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Editorial

Genetic sonography: the historical and clinical role of fetal echocardiography

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INTRODUCTION

The association of congenital heart defects with trisomy 21 was reported over 50 years ago and heart defects remain one of the most common and lethal abnormalities present postnatally in individuals affected by Down syndrome[1–4]. The purpose of this Editorial is to review the use of second-trimester fetal echocardiography as an adjunct to the genetic sonogram from a clinical perspective, based on my experience over the past 20 years[5–13]. The following topics are discussed as they relate to trisomy 21: (1) the incorporation of ultrasound evaluation of the fetal heart as part of the genetic sonogram and comparison of it with other screening modalities from a historical perspective; (2) the postnatal incidence of congenital heart defects; (3) the prenatal incidence of structural and functional heart abnormalities; (4) the relative risk for various cardiac findings; and (5) the suggested use of fetal echocardiography as part of the genetic sonogram given current screening technologies.

HISTORICAL PERSPECTIVE

1985–1990

Second-trimester maternal serum screening

Prior to the 1980s, genetic amniocentesis was offered to women aged 35 years or older for detection of trisomy 21 and other chromosomal abnormalities[14–18]. In the early 1980s, investigators reported the association between decreased levels of maternal serum alpha-fetoprotein (MSAFP) and an increased risk for trisomy 21[19,20]. Cuckle *et al.*[19] suggested that MSAFP screening could detect 21% of fetuses with trisomy 21 at a 5% screen-positive rate. Lustig *et al.*[21] subsequently reported results from implementing MSAFP screening in 275 000 pregnancies, in which the following chromosomal abnormalities were detected: trisomies 13, 18 and 21, 45,X, 47,XXY and triploidy.

Second-trimester genetic sonography

Between 1985 and 1990, investigators reported associations between trisomy 21 and visualization on ultrasound of a shortened femur bone, increased thickness of the nuchal skin fold and major structural malformations[22–35]. Some studies found that these sonographic signs had a higher sensitivity for detection of trisomy 21 than did MSAFP screening, while others did not (Table 1).

1991–1995

Second-trimester maternal serum screening

In the early 1990s, the first reports combining measurement of MSAFP, unconjugated estriol and human chorionic gonadotropin (triple marker screen) with maternal age described detection rates for trisomy 21 of between 45% and 73%[36–38]. When patients were stratified by maternal age, the triple screen identified 60% of fetuses with trisomy 21 in women less than 35 years old, and 75% in women aged 35 years or older[39,40].

First-trimester nuchal translucency (NT) screening

Nicolaides summarized the findings from 16 retrospective studies published between 1992 and 1995 in which 1661 fetuses were identified with an increased NT and found an overall incidence of aneuploidy of 28.3%, with trisomy 21 being the most prevalent (14%)[41]. Up to 1995, there was only one prospective study investigating the relationship

Table 1 Genetic sonography detection rates for trisomy 21 in 1985–1990 (in comparison to maternal serum alpha-fetoprotein screening[19], with 21% sensitivity and 5% false-positive rate (FPR))

		Ultrasound		
Reference	*Trisomy 21 fetuses (n)*	*Markers studied*	*Detection rate (%)*	*FPR (%)*
Detection rate ≤ 21%				
Marquette[32] (1990)	31	BPD/FL	10.0	10
LaFollette[29] (1989)	30	FL	13.3	11.8
Nyberg[33] (1990)	49	BPD/FL	14.3	6.1
Shah[35] (1990)	17	BPD/FL, FL	17.6	6
Dicke[27] (1989)	33	BPD/FL	18.0	4.6
Detection rate > 21%				
Cuckle[26] (1989)	50	FL	24.0	6.3
Nyberg[34] (1990)	94	Major structural defects	33.0	NS
Benacerraf[22] (1987)	10	NSF	40.0	0.14
Benacerraf[25] (1989)	20	BPD/FL, NSF	45.0	5
Hill[28] (1989)	22	BPD/FL, NSF, FL	45.5	7.7
Grist[31] (1990)	6	BPD/FL	50.0	5.9
Lockwood[24] (1987)	55	FL	70.0	4.6
Benacerraf[23] (1987)	28	NSF, FL	75.0	2
Ginsberg[30] (1990)	12	BPD/F, NSF	75.0	6.6

The first author only of each reference is given. BPD, biparietal diameter; FL, femur length; NS, not stated; NSF, nuchal skin fold.

Table 2 Genetic sonography detection rates for trisomy 21 in 1991–1995 (in comparison to maternal serum triple marker screening[39], with 69% sensitivity and 5% false-positive rate (FPR))

		Ultrasound		
Reference	*Trisomy 21 fetuses (n)*	*Markers studied*	*Detection rate (%)*	*FPR (%)*
Detection rate ≤ 69%				
Donnenfeld[48] (1994)	13	NSF	8.0	1.2
Bromley[51] (1995)	22	EIF	18.0	4.7
Nyberg[45] (1993)	45	FL, HL	31.1	7.5
Grandjean[52] (1995)	34	FL, BPD, foot length	35.0	4.6
Grandjean[53] (1995)	44	NSF	39.0	4.9
Biagiotti[46] (1994)	27	HL, FL	44.4	7.6
Campbell[47] (1994)	6	BPD/FL ratio	50.0	8
Bahado-Singh[50] (1995)	8	NSF (positive triple marker screening)	50.0	1.4
Nyberg[55] (1995)	18	Multiple markers	50.0	7.2
Lockwood[44] (1993)	40	BPD, FL, HL	52.4	4.9
Gray[49] (1994)	32	NSF	53.0	6.3
Rodis[56] (1991)	11	FL, HL	64.0	5
Bottalico[66] (2009)	12	Multiple markers	66.0	NS
Detection rate > 69%				
Crane[57] (1991)	16	NSF	75.0	1
Benacerraf[43] (1992)	32	Structural abnormalities, BPD, FL, HL, NSF (scoring index)	81.3	4.4
Nadel[54] (1995)	71	Structural abnormalities, BPD, FL, HL, NSF (scoring index)	83.0	13
DeVore[10] (1995)	15	Multiple markers	87.0	11

The first author only of each reference is given. BPD, biparietal diameter; EIF, echogenic intracardiac focus; FL, femur length; HL, humerus length; NS, not stated; NSF, nuchal skin fold.

between aneuploidy and trisomy 21 and this demonstrated that an increased NT (≥ 2.5 mm) identified 75% of fetuses with trisomy 21[42].

Second-trimester genetic sonography

Between 1991 and 1995, investigators reported the sensitivity of abnormal ultrasound findings for detecting trisomy 21 to be between 8% and 87%[10,43–57]. The combinations of abnormal ultrasound findings listed in Table 1 that outperformed MSAFP screening had lower detection rates when compared with that of triple marker screening (69%). The only studies that found detection rates greater than that of triple marker screening examined a combination of ultrasound markers that included evaluation for major structural malformations of the cardiovascular and non-cardiovascular organ systems (Table 2)[10,43,54,57].

1996–2010

Second-trimester maternal serum screening

In 1992 Van Lith *et al.*[58] reported the association between inhibin-A and trisomy 21. In 1996 two studies examined retrospectively serum from known trisomy 21 cases and controls and concluded that inhibin-A could be added as a fourth marker to the triple marker screen, increasing the sensitivity[59,60]. This became known as the QUAD test, which has a sensitivity of 81% at a 5% screen-positive rate[61].

First-trimester maternal serum and nuchal translucency (NT) screening

Following the reports of associations between increased NT and chromosomal abnormalities, prospective studies were undertaken to validate the retrospective observations. Nicolaides[41] summarized these, reporting an NT screening sensitivity of 76.8%, with a false-positive rate of 4.2%. Subsequent to this observation, investigators have added measurement of serum analytes (beta-human chorionic gonadotropin and pregnancy-associated plasma protein-A) to increase the detection rate to 87%, with a false-positive rate of 5%[41]. In a large prospective study of over 30 000 patients, the FASTER trial[61] reported a sensitivity of 82%, with a false-positive rate of 5%. In 2005, Nicolaides *et al.*[62] reported a two-stage first-trimester screening approach that identified 90% of fetuses with trisomy 21, at a 5% false-positive rate. The improved detection rate required patients with an intermediate risk for trisomy 21 (1 in 101 to 1 in 1000) following first-trimester NT and serum screening to be evaluated for the following fetal abnormalities: absent nasal bone, tricuspid regurgitation and abnormal blood flow through the ductus venosus. If any of these findings was present, the patient was offered invasive testing to detect trisomy 21.

First-trimester combined plus second-trimester QUAD screening

In 2005, Malone *et al.*[61] reported results from the FASTER trial in which a 95% detection rate (5% false-positive rate) for trisomy 21 was reported by integrating the first-trimester combined test with the second-trimester QUAD test.

Second-trimester genetic sonography

Since 1996, various investigators have reported the association between abnormal ultrasound findings and trisomy 21, with detection rates ranging between 53.1% and 92.8% (Table 3)[5,63–66]. The only studies that reported detection rates exceeding 90% included a fetal echocardiogram as part of the ultrasound examination (Table 3)[5,67].

Figure 1 summarizes the detection rates for trisomy 21 between 1985 and 2010 using different testing schemes. Genetic sonography outperformed second-trimester MSAFP, triple marker and QUAD screening. When first-trimester screening was introduced, genetic sonography still had a higher sensitivity than did screening by NT plus serum, and first-trimester NT plus serum, nasal bone and tricuspid regurgitation, but had a higher false-positive rate (13.3–14%, Table 3)[5,67]. It was only when first-trimester NT and serum were combined with second-trimester QUAD screening that genetic sonography detected fewer fetuses with trisomy 21 (Figure 1).

POSTNATAL INCIDENCE OF CONGENITAL HEART DEFECTS IN INDIVIDUALS WITH TRISOMY 21

Table 4 summarizes data from nine studies reported between 1961 and 2008. There are a number of differences between these studies (study duration and geographical distribution, time study was conducted, classification of heart defects, and number of trisomy

Table 3 Genetic sonography detection rates for trisomy 21 in 1996–2010 (in comparison to first- and second-trimester integrated screening, with 90% sensitivity)

		Ultrasound		
Reference	*Trisomy 21 fetuses* (n)	*Markers studied*	*Detection rate (%)*	*FPR (%)*
Detection rate ≤ 90%				
Smith-Bindman[64] (2007)	245	Multiple markers	53.1	14.2
Szigeti[65] (2007)	184	Multiple markers	63.8	NS
Hobbins[63] (2003)	125	Multiple markers	71	NS
Wax[95] (2000)	7	Multiple markers	71	12.1
Vintzileos[109] (1999)	34	Multiple markers	82	8
Vintzileos[110] (2002)	53	Multiple markers	87	11
Detection rate > 90%				
DeVore[5] (2000)	80	Multiple markers	91	14
Vintzileos[67] (1996)	14	Multiple markers	92.8	13.3

The first author only of each reference is given. NS, not stated.

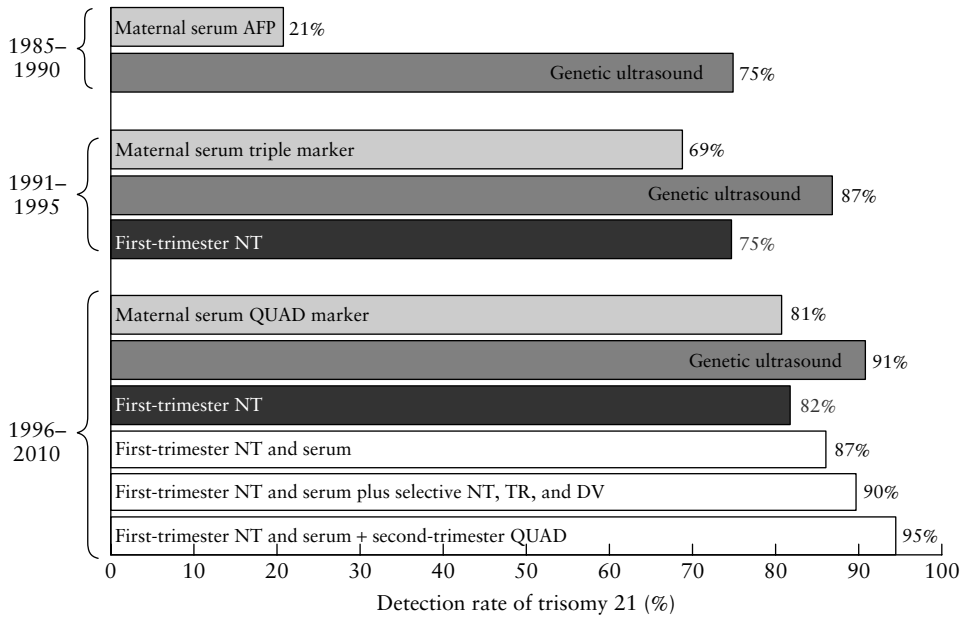

Figure 1 Detection rate of trisomy 21 by different screening tests in three time periods. AFP, maternal serum alpha-fetoprotein; DV, ductus venosus; NT, nuchal translucency; QUAD, second-trimester maternal serum screening; TR, tricuspid regurgitation.

Table 4 Incidence of congenital heart defects (CHD) in liveborn infants with trisomy 21

			CHD (n *(%))*				
Reference	*Study period*	*Trisomy 21 liveborns* (n)	*All CHD*	*Endocardial cushion defect*	*Ventricular septal defect*	*Atrial septal defect*	*Other CHD*
Rowe[69] (1961)	1955–1957	174	70 (40.2)	25 (14.4)	23 (13.2)	6 (3.4)	16 (9.2)
Martin[70] (1989)	NR	137	64 (46.7)	32 (23.4)	14 (10.2)	9 (6.6)	9 (6.6)
Tubman[71] (1991)	1987–1989	81	34 (42.0)	13 (16.0)	5 (6.2)	7 (8.6)	9 (11.1)
Khoury[72] (1992)	1968–1989	532	176 (33.1)	93 (17.5)	30 (5.6)	35 (6.6)	18 (3.4)
Wells[73] (1994)	1988–1992	118	57 (48.3)	22 (18.6)	17 (14.4)	16 (13.6)	2 (1.7)
Freeman[74] (1998)	1989–1995	227	100 (44.0)	45 (19.8)	35 (15.4)	5 (2.2)	15 (6.6)
Stoll[75] (1998)	1979–1996	398	184 (46.2)	79 (19.8)	58 (14.6)	NR	47 (11.8)
McElhinney[76] (2002)	1988–1999	114	75 (65.8)	33 (28.9)	17 (14.9)	NR	25 (21.9)
Nisli[77] (2008)	1994–2006	1042	412 (39.5)	203 (19.5)	92 (8.8)	71 (6.8)	46 (4.4)
Total		2823	1172 (41.5)	545 (19.3)	291 (10.3)	149 (5.3)	187 (6.6)

The first author only of each reference is given. NR, not reported.

21 cases reported). The average incidence of congenital heart defects in individuals with trisomy 21 was 41.5% (range, 33.1–65.8%). The incidence of an endocardial cushion defect was 19.3% (range 14.4% to 28.9%), ventricular septal defect (VSD) 10.3% (range 5.6% to 15.4%), atrial septal defect (ASD) 5.3% (range 2.2% to 13.6%), and other heart defects 6.6% (range 1.7% to 21.9%).

In 2007, Cleves *et al.*[68] reported results from a national hospital discharge database that identified the rate of structural birth defects in liveborn infants with trisomy 21 between 1993 and 2002. The analysis compared findings between 11 372 infants with trisomy 21 and 7 884 209 infants without trisomy 21. From this analysis there are several important observations. First, the incidence of endocardial cushion defects was less than that reported in the studies listed in Table 4 (13.16% vs. 20.2%). Second, there were a number of congenital heart defects observed in trisomy 21 infants (Table 5) that had not been reported in smaller datasets (cf. Table 4)[68–77]. Third, the relative risk for heart defects in trisomy 21 vs. non-trisomy 21 infants was computed and found to be > 5 for all defects listed in Table 5 except for aortic valve stenosis. This suggests that when the cardiovascular system is examined in the fetus at risk for trisomy 21, those malformations listed in Table 5 should be considered as part of the evaluation.

PRENATAL INCIDENCE OF STRUCTURAL AND FUNCTIONAL CONGENITAL HEART DEFECTS IN FETUSES WITH TRISOMY 21

Incidence of structural heart defects detected following genetic amniocentesis

In 2000, Paladini *et al.*[78] examined by ultrasound fetuses with previously identified trisomy 21, having undergone

Table 5 Congenital heart defects (CHD) in 11 372 liveborn infants with trisomy 21[68]

CHD	*% of trisomy 21 liveborns with CHD*	*RR* (95% CI)*
All CHD	35.60	74.51 (70.97, 78.22)
Atrial septal defect	17.36	59.80 (55.63, 64.28)
Ventricular septal defect	13.16	48.39 (45.49, 51.49)
Endocardial cushion defect	10.43	1026.23 (920.65, 1144.80)
Tetralogy of Fallot	2.10	69.34 (59.84, 80.35)
Coarctation of the aorta	0.92	27.02 (21.57, 33.85)
Pulmonary valve stenosis	0.53	8.37 (6.22, 11.26)
Transposition of the great arteries	0.36	10.75 (7.66, 15.08)
Tricuspid valve atresia and stenosis	0.16	18.77 (11.03, 31.92)
Common truncus	0.16	26.65 (15.68, 45.29)
Ebstein's anomaly	0.14	26.99 (15.00, 48.56)
Hypoplastic left ventricle	0.12	5.70 (3.19, 10.20)
Pulmonary valve atresia	0.12	13.78 (7.82, 24.28)
Aortic valve stenosis	0.03	2.72 (1.00, 7.38)

*The relative risk (RR) compares the incidence of CHD in liveborns with and those without trisomy 21.

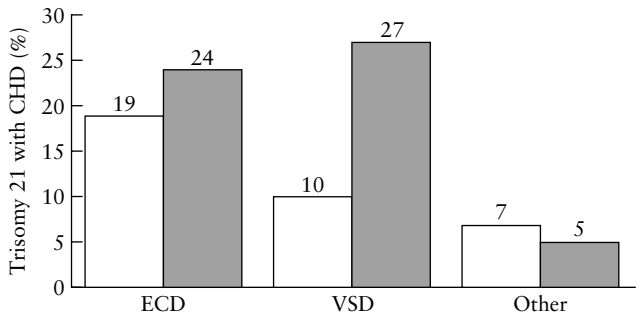

Figure 2 Comparison between incidence of congenital heart defects (CHD) in postnatal (□) and prenatal (■) trisomy 21 studies. Postnatal data are the average from Table 4 and prenatal data are from the study by Paladini *et al.*[78]. ECD, endocardial cushion defect; VSD, ventricular septal defect.

amniocentesis and karyotyping. An echocardiogram was performed in the late second trimester (24 weeks). In this group of fetuses the incidence of congenital heart defects was 56% ($n = 41$), their distribution being as follows: endocardial cushion defect, 24%; ventricular septal defect, 27%; and other defects, 5% (one case of coarctation of the aorta and one of tetralogy of Fallot). Figure 2 compares the combined postnatal data (Table 4) with the prenatal data from Paladini *et al.*[78]: the incidences of endocardial cushion defect and ventricular septal defect were higher in the prenatal than the postnatal analysis. The reasons for this may be the *in-utero* death of fetuses with trisomy 21 who have congenital heart defects, or the spontaneous closure of ventricular septal defects *in utero*[79–81].

Incidence of structural and functional congenital heart defects identified prior to genetic amniocentesis

Table 6 lists studies in which the investigators detected congenital heart defects at the time of the ultrasound examination, prior to discovery that the fetus had trisomy 21. The type of ultrasound evaluation of the cardiovascular system was not reported in three studies, while in others, screening for congenital heart defects ranged from evaluation of the four-chamber view to a full fetal echocardiogram that included examination of the outflow tracts and color Doppler evaluation of blood flow. The detection rate for heart defects ranged from 5.32% to 46.0%. Only studies that included a targeted fetal echocardiogram reported an incidence of congenital heart defects that was similar to that found postnatally (>35%) in individuals with trisomy 21.

In 2000 I reported a number of structural and functional heart abnormalities in fetuses with trisomy 21[5]. Of the cardiac defects listed in Table 7, only mitral regurgitation (1.3%) and isolated outflow tract anomalies (3.8%) had lower sensitivities for the detection of trisomy 21 than had the other findings.

I also found a pericardial effusion in 18.8% of fetuses with trisomy 21[5]. The association between a pericardial effusion and trisomy 21 has been reported by other investigators[82–84]. Tricuspid regurgitation was present in 28.8% of fetuses with Down syndrome[5]. Subsequent to this report, investigators had identified tricuspid regurgitation to be associated with trisomy 21 identified during first-trimester NT screening[62,85–92].

RELATIVE RISK OF CARDIAC ABNORMALITIES ASSOCIATED WITH TRISOMY 21

The relative risk is the probability of a fetus having trisomy 21 when compared with a fetus without this condition. The presence of an ultrasound marker with a relative risk of 4 suggests a four-fold increase from the previous risk, assuming that the ultrasound marker is independent of other risk factors.

Only one study listed in Table 6 reported relative risks for specific cardiac abnormalities detected during the second-trimester genetic sonogram (Table 8, Figures 3–7)[5]. Logistic regression identified an interaction between increased nuchal skin fold and right-to-left disproportion of the heart, defined as right atrium/ventricle larger than left atrium/ventricle. This reaffirmed an association that had been described previously[93]. If the genetic sonogram demonstrated none of the abnormalities listed in Table 8, then the relative risk of a normal examination was 0.11. Figure 8 illustrates the calculation of the risk for trisomy 21 using relative risks from Table 8.

Another cardiac finding that has been associated with trisomy 21 is an echogenic intracardiac focus (EIF), located predominantly in the left ventricular chamber

Table 6 Congenital heart defects (CHD) detected at the time of genetic sonography in fetuses with trisomy 21

Reference	*Study period*	*Trisomy 21 fetuses* (n)	*Type of heart study*	*Fetuses with CHD* (n)	*% of trisomy 21 fetuses with CHD*	*Types of cardiac defect*
Nyberg[34] (1990)	1984–1990	94	NS	5	5.32	ECD (4), VSD (1)
Verdin[111] (1998)	1993–1995	16	NS	1	6.25	ASD, dilated right atrium
Benacerraf[23] (1987)	1983–1987	28	NS	2	7.14	ECD (2)
Hobbins[63] (2003)	NS	125	4-CV, outflow tracts	12	9.60	NS
Nyberg[112] (1998)	1990–1996	142	4-CV	15	10.56	NS
Watson[113] (1994)	NS	15	NS	2	13.00	ECD (1), VSD (1)
Szigeti[65] (2007)	1990–2004	184	4-CV, outflow tracts	33	17.93	NS
Crane[57] (1991)	1988–1990	16	NS	3	18.75	Ebstein anomaly (1), ECD (1), ASD (1)
Benacerraf[43] (1992)	1990–1991	32	NS	7	21.88	NS
Vergani[114] (1999)	1990–1996	22	4-CV, outflow tracts, color Doppler	5	22.73	ECD (4) Ebstein anomaly (1)
Vintzileos[67] (1996)	1992–1995	13	4-CV, outflow tracts	5	35.7	VSD (5)
DeVore[5] (2000)	1990–1999	80	4-CV, outflow tracts, color Doppler	37	41.11	ECD (7), VSD (27), OFT (3)
DeVore[10] (1995)	1990–1992	15	4-CV, outflow tracts, color Doppler	9	46.00	Complex heart defects (7), VSD (2)

The first author only of each reference is given. 4-CV, four-chamber view; ASD, atrial septal defect; ECD, endocardial cushion defect; NS, not stated; OFT, outflow tract abnormalities; VSD, ventricular septal defect.

Table 7 Abnormal ultrasound findings in 80 second-trimester fetuses with trisomy 21[5]

Abnormal ultrasound finding	n	*Sensitivity (%)*	*Specificity (%)*
Structural heart defects			
Ventricular septal defect	27	33.8*	94.6
Endocardial cushion defect	7	8.8*	‡
Right-to-left disproportion†	18	22.5*	98.9
Outflow tract abnormalities	3	3.8*	99.7
Functional heart defects			
Pericardial effusion	15	18.8*	97.6
Tricuspid regurgitation	23	28.8*	98.3
Mitral regurgitation	1	1.3**	99.9

Compared with control patients ($n = 2000$), the listed abnormal ultrasound findings were significantly more frequent in fetuses with trisomy 21: *$P < 0.001$, **$P < 0.01$. †Right atrium/ventricle larger than left atrium/ventricle. ‡There were no fetuses in the control group that had an endocardial cushion defect.

Table 8 Relative risk (RR) for cardiovascular and non-cardiovascular ultrasound markers in 80 second-trimester fetuses with trisomy 21[5] (with sensitivity 91% and false-positive rate 14%)

Abnormal ultrasound finding	*RR (95% CI)*
Head	
CNS abnormalities	24.85 (5.78, 106.78)
Increased NSF (≥ 6 mm)	71.31 (26.19, 194.13)
Chest	
Structural heart defects	
VSD	12.54 (6.16, 25.50)
R-to-L disproportion*	88.29 (29.37, 265.38)
Functional heart defects	
Pericardial effusion	10.02 (3.82, 26.29)
Tricuspid regurgitation	5.89 (2.38, 14.49)
Abdomen	
Hyperechoic bowel	5.65 (2.45, 13.06)
Pyelectasis	4.57 (1.46, 14.25)
Interactions	
Increased NSF and R-to-L disproportion*	0.029 (0.0027, 0.319)

*Right atrium/ventricle larger than left atrium/ventricle. CNS, central nervous system; NSF, nuchal skin fold; R-to-L, right-to-left; VSD, ventricular septal defect.

and thought to be calcification of the papillary muscle[94]. There has been debate regarding the significance of an isolated EIF with respect to the risk for trisomy 21[95–103]. The reported relative risk for an isolated EIF has ranged between 1.6 and 74[95–103]. One of the difficulties in interpreting the data is knowing whether or not the EIF was truly isolated, or whether there were cardiac defects present, but not reported. In 2001, Huggon *et al.*[97] addressed this question in patients referred for fetal echocardiography because of an increased risk for congenital heart defects, and found that an isolated EIF had a relative risk of 5.54 for trisomy 21. In 2006, I reported results from 59 fetuses with trisomy 21 who underwent detailed fetal echocardiography having been referred for advanced maternal age or abnormal second-trimester maternal serum screening[104]. The incidence of fetuses with an EIF (isolated or non-isolated) was significantly higher ($P < 0.001$) in fetuses with trisomy 21 (11.9%; 7/59) than in those without trisomy 21 (0.9%; 29/3311). From this group only one (1.7%) fetus had an isolated EIF and trisomy 21. However, this was significantly different ($P < 0.01$) from the control group results (0.12%; 4/3311). The relative risk for trisomy 21 when an isolated EIF was identified at fetal echocardiography was 1.94[104].

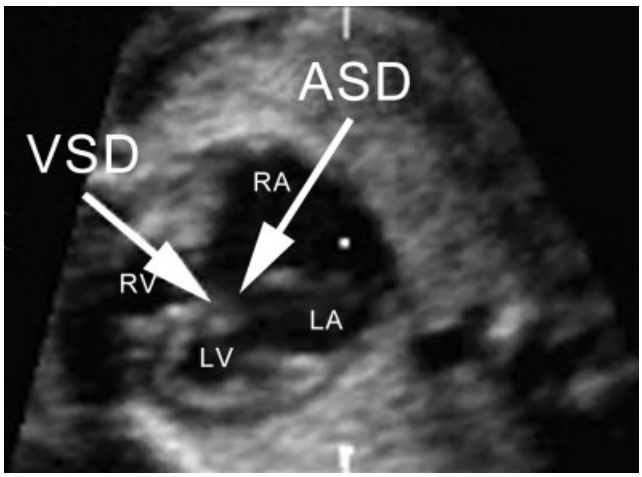

Figure 3 Four-chamber view illustrating an endocardial cushion defect in which a ventricular (VSD) and atrial (ASD) septal defect are present. LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle.

SUGGESTED USE OF FETAL ECHOCARDIOGRAPHY AS PART OF THE GENETIC SONOGRAM GIVEN CURRENT SCREENING TECHNOLOGIES

At present, common screening tests for trisomy 21 may include any of the following: (1) first-trimester combined NT and serum screening, (2) first-trimester combined NT and serum screening plus second-trimester QUAD screening, (3) first-trimester serum and second-trimester serum screening, or (4) second-trimester QUAD screening. Because of the technical skills of the sonographer/sonologist required to detect over 90% of trisomy 21 fetuses using non-cardiac and cardiac markers (Table 8), genetic sonography should only be used as an adjunct to the above screening protocols or in women who register for prenatal care after 20 weeks of gestation. The following two scenarios illustrate when genetic sonography, coupled with fetal echocardiography, should be considered.

Genetic sonography as an adjunct to first-trimester NT and serum and/or second-trimester serum screening

When genetic sonography was first introduced in the early 1990s it was an option for screening for trisomy 21 in women less than 35 years of age for two reasons: (1) the detection rate was similar to or higher than that using MSAFP screening, and (2) the ultrasound exam only required measurements of the biparietal diameter, femur length and nuchal skin fold (Table 1). However, as more analytes were added, second-trimester maternal serum (triple and QUAD) screening increased the detection rate for trisomy 21, was easier to use, and did not require the specialized ultrasound skills needed to keep the genetic sonogram comparable in terms of detection rates (Table 2).

Investigators have reported the use of genetic sonography as an adjunct to other screening protocols. In 2001, Roberto Romero and I[11] reported offering genetic sonography to women considered to be at moderate risk (1:190–1:1000) for trisomy 21

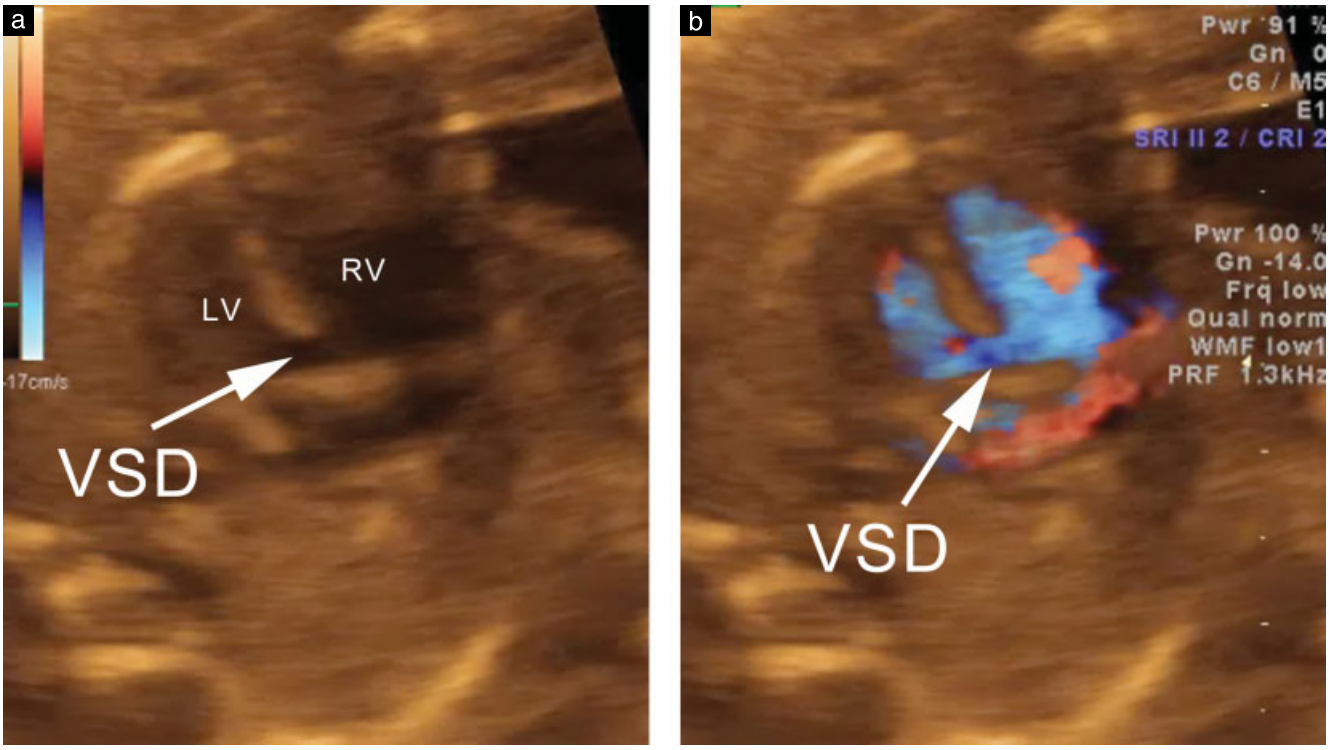

Figure 4 Four-chamber view illustrating a ventricular septal defect (VSD) at the level of the inflow tracts. (a) B-mode image; (b) power Doppler image confirming flow at the level of the VSD. LV, left ventricle; RV, right ventricle.

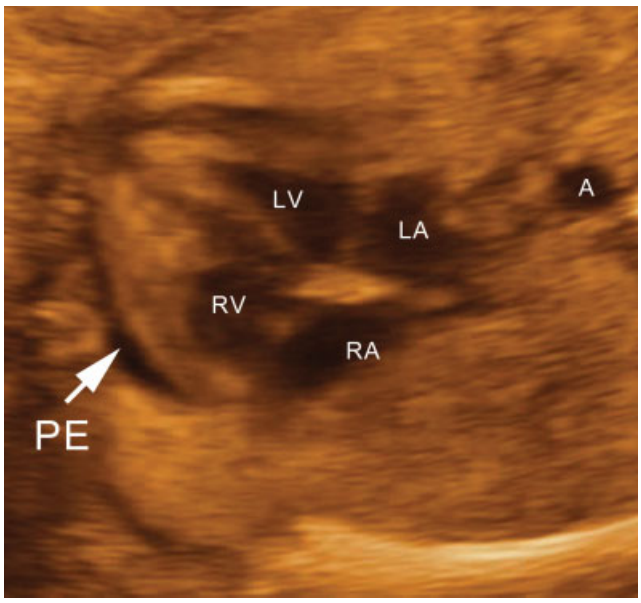

Figure 5 Four-chamber view illustrating a pericardial effusion (PE) along the right ventricular wall. In fetuses with trisomy 21 the PE is almost always along this wall. A, aorta; LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle.

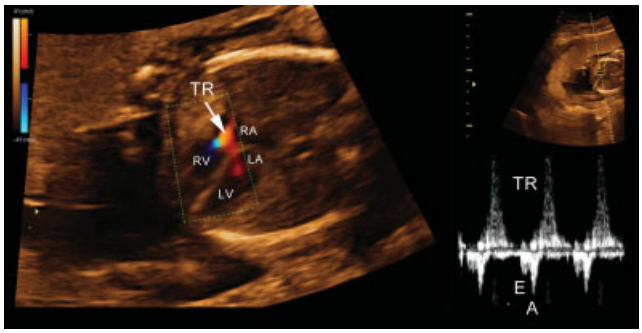

Figure 6 Four-chamber view with pulsed Doppler illustrating tricuspid regurgitation (TR). In fetuses with trisomy 21 the duration of the TR Doppler jet is often greater than 50% of systole. A, A-wave; E, E-wave; LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle.

after maternal triple-marker screening increased the trisomy 21 detection rate from 49% to 68.1–77.8% and reduced the number of amniocenteses required to detect a single fetus with trisomy 21. In 2002, we[12] reported an analysis offering genetic sonography, followed by amniocentesis if abnormalities were detected, to patients 35 years and older who had originally declined invasive testing for the diagnosis of trisomy 21. We found that such a protocol resulted in a higher overall detection rate for trisomy 21 (70–97%) and did not increase the risk of pregnancy loss. In 2003[13], we reported a third application of genetic sonography when we concluded that offering genetic sonography to patients 35 years of age and older following a negative maternal serum triple-marker screening test resulted in an increase in the detection rate of trisomy 21 from 86.3% to 93.2–98.6%. In 2003, I[8] reviewed the data and reported that genetic sonography was cost effective.

Following the widespread use of first-trimester combined NT and serum screening, and first- and second-trimester integrated screening, the question that has recently been posed is whether genetic sonography has any value following normal results using these testing schemes. In 2006, Rozenberg *et al.*[105] reported the detection and screen-positive rates following first-trimester combined screening for trisomy 21 to be 80.4% (41/51) and 2.7%, respectively. When the screen-negative women underwent second-trimester genetic sonography, the overall detection rate for trisomy 21 increased to 90.2% (46/51), with a 4.2%, screen-positive rate. Four of the five fetuses with trisomy 21 and an abnormal ultrasound exam had cardiac defects (endocardial cushion defect ($n = 2$); ventricular septal defect ($n = 1$); and tetralogy of Fallot ($n = 1$)). In 2009, results from the FASTER trial[106] were published, in which the investigators examined the effectiveness of second-trimester genetic sonography in modifying Down syndrome screening results. The study found that the inclusion of genetic sonographic markers increased the detection rate following the first-trimester combined test from 81% to 90%, the first- and second-trimester integrated test from 93% to 98%, and the second-trimester QUAD test from 81% to 90%.

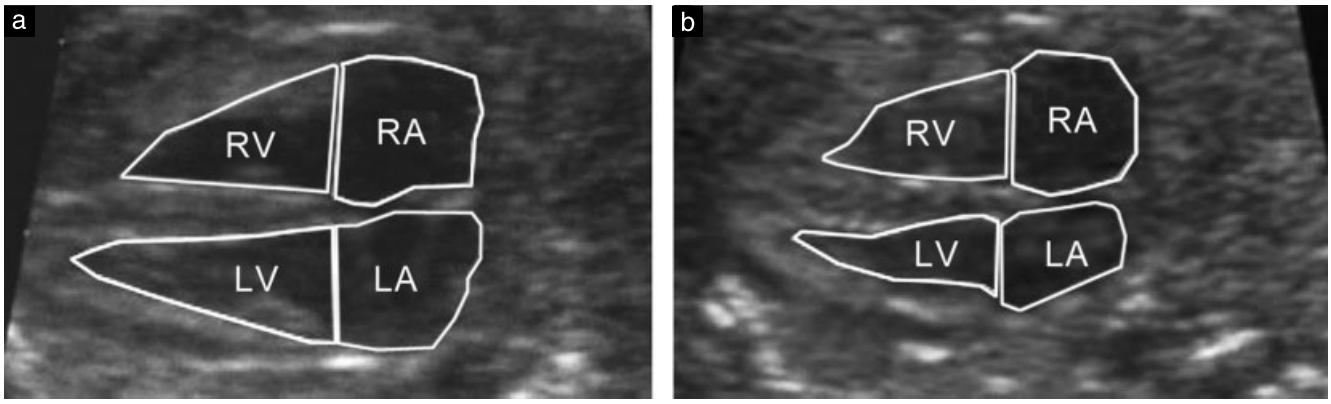

Figure 7 Four-chamber view in two fetuses with trisomy 21, showing the normal appearance (a) and right-to-left disproportion of the atrial and ventricular chambers (b). Note the right atrium and ventricular chambers are larger than the left atrial and ventricular chambers in (b). LA, left atrium; LV, left ventricle; RA, right atrium; RV, right ventricle.

Single abnormal ultrasound finding

Data: The abnormal ultrasound finding is a pericardial effusion with a relative risk of 10.02. Prior risk for trisomy 21 is 1 in 270.

Calculation of Risk:

1. Divide 1/(270−1) = 0.0037
2. Multiply the prior risk (0.0037) by the relative risk for a pericardial effusion (10.02)
3. Calculation = 0.0037 × 10.02 = 0.037
4. Divide 1/0.037 = 28
5. The new risk for trisomy 21 is 1 in 28

Two independent abnormal ultrasound findings

Data: The abnormal ultrasound findings are tricuspid regurgitation and hyperechoic bowel with relative risks of 5.89 and 5.65, respectively.

Prior risk for trisomy 21 is 1 in 270

Calculation of Risk:

1. Divide 1/(270−1) = 0.0037
2. Multiply the risk (0.0037) by the relative risks for both findings, 5.89 and 5.65
3. Calculation = 0.0037 × 5.89 × 5.65 = 0.123
4. Divide 1/0.123 = 8
5. The new risk for trisomy 21 is 1 in 8

Two non-independent ultrasound markers

Data: The abnormal ultrasound findings are right-to-left disproportion of the heart and an abnormal nuchal skin fold. The relative risks are 88.29 and 71.31, respectively. Because there is an interaction between these two variables, the relative risk for the interaction is 0.029.

Prior risk for trisomy 21 is 1 in 270

Calculation of Risk:

1. Divide 1/(270−1) = 0.0037
2. Multiply the prior risk (0.0037) by the relative risks
3. Calculation = 0.0037 × 88.29 × 71.31 × 0.029 = 0.676
4. Divide 1/0.676 = 2
5. The new risk for trisomy 21 is 1 in 2

Normal ultrasound examination study in which none of the ultrasound markers is present

Data: The examiner screened for all ultrasound markers listed in Table 8. No abnormalities were identified. The relative risk following a normal study is 0.11.

Prior risk for trisomy 21 is 1 in 100

Calculation of Risk:

1. Divide 1/(100−1) = 0.01
2. Multiply the prior risk (0.01) by the relative risk of a normal ultrasound study (0.11)
3. Calculation = 0.01 × 0.11 = 0.0011
4. Divide 1/0.0011 = 900
5. The new risk for trisomy 21 is 1 in 900

Figure 8 Calculating the *a posteriori* risk for trisomy 21 following genetic sonography.

Genetic sonography used as an arbitrator when prior screening tests are positive and the patient does not desire invasive testing

Although the false-positive rate for current first- and second-trimester screening, as defined above, is less than 7%, not all patients who are screen-positive desire invasive testing. When confronted by this clinical dilemma, genetic sonography is often useful when the sensitivity is 90% or higher. In 2003, Yeo and Vintzileos[107] reported the use of second-trimester genetic sonography to reduce the need for amniocentesis in the high-risk patient. When the genetic sonogram was normal they reported the following: (1) the amniocentesis rate was only 3%, (2) genetic

sonography was a patient-driven service, and (3) the information obtained at the time of the ultrasound examination made an important contribution to the patient's decision as to whether or not to proceed with invasive testing. Using the relative risks listed in Table 8 and the calculation described in Figure 8, patients with a risk for trisomy 21 of between 1 in 31 and 1 in 270 can decrease their risk to 1 in 272 to 1 in 2545, respectively (Table 9).

While investigators have reported an overall false-positive rate less than 7% for all women studied (see above), the false-positive rate for women over the age of 38 years increases as a function of maternal age[108]. For this reason, fetal echocardiographic genetic sonography, which has a fixed false-positive rate of 14%[5], may be an option for older women, in whom the false-positive rate for first-trimester NT and serum screening as well as second-trimester QUAD screening is greater than 14%[108].

CONCLUSION

After reviewing the data presented in this Editorial, I would like to make the following observations:

Table 9 Adjusted risk for trisomy 21 following a normal genetic sonogram incorporating fetal echocardiography (relative risk following a normal study = 0.11)[5]

Risk (1 in:) for trisomy 21 following:		*Risk (1 in:) for trisomy 21 following:*	
Initial 1st and/or 2nd trimester screening test	*Normal genetic sonogram using criteria in Table 8*	*Initial 1st and/or 2nd trimester screening test*	*Normal genetic sonogram using criteria in Table 8*
5	36	140	1260
10	82	145	1309
15	127	150	1355
20	173	155	1400
25	218	160	1445
30	264	165	1491
35	309	170	1536
40	355	175	1582
45	400	180	1627
50	445	185	1673
55	491	190	1718
60	536	195	1764
65	582	200	1809
70	627	205	1855
75	673	210	1900
80	718	215	1945
85	764	220	1991
90	809	225	2036
95	855	230	2082
100	900	235	2127
105	945	240	2173
110	991	245	2218
115	1036	250	2264
120	1082	255	2309
125	1127	260	2355
130	1173	265	2400
135	1218	270	2845

1. Fetal echocardiography, when coupled with other ultrasound markers, can identify over 90% of fetuses with trisomy 21. This is comparable to first-trimester NT and serum screening as well as integrated screening.
2. Because of the difficulty in detecting structural and functional cardiovascular abnormalities during the second trimester of pregnancy, fetal echocardiography as a component of the genetic sonogram may be difficult to apply as a primary screening tool for trisomy 21.
3. Fetal echocardiographic genetic sonography could be used for patients who present late (after 20 weeks of gestation) for prenatal care and are therefore not eligible for second-trimester QUAD screening.
4. Fetal echocardiographic genetic sonography, when used as an adjunct to first- and/or second-trimester screening for trisomy 21 may increase the detection rate to as high as 99%. This may be advantageous for patients who desire the highest sensitivity for detection before considering invasive testing.
5. Because of the increasing false-positive rate associated with first trimester NT plus serum screening in women 38 years of age and older (false-positive rate 16% at 38 years, false-positive rate 58% at 45 years) fetal echocardiographic genetic sonography may be an alternative option for these patients because of its high sensitivity (91%) and lower false-positive rate (14%)[5,108].

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