Hemochromatosis C282Y Mutation and Histological Fibrosis in Patients With C Virus Chronic Hepatitis

To the Editor:

We read with interest the article recently published in HEPATOLOGY,[1] which suggested that heterozygosity for hemochromatosis C282Y mutation could be associated with more fibrosis in patients with chronic active hepatitis (CAH) owing to hepatitis C virus (HCV). Liver iron accumulation has been observed in patients with HCV infection, and could negatively influence the natural course of HCV-related liver disease.[2] Recently, a homozygous missense mutation, C282Y, within the HFE gene located on the chromosome 6, was found to be present in 82% to 100% of the patients with genetic hemochromatosis.[3,4] The ability to detect this mutation on DNA extracted from peripheral blood cells allows the assessment of its role in diseases associated with liver iron accumulation. We studied 209 consecutive patients with histologically proven CAH caused by HCV (131 men, 78 women, mean age 44.3 ± 12.0 years) including 19 patients with cirrhosis (9.1%). Genomic DNA was extracted from peripheral leukocytes by standard methods. The C282Y mutation was screened for by restriction enzyme analysis of polymerase chain reaction–amplified products and by *Rsa* I digestion as described.[3] Histological fibrosis score was determined according to METAVIR classification. One patient was found to be homozygous for the C282Y mutation, with marked iron overload similar to that reported in genetic hemochromatosis and was therefore excluded for the assessment of the relationship between fibrosis and genotype at the HFE locus. The relationship between the C282Y mutation and fibrosis is shown in Table 1. The overall prevalence of heterozygous state for C282Y mutation (10.6%) was similar to that reported in the general population[1,3] and was not related to the degree of fibrosis. In addition, the prevalence of heterozygosity for C282Y mutation did not differ in patients with or without cirrhosis (15.8% vs. 10.2%, respectively, $P = .71$). Thus, in our experience based on a large number of patients, heterozygosity for C282Y was not related to the severity of fibrosis. The relatively small number of patients who were heterozygous for C282Y in the study of Smith et al.[1] could explain the discrepancy with our data. Our data are consistent with heterozygosity for C282Y having no influence on liver iron accumulation.[5]

TABLE 1. Relationship Between C282Y Mutation and Histological Fibrosis (METAVIR Classification) in Patients With HCV Chronic Hepatitis

C282Y Mutation	Normal	Heterozygous	Overall
Fibrosis			
0-1	112 (89.6%)	13 (10.4%)	125
2	35 (92.1%)	3 (7.9%)	38
3	23 (88.5%)	3 (11.5%)	26
4	16 (84.2%)	3 (15.8%)	19
Overall	186 (89.4%)	22 (10.6%)	208

$P = .83$.

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Flumazenil and Hepatic Encephalopathy

To the Editor:

We read with interest the trial of Barbaro et al.,[1] on flumazenil for hepatic encephalopathy (HE), which may have surpassed former trials[2-4] in its scope and population size but left several questions unanswered. In keeping with previous results,[3,4] clinical benefit is apparent in only about a quarter of those receiving the drug. Therefore, given the cost of flumazenil therapy, its widespread clinical use is questionable unless one can better define the subset of patients who respond. Barbaro et al. found that the number of alcoholic and posthepatic cirrhotics among flumazenil responders was similar. However, the starting study population contained more patients with posthepatic cirrhosis than with alcoholic cirrhosis. Forty-seven of 211 (22.3%) patients with alcoholic cirrhosis showed improved neurological score with flumazenil, as opposed to 35 of 312 (11.2%) patients with posthepatic cirrhosis ($\chi^2_y = 10.3$, $P < .001$). The difference remains statistically significant if patients in grades III and IVa are considered separately ($\chi^2_y = 3.98$, $P < .05$ for grade III and ($\chi^2_y = 6.23$, $P < .025$ for grade IVa). Consistent with the apparently greater efficacy of flumazenil in alcoholic cirrhosis are data indicating that these patients have increased benzodiazepine receptor binding.[5]

A second point concerns measurement of serum benzodiazepines, which was mostly negative. The method used may not detect endogenous nonbenzodiazepine substances with ben-

zodiazepine-like activity, such as the peptides known as endozepines.[6] According to recent studies, these endogenous ligands to the benzodiazepine receptor are responsible for most of the benzodiazepine activity in HE[7]; their measurement might be useful in predicting the response to flumazenil.

Finally, in considering treatment one must take into account the fact that most patients will recover spontaneously in a few days and, more importantly, that both responders and nonresponders have similar mortality rates. Thus, there does not seem to be a rationale for the use of flumazenil in the single dose protocol used by the authors. Further trials should examine multiple-dose regimens and long-term ambulatory prophylaxis of chronic HE, which to our knowledge has been described only in two isolated case reports[8,9] and has never been subjected to controlled studies. Flumazenil might help not only to prevent attacks but also to reverse chronic cognitive deficits, particularly in alcoholic cirrhotic patients in whom the drug has been shown to improve performance in neuropsychologic tests.[10] Cost remains an important issue; however, for patients being maintained while awaiting liver transplantation, this might be balanced by potential savings in hospitalization costs warranting further studies.

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Reply:

Although in alcoholic cirrhosis increased benzodiazepine receptor binding has been described, we would be cautious about inferring that alcoholic cirrhosis with hepatic encephalopathy (HE) is a special indication for flumazenil treatment. According to our experience, the grade of HE, the time of onset of neurological symptoms, and a previous history of benzodiazepine intake are the main variables to be considered. In the studies described, we measured exogenous benzodiazepines only. We agree that future work in this area should include measurement of endogenous substances with benzodiazepine-like activity, such as endozepines.

The cross-over design of our study allowed us to examine the clinical response in relation to time of treatment, and we found an increased rate of response with early administration of flumazenil: Administration within the first 3 hours from the onset of neurological symptoms may be optimal. A single flumazenil dose was used to avoid side effects related to the increased plasma half-life of the drug in cirrhotic patients. Careful monitoring of side effects is required when using large doses or continuous infusion of flumazenil.[1] Administration of the drug to cirrhotic patients with metabolic acidosis may lead to convulsions, the treatment of which may worsen the clinical outcome of HE.

We have not used flumazenil for prophylaxis of chronic HE in ambulatory patients. Our concerns relate to the cost of the drug, its short half-life, and the risk of side effects. If chronic administration is undertaken, it should be only in the setting of controlled clinical trials on selected patients.

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