

## **Diagnosis and therapy Of Genetic Haemochromatosis (Review and 2017 update)**

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### Guideline writing methodology

The original literature review (Worwood and Dooley 2000) was based on a total of forty years experience in genetic haemochromatosis (GH) by the authors, on searching the literature using appropriate keywords (in Medline and BIDS) and a review of the existing guidelines published by Expert Groups. The guideline has now been updated to incorporate new evidence up to and including 2017, to coincide with the development of a Guideline for the diagnosis and management of a raised serum ferritin (Cullis et al (submitted for publication) 2017).

### Evidence and strength of recommendations

#### Randomised controlled trials

In 1976 the results of a study of venesection therapy for the removal of excess iron from patients with GH were published ( Bomford & Williams 1976). The authors compared the survival of treated patients with that of historical controls (patients presenting with GH but not receiving phlebotomy). For patients receiving venesection therapy, life expectancy was significantly improved compared with that for untreated patients. This result has since been confirmed and extended in larger studies ( Adams et al 1991; Niederau et al 1996 ) If phlebotomy is started before cirrhosis and diabetes have developed, life expectancy is normal. However, the obvious success of this treatment has meant that it has not been ethically acceptable to compare venesection with ‘no treatment’

For similar reasons there have been no randomised controlled trials of the effect of treatment in genetically predisposed family members of probands with GH. Phlebotomy is undertaken if there is biochemical evidence of iron overload and it has not been acceptable to delay treatment in order to follow the development of clinical symptoms or assess the value of phlebotomy in preventing disease.

For these reasons there are no recommendations graded A.

### **Summary of recommendations:**

1. *Unselected population screening for HFE gene mutation is not recommended. (1B)*

2. *GH patients who present with SF >1000µg/l and any with raised transaminases should be referred to a hepatologist for fibrosis assessment and exclusion of cirrhosis.(1B)*
3. *Liver biopsy is no longer required for diagnosis of HFE GH but may be required to assess the severity of fibrosis in GH patients with SF >1000µg/l and/or elevated transaminases. Transient elastography could be used to select which patients from this group require liver biopsy.(2C)*
4. *Patients of north European ancestry with clinical features suggestive of GH should have the following laboratory investigations; FBC, LFTs, SF and Tsat. Molecular testing for HFE GH should follow if results fulfil the criteria of recommendation 5 (see below). (1B)*
5. *All adult patients of north European ancestry with raised SF and random Tsat (>300µg/l and > 50% males; > 200µg/l and > 40% females) and normal FBC should have molecular testing for HFE GH.(1B)*
6. *Laboratory screening to include FBC, LFTs, SF, Tsat and HFE should be offered to family members after the diagnosis of HFE GH. Family screening should include parents ( if available), siblings, partner and children (over the age of consent). Extended family screening is not recommended following identification of an individual as a C282Y/H63D compound heterozygote. (1B)*
7. *Investigation of all confirmed C282Y homozygotes should include FBC, LFTs, SF and Tsat. Thereafter further investigation may be required as follows:*
  - a. *SF < 1000µg/l, normal LFTs, normal clinical examination; no further investigation required. Follow recommendation 8.(1C)*
  - b. *SF > 1000µg/l and or abnormal LFTs. All such patients require referral to Hepatology for fibrosis assessment to exclude the presence of cirrhosis. A minimum would be elastography. For patients with confirmed cirrhosis monitor with AFP and hepatic ultrasound every 6 months (recommendation 11). (2C)*
8. *Non C282Y homozygotes with significant iron loading as confirmed by MRI and or liver biopsy should be investigated for rare iron loading genotypes or digenic inheritance.(1C)*
9. *At diagnosis all fit GH patients with biochemical iron loading should undergo weekly venesection until SF < 20-30µg/l and Tsat< 50%. During this phase of venesection FBC should be monitored weekly and SF ± Tsat monitored monthly. Homozygotes with normal iron indices and compound heterozygotes with minimal elevation of iron indices may be suitable for blood donation and annual monitoring of SF and Tsat.(1B)*
10. *During maintenance venesect as required, preferably at a blood donation centre to maintain normal FBC, SF < 50 µg/l and Tsat<50%(1C)*

***11. HFE GH with cirrhosis; Treat as per recommendations 9 and 10 but not suitable for blood donation. Monitor AFP and hepatic ultrasonography every 6 months.(2C)***

**Iron balance**

Body iron content is in the order of 4g and is controlled by the complex regulation of dietary iron absorption from the upper gastrointestinal (GI) tract. There is no regulatory pathway for the excretion of excess iron and daily involuntary losses of ~1mg occur via desquamation of cells in the GI tract. In females additional iron loss occurs due to menstrual loss (20-40mg iron per period) and pregnancy (net iron loss from placental transfer to foetus and blood loss at delivery is ~ 500mg/pregnancy). As a result of these minor losses the adult male/female will only require to absorb 1-2 mg/day of dietary iron to maintain normal iron balance

Most iron (3g) is present in haemoglobin with some 300mg in myoglobin. There are many iron containing enzymes involved in respiration in all tissues (100mg), and there is a small amount of iron (4mg) bound to transferrin (Tf) at any one time in the plasma and extravascular circulation.

Transferrin is a glycoprotein produced in the liver and regulated by body iron stores. In iron deficiency transferrin production increases and in iron overload decreases. Each Tf molecule has 2 iron binding domains. Over the course of 24h however Tf will transport some 20-40mg of iron from the body stores to the tissues. In health Tf is approximately 30% saturated with iron with only some 10% in the diferric state although diferric Tf will appear more frequently as the transferrin saturation (Tsat) increases. All cells accept iron from Tf via interaction with their cell surface transferrin receptors (TfR) and will accept iron more readily from diferric rather than monoferric Tf (Young et al 1984; Heubers et al 1985). Macrophages of the reticuloendothelial system (RES) have an additional and far greater supply of iron from the ingestion and degradation of effete RBCs.

Ferritin is present in all cells and provides safe iron storage in a readily accessible soluble form. In the event of intracellular iron excess ferritin molecules may aggregate to form insoluble haemosiderin. The largest body stores of iron exist within the macrophages of the RES, which in health may store up to 1g of iron as either ferritin or haemosiderin.

A tiny amount of ferritin (serum ferritin, SF) can be detected in the serum. The source of this SF is unknown but it contains little iron and plays no role in iron metabolism. In the vast majority of patients without either tissue damage or inflammation, the SF value is directly proportional to the amount of RES macrophage storage iron (Worwood 1982). It is reduced in states of iron deficiency and raised in states of iron overload. Control of iron metabolism is orchestrated by the hormone hepcidin which together with the iron transporter ferroportin acts to regulate both the absorption of dietary iron and RES macrophage iron release (Nemeth et al 2004).

## Mechanism of iron toxicity

As serum iron levels rise Tf levels fall, T<sub>sat</sub>% rises and highly toxic non transferrin bound iron (NTBI) may appear in the plasma. Parenchymal cells will accept more Tf-iron as the T<sub>sat</sub>% and diferric transferrin levels rise and have no defence against the entry of NTBI (Porter et al 2014). Within the cell iron toxicity is thought to occur via pathways such as the Fenton reaction, free radical formation and lipid peroxidation.

## **Iron loading genotypes**

### HFE GH (Type 1 GH)

The syndrome now recognised as HFE-related haemochromatosis was first described in 1865 by Trousseau and the name was first used by von Recklinghausen in 1889. In 1935 Sheldon reviewed more than 300 published cases and suggested that the disorder resulted from an inherited defect. In 1996 a HLA-class-I-like gene (HFE) was described (Feder et al 1996) in which there were mutations in most patients satisfying the diagnostic criteria for GH. Over 90% of patients with GH carry a mutation of the HFE gene at amino acid 282 which results in the replacement of cysteine by a tyrosine residue (C282Y). They also described a second variant at amino acid 63, in which aspartic acid replaced histidine (H63D). This second mutation is more common in the general population but is not usually associated with iron accumulation.

GH is one of the most frequent genetic disorders found in populations of northern European descent. It seldom causes end organ damage before the age of 30 years, is inherited in an autosomal recessive fashion and is characterised by excessive iron absorption from the GI tract, excessive iron release from RES macrophages, excessive iron saturation of plasma transferrin and consequently parenchymal iron overload and toxicity.

The HFE gene product is a 343 residue type 1 transmembrane glycoprotein. This protein has a complex and as yet imperfectly understood interaction between transferrin receptor 2 (TfR2), hemojuvelin (HJV), bone morphogenic protein (BMP) receptor Alk3 and ligands which control hepcidin expression in hepatocytes (Wu et al 2014). Hepcidin inhibits the efflux of iron from both enterocytes and RES macrophages by inducing the degradation of the iron exporter ferroportin. In HFE GH hepcidin levels are suppressed leading to reduced ferroportin degradation and increased iron efflux from enterocytes and macrophages (Figure 1).

Variants C282Y and H63D of the HFE gene (chromosome 6p21.3) are the 2 polymorphisms most frequently associated with HFE GH. Studies of allele frequencies in northern European populations have reported 10-15% C282Y heterozygotes, 0.25-1.0% C282Y homozygotes, 20-30% H63D heterozygotes and about 2% C282Y/H63D compound heterozygotes (Worwood 2005). Of the various possible genotype combinations C282Y homozygotes are most at risk of iron overload and such homozygotes are responsible for some 90% of clinical cases. However many young genetically predisposed homozygotes will have normal SF and/or T<sub>sat</sub>. Over time some 80% and 50% of these male and female homozygotes

will show biochemical penetrance with raised T<sub>sat</sub> and SF values. Clinical penetrance is considerably lower and the risk profile for iron loading for the possible genotypes can be summarised as follows;

C282Y/C282Y >>> C282Y/H63D > C282Y/-, H63D/H63D, H63D/-

Compound heterozygotes (C282Y/H63D) will seldom develop iron overload without additional factors such as alcohol excess or metabolic syndrome which themselves may contribute more to any hyperferritinaemia than the molecular abnormality itself (Cullis et al 2017). Indeed some recommend that molecular testing for GH should be confined to the C282Y mutation alone (Porto et al 2016). In the UK only C282Y heterozygotes are tested for the H63D mutation and in France laboratories are not reimbursed for H63D genotyping.

GH patients who lack C282Y mutations of the HFE gene may require sample referral to specialist centres for detection of very rare iron loading genotypes. Similarly simple C282Y heterozygotes and C282Y/H63D compound heterozygotes with unexpected severe iron loading should also be investigated for possible digenic inheritance of other contributing iron loading genotypes (Merryweather-Clarke et al 2003). In the UK this service is available at The Institute of Molecular Medicine, University of Oxford, John Radcliffe Hospital, Oxford OX3 9DS; <https://www.imm.ox.ac.uk> and through the clinical service provided by the UK Genetic Testing Network via Molecular Genetics at Cambridge University Hospitals: <https://ukgtn.nhs.uk>.

#### Rare genotypes and rare forms of GH (reviewed by Camaschella and Poggiali 2009)

##### *Type 2 Juvenile GH.*

This is a rare autosomal recessive disorder affecting either the HJV gene encoding hemojuvelin (type 2A) or more rarely the HAMP gene encoding hepcidin (type 2B). Iron overload is severe. It develops in younger patients and commonly causes cardiac failure from iron overload and panhypopituitarism (Porto et al 2016).

##### *Type 3 GH, Transferrin receptor 2 deficiency*

This is a rare autosomal recessive disorder caused by mutations in the TFR2 gene coding for transferrin receptor 2. This has been found in European and Japanese families, leads to high T<sub>sat</sub> and a clinical phenotype similar but intermediate in severity to Type 1 and Type 2 GH (Olynyk et al 2008).

##### *Type 4 GH, Ferroportin disease.*

Type 4a is a rare autosomal dominant disorder with a broad geographical distribution including Asia. Mutations in the ferroportin gene lead to 'loss of ferroportin function', reduced RES macrophage iron release and consequent RES iron overload. Serum ferritin is raised with T<sub>sat</sub> values either low or normal. Significant parenchymal iron overload and organ damage does not occur but iron deficiency anaemia can develop with venesection.

Type 4b is also a rare disorder in which the mutations disrupt the ferroportin hepcidin axis with 'gain of ferroportin function' causing a clinical phenotype similar to Type 1 GH (Olynyk et al 2008).

### *Type 5 GH*

Iron overload, inherited in an autosomal dominant manner from a mutation in FTH 1 affecting the iron responsive element of H ferritin mRNA has been described in a Japanese family (Kato et al 2001)

### Early diagnosis of GH and life expectancy

Early diagnosis is not easy as the symptoms with which many patients present are relatively common and non-specific. Unfortunately many patients in primary care in the UK with raised SF values are not screened for GH (Ogilvie et al 2010). If patients are however diagnosed in the pre-cirrhotic, pre-diabetic stage and treated by venesection to remove the excess iron then life expectancy is normal (Niederau et al 1996). However, once cirrhosis and diabetes mellitus have developed, patients have a shortened life expectancy and, if cirrhosis is present, a high risk of liver cancer even when iron depletion has been achieved. It is therefore important to diagnose the condition as early as possible and multiple studies, as described below have been performed to examine possible approaches to improve the early diagnosis.

### **Biochemical penetrance, population studies and screening for GH**

A study of 65,000 volunteers in Norway found that < 10% of individuals with raised non-fasting transferrin saturation had HFE GH (Asberg et al 2001). As such it is now recognised that the biological variability of serum iron levels and hence Tsat, limits the use of Tsat alone in screening for GH (Adams & Barton 2007).

SF values are raised in about 80% of male and 50% of female homozygotes (Adams & Barton 2007)). Although measurement of SF might then be considered an effective approach for screening for GH the vast majority of patients with raised SF do not have GH. The additional finding of a raised Tsat however does increase the likelihood of GH being the cause of a raised SF. The measurement of fasting morning Tsat values has long been advocated to assist the diagnosis of GH by avoiding the effects of diurnal variation and dietary intake on serum iron levels. However repeated venous sampling of patients is often impractical and although a raised Tsat in non homozygotes may normalise when measured in the fasting state they do not improve the predictive value of random Tsat for diagnosing HFE GH (Adams et al 2005).

Four major studies of volunteer screening for GH have been reported: the Hemochromatosis and Iron-overload Screening (HEIRS) study (Adams et al 2005), the Scripps/Kaiser hemochromatosis screening study (Wallen et al 2008), the Melbourne Collaborative Cohort study-Health Iron (Allen et al 2008) and a study of 10,500 blood donors from South Wales (Jackson et al 2001). A non volunteer study of patients with raised SF values, taken at the initiative of Primary Care physicians, without influence from the laboratories or secondary care has also recently reported (Ogilvie et al 2015). The results of these studies are summarised in Table 1. It is noted however that a limitation of these approaches to improve early diagnosis is that females predominate in both volunteer studies and GP requests for SF (Adams et al 2005), while clinically significant GH predominates in males.

The different mean/median SF and T<sub>sat</sub> values for newly diagnosed GH reflect the different age groups included in the 4 studies. The lowest values were found in the Welsh blood donor study which included subjects as young as 17 with a mean age of 36 years. The highest values were found in the Melbourne study which only accepted volunteers 40-70yrs. The apparent predominance of new female homozygotes in HEIRS, Melbourne and West of Scotland publications simply reflects the predominance of female volunteers and patients. From the West of Scotland study the following predictive algorithm was developed to assist early diagnosis for patients > 30yrs in primary care;

***Males with SF >300µg/l and T<sub>sat</sub>>50% and females with SF >200µg/l and T<sub>sat</sub>>40% will have a 19% and 16% likelihood of being C282Y homozygotes.***

Taken together all studies show that GH is almost exclusively a disease of white races, that >70% of patients/volunteers at the time of diagnosis have SF values < 1000µg/l, that male homozygotes have higher SF and T<sub>sat</sub> values than female homozygotes and are 3-5 times more likely to have SF values >1000ug/l. .

#### GH and Serum Ferritin values > 1000µg/l

Serum ferritin values >1000µg/l in homozygotes at presentation are significant as studies have shown that these patients are at greatest risk of clinical disease, cirrhosis and hepatocellular carcinoma (Powell et al 2006; Allen et al 2008). Most individuals however with SF >1000µg/l do not have GH (see Table 1). The HEIRS study of almost 100,000 multiethnic volunteers found 364 patients (of whom only 29 were C282Y homozygotes) with SF values > 1000ug/l. Such high values were 2-3 times more prevalent in Blacks and Asians than in whites and were not related to HFE genotype.

The prevalence of SF values < 1000µg/l appears to be more common in primary care patients (Ogilvie et al 2010) than in volunteer studies but even in an area of high prevalence only 29 of 180 such patients were found to be C282Y homozygotes (Ogilvie et al 2015).

#### Non progressive GH

These and other studies have shown that SF values in adults with GH do not inevitably show a progressive rise and may remain relatively stable over many years (Olynyk 2004; Waalen et al 2008). Powell et al (2006) observed a subgroup of untreated homozygotes with normal SF values for up to 24yrs. Presumably then a steady (but as yet unexplained) state develops in homozygotes when a certain level of iron is reached but that this level will differ between patients.

### **TABLE 1**

#### **Biochemical Expression C282Y Homozygous Genetic Haemochromatosis in 5 Major Population Studies**

## **Clinical penetrance of C282Y homozygous GH. Effect of venesection on hepatic fibrosis.**

Clinical penetrance is very much less common than biochemical penetrance and for this reason population screening for HFE genotype is not recommended.

### ***Recommendation 1***

***Unselected population screening for HFE gene mutation is not recommended.(1B)***

Different studies report different estimates of clinical penetrance that range from 0% (Jackson et al 2001) and < 1% (Beutler et al 2002) to 28-30% for male homozygotes and 1-11.5% for female homozygotes (Powell et al 2005, 2006; Allen et al 2008) These differences relate to the different study populations and their varying degrees of iron loading.

### Hepatic pathology

Liver biopsy performed on 350 asymptomatic homozygotes (SF values usually > 500 $\mu$ g/l ) found stage 2-4 hepatic fibrosis in 29.7% and 13.3% of male and female patients respectively. The presence of hepatic fibrosis and cirrhosis correlated significantly with hepatic iron concentration and SF in both men and women. No case of cirrhosis was found in patients with SF values < 1000  $\mu$ g/l (Powell et al 2006).

### Role of liver biopsy and Imaging techniques

Liver biopsy is no longer required for GH diagnosis but remains important for both fibrosis assessment and exclusion of additional liver diseases such as alcoholic and non-alcoholic fatty liver. The importance of fibrosis assessment is to select those who require surveillance for hepatocellular carcinoma, essentially those with cirrhosis. Significant fibrosis is excluded in C282Y homozygotes with normal transaminases and SF values <1000 $\mu$ g/l (Guyader et al 1998). In addition, transient elastography (TE and often referred to as FibroScan) can further refine the cohort that require biopsy. This is a non invasive method of assessing liver fibrosis which has been validated in most forms of chronic liver disease. The shear wave velocity correlates with liver stiffness, most accurately at the lower and higher ends of the range. In homozygotes with raised transaminases or SF values >1000 $\mu$ g/l a TE < 6.4 kPa excluded significant fibrosis whereas TE > 13.9 was predictive for severe fibrosis (Legros et al 2015). Serum hyaluronic acid measurement has also shown excellent sensitivity and specificity for cirrhosis when values >46.5 ng/ml are obtained but this assay is not in routine clinical use (Crawford et al 2009).

### ***Recommendation 2***

***GH patients who present with SF >1000µg/l and any with raised transaminases should be referred to a hepatologist for fibrosis assessment and exclusion of cirrhosis.(1B)***

Magnetic resonance imaging (MRI) can be used for iron quantification with reasonable accuracy using the T2\*method (St Pierre et al 2005).The latter is helpful in patients negative for HFE mutations where SF may not correlate directly with the degree of body iron loading. MRI has the advantage over biopsy of being able to identify heterogeneous iron distribution within the liver. It may also obviate the immediate need for liver biopsy to determine the necessity for venesection in non HFE cases. Biopsy however remains the gold standard for iron quantification where iron is typically graded 1-4 or if needed more accurately by measurement of the historical hepatic iron index (HII). Patients with unexplained significant iron overload should be considered for extended genetic testing.

### ***Recommendation 3***

***Liver biopsy is no longer required for diagnosis of HFE GH but may be required to assess the severity of fibrosis in GH patients with SF >1000µg/l and or elevated transaminases. Transient elastography could be used to select which patients from this group require liver biopsy.(2C)***

Phlebotomy programmes deliver significant (7.5 fold) reduction in mean hepatic fibrosis scores. They have also been used to provide quantitative estimates of iron depletion leading to estimates that 1µg/l SF in homozygotes represents approx 3.3 mg of storage iron (Powell et al 2006) and that SF values >900µg/l correlate with > 4g of storage iron in men and >2g in women (Gordeuk et al 2008).

### **Bronzed diabetes and other endocrine disorders**

Although ‘Bronzed Diabetes’ remains the historical moniker for GH, studies of HFE mutation frequencies in diabetes mellitus have not shown any increase in frequency compared with control groups (Frayling et al 1998). These results however more likely reflect the now overwhelming contribution of obesity to the development of diabetes rather than any lack of iron toxicity on the pancreas in GH.

Hypogonadotropic hypogonadism can develop in cases with severe iron loading as the result of iron deposition in pituitary.

### **GH arthropathy**

GH arthropathy typically affects the second and third metacarpophalangeal joints (painful handshake sign). It is unrelated to SF values and indeed seldom responds to venesection and iron depletion. It does however appear to develop more frequently in known homozygotes who fail to maintain Tsat < 50% (Bardou-Jacquet et al 2017)

### **GH cardiac toxicity**

Cardiac toxicity in the forms cardiomyopathy and conduction defects can occur in GH but are more frequently seen in patients with transfusional iron overload

### **Diagnosis of HC; patient selection and laboratory evaluation.**

As clinical penetrance is low and highly variable unselected population screening for HFE-GH is not recommended. As such the appropriate investigation of patients and individuals for GH is currently restricted to;

#### *1. Patients with symptoms in keeping with iron overload*

Symptoms are however often vague and longstanding. Suspicion should however be raised for male subjects of north European ancestry presenting with unexplained weakness or fatigue, abnormal liver function tests, arthralgia/arthritis, impotence, diabetes of late onset, cirrhosis, or bronze pigmentation.

It is recommended that initial screening requests for GH in these patients should include FBC, SF, serum iron and Tsat. There is no place for the single measurement of either SF or Tsat in the preliminary detection of GH. Both measurements are essential. Thereafter for patients >30yrs from Primary care in areas of high prevalence if SF is beyond the normal range (200 and 300 $\mu$ g/l F:M) and Tsat >40% and 50% (F:M) HFE genotyping should be performed with an expected diagnostic yield of almost 20% for HFE-GH (Ogilvie et al 2015). There is no such evidence for patients <30yrs but similar values might be expected, particularly in males.

#### **Recommendation 4**

***Patients of north European ancestry with clinical features suggestive of GH should have the following laboratory investigations; FBC, LFTs, SF and Tsat. Molecular testing for HFE GH should follow if results fulfil the criteria of recommendation 5 (see below). (1B)***

#### *2. Patients with unexplained raised SF (a frequent incidental finding)*

Raised SF values are found frequently in secondary care where they often occur in the context of alcohol abuse, established liver disease, malignant disease, renal dialysis, inflammatory disease and transfusional or secondary iron overload (see Cullis et al 2017).

There is already a high and increasing number of SF requests being made from primary care. These samples can be exploited to detect undiagnosed homozygotes (Ogilvie et al 2015). In primary care SF values above the upper limits of normal (300 $\mu$ g/l for adult males and 200 $\mu$ g/l for adult females) can be found in almost 20% of males > 30yrs and in 10% and 17% of females 50-70yrs and > 70yrs respectively. These raised values frequently appear to be unexplained (Ogilvie et al 2010). For such patients FBC and Tsat values as described above should guide requests for HFE genotype

### **Recommendation 5**

**All adult patients of north European ancestry with raised SF and random T<sub>sat</sub> (>300µg/l and > 50% males; > 200µg/l and > 40% females) and normal FBC should have molecular testing for HFE GH. (1B)**

3. Targeted screening of family members of an index case of C282Y homozygous GH. Siblings, parents and children (over the age of consent) of a patient should be offered testing. Testing of partners can assist in determining the risk for children. It is not recommended that family screening be performed after identification of a heterozygote carrier or indeed a compound heterozygote. Initial family screening should be as above but also include HFE genotype with expected frequencies of 25% and 50% for GH when parents are either both heterozygotes or one heterozygous and the other homozygous

### **Recommendation 6**

**Laboratory screening to include FBC, LFTs, SF, T<sub>sat</sub> and HFE should be offered to family members after the diagnosis of HFE GH. Family screening should include parents (if available), siblings, partner and children (over the age of consent). Extended family screening is not recommended following identification of an individual as a C282Y/H63D compound heterozygote. (1B)**

### **Recommendation 7**

**Investigation of all confirmed C282Y homozygotes should include FBC, LFTs, SF and T<sub>sat</sub>. Thereafter further investigation may be required as follows;**

- c. **SF < 1000µg/l, normal LFTs, normal clinical examination; no further investigation required. Follow recommendation 8. (1C)**
- d. **SF > 1000µg/l and or abnormal LFTs. All such patients require referral to Hepatology for fibrosis assessment to exclude the presence of cirrhosis. A minimum would be elastography. For patients with confirmed cirrhosis monitor with AFP and hepatic ultrasound every 6 months (recommendation 11). (2C)**

### **Recommendation 8**

**Non C282Y homozygotes with significant iron loading as confirmed by MRI and or liver biopsy should be investigated for rare iron loading genotypes or digenic inheritance. (1C)**

### **Treatment: at diagnosis**

Following confirmation of GH the decision to start venesection should be made on the basis of both clinical and laboratory findings. The frequency of venesections may

need to be individualised, taking into account the presenting severity of iron overload, organ damage, age, comorbidities and tolerability of the procedure. The risks of venesection *per se* in a frail newly diagnosed patient with only minor iron loading may well outweigh any potential benefit.

Venesection is indicated for all fit patients with biochemical iron overload with or without clinical features. In line with EASL guidelines (2010) and previous BCSH recommendations (2000) this should initially be performed once weekly (450-500ml, ~200-250mg iron). Monitor Hb levels weekly, SF monthly and reduce rate of venesection if anaemia develops. Transferrin saturation should be measured every 3 months as a disproportionate fall in Tsat and Hb would occur if other factors (such as alcohol or metabolic syndrome) together with GH were contributing to the raised SF. Continue with venesection until SF concentration is 20-50µg/l and Tsat is <50%.

Evidence IIb–IV; Grade B, C

### ***Recommendation 9***

***Treatment at diagnosis. All fit GH patients with biochemical iron loading should undergo weekly venesection until SF < 20-30µg/l and Tsat < 50%. During this phase of venesection FBC should be monitored weekly and SF ± Tsat monitored monthly. Homozygotes with normal iron indices and compound heterozygotes with minimal elevation of iron indices may be suitable for blood donation and annual monitoring of SF and Tsat. (1B)***

### Iron depletion for patients who cannot tolerate venesection

In rare circumstances such as juvenile haemochromatosis or delayed diagnosis it may be necessary to use iron chelation therapy to reverse cardiac damage. Physicians dealing with the treatment of iron overload due to blood transfusion in patients with homozygous beta-thalassaemia have great expertise in dealing with heart failure and should be consulted.

There is limited experience of using the oral iron chelator deferasirox (Exjade) in patients with C282Y homozygous GH and modest iron loading (median SF 645µg/l). SF values were modestly reduced but significant patient withdrawal from gastrointestinal, renal and hepatic toxicities was observed (Phatak et al 2010).

### Homozygotes without iron accumulation

For those subjects identified by family screening as C282Y homozygous but without iron accumulation, it is reasonable to monitor SF and iron status at yearly intervals in order to detect the onset of tissue iron accumulation. These patients may be encouraged to become blood donors (see below).

## **Treatment: in maintenance phase**

Once excess iron has been removed and treatment by phlebotomy has ceased, iron will begin to re-accumulate. The actual rate of re-accumulation cannot be predicted but will occur more rapidly in those with marked iron loading at presentation. Many patients appear to lose some or all iron loading tendency over time and maintain normal iron balance with decreasing need for venesection. Patients usually return to the outpatient clinic every 3-6 months, and further phlebotomy is carried out when necessary.

The SF should be maintained at  $< 50 \mu\text{g/l}$ . The question as to whether C282Y homozygotes with a raised Tsat despite  $\text{SF} < 50 \mu\text{g/l}$  should be treated has not been fully resolved. This situation is frequently found during maintenance. Previous BCSH guidelines (2000) recommended that maintenance venesection programmes should maintain  $\text{SF} < 50 \mu\text{g/l}$  and  $\text{Tsat} < 50\%$ . Guidelines however prepared by the American Association for the study of liver diseases (AASLD; Bacon et al 2011) and European Association for the Study of the Liver (EASL 2010) only recommend keeping SF values between 50-100  $\mu\text{g/l}$  and do not recommend Tsat monitoring. For the reasons given below however we have chosen to follow previous BCSH guidance and recommend monitoring Tsat values during maintenance. Firstly it would seem unwise for a patient with high SF and iron loading at diagnosis not to restart venesection as Tsat starts to rise once again. Furthermore as cellular iron uptake and the risk of NTBI increase as Tsat values rise and as GH arthropathy has recently been shown to develop more often in patients who fail to maintain  $\text{Tsat} < 50\%$  (Bardou-Jacquet 2017) we would recommend maintenance venesection targets for both  $\text{SF} < 50 \mu\text{g/l}$  and  $\text{Tsat} < 50\%$  in all fit patients.

Patients frequently ask about dietary restriction of iron rich foods or the use of proton pump inhibitors (Hutchison et al 2007) to reduce iron absorption. Such measures may offer some benefit particularly for patients who do not tolerate or comply with venesections but venesection remains the mainstay of maintenance treatment.

### ***Recommendation 10***

***During maintenance venesect as required, preferably at a blood donation centre to maintain normal FBC,  $\text{SF} < 50 \mu\text{g/l}$  and  $\text{Tsat} < 50\%$ . (1C)***

#### Management of patients with liver cirrhosis

As these patients have a 100 fold increased risk of developing primary liver cancer,  $\alpha$ -fetoprotein levels and hepatic ultrasonography should be carried out every 6 months. Evidence IIb–IV; Grade D

### ***Recommendation 11***

***HFE GH with cirrhosis; Treat as per recommendations 9 and 10 but not suitable for blood donation. Monitor AFP and hepatic ultrasonography every 6 months. (2C)***

#### Homozygotes and blood donation

In some countries homozygotes are encouraged to donate blood to the transfusion service either after initial venesections have restored normal iron stores or to prevent iron loading in asymptomatic homozygotes with normal SF values. This has now been accepted by UK Transfusion services (Joint Public Advisory Committee (JPAC) at [www.transfusionguidelines.org.uk](http://www.transfusionguidelines.org.uk)) although the responsibility to monitor SF and T<sub>sat</sub> remains with the referring clinician. It would seem reasonable to suggest that such patients with GH be allowed to donate blood three or four times a year and so provide mutual benefit to both patient and society .

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