Diabetes in First-Degree Family Members: A Predictor of Type 2 Diabetes in 159 Nonscreening Alabama Hemochromatosis Probands With *HFE* C282Y Homozygosity

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OBJECTIVE

We sought to identify predictors of diabetes diagnosed before hemochromatosis.

RESEARCH DESIGN AND METHODS

We studied these 16 variables in 159 nonscreening hemochromatosis probands with *HFE* C282Y homozygosity: age; sex; BMI; diabetes reports in first-degree family members (dichotomous); heavy ethanol consumption; cigarette smoking; elevated serum alanine aminotransferase/aspartate aminotransferase levels; nonalcoholic fatty liver; chronic viral hepatitis; cirrhosis; hand arthropathy; iron removed by phlebotomy; and positivity for HLA-A*01, B*08; A*03, B*07; and A*03, B*14 haplotypes. We performed univariable and multivariable analyses.

RESULTS

Twenty-three probands (14.5%) had diabetes; 19 were men. Each of the 23 probands had type 2 diabetes. Mean BMI was greater in probands with diabetes (31.7 ± 8.5 [SD] kg/m^2 vs. 27.6 ± 5.1 kg/m^2; *P* = 0.032). Reports of any first-degree family member with diabetes were more prevalent in probands with than in probands without diabetes (69.6 vs. 17.6%; *P* < 0.0001). In probands with diabetes, the odds ratio (OR) of maternal diabetes was 6.7 (95% CI 2.3–19.7; *P* = 0.0005) and of sibling diabetes was 11.7 (3.0–45.5; *P* = 0.0004). In a logistic regression model, predictors of diabetes at hemochromatosis diagnosis in 159 probands were diabetes reports in family members (OR 8.5 [95% CI 2.9–24.8]; *P* < 0.0001) and BMI (1.1 [1.0–1.2]; *P* = 0.049). This model explained 26.0% of total deviance contributing to diabetes.

CONCLUSIONS

In nonscreening hemochromatosis probands with *HFE* C282Y homozygosity, a heritable factor(s) increases the risk of diabetes diagnosed before hemochromatosis.

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Hemochromatosis due to homozygosity for the C282Y mutation of the *HFE* gene on chromosome 6p21.3 occurs in 0.3–0.6% of people of Northwestern European descent (1–3). Iron overload, especially if severe, may cause cirrhosis, primary liver cancer, diabetes, other endocrinopathy, and cardiomyopathy (3,4). A diagnostic triad for hemochromatosis of hyperpigmentation, diabetes, and cirrhosis was proposed in the 19th century (5). Reports from the late 19th century to the mid-20th century demonstrated that ∼80% of patients with hemochromatosis had diabetes, typically associated with cirrhosis and severe hemosiderin deposition and associated morphologic abnormalities in the pancreatic acini and islets of Langerhans (6,7). Relative specificity of iron deposition for the β-cells of the pancreatic islets in hemochromatosis was described in 1956 (8).

In 1968, Balcerzak et al. (9) reported that the prevalence of diabetes was high (47%) in people with normal iron stores in five hemochromatosis kinships. In 1972, Dymock et al. (10) reported that 25% of 68 hemochromatosis patients with diabetes had first-degree relatives who also had diabetes, whereas only 4% of 42 hemochromatosis patients without diabetes had a first-degree relative with diabetes. These observations, analyzed with univariable techniques, suggested that a heritable factor(s) increases the risk of diabetes in people with hemochromatosis.

The diagnosis of hemochromatosis changed in 1996 with the discovery of the *HFE* gene and the C282Y polymorphism (1). Thereafter, more people with abnormal iron phenotypes were confirmed to have hemochromatosis using a genetic criterion (C282Y homozygosity) than with the traditional diagnostic triad, although ∼10% of Western European whites with hemochromatosis phenotypes do not have C282Y homozygosity (1). In two hemochromatosis case series from the 21st century (11,12), the prevalence of diabetes in people with hemochromatosis phenotypes was much lower, i.e., 23 and 22%, respectively, than typically reported in the 20th century.

In one study, all subjects were referred from a hemochromatosis clinic (11). In the other study, subjects were ascertained due to clinical suspicion, family screening, or detection of hyperferritinemia on routine health checks (12). The proportions of hemochromatosis subjects with *HFE* C282Y homozygosity in the two reports were 87% (26 of 30) and 78% (185 of 237), respectively (11,12). In the second report, neither serum ferritin (SF) level at diagnosis nor cirrhosis nor *HFE* genotype was significantly associated with the occurrence of diabetes in people with hemochromatosis phenotypes by univariable analyses (12). Family history of diabetes was not evaluated in either of these reports (11,12).

Thus, a critical multivariable assessment of the relationships of the type(s) of diabetes, severity of iron overload, occurrence of liver disease, family history of diabetes, and other variables in people with hemochromatosis and *HFE* C282Y homozygosity has not been reported (4). We sought to identify factors associated with increased risk of diabetes at diagnosis in 159 nonscreening hemochromatosis probands, each of whom also had *HFE* C282Y homozygosity. The basis of hemochromatosis diagnosis in each proband was an abnormal iron phenotype discovered in a medical care setting, not in population or clinic screening or in a family study. Available independent variables were age, sex, BMI, proband reports of diabetes in first-degree family members, heavy ethanol consumption, cigarette smoking, concomitant liver disorders, hand arthropathy, quantities of iron removed by phlebotomy (QFe), and common HLA-A and -B haplotypes. Our results are discussed in the context of previous descriptions of the relationships of iron overload, comorbid conditions, and family history in the occurrence of diabetes in *HFE* hemochromatosis.

RESEARCH DESIGN AND METHODS
Selection of Hemochromatosis Probands
The performance of this work was approved by the institutional review board of Brookwood Medical Center. We conducted computerized and manual searches of medical records to identify patients who were evaluated for hemochromatosis because they had elevated values of transferrin saturation (TS) or SF. Each person selected for this study was a white adult (>18 years of age) and the first in his/her respective family to be diagnosed to have hemochromatosis (proband). We included probands who *a*) were diagnosed to have hemochromatosis during routine medical care, not as a consequence of screening; *b*) had elevated SF levels at diagnosis (men >300 μg/L; women >200 μg/L) (2,3); *c*) had genotype *HFE* C282Y/C282Y; *d*) had undergone HLA-A and -B haplotyping; *e*) completed iron depletion therapy by phlebotomy; and *f*) resided in central Alabama. Each proband was diagnosed to have hemochromatosis in the interval 1976–2010. Each proband was evaluated for iron overload and associated complications, as appropriate (3). BMI was computed as described previously (13). Probands with BMI 25–29 kg/m^2 and ≥30 kg/m^2 were defined as overweight and obese, respectively.

Laboratory Methods
Levels of TS, SF, serum alanine aminotransferase (ALT), and serum aspartate aminotransferase (AST) were measured using automated clinical methods. *HFE* mutation analysis was performed as previously described (14) using current or archived DNA specimens. We studied HLA-A and -B haplotypes HLA-A*01, B*08; A*03, B*07; and A*03, B*14 in the present probands because these haplotypes influence iron phenotypes in C282Y homozygotes (15–18) and because other loci in linkage with HLA on chromosome 6p influence risk of conversion from impaired glucose intolerance to type 2 diabetes (19). HLA-A and -B alleles were detected using low-resolution DNA-based typing (PCR/sequence-specific oligonucleotide probe) in probands and family members. In each proband in whom a single A or B allele was detected by DNA-based typing, we verified the allele(s) and set phase to ascertain haplotypes of the proband using HLA analyses of

appropriate first-degree family members (parents, full siblings, and children). Assigning paternal or maternal haplotypes for each proband was not possible because the parents of many probands were dead or otherwise unavailable. For the present analysis, all haplotypes were defined only by A and B alleles (20). Sections of liver biopsy specimens were stained using hematoxylin and eosin, Masson's trichrome, and Perls' Prussian blue techniques. Intrahepatocytic iron was graded according to the method of Scheuer et al. (21). Routine methods were used to detect HB$_s$Ag, HB$_s$Ab, HB$_c$Ab, and hepatitis C antibody.

Diabetes in Hemochromatosis Probands
Probands with diabetes were identified and characterized by referring physicians, our queries regarding diabetes, and medication reviews. Probands were diagnosed to have diabetes before hemochromatosis was diagnosed. Diabetes was defined and subclassified according to the criteria of the American Diabetes Association (22). We excluded one proband with diabetes, a woman who had undergone pancreatectomy for management of adenocarcinoma of the pancreas.

Family History of Diabetes
Reports of first-degree relatives (parents, full siblings, and children) with diabetes were elicited from each proband and documented in his/her medical record at the time of initial evaluation.

Definitions of Smoking and Heavy Ethanol Consumption
Smoking was defined as the self-report of daily cigarette smoking for ≥5 years. Heavy ethanol consumption was defined as the self-reported consumption of ≥60 g ethanol per day for ≥5 years.

Definitions of Liver Conditions
Probands were classified as having elevated serum ALT or AST if either of their levels were higher than the respective upper reference limits (>2 SD above mean; >40 IU/L). Nonalcoholic fatty liver disease (NAFLD) was defined as steatosis or steatohepatitis detected on liver biopsy specimens or by typical increase of hepatic echogenicity detected by ultrasonography, in the absence of self-reports of heavy ethanol consumption. Chronic hepatitis B or C was defined as positivity for HB$_s$Ag or hepatitis C antibody, respectively, in association with other clinical or liver biopsy abnormalities consistent with chronic viral hepatitis. Liver biopsy was performed in all probands with SF >1,000 μg/L and in other probands whose evaluations suggested that they had an undiagnosed liver disorder other than that attributable to iron overload. Cirrhosis was defined by pathologist interpretations of liver biopsy specimens.

Definition of Hemochromatosis Hand Arthropathy
We evaluated this manifestation because it is typically associated with severe iron overload (4). Probands who had symptoms or physical examination findings in hand joints suggestive or typical of hemochromatosis arthropathy at diagnosis were evaluated with hand radiographs and other studies, as appropriate. The objective criteria we used to classify probands as having hemochromatosis hand arthropathy included *1*) symmetrical polyarthropathy, typically with hypertrophy and related abnormalities of metacarpophalangeal and proximal interphalangeal joints; *2*) subchondral cysts with sclerotic margins at the metacarpal heads, especially of the second and third metacarpophalangeal joints; and *3*) joint space narrowing, usually with asymmetric cartilage loss (4,23). Other abnormalities were detected in some probands, especially those with severe arthropathy (4,23).

Iron Removed by Phlebotomy
Iron depletion therapy, defined as the periodic removal of blood to eliminate storage iron, was complete when SF was ≤20 μg/L (3). QFe was estimated as 200 mg Fe per unit of blood (450–500 mL) (3).

Rates of Diabetes, Overweight, and Obesity in Probands and General Population
We compared prevalences of the characteristics diabetes diagnosis, overweight, and obesity in the present probands with those in the general population using 2010 data obtained online from the Behavior Risk Factor Surveillance System of the Centers for Disease Control and Prevention (data from Birmingham-Hoover, Alabama metropolitan area, 2010, http://www.cdc.gov/brfss/data_tools.htm, accessed 27 May 2013).

Statistics
Preliminary analyses revealed that TS, SF, and QFe values were not normally distributed. Accordingly, these data were converted to natural logarithms (ln) to normalize them for analyses, and anti-lns of the results were computed to display mean values (95% CIs). The Pearson correlation coefficient of lnTS with lnQFe was 0.2170 (one-tailed test; P = 0.0030). The Pearson correlation coefficient of lnSF with lnQFe was 0.6950 (one-tailed test; P < 0.0001). Thus, we used lnQFe as the only independent variable of iron overload severity in logistic regression analyses.

The final analytic data set consisted of observations on 159 probands. Fifteen independent variables for analysis were age, sex, BMI, reports of diabetes in first-degree family members, heavy ethanol consumption, cigarette smoking, elevated serum level of ALT or AST, NAFLD, chronic viral hepatitis B or C, cirrhosis, hand arthropathy, and iron removed by phlebotomy (lnQFe). In all analyses, we treated reports of diabetes in first-degree family members as a dichotomous variable (family member [s] with or without diabetes). Other dichotomous variables included sex; heavy ethanol consumption; cigarette smoking; elevated serum level of ALT or AST; NAFLD; chronic viral hepatitis B or C; cirrhosis; hand arthropathy; and positivity for HLA haplotypes A*01, B*08; A*03, B*07; and A*03, B*14.

General descriptive data are presented as enumerations, percentages, mean ± 1 SD, or mean (95% CI). Univariable comparisons between groups were evaluated using Student *t* test, Pearson χ^2 test, and Fisher exact test, as appropriate. Logistic regression analyses were performed to identify variables that are independent predictors of diabetes in probands (significant positive or negative

association). We also expressed these results as odds ratio (OR) (95% CI). Values of $P < 0.05$ were defined as significant and are displayed as two significant figures. A computer spreadsheet (Excel 2000; Microsoft Corp., Redmond, WA) and a statistical program (GB-Stat, version 10.0, 2000; Dynamic Microsystems, Inc., Silver Spring, MD) were used to perform the analyses.

RESULTS

General Characteristics of 159 Hemochromatosis Probands

There were 102 men (64.2%) (Table 1). The proportion of men, mean BMI, and prevalence of diabetes reports in first-degree family members were higher in probands with diabetes than in probands without diabetes. We observed no other significant differences between probands with and without diabetes (Table 1).

Mean SF at diagnosis was greater in men (1,096 μg/L [95% CI 953–1,261]) than women (660 μg/L [549–795]) ($P < 0.0001$). The prevalence of elevated serum levels of AST, reports of heavy ethanol consumption, occurrence of chronic viral hepatitis (six hepatitis C and one hepatitis B), and reports of

cigarette smoking were greater in men than women ($P = 0.016, 0.0002, < 0.0001$, and 0.004, respectively). Mean QFe was greater in men (5.9 g [95% CI 5.2–6.6]) than women (3.0 g [2.5–3.7]) ($P = < 0.0001$).

Diabetes Classification and Treatment

Twenty-three probands (14.5%) had diabetes at diagnosis of hemochromatosis; 19 of the 23 probands (82.6%) were men. There were no significant differences between the general characteristics of the 19 men and the 4 women with diabetes (data not shown).

Each of the 23 probands with diabetes had type 2 diabetes. Diabetes in 19 of the 23 probands (82.6%) was treated with oral hypoglycemic agents only. Three other probands were treated with oral hypoglycemic agents and insulin. One man was treated with insulin alone. Characteristics of the 19 probands treated without insulin and the 4 probands treated with insulin did not differ significantly (data not shown).

Diabetes Reports in First-Degree Family Members

OR is the ratio of the odds of diabetes reports in family members in the two

proband groups (with and without diabetes). The likelihood of probands with diabetes having a paternal history of diabetes did not differ significantly from that of probands without diabetes (OR 1.0 [95% CI 0.2–4.7]). Probands with diabetes were much more likely than probands without diabetes to have a maternal history of diabetes (6.7 [2.3–19.7]) or a sibling history of diabetes (11.7 [3.0–45.5]) (Table 2).

Logistic Regression on Diabetes at Diagnosis

We performed a multiple logistic regression on diabetes at diagnosis of hemochromatosis using 16 independent variables. The only variable significantly associated with diabetes in probands was reports of diabetes in first-degree family members (OR 10.9 [95% CI 3.4–35.2]; $P < 0.0001$). This 16-factor model explained 30.7% of total deviance contributing to diabetes at diagnosis of hemochromatosis ($\chi^2 = 41.39$; $P = 0.0005$).

To obtain the best fitting model while minimizing the number of independent variables, we performed likelihood ratio tests and determined that only age, male sex, BMI, family history of diabetes, and heavy ethanol consumption would be possible significant contributors to a reduced model. Using these five observations as independent variables, a reduced logistic regression model demonstrated that only family history of diabetes and BMI were significant predictors of diabetes ($P < 0.0001$ and $P = 0.049$, respectively). The corresponding ORs and 95% CIs for these independent variables were 8.5 (2.9–24.8) and 1.1 (1.0–1.2). This five-factor model explained 26.0% of total deviance contributing to diabetes at diagnosis of hemochromatosis ($\chi^2 = 34.90$; $P < 0.0001$).

In another analysis, we combined heavy ethanol consumption, elevated serum ALT/AST levels, nonalcoholic fatty liver, chronic viral hepatitis, and cirrhosis observations to create the single aggregate variable "liver-related variables" but we retained all other variables. A regression on diabetes using these 11 variables revealed that OR was significant only for reports of diabetes in

Table 1—Characteristics of 159 nonscreening hemochromatosis probands with *HFE* C282Y homozygosity

Characteristic	Diabetes ($n = 23$)	No diabetes ($n = 136$)	P value
Mean age at diagnosis, years (SD)	53 ± 12	49 ± 13	0.20
Male, % (n)	82.6 (19)	61.0 (83)	0.035
Mean BMI, kg/m² (SD)	31.7 ± 8.5	27.6 ± 5.1	0.032
Overweight (25–29 kg/m²), % (n)	43.5 (10)	39.7 (54)	0.73
Obese (≥30 kg/m²), % (n)	39.1 (9)	27.9 (38)	0.28
Family history of diabetes, % (n)	69.6 (16)	17.6 (24)	<0.0001
Smoking, % (n)	39.1 (9)	22.8 (31)	0.095
Heavy ethanol intake, % (n)	8.7 (2)	15.4 (21)	0.31
Elevated ALT, % (n)	21.7 (5)	20.0 (27)	0.51
Elevated AST, % (n)	26.1 (6)	25.0 (34)	0.91
NAFLD, % (n)	21.7 (6)	25.0 (34)	0.74
Viral hepatitis, % (n)†	4.3 (1)	4.4 (6)	0.73
Cirrhosis, % (n)	17.4 (4)	12.5 (17)	0.36
Hand arthropathy, % (n)	39.1 (9)	25.7 (35)	0.18
Mean QFe, g (95% CI)	4.0 (3.0–5.5)	4.0 (3.5–4.5)	0.59
HLA-A*01, B*08, % (n)	17.4 (4)	7.4 (10)	0.12
HLA-A*03, B*07, % (n)	29.2 (6)	27.2 (37)	0.91
HLA-A*03, B*14, % (n)	4.3 (1)	7.4 (10)	0.49
No designated HLA haplotype, % (n)‡	52.2 (12)	63.2 (86)	0.31

†Chronic viral hepatitis B or C. ‡No HLA haplotype A*01, B*08; A*03, B*07; or A*03, B*14.

Table 2—ORs of diabetes in 159 hemochromatosis probands based on diabetes reports in first-degree relatives

Report of diabetes	Probands with diabetes (n = 23)	Probands without diabetes (n = 136)	OR (95% CI)	P value
Father	2	12	1.0 (0.2–4.7)	0.98
Mother	8	10	6.7 (2.3–19.7)	0.0005
Sibling(s)	6	4	11.7 (3.0–45.5)	0.0004

first-degree family members (OR 10.6 [95% CI 3.4–33.1]; $P < 0.0001$). This 11-factor model explained 19.5% of total deviance contributing to diabetes at diagnosis of hemochromatosis ($\chi^2 = 35.02$; $P = 0.0002$).

Rates of Diabetes, Overweight, and Obesity in Probands and General Population

Comparisons of the present probands with those of a 2010 general population sample from the Birmingham-Hoover metropolitan area of Alabama demonstrate that the prevalence rates of diabetes diagnosis, overweight, or obese in the probands did not differ significantly from those of the general population sample (Table 3).

Diabetes and Overweight/Obesity Prevalence by Year of Diagnosis

Sixty-eight probands were diagnosed to have hemochromatosis during the interval 1976–1995. Twelve (17.6%) had diabetes. Ninety-one probands were diagnosed to have hemochromatosis during the interval 1996–2010. Eleven (12.1%) had diabetes. Proportions of probands in the two subgroups did not differ significantly ($P = 0.32$).

In probands diagnosed to have hemochromatosis during the interval 1976–1995, 31 (45.6%) were overweight. In probands diagnosed during the interval 1996–2010, 33 (36.3%) were overweight ($P = 0.2356$). In probands diagnosed to have hemochromatosis during the interval 1976–1995, 12 (17.6%) were obese. In probands diagnosed during the interval 1996–2010, 35 (38.5%) were obese ($P = 0.0044$).

CONCLUSIONS

The present results demonstrate that each of the 23 probands with diabetes had type 2 diabetes and that there is a significant positive association of type 2 diabetes at diagnosis of hemochromatosis in nonscreening probands with *HFE* C282Y homozygosity with their reports of diabetes in first-degree family members, after correction for other variables. There was no significant statistical contribution of severity of iron overload, cirrhosis, or other liver-related variables to the occurrence of diabetes in this cohort whose diagnoses of hemochromatosis were ascertained in a uniform manner. In the 19th century through the latter 20th century, the predominant etiology of diabetes in whites with hemochromatosis was generally acknowledged to be severe pancreatic and hepatic iron overload. In the present hemochromatosis cohort, we demonstrate that the predominant etiology of type 2 diabetes may be the inheritance of nonhemochromatosis traits shared with first-degree relatives. These results confirm and extend the postulates of Balcerzak et al. (9) and Dymock et al. (10) that a heritable factor(s) increases risk of diabetes in people with and without hemochromatosis. In subjects unselected for hemochromatosis diagnoses, the risk of type 2 diabetes is also increased in those

who have positive family histories of diabetes (24–27).

In the current study, we relied on reports of diabetes in first-degree family members elicited from nonscreening probands at diagnosis of hemochromatosis. In studies of subjects selected without regard to hemochromatosis or iron overload diagnoses, the validity of patient self-reports of diabetes among relatives revealed sensitivities of >60% and specificities as high as 90% (28–31). In the Hemochromatosis and Iron Overload Screening (HEIRS) Study, the analytical validity of self-reported family history of hemochromatosis, iron overload, and related disorders was evaluated in 145 index participants with hemochromatosis or iron overload, 549 family members, and 641 control participants. The sensitivity and specificity for diabetes were 58 and 96%, respectively (32). Accordingly, the true prevalence of diabetes in first-degree family members in the current study may have been higher than that suggested by family history reports.

The OR for diabetes in the present probands was increased in association with reports of maternal diabetes. Maternally transmitted type 2 diabetes in Han Chinese and Finnish subjects has been linked to nonsynonymous mitochondrial DNA variants and rare mitochondrial haplotypes (33,34). Taken together, these observations suggest that mitochondrial DNA variants are one mechanism by which type 2 diabetes could be maternally transmitted to people with *HFE* hemochromatosis. The elevated OR for diabetes in the present probands with sibling reports of diabetes is consistent with the results of family history of diabetes studies in people unselected for hemochromatosis diagnoses (24–27).

Table 3—Prevalence of characteristics of Alabama adults

Characteristic	Population sample†	Hemochromatosis probands	P value
Diabetes diagnosis, % (95% CI) [n]	12.4 (10.2–14.5) [193]	14.5 (9.0–20.0) [159]	0.58
Overweight (25–29 kg/m^2), % (95% CI) [n]	37.0 (32.8–41.1) [399]	40.3 (32.7–47.8) [159]	0.49
Obese (≥30 kg/m^2), % (95% CI) [n]	29.3 (25.7–32.8) [350]	29.6 (22.5–36.7) [159]	0.98

†Data from Birmingham-Hoover, Alabama metropolitan area, 2010, http://www.cdc.gov/brfss/data_tools.htm, accessed 27 May 2013.

lnQFe was not significantly associated with diabetes in the present probands. In a series of *HFE* hemochromatosis cases, cirrhosis and hand arthropathy have significant positive associations with severity of iron overload (4,35,36). In the current study, neither cirrhosis nor hand arthropathy was significantly associated with diabetes at diagnosis of hemochromatosis. In hemochromatosis family members studied by Balcerzak et al. (9), there was little correlation of the severity of iron overload with the occurrence of diabetes. In Irish patients with hemochromatosis, neither SF level nor cirrhosis at diagnosis was significantly associated diabetes (12).

This case-case study evaluated the risk factors for type 2 diabetes in a cohort of hemochromatosis probands diagnosed in the interval 1976–1995. We also compared the prevalence rates of the characteristics diabetes diagnoses, overweight, or obesity in the present probands with those in a 2010 population sample of Birmingham-Hoover, Alabama metropolitan area. We observed no significant differences between the two groups. We were unable to locate other diabetes prevalence data for adult whites in central Alabama that would reveal relationships of diabetes and BMI that were pertinent to either the entire interval 1976–2010 during which we diagnosed the present probands or a midpoint in that interval. The present probands are white, whereas only 69.5% of the 2010 population sample was white. It is widely acknowledged that the prevalence of diabetes in Alabama adults has increased during the 1976–2010 interval during which we diagnosed the present probands.

The *HFE* genotype of people with hemochromatosis phenotypes in clinical and screening studies does not predict the occurrence of diabetes (12,32,37). In the current study, positivity for HLA haplotypes HLA-A*01, B*08; A*03, B*07; and A*03, B*14 was not associated with a significant OR for occurrence of diabetes in multiple logistic regressions, although these haplotypes influence iron phenotypes in C282Y homozygotes (15–18). The frequency of the C282Y or H63D mutations in patients with impaired glucose intolerance or type 2 diabetes was not increased in a population-based sample or in a meta-analysis of eight other studies (38). Taken together, these observations suggest that the role of common *HFE* mutations in the pathogenesis of diabetes in people with hemochromatosis and in the general population is limited and that the chromosome 6p–linked HLA haplotypes we studied may not bear alleles that are important in diabetes pathogenesis in *HFE* C282Y homozygotes.

In the current study, we demonstrate that the prevalence of type 2 diabetes in people with hemochromatosis phenotypes and *HFE* C282Y homozygosity is lower than the prevalence of diabetes reported in people with hemochromatosis phenotypes in the 20th century (4,6,7). We postulate that this is due to changing diagnostic criteria for *HFE* hemochromatosis (39), earlier diagnosis of hemochromatosis (12), greater use of phlebotomy therapy (3), and the awareness of greater heterogeneity in clinical and laboratory manifestations in people with C282Y homozygosity today than was recognized in people with hemochromatosis phenotypes decades ago (4).

Mean BMI of the present hemochromatosis probands with diabetes was significantly greater than that of probands without diabetes. In a reduced logistic regression model, BMI was a significant independent predictor of diabetes, although the OR was relatively low. We observed that some probands with diabetes were either overweight (44%) or obese (39%). We also observed that the prevalence of obesity was greater in probands diagnosed to have hemochromatosis in the interval 1996–2010 than the prevalence of obesity in probands diagnosed in the interval 1976–1995. McClain et al. (11) observed that Utah hemochromatosis patients with diabetes were either overweight (14%) or obese (86%). On the other hand, the proportions of the present probands who were overweight or obese did not differ significantly between those with diabetes and those without diabetes or between the probands and people in the 2010 Birmingham-Hoover population sample unselected for hemochromatosis diagnoses. In adults not selected for hemochromatosis diagnoses, it is generally acknowledged that the risk of type 2 diabetes is greater in subjects with increased BMI (40). It was beyond the scope of our work to compare the BMI of probands at the time of diabetes diagnosis with BMI at the time of hemochromatosis diagnosis.

The significant positive association of diabetes histories in first-degree family members of hemochromatosis probands demonstrated herein does not exclude the possibility that diabetes pathogenesis is heterogeneous in some, if not most, people with *HFE* hemochromatosis. More than 70% of the deviance in the present logistic regression model was not explained by first-degree family histories of diabetes or other independent variables we studied. In individual patients with hemochromatosis, siderosis of pancreatic β-cells, liver disease, inheritance of HLA-linked and non–HLA-linked diabetogenic alleles, and other heritable and acquired factors (4,6,7,22) may contribute to diabetes pathogenesis.

Evaluating observations on diabetes management and complications in probands was beyond the scope of the current study. Likewise, it was not possible to review medical records, BMI, iron measures, and other observations of the first-degree relatives of probands. Thus, we cannot exclude the possibility that BMI in first-degree family members of probands, rather than the proband family histories of diabetes and BMI alone, was a significant independent determinant of diabetes in probands.

We conclude that increased risk for diabetes at hemochromatosis diagnosis in nonscreening *HFE* C282Y homozygotes is associated with reports of diabetes in first-degree family members and BMI but not with cirrhosis or other liver disorders, other comorbid conditions, iron removed by phlebotomy, or common HLA haplotypes linked to *HFE* C282Y on chromosome 6p.

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