Preventing organ damage by genetic testing for hereditary haemochromatosis

Kotze MJ, ***De Villiers JNP***
Genecare Molecular Genetics (Pty) Ltd., Christiaan Barnard Memorial Hospital, Cape Town
Van der Merwe SW
Department of Internal Medicine and Gastroenterology, University of Pretoria,
Unitas hospital, Netcare, Centurion, Pretoria.

Correspondence to: Dr Maritha Kotze, PO Box 15743, Vlaeberg, 8018.
Tel: 021 422 5538 Fax: 021 422 5539 E-mail: mjk@genecare.co.za

Abstract

The rapid discovery of several iron-related genes in the last 10 years has led to the development of cost-effective genetic assays for early diagnosis of hereditary haemochromatosis (HH). A genetic predisposition for this relatively common autosomal recessive disease has been identified in approximately ***1 in 100 South Africans of European descent***. If left untreated, this condition may lead to organ damage presenting as cirrhosis, liver cancer, diabetes, arthritis, impotence, sterility and/or cardiac disease. Due to the fact that serum iron parameters are frequently affected by factors such as liver disease and inflammation, direct mutation detection has become the method of choice for accurate diagnosis of inherited iron overload in patients with elevated iron stores. Haemochromatosis can be prevented by regular blood donation or phlebotomy and therefore detection of a genetic predisposition at an early age, before irreversible damage to cardiac, hepatic and endocrine tissue occurs, represents an important clinical goal. ***(SA Fam Pract 2005;47(2): 44-45)***

Introduction

Hereditary haemochromatosis (HH) is a common genetic condition but remains largely unrecognised or misdiagnosed. This can be ascribed largely to the wide range of conditions and the non-specific symptoms associated with HH complicating early clinical diagnosis. Early features of iron overload such as fatigue, joint pain, abdominal pain and loss of libido are non-specific and not commonly recognised to be associated with HH by primary care physicians (Niederau et al. 1996). Most cases of early iron overload have normal liver function tests, whereas mildly abnormal liver function tests are commonly ascribed to excessive alcohol use.

Conditions and symptoms associated with iron overload include:

- Chronic parenchymal liver disease, cirrhosis, hepatocellular carcinoma
- Cardiomyopathy and arrhythmias
- Diabetes mellitus type I and II
- Infertility, amenorrhoea, impotence, loss of libido, testicular atrophy
- Anterior pituitary failure
- Arthritis, arthralgia, joint pain
- Porphyria cutanea tarda
- Weakness, chronic fatigue
- Mood swings, depression
- Unexplained abdominal pain, frequent diarrhoea
- Skin pigmentation, bronzing of the skin
- Loss of body hair

How does iron accumulation lead to disease?

Iron is absorbed from the gut enterocyte, and transported bound to the carrier protein transferrin to most organs of the body. Because of iron toxicity, stored iron is mainly compartmentalised as ferritin in the bone marrow where it is available for haem synthesis.

If iron absorption is dysregulated iron will accumulate. Iron deposited in organs will lead to organ dysfunction. In haemochromatosis organs most commonly affected include the liver, skin, pancreas, joints, heart and pituitary gland.

Who is at risk?

Genetic studies performed in South Africa by de Villiers et al (1999a,b) have identified the genetic defects in more than 80% of HH patients and have shown that 1 out of 6 Caucasians are carriers of a common iron-related mutation (C282Y) in the HFE gene (Feder et al. 1996). This means that an estimated 1/115 Caucasian individuals in South Africa are homozygous for the C282Y mutation. As mutation carriers do not necessarily develop clinical symptoms the defective gene can be passed on in a family unnoticed. HH is the most common autosomal resessive disorder that affects humans. This means that both genes must be inherited (one from each parent) to develop clinical disease. Offspring of two mutation carriers will have a 25% (1 in 4) chance of inheriting two copies of the defective gene. Since organ damage occur in approximately 40-60% of individuals with a genetic predisposition for haemochromatosis, it is important that testing is offered to all relatives of an HH sufferer (Milani and Kotze 1999). The risk is increased if a family history of arthritis, diabetes, liver disease or heart failure is present.

Diagnosis of haemochromatosis

Determination of transferrin saturation is recommended as a first line screening method for haemochromatosis and can detect cases of iron overload before organ dysfunction has occurred. However, the use of transferrin saturation requires fasting, is relatively non-specific and will also be elevated in chronic liver diseases due to secondary iron overload. DNA testing, on the other hand, provides a definitive diagnosis in the majority of affected cases with elevated transferrin saturation and ferritin levels, without the need to perform a liver biopsy. The ability to perform rapid mutation analysis on samples that are not C282Y homozygotes is becoming increasingly important in the South African population (Zaahl et al. 2004) as more novel mutations are found in an increasing number of genes (Table I).

Genetic testing

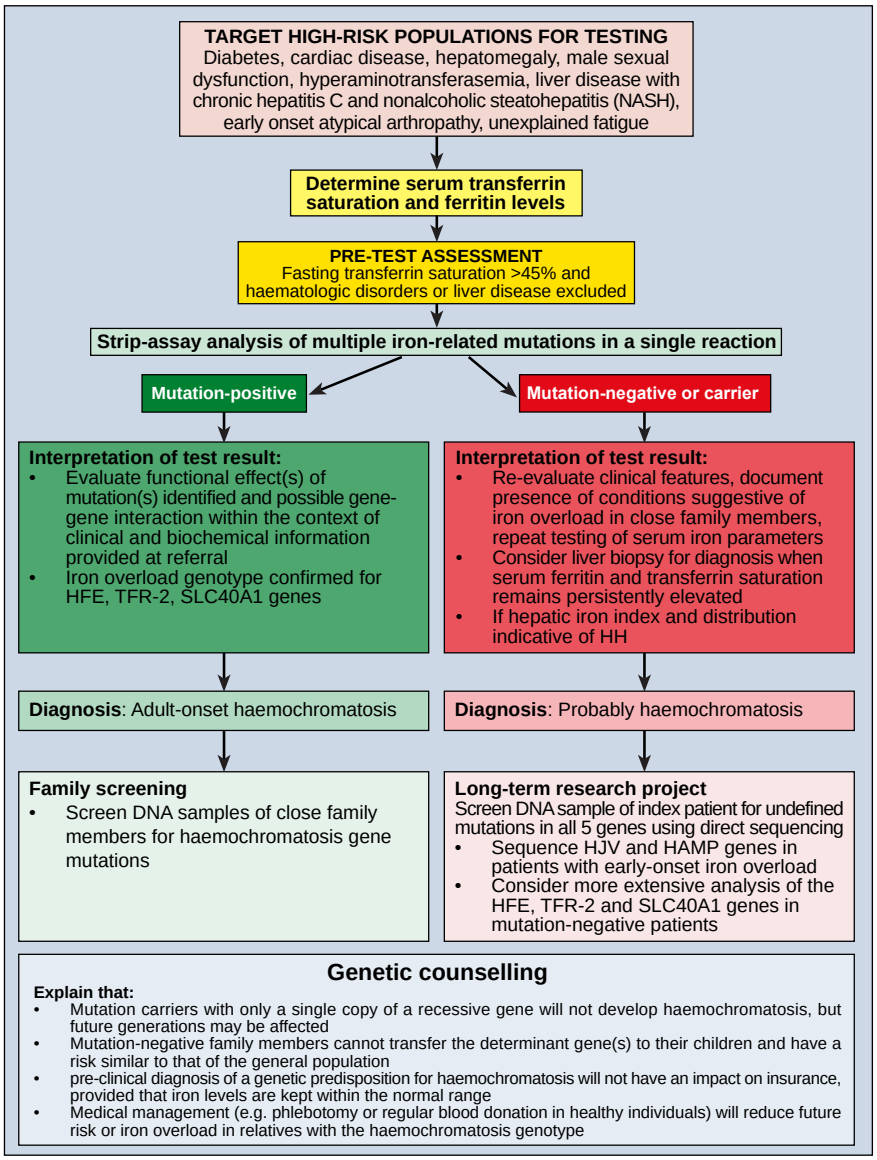
Genetic testing is important since it can provide a definitive diagnosis of inherited iron overload without the necessity of an invasive liver biopsy. Several polymerase chain reaction (PCR)-based methods have been developed for detection of mutations underlying haemochromatosis, including a reverse-hybridisation method that allows simultaneous analysis of multiple mutations in a single reaction (Oberkanins et al. 2000). The haemochromatosis strip-assay currently includes 17 mutations in three genes and validation of this assay in the South African population (Kotze et al. (2004)) accurately produced the correct genotype. This is an important consideration, because the gene regions of relevance to PCR-based tests for HH frequently contain sequence changes that may interfere with the test procedure and data interpretation (de Villiers and Kotze 1999). Guidelines for genetic testing and diagnosis of haemochromatosis are provided in the flow diagram in Figure 1.

Table I: Different genes underlying iron overload disease subtypes

Gene	Subtype	Main Organs affected	Organ damage	Symptomatic
Human Iron (HFE)	Type I, AR	Liver, endocrine glands, heart	Variable	4-5th decade
Hemojuvelin (HJV)	Type II, AR	Liver, endocrine glands, heart	High	2-3rd decade
Hepcidin (HAMP)	Type IIB, AR	Liver, endocrine glands, heart	High	2-3rd decade
Tf receptor (TFR-2)	Type III, AR	Liver, endocrine glands, heart	Variable	4-5th decade
Ferroportin (SLC40A1)	Type IV, AD	Liver spleen	Low	4-5th decade

AR, autosomal recessive inheritance; AD, autosomal dominant inheritance

Figure 1: New guidelines for genetic testing and diagnosis of Haemochromatosis



Treatment

Weekly therapeutic phlebotomy of 500 ml whole blood (equivalent to approximately 250 mg iron) may need to be performed in HH patients with high iron stores (Bacon et al. 1997). Regular venesection should be continued until ferritin levels are <50 ng/ml. Although some patients with HH, for reasons that are unclear at this time, do not reaccumulate iron, most patients will require maintenance phlebotomy of 1 unit of blood to be removed every 2-3 months.

Conclusions

With patients becoming increasingly aware of available genetic testing options, it is important that the family doctor becomes knowledgeable in identifying and advising patients at increased risk of iron overload. To facilitate interpretation of genetic test results, information on clinical symptoms and iron parameters has to be provided when patients are referred for exclusion/confirmation of a clinical diagnosis of HH, or for pre-clinical diagnosis and determination of carrier status in affected families.

References

1. Bacon BR. Diagnosis and management of hemochromatosis. Gastroenterology 1997; 113: 995-999.
2. De Villiers JNP, Hillermann R, de Jong G, Langenhoven E, Rossouw H, Marx MP, Kotze MJ. High prevalence of the Cys282Tyr HFE mutation facilitates an improved diagnostic service for hereditary haemochromatosis in South Africa. S Afr Med J 1999; 89: 279-282.
3. De Villiers JNP, Hillermann R, Loubser L, Kotze MJ. Spectrum of mutations in the HFE gene implicated in haemochromatosis and porphyria. Hum Mol Genet 1999b; 8: 1517-1522.
4. De Villiers JNP, Kotze MJ. Significance of linkage disequilibrium between mutation C282Y and a MseI polymorphism in population screening and DNA diagnosis of hemochromatosis. Blood Cells Mol Dis 1999; 15: 250-252.
5. Feder JN, Gnirke A, Thomas W, et al. A novel MHC class I-like gene is mutated in patients with hereditary haemochromatosis. Nature Genet 1996: 13: 399-408.
6. Niederau C, Fischer R, Purschel A, Stermmel W, Haussinger D, Stohmeyer G. Long-term survival in patients with hereditary hemochromatosis. Gastroenterology 1996: 110: 1107-1119.
7. Kotze MJ, de Villiers JNP, Bouwens CSH, Warnich L, Zaahl MG, van der Merwe S, Oberkanins C. Molecular diagnosis of hereditary haemochromatosis: application of a newly-developed reverse-hybridisation assay in the South African population. Clin Genet 2004; 65: 317-321.
8. Milani MY, Kotze MJ. Molecular diagnosis of hereditary haemochromatosis: identify an affected person and save a family. S Afr Med J 1999; 89: 263-264.
9. Oberkanins C, Moritz A, de Villiers JNP, Kotze MJ, Kury F. A reverse-hybridization assay for the rapid and simultaneous detection of nine HFE gene mutations. Genet Testing 2000; 4: 121-124.
10. Zaahl MG, Merryweather-Clarke AT, Kotze MJ, van der Merwe S, Warnich L, Robson KJ. Analysis of genes implicated in iron regulation in individuals presenting with primary iron overload. Hum Genet 2004; 115: 409-417.