I. Introduction

Since the introduction of the first commercially available cell-free DNA screening for Down syndrome in 2011, the content available to patients has expanded to include trisomy 18, trisomy 13, fetal sex and sex chromosome aneuploidies, and select microdysgenosis syndromes. Most recently, this technology has expanded to allow for genome-wide screening for any aneuploidy and sub-chromosomal events 7 Mb and larger. Genome-wide cfDNA analysis has allowed for more clinically relevant information through screening, but occasionally such results can have additional important implications for the pregnant woman, her partner, or for the fetus. This case series examines some of the complex possible results and important points raised by these cases.

II. Methods

Maternal blood samples submitted to Sequenom Laboratories® for MaterniT®-GENOME testing were subjected to DNA extraction, library preparation, and genome-wide massively parallel sequencing as described by Jensen et al. Sequencing data were analyzed using a novel algorithm to detect trisomies and sub-chromosomal, genome-wide events 7Mb and larger, as described by Lefkowitz et al. Follow-up information and pregnancy outcomes were elicited from the clinicians as part of routine, ongoing laboratory protocol for positive cases.

III. Cases

Case 1: A primagravida woman in her mid-twenties was referred to maternal fetal medicine at 21+3 weeks due to anomalies identified on a routine anomaly scan with her obstetrician. Her ultrasound at maternal fetal medicine showed left renal agenesis, micrognathia, hypoplastic cerebellum, and generous cerebral ventricles. After declining amniocentesis, MaterniT®-GENOME was ordered by a genetic counselor who showed an approximately 60 Mb duplication of 4q13.32-q21.2. Follow-up ultrasound at 26+2 weeks showed growth restriction. A fetal echocardiogram was also performed at this time due to concern for a fetal heart defect but the cardiac anatomy was determined to be normal. After counseling regarding the cfDNA results and the ultrasound findings, the patient elected to forego amniocentesis and proceeded with amniocentesis. Karyotyping was 46,XY,der(Y)t(Y;4)(p11.32;q25), indicating that the duplicated piece of chromosome 4 was on the terminal end of the Y chromosome with the Y chromosome apparently intact. Microarray results confirmed the 60.4 Mb duplication of 4q13.32-q21.2 indicative of an unbalanced reciprocal translocation along with a common benign chromosome 9 inversion. Microarray confirmed the 65.5 Mb duplication of 8q21.33p24.3. Neither microarray nor MaterniT®-GENOME suggested chromosome 15 imbalance, as the implicated segment of chromosome 15 is comprised of non-coding p arm satellite material. Maternal microarray was subsequently ordered and confirmed a 19.9 Mb duplication of 8q11.21-q21.33, which was classified as a variant of uncertain significance that was likely benign. Two OMIM genes are present in the region, HEY1 and ZFAND1. While the patient herself was phenotypically normal, her other child had questionable developmental delays, familial history was otherwise unremarkable as reported by the patient. Parental karyotyping were not completed but were inferred to be normal. Delivery was induced at 38+2 weeks due to fetal anomalies. Additional neonatal findings included hypertelorism, low-set ears, severe retinomicrophthalmia, undescended testes, hypertonia, a sacral dimple, long and broad big toes, and feeding dysfunction requiring G-tube placement. The child spent nearly 2 months in the NICU and was placed for adoption after birth. No additional follow-up information is available for the patient or the infant after discharge from the hospital.

Case 2: A G2P1 woman in her early 20s was referred to maternal fetal medicine at 24 weeks due to ventriculomegaly and a single umbilical artery identified on routine ultrasound. Ultrasound at maternal fetal medicine confirmed ventriculomegaly and the single umbilical artery and also identified a bilateral clift lip and palate, growth restriction, and hypoplastic left heart. Cardiac views were poor and she was scheduled for a fetal echo. She met with a genetic counselor, at which time she declined amniocentesis. She accepted MaterniT®-GENOME, which reported a 63.70 Mb duplication of 8q, and discussion with the laboratory indicated concern for an adjacent maternal duplication of 1.95 Mb. Although allowed to choose the best diagnostic tool for her, the patient did not want to pursue amniocentesis. After declining amniocentesis, MaterniT®-GENOME was ordered by a genetic counselor which showed an 80.4 Mb duplication of 4q25q35.2. No loss of any Y material was noted on microarray. Paternal chromosomes were ordered and were normal. Non-paternity was denied. Unfortunately, the pregnancy ended in an IUFD at the beginning of the third trimester.

Case 3: A 30-year-old G3P2 was referred to maternal fetal medicine at 23+1 weeks for a level II ultrasound after routine ultrasound showed questionable enlarged lateral ventricles in the brain and poor views of the cardiac anatomy. Her ultrasound scan at maternal fetal medicine showed a significantly abnormal facial profile with severe frontonasal bossing and mediolateral hypoplasia in a male fetus. The patient was referred to maternal fetal medicine, where the patient attended the maternal appointment with her partner. Ultrasound also noted minimal bilateral cranial vault uromegaly and the cardiac anatomy was not well visualized due to maternal body habitus. A subsequent fetal echocardiogram at 25 weeks was normal. The patient received genetic counseling and testing options were discussed. The declined amniocentesis but accepted MaterniT®-GENOME, which reported a 19.9 Mb deletion of 8p12.11p23.1. When the patient returned for her 20-week ultrasound, the karyotype was normal. When paternal karyotype was normal, the clinician had a conversation with the family about non-paternity. Although non-paternity was denied, unbalanced, non-reciprocal Y-autosome translocations are rare and recurrence risk would be significant if the Y-autosome translocation were inherited from a man carrying a balanced arrangement. The clinical and psychosocial implications of the fetal results impacted how the provider managed the follow-up in this case.

IV. Discussion

In case 1, the abnormality identified by MaterniT®-GENOME prompted the provider to order a karyotype in addition to microarray when the patient wished to pursue diagnostic testing. When the diagnostic testing showed involvement of the Y chromosome, the finding directed the clinician to only pursue paternal karyotype when insurance and logistical issues might have otherwise prompted starting with maternally derived karyotype. When paternal karyotype was normal, the clinician had a conversation with the family about non-paternity. Although non-paternity was denied, unbalanced, non-reciprocal Y-autosome translocations are rare and recurrence risk would be significant if the Y-autosome translocation were inherited from a man carrying a balanced arrangement. The clinical and psychosocial implications of the fetal results impacted how the provider managed the follow-up in this case.

In case 2, the cfDNA results directed additional testing for the pregnant woman, providing important information about recurrence risk for future pregnancy.

In case 3, what initially appeared to be a simple deletion ended up being part of a very complicated result. Given the abnormal phenotype in another family member, the provider did discuss testing for both the paternal and maternal karyotype. When maternal karyotype was normal, the provider did not have a conversation with the family about non-paternity. Although non-paternity was denied, unbalanced, non-reciprocal Y-autosome translocations are rare and recurrence risk would be significant if the Y-autosome translocation were inherited from a man carrying a balanced arrangement. The clinical and psychosocial implications of the fetal results impacted how the provider managed the follow-up in this case.

V. References