JGH Journal of Gastroenterology and Hepatology

JGHF

doi:10.1111/jgh.16284

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BILIARY AND PANCREATIC

Clinical and genetic determinants of severe acute pancreatitis: A genetic association study in the UK Biobank

Wing Kiu Chou,* Stephen Lam*,†1 and Bhaskar Kumar*,†1

*Norwich Medical School, University of East Anglia and †Department of Upper Gastrointestinal Surgery, Norfolk and Norwich University Hospital Norwich, UK

Key words
disease severity, inflammation, mutation, population-based genetic association study, risk factors.

Accepted for publication 26 June 2023.

Correspondence
Mr Bhaskar Kumar, Department of Upper Gastrointestinal Surgery, Norfolk and Norwich University Hospital, Norwich, UK.
Email: bhaskar.kumar@nnuh.nhs.uk

Declaration of conflict of interest: W. K. C. received the Guts UK/Dr Falk Pharma Medical Student Prize 2022 and the Wolfson Foundation Intercalation Prize, which supported his degree. The remaining authors, S. L. and B. K., disclose no conflict of interest.

Financial support: This study was funded by Norfolk and Norwich University Hospital Charity Grant 51007/F610. The funder was not involved in the planning, design, implementation, statistical analysis, interpretation, or any other aspect of the study including preparation of the paper or knowledge of its contents.

Abstract

Background and Aim: The clinical severity of acute pancreatitis is unpredictable, ranging from self-limiting disease to life-threatening inflammation. The determinants of severe acute pancreatitis (SAP) are unclear. We aim to identify clinical variables and single nucleotide polymorphisms (SNP) associated with SAP.

Methods: We used UK Biobank data to conduct a case–control clinical and genetic association study. Pancreatitis patients were identified through national hospital and mortality records across the United Kingdom. Clinical covariates and SAP were analyzed for associations. Genotyped data that included 35 SNPs were assessed for independent associations with SAP and SNP to SNP interaction.

Results: A total of 665 patients with SAP and 3304 non-SAP patients were identified. Male sex and older age increased odds of developing SAP (odds ratio [OR] 1.48; 95% confiden interval [CI] 1.24–1.78, $P < 0.0001$) and (OR 1.23; 95% CI 1.17–1.29), $P < 0.0001$), respectively. SAP was associated with diabetes (OR 1.46; 95% CI 1.15–1.86, $P = 0.002$), chronic kidney disease (OR 1.74; 95% CI 1.26–2.42, $P = 0.001$), and cardiovascular disease (OR 2.00; 95% CI 1.54–2.61, $P = 0.0001$). A significant association was established between IL-10 rs3024498 and SAP (OR 1.24; 95% CI 1.09–1.41, $P = 0.0014$). Epistasis analysis revealed that the odds of SAP was greater by an interaction between TLR 5 rs5744174 and Factor V rs6025 (ORinteraction 7.53; $P = 6.64 \times 10^{-5}$).

Conclusion: This study reports clinical risk factors for SAP. We also show evidence for an interaction between rs5744174 and rs6025 as determinants for SAP in addition to rs3024498 independently altering the severity of acute pancreatitis.

[1]Stephen Lam and Bhaskar Kumar are joint senior authors.

Introduction

Acute pancreatitis (AP) is a leading cause of emergency surgical and medical gastrointestinal hospitalizations in the United Kingdom.[1,2] The mortality and morbidity burden of AP are significant, yet our understanding of the mechanisms that dictate disease severity is poorly understood. A subset of individuals develop severe or fulminant disease, which is associated with significant morbidity and mortality.[3] This burden is often reflected by prolonged hospital stay, critical care admission, and surgical intervention.[3]

Mechanistically, it is widely accepted that AP is induced by sudden premature activation of pancreatic enzymes leading to localized autodigestion of the gland.[4] Serum biomarkers such as serum C-reactive protein, procalcitonin, and interleukin-8 have been associated with a more severe form of the disease.[5] Although associations between clinical patient characteristics such as body mass index (BMI),[6] and age[7] have been reported to independently increase the risk of severe acute pancreatitis (SAP) and its complications, our understanding of specific blood biomarkers and clinical parameters that may be predictors for severe disease is limited.

SAP is defined by the revised Atlanta Classification as the presence of persistent organ failure.[7] Treatment is supportive and centered around patient analgesia, nutritional support, and monitoring.[7] This may suffice for patients with non-SAP, but individuals that have SAP can develop acute lung injury, disseminated intravascular coagulopathy, and sepsis.[8] Up to half of such patients will not survive.[8] To date, there are no novel therapeutic agents developed specifically for SAP and pharmacological regimes have remained unchanged for over half a century.[9] A better understanding of the drivers of this unpredictable inflammation of the pancreas may help future drug repositioning and discovery.

Genetic association studies may provide the basis for further understanding specific predictors of SAP, which may identify genetic variants and their relationships with disease severity. A sustained

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imbalanced production of pro-inflammatory cytokines such as tumor necrosis factor-alpha (TNF-α) and interleukin (IL) IL-6 raised in the acute phase of AP[10] can result in multiorgan dysfunction, acute respiratory distress syndrome, and septic shock.[11] Genetic polymorphisms of acute inflammatory mediators and innate immunity processes have been identified as candidate genes by researchers to explore. A case–control study of 88 Spanish patients with pancreatitis investigating the TNF-α 238 polymorphism concluded that individuals carrying the heterozygous genotype G/A had a greater risk of systemic complications including organ failure, shock, and respiratory failure.[12] While a statistically significant association was reported in this demographic of Spanish patients, when the TNF-α variant at the −238 locus was investigated on 211 patients in an American population,[13] researchers did not report an association with organ failure. The conclusions drawn from these genetic association studies are varied, potentially due to different methods used to define severe disease and small sample sizes which may lead to type I and type II error. Furthermore, due to the lack of an a priori statistical analyses, post hoc selective reporting of genetic comparison models cannot be excluded.[14]

We aimed to clarify the relationship between patient clinical characteristics and single nucleotide polymorphisms and their relationship with AP severity using a large population-based case–control design using data from the UK Biobank.

Methods

This genetic association study is reported in accordance with Strengthening the Reporting of Genetic Association studies guidance, an extension of Strengthening the Reporting of Observational Studies in Epidemiology statement.[15,16]

Study design and participants. This study used the UK Biobank to investigate clinical and genetic determinants of SAP with a case–control study design. The UK Biobank is a large prospective cohort study of 502 628 participants aged 40 to 69 years of age across 22 centers in Scotland, England, and Wales.[17] Physical measurements, biological samples, and sociodemographic information were obtained at assessment centers through questionnaires, interviews, and biobanking of blood, urine, and saliva.[17] Hospital inpatient data were also linked to individual UK Biobank participants through national databases which included Hospital Episode Statistics for England, Scottish Morbidity Record, and Patient Episode Database for Wales.[17] Patient admission, diagnosis, and operative procedures were coded using the International Classification of Diseases (ICD) ICD-9, ICD-10, and Classification of Interventions and Procedures (OPCS) OPCS-3 and OPCS-4. All participants provided signed consent forms and ethical approval was granted by the North West Multi-Centre Research Ethics Committee (approval number: 11/NW/0382). This present study received approval under UK Biobank application number 82952 on August 10, 2021.

We included all patient with a diagnosis of pancreatitis in the cohort. Cases were defined as patients who developed SAP and identified through a multi-step process. First, participants with AP in the UK Biobank were identified using ICD-9 code "5770" and ICD-10 code "K85" (K85.0 for idiopathic AP; K85.1 for biliary AP; K85.2 for alcohol-induced AP; K85.3 for drug-induced AP; K85.8 other AP; K85.9 unspecified AP). Patients with AP were also captured through national death registry record linkage, where the cause of death was reported as AP. Following this, we classified patients as SAP, if they received admission to critical care during their AP hospital episode, developed 30-day severe complications (including cardiac failure, renal failure, pulmonary edema, and disseminated intravascular coagulation [Table S2]), or required invasive procedures or surgical intervention such as a pancreatic necrosectomy (Table S3). Patients were also classified as cases if they died from AP, defined as 90-day all-cause mortality following admission or identified through death certificates. Controls were non-SAP and defined as any patient with AP but do not fall under any of the aforementioned definitions for severe disease.

Clinical characteristics. Patient comorbidities were identified through ICD-9 and ICD-10 codes. We compared specific comorbidities between cases and controls for diabetes, chronic obstructive pulmonary disease, chronic kidney disease, cerebrovascular accident, myocardial infarction, congestive cardiac failure, and hyperlipidemia. Anthropometric data such as BMI, calculated during recruitment, were also compared between groups. Biomarker measurements including serum cholesterol, triglycerides, HbA1c, white cell count, and calcium were ascertained at recruitment assessment centers and tabulated between cases and controls.

Selection of single nucleotide polymorphisms and genotyping/quality control. Over 488 377 participants in the UK Biobank were genotyped by Affymetrix Research Services. Blood samples were sent for whole-genome genotyping using the Applied Biosystems™ UK Biobank Axiom™ Array and UK BiLEVE Axiom™ Array, both purposefully designed for UK Biobank and share 95% marker content. A robust genetic quality control pipeline was implemented, which includes testing for plate effects, batch effects, sex effects, array effects, discordance and departure from Hardy Weinberg Equilibrium (HWE). To control for study population homogeneity, genotyped data included in analyses were limited to individuals self-reporting as White-British with similar genetic ancestry revealed through principal component analysis.[18] Thirty-five candidate SNPs identified from previous literature and systematic reviews were selected for analysis in the present study (Table 1).

Statistical analysis. Descriptive statistical analysis was conducted on cases and controls of SAP and non-SAP. Baseline clinical and demographic characteristics were presented as median and interquartile range or as a mean and standard deviation. Categorical variables were expressed as frequency (percentage). To investigate the association between clinical variables and SAP, we used a multivariable logistical regression model to estimate odds ratios (OR) using non-SAP as the comparator. Our regression model was adjusted for age (per 5-year increase), sex (binary), BMI (continuous), smoking status (current, never), Townsend deprivation index 5 (most deprived), chronic obstructive pulmonary disease (binary), diabetes (binary), chronic kidney disease (binary), cerebrovascular accident (binary), cardiovascular disease

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Table 1 SNP selected for association analysis in the UK Biobank

Gene	Gene symbol	rsID	SNP position (major allele/minor allele)
Tumor necrosis factor-α	TNF-α	rs1800629	31575254 G/A
Tumor necrosis factor receptor 2	TNFRSF1B	rs1061622	12192898 T/G
Prostaglandin-endoperoxide synthase 2	PTGS2	rs5275	186673926 A/G
Interleukin-3	IL-3	rs40401	132060785 C/T
Interleukin-4	IL-4	rs2243250	132673462 C/T
Interleukin-6	IL-6	rs1800795	22727026 G/C
Interleukin-8	IL-8	rs10191411	148630399 C/T
Interleukin-8/chemokine CXC ligand 8	IL-8	rs4073	73740307 T/A
Interleukin-10	IL-10	rs3024493	206770623 C/A
Interleukin-10	IL-10	rs3024505	206766559 G/A
Interleukin-10	IL-10	rs3024498	206768184 T/C
Interleukin-10	IL-10	rs1518111	206771300 C/T
Interleukin-10	IL-10	rs1800871	206773289 G/A
Interleukin-13	IL-13	rs1800925	132657117 C/T
Interleukin-23 receptor	IL-23R	rs11209026	67240275 G/A
Toll-like receptor 1	TLR1	rs5743611	38798593 C/G
Toll-like receptor 2	TLR2	rs5743704	153704799 C/A
Toll-like receptor 4	TLR4	rs4986790	117713024 A/G
Toll-like receptor 4	TLR4	rs4986791	117713324 C/T
Toll-like receptor 5	TLR5	rs5744174	223111186 A/G
Factor V	F5	rs6025	169549811 C/T
Glutathione synthetase	GSS	rs121909308	34935611 G/A
Glutathione synthetase	GSS	rs121909309	34932121 G/A
Glutathione s-transferase pi	GSTP1	rs1695	67585218 A/G
Caspase-9	CASP9	rs1052576	15506048 C/T
Caspase-10	CASP10	rs13006529	201217736 T/A
Catalase	CAT	rs7943316	34438925 T/A
Vitamin D Receptor	VDR	rs731236	47844974 A/G
Receptor interacting protein kinase 2	RIPK2	rs42490	89766285 A/G
Nucleotide-binding oligomerization domain-containing protein 2	NOD2	rs9302752	50685192 C/T
Superoxide dismutase 2	SOD2	rs4880	159692840 G/A
Myosin IXB	MYO9B	rs2305767	17183487 T/C
Apolipoprotein E	APOE	rs7412	44908822 C/T
Chemokine (C-X-C motif) ligand 1	CXCR1	rs2234671	218164385 C/G
Chemokine (C-X-C motif) ligand 1	CXCR1	rs16858811	218165120 A/C

(binary), and hyperlipidemia (binary). A P value of < 0.05 was considered statistically significant.

We conducted association analysis of individual SNPs between cases and controls under four genetic models: additive, codominant, dominant, and recessive models. The Akaike information criteria was used to select the best-fitting model for each SNP. Candidate SNPs were first analyzed for departure from HWE using an exact test. Only SNPs with HWE ≥ 0.05 were retained for univariate association analysis, which was presented as a crude OR. A Manhattan Plot was used for data visualization of individual association analysis of SNPs. SNP to SNP interaction (genetic epistasis) was analyzed using pairwise log-likelihood ratio tests and conditional regression association tests. A heatmap of P values was used to visualize magnitudes of epistasis associations. Control for multiple hypothesis testing was considered using Bonferroni correction, and a P value of < 0.00016 was considered statically significant. All statistical analysis was performed on STATA version 16.0 for Windows (StataCorp LLC, College Station, TX, USA) and R Statistical Software (v4.1.2). Genetic association analysis was performed using the "SNPAssoc" R package (v2.0-11).[19]

Results

Patient characteristics. Of the 502 536 participants enrolled onto the UK Biobank, and 3969 had a diagnosis of acute pancreatitis. A total of 655 participants (16.75%) were identified to have SAP, and 3304 participants (83.25%) had non-SAP. Table 2 summarizes patient demographic and clinical characteristics stratified by cases and controls.

Clinical covariates. The median age of patients with SAP was higher compared with non-SAP at 67.3 and 61.6 years, respectively. Comparing cases with controls, a greater proportion of patients were male (59.8% *vs* 46.7%), current smokers (16.5% *vs* 13.9%), dependent on alcohol (7.4% *vs* 5.5%) and a higher proportion were socioeconomically deprived (Townsend Index 5, 10.1% *vs* 8.6%). Both cases and controls were predominately of "White-British" ethnicity (96.2% and 96.7%). Diabetes at the time of diagnosis was almost double in patients with SAP (20.5% *vs* 10.8%), prevalent diagnosis of chronic kidney disease was three

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Table 2 Baseline characteristics of the patients included in the study

Variables	Severe acute pancreatitis	Non-severe acute pancreatitis	No pancreatitis
Number (%)	665 (16.75)	3304 (83.25)	498 669
Age (years), median IQR	67.3 (60.3–73.4 [39.3–82.5])	61.6 (54.6–69.2 [31.4–82.9])	56.5, (50–63 [39–71])
Sex (male), no. (%)	398 (59.85)	1545 (46.76)	227 145 (45.57)
Cholesterol, mmol/L	5.3 (4.4–6.1 [2.6–8.8])	5.5 (4.7–6.3 [2.5–9.8])	5.6 (4.9–6.4 [1.5–13.5])
HDL cholesterol, mmol/L	1.3 (1.0–1.5 [0.6–2.7])	1.3 (1.1–1.5 [0.6–3.0])	1.3 (1.1–1.6 [0.2–4.0])
LDL cholesterol, mmol/L	3.3 (2.6–3.9 [1.4–6.1])	3.5 (2.8–4.1 [1.3–6.8])	3.5 (2.9–4.1 [0.5–9.6])
Triglycerides, mmol/L	1.8 (1.3–2.5 [0.5–7.0])	1.7 (1.2–2.4 [0.4–10.1])	1.4 (1.0–2.1 [0.2–11.2])
HbA1c, mmol/L	37.0 (34.4–40.7 [26.7–88.4])	36.1 (33.4–39.4 [20.3–104.3])	35.2 (32.8–37.9 [15.3–265.6])
White cell count, 10^9/L	7.4 (6.2–8.7 [3.4–15])	7.0 (5.9–8.3 [3.1–19.1])	6.6 (5.6–7.8 [0.04–145.8])
Calcium, mmol/L	2.4 (2.3–2.4 [2.1–2.7])	2.4 (2.3–2.4 [2.1–2.9])	2.3 (2.3–2.4 [1.4–3.4])
Systolic blood pressure, mmHg	142 (128–164 [122–176])	140 (126–157 [106–193])	139 (126–153 [86–240])
Alcohol; daily/almost daily, no. (%)	109 (16.4)	538 (16.3)	104 058 (20.8)
Alcohol dependent, no. (%)	49 (7.4)	183 (5.5)	8630 (1.7)
Smoking status; current, no. (%)	110 (16.5)	459 (13.9)	52 393 (10.5)
White ethnicity, no. (%)	640 (96.2)	3197 (96.7)	468 865 (94.0)
Asian ethnicity, no. (%)	19 (2.9)	70 (2.1)	11 352 (2.2)
Townsend deprivation index 5 (lowest), no. (%)	67 (10.1)	283 (8.6)	28 138 (5.6)
BMI (kg/m^2), mean (SD)	29.4 (5.7)	29.4 (5.6)	27.4 (4.7)
Weight (kg), mean (SD)	84.3 (18.3)	82.9 (17.3)	77.9 (15.8)
Male	88.5 (16.4)	88.6 (16.3)	85.8 (14.3)
Female	78.1 (19.2)	78.0 (16.7)	71.4 (14.0)

Table 3 Prevalent comorbidities at the time of diagnosis in patients with severe acute pancreatitis and non-severe acute pancreatitis

Specific comorbidities, no. (%)	Severe acute pancreatitis	Non-severe acute pancreatitis
Diabetes	136 (20.5)	358 (10.8)
COPD	78 (11.7)	182 (5.5)
Chronic kidney disease	76 (11.4)	126 (3.8)
Cerebrovascular accident	44 (6.6)	96 (2.9)
Cardiovascular disease (MI CCF)	131 (19.7)	222 (6.7)
Hyperlipidemia	168 (25.3)	547 (16.6)

times higher in SAP patients (11.4% *vs* 3.8%) and both COPD and CVA had approximately twice the prevalence in participants with SAP (11.7% *vs* 5.5% and 6.6% *vs* 2.9%) (Table 3). Notably, the prevalence of cardiovascular disease was much greater in SAP at almost triple the proportion and the prevalence of hyperlipidemia was approximately present in a quarter of individuals with SAP compared to 16.6% in non-SAP (Table S4).

Table 4 describes effect size estimates for clinical variables and the development of SAP, using non-SAP as a reference. A regression model estimated the odds age (per 5-year increase) and developing SAP was 1.23 (95% CI 1.17–1.29, $P < 0.0001$). Male sex was also strongly associated with SAP (OR 1.48; 95% CI 1.24–1.78, $P < 0.0001$). Regression analysis did not establish any statistically significant association for BMI, smoking, alcohol dependency or Townsend index score, and disease severity. However, the odds of SAP was 46% and 74% higher in individuals with prevalent diabetes and chronic kidney disease, respectively. No other factors reached statistical significance (Table S5).

Table 4 Multivariable logistical regression model of age (per 5-year increase), sex (male *vs* female), BMI (per unit increase), smoking status (current), Townsend deprivation index 5 (most deprived), chronic obstructive pulmonary disease, diabetes, chronic kidney disease, cerebrovascular accident, cardiovascular disease (myocardial infarction and congestive cardiac failure), hyperlipidemia, and the odds of severe acute pancreatitis *versus* non-severe acute pancreatitis

Variable	Odds ratio (95% CI), *P* value
Age	1.23 (1.17–1.29), $P < 0.0001$
Sex	1.48 (1.24–1.78), $P < 0.0001$
BMI	0.99 (0.97–1.01), $P = 0.64$
Current smoker	1.24 (0.96–1.61), $P = 0.09$
Alcohol dependent	1.29 (0.87–1.90), $P = 0.20$
Townsend deprivation index 5 (lowest)	1.15 (0.85–1.56), $P = 0.36$
COPD	1.16 (0.85–1.59), $P = 0.35$
Diabetes	1.46 (1.15–1.86), $P = 0.002$
Chronic kidney disease	1.74 (1.26–2.42), $P = 0.001$
Cerebrovascular accident	1.38 (0.92–2.05), $P = 0.12$
Cardiovascular disease	2.00 (1.54–2.61), $P = 0.0001$
Hyperlipidemia	0.92 (0.73–1.51), $P = 0.46$

Single nucleotide polymorphisms analysis. Genotyped data were available for 646/665 participants with SAP and 3188/3304 participants with non-SAP. Among 35 SNPs (shown in Table 1) retrieved from UK Biobank, rs2243250 in the IL-4 gene, rs5743704 in the TLR-2 gene, rs121909308 and rs121909309 in the GSS gene, rs2234671 in the CXCR1 gene, violated HWE (≤ 0.05) and were excluded from the analysis. Figure 1 illustrates the association between individual SNPs and disease severity based on five genetic models. An assessment

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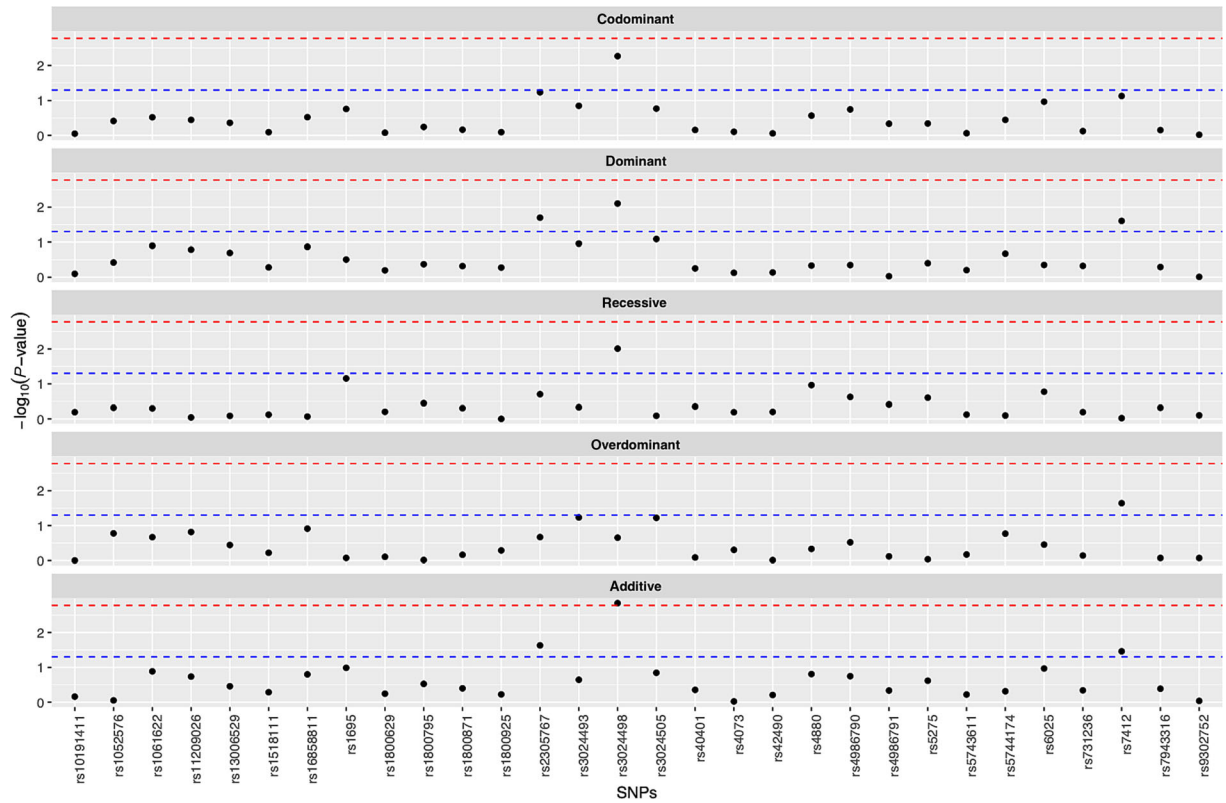

Figure 1 Manhattan plot of individual SNP associations under the codominant, dominant, recessive, overdominant, and additive models. Significance: - -, Bonferroni; - -, Nominal.

of 30 SNPs for the additive model revealed only rs3024498 reaching statistical significance following Bonferroni correction (*P* value of 0.0014). Logistical regression of genotypes found that a carrier of the risk C allele for this genetic polymorphism of the IL-10 gene increased the odds of more severe disease by 24% (OR 1.24; 95% CI 1.09–1.41) assuming the best fit model (additive). For the other genetic comparison models, no associations were found between SNPs and greater disease severity. Each SNP and its association with SAP are described in Table S1.

Single nucleotide polymorphisms to single nucleotide polymorphisms analysis. Pairwise SNP–SNP analysis was also explored. Comparisons of SNPs were performed for all 30 SNPs included in analysis (Fig. 2). The most significant association was between a variant of TLR5 and factor V (rs5744174 and rs6025) under an additive model. A log-likelihood ratio test of interaction revealed a significant association between both SNPs. This was further confirmed by conditional logistical regression, which found that individuals homozygous for the risk allele G for rs5744174 and had the T/C–T/T genotype for rs6025 had 7.53 times the odds of having SAP (95% CI 2.37–23.90, $P = 6.6476 \times 10^{-5}$). There were no other significant pairwise associations.

Discussion

We investigated the clinical and genetic predictors for SAP based on a large sample in a UK population. To the best of our knowledge, this is the first-time determinants for disease severity have been studied in the UK Biobank.

Our study confirms the findings from previous work, suggesting an association between diabetes and the development of more severe disease.[20] We found that a pre-existing diagnosis of diabetes was independently associated with the SAP (1.46 (1.15–1.86), $P = 0.002$). Our findings also suggest that the odds of developing SAP were greater given a pre-existing diagnosis of chronic kidney disease and prevalent cardiovascular disease.

A key finding in our study is a novel association between rs3024498 of IL-10 and severe pancreatitis. The IL-10 cytokine rises during the acute phase of inflammation and is a potent anti-inflammatory mediator. It attenuates the progression of inflammation by inhibiting macrophages and monocytes from releasing pro-inflammatory mediators namely IL-1, IL-6, and TNF-α, which are proportional to disease severity. In recent years, animal studies[21,22] found administration of IL-10 or augmentation of IL-10 was successful in modulating the degree of pancreatic inflammation—preventing cellular necrosis, reducing acinar edema and vacuolization. Support for an IL-10-mediated modifier of disease severity is also highlighted in recent work investigating severe COVID-19 infection.[23] Where early elevation of serum IL-10 levels predicts a more potent COVID-19 inflammatory reaction and "cytokine storm." Similarly, in SAP, a pronounced inflammatory cascade occurs, but whether IL-10 polymorphisms are determinants of a more severe disease has largely been unknown. Our finding revealed that carriage of risk C allele increased the odds

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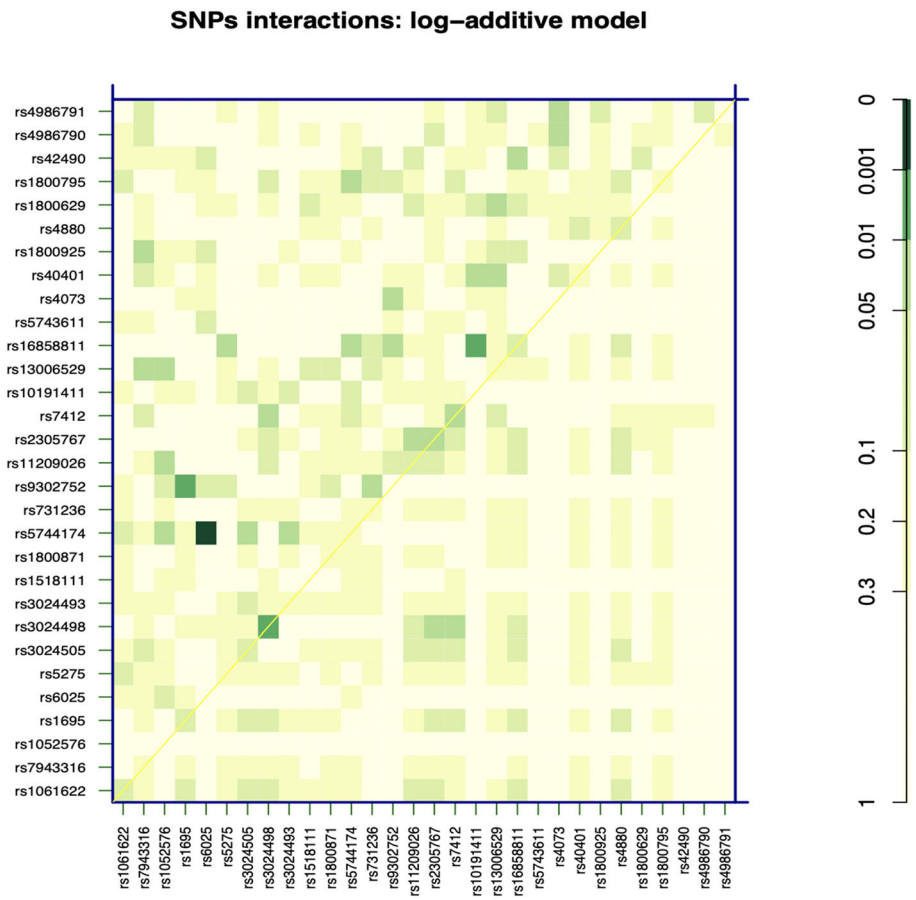

Figure 2 SNP to SNP interaction analysis results for severe acute pancreatitis. A heatmap plot for each SNP using the additive genetic model. Each plot contains the *P* values calculated from different likelihood ratio tests. Different colors indicate differing statistical significance levels. The yellow diagonal line contains *P* values from likelihood ratio tests for the crude effect of each SNP. The upper triangle in the matrix contains the *P* values for the interaction (epistasis) log-likelihood ratio tests. The lower triangle contains *P* values from likelihood ratio test comparing the two-SNP additive likelihood to the best of the single-SNP models.

of SAP. This finding may, in part, explain whether individuals included in our study were genetically predisposed to severe disease. The exact downstream effects of rs3024498 are unknown, but this 3′ untranslated region variant may contribute to complex gene expression or regulatory functions of IL-10.[24] The pathomechanstic consequences of polymorphisms within the IL-10 family of cytokines and IL-10 haplotypes on serum IL-10 levels and linking this to disease severity need to be clarified. This is particularly desirable for future risk stratification of individuals through polygenic risk scores and may also lend credence to future human trials with recombinant IL-10 and drug repositioning, although we should be cautious about overestimating the impact of IL-10 targeted drugs as previous trials have not consistently demonstrated improvement in outcomes.[25]

To the best of our knowledge, the current study is the first to investigate the significance of SNP to SNP interactions in the context of AP disease severity. In our analysis, the key finding was an interaction between TLR 5 and Factor V genes. TLR5 is involved in driving early inflammation and recognition of bacterial pathogens and the Factor V mutation results in the hypercoagulable state seen in Factor V Leiden. Although no crude associations were detected for each SNP independently, individuals were at a much greater risk of SAP if they carried both risk alleles of rs5744174 and rs6025. This is compelling evidence for the presence of a multiplicative complex genetic architecture in SAP. We believe that it is unlikely that the risk of SAP is solely dependent on individual SNPs; it is more plausible that the risk is also influenced by an interconnected network of SNP to SNP interactions.

There are several strengths to our study, notably, a population-based well-curated dataset, candidate SNP analysis using the largest sample of patients to date and the application of stringent genetic quality control. Additionally, this is the first time that the severity of AP has been classified in the UK Biobank. However, our study is subject to some limitations that should be considered during the interpretation of our findings. Firstly, subgroup analysis dependent on etiology was not analyzed. Previous published work suggests potential measurement bias, whereby ICD codes were inputted as "unspecified" or not entered.[26] Therefore, a stratified etiology-dependent analysis may result in biased findings not representative of true incidence rates. Furthermore, cases were not defined by the modified Atlanta Classification

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due to data availability limitations but rather based on composite codes that act as proxies for a severe disease course. In this case–control study, our definition of cases most closely matches moderately severe and severe disease courses and controls with mild pancreatitis. Baseline biochemical results collected at baseline should also be interpreted with caution as 68% of participants developed pancreatitis after recruitment into the UK Biobank (median 7.1 years, interquartile range 3.9–10). In view of our genetic analysis of SNPs, despite tightly controlled genotyping considerations on a Caucasian population, intrapopulation heterogeneity and cryptic relatedness cannot be excluded.[27]

In summary, clinical variables identified in our study such as age, sex, and specific comorbidities may aid in early prognostication of AP. Genetic determinants of disease severity such as IL-10 rs3024498 and SNP to SNP interactions may also uncover putative mechanisms that govern disease severity and ultimately inform future therapeutics used to attenuate the progressive inflammatory cascade in this potentially fatal disease.

Data availability statement. The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Supporting information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Table S1. Association tests for each SNP under codominant, dominant, recessive, overdominant and additive genetic models.
Table S2. Pancreatitis 30-day complication coding.
Table S3. Procedures with or within 30-day of acute pancreatitis diagnosis.
Table S4. Logistical regression model of hyperlipidaemia and the odds of severe acute pancreatitis vs non-severe acute pancreatitis. With additional adjustment of covariates for age (per five year increase), sex (male vs female), BMI (per unit increase), smoking status (current), Townsend deprivation index 5 (most deprived), chronic obstructive pulmonary disease, diabetes, chronic kidney disease, cerebrovascular accident, and cardiovascular disease (myocardial infarction and congestive cardiac failure).
Table S5. Association between SNPs and comorbid disease.

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