Table 2 *MYO9B* haplotypes in Spanish patients with ulcerative colitis, Crohn's disease and controls

Rs2305767/ rs1457092/ rs2305764	UC (n = 1356) n (%)	CD (n = 1254) n (%)	Controls (n = 1980) n (%)	UC vs controls	CD vs controls	UC vs CD
GCG	516 (38)	563 (45)	844 (43)	p = 0.008 OR = 0.83 (95% CI 0.72 to 0.95)	p = 0.20 OR = 1.10 (95% CI 0.95 to 1.27)	p = 0.0004 OR = 0.75 (95% CI 0.64 to 0.89)
AAA	549 (40)	424 (34)	713 (36)	p = 0.009 OR = 1.21 (95% CI 1.05 to 1.40)	p = 0.20 OR = 0.91 (95% CI 0.78 to 1.06)	p = 0.0004 OR = 1.33 (95% CI 1.13 to 1.57)
ACG	226 (17)	223 (18)	347 (18)	p = 0.52 OR = 0.94 (95% CI 0.78 to 1.14)	p = 0.85 OR = 1.02 (95% CI 0.84 to 1.23)	p = 0.45 OR = 0.92 (95% CI 0.75 to 1.14)
ACA	42 (3.1)	24 (1.9)	46 (2.3)	p = 0.17 OR = 1.34 (95% CI 0.86 to 2.10)	p = 0.44 OR = 0.82 (95% CI 0.48 to 1.39)	p = 0.05 OR = 1.64 (95% CI 0.96 to 2.81)
Rare haplotypes	23 (1.7)	20 (1.6)	30 (1.5)	p = 0.68 OR = 1.12 (95% CI 0.63 to 2.00)	p = 0.86 OR = 1.05 (95% CI 0.57 to 1.93)	p = 0.84 OR = 1.06 (95% CI 0.56 to 2.03)

Haplotypes were estimated by the expectation-maximisation algorithm implemented in the Arlequin software.
CD, Crohn's disease; UC, ulcerative colitis.

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Cathepsin B gene polymorphism Val26 is not associated with idiopathic chronic pancreatitis in European patients

Pancreatitis is thought to be a disease of autodigestion triggered by premature and intracellular activation of digestive proteases.[1] We and others have shown that lysosomal cathepsin B can activate trypsinogen intracellularly[2] and that a large proportion of pancreatic cathepsin B is physiologically sorted into the secretory compartment.[3] Further support for a role of cathepsin B in pancreatitis came from a recent study published by Mahurkar and coworkers in *Gut*[4] in which the authors reported that a leucine to valine mutation at position 26 of cathepsin B (L26V) is associated with tropical calcifying pancreatitis (odds ratio ~2.2) in patients from southern India. Tropical calcifying pancreatitis is also associated (in up to 50% of cases) with mutations in the SPINK1 gene[5]—with N34S being the most common mutation. In the study by Mahurkar *et al* the cathepsin B L26V mutation was, however, equally as common in SPINK1 N34S patients as in SPINK1 wild type patients, which suggests that cathepsin B is involved in an independent disease causing mechanism for pancreatitis.

As idiopathic chronic pancreatitis in Western countries and tropical calcifying pancreatitis in India share a high prevalence of SPINK1 mutations,[5] we investigated whether the former is also associated with the L26V cathepsin B mutation. We studied 64 patients with idiopathic chronic pancreatitis (ICP, defined as having unequivocal morphological evidence of chronic pancreatitis on computed tomography or endoscopic retrograde cholangio-pancreatography, or both) from northern Germany (aged 3 to 68 years; 38 male, 26 female) and 100 locally recruited healthy control subjects according to a recently reported ethics committee approved protocol.[6] Patients with known risk factors for pancreatitis, such as a history of regular alcohol consumption (more than two drinks or 20 g a day), or with biliary, metabolic, or endocrine disorders, cystic fibrosis, or hereditary pancreatitis were excluded.

Genomic DNA was extracted from blood leucocytes and polymerase chain reaction amplification of exons 2 and 3 of the cathepsin B gene was undertaken using specific oligonucleotides (sense: CGA GAC GGT GCC CCT GTG TGT G; antisense: GAG GCC TTC ACT CTC CCA CTT CC). Sequencing of cathepsin B exons in ICP patients identified 31 heterozygous and 10 homozygous Val26 alleles (allele frequency 0.398), while in the control cohort we found 46 heterozygous and 25 homozygous Val26 mutations (allele frequency 0.48; table 1). To our surprise and in contrast to the study by Mahurkar *et al*,[4] the allele frequency of cathepsin B Val26 appeared to be even higher among controls than among ICP patients.

Being faced with a Val26 allele frequency higher in Western control subjects than in Indian pancreatitis patients, we searched the SNP Genbank of NCBI for ethnic cohorts in whom the frequency of the Val26 variant had been reported. From the reported 16 groups of diverse ethnic backgrounds, the nine largest cohorts (n >40) were selected, comprising 1198 individuals. The according frequencies for C–G mutations (C is replaced by G in Val26) at codon 26 are given in table 2.

Table 1 Allele frequency of the Val26 mutation in German cohorts of ICP patients and controls

	n	26V allele frequency	OR	95% CI	p Value
Controls	100	0.48	0.718	0.45 to 1.15	0.147
Patients	64	0.398			

CI, confidence interval; ICP, idiopathic chronic pancreatitis; OR, odds ratio.

Table 2 Allele frequencies of Val26 mutations in individual ethnic groups

Population	Ethnic origin	(n)	pG (Val26)
CEPH	Mixed white	92	0.320
Pooled_CEPH	White	94	0.323
HapMap-CEU	European	55	0.355
SC_95_C	White	45	0.367
HapMap-YRI	Sub-Saharan African	52	0.394
TSC_42_C	White	41	0.430
JBIC-allele	Japanese	732	0.493
HapMap-JPT	Asian	44	0.523
HapMap-HCB	Asian	43	0.547
Total		1198	0.452±0.042

see refSNP ID: rs12338 at http://www.ncbi.nlm.nih.gov/SNP/snp_ref.cgi?rs=12338.

Table 2 shows wide variation in frequency (0.32 to 0.547), with Val26 being the commonest allele in two of the cohorts. The mean Val26 frequency of 0.452 (n = 1198; 95% confidence interval, 0.410 to 0.494) approaches that of our German control subjects (0.48) and is not different from our pancreatitis patients.

We conclude that a high frequency of Val26 in a given cohort merely represents a polymorphic allele variation and not a susceptibility factor for idiopathic chronic pancreatitis. While this is likely to be the case in most ethnic groups it does not explain the findings of Mahurkar and coworkers,[4] who found significantly fewer Val26 alleles in two Indian control cohorts. Their findings may either indicate subtle differences in the ethnic background of their patients and controls or they could be explained by a specific role for cathepsin B in tropical rather than idiopathic chronic pancreatitis. Further investigations addressing the role of cathepsin B in more common varieties of pancreatitis and in different ethnic groups are still urgently needed.

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Familial association of benign pancreatic hyperenzymaemia and pancreatic cancer

Benign pancreatic hyperenzymaemia (BPH) is a syndrome characterised by a chronic increase in serum pancreatic enzymes in the absence of pancreatic disease.[1–5] In March 2000 we saw a woman who had this form of hyperenzymaemia, whose mother had died of pancreatic cancer. Prompted by this observation, we undertook a prospective study to determine whether there is a familial association between BPH and pancreatic cancer. Between June 2000 and October 2006, we saw 68 subjects with BPH (42 male, 26 female; age range 8 to 72 years, mean age 45.7 years). They were questioned about the presence of pancreatic cancer in members of their families. The subjects who had relatives with pancreatic cancer and their immediate family members were included in the study. In addition to serum pancreatic enzymes, all underwent magnetic resonance imaging (MRI) with secretin stimulation and endoscopic ultrasound for detailed study of the Wirsung duct and pancreatic parenchyma.

Among the 68 subjects who had BPH, six had relatives who had died of pancreatic cancer. The demographic and clinical characteristics of these six subjects are shown in table 1. MRI was normal in all six, while endoscopic ultrasonography was normal in five; in the sixth (subject 6), parenchymal abnormalities (lobularity and hyperechoic strands) were present in the head of the pancreas. Two of the 10 relatives of these six subjects had pancreatic hyperenzymaemia; in both these subjects, MRI and endoscopic ultrasonography results were normal (table 1). Among the six study subjects there were nine relatives who had died of pancreatic ductal cancer (table 1). Four of the six had only one relative; one (subject 2), had two family members with pancreatic cancer, and another (subject 6) had three relatives with pancreatic cancer. The diagnosis of pancreatic cancer was based on histology in seven of the nine subjects, and on clinical and imaging findings in the other two.

Our findings show that there may be a familial association between BPH and pancreatic cancer. The significance of this association is not clear. BPH probably results from a defect in the passage of enzymes from acinar cells into the blood which results in greater than normal quantities in the circulation.[4] The anomaly is benign, generally being discovered during a routine blood analysis which includes amylase determination. At this point it is not known whether the occurrence of BPH in a family member or a relative of someone who has pancreatic cancer is incidental or the result of an underlying connection between the two conditions.

In order to see whether there were pancreatic lesions in subjects included in the study, they underwent MRI with secretin stimulation as well as endoscopic ultrasonography, which are generally sensitive for the detection of even small pancreatic lesions.[6] MRI was normal in all six subjects, while endoscopic ultrasonography was normal in all but one; this subject had three family members who had died of pancreatic cancer. The parenchymal abnormalities were lobularity and hyperechoic strands in the pancreatic head—lesions that have been seen by other investigators[7–9] in relatives of patients with pancreatic cancer, primarily subjects who had two or more relatives with the tumour. These changes, together with others,[7–9] have been shown to be associated with pancreatic dysplasia, a precursor lesion of pancreatic cancer.[10] Our study subject who had pancreatic abnormalities on endoscopic ultrasonography will be carefully followed, both clinically and ultrasonographically.

In conclusion, our study shows that persistent pancreatic hyperenzymaemia may be encountered in family members of patients with pancreatic cancer. The significance of this association is not clear, but it is possible that these individuals could be at increased risk of pancreatic cancer.

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