Prenatal diagnosis of single-gene disorders: case studies

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Abstract

In volume 13 issue 3 of this journal, various methods for diagnosing single-gene disorders in the antenatal period were reviewed. The general principles remain the same and have not been discussed again in detail here. Instead, four cases are described that illustrate the points made in this original article, with reference to background information where necessary. Molecular biology is playing an increasingly important role in the prenatal diagnosis of single-gene disorders. In the absence of a family history, or a previously affected sibling, it is usually ultrasound, however, that first raises the possibility of one of these diagnoses in the prenatal period. Clinical investigations, histopathology and molecular testing following birth, or termination of pregnancy, usually combine to provide a precise diagnosis, after which molecular testing in a future pregnancy often becomes possible if the recurrence risks are high.

Keywords prenatal diagnosis; single gene; molecular; amniocentesis; thalassaemia; IEOM; urea cycle; thanatophoric dysplasia; polycystic kidneys

Introduction

More than 5000 human diseases are caused by mutations within single genes. In many cases the genomic location of the affected gene is known, and this may allow some form of molecular testing to determine whether a disease-causing mutation has been inherited by a pregnancy. Indeed, the gene may have been studied in such detail that mutations may be relatively easy to recognise.

There is no doubt that molecular biology has transformed prenatal diagnosis. Testing for these mutations, however, often requires DNA from a prior case within the family, and molecular testing can even then take many months and is not always successful. The exact diagnosis in the affected individual is frequently called into question, making prenatal molecular testing hazardous or impossible. There also remain a number of congenital abnormalities, syndromes and metabolic disorders in which patterns of inheritance suggest a single-gene defect but in which identification of the gene responsible, or its mutations, has so far proved elusive. For all of these reasons, alternative forms of prenatal diagnosis are still required. The options for prenatal diagnosis of single-gene disorders have not changed over the past 3 years, although the ability to make a molecular diagnosis for many conditions has improved.

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The prenatal diagnostic possibilities for single-gene disorders include:

- ultrasound;
- enzyme and metabolite analysis;
- molecular (DNA) testing of chorionic villus or amniotic cells;
- direct biopsy of fetal tissues;
- non-invasive testing of free fetal DNA or fetal cells in maternal blood.

This article examines four cases in detail to illustrate the points made in the previous review. This is a complex field, and the general principles are more important than the specific details.

Case 1

Sue Smith, a 27-year-old woman in her second pregnancy, is scanned routinely at 21 weeks' gestation. The fetal kidneys are both enlarged and echogenic (Figure 1). The amniotic fluid volume is moderately reduced below the normal range, but the maximum vertical pool is 3 cm. No other structural abnormalities are noted on detailed scanning. She has no family or personal history of renal disease or hypertension, and her first child Catherine is fit and well at 4 years of age.

Sue is seen and counselled by a fetal medicine specialist and a paediatric nephrologist, who jointly discuss the differential diagnosis (Table 1). Sue and her partner Bill themselves undergo renal scanning, and no abnormalities are found. This, the oligohydramnios and the absence of a positive family history makes autosomal dominant polycystic kidney disease unlikely.

Indeed, the most likely diagnosis is autosomal recessive polycystic kidney disease, but this cannot be made with any degree of certainty without pathological confirmation, unless there has been a previous confirmed case within the family. There remains the possibility of normal kidneys, although their size and the reduction in liquor volume mean that this is less likely. Even if it were possible to make a precise diagnosis prenatally, most conditions demonstrate a wide spectrum of disease severity. However, 30–50% of children with autosomal recessive polycystic kidney disease die within the first few weeks of life from pulmonary hypoplasia or renal failure. Of the neonatal survivors, 80% are alive still by 10 years of age, although the mean age for diagnosis of chronic renal failure is 4 years. The majority are on dialysis or have undergone transplantation by 20 years of age. Almost half will suffer with hepatic complications (fibrosis and portal hypertension) by adulthood.

Autosomal dominant polycystic kidney disease is often considered to be a less severe condition with a much later age of onset, but cases presenting before birth have a worse outcome, with 40–50% dying before 1 year of age. The neonatal survivors have a significant risk of end-stage renal failure during childhood.

Bill and Sue feel they cannot terminate the pregnancy with this degree of uncertainty and choose to continue. By 26 weeks' gestation the liquor volume has reduced further and the offer of late termination is made but declined. Spontaneous labour ensues at 34 weeks' gestation, and the child, Jamie, dies 14 days after birth with failure of ventilation secondary to pulmonary hypoplasia, and abnormal renal function. Bill and Sue consent to a post-mortem, and the characteristic histopathological features

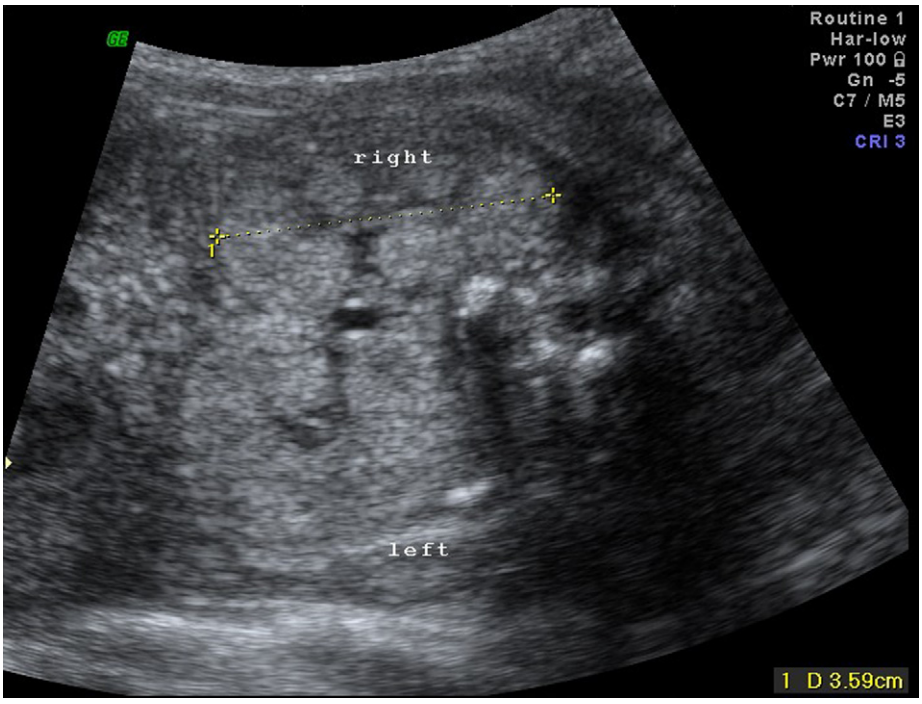

Figure 1 An ultrasound image of bilaterally enlarged echogenic fetal kidneys (case 1).

Differential diagnosis of echogenic fetal kidneys

(A) Genetic renal disorders

Isolated

- *Autosomal recessive polycystic kidney disease*
- Autosomal dominant polycystic kidney disease
- Renal tubular dysgenesis
- Congenital nephrotic syndrome

Syndromic

- *Bardet–Biedl syndrome*
- Meckel syndrome
- Joubert syndrome
- Beckwith–Wiedemann syndrome

(B) Metabolic syndromes associated with renal cysts
e.g. Zellweger syndrome, Smith–Lemli–Opitz syndrome

(C) Non-hereditary causes

- Ureteral obstruction/atresia
- Trisomies

(D) Normal kidneys

Table 1

of autosomal recessive polycystic kidney disease are found, confirming the diagnosis. DNA from Jamie is stored with Bill and Sue's permission.

They return a year later for pre-pregnancy counselling. The clinical geneticist informs them again of the 1 in 4 risk of recurrence for this condition. They decide that they will only pursue another pregnancy if prenatal diagnosis is possible. The fetal medicine specialist advises that, in view of their history, scanning would be an effective way of diagnosing or excluding a recurrence of the condition. Definite scan appearances might not, however, be present until well after 18 weeks' gestation.

The geneticist contacts a laboratory in Europe that is testing for mutations that cause autosomal recessive polycystic kidney disease as part of a research programme. They inform her that the gene underlying this condition (PKHD1) is found on chromosome 6p12 and consists of 86 exons that normally undergo complex splicing. Most PKHD1 mutations are unique to single families ('private'), and mutation testing is difficult and time-consuming. The laboratory staff are currently finding specific mutations in approximately 80% of cases; in the remainder, no mutations are forthcoming. Furthermore, the testing process is taking many months. They are happy to have a sample of the DNA for testing but recommend a linkage study in the meantime.

Linkage analysis

Linkage analysis is still important in cases in which the specific mutation cannot be identified. Linkage analysis relies on the presence of *polymorphisms*. Polymorphisms are variations in the genomic sequence that, unlike mutations, do not disrupt the function of the genes that they are close to or even within. All organisms carry many thousands of polymorphisms, and these differ greatly between individuals. It is sometimes the case that a particular polymorphism is 'linked' to a mutation, i.e. they are inherited together. The presence of the polymorphism is then usually an indicator that the mutation is also present. Restriction fragment length polymorphisms are polymorphisms that alter restriction enzyme cutting sites. Short tandem repeats are short sequences of DNA that are repeated a number of times. This number varies between individuals and can sometimes be used as a marker for the mutation.