs141771937	46090283	0.011	0.73 [0.27-1.98]	0.54
19	rs559050154	49909139	0.003	0.82 [0.20-3.33]	0.78
20	rs78188241	10720969	0.027	1.47 [0.85-2.53]	0.17
20	rs117414348	11731982	0.006	0.81 [0.2-3.26]	0.76
20	rs77942623	24359932	0.004	1.69 [0.62-4.61]	0.31
20	rs76505427	36479360	0.007	1.15 [0.28-4.75]	0.84
20	rs118012081	39496129	0.007	0.45 [0.11-1.81]	0.26
20	rs142276787	40341113	0.010	0.85 [0.31-2.32]	0.75
20	rs12480648	42789152	0.008	0.82 [0.30-2.21]	0.69
20	rs76672992	48957748	0.011	1.88 [0.92-3.82]	0.08

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Table E2. Stage II validation of survival-associated variants identified in stage I.

Chr	SNP rs ID	Position	EAF	HR [95%CI]	*p*
20	rs71325411	61469020	0.055	1.30 [0.92-1.83]	0.14
22	rs362240	19257205	0.008	1.62 [0.66-3.99]	0.29
22	rs187511566	34904345	0.007	1.75 [0.71-4.29]	0.22
22	rs9306307	36599507	NA	NA	NA
22	rs79385984	38095241	0.017	1.41 [0.72-2.76]	0.31

*p<0.05 but with an opposite direction of effect compared to Stage I

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Table E3. Survival association for top variants before and after multivariable adjustment

Variant location (hg19) and nearest gene						Unadjusted		Adjusted model 1*		Adjusted model 2**	
Chr.	**Position**	**SNP rsID**	**Gene**	**REF**	**EA**	**HR [95% CI]**	***P-value***	**HR [95 % CI]**	***P-value***	**HR [95 %CI]**	***P-value***
2	84291167	rs184498750	*SUCLG1*	G	T	3.11 [1.88-5.15]	$9.83x10^{-6}$	1.84 (1.13-2.99)	0.013	2.02 (1.24-3.28)	0.005
15	76081200	rs60514164	*UBE2Q2*	C	T	1.6 [1.32-1.95]	$2.35x10^{-6}$	1.48 (1.20-1.84)	$3.11x10^{-4}$	1.47 (1.19-1.83)	$3.83x10^{-5}$
15	101914234	rs35647788	*PCSK6*	C	T	4.76 [2.62-8.64]	$2.96x10^{-7}$	4.64 (2.38-9.04)	$6.32x10^{-6}$	4.18 (2.15-8.15)	$2.45x10^{-5}$
19	1412985	rs3893252	*DAZAP1*	C	T	3.57 [1.97-6.49]	$2.91x10^{-5}$	1.39 (0.74-2.61)	0.304	1.29 (0.68-2.44)	0.432

* Adjusted for baseline GAP stage

** Adjusted for sex and baseline age, FVC (% predicted) and DLCO (% predicted)

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Table E4. Characteristics of patients with PCSK6 variant

Patient	Study	Centre	Risk Alleles	Age	Sex	Outcome	Survival (months)
1	UK	Edinburgh	1	78	Male	Death	9.63
2	UK	Edinburgh	1	64	Male	Death	38.83
3	UK	Edinburgh	1	78	Male	Death	26.2
4	UK	PROFILE	1	76	Male	Death	5.62
5	UK	PROFILE	1	71	Female	Death	7.92
6	UK	Trent Lung	1	77	Male	Death	38.79
7	UK	Trent Lung	1	70	Male	Death	19.13
8	UUS	Brompton	1	77	Male	Death	10
9	UUS	Brompton	1	74	Male	Death	8
10	UUS	Brompton	1	62	Female	Death	2
11	UUS	Brompton	1	72	Female	Alive	10
12	UUS	Chicago	1	78	Male	Alive	1.18
13	UUS	Chicago	1	53	Male	Alive	16.79
14	UUS	Chicago	1	71	Male	Alive	5.36
15	UUS	Chicago	1	68	Male	Death	20.4
16	UUS	Chicago	1	67	Male	Alive	7.43
17	UUS	Nottingham	1	85	Male	Death	6.44
18	UUS	UCD	1	77	Female	Death	11.89
19	UUS	UCD	1	69	Female	Alive	22.8

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Table E5. Sensitivity analysis comparing PCSK6 results with censoring lung transplant versus considering this an event

	UK		UUS		Meta	
	OR [95% CI]	**p**	**OR [95% CI]**	**p**	**OR [95% CI]**	**p**
Transplant as event	5.18 [2.35, 11.42]	4.60E-05	4.25 [1.78, 10.17]	1.14E-03	4.76 [2.62, 8.64]	2.96E-07
Transplant censored	5.23 [2.41, 11.35]	2.83E-05	4.48 [1.87, 10.74]	7.67E-04	4.89 [2.73, 8.73]	8.04E-08

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Table E6. *In silico* functional assessment variants associated with TFS after stage II				
	rs184498750 *(SUCLG1)*	**rs60514164** *(UBE2Q2)*	**rs35647788** *(PCSK6)*	**rs3893252** *(DAZAP1)*
RegulomedB rank v2.0.3 (Score)	5 - TF binding or Dnase peak (0.0)	5 - TF binding or Dnase peak (0.17)	4 - TF binding + Dnase peak (0.61)	3a - TF binding + any motif + DNase peak (0.73)
Enhancer histone marks [HaploReg v4.1]	None	H3K4me1 (Epithelial)	H3K4me1[a], H3K27ac[b]	H3K4me1[c], H3K27ac[d]
Promoter histone marks [HaploReg v4.1]	H3K9ac (Fetal lung)	None	H3K4me3[e], H3K9ac[f]	H3K4me3[g], H3K9ac[h]
DNAse [HaploReg]	None	None	Digestive	HSC & B-cell, Muscle, Fetal Adrenal Gland, Pancreas, Monocytes-CD14+ RO01746 Primary Cells
Altered regulatory motifs [HaploReg v4.1]	CIZ, Mef2	AIRE_2, AP-3	ZBTB33	None
Proteins bound [HaploReg v4.1]	None	None	None	None
Hi-C interactions [HUGIn v1] A *P* ≤ Bonferroni corrected Pvalue was considered to select interactions	None	Fetal Lung fibroblast Cell: *NEIL1*, *MAN2C1*, *SIN3A*, *PTPN9*, *SNUPN*, *UBE2Q2*, *FBXO22*, *NRG4*, *C15orf27*, (*TMEM266*).	Fetal Lung fibroblast Cell: *CERS3*, *LINS*, *ASB7*, *VIMP* (*SELENOS*), *SNRPA1*, *PCSK6*, *TM2D3*, *TARSL2* (*TARS3*), *OR4F6*, *OR4F15* Lung: *TM2D3*, *TARSL2* (*TARS3*)	None
Open Targets Genetics v22.02 Top ranked genes based on the overall V2G score	*SUCLG1* (top ranked), *DNAH6*	*MAN2C1* (top ranked), *SNUPN*, *IMP3, ODF3L1*, *UBE2Q2*	*PCSK6* (top ranked), *SELENOS*, *SNRPA1, CHSY1*, *TM2D3*	*DAZAP1* (top ranked), *GAMT*, *NDUFS7*, *PWWP3A*, *C19orf25*, *APC2*, *RPS15*
sQTL [GTEx v8] Tissue-specific *P* ≤ 0.05	None	*MAN2C1* (Adipose, skin, artery, esophagus)	None	None
eQTL [GTEx v8] Tissue-specific *P* ≤ 0.05	None	*MAN2C1* (lung, cultured fibroblasts, adipose, skin, artery, esophagus, among others)	None	*LLNLR-307A6.1* (skin)
Score CAPE dsQTL >=0.5 [SNPDelScore]	NA	None	NA	H1 BMP4 derived trophoblast cultured cells, IMR90 fetal lung fibroblasts cell line, A549 EtOH 0.02pct lung carcinoma cell line, GM12878 lymphoblastoid cells, HeLa-S3 Cervical carcinoma cell line, Monocytes-CD14+ RO01746 Primary cells, NH-A Astrocytes primary cells, NHEK-Epidermal keratinocyte primary cells
Score CAPE eQTL >=0.5 [SNPDelScore]	NA	iPS DF 6.9 Cells, primary T cells from peripheral blood, foreskin fibroblast primary cells, gastric, ovary, GM12878 lymphoblastoid cells, HSMM skeletal muscle myoblasts cells, HSMM cell derived skeletal muscle myotubes cells, NH-A Astrocytes primary cells	NA	H1 cells, H9 cells, iPS DF 6.9 Cells, iPS DF 19.11 Cells, fetal intestine small, fetal muscle leg, placenta, ovary, HMEC mammary epithelial primary cells, HSMM skeletal muscle myoblasts cells, HSMM cell derived skeletal muscle myotubes cells, NHDF-Ad Adult Dermal Fibroblast primary cells
PheWAS [PhenoScanner v2]	Allele T has positive/increased effect association with:	Allele T has positive/increased effect association with:	Allele T has positive/increased effect association with:	Allele T has positive/increased effect association with:

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Table E6. *In silico* functional assessment variants associated with TFS after stage II

	rs184498750 *(SUCLG1)*	**rs60514164** *(UBE2Q2)*	**rs35647788** *(PCSK6)*	**rs3893252** *(DAZAP1)*
	-Cause of death: hodgkins disease, unspecified (P=1.51x10^{-6}) -Self-reported gall bladder disease (P=1.50 x10^{-5}) -Cause of death: ischaemic cardiomyopathy (P=3.39x10^{-4}) - Treatment with dexamethasone (P=3.65x10^{-4})	-Self-reported nasal or sinus disorder (P=3.98x10^{-5}) -Zoster (P=2.17x10^{-4}) -Treatment with logynon tablet (P=6.84x10^{-4}) -Duration of other exercises (P=9.17x10^{-4}) Allele T has negative/decreased effect association with: - Treatment with becotide 50 inhaler (P=5.84x10^{-4})	-Self-reported fracture lower leg or ankle (P=4.39x10^{-5}) -Self-reported acute infective polyneuritis or Guillain-Barre syndrome (P=1.16x10^{-4}) -Self-reported multiple sclerosis (P=2.27x10^{-4}) -Cause of death: epilepsy, unspecified (P=3.61x10^{-4}) -Polyarthrosis (P=5.07x10^{-4})	-Home area population density: Scotland large urban area (P=5.09x10^{-5}) -Self-reported unclassifiable non-cancer illness (P=1.07x10^{-4}) -Cause of death: duodenum (P=1.24x10^{-4}) -Self-reported thyroid problem (P=1.74x10^{-4}) -Cause of death: tongue, unspecified (P=2.70x10^{-4}) -Cause of death: vascular dementia, unspecified (P=3.18x10^{-4}) - Benign neoplasm of major salivary glands (P=6.06x10^{-4})

CAPE, cellular dependent deactivating mutations; dsQTL, DNase I-sensitive quantitative trait loci; eQTL, expression quantitative trait loci; ESC, embryonic stem cells; GM12878, lymphoblastoid cell line; HSC, hematopoietic stem cells; HSMM, human skeletal muscle myoblasts; Hi-C, Hydrophobic interaction chromatography; HUGIn, Hi-C Unifying Genomic Interrogator; HUVEC, human umbilical vein endothelial cell; iPS DF 6.9 Cells, human induced pluripotent stem cell line derived from foreskin fibroblasts; IMR90, human fetal lung cells; iPSC, induced pluripotent stem cells; LD, linkage disequilibrium; NH-A, normal human Astrocytes; NHDF, normal human dermal fibroblasts; NHLF, normal human lung fibroblasts; TF, transcription factor; V2G, variants to genes.

[a]ESC, iPSC, ES-derive, Blood & T-cell, HSC & B-cell, Epithelial (Foreskin Keratinocyte Primary Cells skin03), Brain, Muscle, Heart, Digestive, Fetal Adrenal Gland, Liver, Pancreas, Lung, Spleen; [b]ES-derive, Brain, Digestive, Liver; [c]IMR90, ESC, iPSC, ES-derive, Blood & T-cell, HSC & B-cell, Mesench, Myosat, Epithelial, Neurosph, Thymus, Brain, Adipose, Muscle, Heart, Sm. Muscle, Digestive, Placenta Amnion, Fetal Lung, Ovary, Fetal Adrenal Gland, Placenta, Liver, Pancreas, Lung, Spleen, A549 EtOH 0.02pct Lung Carcinoma Cell Line, Dnd41 T Cell Leukemia Cell Line, GM12878 Lymphoblastoid Cells, HeLa-S3 Cervical Carcinoma Cell Line, HepG2 Hepatocellular Carcinoma Cell Line, HMEC Mammary Epithelial Primary Cells, HSMM Skeletal Muscle Myoblasts Cells, HUVEC Umbilical Vein Endothelial Primary Cells, K562 Leukemia Cells, NH-A Astrocytes Primary Cells, NHEK-Epidermal Keratinocyte Primary Cells, NHLF Lung Fibroblast Primary Cells, Osteoblast Primary Cells; [d]ESC, iPSC, ES-derive, Blood & T-cell, HSC & B-cell, Mesench, Epithelial, Thymus, Brain, Adipose, Muscle, Heart, Sm. Muscle, Digestive, Ovary, Pancreatic Islets, Fetal Adrenal Gland, Placenta, Liver, Pancreas, Lung, Spleen, Dnd41 T Cell Leukemia Cell Line, GM12878 Lymphoblastoid Cells, HeLa-S3 Cervical Carcinoma Cell Line, HUVEC Umbilical Vein Endothelial Primary Cells, K562 Leukemia Cells,Monocytes-CD14+ RO01746 Primary Cells, NH-A Astrocytes Primary Cells, NHDF-Ad Adult Dermal Fibroblast Primary Cells, NHLF Lung Fibroblast Primary Cells, Osteoblast Primary Cells; [e]iPSC, Digestive; [f]Blood & T-cell, Epithelial, Digestive; [g]iPSC, ES-derive, Blood & T-cell, HSC & B-cell, Mesench, Epithelial, Neurosphil, Brain, Adipose, Muscle, Heart, Sm. Muscle, Digestive, Placenta Amnion, Pancreas, Spleen, Dnd41 T Cell Leukemia Cell Line, GM12878 Lymphoblastoid Cells, HeLa-S3 Cervical Carcinoma Cell Line, HepG2 Hepatocellular Carcinoma Cell Line, Monocytes-CD14+ RO01746 Primary Cells, NHLF Lung Fibroblast Primary Cells, Osteoblast Primary Cells; [h]IMR90, ESC, iPSC, ES-derive, Blood & T-cell, Mesench, Epithelial, Brain, Adipose, Muscle, Heart, Sm. Muscle, Digestive, Fetal Lung, Liver, Dnd41 T Cell Leukemia Cell Line, HeLa-S3 Cervical Carcinoma Cell Line, HepG2 Hepatocellular Carcinoma Cell Line, Monocytes-CD14+ RO01746 Primary Cells, NH-A Astrocytes Primary Cells, NHEK-Epidermal Keratinocyte Primary Cells, NHLF Lung Fibroblast Primary Cells

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Table E7. Sentinel eQTL variants for PCSK6 expression in GTEx (P-value and effect size refer to the association of the variant with PCSK6 expression)

Variant Id	rsid	P-Value	Normalised effect size	Tissue	D' with rs35647788
chr15_101450376_G_A_b38	rs12594107	2.2×10^{-10}	0.29	Adipose - Visceral (Omentum)	0.333
chr15_101451106_A_G_b38	rs7172696	3.0×10^{-9}	0.3	Artery - Aorta	0.274
chr15_101314021_G_A_b38	rs1871974	5.8×10^{-5}	-0.23	Artery - Aorta	1
chr15_101457731_C_T_b38	rs903551	1.0×10^{-5}	0.39	Artery - Coronary	1
chr15_101298620_C_T_b38	rs2412069	3.10×10^{-8}	-0.22	Artery - Tibial	0.24
chr15_101275527_CA_C_b38	rs5815005	5.2×10^{-5}	-0.48	Artery - Tibial	1
chr15_101412376_T_G_b38	rs12901903	5.9×10^{-5}	0.31	Artery - Tibial	1
chr15_100951683_G_A_b38	rs111324676	2.0×10^{-5}	-0.55	Brain - Cerebellum	1
chr15_101365761_T_G_b38	rs8029570	2.5×10^{-5}	-0.18	Brain - Cerebellum	1
chr15_101296232_G_A_b38	rs8029790	4.30×10^{-10}	-0.31	Cells - Cultured fibroblasts	0.271
chr15_101298813_G_A_b38	rs35193516	6.9×10^{-5}	-0.71	Cells - Cultured fibroblasts	1
chr15_101451106_A_G_b38	rs7172696	1.30×10^{-7}	0.26	Esophagus - Gastroesophageal Junction	0.274
chr15_101554156_T_C_b38	rs117006479	4.70×10^{-7}	0.48	Esophagus - Mucosa	1
chr15_101503557_T_C_b38	rs28735675	6.7×10^{-6}	0.15	Esophagus - Mucosa	0
chr15_101451568_T_C_b38	rs7178458	2.10×10^{-11}	0.25	Esophagus - Muscularis	0.274
chr15_101437584_T_G_b38	rs11638957	2.9×10^{-6}	0.16	Esophagus - Muscularis	0.365
chr15_101612429_G_A_b38	rs113387732	3.0×10^{-5}	0.39	Esophagus - Muscularis	1
chr15_101473678_G_C_b38	rs1973403	5.7×10^{-5}	0.15	Esophagus - Muscularis	0.595
chr15_101456239_GT_G_b38	rs71154332	3.2×10^{-14}	0.37	Heart - Atrial Appendage	0.256
chr15_101495064_T_C_b38	rs2047222	4.6×10^{-5}	-0.19	Heart - Atrial Appendage	0.143
chr15_101451106_A_G_b38	rs7172696	2.3×10^{-8}	0.27	Heart - Left Ventricle	0.274
chr15_101444144_C_T_b38	rs145005722	1.1×10^{-5}	-0.77	Heart - Left Ventricle	1

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chr15_101480670_G_C_b38	rs7165319	6.6×10^{-5}	-0.28	Heart - Left Ventricle	0.15
chr15_101616653_G_A_b38	rs72761618	4.2×10^{-6}	0.33	Muscle - Skeletal	1
chr15_101329399_G_A_b38	rs72770737	1.2×10^{-8}	0.22	Nerve - Tibial	1
chr15_101309684_C_T_b38	rs4965377	4.8×10^{-8}	-0.26	Nerve - Tibial	1
chr15_101423871_T_C_b38	rs7169453	1.8×10^{-5}	-0.13	Nerve - Tibial	0.412
chr15_101370059_A_G_b38	rs114524031	2.7×10^{-5}	-0.54	Nerve - Tibial	1
chr15_101446601_G_A_b38	rs7402924	1.3×10^{-6}	0.37	Ovary	0.072
chr15_101500198_C_T_b38	rs11247301	3.7×10^{-8}	-0.41	Pancreas	1
chr15_100934663_G_A_b38	rs60722485	4.0×10^{-6}	1.5	Pituitary	0
chr15_101488478_G_C_b38	rs12437484	2.9×10^{-7}	0.13	Skin - Sun Exposed (Lower leg)	1
chr15_101456364_T_A_b38	rs4547317	2.0×10^{-5}	0.18	Thyroid	0.051

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Table E8. Survival association for fifteen genetic variants previously linked to IPF susceptibility

							Risk GWAS		Survival GWAS	
Chr.	**Position**	**SNP rsid**	**Locus**	**Ref. allele**	**Effect allele**	**EAF**	**OR* [95% CI]**	***P-value***	**HR [95% CI]**	***P-value***
3	44902386	rs78238620	*KIF15*	T	A	0.08	1.58 [1.37, 1.83]	5.12×10^{-10}	0.92 [0.76, 1.13]	0.444
3	169481271	rs12696304	*LRRC34 /TERC*	C	G	0.31	1.31 [1.21, 1.40]	7.09×10^{-13}	0.95 [0.86, 1.06]	0.343
4	89885086	rs2013701	*FAM13A*	T	G	0.575	1.28 [1.19, 1.35]	3.30×10^{-13}	1.01 [0.92, 1.12]	0.792
5	1282414	rs7725218	*TERT*	A	G	0.734	1.39 [1.30, 1.49]	1.54×10^{-20}	0.97 [0.86, 1.09]	0.622
5	169015479	rs1164837831	*SPDL1*	G	A	0.013	2.40 [1.70-3.40]	$7.55\text{x}10^{-7}$	1.23 [0.81-1.87]	0.327
6	7563232	rs2076295	*DSP*	T	G	0.573	1.46 [1.37, 1.56]	2.79×10^{-30}	1.07 [0.97, 1.18]	0.153
7	1909479	rs12699415	*MAD1L1*	G	A	0.469	1.28 [1.19, 1.37]	7.15×10^{-13}	0.98 [0.89, 1.09]	0.765
7	99630342	rs2897075	7q22.1	C	T	0.408	1.30 [1.21, 1.38]	3.10×10^{-14}	1.08 [0.98, 1.20]	0.127
8	120934126	rs28513081	*DEPTOR*	G	A	0.631	1.22 [1.15, 1.32]	1.20×10^{-9}	0.97 [0.88, 1.08]	0.559
11	1241221	rs35705950	*MUC5B*	G	T	0.321	4.84 [4.37, 5.36]	1.18×10^{-203}	0.78 [0.64, 0.94]	0.008
13	113534984	rs9577395	*ATP11A*	G	C	0.827	1.30 [1.20, 1.41]	1.34×10^{-10}	0.98 [0.85, 1.11]	0.723
15	40720542	rs59424629	*IVD*	G	T	0.573	1.30 [1.22, 1.41]	7.30×10^{-16}	1.12 [1.01, 1.23]	0.028
15	86097216	rs62023891	*AKAP13*	G	A	0.343	1.27 [1.18, 1.36]	1.27×10^{-10}	0.98 [0.88, 1.09]	0.655
17	44214888	rs2077551	*MAPT*	C	T	0.828	1.41 [1.30, 1.54]	2.83×10^{-16}	0.97 [0.84, 1.12]	0.675
19	4717672	rs12610495	*DPP9*	A	G	0.371	1.31 [1.22, 1.42]	2.92×10^{-12}	0.99 [0.87, 1.11]	0.394

Abbreviations: EAF = effect allele frequency; OR = odds ratio; HR = hazard ratio; CI = confidence interval
*Odds ratios are given with respect to the allele that is associated with increased disease risk

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Table E9. Risk association for the four genetic variants linked to IPF survival in the stage I

							Risk GWAS	
Chr.	**Position**	**SNP rsid**	**Locus**	**Ref. allele**	**Effect allele**	**EAF**	**OR* [95% CI]**	***P-value***
2	84291167	rs184498750	*SUCLG1*	G	T	0.01	0.90 [0.66, 1.24]	0.531
15	76081200	rs60514164	*UBE2Q2*	C	T	0.07	0.93 [0.82, 1.06]	0.298
15	101914234	rs35647788	*PCSK6*	C	T	0.01	1.01 [0.67, 1.53]	0.947
19	1412985	rs3893252	*DAZAP1*	C	T	0.01	0.74 [0.50, 1.11]	0.146

Abbreviations: EAF = effect allele frequency; OR = odds ratio; HR = hazard ratio; CI = confidence interval
*Odds ratios are given with respect to the allele that is associated with increased disease risk

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