VCGS Leaders in Genetic Health

the partumpost

NIPT: twins, triplets & co-demise

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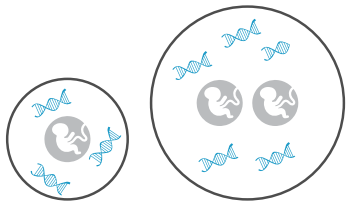
Why screening in twin pregnancies is challenging

Cell-free (cf)DNA screening in twin pregnancies presents unique challenges. Although the total fetal fraction in twins is approximately 1.6 times that reported in singletons, the average fetal fraction per twin is decreased (1). This makes sensitive screening in twins, which represent 3% of all live births, problematic (2). Additionally, some NIPT methodologies are not capable of screening twin pregnancies.

Sensitive screening in twins is challenging: while total fetal fraction is higher, fetal fraction per twin is lower.

Placental volume, chorionicity & fetal fraction

Fetal fraction is known to increase with placental size. Total placental volumes and weights in twin pregnancies are approximately 1.6 to 1.7 times that of singletons (3, 4). These values are reflected in the observed 1.6 fold increase in fetal fraction.



fetal fraction for each twin is lower when compared with singleton pregnancies

Monochorionic (MC) and dichorionic (DC) placentas have comparable total volumes and weights. Theoretically, the sensitivity for trisomy screening in MC twins should be as good as or better than screening in singletons.

This is because MC twins are almost certainly genetically identical and the combined fetal fraction (1.6x singletons) is representative of both twins. In contrast, the sensitivity for DC (usually genetically non-identical) twins may be reduced because the average fetal fraction per twin is 0.8x that of singletons.

When a trisomy is present in DC twins, usually only one twin is affected. Therefore, NIPT must be sufficiently sensitive to detect this trisomy at lower than average fetal fractions (1-3, 5).

One recent meta-analysis study found sensitivity for trisomy 21 and trisomy 18 in twin pregnancies to be reduced by 9% and 22% respectively, although the dataset used for the twin analysis was not large (2).

NIPT failure in twin pregnancies

For NIPT methods that require at least 4% fetal fraction for sensitive screening, test failure rates in twins can be as high as 13.2% (6). One large prospective study of over 400 twin pregnancies had a test failure rate at first blood draw of 9.3%, increasing to 56.2% in IVF conceived twins (7). Even on second blood draw the failure rate remained above 5%.

As percept NIPT is highly sensitive at low fetal fractions, it's ideal for screening twins, with very low test failure.

VCGS has screened 464 twin pregnancies to December 2016, with only 1.6% requiring a second blood draw. Four pregnancies with trisomy have been identified (two MCDA; two DCDA).

Test sensitivity for trisomy screening in twins using *percept* NIPT is 100% with a false positive rate of just 0.2% (1/464) and no known false negatives. All women requiring a redraw received an informative result. This is consistent with the high success rate reported in a recent publication using a similar method of NIPT (8).

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NIPT screening in triplet pregnancies

VCGS is the only NIPT provider in Australia that offers screening in triplet pregnancies. We are committed to providing these women with screening options where few have previously existed. Seven low risk pregnancies have been successfully screened to date (Figure 1).

Please contact VCGS prior to blood draw if you are caring for a woman with a triplet pregnancy, as prior approval is required. Screening is available from 12 weeks of gestation.

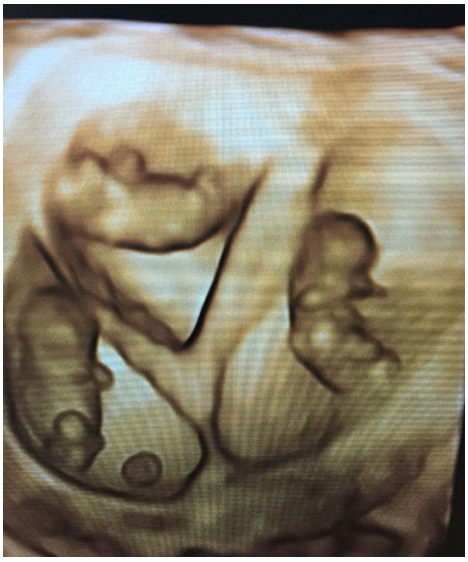

Figure 1. Trichorionic triamniotic (TCTA) triplet pregnancy screened using percept NIPT. Ultrasound image courtesy of Dr Emily Olive, with patient consent.

Co-twin demise & NIPT

Co-twin demise is usually a contraindication for NIPT. It is problematic because the placental trophoblast from the demised twin continues to release cfDNA into the maternal circulation (9). This can lead to a false positive result if the demised twin carries a chromosome condition being screened for. Co-twin demise may also lead to misreporting of the fetal sex, when the demised twin is male and the live twin is female (10).

VCGS has screened many pregnancies with ultrasound evidence for a demised twin. By retesting pregnancies over many weeks we have monitored the fall in fetal fraction from the demised twin, thereby enabling accurate screening in the live twin.

The data collected now informs our advice on the management of these pregnancies. The gestation at demise is important for uncomplicated screening and becomes more difficult after 8 weeks of pregnancy.

We recommend contacting our laboratory if there is evidence for a twin demise. We can then provide specific advice on the best gestation to undertake screening.

Please note this advice is not applicable to other providers of NIPT as screening requires the application of additional bioinformatics.

Please email vcgs@vcgs.org.au for any questions, topic suggestions or to subscribe to the partum**post**.

References

1. Y. Zhou, Z. Zhu, Y. Gao, Y. Yuan, Y. Guo, L. Zhou, K. Liao, J. Wang, B. Du, Y. Hou, Z. Chen, F. Chen, H. Zhang, C. Yu, L. Zhao, T. K. Lau, F. Jiang, W. Wang, Effects of Maternal and Fetal Characteristics on Cell-Free Fetal DNA Fraction in Maternal Plasma. Reproductive Sciences 22, 1429-1435 (2015).
2. S. Taylor-Phillips, K. Freeman, J. Geppert, A. Agbebiyi, O. A. Uthman, J. Madan, A. Clarke, S. Quenby, A. Clarke, Accuracy of non-in vasive prenatal testing using cell-free DNA for detection of Down, Edwards and Patau syndromes: a systematic review and meta-analysis. BMJ Open 6, e010002 (2016).
3. P. Wegrzyn, C. Fabio, A. Peralta, C. Faro, M. Borenstein, K. H. Nicolaides, Placental volume in twin and triplet pregnancies measured by three-dimensional ultrasound at 11 + 0 to 13 + 6 weeks of gestation. Ultrasound in Obstetrics & Gynecology 27, 647-651 (2006).
4. H. Pinar, C. J. Sung, C. E. Oyer, D. B. Singer, Reference Values for Singleton and Twin Placental Weights. Pediatric Pathology & Laboratory Medicine 16, 901-907 (2010).
5. M. D. Pertile, T. Charles, T. Burgess, Fetal fraction facts. partum**post** 1 (1), (2017).
6. M. del Mar Gil, M. S. Quezada, B. Bregant, A. Syngelaki, K. H. Nicolaides, Cell-free DNA analysis for trisomy risk assessment in first-trimester twin pregnancies. Fetal Diagnosis and Therapy 35, 204-211 (2014).
7. L. Sarno, R. Revello, E. Hanson, R. Akolekar, K. H. Nicolaides, Prospective first-trimester screening for trisomies by cell-free DNA testing of maternal blood in twin pregnancy. Ultrasound in Obstetrics & Gynecology 47, 705-711 (2016).
8. L. Fosler, P. Winters, K. W. Jones, K. J. Curnow, A. J. Sehnert, S. Bhatt, L. D. Platt, Aneuploidy screening by non-invasive prenatal testing in twin pregnancy. Ultrasound in Obstetrics & Gynecology 49, 470-477 (2017).
9. K. J. Curnow, L. Wilkins-Haug, A. Ryan, E. Kirkizlar, M. Stosic, M. P. Hall, S. Sigurjonsson, Z. Demko, M. Rabinowitz, S. J. Gross, Detection of triploid, molar, and vanishing twin pregnancies by a single-nucleotide polymorphism-based noninvasive prenatal test. American Journal of Obstetrics and Gynecology 212, 79 e71-79 (2015).
10. B. Vlkova, J. Hodosy, Vanishing twin as a potential source of bias in non-invasive fetal sex determination: a case report. The Journal of Obstetrics and Gynaecology Research 40, 1128-1131 (2014).