Singapore Journal of Obstetrics and Gynaecology
2006; 37(1): 36-40

Review Article

Down Syndrome Screening in Twin Pregnancies

XH Ong, TYT Tan, FM Lai, GSH Yeo

INTRODUCTION

The routine offer of Down syndrome (DS) screening for antenatal patients who booked before 20 weeks gestation is now a standard of care. Common methods of screening available for singleton pregnancies include:

(a) Historical methods – Family history of translocation DS, past obstetric history of fetal chromosomal abnormalities, advanced maternal age
(b) Ultrasound methods – nuchal translucency (NT) and/or nasal bone (NB) detection at 11-14 weeks, second trimester genetic sonogram
(c) Serum biochemical methods – free beta-human chorionic gonadotrophin (b-hCG) and pregnancy associated plasma protein A (PAPP-A) at 11-14 weeks, and alpha fetoprotein (AFP) and beta-hCG at 15-20 weeks
(d) Integrated tests – combination of ultrasound methods and/or maternal serum biochemical methods have been reported to give higher detection rates albeit at higher costs

Most units that offer DS screening programs use cut-offs of about 1:250 to 1:300 to offer invasive testing, a practice that has been passed down since the early days. It also fits quite nicely with what we know about procedural loss rates of amniocentesis for singletons, which is 0.3 to 0.5%.

ADDITIONAL CONSIDERATIONS IN DS SCREENING IN TWINS

However, there are added considerations in the screening for DS in twins. Firstly, the chorionicity of a twin pregnancy may indicate the zygosity. All monochorionic twins are monozygotic while the majority of dichorionic twins are dizygotic. Hence all monochorionic twins are concordant for chromosomal make-up with exceptions possible when there is discordance in anomalies. Dichorionic twins, on the other hand, should be assumed to be discordant in chromosomal make-up for practical purposes. Secondly, twin and higher order multiple pregnancies are increasingly contributed by increasing use of assisted reproductive techniques for subfertile couples. To couples with twins from natural conceptions or assisted reproductive techniques, the tolerance level for procedure-related pregnancy miscarriage rates may be different from those with singleton pregnancies. Thirdly, invasive karyotyping in twins is associated with a 2 to 3% procedure-related miscarriage rate, higher than that for singletons.[1]

XH Ong
Medical Student
National University of Singapore

TYT Tan
Consultant
Department of Maternal Fetal Medicine
KK Women's and Children's Hospital

FM Lai
Head and Consultant
Antenatal Risk Assessment Unit
Department of Maternal Fetal Medicine
KK Women's and Children's Hospital

GSH Yeo
Head and Senior Consultant
Department of Maternal Fetal Medicine
KK Women's and Children's Hospital
Correspondence to:

Dr Tony Tan
Department of Maternal Fetal Medicine
KK Women's and Children's Hospital
100 Bukit Timah Road
Singapore 229899
E-mail: Tony.Tan.YT@kkh.com.sg

Prior to any DS screening test, the following facts should be made known to the couple expecting twins:

1. DS is not usually a lethal condition. If one twin is normal and the other is a DS fetus, expectant management usually results in the birth of a DS child.
2. If the karyotyping results are likely to be concordant (i.e. both are DS fetuses or both are normal fetuses), the decision for prenatal testing and diagnosis is essentially similar to a singleton pregnancy. Hence, the decision for testing in monochorionic twins is fairly straightforward.
3. If the karyotyping results could turn out discordant (i.e. one normal and the other a DS fetus, the couple may face a dilemma:
   (a) If the couple absolutely does not wish to risk losing both fetuses even if one is normal, they may not want to have the screening test.
   (b) If the couple accepts the higher procedure-related miscarriage rate of invasive karyotyping and the possibility of selective fetocide/termination of pregnancies in affected twin pregnancies, they may opt for the DS screening or diagnostic test.
4. Selective reduction of an abnormal fetus in twin pregnancies is associated with a lower proportion with preterm delivery if performed in the first trimester (usually before 14 weeks) than in the second trimester.[2] Hence a strategy that allows karyotyping to be performed in the first trimester may be preferred.
5. Karyotyping in the first trimester by chorionic villus sampling (CVS) is technically more demanding for dichorionic twins and may result in inaccurate sampling.

INVASIVE TESTING IN TWIN PREGNANCIES

Procedural loss rates of amniocentesis in twins reach 2 to 3%.[1,3] In a cohort study involving dichorionic twin pregnancies,[1] the risk of early fetal loss in twins undergoing second trimester amniocentesis (2.73%) was greater than that of exposed singletons (0.60%) and unexposed twins (0.63%). Considerations of amniocentesis in twins should therefore be different from singletons. A higher threshold for invasive karyotyping may be appropriate in twins.

The method of karyotyping in twins depends on the gestational age, chorionicity and the concordance of ultrasound abnormalities in both twins. In our centre, single uterine entry and sampling of both sacs is preferred for dichorionic twins, while sampling of one sac in monochorionic twins is adequate except in those cases with discordant ultrasound findings when double puncture may be preferred. This is to avoid the complications of septostomy including cord accidents that may be more likely in monochorionic twins with thinner intertwin membranes than in dichorionic twins with thicker intertwin membranes.

In the first trimester, CVS may be considered. For dichorionic twins, both fetuses should be karyotyped and attempts should be made to reach the extreme ends of the placentas through a single uterine entry of one needle, or two separate needle insertions are necessary. Operators should be well trained with both the transabdominal and transvaginal routes of CVS.[5-6] In about 5% of cases, there is uncertainty that both placentas have been sampled, especially in cases where the placentas are on the same side of the uterus.[6-7] Hence most centres consider this an unsuitable test for dichorionic twins. In monochorionic twins with concordant ultrasound findings, a single sample by CVS is adequate.

MATERNAL AGE

In singleton pregnancies, the risk of DS may be estimated based on maternal age. In twin pregnancies, the risk of DS according to maternal age is calculated based on singleton data. Calculations by Meyers et al8 worked out that the risk of aneuploidy at amniocentesis in at least one of the twin fetuses is 1/193 in white and African-American women at maternal age 31. This is comparable to the risk of aneuploidy at amniocentesis in singletons at age 35 (1/192) and the authors recommend that amniocentesis should be offered to all twin pregnancies when the maternal age is 31 or above. We do not deem this to be an appropriate practice as DS risk in twins may actually be less3 than that for singletons when the maternal age is standardized and the procedure-related loss rates are much higher for twins than for singletons. However, in the absence of good data, we still use singleton maternal age data to estimate the maternal age related DS risks in twins.

ULTRASOUND SCREENING METHODS

First trimester Nuchal translucency

NT screening involves the sonographic measurement of a translucent space on the neck of the fetus at 11-14 weeks' gestation by trained and accredited individuals. The measurement is increased in a large number of aneuploid fetuses in the first trimester of pregnancy. In singletons, NT measurement is a standard screening test for DS, with high sensitivity (82%) and low false-positive rates (2.5-8%).[9-13] Observational studies11,14 have also shown that the detection rate of NT in twin fetuses for DS are similar to that found in singleton pregnancies.

The preferred screening method for DS in dichorionic

twins should be NT measurement.[11,15-16] Although the detection rate of DS in monochorionic twins with NT is about 80%, similar to that of dichorionic twins, a higher false-positive rate was seen in monochorionic gestations (8.4%) than with dichorionic gestations (5.4%).[14] The higher false-positive rates in monochorionic gestations noted may be secondary to discordant NT measurements in monochorionic pregnancies with early twin-to-twin transfusion syndrome (TTTS). For the estimation of risk in monochorionic twins, there is controversy whether the largest or the smallest measurement or the average of the two should be used.

Second trimester genetic sonogram

A standard second trimester genetic sonogram in singleton pregnancy includes the documentation of the various soft tissue markers which include nuchal fold, echogenic bowel, intracardiac focus (ECF), choroid plexus cyst (CPC), length of the femur and humerus, and renal pyelectasis.

In monochorionic twins, discordance in nuchal fold thickness may be another manifestation of TTTS and hence should be used with caution for estimation of Down syndrome. For echogenic bowel, intracardiac focus, choroid plexus cyst, and renal pyelectasis, one would expect that the likelihood ratios of singletons be applicable for use in both monochorionic and dichorionic twin pregnancies. The intracardiac focus is not a useful soft marker in Asian fetuses. Shortening in humeral and femur lengths may be due to intrauterine growth restriction (IUGR). As both monochorionic and even dichorionic twins have a high rate of IUGR, the use of these latter two soft markers may be associated with high false positive rates.

Nasal bone

NB detection can also be performed at the 11-14 week nuchal translucency scan. Absent or hypoplastic NB has been reported to be one of the strongest soft tissue markers associated with Down Syndrome (DS), as reviewed by Zuzarte et al in a recent paper in this journal.[17] Likelihood ratios of absent NB for DS in first trimester fetuses ranged from 25-50, while ratios of absence of NB in normal karyotype is about 0.2 in various studies involving high risk and low risk populations.[18-22] The NB performs equally well as a soft marker if not better in the second trimester compared to the first trimester as it is technically easier to visualize it in a larger fetus. It has been speculated that the combination of NT and NB could increase the sensitivity of detecting DS from 75 to 93% for a 5% false positive rate.[20] Though this conclusion is from singleton data, one would expect that ultrasound detection of NB to perform equally well in both dichorionic and monochorionic twins.

While the finding of structural anomalies will increase the background risk for DS in such fetuses,[23-24] the NB appears to be the single most powerful soft tissue marker for DS in both first and second trimester ultrasound screening.

SERUM BIOCHEMISTRY MARKERS

Maternal serum screening for DS can be performed in both the first and second trimester. In the first trimester, levels of free beta-hCG and PAPP-A are measured. Various combinations of AFP, free and total hCG, unconjugated estriol and dimeric inhibin A are measured in the second trimester as the double, triple and quadruple test.25

Serum markers in the mother are contributed by two separate fetoplacental units in twins and hence the results derived from maternal serum screening cannot be attributed specifically to any one of the twins. Appropriate adjustments must be made when calculating the risk for DS in twin pregnancies. Both AFP and beta-hCG values were found to be twice as high in the spontaneous twin pregnancies as in the singleton group.[26] Chorionicity[27] does not appear to affect the distribution of free beta-hCG and PAPP-A in unaffected twin pregnancies although miscarriage or intrauterine death of one fetus (increases the inhibin A levels),[28] IVF or spontaneous (IVF twin pregnancies have higher beta-hCG levels,[26] and race are known to cause variations in maternal serum markers.[29] Fraught with such diffculties, the detection rate for DS is only about 40% in twin pregnancies compared to 60% in singletons.[11]

INTEGRATED SCREENING TESTS FOR TWIN PREGNANCIES: NT AND FIRST TRIMESTER MATERNAL SERUM SCREENING

Some studies have shown that integrated screening tests may improve the detection rate of DS in twin pregnancies.[13,25,30] Spencer et al30 reported that the addition of first trimester maternal serum biochemistry to NT increased the detection rate by a further 5-6% from that estimated by NT alone (75.2%). Wald et al13 reported that the estimated detection rates for a 5% false-positive rate of combined testing (NT, free beta-hCG and PAPP-A in the first trimester with maternal age) are 84% for monochorionic twins, 70% for dichorionic twins and 72% for all twins. The integrated test may still offer the most effective method of DS screening in twins though more data is required in this area.

CONCLUSION

A variety of DS screening tests are effective for twin pregnancies. An increase in the threshold for invasive prenatal diagnostic tests may be appropriate as the procedural loss rates associated with karyotyping may

be higher in twins. If a single screening test is to be used for DS risk assessment in twins, an ultrasound scan for NT with or without the NB in the first trimester is preferred to maternal serum screening alone in the first or second trimesters. Integrated tests using NT and first trimester maternal serum screening may further increase the detection rates of DS for twin pregnancies.

REFERENCES

1. *Yukobowich E, Anteby EY, Cohen SM, Lavy Y, Granat M, Yagel S. Risk of fetal loss in twin pregnancies undergoing T2 amniocentesis. Obstet Gynecol 2001; 98: 231-4.*

2. *Yaron Y, Johnson KD, Bryant-Greenwood PK, Kramer RL, Johnson MP, Evans MI. Selective termination and elective reduction in twin pregnancies: 10 years experience at a single centre. Human Reproduction 1998. 13(8): 2301–2304.*

3. *Jamar M, Lemarchal C, Lemaire V, Koulischer L, Bours V. A low rate of trisomy 21 in twin-pregnancies: a cytogenetics retrospective study of 278 cases. Genet Couns. 2003; 14(4):395-400.*

4. *Pruggmayer MR, Jahoda MG, Van der Pol JG, Baumann P, Holzgreve W, Karkut G, et al. Genetic amniocentesis in twin pregnancies: results of a multicenter study of 529 cases. Ultrasound Obstet Gynecol. 1992 1;2(1):6-10.*

5. *De Catte L, Liebaers I, Foulon W. Outcome of twin gestations after •rst trimester chorionic villus sampling. Obstet Gynecol. 2000;96(5 Pt 1):714-20.*

6. *Van den Berg C, Braat APG, Opstal DV, Halley DJJ, Kleijer WJ, Den Hollander NS et al. Amniocentesis or Chorionic Villus Sampling in Multiple Gestations? Experience with 500 Cases. Prenat. Diagn. 1999; 19: 234–244.*

7. *Nicolaides KH, Sebire NJ, Snijders RJM. Multiple pregnancy. In: Nicholaides KH eds. The 11-14-week scan The diagnosis of fetal abnormalities. USA, UK, Europe: The Parthenon Publishing Group Ltd, 1999: 171-173.*

8. *Meyers C, Adam R, Dungan J, Prenger V. Aneuploidy in multiple gestations: When is maternal age advanced? Obstet Gynecol 1997; 89:248-51.*

9. *Snijders RJM, Noble PL, Sebire NJ, Souka A, Nicolaides KH. UK multicentre project on assessment risk of trisomy 21 by maternal age and fetal nuchal translucency thickness at 10-14 weeks of gestation. Lancet 1998; 352: 343-346.*

10. *Spencer K, Souter V, Tul N, Snijders R, Nicolaides KH. A screening program for trisomy 21 at 10-14 weeks using fetal nuchal translucency, maternal serum free beta-human chorionic gonadotropin and pregnancy-associated plasma protein-A. Ultrasound Obstet Gynecol 1999;13(4):231-7.*

11. *Maymon R, Dreazen E, Rozinsky S, Bukovsky I, Weinraub Z, Herman A. Comparison of Nuchal Translucency Measurement and Second-trimester Triple Serum Screening in Twin versus Singleton Pregnancies. Prenat Diagn 1999; 19: 727-731.*

12. *Nicolaides KH, Heath V, Cicero S. Increased fetal nuchal translucency at 11-14 weeks. Prenat Diagn 2002; 22: 302-315.*

13. *Wald NJ, Rish S, Hackshaw AK. Combining nuchal translucency and serum markers in prenatal screening for Down syndrome in twin pregnancies. Prenat Dian 2003; 23: 588-592.*

14. *Sebire NJ, Snijders RJM, Huges K, Sepulveda W, Nicolaides KH. Screening for trisomy 21 in twin pregnancies by maternal age and fetal nuchal translucency thickness at 10-14 weeks of gestation. B J Obstet Gynaecol 1996a; 103: 999-1003.*

15. *Neilson JP. Assessment of fetal nuchal translucency test for Down's syndrome. Lancet 1997; 350: 754–755.*

16. *Cuckle H. Antenatal screening for Down's syndrome. Lancet. 1998; 352(9134): 1144-5.*

17. *Zuzarte R, Tan TYT, Yan YL, Lai FM, Yeo GSH. Absent fetal nasal bone – strongest soft marker for Down syndrome yet? KK Review 2005; 8(2): 27-31.*

18. *Cicero S, Curcio P, Papageorghiou A, Sonek J, Nicolaides K. Absence of nasal bone in fetuses with trisomy 21 at 11-14 weeks of gestation: an observational study. Lancet 2001; 17; 358 (9294): 1665-7.*

19. *Otano L, Aiello H, Igarzabal L, Matayoshi T, Gadow EC. Association between first trimester absence of fetal nasal bone on ultrasound and Down syndrome. Prenat Diagn. 2002; 22(10): 930-2.*

20. *Cicero S, Rembouskos G, Vandecruys H, Hogg M, Nicolaides KH. Likelihood ratio for trisomy 21 in fetuses with absent nasal bone at the 11-14-week scan.Ultrasound Obstet Gynecol. 2004; 23(3): 218-23.*

21. *Orlandi F, Bilardo CM, Campogrande M, Krantz D, Hallahan T, Rossi C, Viora E. Measurement of nasal bone length at 11-14 weeks of pregnancy and its potential role in Down syndrome risk assessment. Ultrasound Obstet Gynecol. 2003; 22(1): 36-9.*

22. *Zoppi MA, Ibba RM, Axiana C, Floris M, Manca F, Monni G. 2003. Absence of fetal nasal bone and aneuploidies at first-trimester nuchal translucency screening in unselected pregnancies. Prenat Diagn. 2003 Jun;23(6):496-500.*

.

23. *Bromley B, Lieberman E, Shipp TD, Benacceraf BR. The Genetic Sonogram A method of risk assessment for Down syndrome in the second trimester. J Ultrasound 2002; Med 21: 1087-1096.*

24. *Devore GR. Trisomy 21: 91% detection rate using second-trimester ultrasound markers. Ultrasound Obstet Gynaecol 2000; 16: 133-141.*

25. *Odibo AO, Lawrence-Cleary K, Macones GA. Screening for Aneuploidy in Twins and Higher-Order Multiples: Is First-trimester Nuchal Translucency the Solution? Obstet and Gynae Survey 2003; 58(9): 609-614.*

26. *Raty R, Virtanen A, Koskinen P, Laitinen P, Forsstrom J, Salonen R et al. Maternal midtrimester serum AFP and free b-hCG levels in in vitro fertilization twin pregnancies. Prenat Diagn 2000; 20: 221-223.*

27. *Spencer K. Screening for trisomy 21 in twin pregnancies in the first trimester: does chorionicity impact on maternal serum free beta-hCG or PAPP-A levels? Prenat Diagn 2001; 21: 715–717.*

28. *Goodwin KM, Sweeney PJ, Lambert-Messerlian GM, Canick JA. High maternal serum inhibin A levels following the loss of one fetus in a twin pregnancy. Prenat Diagn 2000; 20: 1015-1017.*

29. *O'Brien JE, Dvorin E, Yaron Y, Ayoub M, Johnson MP, Hume Jr RF, Evans MI. Differential Increases in AFP, hCG, and uE3 in Twin Pregnancies: Impact on Attempts to Quantify Down Syndrome Screening Calculations. American Journal of Medical Genetics 1997; 73: 109–112.*

30. *Spencer K. Screening for trisomy 21 in twin pregnancies in the first trimester using free beta-hCG and PAPP-A, combined with fetal nuchal translucency thickness. Prenat Diagn 2000; 20: 91-95.*

Editorial Note: There is higher procedural loss rates associated with karyotyping may be higher in twins. An ultrasound scan for NT in the first trimester is preferred to maternal serum screening alone in the first or second trimesters. Integrated tests using NT and first trimester maternal serum screening may further increase the detection rates of Down Syndrome for twin pregnancies.

Reprinted from KK Hospital Review: Special Issue on Antenatal Screening. May 2006; 9(01) with permission from Editor.