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Independent learning program for GPs

Unit 475 October 2011

Genetic disorders

The Royal Australian College of General Practitioners

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Genetic disorders

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This unit of *check* looks at genetic disorders. There is a range of conditions that can be classified as genetic in origin. Genetic disorders include conditions encoded for by a single gene or multiple genes, conditions due to a chromosomal abnormality, conditions with an appreciable genetic component but whose pattern of inheritance has not yet been identified, and congenital malformations. Genetic disorders which are not uncommonly seen in general practice have been selected for this unit of *check*.

Knowledge of the pattern of inheritance of specific genetic disorders, when to consider testing, and what investigations to perform, allows for identification of these disorders or the predisposition to them. The general practitioner and the familial clinic play important roles with respect to testing for genetic disorders, treatment of genetic disorders, screening for complications and genetic counselling about the implications for other family members including children.

The authors of this unit bring a wealth of knowledge and experience in the area of genetic disorders.

The authors of this unit are:

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The learning objectives of this unit are to:

- display increased knowledge of the pattern of inheritance of particular genetic disorders such as haemochromatosis, factor V Leiden mutation, alpha thalassaemia, breast cancer due to the BRCA gene mutation, and cystic fibrosis; the indications for genetic testing and the specific genetic tests available for diagnosis of these disorders or the predisposition to these disorders
- understand the importance of obtaining a family history and referring to a familial clinic where appropriate
- understand the importance of offering screening for Down syndrome to pregnant women of all ages
- effectively counsel couples with a high risk first trimester screening result and provide them with accurate information about prenatal diagnostic testing for chromosomal abnormalities
- display increased awareness of the screening implications for an individual with a positive genetic test
- display increased knowledge of some of the treatments available for haemochromatosis, factor V Leiden mutation, alpha thalassaemia, breast cancer due to the BRCA gene mutation and cystic fibrosis.

We hope that this unit of *check* will assist you to confidently assess and manage patients who present with particular genetic disorders in general practice.

Kind regards

Catherine Dodgshun
Medical Editor

CASE 1

DOES WARREN'S FAMILY HISTORY PUT HIM AT RISK?

Warren is a plumber, aged 47 years, who presents to you as a new patient. He has tiredness and joint pain.

Warren says that he has been feeling exhausted at the end of each day for the last year, despite not working any harder than usual. The joints of the second and third digits of his right hand became swollen and painful about a year ago and the same joints of his left hand developed similar symptoms about 6 months ago. Warren has no other past medical problems and is not on any medications. He reports that he drinks an average of two cans of beer per day and eats red meat almost every day.

Warren's father was born in France and died at the age of 59 years from myocardial infarction. Warren's mother, whose ancestry is English, is alive and apparently healthy, as is his brother, who is 50 years of age. Warren's sister died recently at 54 years of age from liver cancer. Warren has two children, a boy 15 years of age and a girl aged 13 years.

On examination the second and third metacarpophalangeal joints of Warren's right hand are more swollen than on his left. Mild hepatomegaly is present. Blood pressure is within the normal range. His height is 168 cm and his weight is 85 kg.

You request the following blood tests:

- full blood examination (FBE)
- urea, electrolytes and creatinine (UEC)
- liver function tests (LFTs)
- fasting blood glucose level (BGL)
- testosterone
- follicle stimulating hormone (FSH)
- luteinising hormone (LH), and
- fasting iron studies.[1]

Results are all normal apart from the iron studies, which show a transferrin saturation (TS) of 54% and a serum ferritin (SF) of 800 µg/L.

QUESTION 1

How should these results be interpreted?

QUESTION 2

What is the mechanism of iron overload (haemochromatosis)?

QUESTION 3

What features in the history suggest a diagnosis of hereditary haemochromatosis (HH)?

FURTHER HISTORY

You request further fasting iron studies 1 month later. The results reveal a TS of 56% and a SF of 1100 µg/L; LFTs remain in the normal range.

QUESTION 4

What investigations would you request in order to determine if the iron overload is due to HH?

FURTHER INFORMATION

Investigations reveal that Warren is C282Y homozygous.

QUESTION 5

What treatment and follow up would you advise for Warren? What would you advise him about alcohol intake, dietary iron intake and vitamin C supplements?

QUESTION 6

If the haemochromatosis (HFE) gene test for Warren was negative but the iron studies persistently demonstrated iron overload, what follow up would you advise?

FURTHER INFORMATION

Warren informs his brother, Richard, that he is also at risk of having HH. Richard comes to see you and you organise HFE genetic testing. He has no clinical symptoms.

QUESTION 7

What advice would you give to Richard if his test results showed that he is a C282Y heterozygote?

QUESTION 8

What advice would you give to Richard if he were a C282Y homozygote?

QUESTION 9

Warren requests that you organise genetic testing for his children. What do you advise?

CASE 1 ANSWERS

ANSWER 1

A fasting TS >45% in combination with an elevated SF suggests early iron overload and hereditary haemochromatosis (HH). The combination of TS and SF is more sensitive than requesting an SF alone as the TS reflects increased iron absorption. The SF is also an acute phase reactant and can therefore be influenced by other factors.

Serum ferritin is abnormal when it is >250 µg/L in premenopausal women and >300 µg/L in men and postmenopausal women. It is often elevated in the setting of alcohol consumption, inflammation, malignancy and liver conditions, including fatty liver disease. Liver function tests may or may not be normal in early stages of HH.[2]

Iron studies need to be abnormal on more than one occasion to be further suggestive of HH as possible underlying causes for iron overload and storage also include:[3]

- nonalcoholic steatohepatitis
- alcoholic liver disease
- chronic viral hepatitis.

ANSWER 2

Iron accumulation occurs when there is abnormally elevated absorption of iron across the intestinal mucosa, together with an inactive iron excretory mechanism.[3]

People with HH have a mutation in their HFE gene which causes the HFE protein (located on intestinal and hepatic cells and which regulates absorption of iron) to malfunction.[3] As a result, they absorb and store dietary iron at 2–3 times the normal rate.[4] This leads to the accumulation of 0.5–1.0 g of iron per year. Once absorbed, there is no physiologic mechanism for excretion of excess iron from the body other than pregnancy or menstrual and other bleeding. The deposition of iron occurs in parenchymal cells of the liver, heart, pancreas and other organs.[4]

ANSWER 3

Clinical features in Warren's case that suggest HH include tiredness and arthralgia with joint swelling/tenderness in the second and third metacarpophalangeal joints, which is the most common pattern in HH[3] (*Figure 1*).

Common clinical features of HH include one or more of the following:[2]

- lethargy and weakness
- arthralgia
- loss of libido
- upper abdominal discomfort
- hepatomegaly
- grey/bronze skin pigmentation
- testicular atrophy
- joint swelling/tenderness, most often in the second and third metacarpophalangeal joints.

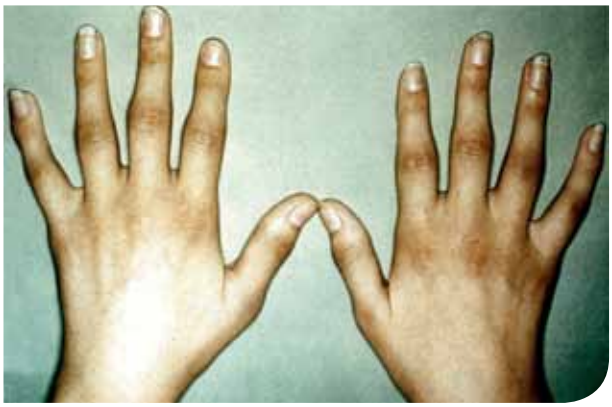

Figure 1. HH associated arthritis with swelling of the second and third metacarpophalangeal joints

Symptoms usually develop after decades of increased iron absorption. Symptoms usually appear after 15–20 g of iron accumulates in the body. Men therefore tend to become symptomatic in their 40s and women who stop menstruating develop symptoms about 15 years later.[4]

Warren's ancestry and family history supports a diagnosis of HH.

HH is a common condition most prevalent in people of northern European backgrounds (about 1 in 250).[2]

HH is an inherited autosomal recessive condition. While in this case, Warren had a sister who had liver cancer (which can develop in individuals who have HH complicated by cirrhosis of the liver), there is often no family history of the condition or affected family members may appear to be scattered among, or within, generations.

Where both parents are heterozygous (carriers for a mutation in the HFE gene), there is a 25% chance that each of their children will inherit a gene mutation in both copies of their HFE gene and be genetically predisposed to HH.

If only one parent is heterozygous, there is a 50% chance that each of their children will be an HFE gene mutation carrier.

ANSWER 4

Given that the fasting TS and SF levels are increased again, the genetic test for HH should be requested. The test is covered by Medicare if:

- there is elevated TS or elevated SF on testing of repeated specimens, or
- there is a first degree relative with haemochromatosis.

The genetic test tests for mutations in the HFE gene. Two mutations in the gene are believed to be the most common cause of HH: C282Y and H63D. This nomenclature is based on changes in the gene that then cause changes to the amino acid structure of the HFE protein.

Possible results of HFE gene testing include:

- C282Y homozygote:
  - C282Y mutation in both alleles (copies) of the HFE gene
  - about 90% of people of northern European ancestry with symptoms of HH are C282Y homozygotes
  - about 60–70% will develop iron overload during their lifetime[5]

- Compound heterozygote (C282Y/H63D):
  - a C282Y mutation in one allele of the HFE gene and a H63D mutation in the other allele of the HFE gene
  - only about 1% of people with this genotype develop HH
  - iron status should be monitored every 2–5 years
- C282Y heterozygote, H63D heterozygote or H63D homozygote:
  - patients should be reassured that their own risk of developing symptomatic HH is extremely small. However, they should be informed of the need to discuss with their relatives the need for testing
  - some patients may have minor abnormalities in iron studies
  - there is no need to monitor iron status unless initial TS or SF levels are abnormal or symptoms are present.

Frequency of HFE genotypes in the Australian population is shown in *Table 1*.

Other mutations have been described but their clinical significance is unknown.

ANSWER 5

Warren has HH. As iron overload is present, lifelong venesection and monitoring of iron studies is required.[1]

- An initial course of 1 or 2 venesections per week is usually required until excess iron stores are removed. (There is currently controversy as to an ideal target range for SF from venesection therapy, which varies from 50–300)
- Once iron levels are at low normal levels, patients usually require one venesection every 3–4 months to keep levels low without rendering the patient iron deficient
- The response to venesection treatment depends on the presenting symptoms and the stage of the condition at the time of diagnosis Venesection can be performed in a number of settings, including:
  - The Australian Red Cross Blood Service, which offers a therapeutic venesection service for patients with HH, provided that they do not suffer from a transfusion transmissible condition. The referring doctor is required to review the ongoing need for venesection at least every 12 months
  - some general practices (Medicare Item 13757 applies) in association with a hepatology clinic
  - some private pathology services.

Table 1. Frequencies of HFE genotypes in the Australian population[2]

HFE genotype	Frequency
No gene mutation found	2/3
Homozygous C282Y	1/200
Compound heterozygote (C282Y/H63D)	1/50
Heterozygous C282Y	1/10
Heterozygous H63D	1/6
Homozygous H63D	1/100

Given Warren's SF level and the presence of hepatomegaly, liver biopsy should be performed to establish or exclude the presence of cirrhosis.

There is no value in a low iron diet in the management of hemochromatosis.[2] Vitamin C (ascorbic acid) supplements should be avoided, since vitamin C increases iron absorption.

Warren should abstain from alcohol consumption until iron levels are normalised through venesection.

ANSWER 6

All patients with iron overload require follow up regardless of the HFE gene test result, because in a small percentage of cases of HH, a different, rarer gene mutation may be responsible.

Liver biopsy and histopathology examination is the most validated method for establishing or excluding hepatic cirrhosis and determining prognosis and risk of hepatocellular carcinoma. However, there is now a noninvasive, reasonably accurate measure of liver iron loading status through magnetic resonance imaging (MRI) of the liver using a newly patented technique called Ferriscan®.[3] The availability of this is currently limited. A flow chart for managing iron overload can be found on the Royal College of Pathologists of Australasia's website (see *Resources*).

ANSWER 7

If Richard is C282Y heterozygous, baseline iron studies should be performed. Iron status does not need to be monitored unless initial iron studies are abnormal, or symptoms or signs that suggest HH develop. However, Richard needs to be aware of the possible implications for his children.

ANSWER 8

Individuals who are C282Y homozygotes who have no clinical symptoms of HH should have their iron studies tested; 60–70% will develop iron overload in their lifetime but not all will develop iron overload related disease. If iron studies are normal on initial testing, they should be repeated every 2–5 years.

If iron studies are abnormal on initial testing, venesections should be performed until SF levels return to normal.

ANSWER 9

There have been no reports of iron overload from C282Y homozygosity occurring before the age of 18 years, therefore children should not be offered testing until they have turned 18 years of age.[2]

However, if Warren is anxious then testing his wife would be informative of their children's risk. If she does not carry the C282Y gene mutation – or the H63D gene mutation – all their children would be carriers and their risk of developing symptomatic HH is extremely small.

CASE 2

IS DEBORAH AT RISK FOR CLOTTING?

Deborah is 33 years of age and presents to you with a request for a repeat prescription for the combined oral contraceptive pill (COCP).

Deborah had sporadically used the COCP in her 20s, but stopped when she wanted to start a family a couple of years ago. Deborah was recently pregnant but had a fetal death in utero 2 months ago at about 35 weeks gestation (with an apparent unknown cause). She would like to conceive again but feels that it would be better to wait a few months.

On further questioning, Deborah says that she might have had a very early miscarriage a few years earlier but wasn't sure about her dates. Deborah has no past history of a deep venous thrombosis and is a nonsmoker. She informs you that her mother had a deep vein thrombosis after a long aeroplane flight when aged in her 40s.

On examination, Deborah has a blood pressure of 115/74 and a body mass index (BMI) of 22.3.

QUESTION 1

What features in Deborah's history act as a prompt for you to perform thrombophilia screening?

QUESTION 2

What other features in an individual's history might prompt a thrombophilia screen?

QUESTION 3

List four thrombophilic factors that are included in a thrombophilia screen.

QUESTION 4

Which of the thrombophilic factors you listed are strongly thrombophilic and which are weakly thrombophilic?

FURTHER INFORMATION

A thrombophilia screen shows that Deborah is heterozygous for the factor V Leiden mutation.

QUESTION 5

How does this finding influence the decision of whether to prescribe Deborah the COCP?

QUESTION 6

Would Deborah require thromboprophylaxis in her next pregnancy or in the postpartum period?

QUESTION 7

What are the implications for Deborah's first degree relatives regarding their risk of venous thromboembolism (VTE)?

CASE 2 ANSWERS

ANSWER 1

Features in Deborah's history which place her at risk of VTE, and which should prompt you to perform a thrombophilia screen, are:

- her pregnancy with an unexplained fetal death in utero in late gestation
- a family history of VTE as she has a first degree relative who experienced a first VTE before the age of 50 years.

ANSWER 2

In addition to the features mentioned for Deborah, other features that are indications for a thrombophilia screen are the occurrence of:

- spontaneous VTE in the absence of recognised risk factors
- VTE at an age less than 50 years
- recurrent (more than one) VTE
- venous thrombosis in unusual sites such as the central nervous system, abdominal veins or upper limb veins
- VTE during pregnancy or the puerperium
- VTE associated with the use of oestrogen containing contraception or hormone replacement therapy (HRT)
- recurrent (more than two) unexplained first trimester miscarriage, stillbirth, unexplained severe pre-eclampsia, placental abruption, or a fetus with severe intrauterine growth restriction.

ANSWER 3

A thrombophilia screen includes factors categorised into two groups of hereditary thrombophilic conditions:

- Group 1 conditions are caused by a defect or deficiency of an anticoagulant protein and include antithrombin deficiency, protein S deficiency and protein C deficiency
- Group 2 conditions are due to genetic mutations that result in an increased tendency towards thrombosis and include factor V Leiden (causes 90% of cases of activated protein C resistance), prothrombin gene variant and elevated levels of factors VIII, IX and XI (these are determined by clotting assays).

ANSWER 4

Group 1 conditions tend to be moderately to strongly thrombophilic leading to a greater risk of VTE, while those in Group 2 are weakly thrombophilic. However, Group 2 conditions are about five times more frequent than Group 1 conditions.

Some characteristics of Group 1 and Group 2 inherited thrombophilic conditions are shown in *Table 2*.

ANSWER 5

As Deborah has now been found to have a hereditary thrombophilia, she is at further risk of developing a VTE in the presence of acquired risks (*Table 3*). In particular, the COCP increases her risk by 2–4 fold, and is relatively contraindicated in women with a hereditary thrombophilia. Third generation COCPs are more thrombogenic than second generation COCP preparations. However, as Deborah is heterozygous for a weak thrombophilia, and in the absence of other risk factors such as smoking or obesity, her absolute risk of a VTE on the COCP is still low.

Decisions regarding use of the COCP should be made on an individual basis, after a risk-benefit analysis has been performed and in consultation with the patient. In addition to the remaining acquired risk factors other considerations are:

- suitability of alternative means of contraception
- patient preference
- patient compliance.

Advice should be sought from a specialist haematologist and/or family planning expert if required.

Table 3 lists acquired risk factors for thromboemobolism for individuals with a hereditary thrombophilia.

FEEDBACK

Screening all women for hereditary thrombophilias is not routinely recommended before commencing the COCP, HRT or tamoxifen (and other selective oestrogen receptor modulators). Routine screening for hereditary thrombophilias is also not recommended for patients with a personal or family history of

Table 2. Some characteristics of Group 1 and Group 2 inherited thrombophilic conditions[6]

Conditions and their testing	Heterozygote frequency in the general population	Prevalence in individuals with a VTE	Clinical state
Group 1 conditions			
Antithrombin deficiency (functional/antigenic assay)	1 in 500	1 in 20	• Severe thrombophilia • 60% of heterozygotes develop VTE by age 60 years • Homozygosity generally incompatible with life
Protein C deficiency (functional/antigenic assay)	1 in 500	1 in 25	• Moderate to severe thrombophilia • Up to 50% of heterozygotes develop VTE by age 60 years • Homozygotes develop severe thrombophilia: neonatal purpura fulminans; disseminated intravascular coagulation
Protein S deficiency (functional/antigenic assay)	3–13 in 1000	1 in 25	• Moderate thrombophilia • 30% of heterozygotes develop VTE by age 60 years • Homozygotes develop severe thrombophilia: neonatal purpura fulminans
Group 2 conditions			
Factor V Leiden mutation (DNA assay)	1 in 20*	3–5 in 10	• Mild thrombophilia • 6% of heterozygotes develop VTE by age 65 years • Homozygotes develop moderate thrombophilia
Prothrombin gene variant (DNA assay)	2–3 in 100*	1.5 in 10	• Mild thrombophilia • <5% of heterozygotes develop VTE by age 60 years • Homozygotes develop moderate thrombophilia

* In Caucasian populations. It is rare in Asian or African populations

Table 3. Acquired risk factors for thromboembolism for individuals with a hereditary thrombophilia[6]

Acquired risk factors
• Obesity
• Increasing age
• Prolonged immobilisation (>10 days)
• Surgery or trauma
• Pregnancy and the puerperium
• Smoking
• Combined oral contraceptive pill or hormone replacement therapy
• Active cancer
• Antiphospholipid antibodies
• Acquired activated protein C resistance

arterial thrombosis (acute coronary syndrome or stroke).[7,8]

With regard to factor V Leiden mutation, 20 000 women would need to be screened in order to prevent one DVT, while 2 million women require screening to prevent one death from pulmonary embolism. Instead of performing routine screening for hereditary thrombophilias before prescribing oestrogen containing preparations, practitioners should take careful past medical and family histories, including asking about additional risk factors for thrombosis. Screening can then be targeted to those women who appear to be most at risk of having a hereditary thrombophilia.[6]

ANSWER 6

Deborah may require postpartum thromboprophylaxis. Periods of increased thrombotic tendency due to a risk factor, such as pregnancy, immobilisation or surgery, temporarily increase the baseline risk and, together with age, should be taken into account in determining Deborah's need for thromboprophylaxis. Decisions regarding the management of hereditary thrombophilias in pregnancy should be made in consultation with a specialist haematologist, obstetric physician or obstetrician.

FEEDBACK

Primary prophylaxis for individuals with hereditary thrombophilia is not recommended, as the lifetime risk of death from bleeding on anticoagulants is greater than the risk of death from thrombosis in previously asymptomatic individuals.

ANSWER 7

Deborah should be encouraged to inform her first degree relatives that they may be at risk of inheriting the factor V Leiden mutation. Her relatives should consider discussing this risk with their own GP, who may also refer them to a haematologist.

Factor V Leiden (and prothrombin gene variant) genetic testing is available under Medicare to patients with:

- a personal history of DVT, or
- a family history of a diagnosed hereditary thrombophilia.

Testing for thrombophilias is a complex issue and should be guided by whether testing will inform management of the patient.[9]

CASE 3

SONIA AND GARY WISH TO CONCEIVE

Sonia is 24 years of age and recently migrated from Thailand with her partner Gary. Both Sonia and Gary are of Thai ancestry. She is nulliparous and came to see you 2 weeks ago because she is planning to get pregnant. She has no past medical history, is not taking any regular medications, is a nonsmoker and does not drink any alcohol. A PAP smear taken several months ago was normal. Physical examination, including her blood pressure, abdominal examination and BMI, was normal.

You advised her to commence folic acid supplements, administered a diphtheria/pertussis/tetanus vaccine and requested the following blood tests: FBE, iron studies, haemoglobin electrophoresis, rubella IgG and varicella zoster virus IgG.

Sonia's FBE shows her haemoglobin (Hb) to be 10.5g/dL with a microcytosis (67fl) and her Hb electrophoresis and iron studies are normal. Results also reveal that Sonia is immune to rubella and has antibodies to varicella zoster virus.

She comes to see you today with her partner Gary for the results of her blood tests.

QUESTION 1

What is the most likely cause of Sonia's microcytic anaemia?

QUESTION 2

What is the next most appropriate test?

FURTHER INFORMATION

The DNA studies for Sonia show that she is heterozygote for the two gene deletion alpha (α) thalassaemia (--/$\alpha\alpha$) mutation.

QUESTION 3

What is the next most appropriate step in management of this situation?

FURTHER INFORMATION

You also arrange investigations for Gary. The FBE demonstrates a normal haemoglobin concentration but a microcytosis at 78 fL. Iron studies and Hb electrophoresis are both normal.

QUESTION 4

Does the normal Hb electrophoresis result exclude α thalassaemia trait in Gary?

FURTHER INFORMATION

You request DNA studies, which show that Gary is heterozygote for single gene deletion α thalassaemia (-α/$\alpha\alpha$) mutation.

QUESTION 5

What are the implications for Sonia and Gary in planning their pregnancy?

QUESTION 6

If Gary had the two gene deletion α thalassaemia (--/αα) mutation, what would the implications be?

QUESTION 7

If Sonia and Gary were beta (β) thalassaemia heterozygotes, what would the implications be?

QUESTION 8

What can be offered to Sonia and Gary before they conceive?

CASE 3 ANSWERS

ANSWER 1

The most likely cause of Sonia's microcytic anaemia is α thalassaemia minor.

Normal haemoglobin A (HbA), which makes up to 98% of normal adult haemoglobin, is composed of two α and two β globin chains. The β globin genes are located on chromosome 11 while the α globin genes are located on chromosome 16. Importantly, there are two copies of the β globin gene (one on each chromosome) while there are four copies of the α globin genes (two on each chromosome).

The thalassaemia syndromes result from the reduced production of one or more of the globin chains of haemoglobin.[10] Reduced production of α chains leads to α thalassaemia; underproduction of β chains leads to β thalassaemia. The reduced production of a specific globin chain leads to an imbalance in the α and β chains. The unpaired chains precipitate in red blood cells.[11]

The two types of α thalassaemia trait are α^+ and α^0 trait. Alpha$^+$ trait refers to the deletion or a mutation in one of the paired α genes (eg. -α/αα). This carrier state may be associated with normal haematological parameters or a mild microcytosis.

Alpha^0 trait refers to the deletion or a mutation in both of the α globin genes (eg. --/αα). This carrier state is usually associated with a mild anaemia and microcytosis out of proportion to the degree of anaemia. The only implication for the carrier is the need for partner testing to define the risk of having a child with a more severe thalassaemia syndrome.

Table 4 lists the mean haematological parameters seen in various genotypes of α thalassaemia trait.

Table 4. Mean haematological parameters seen in various genotypes of α thalassaemia trait[12]

Genotype	Mean haemoglobin concentration (g/dL)	Range of haemoglobin concentration (g/dL) (normal range 13–18 g/d/L)	Mean corpuscular volume (fl) (normal range 78–98 fl)
-α/αα	Male: 14.3 Female:12.6	13–16.5 11–14	81.2
-α/-α	Male: 13.9 Female:12.0	12.0–15.5 10.5–13.0	71.6
--/αα	Male: 13.7 Female:12.1	12.5–15.0 10.5–13.0	69.1

ANSWER 2

The next most appropriate test is to arrange DNA studies for α thalassaemia in Sonia.

ANSWER 3

Testing for her partner Gary should be arranged, which should include FBE, iron studies, Hb electrophoresis and DNA studies.

ANSWER 4

A normal Hb electrophoresis result does not exclude thalassaemia. Alpha thalassaemia minor cannot be diagnosed on haemoglobin electrophoresis. Therefore DNA studies are required.

ANSWER 5

The risks to the child are: a 25% risk of HbH disease, a 50% chance of α thalassaemia trait, and 25% chance of being genotypically normal.

Figure 2 shows the expected pregnancy outcomes where one partner is a carrier of one gene mutation thalassaemia (α^+ thalassaemia trait) and one partner is a carrier of two gene mutation thalassaemia (α^0 thalassaemia trait).

HbH disease (--/-α) has a variable phenotype. Due to the lack of α globin chain production, tetramers of α globin chains develop (β^4 = HbH). These tetramers have a very high oxygen affinity and thus are functionally useless forms of haemoglobin. Further, they are relatively insoluble, ie. they behave as an unstable haemoglobin. This causes chronic haemolysis, which in addition to a degree of dyserythropoiesis (defective development of red blood cells), means that HbH disease can result in a mild to potentially severe chronic anaemia. Patients are usually not transfusion dependent, with haemoglobin concentrations in the range of 80–90 g/L.[14]

Patients usually develop splenomegaly and may develop gallstones – both a result of chronic haemolysis. Over time, patients may require more frequent blood transfusions and acute insults (eg. infections or exposure to oxidising drugs) may worsen the anaemia. Splenectomy is often a therapeutic option in those whose anaemia has become more severe, and in a proportion of cases this will improve the haemoglobin enough to obviate the need for regular transfusion. Iron overload, due blood transfusions and increased intestinal absorption, and skeletal changes may also be a problem in those with HbH disease.

ANSWER 6

If both Sonia and Gary were carriers of two gene deletion α thalassaemia (--/$\alpha\alpha$) then there would be a 25% chance that the pregnancy would be haemoglobin Barts hydrops fetalis, a 50% chance of α thalassaemia trait, and a 25% chance of it being genotypically normal.

Haemoglobin Barts (--/--) occurs where all four α globin genes are deleted or dysfunctional. This is very important as it has implications for both the fetus and mother. Haemoglobin Barts is composed of tetramers of γ globin chains (γ^4) and this represents the most severe form of α thalassaemia. The fetus is profoundly anaemic and this leads to heart failure with pleural effusions, ascites and hepatosplenomegaly. This usually results in stillbirth or death early in the postnatal period. Maternal complications may include severe pre-eclampsia and preterm labour.

Figure 3 shows the expected pregnancy outcomes where both partners are carriers of two gene deletion α thalassaemia (ie. α^0 thalassaemia trait).

Given there is no effective treatment for haemoglobin Barts hydrops fetalis, it is imperative that at risk couples be identified and referred for genetic counselling.

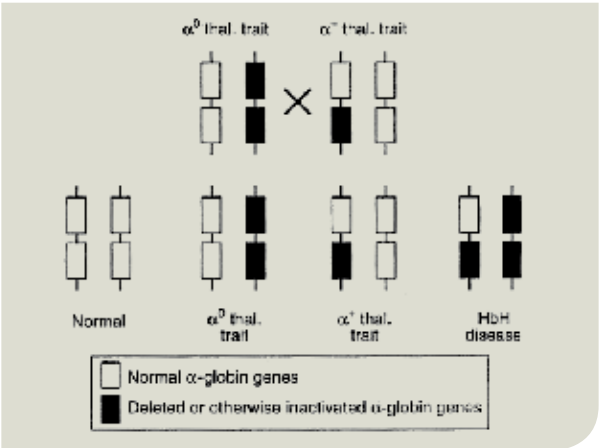

Figure 2. Expected pregnancy outcomes from α^+ and α^0 thalassaemia trait.

Reproduced with permission from Wiley–Blackwell[13]

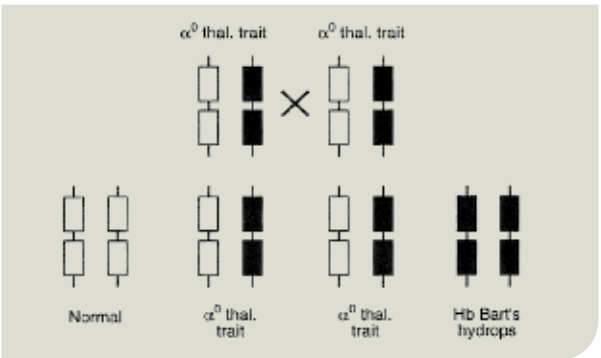

Figure 3. Expected pregnancy outcomes where both partners are carriers of two gene deletion α thalassaemia (ie. α^0 thalassaemia trait)

Reproduced with permission from Wiley-Blackwell[13]

ANSWER 7

If Sonia and Gary were β thalassaemia heterozygotes, there would be a 25% risk of β thalassaemia major.

Beta thalassaemia major is usually associated with a severe transfusion dependent anaemia. Therapy in the form of regular blood transfusions is directed at ameliorating the symptoms associated with severe anaemia. This also helps prevent some of the skeletal changes that can occur as a consequence of compensatory bone marrow expansion. However, iron overload occurs as a result of a regular chronic transfusion program. Iron loading tends to affect the anterior pituitary, liver, heart and pancreas.

Therefore, endocrinopathies, liver cirrhosis, cardiomyopathy and diabetes may all occur. Regular iron chelation therapy is required to prevent these complications.

ANSWER 8

Genetic counselling could be offered to explain the potential risks and reproductive choices including the role of prenatal diagnosis.

CASE 4

CHLOE'S FAMILY HISTORY OF CANCER

Chloe, 35 years of age, comes to see you for advice about breast cancer screening. She has no medical problems and is married with two young children. She is concerned about her risk of breast cancer because her mother recently developed breast cancer at 52 years of age.

You ask about additional family history. Her mother has no relatives with cancer. Chloe has two paternal aunts who were diagnosed with cancer; one aunt had breast cancer at 37 years of age and another aunt had ovarian cancer in her 40s. Chloe's paternal grandmother was diagnosed with breast cancer at 45 years of age.

QUESTION 1

What feature/s of the family history of cancer suggest/s that Chloe may be at increased risk of breast cancer?

QUESTION 2

What Australian risk assessment tools are available for GPs to assess family history of breast cancer and ovarian cancer?

QUESTION 3

Who can arrange genetic testing for an inherited predisposition to breast and ovarian cancer?

QUESTION 4

Which genes have a high risk of breast cancer (if an inherited mutation is present)?

QUESTION 5

What type of genetic test is offered first to investigate for an inherited predisposition to breast/ovarian cancer?

FURTHER INFORMATION

Chloe's paternal aunt was offered genetic testing and an inherited mutation in BRCA1 was identified.

QUESTION 6

What is a predictive genetic test?

QUESTION 7

What screening is recommended for women at high risk of breast cancer (for example, BRCA1 mutation carriers)?

QUESTION 8

What risk-reducing surgery may be considered for women at high risk of breast and ovarian cancer?

CASE 4 ANSWERS

ANSWER 1

The maternal and paternal family histories are both important to assess Chloe's risk of breast cancer. Each side of the family is assessed separately and is not added together to assess risk.

Chloe's mother was diagnosed with breast cancer after age 50 years – this maternal history of cancer does not suggest an inherited cause.

Her father's history of cancer is concerning, as there is a known inherited predisposition to both breast and ovarian cancer in some families. There are three women diagnosed with these cancers, and they were diagnosed at young ages.

ANSWER 2

Australian guidelines to assess the risk of breast and ovarian cancer based on family history have been developed by the National Breast and Ovarian Cancer Centre (NBOCC) and were revised in 2010. In addition, an online risk calculator is available for use by primary care physicians called FRA-BOC (see *Resources*). This includes questions about the family history of breast and/or ovarian cancer (and sometimes asks for age at diagnosis). Information known about genetic testing for the family can be included. Using FRA-BOC, Chloe is considered at potentially high risk of breast/ovarian cancer and referral to a family cancer clinic is advised.

ANSWER 3

Genetic testing needs to be arranged by a family cancer clinic. These clinics are located in most large Australian cities, and some centres provide outreach clinics to rural centres. A list of family cancer clinics in Australia can be found on the Centre for Genetics Education website (see *Resources*).

The family cancer clinic may use risk assessment tools such as BOADICEA (a computer program), BRCA-PRO (a statistical model with associated computer program) or the Manchester Score (an empirical scoring system) which may incorporate information about the pathology of cancers as well as the family history to determine whether genetic testing for BRCA1/BRCA2 carrier status is appropriate or not. Testing is offered following genetic counselling and results may clarify the risk of cancer for family members.

ANSWER 4

An inherited mutation in either BRCA1 or BRCA2 is present in around 5% of women diagnosed with breast cancer. These mutations are autosomal dominantly inherited. BRCA1 and BRCA2 mutation carriers have an increased risk of breast cancer (40 to 80% risk by age 70) and an increased risk of ovarian cancer (10 to 60% risk by age 70).[15] Men with an inherited mutation in BRCA1 or BRCA2 are at increased risk of breast cancer (up to 6% risk for BRCA2 carriers) and possibly at increased risk of prostate cancer.

Other rare syndromes (which have additional clinical features to suggest their diagnosis) associated with potentially high risk of breast cancer are: Li-Fraumeni syndrome (p53 gene), Cowden syndrome (PTEN gene), Peutz-Jeghers syndrome (STK11 gene) and hereditary diffuse gastric cancer (E-cadherin gene).

ANSWER 5

There are two different types of genetic tests. The genetic test offered first to a family is a mutation search, to determine whether an inherited mutation can be found within a known cancer predisposition gene. This process commences with one person who has usually had either breast or ovarian cancer in the family. This first genetic test, a mutation search, identifies a causative mutation in some families, but not all high risk families. If this first test does not identify a mutation, the result is considered inconclusive. In this situation, all at risk women are advised to have breast cancer screening and sometimes risk-reducing procedures (depending upon the family history).

The second type of genetic test is discussed in *Answer 6.*

ANSWER 6

Once an inherited mutation is found for a family, a predictive genetic test can be offered to at risk family members to determine whether they have inherited the family mutation or not.

Given that Chloe's paternal aunt has an inherited mutation in BRCA1, Chloe can be offered a predictive genetic test.

If Chloe has inherited the family mutation, then she will be advised about her options to reduce her risk of breast and ovarian cancer and options for screening.

If Chloe has not inherited the family mutation, she is at close to the population risk of breast and ovarian cancer and will be advised to follow population screening.

ANSWER 7

Annual breast cancer screening is advised, with digital mammography and/or magnetic resonance imaging (MRI). The age to commence screening depends on the youngest age of breast cancer in the family or commences by age 30 years. Breast MRI is funded by Medicare but requires a specialist to request the MRI on a machine with the correct equipment and licence.

MRI screening for high risk women has been shown to find breast cancer at an earlier stage than mammography.[16] There is no long term data available to assess whether this improves survival following early detection of breast cancer yet, but studies are ongoing.
There is no proven benefit for ovarian cancer screening with either transvaginal ultrasound or serum CA125 levels and no screening is advised for ovarian cancer (NBOCC).

ANSWER 8

Risk reducing mastectomy is an option for high risk women. This reduces the risk of developing breast cancer by at least 90%.[17]

Following completion of child bearing and around the age of 40 years, bilateral salpingo-oophorectomy (BSO) is advised. This reduces the risk of ovarian cancer by 98% and if performed at age 40, will also reduce the risk of breast cancer by 50%. Following surgery, HRT can be considered until age 50, as there is no evidence this increases the risk of breast cancer.[18,19] Performing BSO has been shown to reduce the all-cause mortality, breast cancer mortality and ovarian cancer mortality for BRCA1 and BRCA2 mutation carriers (Prevention and Observation of Surgical End Points [PROSE] study).[20]

CASE 5

JANE'S COMBINED FIRST TRIMESTER SCREENING

Jane, 31 years of age, is in her third pregnancy. She has a healthy 2 year old son and had a miscarriage in her first pregnancy at 9 weeks gestation. She has no significant past or family history of genetic abnormality.

She presents at 12 weeks in an uncomplicated pregnancy to discuss the results of her combined first trimester screening.

Ultrasound reveals a single fetus with no obvious structural anomalies but an enlarged nuchal translucency (NT) measurement of 3.6 mm (*Figure 4*) where the upper limit of the normal range at this gestation is 2.4 mm (*Figure 5*).

Serology performed at 11 weeks gestation shows a normal free β hCG level of 1.15 multiples of the median (MoM) but a low pregnancy associated plasma protein A (PAPP-A) level of 0.23 MoM.

The combined risk for trisomy 21 is 1 in 4, which is elevated from a maternal age related risk of 1 in 494. The risks for trisomy 18 (1 in 47) and trisomy 13 (1 in 46) are also elevated.

Figure 4 shows the fetal NT enlarged to 3.6 mm.

Figure 5 shows the normal range for NT measurement at 11–13^{+6} weeks gestation.

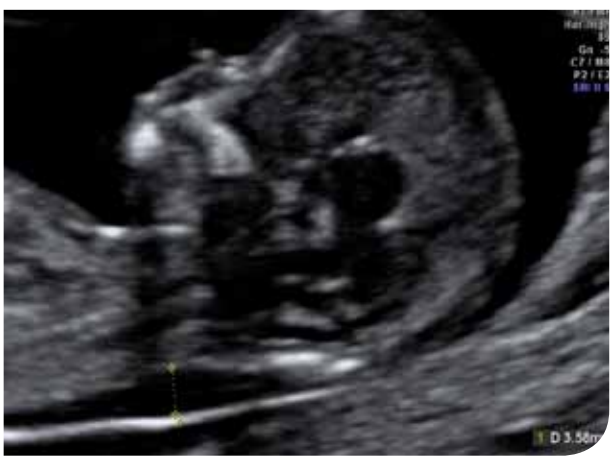

Figure 4. Jane's first trimester ultrasound

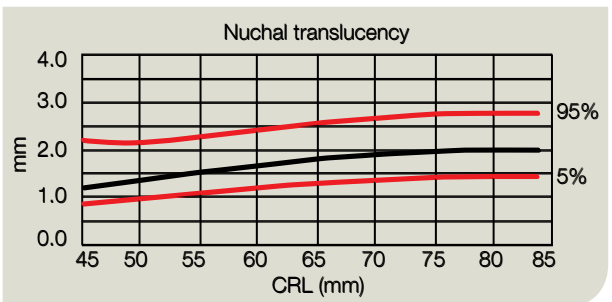

Figure 5. Normal range for NT measurement at 11–13^{+6} weeks gestation

QUESTION 1

Who should be offered prenatal aneuploidy screening?

QUESTION 2

What is your approach to counselling Jane about this result? What is the significance of these results?

QUESTION 3

What management options are available?

FURTHER INFORMATION

Jane decides to have CVS which is performed at 13 weeks and shows the karyotype of the fetus is 46 XX.

QUESTION 4

What are the associations with an enlarged NT measurement in a fetus with normal chromosomes?

QUESTION 5

What is the significance of a low PAPP-A level? How does Jane's PAPP-A level influence your management of her pregnancy?

FURTHER INFORMATION

Jane has a 20 week ultrasound at the fetal medicine unit of the nearest tertiary hospital. Fetal morphology and echocardiography assessment are found to be normal.

QUESTION 6

What is the prognosis for this child?

QUESTION 7

What is new in the field of prenatal screening and diagnosis of chromosomal abnormalities?

CASE 5 ANSWERS

ANSWER 1

Prenatal screening involves the use of maternal age, ultrasound and/or biochemical tests to identify 'at risk' individuals who may benefit from definitive prenatal diagnostic testing (by chorionic villus sampling or amniocentesis). It is appropriate to offer screening for Down syndrome to pregnant women of all ages.[21–23] Formal training in the performance of NT measurement and continuous operator audit are crucial to the accurate performance of this screening test.[24–27]

FEEDBACK

First trimester ultrasound has the additional benefit of providing accurate pregnancy dating, exclusion of unrecognised early pregnancy failure, accurate chorionicity assessment in twins and the detection of many severe fetal morphological anomalies (such as anencephaly).

A combination of patient characteristics, ultrasound and biochemical parameters at the 11–13 week scan has the potential to select out a small group of women at high risk for fetal aneuploidy, fetal structural abnormalities, miscarriage and stillbirth, preterm delivery, pre-eclampsia, gestational diabetes and fetal growth anomalies.[28]

ANSWER 2

Nondirective empathic counselling is vital. While the risk of aneuploidy is high due to the elevated NT measurement and the low PAPP-A level, counselling should focus on the positive aspects of the situation. The accuracy of the risk estimate has been confirmed in large prospective datasets, so in this case there is a 75% chance that the fetus does not have trisomy 21 and a 98% chance that it does not have trisomy 18 or 13. Other positive features are that the fetus appears structurally normal, appropriately grown and the β hCG level is normal.

ANSWER 3

The primary management issue is to discuss definitive prenatal testing, either personally or in conjunction with fetal medicine or genetic specialists. There is value in offering this to all patients with an increased risk as information regarding the karyotype provides relief from anxiety (in the majority of cases) or allows personal and family preparation for decisions regarding termination or continuance of the pregnancy.

Prenatal diagnostic testing is either by chorionic villus sampling (CVS) at 11–13 weeks gestation or by amniocentesis at 15–17 weeks. The only significant complication of these tests is a small increase in risk of miscarriage above the background rate of spontaneous pregnancy loss. The exact risk increase remains unclear as randomised controlled trials are ethically difficult, but the rate is known to be operator dependent. Uncomplicated transabdominal CVS has an additional miscarriage risk in the order of 1% and approximately 0.5% for amniocentesis.

The original fear of limb reduction abnormalities in association with CVS is unfounded when CVS is performed after 10 weeks gestation. Amniocentesis is technically possible prior to 15 weeks, but problems with culture failure, an increase in talipes and a higher miscarriage rate due to membrane rupture prevent its early use.

Other management options include:

- ultrasound review at 15–16 weeks to ensure fetal survival, appropriate growth and exclusion of hydrops fetalis
- formal fetal morphology assessment at 18–20 weeks to exclude structural anomalies that can be associated with enlarged NT measurements (see *Answer 4*)
- formal fetal echocardiography, and
- growth and welfare assessment in the second half of the pregnancy given the association of low PAPP-A and adverse pregnancy outcome (see *Answer 5*).

ANSWER 4

A large NT measurement is associated with a high risk for chromosomal abnormality. Other associations include spontaneous fetal loss, cardiac abnormalities and rare genetic syndromes.[29] Serial ultrasounds including assessing fetal morphology and fetal echocardiography should be performed. Even a subtle variation in fetal morphology should be taken seriously, as this may be the only sign of an underlying genetic syndrome. Postnatal echocardiography should also be performed.

Table 5 lists some possible associations with a karotypically normal fetus with a NT measurement of 3.5 mm or more.

ANSWER 5

Low levels of PAPP-A are associated with adverse outcomes in later pregnancy. The lower the PAPP-A level, the higher the incidence of associated spontaneous fetal demise, stillbirth, growth restriction, pregnancy induced hypertension and premature delivery.[30]

Low levels of PAPP-A are associated with a high risk for Down syndrome. The choice of subsequent prenatal diagnostic test may be influenced by the higher natural fetal loss rate, which is unrelated to the procedure related loss rate of CVS or amniocentesis. PAPP-A levels less than 0.3 MoM justify monitoring fetal growth and welfare in the second half of the pregnancy.

ANSWER 6

If the increased NT resolves and there is no structural anomaly, long term studies have shown no increased risk of adverse neonatal outcome or developmental delay compared with the normal paediatric population.[31] Caution is urged in the counselling of parents when the NT measurement exceeds 6.5 mm, as there may be a poorer outcome in this group despite an apparently normal mid-trimester scan.

Table 5. Possible associations with a karyotypically normal fetus with an NT measurement of 3.5 mm or more

Miscarriage/fetal death	Risk 2.7% for NT 3.5–4.4 mm Risk 19% for NT >6.5 mm
Major fetal anomaly • Cardiac anomalies (1 in 16 overall) • Diaphragmatic hernia • Exomphalos • Body stalk anomaly • Thoracic mass • Skeletal anomalies	Risk 10% for NT 3.5–4.4 mm Risk 46% for NT >6.5 mm
Genetic syndromes • Fetal akinesia deformation sequence • Noonan syndrome • Smith-Lemli-Opitz syndrome • Type 1 spinal muscular atrophy • Congenital adrenal hyperplasia	Risk 1.4% for NT 3.5–4.4 mm Risk 9.4% for NT >6.5 mm

ANSWER 7

The following tests are new in the field of prenatal screening and diagnosis of chromosomal abnormalities.

New first trimester ultrasound markers

Several novel first trimester ultrasound features have been proposed in an effort to improve trisomy 21 detection but, more importantly, to lower the false-positive rate of combined screening, which will expose fewer women to unnecessary invasive diagnostic tests. Assessment of the nasal bone is the best studied and is currently recommended for screening in Australia.

The nasal bone is known to be 'absent' in about 60% of Down syndrome fetuses and in 1–2% of euploid fetuses. Several large prospective studies have shown that adding nasal bone assessment to combined first trimester screening maintains a 90% Down syndrome detection rate but lowers the false positive rate to 2–3%. There are other means for assessing Down syndrome risk at the 12-week scan such as Doppler interrogation of the tricuspid valve (regurgitation in 55% of trisomy 21 fetuses) and the ductus venosus (absent or reversed a-wave in 65%).[32–34]

Fetal DNA in maternal blood

Fetal DNA can be found in maternal plasma. DNA sequences that are unique to the fetus can potentially be amplified and quantified. However, gene dosage variation is currently too nonspecific to clinically test for chromosome abnormality in maternal serum.

Microarray comparative genomic hybridisation (aCGH)

This exciting development in molecular genetics enables prenatal diagnosis of aneuploidy as well as microduplications and microdeletions that cannot currently be diagnosed by routine karyotyping. It is being applied to amniocentesis and CVS samples in research settings to identify more than 120 rare conditions such as Prader-Willi syndrome.

CASE 6

SARAH PRESENTS WITH A PERSISTENT COUGH

Sarah, 4 years of age, is brought in to see you by her mother. Sarah has a chronic productive sounding cough which has been present for about 12 months but has been increasing in frequency and severity over that time. She is often wheezy, particularly with viral respiratory infections, but has not responded to salbutamol administered via a spacer, and there is no family history of asthma. Her parents complain that she often has frequent, offensive smelling stools but they have not been concerned about her nutrition.

Sarah was born in Victoria and her parents did not hear anything about the results of her routine newborn screening test. However, Sarah's first cousin was identified as a cystic fibrosis (CF) carrier following newborn screening. Sarah's mother did not hear the results of any further genetic testing in that family. Sarah's mother was not offered carrier screening prior to, or during, her pregnancy with Sarah.

QUESTION 1

What are the clinical features that suggest CF in Sarah? What should be asked about on further history taking?

QUESTION 2

What does newborn screening for CF specifically test for and what further testing is performed to make a definitive diagnosis of CF?

QUESTION 3

What is the inheritance of CF?

QUESTION 4

Are all carriers for CF gene mutations healthy or can they have a minor variant of CF?

QUESTION 5

How would you make a diagnosis of CF in Sarah? What further investigations would you request?

QUESTION 6

What else should be done to manage Sarah's presenting complaint?

QUESTION 7

It may seem sensible to immediately order a CFTR gene test, but what are the limitations of this test for Sarah?

QUESTION 8

There are a number of genetic counselling issues in this case. Please discuss these.

QUESTION 9

Sarah's mother has not been offered carrier screening. What are the possibilities for CF carrier screening in Australia?

CASE 6 ANSWERS

ANSWER 1

Sarah has the following two clinical features that suggest CF:

- a prolonged productive cough that suggests suppurative bronchitis
- frequent offensive stools.

Other features to ask about include:

- features that can be associated with asthma such as atopic eczema and hay fever. It is common for children who present with suppurative bronchitis to be wheezy. In the absence of risk factors for asthma and in the absence of response to a bronchodilator, it is likely that the wheeze is secondary to airways inflamed by infection and obstructed by infected mucus. It is common for patients with suppurative bronchitis to be mislabelled with asthma and the history of productive cough is very important to separate the two conditions
- recurrent infections such as otitis media and skin infections which, in combination with suppurative bronchitis, suggest immunodeficiency
- purulent rhinitis and suppurative otitis media which, in combination with suppurative bronchitis, dextrocardia and

neonatal distress, suggest primary ciliary dyskinesia

- neonatal meconium obstruction, prolonged neonatal jaundice, rectal prolapse and nasal polyps, which suggest CF
- other features of the stool, such as paleness and floating in the toilet bowl, that suggest steatorrhoea
- failure to thrive (by growth percentiles), which can occur in CF. The absence of recorded failure to thrive by growth percentiles makes it less likely that the stool history is that of steatorrhoea but formal testing of the stool is required.

A combination of suppurative bronchitis and pancreatic exocrine insufficiency makes CF the most likely clinical diagnosis. CF has an incidence of 1 in 2500 and is much more common than these other causes of suppurative bronchitis.

Most (85–90%) CF patients have pancreatic exocrine insufficiency. However, the remaining 10–15% of CF patients are pancreatic sufficient and this group is much more likely to present in childhood or adolescence.

ANSWER 2

Newborn screening for CF is now universal in Australia (New South Wales was the first state to commence screening in 1981 and Western Australia was the last to do so, in 2002).[35] All infants have a heel-prick taken on day 2–4 of life. For CF, serum trypsinogen by immunoreactive assay (IRT) is measured. Most infants with CF have elevated levels of trypsinogen in the blood. Infants with the highest level of trypsinogen (top 1% of results) have cystic fibrosis transmembrane conductance regulator (CFTR) gene mutation analysis performed from the day 2–4 blood spot. The CFTR gene encodes for an epithelial ion transport protein.

Laboratories around Australia test for a different number of gene mutations, depending on the frequency of those in the community and costs to the laboratory. Infants may have one of the following results on gene mutation analysis:

- two identified CFTR mutations – these infants have CF and should be referred to a CF clinic
- one identified CFTR mutation – these infants will either be healthy carriers or have CF, as the absence of an identified mutation on the second allele is not proof that that allele is normal. All infants with one CFTR gene mutation identified on screening require a sweat chloride test. Infants may have one of the following results on sweat chloride testing:
  - an elevated sweat chloride (>60 mmol/L) – these infants have CF and there must be a second mutation which may be discovered on further testing
  - borderline (or indeterminate) range of sweat chloride (30–59 mmol/L) – these infants require further testing
  - normal range sweat chloride (<30 mmol/L) – these infants are healthy carriers
- no identified gene mutations – infants who have an elevated IRT but no identified gene mutations are at low risk of having CF and their parents are not notified.

Infants with CF can be missed by newborn screening by not having an initial elevated IRT or they may not have an identified CFTR mutation. Most infants missed by screening who have pancreatic exocrine insufficiency present in the first 6 months of life with failure to thrive. Infants with CF who are pancreatic sufficient often do not present until childhood or adolescence.

ANSWER 3

CF is an autosomal recessive condition. People with CF have two mutations in the CFTR gene. A CFTR gene mutation is inherited from each parent and there are no new (de novo) mutations.

There are over 1500 CFTR gene mutations known to be associated with CF and over 200 polymorphisms of uncertain significance. The commonest CFTR gene mutation is called p.F508del (formerly known as ΔF508) which represents 70% of CF alleles. This is most common in the northern European and Caucasian populations. The next most common mutation represents 4–5% of mutations; other mutations are less frequent. A 12 mutation panel will cover 84% of mutations in the Australian population. There is diminishing return when testing for an increasing number of mutations as most are extremely rare.

ANSWER 4

Carriers of one CFTR gene mutation (like any carrier of an autosomal recessive condition) are always healthy.

ANSWER 5

The diagnosis of CF is based on clinical features (including the newborn screening result) with evidence of CFTR gene mutations and abnormal sweat chloride results.[36]

The clinical features of CF were explored further in *Question 1.* It would be very helpful to know whether Sarah has pancreatic exocrine insufficiency. Evidence of fat maldigestion can be sought by stool fat analysis looking for fat globules representing undigested fat (as opposed to fat crystals which represent digested but unabsorbed fat) or faecal elastase (low or absent in people with pancreatic exocrine insufficiency).

Sputum microbiology can sometimes be helpful for diagnosis and certainly to guide treatment. Children with idiopathic suppurative bronchitis commonly have no organisms cultured, but may have *Haemophilus influenzae* (nontypeable), *Staphylococcus aureus* or *Moraxella catarrhalis*. Identification of *Pseudomonas aeruginosa* is rare in children without CF.

To confirm the diagnosis of CF, the sweat chloride test is the most appropriate test to order next. This should be done at a laboratory familiar with performing sweat chloride tests, usually in a large children's hospital attached to a CF clinic. The values of sweat chloride in most children with CF will be >60 mmol/L. A handful of children have indeterminate sweat chloride results (1–2% of tests) and some of these children have clinical variants of CF with mutations that confer some activity of the chloride channel. This is best determined by a CF physician familiar with the diagnosis of CF.

ANSWER 6

Sarah should have a sputum test (microscopy, culture and sensitivities) to guide antibiotic therapy. The common bacteria in patients with suppurative bronchitis are discussed in *Answer 5*. Amoxycillin/clavulanate is a good antibiotic for treatment of these bacteria while waiting for a result. Children with suppurative bronchitis and certainly those with CF require prolonged courses of antibiotics and usually more than a week is required. An airway clearance regimen (chest physiotherapy) can help to clear infected secretions.

A chest X-ray to look for bronchiectasis would be advisable. Spirometry (in children older than 6 years of age) to measure lung function can be helpful; patients with suppuratives bronchitis often demonstrate an obstructive pattern, without bronchodilator response.

ANSWER 7

Most laboratories testing for CFTR gene mutations in Australia will only test for a maximum 12 mutations covering up to 84% of the possible gene mutations. A negative mutation result may still leave some uncertainty. Discovery of a single CFTR gene mutation leaves the dilemma as to whether Sarah has CF with an unidentified second mutation, or is truly a carrier (one mutation) with bronchitis. There is no advantage to Sarah, at 4 years of age, to know whether she is a carrier, and testing now removes her option of making that decision herself as an adult. The key piece of information is whether Sarah has CF or not, and this can be answered by a sweat chloride test.

ANSWER 8

Sarah's family history is interesting given her mother's sister (Sarah's aunt) had a baby identified as a carrier by newborn screening. Identification of carriers by newborn screening is an unintended consequence of the protocol that uses IRT followed by CFTR gene mutation analysis. Infants identified as carriers with one gene mutation and a normal sweat chloride test should have family genetic counselling. In most situations, one parent is a carrier but it is possible that both parents are carriers and on this occasion only one mutation was passed on. In this particular case scenario we don't have information as to whether Sarah's aunt was identified as a carrier.

The usual practice for most genetic counselling services in Australia would be to offer cascade family testing. The practical difficulty for cascade family testing is that it is the responsibility of Sarah's aunt to inform her sister of a genetic risk. Beyond the parents of the identified carrier (or infant affected with CF) few family members take up the offer of cascade family testing. This can be explained by a combination of the wider family members not being informed or being informed and not perceiving their increased risk, or simply being at a different life stage for which testing is irrelevant (eg. have completed their family or not ready to start a family). Most families are quite anxious around the time of identification of the carrier infant, although this is generally short lived.

ANSWER 9

Community based CF carrier screening is available in Victoria, New South Wales and Queensland, through John Hunter Hospital Genetics, Sydney IVF and Queensland Fertility Services.[37] The program in Victoria is a fee-for-service (current cost $220) and is offered by a cheek swab through obstetricians and shared care GPs.

Pre-test information is available from the doctor, by brochure and via the Victorian Clinical Genetics Services (see *Resources*). The cheek swab can be mailed to the laboratory and the results faxed to the referring doctor. Most genetic counselling for identified carriers can be done by telephone with partners of carriers being strongly recommended to have CF carrier testing. Patients and doctors wishing to request CF carrier screening living in states other than Victoria can do so through the above services, although genetic counselling when carrier couples are identified would need to proceed through local genetic services.

Carrier screening for CF offers couples choice prior to having a baby with CF. Carrier screening can be performed prior to pregnancy (pre-conception) or in the early stages of pregnancy. Carrier screening performed prior to pregnancy offers the greatest range of reproductive options, including IVF and pre-implantation genetic diagnosis. Carrier screening performed in the early stages of pregnancy, ideally before 14 weeks gestation, allows time for partner testing and counselling about options.

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PROFESSIONAL RESOURCES

General

- The Genetics Education in Medicine Consortium has produced a publication called *Genetics in Family Medicine: The Australian Handbook for General Practitioners*. This publication has information for both doctors and patients on a range of genetic conditions including each of those covered in this unit of *check* as well as contact details of genetic services throughout Australia. Available at www.nhmrc.gov.au/your-health/egenetics/health-practitioners/genetics-family-medicine-australian-handbook-general-prac

Cancer

- Information on familial cancer services, specific cancers and evidence for screening is produced by the Australian Cancer Network and NHMRC. *Clinical practice guidelines. Familial aspects of cancer: a guide to clinical practice*. This is currently under review. Available at www.nhmrc.gov.au/_files_nhmrc/publications/attachments/cp67.pdf
- The National Breast and Ovarian Cancer Centre website provides information on familial aspects of breast cancer and ovarian cancer and provides a unique breast cancer risk calculator. Available at www.nbocc.org.au

Cystic fibrosis

- Report from the Australasian Association of Clinical Biochemists Sweat Testing Working Party. Australian guidelines for the performance of the sweat test for the diagnosis of cystic fibrosis. Clin Biochem Rev 2006;(Suppl 1). Available at www.aacb.asn.au/admin/?getfile=700

Haemochromatosis

- Royal College of Pathologists of Australasia. Flow chart for managing iron overload. Available at www.rcpamanual.edu.au/staging_area/index.php?option=com_pdst&task=show_pdst&id=71&Itemid=103

Prenatal screening

- The Victorian Clinical Genetics Service provides information on maternal serum screening for doctors. Available at www.vcgspathology.com.au/downloads/mss/MaternalSerumScreening.pdf.

RESOURCES FOR PATIENTS

General

- Centre for Genetics Education, NSW Health has a range of fact sheets with an overview of genetic conditions, genetic counselling and specific genetic conditions, including each of those covered in this unit of *check*, as well as a family history questionnaire and a list of familial cancer services throughout Australia. Available at www.genetics.edu.au
- The Australasian Genetic Alliance provides support for individuals affected by genetic conditions. Available at www.australasiangeneticalliance.org.au/home

Cystic fibrosis

- The Victorian Clinical Genetics Services provides information on screening for carriage of cystic fibrosis genes. Available at www.cfscreening.com.au
- Cystic Fibrosis Australia provides clinical information and information about events and resources. Available at www.cysticfibrosis.org.au
- Cystic Fibrosis Medicine provides clinical information. Available at www.cysticfibrosismedicine.com

Haemochromatosis

- Haemochromatosis Society Australia. Telephone 1300 019 028. Available at www.haemochromatosis.org.au

Prenatal screening

- The Victorian Clinical Genetics Service provides information on maternal serum screening for patients in a range of languages. Available at www.vcgspathology.com.au

Thalassaemia

- Thalassaemia Australia. Available at www.thalassaemia.org.au.

Genetic disorders

In order to qualify for 6 Category 2 points for the QI&CPD activity associated with this unit:

- read and complete the unit of *check* in hardcopy or online at the *gplearning* website at www.gplearning.com.au, and
- log onto the *gplearning* website at www.gplearning.com.au and answer the following 10 multiple choice questions (MCQs) online
- complete the online evaluation.

If you are not an RACGP member, please contact the *gplearning* helpdesk on 1800 284 789 to register in the first instance. You will be provided with a username and password that will allow you access to the test.

The expected time to complete this activity is 3 hours.

Do not send answers to the MCQs into the *check* office. This activity can only be completed online at www.gplearning.com.au.

If you have any queries or technical issues accessing the test online, please contact the *gplearning* helpdesk on 1800 284 789.

QUESTION 1

Maikel is 45 years of age and presents with 3 months of tiredness. He has no relevant family history. Investigations reveal an elevated transferrin and serum ferritin and you consider the causes of such abnormalities including a diagnosis of hereditary haemochromatosis. Which of the following is necessary in order to request a genetic test for hereditary haemochromatosis under Medicare?

A. Abnormal fasting iron studies
B. Abnormal fasting iron studies on more than one occasion
C. Abnormal liver function tests
D. Abnormal ultrasound of the liver
E. Abnormal Ferriscan® (MRI of the liver).

QUESTION 2

Thomas is 32 years of age and is found to be a C282Y homozygote for hereditary haemochromatosis. His wife is a carrier for the same gene mutation. Thomas is concerned about the risk to his unborn children. Which of the following estimates the risk to his unborn children with respect to hereditary haemochromatosis due to the C282Y gene mutation?

A. There is a 25% chance of them being carriers
B. There is a 25% chance of them being predisposed to hereditary haemochromatosis
C. There is a 50% chance of them being predisposed to hereditary haemochomatosis
D. There is a 50% chance of them being genotypically normal
E. There is a 75% chance of them either being carriers or being predisposed to hereditary haemochromatosis.

QUESTION 3

After seeing a patient with thromboembolism in your practice, a medical student who observed the consultation asks you to consider scenarios in which you would perform a thrombophilia screen. All of the following are indications for performing a thrombophilia screen **except**:

A. venous thromboembolism occurring while taking the combined oral contraceptive pill
B. venous thromboembolism occurring following an open cholecystectomy
C. venous thromboembolism occurring in the setting of pre-eclampsia at 26 weeks gestation
D. venous thromboembolism at 48 years of age
E. venous thrombosis of the cavernous sinus.

QUESTION 4

Vo Binh Lo is 27 years of age and is planning a pregnancy. Investigations reveal a mild microcytosis with no evidence of anaemia. You tell her that:

A. if she has no evidence of anaemia, she does not have any form of thalassaemia
B. if her iron studies reveal iron deficiency, investigations for thalassaemia are not necessary
C. if her haemoglobin electrophoresis is normal, this excludes thalassaemia
D. if her DNA studies for thalassaemia are normal, iron deficiency is likely to be the cause for her microcytosis
E. if her DNA studies and those of her partner both reveal α^0 thalassaemia trait (two gene deletion or mutation thalassaemia), there is a 25% chance of their pregnancy having HbH disease.

QUESTION 5

Sandra is 34 years of age and asks you about her risk of breast cancer based on her family history. There are multiple cases of breast cancer in her family and the family are waiting to be seen by a family cancer clinic. Which of the following applies to calculation of risk of breast cancer based on family history?

A. Maternal and paternal sides of the family history are added together to assess risk
B. Diagnosis of breast cancer in a first degree relative at 55 years of age suggests an inherited gene mutation is present
C. A mutation search allows for identification of a specific gene mutation in asymptomatic individuals at high risk of breast cancer based on family history
D. A predictive genetic test can be performed on those affected by breast cancer
E. Given the inheritance of the BRCA mutation, most generations will have at least one individual carrying the gene.

QUESTION 6

Samantha is 38 years of age and has a family history of breast and ovarian cancer. Her mother died at the age of 40 from breast cancer and her aunt died from ovarian cancer at age 36 years. You refer her to a family cancer clinic where testing reveals that she carries the mutation for the BRCA1 gene. You advise her that:

A. screening breast MRI has been shown to improve survival in high risk women

B. you can request Medicare funded screening with MRIs in addition to mammograms

C. bilateral salpingo-oophorectomy (BSO) performed at age 40 years reduces the risk of breast cancer by 98%

D. bilateral mastectomies reduce the risk of developing breast cancer by at least 90%

E. hormone replacement therapy following BSO used until the age of 50 has been shown to increase the risk of breast cancer.

QUESTION 7

Michelle is 32 years of age and is 10 weeks pregnant. She asks you about screening and testing for Down syndrome. Which of the following is true of prenatal screening and prenatal diagnostic testing in general?

A. Prenatal screening for Down syndrome should be targeted towards those most at risk

B. Prenatal screening for Down syndrome should be offered to pregnant women of all ages

C. Prenatal diagnostic testing for Down syndrome should be offered to all pregnant women

D. Prenatal diagnostic testing should not be offered to all women at increased risk

E. Prenatal screening for Down syndrome revealing a nuchal translucency in the normal range on ultrasound at 11–13^{+6} weeks excludes Down syndrome.

QUESTION 8

Samneth is 36 years of age and has a high risk combined first trimester screening result. Which of the following is true regarding subsequent definitive testing for aneuploidy (abnormal number of chromosomes)?

A. Amniocentesis is usually performed at a minimum gestation of 15 weeks, whereas CVS is performed at a minimum gestation of 11 weeks

B. Amniocentesis is performed transvaginally whereas CVS cannot be performed transvaginally

C. Amniocentesis has a miscarriage rate of around 1% above the background rate and transabdominal CVS has a miscarriage rate of around 0.5% above the background rate

D. Amniocentesis allows for termination via surgical means, whereas an abnormality detected by CVS does not usually allow for termination in this way

E. Amniocentesis must be performed in hospital whereas CVS does not need to be performed in hospital.

QUESTION 9

Ashley is 9 years of age and is a new patient who presents with 6 months of a productive cough on a background of frequent respiratory infections throughout his life. You consider that he may have cystic fibrosis (CF). Which of the following is true with respects to diagnosing CF in general?

A. Pancreatic exocrine insufficiency is necessary for a diagnosis of CF

B. An elevated serum trypsinogen on newborn screening is necessary for a diagnosis of CF

C. An elevated or indeterminate sweat chloride result is necessary for the diagnosis of CF

D. Two identified CFTR mutations are necessary for a diagnosis of CF

E. A family history of CF is necessary for the diagnosis of CF.

QUESTION 10

You read an article in a journal about inheritance of genetic diseases. Which of the following conditions is inherited in an autosomal dominant fashion?

A. CF

B. Down syndrome

C. Haemochromatosis

D. Inherited mutations in the BRCA1 and BRCA2 genes

E. Thalassaemia.