INTRODUCTION

The implementation of chromosomal microarray (CMA) technology in the clinical prenatal setting has achieved consensus for use in cases with fetal defects in ultrasound examinations. The expansion of CMA as a standard examination for all patients undergoing invasive procedures has not achieved the same level of consensus. The main arguments against it include (1) counseling challenges related to the identification of so-called ‘variants of unknown significance’ (VUS) or with inconclusive lower penetrance and (2) costs involved, especially when weighed against the relatively smaller benefit for ‘lower risk’ indications, where healthcare is mainly funded by the government.

Lower cost testing alternatives, although less comprehensive but less challenging, have thus been proposed as “all-in-one” tests with “lower risk” indication. Prenatal BAcS-on-Beads™ (PNBoBs™) is a new emerging targeted isolation-based technology used to detect rapid DNA copy number gains and losses. PNBoBs™ is a well suited alternative for rapid FISH or QF-PCR as, unlike those technologies, it also has the ability to detect common microdeletions aside of the common trisomies. The microdeletion regions reported by PNBoBs are recurrent dominant and relatively well-characterized microdeletion syndromes which may be easily missed by prenatal cytogenetics, each with reasonably well-known genotype-phenotype correlation and associated with relatively minor challenges in terms of genetic counseling.

AIM OF THE STUDY

Comparison of the diagnostic performance of PNBoBs™ and QF-PCR assays for prenatal detection of chromosome abnormalities.

MATERIALS AND METHODS

- Description study based on a retrospective anaemized cohort (May 2015 – December 2013)
- IRB approved: TomaBiome® IRB protocol 00000005
- All patients gave consent for the analyses
- 2784 (heuristic (CH) and maternal cell-free DNA samples were prospectively analyzed with conventional karyotyping and PNBoBs™
- Fetal tissue analysis and PNBoBs™ analyses were performed on 1287 samples
- Classification of samples
  1. Classification by indication for prenatal diagnosis
     - True positive results: diagnostic findings (risk cell free DNA testing result (US-LR)
     - low risk population group could not be detected by QF-PCR.
     - high risk cell free DNA testing result (US-LR)
     - abnormal ultrasound (US) (subdivided into: low risk (US-LR), high risk for submicroscopic copy number abnormalities (CNV) (US-HR) and fetal defect not detected (US-LR/GA))

- Other considerations
  - Low a priori risk for CNV: AMA, IMSS-DS, MA, PFA, US-LR and Other
  - High a priori risk for CNV: PCCA, MS, US-HR, US-Unk and Unknown
  - Other indications (Other)
  - Parent carrier of a chromosome abnormality (PCCA)
  - Maternal anxiety (MA)
  - Positive ultrasound (US) 
    - diagnosis of congenital anomalies
    - fetal defect not reported
    - abnormal ultrasound (US) subdivided into: low risk (US-LR), high risk for submicroscopic copy number abnormalities (CNV) (US-HR)

- Diagnostic yield and performance of PNBoBs™ and QF-PCR

- PNBoBs™ for prenatal diagnosis
  - shows a higher diagnostic yield compared to QF-PCR in each indication due to the additional detection ability of microdeletion/applications (Table 2). 11 abnormal cases detected by PNBoBs™ in the low risk and 1 case in the high risk prenatal population group could not be detected by QF-PCR. These included cases 1-3.

CONCLUSIONS

Although PNBoBs™ may not have the breadth and scope to replace chromosomal microarrays in the prenatal setting, particularly when a fetal anomaly has been detected by US analysis, it is a well suited alternative for QF-PCR as PNBoBs™ is superior in terms of sensitivity for a wider set of chromosome abnormalities in all indications, specifically for those pregnancies without a high risk chromosome abnormality risk cell free DNA testing result (US-LR) and who were not referred for a balanced risk of trisomy (increased MCH for DS, AMA and Anxiety >75) and in which, in some countries, QF-PCR is used as a stand-alone test (without karyotype).

RESULTS

- For the low risk cell free DNA testing result (US-LR), the diagnostic yield was reported as 27.5% (95% CI: 23.29-32.84) for karyotype, 24.6% (95% CI: 17.51-33.34) for PNBoBs™ and 20.2% for QF-PCR (95% CI: 15.75-25.62) (Table 1).
- For high risk cell free DNA testing result (US-HR), this was equated to 24.6% (95% CI: 17.51-33.34), 15.2% (95% CI: 11.02-20.10), and 15.2% (95% CI: 11.02-20.10) respectively (Table 1).

Figure 2 - Case 4 results: A) abnormal cell line carrying a dic(22;22)(q11.2;q11.2) chromosome abnormality. B) arrayCGH results: arr[hg19],D8S2333+.ish 4p16.3(WHSC1,WHSC2) t(4;8)(p15.32;p22)dn.ish der(4)t(4;8)

Figure 3 - Case 5 results: A) karyotype on AF: 46,XY,der(4) t(4;8)(p15.32;p22)dn.ish der(4)t(4;8). B) arrayCGH results: arr[hg19],D8S2333+.ish 4p16.3(WHSC1,WHSC2) t(4;8)(p15.32;p22)dn.ish der(4)t(4;8)

Figure 4 - Case 6 results: A) abnormal cell line carrying a dic(22;22)(q11.2;q11.2) chromosome abnormality. B) arrayCGH results: arr[hg19],D8S2333+.ish 4p16.3(WHSC1,WHSC2) t(4;8)(p15.32;p22)dn.ish der(4)t(4;8)

REFERENCES

1. Caldas A, Gravina E, D'Urso E, et al. The clinical utility of microarray technologies in the prenatal setting, particularly when a fetal anomaly has been detected by US analysis, it is a well suited alternative for QF-PCR as PNBoBs™ is superior in terms of sensitivity for a wider set of chromosome abnormalities in all indications, specifically for those pregnancies without a high risk chromosome abnormality risk cell free DNA testing result (US-LR) and who were not referred for a balanced risk of trisomy (increased MCH for DS, AMA and Anxiety >75) and in which, in some countries, QF-PCR is used as a stand-alone test (without karyotype).

POSTER LEGEND

Ultrasound findings (US), abnormal maternal age (AMA), increased maternal serum screening for Down syndrome (MSS-DS), high risk cell free DNA testing result (US-LR), maternal anxiety (MA), microarray/invasive prenatal test (MIP), previous fetal history (PFA) and a monosomy X cell line (US-LR) are the major factors that enhance the risk of CNV aneuploidy.

Acknowledgments: Stefano Di Stefano, et al. The clinical utility of microarray technologies in the prenatal setting, particularly when a fetal anomaly has been detected by US analysis, it is a well suited alternative for QF-PCR as PNBoBs™ is superior in terms of sensitivity for a wider set of chromosome abnormalities in all indications, specifically for those pregnancies without a high risk chromosome abnormality risk cell free DNA testing result (US-LR) and who were not referred for a balanced risk of trisomy (increased MCH for DS, AMA and Anxiety >75) and in which, in some countries, QF-PCR is used as a stand-alone test (without karyotype).

TOMA ADVANCED BIOMEDICAL ASSAYS S.p.A. • Via F. Ferrer 25/27 • 20152 Busto Arsizio (VA) Italy • Tel +39 0331 652911 • Fax +39 0331 652919 • www.tomalab.com