Diagnosis and Management of Hemochromatosis

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PREAMBLE

Practice guidelines, intended for use by physicians, suggest preferable approaches to the diagnostic, therapeutic, and preventative aspects of care. These guidelines are intended to be flexible, in contrast with "standards of care," which are inflexible policies to be followed in almost every case.[1] They are developed in a manner consistent with the American Gastroenterological Associations' Policy Statement on Development and Use of Practice Guidelines.[2]

Specific recommendations are based on relevant published information. In an attempt to standardize recommendations, the Practice Guidelines Committee of the American Association for the Study of Liver Diseases modified the categories of the Infectious Diseases Society of America's Quality Standards.[3] These categories are reported with each recommendation, using the letters A through E to determine the strength of recommendation (Table 1) and Roman numerals I through IV to determine quality of evidence upon which recommendations are based (Table 2).

These guidelines provide data-supported peer-reviewed recommendations for the care of patients with hemochromatosis. They are based on the following: (1) a formal review and analysis of the recent published literature on hemochromatosis (Medline Search from 1990-2000); (2) the American College of Physicians' Manual for Assessing Health Practices and Designing Practice Guidelines; (3) several published guidelines, including the American Association for the Study of Liver Diseases' *Policy Statement on Development and Use of Practice Guidelines* and the American Gastroenterological Association's *Policy Statement on Guidelines*[2]; and (4) the experience of the author in the clinical care of patients with hemochromatosis.

BACKGROUND

Hereditary hemochromatosis (HH) is the most common, identified, genetic disorder in the Caucasian population. Although its geographic distribution is worldwide, it is concentrated in individuals of northern European origin, particularly of Nordic or Celtic ancestry, in whom it occurs with a prevalence close to 1 per 200 of the population.[4-6] The pathophysiologic predisposition to increased and inappropriate absorption of dietary iron may lead to the progressive development of life-threatening complications of cirrhosis, hepatocellular cancer, diabetes, and heart disease. The gene defect described in 1996[7] is a G to A missense mutation (C282Y) leading to the substitution of tyrosine for cysteine at the 282 amino acid position of the protein product of the newly discovered *HFE* gene located on the short arm of chromosome 6 (6p). Another mutation (H63D) in which aspartic acid is substituted for histidine at position 63 has also been associated as a cofactor in some cases of hemochromatosis. The homozygous state in which both alleles of chromosome 6 possess the C282Y mutation or the compound heterozygous state with C282Y on one chromosome and H63D on the other, are the predominant genetic abnormalities associated with phenotypic HH. In most studies to date, C282Y/C282Y homozygosity has been found in more than 90% of patients with hemochromatosis, while compound heterozygosity (C282Y/H63D) accounts for 3% to 5% of such cases in published series. Possession of the C282Y mutation on both alleles of the chromosome pair has a high positive predictive accuracy for phenotypic HH. In the only large population study published to date in which penetrance of the *HFE* gene mutation has been studied comprehensively, all C282Y homozygotes had elevated transferrin saturation (100% positive predictive accuracy). However, full expression as defined by progressive tissue iron overload occurred in only 58% of these homozygotes.[6]

Although the vast majority of familial cases of hemochromatosis in the Anglo-Celtic population are associated with the described pathogenic mutations of the *HFE* gene, it is highly probable that genes other than *HFE* play a role in familial iron overload in other populations. In particular, there are well documented families in Italy with iron overload comparable with *HFE*-related hemochromatosis,[8-10] in whom neither the C282Y nor H63D mutation existed, and in whom the genetic abnormality could not be located to chromosome 6p.

The clinical condition of hereditary hemochromatosis evolves in a series of stages beginning with clinically insignificant iron accumulation (0-20 years of age, 0-5 g parenchymal iron storage). This evolves to a stage of iron overload without disease (approximately 20-40 years of age, 10-20 g parenchymal iron storage), which if left untreated, may progress to a stage of iron overload with organ damage (usually more than 40 years of age and >20 g parenchymal iron storage).[11,12] Ideally, any strategy for diagnosis should identify cases before

Abbreviations: HH, hereditary hemochromatosis; HIC, hepatic iron concentration; HII, hepatic iron index; HCC, hepatocellular carcinoma; TIBC, total iron-binding capacity; UIBC, unsaturated iron-binding capacity.

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TABLE 1. Categories Reflecting the Evidence to Support the Use of a Guideline Recommendation

Category	Definition
A	Survival benefit
B	Improved diagnosis
C	Improvement in quality of life
D	Relevant pathophysiologic parameters improved
E	Impacts cost of health care

Adapted and modified from Gross et al.[3]

the third stage of disease has developed so that therapy to remove iron can prevent progression to irreversible tissue damage. Fortunately, biochemical serum testing with indirect iron markers is capable of identifying most cases of iron overload well before tissue damage has become irreversible. Therefore, these guidelines will emphasize the fundamental objective of detection of HH before organ damage has occurred (Table 3).

Current clinical practice in diagnosis and management of HH has evolved from experience in screening healthy blood donors or selected populations and managing patients and their discovered relatives with phenotypic HH.[6,12-14] Early institution of phlebotomy has proven to be a highly effective therapy for HH, which prevents morbidity and promotes normal longevity.[15] As a result, randomized, controlled trials of other therapies or observation have been regarded as unethical and have not been done.

The development of liver injury in those with HH is related to the progressive accumulation of hepatic iron.[15-18] Hepatic iron concentration increases with age in most homozygotes. In HH patients over the age of 40 years, hepatic iron concentration is likely to exceed 10,000 μg/g dry weight and liver biopsy results are more likely to show fibrosis or cirrhosis.[16-18] It was the observation that the hepatic iron concentration (HIC) increased with age that led to the concept of hepatic iron index (HIC in micromoles per gram dry weight divided by age in years). A hepatic iron index (HII) in excess of 1.9 μmol/g per year of life was found to effectively distinguish homozygous hemochromatosis from heterozygotes and patients with alcohol-induced liver disease. However, it is now clear that the rate of iron accumulation is variable and exceptions may occur in between 8% and 50% of individuals with HH.[6,12,14,19] Therefore, while an HII less than 1.9 does not entirely exclude HH, a value greater than this certainly documents significant iron overload in the C282Y homozygote and in individuals with certain forms of secondary iron overload.

TABLE 2. Quality of Evidence on Which Recommendation Is Based

Grade	Definition
I	Evidence from multiple well-designed randomized controlled trials each involving a number of participants to be of sufficient statistical power
II	Evidence from at least one large well-designed clinical trial with or without randomization, from cohort or case-control analytic studies, or well-designed meta-analysis
III	Evidence based on clinical experience, descriptive studies, or reports of expert committees
IV	Not rated

Adapted and modified from Gross et al.[3]

TABLE 3. Management Objectives for HH

Management Objectives
Early diagnosis to prevent organ damage and dysfunction due to tissue iron toxicity
Screening and early detection of asymptomatic HH cases to reduce mortality
Recognition and diagnosis of symptomatic cases of HH, to minimize progression and complications of the disease
Adequate treatment of HH to promote rapid, safe, and effective removal of iron
Vigilant follow-up and maintenance treatment of all cases of HH

The degree of iron overload has a direct impact on life expectancy of the individual with HH. The major causes of death are decompensated cirrhosis, hepatocellular carcinoma (HCC), diabetes mellitus, and cardiomyopathy.[15] These occurred with a frequency 10- to 119-fold higher than expected in an age- and sex-matched population without HH. Survival was normal in HH patients in whom treatment was initiated before the development of cirrhosis or diabetes, confirming the importance of early diagnosis and treatment.

DIAGNOSIS OF HEREDITARY HEMOCHROMATOSIS

Target Populations

Target populations are shown in Table 4. The diagnosis of hemochromatosis is based on documentation of increased iron stores, namely increased hepatic iron concentrations associated with elevated serum ferritin levels. HH can be further defined genotypically by the familial occurrence of iron overload associated with C282Y homozygosity or C282Y/H63D compound heterozygosity.[20] As serologic iron markers have become more widely available over the last several years, the majority of patients with HH are now identified while still asymptomatic and without evidence of hepatic fibrosis or cirrhosis.[15] Diagnostic screening strategies should target high-risk groups such as those with suspicious organ involvement, a familial history of HH, and those with chance detection of biochemical or radiologic abnormalities suggestive of the possibility of iron overload.

TABLE 4. Target Populations for Screening for HH

Target Populations for Hemochromatosis Evaluation
Symptomatic patients
Unexplained manifestations of liver disease or a presumably known cause of liver disease with abnormality of one or more indirect serum iron markers
Type 2 diabetes mellitus, particularly with hepatomegaly, elevated liver enzymes, atypical cardiac disease or early-onset sexual dysfunction
Early-onset atypical arthropathy, cardiac disease, and male sexual dysfunction
Asymptomatic patients
Priority groups
First-degree relatives of a confirmed case of hemochromatosis
Individuals with abnormal serum iron markers discovered during routine testing
Individuals with unexplained elevation of liver enzymes or the serendipitous finding of asymptomatic hepatomegaly or radiologic detection of enhanced computed tomography attenuation of the liver
General population
See Fig. 1.

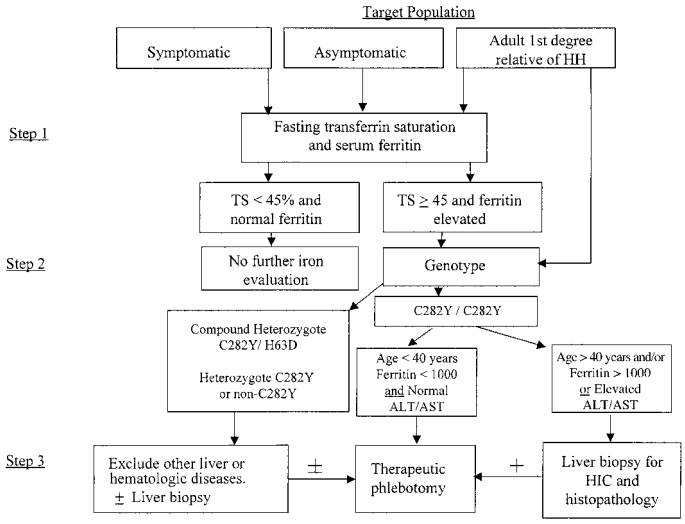

FIG. 1. Proposed algorithm for management of HH.

Evidence is accumulating to support the cost effectiveness of serologic strategies for screening the general population for iron overload.[13,21-25] Most of these reports have only assessed the usefulness of standard serologic tests such as serum iron, transferrin saturation, or serum ferritin; only one study has included the recently discovered *HFE* gene mutation.[25] This latter study compared screening of blood donors by phenotypic or genotypic methods. It was concluded that the most cost-effective strategy for identifying cases in the general population was phenotypic screening (standard iron markers) with genotypic confirmation of homozygosity in those with indirect markers of iron overload ($2,700 per case). This strategy had a high predictive value for the detection of homozygotes with iron overload and remained cost effective even when it was assumed that as few as 20% of cases would ever develop life-threatening complications of the disease.[25,26] In contrast, genotypic screening (by mutation analysis) of the general population would be prohibitively expensive ($110,000 to detect one case) and the strategy would have specificity limitations in the light of accumulating evidence for the incomplete penetrance of the gene mutations.[6] These limitations may become less important as newer and less expensive techniques of mutation analysis are developed. At this time we are supportive of a low-cost phenotypic approach for screening the general population.

Data on sensitivity, specificity, and predictive value of phenotypic screening tests have been provided by studies both in asymptomatic populations (*e.g.*, healthy blood donors and large-scale screening of a healthy population) and in families of detected homozygotes.[6,27-29] More recent studies are available for sensitivity and specificity of genotyping studies.[6,12,20]

The following diagnostic algorithm proceeds in 3 steps, beginning with phenotypic evaluation followed by genotyping of those with elevated iron markers (Fig. 1). The proposed algorithm is constructed to detect iron overload caused by HH with a high degree of accuracy, while providing a pathway for those cases of iron overload unassociated with the *HFE* mutation.

ALGORITHM STEP 1

Indirect Serologic Markers of Iron Stores

The initial approach to diagnosis of HH is by indirect serologic markers of iron stores. Transferrin saturation (TS) is derived by dividing the serum iron by the total iron binding capacity. When the fasting value exceeds 50% for women and 60% for men, TS has a sensitivity of 0.92, a specificity of 0.93, and a positive predictive value of 86% for the diagnosis of HH.[27-29] Overnight fasting avoids circadian or postprandial variations and eliminates 80% of false-positive TS results.[27] Lowering the cutoff TS value to 45% increases sensitivity, but reduces specificity and positive predictive value. In a recent study, values exceeding 45% correctly identified 97.9% of homozygotes with no false positives among the normal population.[30] However, this cutoff did include 22.2% of the heterozygote population, a group recognized to occasionally have phenotypic markers of iron overload. In another report, this cutoff was 100% sensitive for the detection of C282Y homozygotes; however, only 44% of those with TS more than 45% were genetic homozygotes.[6] Thus, lowering the threshold for TS to 45% will also identify other groups with relatively minor degrees of secondary iron overload (*e.g.*, alcohol-induced liver disease, steatohepatitis, chronic hepatitis C, previous surgical portacaval shunt, etc.) and these cases will require further evaluation by the clinician.

In most clinical laboratories TS was customarily measured by determining two serum iron measurements; the first a measurement of the subjects' fasting serum iron, the second a repeat measurement after adding exogenous iron to saturate the serum transferrin followed by removal of the nontransferrin-bound iron. The latter determines the total iron binding capacity (TIBC). The ratio serum iron to TIBC gives the transferrin saturation (TS in percent). Although the serum iron is an automated test, the TIBC is not, making the traditional method for deriving TS relatively expensive. Alternatively, it has been proposed that costs could be reduced by using unsaturated iron binding capacity (UIBC).[31] Values for UIBC less than 28 μmol/L are indicative of iron overload. In fact, many laboratories now determine TIBC by summing serum iron and UIBC (both automated methods). TS is then expressed as the ratio of serum iron to the calculated TIBC: Fe/(Fe + UIBC). This allows the clinician to judge the significance of a raised TS, by noting those that might be spuriously elevated by a low TIBC (low serum transferrin concentration). It is recommended that TS be calculated in this way to reduce costs, particularly for large-scale screening.

Other indirect markers of iron stores such as serum iron or ferritin lack specificity when used alone. The serum iron has positive and negative predictive values for HH of 61% and 87%, respectively, compared with 74% and 93% for TS.[29] Serum ferritin is also nonspecific particularly in the face of inflammatory conditions, chronic hepatitis C, alcohol-induced liver disease, and neoplastic diseases. However, a serum ferritin level in combination with TS has a negative predictive value of 97% and exceeds the accuracy of any of the indirect tests used in isolation.[29] In confirmed HH, a level of serum ferritin >1,000 ng/mL is an accurate predictor of the degree of hepatic fibrosis (cirrhosis).[32]

Recommendation 1. Initial screening of individuals with suspected iron overload and those over the age of 20 years who are first-degree relatives of known cases of HH should be done by measurement of transferrin saturation after an overnight fast. Simultaneous serum ferritin determination increases the predictive accuracy for diagnosis of iron overload. TS is also the test of choice for screening the general adult population for iron overload states (Fig. 1) (rating: II A, B, C, D, and E).

ALGORITHM STEP 2

Genotypic Testing: Mutation Analysis

Fasting transferrin saturation less than 45% and a normal serum ferritin would require no further evaluation. Elevation of TS and serum ferritin would require genotypic testing as indicated in step 2 of the diagnostic algorithm in Fig. 1. The presence of the *HFE* mutations C282Y and H63D can now be detected by polymerase chain reaction using whole blood samples.[7] Individuals with serum indicators of iron overload who are homozygous for the C282Y mutation require phlebotomy therapy. Those who are unlikely to have significant hepatic injury may be offered therapeutic phlebotomy without the necessity for a liver biopsy. This includes individuals less than 40 years of age who have no clinical evidence of liver disease (raised alanine transaminase, hepatomegaly, etc.) and whose serum ferritin is less than 1,000 ng/mL. Higher values of serum ferritin are associated with an increased likelihood of significant hepatic fibrosis or cirrhosis.[20,32] On the other hand, liver biopsy should be offered to document the degree of fibrosis in all homozygotes who are over the age of 40 years or those who have an elevated serum alanine transaminase level, have clinical evidence of liver disease, or have a serum ferritin greater than 1,000 ng/mL. Since these are likely to be individuals over the age of 40 years, discretion is appropriate in recommending liver biopsy on the basis of age alone. In the absence of the above indicators of cirrhosis, other risk factors (*e.g.*, alcohol abuse, or coexisting clinical features of HH, such as diabetes, impotence, etc.) may play a role in making this recommendation. Liver biopsy and hepatic iron evaluation are also recommended in compound heterozygotes (C282Y/H63D), C282Y heterozygotes, or non-*HFE* mutated individuals who have indirect markers of iron overload, particularly if they also have abnormal liver enzymes or clinical evidence of liver disease. Although these individuals account for a small proportion of phenotypic hemochromatosis, they have a low likelihood of significant iron overload, and elevated iron tests are often due to other causes of liver disease.[33]

Although recognizing that the penetrance of the C282Y mutation is variable, the option is provided in step 1 of the diagnostic algorithm to proceed to gene mutation analysis regardless of the TS or serum ferritin in first-degree relatives of a known HH individual. In the case of the children of an HH patient, mutation analysis in the spouse allows for assessment of the genotypic status of the children.[14] If the spouse possesses no C282Y mutation, the offspring can only be heterozygous. An HH patient with a spouse who is a heterozygote for C282Y has a 50% chance of having homozygous offspring. Because organ damage is virtually unknown in HH before adult life, evaluation of first-degree relatives can be postponed until about 20 years of age.

Recommendation 2. Genotyping to detect HFE mutations should be performed for all individuals who have abnormal iron studies and on those who are first-degree relatives of identified homozygotes as detailed in step 1 of the diagnostic algorithm. In the absence of indicators suggestive of significant liver disease, C282Y homozygotes under the age of 40 years may be treated by therapeutic phlebotomy without the need for liver biopsy. Liver biopsy is recommended in all homozygotes with clinical evidence of liver disease, serum ferritin greater than 1,000 ng/mL, and particularly in those greater than 40 years of age with other risk factors for liver disease. Liver biopsy should also be considered in compound or C282Y heterozygotes with elevated TS, particularly those who have had abnormal liver enzyme levels or clinical evidence of liver disease (rating: II A, B, C, D, and E).

ALGORITHM STEP 3

Liver Biopsy for HIC

Liver biopsy is useful to document the presence of cirrhosis (if not evident from radiologic studies) to rule out significant iron overload when iron markers are equivocal, or to investigate other possible causes of liver disease. Histopathology and staging of fibrosis is best determined with hematoxylin-eosin and Masson trichrome staining, respectively. The liver is the most easily accessible tissue for accurately assessing iron stores. The degree and cellular distribution of iron stores is best assessed using a Perls' Prussian blue stain. Before 1985, the extent of iron deposition was judged exclusively by this method and, in fact, a qualitative assessment of iron stores was derived based on stainable iron.[34] Two qualitative scales have been proposed.[35,36] The most commonly used of these, the Ludwig-Batts system, estimates the proportion of hepatocytes that stain for iron, recognizing the progressive nature of iron accretion through the hepatic acinus from Rappaport zone 1 (periportal) to zone 3 (pericentral).[36] Although grade 4 iron deposition (panacinar) usually indicates HH range quantitative iron levels, grades 2 and 3 correlate poorly with quantitative iron content. For this reason the quantitative, biochemical HIC has become the preferred method for evaluating the hepatic iron stores. Quantitative iron determinations from fresh frozen and formalin-fixed, paraffin-embedded samples are comparable.[36,37] Accordingly, a biopsy core at least 2.5 to 3.0 cm in total length should be obtained. A 0.5- to 1.0-cm piece of the tissue core should be removed and placed in a dry tube or in 10% formalin (not in saline, which may leach out iron). The remainder of the fixed tissue is processed for routine histopathologic evaluation and a Perls' Prussian blue stain. If tissue was not separated and saved before fixation and embedding, the remaining tissue can be removed from the paraffin block and sent for quantitative iron.

The normal HIC is less than 1,800 μg/g dry weight (equivalent to 32 μmol/g). It is now clear from several studies that most patients with homozygous HH steadily and inexorably accumulate iron at least through early adult and middle life, unless they have had blood loss or have been blood donors, in contrast to patients with secondary iron overload caused by other chronic liver diseases. The concept of the HII as a measure of the iron accretion rate was developed to distinguish HH from these other potentially confounding clinical situations, particularly alcohol-induced liver disease.[16-18] A rate in excess of 1.9 μmol/g/y is strong evidence for homozygous hemochromatosis. However, it has recently been shown that up to 15% of genotypic homozygotes for HH do not meet the previously defined rate of at least 1.9 μmol/g/y. Thus, an elevated HII is no longer considered essential for diagnosis.[20] Yet, even these individuals with partial expression of homozygous HH have HIC at least 3 times the upper limit of normal if they are more than 20 years old. Finally, it should be emphasized that secondary iron overload due to dyserythropoietic or hemolytic anemia may have HIC comparable with that seen in HH, particularly in those who require repeated blood transfusions. These causes should be easily distinguishable by other clinical criteria.

Although the rate of hepatic iron accumulation (HII) has lost some of its importance in the diagnosis of HH, it is nevertheless the correlation between HIC and age that determines the age at which fibrosis will develop. Sallie et al.[18] found no patient who developed hepatic fibrosis before the HIC levels exceeded 14,000 μg/g dry weight. Hepatic fibrosis was not present in any patient less than 40 years of age in the series reported by Bacon et al.[20] and Guyader et al.[32] and occurred only at a younger age or lower levels of HIC in individuals who also abuse alcohol.[11] The latter is the basis for recommendation 2 (Fig. 1) regarding the lack of need for liver biopsy in some patients. Indeed, Bacon et al. retrospectively applied this algorithm to 66 patients who were C282Y homozygotes and found that 19 of the 66 would not have required the liver biopsy, which otherwise would have been necessary to determine HIC.[20]

The value of liver biopsy is not limited to determination of HIC. Documentation of extensive bridging fibrosis or cirrhosis by liver biopsy has a profound impact on the prognosis in HH patients. Serum aminotransferase levels may be helpful in identifying chronic liver disease but lack negative predictive accuracy since half of cirrhotic HH patients have normal alanine transaminase or aspartate transaminase values.[20] Survival in noncirrhotic HH patients is similar to the normal control population, while those with cirrhosis have significantly increased mortality. Cirrhosis or its complications, particularly hepatocellular cancer, account for three quarters of HH-related deaths.[15] Thus, close surveillance for HCC has been proposed for cirrhotic individuals, although there are currently no data to guide the optimal method or interval for such screening in HH. Further studies are needed.

Recommendation 3. Liver biopsy is helpful in suspected HH when documentation of HIC and the stage of fibrosis is necessary (see recommendation 2) or to rule out other causes of liver disease. In addition to routine histologic assessment, qualitative hepatic iron determination should be performed by Perls' staining. If this suggests increased iron stores, this should be confirmed by a quantitative iron measurement in stored tissue (rating: II A, B, C, D, and E).

TREATMENT OF HEMOCHROMATOSIS

Hereditary Hemochromatosis

There is overwhelming evidence that institution of phlebotomy therapy before cirrhosis and/or diabetes develop will significantly reduce the morbidity and mortality of HH.[15] Therefore, early identification (step 1 in algorithm, Fig. 1) and preemptive treatment of those at risk is required. This includes treatment of asymptomatic individuals with homozygous HH and markers of iron overload, as well as others with evidence of potentially toxic levels of hepatic iron. In symptomatic patients treatment is also advocated to mitigate as much of the organ damage as possible. Certain clinical features may be ameliorated by phlebotomy (malaise, fatigue, skin pigmentation, insulin requirements in diabetes, abdominal pain), whereas other features are either less responsive to iron removal or do not respond at all (arthropathy, hypogonadism, cirrhosis). The life-threatening complications of cirrhosis, particularly HCC, continue to be a threat to survival even after adequate phlebotomy. HCC accounts for about 30% of all deaths in HH, whereas other complications of cirrhosis account for an additional 20%.[11,15] The observation that HCC is exceedingly rare in noncirrhotic HH provides an additional and powerful argument for preventive therapy prior to the development of cirrhosis.[38]

TABLE 5. Treatment of Iron Overload

Treatment of Hemochromatosis
Hereditary hemochromatosis
One phlebotomy (removal of 500 mL of blood) weekly or biweekly
Check hematocrit prior to each phlebotomy; allow hematocrit to fall by no more than 20% of prior level
Check serum ferritin level every 10-12 phlebotomies
Stop frequent phlebotomy when serum ferritin falls below 50 ng/mL
Continue phlebotomy at intervals to keep serum ferritin to between 25 and 50 ng/mL
Avoid vitamin C supplements
Secondary iron overload due to dyserythropoiesis
Deferoxamine (Desferal) at a dose of 20-40 mg/kg body weight per day
Consider follow-up liver biopsy to ascertain adequacy of iron removal
Avoid vitamin C supplements

The mainstay of treatment for HH remains phlebotomy (Table 5). One unit of blood (equal to about 250 mg of iron, depending on the hematocrit) should be removed once or twice per week as tolerated. In HH patients who may have total body iron stores greater than 30 g, this phlebotomy regimen may take up to 2 to 3 years to adequately reduce iron stores to the desired end point just short of iron deficiency. Each venesection should be preceded by measurement of the hematocrit. The hematocrit should have returned to within 10 points of or no lower than 20% below its starting value. Transferrin saturation usually remains elevated until iron stores are depleted. Serum ferritin may initially fluctuate, but eventually begin to fall progressively with iron mobilization. Serum ferritin should only be done after every 10 to 12 phlebotomies in the initial stages of treatment. It can be confidently assumed that excess iron stores have been mobilized when the serum ferritin falls below 50 ng/mL. As the target figure of 50 ng/mL is approached, it may be repeated more frequently to preempt the development of overt iron deficiency. Levels less than 25 ng/mL indicate iron deficiency and require a temporary hold on further phlebotomies. Iron deficiency anemia should be avoided. At the point at which the above-mentioned criteria indicate incipient iron deficiency, frequent phlebotomy can be stopped and a maintenance schedule started. The frequency of maintenance phlebotomies varies among individuals, as might be expected given the variable rate of iron accumulation in HH. Certain persons (either male or female) require phlebotomy every month, whereas others who presumably reaccumulate iron at a slower rate may need only 3 to 4 units of blood removed per year. Currently, in the United States, blood acquired by therapeutic phlebotomy cannot be used for blood donation, a ruling that is under scrutiny. Therefore, phlebotomy remains a therapeutic procedure with a coding recognized by the Health Care Finance Administration and third-party insurers.

Cardiac dysrhythmias and cardiomyopathy are the most common causes of sudden death in iron overload states. Since the risk of these complications may increase during rapid mobilization of iron, certain additional precautions and therapy may be required. Pharmacologic doses of vitamin C accelerate mobilization of iron to a level that may saturate circulating transferrin, which results potentially in an increase in pro-oxidant and free-radical activity. Therefore, supplemental vitamin C should be avoided by patients undergoing phlebot-

omy. Patients receiving iron chelators should not exceed 200 mg of vitamin C intake daily.[39]

Cirrhosis does not reverse with iron removal and the development of decompensated liver disease is an indication to consider orthotopic liver transplantation. However, survival of transplanted HH patients is lower than in those transplanted for other causes of liver disease.[40,41] Most posttransplantation deaths in HH patients occur in the perioperative period from cardiac or infection-related complications. Whether these problems are related to inadequate removal of excess iron stores before orthotopic liver transplantation is not known. However, this is certainly the case in some patients in whom HH is not or cannot be diagnosed before orthotopic liver transplantation. Persistent tissue iron toxicity may lead to posttransplantation end-organ problems in these cases despite removal of the bulk of parenchymal iron stores with the explanted liver.

Early studies reported that HCC accounted for 30% of all deaths in HH patients and that the risk did not decrease after adequate iron removal by phlebotomy.[41] However, these data were obtained before institution of screening programs that promote earlier identification and treatment of cases. Nonetheless, close life-long observation of HH patients with significant fibrosis or cirrhosis would seem prudent. However, there are currently no data on the optimal method or frequency for such surveillance for the development of HCC, nor are there data supporting the cost effectiveness of such a practice in HH. Until there is information specific to HH on this life-threatening complication, the recommendation for surveillance of HCC is based on analogy to other causes of cirrhosis, particularly chronic viral hepatitis.

Recommendation 4. All patients with HH who have evidence of iron overload should be strongly encouraged to undergo regular phlebotomies until iron stores are depleted. These should be continued for life, gauging the frequency of maintenance therapy on the serum ferritin level. Vitamin C supplements should be avoided. HH patients with cirrhosis should undergo regular screening for HCC (rating: II A, B, C, D, and E).

Secondary Iron Overload

Although these guidelines have concentrated on the management of HH it is pertinent to review the treatment of nonhereditary forms of secondary iron overload. The causes of secondary iron overload are listed in Table 6.

Phlebotomy is useful only in certain forms of secondary iron overload (Table 5). It has been used in African iron overload and porphyria cutanea tarda with reduction in morbidity and mortality. No systematic, randomized controlled studies have been done in secondary iron overload states associated with chronic liver diseases to evaluate long-term outcomes of iron removal therapy. In secondary iron overload associated with ineffective erythropoiesis, iron chelation therapy with parenteral deferoxamine is the treatment of choice. Multiple studies have documented the efficacy of deferoxamine in preventing the complications of iron overload in β-thalassemia.[42] Deferoxamine mesylate (Desferal; Novartis Pharmaceuticals Corporation, East Hanover, NJ) is the only approved iron chelation agent that is widely available. Usually it is administered subcutaneously, using an implanted minipump, by continuous infusion over a 24-hour cycle at a dose of 20 to 40 mg/kg/d. A total dose of about 2 g per 24 hours usually achieves maximum urinary iron excretion. Chelation therapy to reduce hepatic iron concentrations below 15,000 μg/g dry weight significantly reduces the risk of clinical disease.[43] However, the aim of chelation therapy should be more ambitious and should attempt to achieve and maintain near normal hepatic iron concentrations. The application of deferoxamine therapy is limited by cost (particularly in developing countries), the need for a parenteral route of therapy, discomfort and inconvenience (a challenging prospect in young patients), and neurotoxicity. In addition, a variety of opportunistic bacterial infections have been described after prolonged chelation therapy.[44]

TABLE 6. Iron Overload States

Classification
Hereditary hemochromatosis
HH: *HFE* related
C282Y homozygosity
C282Y/H63D compound heterozygosity
Other mutations of *HFE*
HH: non-*HFE* related; other gene mutations
Juvenile hemochromatosis
Autosomal dominant hemochromatosis (Solomon Islands)
Secondary iron overload
Iron-loading anemias ± transfusion
Thalassemia major
Sideroblastic anemia
Chronic hemolytic anemias
Dietary iron overload
Chronic liver diseases
Hepatitis C and B
Alcohol-induced liver disease
Porphyria cutanea tarda
Fatty liver disease
Miscellaneous causes of iron overload
African iron overload
Neonatal iron overload
Aceruloplasminemia
Congenital atransferrinemia

Monitoring iron reduction in patients with secondary iron overload is a challenge. In contrast to HH, where serum ferritin reliably reflects iron burden during therapy, ferritin levels are misleading in secondary overload cases. In many patients, it may be necessary to repeat the liver biopsy to assess the progress of therapy and ensure adequate chelation.[45] Superquantum magnetic susceptibility determinations are capable of measuring hepatic iron concentrations over a wide range, but it is expensive and available only at two centers worldwide. The recent advent of high Tesla magnetic resonance imaging instruments showed some initial promise as a noninvasive way to estimate hepatic iron levels, but it has proven to be inaccurate when there is marked iron overload or hepatic fibrosis/cirrhosis.[46] In selected patients with thalassemia major, allogeneic bone marrow transplantation offers the possibility of permanent cure of the hematologic disorder.[47] However, the preexisting iron overload persists after the transplantation and such patients may be recommended for phlebotomy with the expectation that the bone marrow is capable of enhanced erythropoiesis. As in patients treated by chelation, the liver iron concentration provides an accurate, quantitative means for monitoring iron balance.[48]

Recommendation 5. Treatment of secondary iron overload should be tailored to the underlying cause. Phlebotomy using a regimen similar to HH (see recommendation 4) may be

tolerated in some forms of secondary iron overload without preexisting anemia. Parenteral chelation therapy with deferoxamine is currently the treatment of choice in patients with chronic dyserythropoietic syndromes or chronic hemolytic anemia. Monitoring of the efficacy of therapy during chelation may require repeat liver biopsies to confirm adequate reduction of HIC (rating: II A, B, C, D, and E).

SUMMARY

HH is one of the few genetic disorders in which phenotypic manifestations (organ damage) are delayed to adult life. However, sensitive and specific phenotypic and genotypic testing now allows diagnosis of HH while it is still a disorder of iron metabolism and before it results in end-organ damage.[49] The practical clinical guidelines provided here offer a cost-effective, preemptive diagnostic approach whereby early identification of individuals at risk and initiation of life-saving phlebotomy therapy in the presymptomatic stage of the disorder are facilitated.

ADDENDUM

Since the development of these practice guidelines was initiated by the AASLD, an International Consensus Conference on Haemochromatosis was conducted by the European Association for the Study of Liver (EASL), cosponsored by the WHO, NIH, and CDC, and was recently published as a conference report.[50] Although many of the conclusions and recommendations of this conference accord with the AASLD Practice Guidelines on hemochromatosis, others remain tentative for the international group, particularly those relating to screening for iron overload. However, we share with the international group an awareness of the need for information derived from well-designed screening programs that incorporate careful follow-up of identified HH patients and matched controls.

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REFERENCES

1. Eddy DM. A Manual for Assessing Health Practices and Designing Practice Guidelines. Philadelphia: American College of Physicians 1996:1-126.
2. Position and policy statement. American Gastroenterological Association policy statement on the use of medical practice guidelines by managed care organizations and insurance carriers. Gastroenterology 1995;108: 925-926.
3. Gross PA, Barrett TL, Dellinger EP, Krause PJ, Martone WJ, McGowan JE, Sweet RL, et al. Infectious Diseases Society of America quality standards for infectious diseases: purpose of quality standards for infectious diseases. Clin Infect Dis 1994;18:421.
4. Merryweather-Clarke AT, Pointon JJ, Shearman JD, Robson KJ. Global prevalence of putative haemochromatosis mutations. J Med Genet 1997; 34:275-278.
5. Lucotte G. Celtic origin of the C282Y mutation of hemochromatosis. Blood Cells Mol Dis 1998;24:433-438.
6. Olynyk JK, Cullen DJ, Aquila S, Rossi E, Summerville L, Powell LW. A population based study of the clinical expression of the hemochromatosis gene. N Engl J Med 1999;341:718-724.
7. Feder JN, Gnirke A, Thomas W, Tsuchihashi Z, Ruddy DA, Basava A, Dormishian F, et al. A novel MHC class 1-like gene is mutated in patients with hereditary haemochromatosis. Nat Genet 1996;13:399-408.
8. Carella M, D'Ambrosio L, Totaro A, Grifa A, Valentino MA, Piperno A, Girelli D, et al. Mutation analysis of the *HLA-H* gene in Italian hemochromatosis patients. Am J Hum Genet 1997;60:828-832.
9. Cameschella C, Fargion S, Sampietro M, Roetto A, Bosio S, Garozzo G, Arosio C, et al. Inherited *HFE*-unrelated hemochromatosis in Italian families. HEPATOLOGY 1999;29:1563-1564.
10. Pietrangelo A, Montosi G, Totaro A, Garuti C, Conte D, Cassanelli S, Fraquelli M, et al. Hereditary hemochromatosis in adults without pathogenic mutations in the hemochromatosis gene. N Engl J Med 1999;341: 725-732.
11. Adams PC, Deugnier Y, Moirand R, Brissot P. The relationship between iron overload, clinical symptoms, and age in 410 patients with genetic hemochromatosis. HEPATOLOGY 1997;25:162-166.
12. Crawford DHG, Jazwinska EC, Cullen LM, Powell LW. Expression of HLA-linked hemochromatosis in subjects homozygous for the C282Y mutation. Gastroenterology 1998;114:1003-1008.
13. Adams PC, Gregor JC, Kertesz AE, Valberg LS. Screening blood donors for hereditary hemochromatosis. Decision analysis model based on a 30 year database. Gastroenterology 1995;109:177-188.
14. Adams PC, Chakrabarti S. Genotypic/phenotypic correlations in genetic hemochromatosis: evolution of diagnostic criteria. Gastroenterology 1998;114:319-323.
15. Niederau C, Fischer R, Purschel A, Stremmel W, Haussinger D, Strohmeyer G. Long-term survival in patients with hereditary hemochromatosis. Gastroenterology 1996;110:1107-1119.
16. Bassett ML, Halliday JW, Powell LW. Value of hepatic iron measurement in early hemochromatosis and determination of the critical iron level associated with fibrosis. HEPATOLOGY 1986;6:24-29.
17. Summers KM, Halliday JW, Powell LW. Identification of homozygous hemochromatosis subjects by measurement of hepatic iron index. HEPATOLOGY 1990;12:20-25.
18. Sallie RW, Reed WD, Shilkin KB. Confirmation of the efficacy of hepatic tissue iron index in differentiating genetic hemochromatosis from alcoholic liver disease complicated by alcoholic haemosiderosis. Gut 1991; 32:207-210.
19. Kowdley KV, Trainer TD, Saltzman JR, Pedrosa M, Krawitt EL, Knox TA, Susskind K, et al. Utility of hepatic iron index in American patients with hereditary hemochromatosis: a multicenter study. Gastroenterology 1997;113:1270-1277.
20. Bacon BR, Olynyk JK, Brunt EM, Britton RS, Wolff RK. HFE genotype in patients with hemochromatosis and other liver diseases. Ann Int Med 1999;130:953-962.
21. Balan V, Baldus W, Fairbanks V, Michels V, Burritt M, Klee G. Screening for hemochromatosis: a cost-effectiveness study based on 12,258 patients. Gastroenterology 1994;107:453-459.
22. Phatak PD, Guzman G, Woll JE, Robeson A, Phelps CE. Cost-effectiveness of screening for hereditary hemochromatosis. Arch Int Med 1994; 154:769-776.
23. Baer DM, Simons JL, Staples RL, Rumore GJ, Morton CJ. Hemochromatosis screening in asymptomatic ambulatory men 30 years of age and older. Am J Med 1995;98:464-468.
24. Bassett ML, Leggett BA, Halliday JW, Webb S, Powell LW. Analysis of the cost of population screening for hemochromatosis using biochemical and genetic markers. J Hepatol 1997;27:517-524.
25. Adams PC, Valberg LS. Screening blood donors for hereditary hemochromatosis: Decision analysis model comparing genotyping to phenotyping. Am J Gastroenterol 1999;94:1593-1600.
26. Tavill AS. Screening for hemochromatosis: phenotyping or genotyping or both? Am J Gastroenterol 1999;94:1430-1433.
27. Edwards CQ, Griffen LM, Goldgar D, Drummond C, Skolnick MH, Kushner JP. Prevalence of hemochromatosis among 11,065 presumably healthy blood donors. N Engl J Med 1988;318:1355-1362.
28. Borwein S, Ghent CN, Valberg LS. Diagnostic efficacy of screening tests for hereditary hemochromatosis. Cen Med Assoc 1984;131:895-901.
29. Bassett ML, Halliday JW, Ferris RA, Powell LW. Diagnosis of hemochromatosis in young subjects: Predictive accuracy of biochemical screening tests. Gastroenterology 1984;87:628-633.
30. McLaren CE, McLachlan GJ, Halliday JW, Webb SI, Leggett BA, Jazwinska EC, Crawford DH, et al. Distribution of transferrin saturation in an

Australian population: relevance to the early diagnosis of hemochromatosis. Gastroenterology 1998;114:543-549.
31. Adams PC, Kertesz AE, McLaren CE, Barr R, Bamford A, Chakrabarti S. Population screening for hemochromatosis: a comparison of unbound iron binding capacity transferrin saturation, and C282Y genotyping in 5,211 voluntary blood donors. HEPATOLOGY 2000;31:1160-1164.
32. Guyader D, Jacquelinet C, Moirand R, Turlin B, Mendler MH, Chaperon J, David V, et al. Non-invasive prediction of fibrosis in C282Y homozygous hemochromatosis. Gastroenterology 1998;115:929-936.
33. Bacon BR, Powell LW, Adams PC, Kresina T, Hoofnagle JH. Molecular medicine and hemochromatosis: at the crossroads. Gastroenterology 1999;116:193-207.
34. Deugnier YM, Turlin B, Powell LW, Summers KM, Moirand R, Fletcher L, Loreal O, et al. Differentiation between heterozygotes and homozygotes in genetic hemochromatosis by means of a histological hepatic iron index: a study of 192 cases. HEPATOLOGY 1993;17:30-34.
35. Scheuer PJ, Williams R, Muir AR. Hepatic pathology in relatives of patients with hemochromatosis. J Pathol 1962;84:53-64.
36. Ludwig J, Batts K, Moyer T, Baldus W, Fairbanks V. Liver biopsy diagnosis of homozygous hemochromatosis: a diagnostic algorithm. Mayo Clin Proc 1993;68:263-267.
37. Olynyk JK, O'Neill R, Britton RS, Bacon BR. Determination of hepatic iron concentration in fresh and paraffin-embedded tissue: diagnostic implications. Gastroenterology 1994;106:674-677.
38. Deugnier YM, Gruyader D, Crantock L, Lopez J-M, Turlin B, Yaouang J, Jouanolle H, et al. Primary liver cancer in genetic hemochromatosis: a clinical pathological and pathogenic study of 54 cases. Gastroenterology 1993;104:228-234.
39. Cao A, Gabutti V, Galanello R, et al. Management Protocol for the Treatment of Thalassaemia Patients. Nicosia, Cyprus: Thalassaemia International Federation, 1997.
40. Farrell FJ, Nguyen KM, Woodley S, Imperial JC, Garcia-Kennedy R, Man K, Esquivel CO, et al. Outcome of liver transplantation in patients with hemochromatosis. HEPATOLOGY 1994;20:404-410.
41. Kowdley KV, Hassanein T, Kaur S, Farrell FJ, Van Thiel DH, Keefe EB, Sorrell MF, et al. Primary liver cancer and survival in hereditary hemochromatosis patients undergoing orthotopic liver transplantation. Liver Transplant Surg 1995;1:237-241.
42. Oliveri NF. The β-thalassemias. N Engl J Med 1999;341:99-109.
43. Brittenham GM, Griffith PM, Nienhuis AW, McLaren CE, Young NS, Tucker EE, Allen CJ, et al. Efficacy of deferoxamine in preventing complications of iron overload in patients with thalassemia major. N Engl J Med 1994;331:567-573.
44. Porter JB. A risk-benefit assessment of iron chelation therapy. Drug Saf 1997;17:407-421.
45. Brittenham GM, Cohen AR, McLaren CE, Martin MB, Griffith PM, Nienhuis AW, Young NS, et al. Hepatic iron stores and plasma ferritin concentration in patients with sickle cell anemia and thalassemia major. Am J Hematol 1993;42:81-85.
46. Angelucci E, Giovagnoni A, Valeri, Paci E, Ripalti M, Muretto P, McLaren C, et al. Limitations of magnetic resonance imaging in measurement of hepatic iron. Blood 1997;90:4736-4742.
47. Angelucci E, Muretto P, Lucarelli G, Ripalti M, Barconciani D, Erer B, Galimberti M, et al. Phlebotomy to reduce iron overload in patients cured of thalassemia by some marrow transplantation. Blood 1997;90:994-998.
48. Angelucci E, Brittenham GM, McLaren CE, Ripalti M, Baronciani D, Giardini C, Galimberti M, et al. Hepatic iron concentration and total body iron stores in thalassemia major. N Engl J Med 2000;343:327-331.
49. Tavill AS. Clinical implications of the hemochromatosis gene. N Engl J Med 1999;341:755-757.
50. EASL International Consensus Conference on Haemochromatosis. J Hepatol 2000;33:485-504.