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Common Sense Pathology

A REGULAR CASE-BASED SERIES ON PRACTICAL PATHOLOGY FOR GPs

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Antenatal screening for Down syndrome

A JOINT INITIATIVE OF



Australian
Doctor.

Antenatal screening for Down syndrome



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Introduction

Down syndrome (Trisomy 21) is one of the most common birth defects in Australia and the most common cause of intellectual disability. The proportion of pregnancies involving women over the age of 35 has increased from about 5% to 20% over the past 20 years, and for this reason Down syndrome is one of the few congenital defects with an increasing prevalence. The previously quoted incidence of 1:750 has now increased to more than 1:500.

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Diagnostic antenatal chromosome testing for Down syndrome has been available by amniocentesis and chorionic villus sampling for decades. However, these invasive procedures were generally only offered to those women at high risk according to their older age, where the likelihood of chromosomal abnormalities was more than the risk of the procedure itself causing miscarriage.

At present, there are two recommended screening tests for Down syndrome that have better diagnostic reliability than relying on age alone:

- Combined (ultrasound and serum) first-trimester screening.
- Second-trimester serum screening.

These procedures have improved antenatal diagnosis.

Twenty years ago, the majority of Down syndrome pregnancies were diagnosed at birth but, due to screening, this has continued to fall to about one-quarter of all diagnoses today. The net consequence of the increasing proportion of older mothers and the lower proportion of affected pregnancies continuing to birth is that the birth rate for Down syndrome is only slightly less than it was 20 years ago.

This article discusses the value and application of antenatal screening for Down syndrome.

Case study 1

AS is a 43-year-old professional woman who has finally been successful in becoming pregnant after trying to conceive for three years. She is concerned about her age-related risk of Down syndrome and would like advice on the testing available. Her last normal menstrual period was two months ago.

What advice should you offer?

This woman has a high risk of Down syndrome because of her age. Once a woman is over the age of 35, her risk of a Down syndrome pregnancy is 1:300 and by the age of 43 her risk increases to about 1:30 (figure 1). Any ethnic variation affecting this age-related risk is relatively insignificant.

Although many women in Australia want to know if they have an affected pregnancy because they would consider termination, this should certainly not be assumed. Women may choose not to

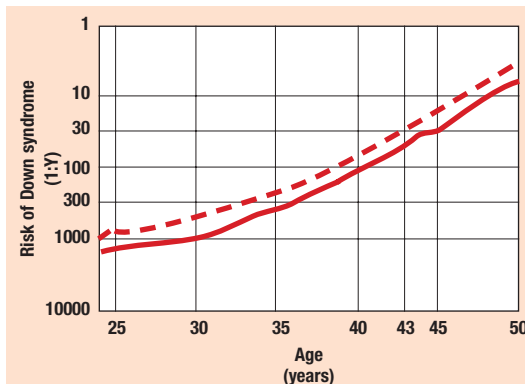


Figure 1. Age-related risk of Down syndrome. The broken line is overall risk. The solid line is a slightly lower risk at 12 weeks when screening tests are performed because some pregnancies will have aborted spontaneously by then.

test or terminate because of religious or moral beliefs. A woman may wish to know if she is having an affected child so that she can be prepared for that challenge. This can be an even more difficult decision for an older woman who may not be able to conceive again.

Fortunately, AS has presented early enough to seek counselling while she considers her options.

Clinically, the situation is much clearer. Offering antenatal testing is recommended for high-risk women over the age of 35 by the RACGP, the Human Genetics Society of Australasia (HGSA) and the Royal Australian and New Zealand College of Obstetricians and Gynaecologists (RANZCOG).

What tests should you offer?

Clearly as her age-related risk is already high (1:30), she should be immediately offered the diagnostic test. Diagnosis requires chromosomal analysis and cells for this test can be obtained by chorionic villus sampling (at 12-14 weeks) or amniocentesis (at 15-18 weeks). The risk of pregnancy loss with chorionic villus sampling is considered to be 1% whereas the risk with amniocentesis is lower at 0.5%. As many of these procedures are now performed by specialist obstetricians with modern ultrasound guidance and smaller needles, loss rates may be much lower than previously quoted. Test results can take a



couple of weeks but provisional results can be available much earlier using expensive fluorescence in-situ hybridisation (FISH) analysis or, in some centres, quantitative fluorescence polymerase chain reaction (QF-PCR).

Offering antenatal screening for Down syndrome in a 43-year-old woman such as AS is quite likely to result in a continued assessment of high risk. About half of pregnant women over the age of 40 will remain at high risk (>1:300) after screening tests and a large majority of women over the age of 45 will similarly remain at high risk.

Case study conclusion

This woman was reluctant to risk this pregnancy with invasive diagnostic procedures and opted for combined first-trimester screening with ultrasound and serum screening. The results of these screening tests showed that her risk had increased from 1:33 to 1:2 and she proceeded to amniocentesis which confirmed Trisomy 21 (47XY +21; figure 2).

Case study 2

KC is a 30-year-old woman who is expecting her third child and, although she wasn't aware of screening for Down syndrome in previous pregnancies, she is asking if you would recommend screening in this pregnancy.

Would you offer screening for Down syndrome?

Knowledge of Down syndrome can be variable in both patients and health care providers and about one-third of women can miss being offered maternal serum screening. At the age of 30, the risk of a Down syndrome pregnancy is 1:1000 compared with the high risk of fetal loss with invasive diagnostic testing, which is 1:100 for chorionic villus sampling and 1:200 for amniocentesis. Therefore, diagnostic testing at this age would not be promoted as first-line investigation because of the higher risk of fetal loss compared with the risk of an affected child.

About one-third of Down syndrome births in Australia occur in women under the age of 35 and, although this is a lower proportion than in the past, public health policy should be aimed at

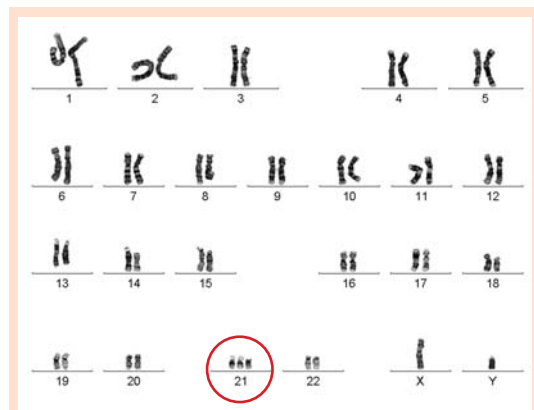


Figure 2. Karyotype obtained after amniocentesis of case study 1 indicating trisomy of chromosome 21 (Down syndrome).

offering antenatal screening to all pregnant women regardless of age.

The 3Centres Collaboration produced consensus guidelines in December 2001 that recommended written information about Down syndrome screening should be available to all pregnant women (level II evidence). There was also level III evidence that the offer of Down syndrome screening should be made available to all pregnant women, irrespective of age.

More recently in July 2007, RANZCOG issued a joint statement with the HGSA that stated: "All pregnant women should be advised of the availability of prenatal screening as early as possible in pregnancy to allow time to discuss the options available and facilitate an informed choice."

What screening tests could be offered?

Combined first-trimester screening

The screening test of choice is combined first-trimester screening. Individually first-trimester ultrasound of the nuchal translucency (measurement of the translucent area between the fetal skin and the underlying tissue at the nape of the neck) has a sensitivity of 60-80% and first-trimester serum screening has a sensitivity of 60-65%. However, in combination they can detect 90% of Down syndrome pregnancies (90% sensitivity), while maintaining 95% specificity (ie, only 5% of normal pregnancies will give a positive screen).

Prediction algorithm

The prediction algorithm is influenced by several factors and it is important that the following information is provided:

- Maternal weight (extremes affect risk).
- Any previous history of Down syndrome pregnancies.
- If the current pregnancy is multiple.
- If the current pregnancy was achieved by assisted reproduction techniques.

The optimum period for testing differs for serum testing (at 9-12 weeks) and ultrasound (at 11 weeks to 13 weeks and six days). One-stop clinics for the assessment of risk that do both tests at the time of ultrasound are convenient, and sometimes necessary, but may result in some loss of diagnostic sensitivity.

The two integral components of combined first-trimester screening are described below. These are rarely applied alone because of their individually lower detection rates.

Nuchal translucency

The nuchal translucency is measured by ultrasound in the first trimester (figure 3). It is one of the strongest indicators of Down syndrome, being present in more than 60% of Down syndrome fetuses at 11 weeks to 13 weeks and six days' gestation.

First-trimester ultrasound also assists in the diagnosis of other fetal abnormalities (eg, anencephaly) and, importantly, confirms gestational age through the measurement of crown-rump length. Early diagnosis of multiple gestation is also assisted during this period when the characteristics of the amnion and chorion are best discerned. This is important because a multiple gestation sharing the same amniotic sac or placenta affects maternal levels of placental proteins. Differentiation between cystic hygroma and enlarged simple nuchal translucency ($\geq 5.0\text{mm}$) is a moot point as a true cystic hygroma is rarely seen in the first trimester but both are sufficiently high-risk situations to warrant immediate chorionic villus sampling.

As the nuchal translucency measurement is critical to the calculation of risk, it should be performed by properly trained ultrasound operators. In



Figure 3a. Nuchal translucency (arrow) is the measurement of the translucent area between the fetal skin and the underlying tissue at the nape of the neck.

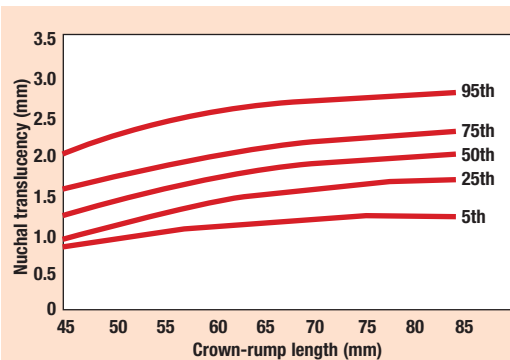


Figure 3b. Normal variation of nuchal translucency against crown-rump length (fetal age).

Source for figures 3a and b: Nicolaides K, et al. *The 11-14 Week Scan: The Diagnosis of Fetal Abnormalities*. Parthenon Publishing Group, New York, 1999.

Australia, ultrasonographers and sonologists should be registered and certification requirements include attendance at a course, log book submission of 25 images and practical examination. In Australia, certification of operators and audit of the performance of the nuchal translucency scan is conducted by the Nuchal Translucency — Ultrasound, Education and Monitoring Program run by RANZCOG.

Worldwide this accreditation is conducted under the auspices of the Fetal Medicine Foundation (UK) established by the founder of the technique, Professor Kypros Nicolaides. All imaging centres in Australia that have the Fetal Medicine Foundation software for the estimation of risk for Down syndrome are audited annually.



Fetus with obvious increased nuchal translucency.
 Source: Nicolaides K, et al. *The 11-14 Week Scan: The Diagnosis of Fetal Abnormalities*. Parthenon Publishing Group, New York, 1999.

present in the maternal circulation and this may become available over the next few years.

What does a positive (high-risk) screen mean?

Screening tests are not diagnostic tests. They imply a high risk of Down syndrome, which is arbitrarily defined as a risk more than 1:300 (some laboratories choose different limits such as 1:200, 1:250 or 1:350).

Most women with a positive screen will not have a Down syndrome pregnancy on diagnostic chromosomal testing. This is an important point to highlight both before and after the screening test is performed.

What does a negative (low-risk) screen mean?

As the sensitivity of screening tests can vary between 60% and 90%, a negative screen means that the likelihood of having a Down syndrome pregnancy is small (<1:300).

Case study conclusion

The GP should make a point of telling KC that, even with a negative screen, there is still a small chance (<1:300) that the baby could have Down syndrome. It is important to highlight this small residual risk for women as they may choose to have further screening tests (eg, combined first- and second-trimester testing) for greater reassurance of the Down syndrome risk.

Case study 3

CR is a 37-year-old woman with no relevant past history who has the following results:

Blood test at 10 weeks and two days:

PAPP-A = 0.38 MOM

beta-hCG = 0.15 MOM

Ultrasound at 12 weeks and six days:

nuchal translucency = 1.9mm

Trisomy 21 risk according to age: 1:240

Trisomy 21 risk including nuchal translucency: 1:161

Trisomy 21 risk combined with blood test: 1:1260

Trisomy 18 risk combined with blood test: 1:46

Would you advise chromosomal testing of the pregnancy?

Although the risk of Down syndrome is now low for this woman, any major abnormality in first-trimester screening is associated with a high risk of aneuploidy. In this example, the very low level of beta-hCG made Trisomy 21 very unlikely.

However, this abnormality is often found in Trisomy 18. Combined first-trimester screening cannot only signal 90% of Down syndrome pregnancies but will also detect similarly high proportions of Trisomy 18, Trisomy 13, Turner's syndrome, and maternal and paternal triploidy (table 2, page 8).

In this case study, CR went on to have an amniocentesis that showed Trisomy 13 (47XX+13; figure 5, page 8).

Any significant decrease in PAPP-A, increases the risk of adverse fetal outcome. These outcomes include intrauterine growth retardation, pre-term birth and pregnancy loss. Follow-up offered may include increased surveillance, growth scans, uterine artery Doppler ultrasound and measurement of cervical length at 22 weeks' gestation.

Any significant increase in nuchal translucency (not seen in this case) may identify fetuses at risk of other adverse outcomes and particularly of congenital heart defects. When nuchal translucency is more than 5mm, the risk of cardiac defects is 5% and when nuchal translucency is more



Table 2. Genetic abnormalities associated with combined first-trimester screening.

Aneuploidy	Prevalence	NT	PAPP-A	Beta-hCG
Trisomy 21 (Down)	1:500	↑↑	↓↓	↑↑
Trisomy 18 (Edward)	1:1200	↑↑	↓↓↓	↓
Trisomy 13 (Patau)	1:3300	↑↑	↓↓↓	↓↓
45X (Turner's)	1:2000 (female)	↑↑↑	↓↓	↑
Triploidy (Maternal)	1:10,000	↓	↓↓↓	↓↓↓
Triploidy (Paternal)	1:10,000	↑↑	↓	↑↑↑

↑, ↓ = Increased/decreased values.

than 6mm, about 25% have cardiac defects.

Abnormalities in second-trimester serum screening can similarly provide additional information. Isolated elevation of alpha-fetoprotein can warn of 90% of neural tube defects. Isolated low unconjugated oestriol gives an increased risk of intrauterine growth retardation and oligohydramnios (or fetal steroid sulphatase deficiency). Low total hCG is also associated with Trisomy 18 and triploidy. Furthermore, while isolated abnormalities of second-trimester screening can be associated with an increased risk of adverse fetal outcome, combined abnormalities are even more likely to be associated with outcomes including pre-eclampsia, placental abruption and fetal death.

Further reading available on request.

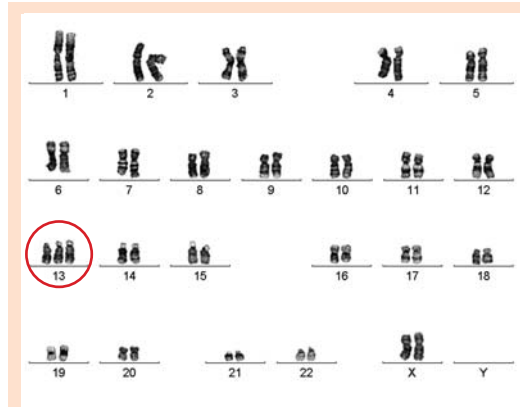


Figure 5. Karyotype obtained after amniocentesis of case study 3 indicating trisomy of chromosome 13 (Patau syndrome).

Key points

- Diagnosis of Down syndrome is increasing with the trend of advancing maternal age.
- One-third of Down syndrome diagnoses are made in pregnant women under the age of 35.
- The prevalence of babies born with Down syndrome has not fallen significantly over the past 20 years.
- All pregnant women should have written information regarding Down syndrome testing and be offered screening early in the pregnancy to allow time for pre-test counselling.
- The preferred screening test is combined first-trimester screening using ultrasound for nuchal translucency and maternal blood testing for PAPP-A and beta-hCG.
- A negative screen indicates a low risk but, even with combined first-trimester screening, 10% of Down syndrome pregnancies will be missed.
- A positive screen indicates a high risk, but most women will not have Down syndrome on diagnostic testing.
- Any major variation from the normal screening results should alert the clinician to other aneuploidies or high-risk pregnancies.