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Clinical outcomes of genome-wide cell-free DNA (cfDNA) for cases screening positive for trisomy 9

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I. Objective

While cell-free DNA (cfDNA) testing for common aneuploidies has been integrated into routine prenatal care, expanded cfDNA content including select microdeletions, large copy number variants, and esoteric aneuploidies has not been routinized. Initial data regarding outcomes from a commercial genome-wide cfDNA test has been described.[1,2,3] The objective is to describe the clinical outcomes observed specifically for cases screening positive for trisomy 9 (T9) on genome-wide cfDNA.

II. Study Design

A retrospective analysis was performed on over 50,000 maternal blood samples submitted for MaterniT® GENOME (genome-wide cfDNA analysis) at Sequenom Laboratories®. Samples were subjected to DNA extraction, library preparation, and genome-wide massively parallel sequencing.[4] Sequencing data were analyzed using a novel algorithm to detect trisomies and subchromosomal, genome-wide copy number variants 7Mb and larger.[1] The cases that screened positive for trisomy 9 were reviewed. Clinical outcomes were requested from ordering providers as part of routine follow-up of positive cases.

III. Results

Sixteen cases screened positive for full trisomy 9, or overrepresentation of the entire length of chromosome 9. Cases that were positive for partial trisomies or segmental trisomies, such as a duplication of the entire p arm of chromosome 9 (e.g. isochromosome), were excluded from this analysis.

Mosaicism ratio is utilized by the laboratory to determine cases which have sequencing data suggestive of mosaicism.[5] Mosaicism ratio is calculated by dividing the fetal fraction estimated for the abnormal event (aneuploid chromosome) by the fetal fraction estimated across all chromosomes. For esoteric aneuploidies, a case with a mosaicism ratio between approximately 0.2 and 0.7 may garner a mosaic comment on the report. All but one of the discordant cases tended to have a lower mosaicism ratio (**Figure 3**, red dots); the outlying discordant case is an ongoing pregnancy, so final obstetric outcome is still unknown.

The most common indication for testing was maternal age, followed by a relatively even divide between positive serum screening and ultrasound finding.

Figure 1:
Distribution of indications for testing for cases screeing postive for trisomy 9.

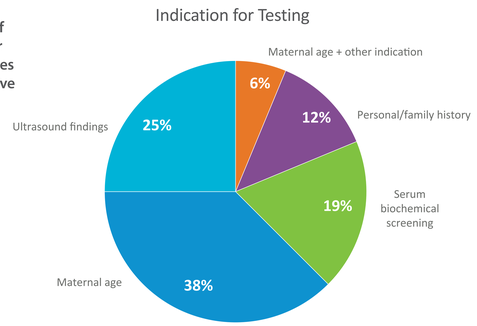


Figure 2:
Adverse clinical outcomes were defined as growth restriction, preterm labor/delivery, miscarriage (SAB) or fetal demise (IUFD), or ultrasound anomalies (not including soft markers for aneuploidy).

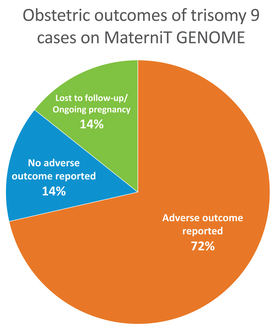
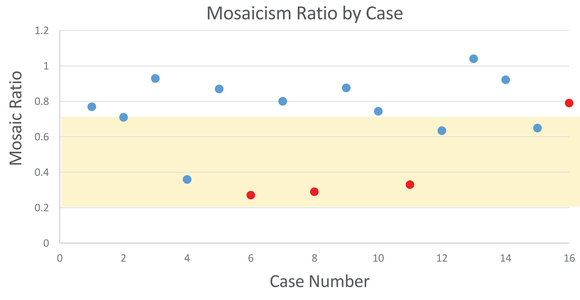


Table 1:
Genetic outcomes of cases receiving a positive result for trisomy 9 on MaterniT GENOME.

	Total number of positive T9 cases on MaterniT GENOME	Confirmed in fetus or placenta	Miscarriage or fetal demise with no testing	Discordant diagnostic testing	Lost to follow-up or ongoing pregnancy
Number of cases	16	7 (43.75%) (6 fetal, 1 confined placental mosaicism)	4 (25%)	4 (25%)	1 (6.25%)

Figure 3:
Distribution of mosaicism ratio by case. Red dots indicate discordant cases. The yellow box signifies the range (0.2-0.7) utilized for including mosaicism comments on the test report by Sequenom Laboratories®.



IV. Conclusion

Mosaicism ratio may give insight into cases that are likely to have a lower positive predictive value[5] but this series also shows that the mosaicism ratio may be helpful in ascertaining the clinical impact, including likelihood of discordant results. While this cohort is too small to draw conclusions about the utility of mosaicism ratio to predict adverse outcomes, it is certainly an area for additional study.

A positive result for trisomy on cfDNA may carry residual risk for fetal aneuploidy (full or mosaic), confined placental mosaicism, and adverse obstetric outcome. Other biological explanations, such as cotwin demise or maternal events might also be considered, although not represented in this cohort to our knowledge nor overtly suggested by the sequencing data.

Adverse outcomes have been reported in other cohorts of trisomy 9 cfDNA cases.[6,7] While the number of cases in this cohort is small, it is worth noting that nearly half (43.75%) of the cases were confirmed in the fetus (n=6) or the placenta (n=1). Another 25% of the cases ended in pregnancy loss without additional testing, raising strong suspicion that the trisomy 9 event exists in the fetus or placenta. Cases of both full and mosaic trisomy have been reported in livebirths, often with significant structural anomalies.[8] Mosaic trisomy 9 ascertained at amniocentesis is associated with a significant risk for structural anomalies, growth restriction, or abnormal outcome at birth.[9] Discrepancies in cytogenetic results between different tissues, such as amniotic fluid and postnatal blood, may complicate prenatal and postnatal diagnosis of this condition.[10] Because of the demonstrated risk and phenotype, providers may find clinical utility in screening for trisomy 9, especially in patients with high risk indications. Consideration of the potential outcomes will be an important counseling tool for providers and patients alike.

V. References

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