Table 2. Multivariable logistic regression analysis, sensitivity, specificity, PPV, and positive likelihood ratio of different HDL cutoff values predicting the presence of celiac disease

HDL cutoff value	Adjusted[a] OR	ROC curve analysis for CD prediction	Sensitivity (%)	Specificity (%)	PPV (%)	LR (+)
≤25 mg/dl	NC[b]	AUC=0.9, CI (0.84–0.95) $P<0.0001$	16	100	100	+
≤30 mg/dl	OR=8, CI (2–32) $P=0.004$	AUC=0.7, CI (0.57–0.84) $P=0.004$	22	96	61	6
≤35 mg/dl	OR=4, CI (1–11) $P=0.01$	AUC=0.61, CI (0.5–0.72) $P=0.04$	30	87	42	3
≤40 mg/dl	OR=5, CI (2–12) $P=0.001$	AUC=0.61, CI (0.5–0.7) $P=0.01$	48	77	39	2

AUC, area under the curve; CD, celiac disease; CI, confidence interval; LR, likelihood ratio; NC, not calculated; OR, odds ratio; PPV, positive predictive value; ROC, receiver operating characteristic.

[a]Adjusted for body mass index and the use of lipid-lowering medication.

[b]Since none of the patients in the non-celiac group had HDL ≤25 mg/dl and the specificity was 100%, the multiple logistic regression analysis for this HDL cutoff value could not be calculated.

tive predictive value of 61% and a positive likelihood ratio of 6.

Our study shows that IDA patients with CD have significantly lower HDL levels compared with IDA patients without CD, and that this finding could be used to improve identification of CD. Screening for CD should at least be done for IDA patients who present with HDL levels ≤40 mg/dl with an expectation that those with levels ≤30 mg/dl would have a greatly increased likelihood of CD positivity.

It is notable that total plasma cholesterol levels (TC) were significantly lower in IDA patients with CD than in IDA patients without CD. This is in accordance with previous observations correlating low TC levels with the presence of CD in hypochromic anemia patients, suggesting that it could be used to select anemic patients for CD screening (6). However, after adjusting for age, gender, BMI, and the use of lipid-lowering medications in our study, TC levels, unlike HDL levels, were not significantly associated with the presence of CD in IDA patients.

The limitations to this study include its single-center setting at a tertiary referral center and lack of a validation cohort. The strengths include the use of biopsy-proven CD patients, a sample size providing >90% power, and that the values reported are those at diagnosis. Future studies should prospectively test this approach of case-finding patients with IDA at high risk for CD.

CONFLICT OF INTEREST

Guarantor of the article: Peter H. Green, MD.

Specific author contributions: Hussein Abu Daya: concept and design, acquisition of data, analysis and interpretation of data, drafting of the manuscript, critical revision of the manuscript for important intellectual content, statistical analysis, final approval of the version to be published; Benjamin Lebwohl, Peter H. Green: concept and design, analysis and interpretation of data, critical revision of the manuscript for important intellectual content, final approval of the version to be published; Scott Smukalla: acquisition of data, analysis and interpretation of data, critical revision of the manuscript for important intellectual content, final approval of the version to be published; Suzanne K. Lewis: analysis and interpretation of data, critical revision of the manuscript for important intellectual content, final approval of the version to be published.

Financial support: None.

Potential competing interests: None.

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Part of this research was submitted to the AGA meeting (DDW) in Orlando 2013.

Genome-Wide Association Study Identifies Two Novel Genomic Regions in Irritable Bowel Syndrome

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doi:10.1038/ajg.2014.56

To the Editor: Irritable bowel syndrome (IBS) is a common, poorly understood gastrointestinal disorder. Although family

and twin studies broadly support the existence of a genetic component of IBS risk (1), previous candidate gene association studies had limited capacity to identify novel genes. Genome-wide association studies (GWAS), which have revealed new risk genes for various complex diseases, have not yet been published for IBS.

To assess the utility of GWAS for IBS, we conducted a pilot study in an Australian cohort (2). IBS was defined using Rome III criteria, with genome-wide genotyping performed using an Affymetrix Kaiser Axiom array. Quality control (QC) excluded single-nucleotide polymorphisms (SNPs) with a call rate <95%, minor allele frequency <1%, or Hardy–Weinberg equilibrium P value $<1\times10^{-6}$. Excluded samples demonstrated a call rate <95%, discrepant clinical and genotypic gender, outlying autosomal heterogygosity, cryptic relatedness (first degree) with a second sample yielding higher quality data, or non-European ancestry based on Eigenstrat principal components analysis. Following QC and genotype imputation using HapMap Phase II European reference data, ~2.5 million SNPs were available for testing in 172 IBS cases and 1,398 controls. Statistical analyses used an additive logistic model adjusted for the first two ancestry principal components; test statistics were also corrected for potential population stratification by genomic control (3). Results were compared with a prespecified significance threshold of 5×10^{-8}.

We found no evidence of confounding by population substructure or other artifacts (genomic inflation $\lambda_{GC}=1.000$). Peak association was observed for a cluster of 21 perfectly correlated SNPs on chromosome 10 (**Figure 1**), each of which showed genome-wide significant association with IBS ($P\sim9\times10^{-9}$). These SNPs span a 9-kb region centered on exon 11 of the protocadherin 15 (*PCDH15*) gene, encoding an integral membrane protein mediating calcium-dependent cell–cell adhesion. The peak genotyped SNP (rs10825269) is a nonsynonymous variant (p. Gly380Ser) within one of the extracellular cadherin (CA) repeats mediating cell–cell contact. The risk allele (T) has a frequency of 12% in European populations (per-allele odds ratio (OR) = 2.25; 95% confidence interval (CI): 1.71–2.98). Sequence conservation analysis (SIFT) suggests that the Gly380Ser amino-acid substitution is tolerated, but it identifies two variants in very close proximity (p. Ala379Thr and Asn389Ser) as deleterious to PCDH15 protein function; these were not tested in our study but are candidate pathogenic variants that may explain the observed association.

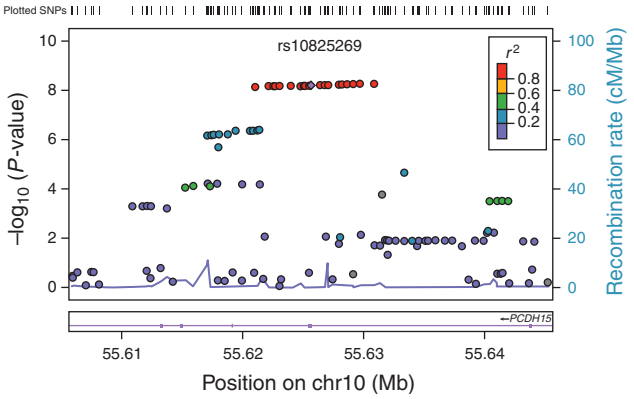

Figure 1. Regional association plot for the chromosome 10 region associated with overall irritable bowel syndrome. SNP, single-nucleotide polymorphism.

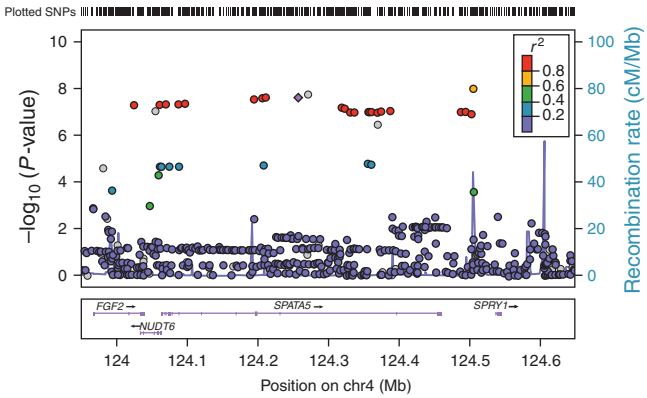

Figure 2. Regional association plot for the chromosome 4 region associated with irritable bowel syndrome-diarrhea. SNP, single-nucleotide polymorphism.

We attempted to replicate this association via *de novo* custom genotyping (Sequenom platform) in two European-ancestry IBS samples: (i) 390 IBS cases and 393 controls from the United States, and (ii) 95 IBS cases and 323 controls from Sweden. A directionally consistent effect was observed in the US sample, whereas the Swedish sample showed no effect. Meta-analysis of the three samples yielded an overall P value of 5×10^{-6} and summary OR of 1.55 (95% CI: 1.28–1.87).

Expression of *Pcdh15* mRNA has been detected in the sensory and neural epithelium and in both gastrointestinal epithelium and surrounding muscle during murine development (4). In humans, *PCDH15* mutations are involved in Mendelian syndromes of cochlear and retinal defects. Their potential role in IBS pathophysiology remains to be confirmed.

Pilot analyses of the diarrhea predominant subtype (IBS-D) were also performed using 56 cases and 1398 controls ($\lambda_{GC}=1.001$). A group of correlated SNPs spanning a 500-kb region on chromosome 4 showed genome-wide significant association with IBS-D (peak $P=2.5\times10^{-8}$ at rs9999118). Replication data for this

SNP were available in the Swedish sample (45 IBS-D cases and 323 controls), which showed a directionally consistent, although attenuated, effect (meta-analysis summary $P=7.9\times10^{-8}$). The risk allele (G) has an allele frequency of 2.5% in Europeans, and was associated with a large summary OR of 7.3, although the 95% CI was wide (3.53–15.03). This chromosome 4 region (see **Figure 2**) contains several genes including fibroblast growth factor 2 (*FGF2*), the overlapping *NUDT6* gene, thought to regulate FGF2 expression, and *SPRY1*, encoding a negative regulator of FGF signaling. FGF2 has a major role in stimulating the regeneration of gastric and colorectal mucosa following inflammation or bacterial infection (5). Variants in this region may thus moderate local FGF2 levels and thus FGF2-mediated gastrointestinal tissue regeneration following infection or inflammation.

Our pilot GWAS was limited by its modest size, and these results require validation in larger samples. However, our findings suggest the presence of common, biologically plausible genetic risk variants for IBS. Future, well-powered GWAS of IBS are likely to reveal new etiological mechanisms for this disorder.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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A Comparison of Patient Satisfaction With Sedation Between Fentanyl/Midazolam and Meperidine/Midazolam in Patients Undergoing Endoscopy

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doi:10.1038/ajg.2014.31

To the Editor: The use of an opioid combined with midazolam to achieve conscious sedation in gastrointestinal endoscopy is widely practiced, producing a hypothetical synergistic effect and allowing for the use of smaller doses of benzodiazepines during endoscopic procedures (1). Data suggest that endoscopists in the United States are nearly evenly split in their use of fentanyl or meperidine (56 vs. 53%) (2). Recent literature has attributed shorter procedure and recovery times to the use of fentanyl; yet, little study has been done to evaluate patient satisfaction (3).

The primary aim of this study was to evaluate whether differences exist in patient satisfaction between groups receiving fentanyl/midazolam and those receiving meperidine/midazolam for general endoscopy conscious sedation. In this quality survey study, answers to the 20-question Patient Satisfaction with Sedation Instrument (PSSI), which has been previously validated (4), were collected during the spring of 2010 from 198 patients who received either fentanyl/midazolam ($N=91$) or meperidine/midazolam ($N=107$) during endoscopy. Sedation was selected on the basis of the endoscopist's preference. Patients were handed a paper copy of the voluntary questionnaire after the procedure, which was returned by posted mail. The total score and sedation subscale scores were calculated according to the PSSI guidelines; higher scores indicate increased satisfaction.

There were two significant differences in PSSI responses between the two groups (**Table 1**). Patients in the meperidine/midazolam group were more often "very satisfied" with the length of time for which they felt the effects of the sedation (77 vs. 65%, $P=0.048$). Also, patients in the meperidine/midazolam group responded "very satisfied" more often to how they would compare their satisfaction with this experience with that of their previous experience with endoscopy (86 vs. 71%, $P=0.014$). In the PSSI subscale scores between the two sedation groups (**Table 2**), results were similar except for the sedation-delivery subscale score, which was significantly higher in meperidine/midazolam patients compared with fentanyl/midazolam patients ($P=0.034$).

We observed that patients were overall satisfied with sedation in both groups. However, there were several questions for which answers showed that patients who received meperidine/midazolam were more satisfied than were patients who received fentanyl/midazolam, and where differences either reached statistical significance or trended toward significance. Patients receiving meperidine were significantly more satisfied with the length of time for which sedation effects were felt, following both esophagogastroduodenoscopy (EGD) and colonoscopy. Therefore, although fentanyl may be associated with a shorter recovery time following endoscopy, this brings into question whether its shorter half-life correlates with decreased patient satisfaction. In addition, we found that patients who had undergone previous endoscopic procedures were more satisfied with their endoscopic experiences after receiving meperidine. This difference, in favor of meperidine use, was also found to be statistically significant. In addition, the meperidine/midazolam group required on average less midazolam, both overall and when sorted by procedure type (EGD, colonoscopy, or both).