Impact of holoprosencephaly, exomphalos, megacystis and high NT in first trimester screening for chromosomal abnormalities

Short title: First-trimester fetal defects and aneuploidies

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ABSTRACT

Objective: To examine the prevalence of alobal holoprosencephaly, exomphalos, megacystis and nuchal translucency thickness (NT) ≥3.5 mm, the incidence and types of associated chromosomal abnormalities and the overall impact on the rate of invasive testing and performance of screening for chromosomal abnormalities of offering invasive testing for these four fetal conditions.

Methods: Prospective screening study for trisomies 21, 18 and 13 by the first-trimester combined test in three maternity units in England.

Results: In the study population of 108,982 cases there were 870 (0.8%) with abnormal karyotype including 654 (75.2%) with trisomies 21, 18 or 13 and 216 (24.8%) with other chromosomal abnormalities. The prevalence of alobal holoprosencephaly, exomphalos, megacystis and NT ≥3.5 mm was 1 in 2,945, 1 in 419, 1 in 1,345 and 1 in 119, respectively. Chromosomal abnormalities were observed in 78.4% of cases of holoprosencephaly, 40.8% of exomphalos, 18.5% of megacystis and 48.5% of those with NT ≥3.5 mm. The most common chromosomal abnormality was trisomy 13 for holoprosencephaly, trisomy 18 for exomphalos and megacystis and trisomy 21 for high NT. Fetal karyotyping for major fetal defects or high NT would potentially detect 57% of all chromosomal abnormalities at an invasive testing rate of 1.1%.

Conclusions: Major fetal defects and high NT at 11-13 weeks' gestation are associated with a high risk of chromosomal abnormalities and merit invasive fetal testing.

INTRODUCTION

Effective screening for trisomies 21, 18 and 13 is provided by the first-trimester combined test with detection rates (DR) of 90%, 97% and 92%, at false positive rate of 4%.[1] Recent evidence suggests that the performance of screening can be improved further by analysis of cell-free (cf) DNA in maternal blood; a meta-analysis of clinical validation and implementation studies has reported that with cfDNA testing the DR for trisomies 21, 18 and 13 were 99%, 96% and 91%, respectively, at overall FPR of 0.35%.[2] However, the cfDNA test is too expensive for universal screening and it is likely to be offered to a subgroup of women identified by the combined test as being at increased risk for trisomies. In the UK, the National Screening Committee (NSC) recommended that the cfDNA test is offered as an alternative to invasive testing to women with a combined test risk of $\geq$1 in 100 at the time of screening.[3] In addition, pregnancies with fetal nuchal translucency thickness (NT) of $\geq$3.5 mm and those with major defects detected at the 11-13 weeks scan should be examined in specialist units for their further management. We have previously reported that many fetal defects can be diagnosed by the first-trimester scan, but specifically, alobal holoprosencephaly, exomphalos and megacystis are always detectable.[4]

The aims of this large screening study are to examine the prevalence of alobal holoprosencephaly, exomphalos, megacystis and NT $\geq$3.5 mm, the incidence and types of associated chromosomal abnormalities and the overall impact on the rate of invasive testing and performance of screening for chromosomal abnormalities of offering invasive testing for these four fetal conditions.

METHODS

This was a prospective screening study for trisomies 21, 18 and 13 by a combination of maternal age, fetal NT, fetal heart rate and serum free ß-hCG and PAPP-A at 11^{+0} -13^{+6} weeks' gestation in women booking for routine pregnancy care at King's College Hospital, London (March 2006 to May 2015), University College London Hospital, London (April 2009 to July 2013) and Medway Maritime Hospital, Gillingham (April 2010 to May 2015).[5] The eligibility criteria were singleton pregnancies with live fetus demonstrated on the 11-13 weeks' scan.

Maternal demographic characteristics, ultrasonographic measurements and biochemical results were recorded in a computer database. Karyotype results and details on pregnancy outcomes were added to the database as soon as they became available. Diagnosis of chromosomal abnormalities was based on prenatal fetal karyotype by chorionic villous sampling or amniocentesis or postnatal karyotype from neonatal blood; phenotypically normal neonates were assumed to be chromosomally normal.

The diagnosis of alobar holoprosencephaly was based on the fusion of the anterior horns of the lateral ventricles and the absence of the butterfly sign in a cross-sectional view of the fetal brain.[6] Exomphalos was diagnosed if there was herniation of bowel or liver in the base of the umbilical cord. Megacystis was defined as enlarged bladder with a longitudinal diameter of ≥7 mm.[7]

RESULTS

During the study period, we examined 115,838 singleton pregnancies. We excluded 6,856 (5.9%) cases because they had missing outcome data (n=5,150) or the fetal karyotype was not known and the pregnancies resulted in termination, miscarriage or stillbirth (n=1,706).

In the study population of 108,982 cases the median maternal age was 31.5 (interquartile range 27.2, 35.2) years and the median gestational age was 12.7 (interquartile range 12.3, 13.1) weeks.[1] There were 108,112 (99.2%) cases with normal fetal karyotype or the birth of a phenotypically normal neonate and 870 (0.8%) cases with abnormal karyotype including trisomy 21 (n=432), trisomy 18 (n=166), trisomy 13 (n=56), mosomy X (n=63), triploidy (n=35) or other aneuploidy [n=118; sex chromosome aneuploidies (n=15), deletions or duplications (n=68), mosaic sex aneuploidies (n=11), mosaic deletions or duplications (n=6), and mosaic trisomies 2, 4, 8, 9, 13, 16, 21 or 22 (n=18)].

The prevalence of alobal holoprosencephaly, exomphalos, megacystis and NT ≥3.5 mm was 1 in 2,945, 1 in 419, 1 in 1,345 and 1 in 119, respectively. In the case of megacystis the bladder length was 7-15 mm in 63 (77.8%) cases and >15 mm in 18 (22.2%). Exomphalos containing liver was observed in 33 (12.7%) cases and the prevalence was unrelated to fetal CRL. Exomphalos containing only bowel was observed in 227 (87.3%) cases and the prevalence increased from 1 in 114 for CRL 45-54 mm to 1 in 953 for CRL 55-84 mm. The incidence and types of associated chromosomal abnormalities for each fetal defect and high NT are shown in Table 1. Chromosomal abnormalities were observed in 78.4% of cases of holoprosencephaly, 40.8% of exomphalos, 18.5% of megacystis and 48.5% of those with NT ≥3.5 mm. The incidence of chromosomal abnormalities was similar in cases of exomphalos containing bowel only with CRL 45-54 mm (41.4%) and those with CRL 55-84 mm (38.4%). The incidence of chromosomal abnormalities was similar in cases of megacystis with bladder length 7-15 mm (19.0%) and those with bladder length >15 mm (16.7%). The most common chromosomal abnormality was trisomy 13 for holoprosencephaly, trisomy 18 for exomphalos and megacystis and trisomy 21 for high NT.

The incidence of chromosomal abnormalities in the presence of each defect was related to the estimated risk from the combined test (Table 2). In holoprosencephaly, exomphalos containing liver, megacystis and high NT all cases of trisomies 21, 18 and 13 were in the subgroups with estimated risk for these trisomies from the combined test of ≥1 in 100. In the case of exomphalos containing bowel only, the rate of trisomies 21, 18 and 13 were 70.2% in those with estimated risk from the combined test of ≥1 in 100 and only 2.4% in those with a risk of <1 in 100. Similarly, the rate of chromosomal abnormalities other than trisomies 21, 18 and 13 in fetuses with holoprosencephaly, exomphalos and high NT was substantially higher in those with estimated risk from the combined test of ≥1 in 100 than in those with a risk of <1 in 100.

Fetal karyotyping for major fetal defects or high NT would potentially detect 57% of all chromosomal abnormalities at an invasive testing rate of 1.1%. If karyotyping was restricted to the subgroup with estimated risk for trisomies from the combined test of ≥1 in 100, 56% of all chromosomal abnormalities would be detected at an invasive testing rate of 0.8%.

DISCUSSION

Main findings

The findings of this large prospective study at 11-13 weeks' gestation demonstrate that the prevalence of alobal holoprosencephaly, exomphalos, megacystis or NT ≥3.5 mm is about 1% and in this group of fetuses the incidence of a wide range of chromosomal abnormalities is more than 40%. Chromosomal abnormalities were observed in 78% of cases of holoprosencephaly, 41% of exomphalos, 19% of megacystis and 49% of those with NT ≥3.5 mm and the most common abnormality was trisomy 13 for holoprosencephaly, trisomy 18 for exomphalos and megacystis and trisomy 21 for high NT. The incidence of chromosomal abnormalities was similar in exomphalos containing bowel only between those with CRL 45-

54 mm and those with CRL 55-84 mm and in megacystis between those with bladder length 7-15 mm and those with bladder length >15 mm.

The rate of trisomies 21, 18 and 13 and other chromosomal abnormalities in fetuses with major fetal defects or high NT was substantially higher in those with estimated risk from the combined test of ≥1 in 100 than in those with a risk of <1 in 100. Fetal karyotyping for major fetal defects or high NT would potentially detect 57% of all chromosomal abnormalities at an invasive testing rate of 1.1%. If in the presence of these defects or high NT the decision in favour or against invasive testing was based on the results of the combined test the rate of invasive testing would be reduced from 1.1% to 0.8% and the DR of chromosomal abnormalities would be reduced from 57% to 56%.

Study limitations

The main limitation of the study relates to ascertainment of pregnancy outcome. Unlike the situation with trisomies 21, 18 and 13, most neonates with sex chromosome aneuploidies and those in the heterogeneous group classified as other chromosomal abnormalities are often phenotypically normal. Consequently, studies that do not involve karyotyping of the whole population will inevitably underestimate the true prevalence of these abnormalities and overestimate the potential sensitivity of a prenatal screening test.[8]

Comparison with findings of previous studies

The findings on the prevalence of major defects and associations with chromosomal abnormalities are in broad agreement with our results from a previous screening study at 11-13 weeks' gestation involving 57,119 pregnancies and several other small studies.[9]

Implications for practice

Holoprosencephaly, exomphalos containing liver, megacystis with bladder length >15 mm and NT ≥3.5 mm are associated with a high rate of perinatal death and high incidence of chromosomal and abnormalities and genetic syndromes; in these conditions invasive testing for fetal karyotyping is an essential step in the investigations that would not only help in defining the prognosis for the index pregnancy but also the risk of recurrence in subsequent pregnancies. In contrast, megacystis with bladder length 7-15 mm and exomphalos containing bowel only, particularly at CRL 45-54 mm, are transient findings with good prognosis [9] and it could be argued that in these cases the decision in favour or against invasive testing should be based on the results of the combined test; alternatively, these conditions could be investigated by cfDNA testing.

Table 1. Prevalence of alobal holoprosencephaly, exomphalos, megacysis and nuchal translucency thickness ≥3.5 mm and incidence of associated chromosomal abnormalities.

Group	n	Abnormal karyotype						
		Total	Trisomy 21	Trisomy 18	Trisomy 13	Triploidy	45,XO	Other
Holoprosencephaly	37	29 (78.4%)	-	5 (17.2%)	18 (62.1%	5 (17.2%)	-	1 (3.4%)
Exomphalos	260	106 (40.8%)	6 (5.7%)	58 (54.7%)	25 (23.6%)	5 (4.7%)	7 (6.6%)	5 (4.7%
Liver	33	15 (45.5%)	-	9 (60.0%)	4 (16.0%)	1 (6.7%)	1 (6.7%)	-
Only bowel	227	91 (40.1%)	6 (6.6%)	49 (53.8%)	21 (23.1%)	4 (4.4%)	6 (6.6%)	5 (5.5%)
CRL 45-54 mm	128	53 (41.4%)	2 (3.8%)	35 (66.0%)	5 (9.4%)	3 (5.7%)	4 (7.5%)	4 (7.5%)
CRL 55-84 mm	99	38 (38.4%)	4 (10.5%)	14 (36.8%)	16 (42.1%)	1 (2.6%)	2 (5.3%)	1 (2.6%)
Megacystis	81	15 (18.5%)	4 (26.7%)	5 (33.3%)	3 (20.0%)	-	-	3 (20.0%)
Bladder 7-15 mm	63	12 (19.0%)	3 (25.0%)	3 (25.0%)	3 (25.0%)	-	-	3 (25.0%)
Bladder >15 mm	18	3 (16.7%)	1 (33.3%)	2 (66.7%)	-	-	-	-
NT ≥3.5 mm	919	446 (48.5%)	227 (50.9%)	101 (22.6%)	32 (7.2%)	6 (1.3%)	59 (13.2%)	21 (4.7%)
Any of the above	1,175	495 (42.1%)	230 (46.5%)	119 (24.0%)	49 (9.9%)	12 (2.4%)	59 (11.9%)	27 (5.5%)
Total population	108,982	870	432	166	56	35	63	118
Defects or high NT	1,175 (1.1%)	495 (56.9%)	230 (53.2%)	119 (71.7%)	48 (85.7%)	12 (34.3%)	59 (93.7%)	27 (22.9%)
CT risk ≥1: in 100	924 (0.8%)	488 (56.1%)	229 (53.0%)	118 (71.1%)	47 (83.9%)	11 (31.4%)	59 (93.7%)	24 (20.3%)

CRL = crown-rump length, NT = nuchal translucency, CT = combined test

Table 2. Incidence of chromosomal abnormalities in the presence of a major defect or high nuchal translucency thickness according to the estimated risk of trisomies 21, 18 or 13 from the combined test.

Group	Trisomies 21, 18 or 13			Other chromosomal defects		
	Total	Risk from combined test		Total	Risk from combined test	
		≥1 in 100	<1 in 100		≥1 in 100	<1 in 100
Holoprosencephaly	23/37 (62.2%)	23/31 (74.2%)	0/6 (0%)*	6/37 (16.2%)	6/31 (19.4%)	0/6 (0%)*
Exomphalos	89/260 (34.2%)	86/126 (68.3%)	3/134 (2.2%)*	17/260 (6.5%)	15/126 (11.9%)	2/134 (1.5%)*
Liver	13/33 (39.4%)	13/22 (59.1%)	0/11 (0%)*	2/33 (6.1%)	2/22 (9.1%)	0/11 (0%)*
Only bowel	76/227 (33.5%)	73/104 (70.2%)	3/123 (2.4%)*	15/227 (6.6%)	13/104 (12.5%)	2/123 (1.6%)*
CRL 45-54 mm	42/128 (32.8%)	42/62 (67.7%)	0/66 (0%)*	11/128 (8.6%)	10/62 (16.1%)	1/66 (1.5%)*
CRL 55-84 mm	34/99 (34.3%)	31/42 (73.8%)	3/57 (5.3%)*	4/99 (4.0%)	3/42 (7.1%)	1/57 (1.8%)*
Megacystis	12/81 (14.8%)	12/29 (41.4%)	0/52 (0%)*	3/81 (3.7%)	1/29 (3.4%)	2/52 (3.8%)
Bladder 7-15 mm	9/63 (14.3%)	9/22 (40.9%)	0/41 (0%)*	3/63 (4.8%)	1/22 (4.5%)	2/41 (4.9%)
Bladder >15 mm	3/18 (16.7%)	3/7 (42.9%)	0/11 (0%)*	0/18 (0%)	0/7 (0%)	0/11 (0%)
NT ≥3.5 mm	360/919 (39.2%)	360/857 (42.0%)	0/62 (0%)*	86/919 (9.4%)	86/857 (10.0%)	0/62 (0%)*

* P<0.05 from chi square test or Fisher exact test for comparison of incidence of chromosomal abnormalities between those with a combined test risk for trisomies of ≥1 in 100 and those with a risk of <1 in 100.

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