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Original Article

Single nucleotide polymorphism array in genetic evaluation of fetal ultrasound abnormalities: a retrospective follow-up study

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Abstract: Fetal ultrasound abnormalities may be complicated by cognitive dysfunction or developmental retardation, and ultrasonography cannot detect these problems; therefore, chromosome detection is required in fetuses with ultrasound abnormalities. To examine the effectiveness of single nucleotide polymorphism (SNP) array in genetic diagnosis of fetal ultrasound abnormalities, the prenatal samples of 805 pregnant women with fetal ultrasound abnormalities were collected for SNP array and karyotyping analysis. A 95.5% percentage of normal karyotypes and 4.5% percentage of abnormal karyotypes were observed, and aneuploidy was detected in 28 fetuses with abnormal karyotypes. SNP array identified 89 positives, including 55 cases (6.8%) with pathogenic copy number variation (CNVs) and 34 (4.2%) with variants of unknown significance (VOUS). In addition to 36 cases showing consistent results with karyotyping, SNP array detected 19 additional cases with pathogenic CNVs, including microdeletion/microduplication syndromes in 18 cases and uniparental disomy in one case. The detection rate of pathogenic CNVs was highest in fetuses with structural abnormalities of multiple systems complicated by non-structural abnormalities (13.7%) and lowest in those with structural abnormalities of a single system (4.2%). Presence of pathogenic CNVs was 12.2% in fetuses with structural abnormalities in the urinary system, followed by in the skeletal system (10.3%), while no pathogenic CNVs were identified in fetuses with structural abnormalities in the head and face, the respiratory system or the digestive system. An 89.6% follow-up rate was seen in the study sample, and 55 fetuses with pathogenic CNVs identified by SNP array were all given induction of labor. Our data demonstrate that SNP array improves the detection of genetics aberrations in fetuses with prenatal ultrasound abnormality relative to karyotyping.

Keywords: Single nucleotide polymorphism array, prenatal diagnosis, prenatal ultrasound abnormality, copy number variation

Introduction

Birth defects are structural, functional, or metabolic abnormalities that occur at birth and are caused by environmental or genetic factors, or a combination of the two, or other undiscovered factors [1]. In China, birth defects are highly prevalent, with a prevalence rate of approximately 5.6%, with an estimated 900 thousand new cases each year [2]. Chromosome abnormality, a leading cause of birth defects, occurs in 0.5% to 0.8% of all birth defects [3]. Since fetal ultrasound abnormalities caused by variation of chromosomes may be complicated by cognitive dysfunction or developmental retardation, and ultrasonography cannot detect these problems, chromosome detection is required in fetuses with ultrasound abnormalities [4-6].

Currently, G-band karyotyping is the gold standard for cytogenetic prenatal diagnosis; however, this method has limitations such as a long period of cell culture, low resolution, and high human resources consumption [7]. Chromosomal microarray analysis (CMA) is effective to detect the copy number of variations (CNVs) of chromosome imbalance, which has a prominent advantage of detecting chromosome microdeletions and microduplications [8]. For

high-throughput, high-resolution, and high-automatic detection, CMA has been proven superior to karyotyping [9]. According to the design of microarrays and the principle of detection, CMA is classified into array-based comparative genomic hybridization (aCGH) and single nucleotide polymorphism array (SNP array) [10]. In addition to CNVs, SNP array also is effective to detect uniparental disomy (UPD), triploidy, and chimera [11].

In 2013, the American College of Obstetricians and Gynecologists (ACOG) recommended the replacement of G-band karyotyping by CMA if ultrasound examinations identified fetal structural anomalies, while both CMA and karyotyping were recommended for prenatal diagnosis among fetuses without structural anomalies [12]. In addition, fetal ultrasound abnormality is considered as an indication for CMA testing, as proposed in the national guidelines for the application of CMA in prenatal diagnosis in China [13]. The purposes of this study were to examine the effectiveness of SNP array in genetic diagnosis of different fetal ultrasound abnormalities and unravel the associations of ultrasonographic findings with chromosomal abnormalities.

Methods

Subjects

A total of 805 pregnant women with prenatal diagnosis of fetal ultrasound abnormalities at the Medical Genetic Diagnosis and Therapy Center, Fujian Maternity and Child Health Hospital during the period from May 2015 through December 2018 were recruited. The pregnant women had a mean age of 30 ± 4.8 years (range, 16 to 45 years) and a mean gestational age of 24.9 ± 4 weeks (range, 17 to 35 + 6 weeks). All pregnant women were classified into 4 groups according to the ultrasonographic findings and number of systems involved. Subjects with a single or multiple structural abnormalities of a single system and without non-structural abnormalities were assigned into Group A (n = 383), subjects with a single or multiple structural abnormalities of multiple systems and without non-structural abnormalities were assigned into Group B (n = 52), and subjects with a single or multiple structural abnormalities of a single system and with non-structural abnormalities were assigned into Group C (n = 319), while subjects with a single or multiple structural abnormalities of multiple systems and without non-structural abnormalities were assigned into Group D (n = 51). The subjects included 93 cases with structural abnormalities of the nervous system, 32 cases with structural abnormalities of the head and face, 24 cases with structural abnormalities of the respiratory system, 466 cases with structural abnormalities of the cardiovascular system, 67 cases with structural abnormalities of the digestive system, 74 cases with structural abnormalities of the urinary system, 29 cases with structural abnormalities of the skeletal system and 20 cases with other structural abnormalities (fetal edema, tumor and structural abnormality of the abdominal wall).

Karyotype analysis

Approximately 30 ml of amniotic fluid specimens were collected via B-mode ultrasound-guided abdominal puncture, with 10 ml used for SNP array and 20 ml for cell culture and karyotype analysis. 2.5 ml of umbilical cord blood samples were collected through B-mode ultrasound-guided cordocentesis, with 1 ml used for SNP array and 1.5 ml for cell culture and karyotype analysis. In addition, 2 ml of peripheral blood was sampled from all pregnant women and their spouses and transferred to EDTA-anticoagulated tubes. All prenatal samples were routinely cultured, mounted on slides and subjected to G-banding. Karyotype analysis was performed on a GSL-120 Streamlines Cytogenetic Analysis System (Leica Microsystems; Mannheim, Germany). At least 40 metaphases were counted for each case, and 5 karyotypes were randomly selected for analysis. The results were interpreted according to the 2016 International System for human Cytogenomic Nomenclature (ISCN) [14].

SNP array

SNP array was strictly performed following the standard operating procedures provided by Affymetrix (Affymetrix; Santa Clara, CA, USA). Briefly, amniotic fluid was sampled, and genomic DNA was extracted from amniotic fluid cells using the QIAamp DNA Blood Mini Kit (Qiagen, Hilden, Germany), digested, amplified, purified, fragmented, labeled and hybridized to the array on the Affymetrix SNP Array 6.0 (Affymetrix; Santa Clara, CA, USA). The CytoScan HD array, including the CNV probe and SNP probe, may

detect CNV, mosaic (mosaic proportion > 10%) and loss of heterozygosity (LOH). All data analyses were performed using the software Chromosome Analysis Suite (ChAS) version 3.2 (Affymetrix; Santa Clara, CA, USA), All nucleotide positions refer to the Human Genome Feb 2009 Assembly (GRCh37/hg19). The interpretation of CNV, which was classified as pathogenic, variants of uncertain significance (VOUS) and benign, was identified using online public databases, including the database of genomic variants (DGV, http://projects.tcag.ca/variation), the DECIPHER database (htts://decinher.sanger.ac.uk/), the OMIM database (http://www.omim.org), the International Standards for Cytogenomic Arrays (ISCA) Consortium and Public Database (https://www.iscaconsortium.org/), the CAGdb database (http://www.cagdb.org/), the CHDWiki database and the NCBI database. For fetuses identified with VOUS, parental testing was recommended for pregnant women and their spouses. If the CNV was inherited from parents with a normal phenotype, VOUS (possibly benign) was defined, and if *de novo* mutations were detected, VOUS (unclear significance or possibly pathogenic) was defined.

Follow-up of pregnancy outcome

The pregnancy outcomes and fetal birth were investigated among all pregnant women through a telephone follow-up.

Data management and analysis

All statistical analyses were entered into Microsoft Excel version 2003 (Microsoft Corporation; Redmond, WA, USA) and performed using the statistical software SPSS version 19.0 (SPSS, Inc.; Chicago, IL, USA). All measured data were expressed as mean ± standard deviation (SD), and all categorical data were described as percentage. Differences of proportions were tested for statistical significance with a chi-square test or Fisher's exact test, with a P value < 0.05 indicative of significance.

Ethical statement

This study was reviewed and approved by the Ethics Review Committee of Fujian Maternity and Child Health Hospital (approval number: 2014042). All experimental procedures were performed following the Declaration of Helsinki, the Regulations for Management of Medical Science Research Involving Humans, and the Technical Guidelines of Cytogenetic Prenatal Diagnosis for Fetal Chromosomal Abnormalities. Signed informed consent was obtained from all pregnant women and their spouses with a detailed description of the purpose of the study.

Results

Karyotypes of fetuses with structural abnormalities

Karyotyping was successfully performed in 803 prenatal samples (99.8%), and 2 samples failed in cell culture. Normal karyotype was identified in 767 samples (95.5%) and abnormal karyotype in 36 samples (4.5%). The abnormal karyotypes included chromosomal aneuploidy in 28 samples (6 samples with trisomy 21, 8 samples with trisomy 18, one sample with trisomy 13, 4 samples with sex chromosome abnormality, 3 samples with an extra chromosome and 6 samples with mosaicism) and ultrasound abnormalities in 8 samples (partial deletion or duplication of the chromosome).

SNP array results of fetuses with structural abnormalities

Of the 805 prenatal samples, SNP array detected 716 negatives (88.9%) and 89 positives (11.1%), which included 55 samples with pathogenic CNVs (6.8%) and 34 samples with VOUS (4.2%). In addition to 36 samples showing consistent results with karyotyping, SNP array detected additional 19 samples with pathogenic CNVs, including microdeletion/microduplication syndromes in 18 cases and pathogenic UPD in one case (**Table 1** and Figure S1). The detection rates of pathogenic CNVs were 4.2% (16/383), 7.7% (4/52), 8.8% (28/319), and 13.7% (7/51) in groups A, B, C and D, respectively, and there was a significant difference in the prevalence of pathogenic CNVs among these four groups (χ^2 = 10.266, P = 0.013). The prevalence of pathogenic CNVs was highest in fetuses with structural abnormalities in the urinary system (12.2%, 9/74), followed by in the skeletal system (10.3%, 3/29), the nervous system (8.6%, 8/93), the cardiovascular system (7.3%, 34/466), and those with other structural abnormalities (5%, 1/20), while no pathogenic CNVs were identified in fetuses with structural abnormalities in the head and face, the respiratory system, or the digestive system.

Table 1. Clinical diagnosis and pregnancy outcomes of fetuses with pathogenic copy number variations and normal karyotype

Fetus number	Ultrasound findings	SNP array				Karyotype	Clinical diagnosis	Pregnant outcomes
		Locus	Genetic alterations	Copy number variation size (Mbp)	Number of OMIM genes			
1	Ectopic right kidney complicated by multicystic dysplastic kidney	22q11.21	Loss	2.8	41	46, XN	DiGeorge syndrome	Induction of labor
2	Tetralogy of Fallot and thymic hypoplasia	22q11.21	Loss	3.1	44	46, XN	DiGeorge syndrome	Induction of labor
3	Ventricular septal defect, right-sided aortic arch and aberrant left subclavian artery	22q11.21	Loss	3.1	43	46, XN	DiGeorge syndrome	Induction of labor
4	Fetal growth restriction and ventricular septal defect	22q11.21	Loss	3.1	87	46, XN	DiGeorge syndrome	Induction of labor
5	Fetal growth restriction and pulmonary valve stenosis	4p16.3p16.1	Loss	6.5	96	46, XN	Wolf-Hirschhorn syndrome	Induction of labor
6	Small cavum septum pellucidum and polyhydramnios	17p12	Loss	1.3	5	46, XN	Hereditary neuropathy with liability to pressure palsies	Delivery of a son with developmental retardation
7	Absence or dysplasia of the right kidney	17p12	Loss	1.4	4	46, XN	Hereditary neuropathy with liability to pressure palsies	Loss to follow-up
8	Dysgenesis of the corpus callosum	17p13.3	Loss	2.2	40	46, XN	Miller-Dieker syndrome	Induction of labor
9	Fetal growth restriction, thickening of the nuchal translucency, and thicker pulmonary artery than aorta	7q11.23	Loss	1.4	29	46, XN	Williams-Beuren syndrome	Induction of labor
10	Foramen ovale aneurysm, smaller left heart than right heart and small ascending aorta	22q11.21	Gain	3.1	43	46, XN	22q11.2 duplication syndrome	Induction of labor
11	Ventricular septal defect and absence or dysplasia of the left kidney	7q11.23	Gain	1.3	22	46, XN	7q11.23 duplication syndrome	Induction of labor
12	Fetal growth restriction, ventricular septal defect, pulmonary valve stenosis complicated by insufficiency, and absence or dysplasia of the left kidney	15q24.1q24.2	Loss	2.6	30	46, XN	Overlap with 15q24 microdeletion syndrome	Loss to follow-up
13	Pulmonary artery atresia with intact ventricular septum, tricuspid valve regurgitation and hydropericardium	17p11.2	Gain	2.1	21	46, XN	17p11.2 duplication syndrome	Induction of labor
14	Right aortic arch with mirror image branching and Right-sided ductus arteriosus	17p11.2	Gain	2.3	51	46, XN	Potocki-Lupski syndrome (17p11.2 duplication syndrome)	Induction of labor
15	Aberrant right subclavian artery and right talipes equinovarus	17p12p11.2	Gain	4.7	46	46, XN	17p11.2 duplication syndrome	Induction of labor
16	Left renal hydronephrosis and right renal separation	Xp22.31	Loss	1.6	4	46, XN	Recessive X-linked ichthyosis	Delivery of a son with normal phenotype
17	Spinal dysplasia, L4 hemirertebra deformity, irregular arrangement of sacrococcygeal vertebrae	16p11.2	Loss	0.7	44			Induction of labor
18	Fetal hydrocephalus	16p11.2	Loss	5.7	20			Induction of labor
19	Fetal growth restriction, ventricular septal defect, aortic stenosis, absence or dysplasia of the left kidney and echogenic bowel	16p13.3p12.3	Loss of heterozygosity	10.3	71	46, XN	Maternal uniparental disomy	Induction of labor

Pregnancy outcomes of fetuses with abnormal SNP array results

Among the 805 subjects, there were 84 cases lost to follow-up, and 721 cases were successfully followed up (89.6% follow-up rate), including 534 survived newborns (66.3%), 124 cases with induction of labor (15.4%), 2 cases with abortions (0.3%), 6 cases with intrauterine fetal death (0.8%), 10 cases with stillbirth (1.2%), and 45 cases without delivery (5.6%). The 55 fetuses with pathogenic CNVs identified by SNP array were all given induction of labor.

There were 13 parental samples subjected to SNP array, and 2 samples were identified with *de novo* mutations, showing VOUS (possibly pathogenic), with fetuses undergoing induction of labor. A sample was identified with maternal UPD, and the fetus survived with normal phenotypes. In addition, 9 samples were identified with parental-derived CNVs, and all showed VOUS (possibly benign), including 5 survival fetuses with normal phenotypes, one case with stillbirth, one case with induction of labor and 2 cases lost to follow-up (**Table 2** and Figure S2).

Discussion

Because of its safety, convenience, and non-invasiveness, ultrasonography plays an important role in prenatal screening and diagnosis, and is the first choice in prenatal diagnosis [16]. During prenatal diagnosis, ultrasound examinations detect an increasing number of fetal structural abnormalities and may identify subtle abnormalities [17-19]. Nevertheless, fetal ultrasound abnormalities, which are caused by variation of chromosomes, may be complicated by cognitive dysfunction or developmental retardation, which is undetectable by ultrasonography. Therefore, chromosomal testing is required among fetuses with ultrasound abnormalities [4-6].

In 2012, microarray analysis was reported to identify microdeletions/microduplications and detect all aneuploidies and unbalanced rearrangements, and in samples with a normal karyotype, microarray analysis revealed clinically relevant deletions or duplications in 6.0% with a structural anomaly and in 1.7% of those whose indications were at an advanced maternal age or had positive screening results [9]. Since then, the effectiveness of microarray analysis has been validated in prenatal diagnosis. It has been shown that SNP array detects 6% to 18.7% chromosomal abnormalities among fetuses with ultrasound aberrations [20, 21], and may identify 1.5% to 7.4% of pathogenic microdeletions/microduplications in individuals with ultrasound abnormalities and normal karyotypes [22-24]. In this study, SNP array was found to identify 6.8% pathogenic CNVs among the study samples, and detect 2.4% pathogenic CNVs in cases with ultrasound abnormalities and normal karyotypes, which was consistent with previous reports [20-24].

Because some critical chromosomal abnormalities may not be discovered by karyotyping alone, CMA is introduced if fetuses display vital structural deformities or multiple malformations [25]. If fetuses have rectifiable anomalies after birth except for diagnosis of other chromosomal disorders, this may enhance the decision for fetal preservation. It was reported that a 5.6% prevalence rate of pathogenic CNVs was detected in fetuses with ultrasound abnormalities of a single system and with normal karyotypes, and the prevalence increased to 13.6% in fetuses with ultrasound abnormalities of multiple systems [26]. In this study, the detection rate of pathogenic CNVs was highest in fetuses with structural abnormalities of multiple systems complicated by non-structural abnormalities (13.7%), followed by in those with structural abnormalities of a single system complicated by non-structural abnormalities (8.8%) and those with structural abnormalities of multiple systems (7.7%), and the prevalence was lowest in those with structural abnormalities of a single system (4.2%).

Some genetic syndromes may manifest corresponding abnormal ultrasound phenotypes [27]. There are approximately 77% of fetuses with DiGeorge syndrome (also termed 22q11.2 deletion syndrome) presenting with ultrasound cardiac abnormalities, since this disorder is associated with loss of a portion of the proximal long arm of chromosome 22 [28]. Previous studies have shown that the detection rate of chromosomal abnormality varies among fetal ultrasound abnormalities identified by microarray analysis, and CMA detects a relatively high prevalence rate of chromosomal abnormalities in the cardiovascular system, the nervous system, the musculoskeletal system, the genitourinary symptom and the renal system [29, 30]. In the current study, the detection rates of

Table 2. Identification of variants of unknown significance (VOUS) in 13 parental samples and fetal pregnancy outcomes

Sample no.	Ultrasonographic findings	SNP array results	Interpretation of VOUS	Pregnancy outcomes
1	Hemivertebral deformity of fourth lumbar spine vertebra and irregular arrangement of the sacrococcygeal vertebrae	Fetus: 16p11.2 (29,428,531-30,190,029) × 1 dn; father: 46, XY; mother: 46, XX	VOUS (possibly pathogenic)	Induction of labor
2	Right-sided aortic arch	Fetus: 17p11.2 (16,615,982-18,922,171) × 3 dn; father: 46, XY; mother: 46, XX	VOUS (possibly pathogenic)	Induction of labor
3	Ventricular septal defect, widened interior diameter of the aorta, mitral regurgitation and separation of the bilateral renal collecting system	Fetus: 10q21.1 (59,059,330-60,684,488) × 1 mat; father: 46, XY; mother: 10q21.1 (59,118,220-60,684,488) × 1	VOUS (possibly benign)	Stillbirth
4	Dandy-Walker malformation and ventricular septal defect	Fetus: 15q11.2 (22,770,421-23,277,436) × 1 pat; father: 15q11.2 (22,770,421-23,615,769) × 1; mother: 46, XX	VOUS (possibly pathogenic)	Induction of labor
5	Fetal growth restriction, persistent left superior vena cava, stenosisof the interior diameter of the aortic arch, and hyperechoicbilateral renal parenchyma	Fetus: 2p25.3p11.2 (50,813-87,053,152) hmz 2q11.1q37.3 (95,550,957-242,773,583) hmz; father: 46, XY; mother: 46, XX	Maternal UPD at chromosome 2	Survival with normal phenotype
6	Thickening of right ventricular and pericardial effusion	Fetus: 3p22.3 (33,805,560-35,318,562) × 3 mat; father: 46, XY; mother: 3p22.3 (33,805,560-35,281,232) × 3	VOUS (possibly benign)	Survival with normal phenotype
7	Absence of corpus callosum	Fetus: 5q35.3 (179,194,643-179,767,135) × 3 mat; father: 46, XY; mother: 5q35.3 (179,194,643-179,767,135) × 3	VOUS (possibly benign)	Survival with normal phenotype
8	Ventricular septal defect and tricuspid regurgitation	Fetus: 5q14.1 (76,983,283-77,512,158) × 3 mat; father: 46, XY; mother: 5q14.1 (76,993,054-77,512,158) × 3	VOUS (possibly benign)	Survival with normal phenotype
9	Thickening of the nuchal translucency by 0.64 cm, abnormal posture of both hands and bilateral strephenopodia	Fetus: 8q24.22q24.3 (135,106,599-140,610,869) × 3 mat; father: 46, XY; mother: 8q24.22q24.3 (135,139,947-140,597,017) × 3	VOUS (possibly benign)	Induction of labor
10	Bilateral strephenopodia	Fetus: Xp22.31 (6,449,558-8,135,568) × 3 pat; father: Xp22.31 (6,449,558-8,135,568) × 3; mother: 46, XX	VOUS (possibly benign)	Survival with normal phenotype
11	Nasal bone hypoplasia and ventricular septal defect	Fetus: 15q26.1 (90,211,822-91,080,606) × 1 pat; father: 15q26.1 (90,211,822-91,080,606) × 1; mother: 46, XX	VOUS (possibly benign)	Survival with normal phenotype
12	Right-sided multicystic dysplastic kidney	Fetus: 2p15 (62,195,812-62,697,481) × 1 fat; father: 2p15 (62,195,812-62,697,481) × 1; mother: 46, XX	VOUS (possibly benign)	Lost to follow-up
13	Persistent left superior vena cava and single umbilical artery	Fetus: 8q24.13 (126,044,027-126,414,021) × 3 mat; father: 46, XY; mother: 8q24.13 (126,044,027-126,414,021) × 3	VOUS (possibly benign)	Lost to follow-up

dn, *do novo* mutation; hmz, loss of heterozygosity; mat, mother-inherited; fat, father-inherited; par, parental-inherited; UPD, uniparental disomy.

pathogenic CNVs were 12.2%, 10.3%, 8.6%, 7.3%, and 5% in fetuses with ultrasound abnormalities in the urinary system, the skeletal system, the nervous system, the cardiovascular system, and other systems, while no pathogenic CNVs were identified in fetuses with structural abnormalities in the head and face, the respiratory system, or the digestive system. These findings suggest that SNP array should be strongly recommended for detection of chromosomal abnormalities in fetuses with ultrasound abnormalities in the urinary system, the skeletal system, the nervous system, and the cardiovascular system.

Since SNP array exhibits a high efficiency for diagnosis of fetal chromosomal disorders, multiple VOUS with unclear relevance with clinical phenotypes are detected [31]. The identification of VOUS during prenatal diagnosis may result in a difficulty in clinical genetic counseling, cause pressures on pregnant women and their families, and even lead to excessive induction of labor. VOUS has been identified in less than 5% of all prenatal samples [32, 33]. In this study, a 4.2% prevalence rate of VOUS was detected in the 805 prenatal samples, which was in agreement with previous reports [32, 33]. However, further studies are required for precise assessment of fetuses with VOUS.

In the current study, a fetus identified who had 10.q21.1 deletion by SNP array was found to have inherited it from the mother with a normal phenotype, and VOUS was interpreted as possibly benign. The fetus survived for 20 days, followed by stillbirth, and a deletion of 1.5 Mb was detected in this case, which contained 5 *OMIM* genes. There have been no reports related to pathogenicity of deletion of this fragment; however, this region contained a *BICC1* gene, which is associated with cystic renal dysplasia [34]. The fetus was identified with VOUS (possibly benign); poor pregnancy outcome remained possible in this fetus. In the present study, a fetus identified with 16p13.11 duplications by SNP array was found to have inherited it from the father with normal phenotype, and VOUS was interpreted as possibly pathogenic. The fetus survived with normal phenotype, and a duplication of 1.25 Mb was detected in this case, which contained 19 *OMIM* genes. Subsequent microarray analysis identified 16p13.11 microduplications in this case, and the prevalence of this microduplication is less than 1% in normal populations [35]. The 16p13.11 microduplication region is a susceptible focus of neurocognitive disorders, which may present as developmental retardation, learning difficulty, linguistic difficulties and behavioral abnormalities [35]. 16p13.11 microduplication may be inherited from parents with normal phenotypes or caused by *de novo* mutations [36, 37]. This fetus inherited it from the father with a normal phenotype, and was found to have fetal lateral ventricle broadening on ultrasound, with VOUS interpreted as possibly pathogenic. These findings suggest that the fetal pregnant outcomes remain to be satisfactory even if the VOUS is interpreted as possibly pathogenic. In addition, there were 10 fetuses identified with VOUS (possibly pathogenic) among these fetuses without SNP array of parental samples, including 9 fetuses with induction of labor and a fetus with VOUS (possibly benign) and induction of labor. It is considered that excessive induction of labor may occur in these fetuses. Parental testing is therefore recommended in fetuses with VOUS by SNP array to identify the type and origin of CNVs. In addition, the subsequent follow-up is of great importance to facilitate genetic counseling.

Results of the present study demonstrate that SNP array improves the detection of genetic aberrations in fetuses with prenatal ultrasound abnormality relative to conventional karyotyping, and the risk of CNVs shows a tendency towards a rise with the increase in the number of abnormal ultrasound items. SNP array is strongly recommended for detection of fetal ultrasound abnormalities.

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Disclosure of conflict of interest

None.

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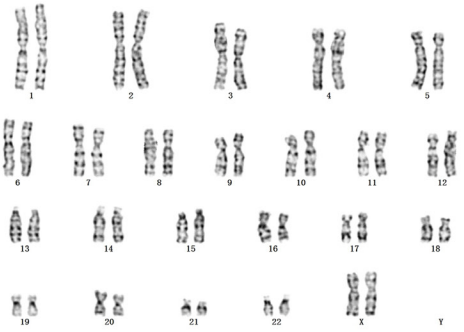
Fetus 1



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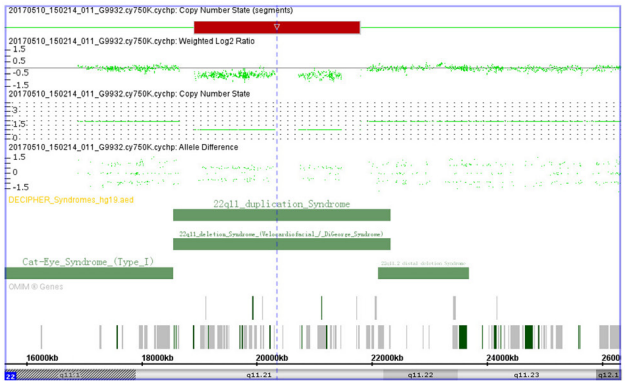
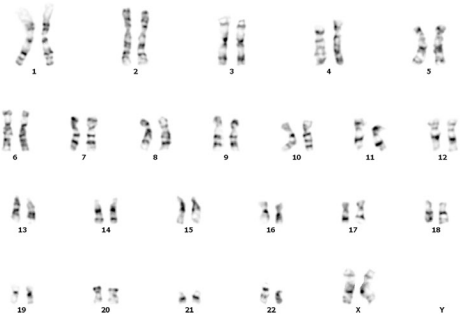


SNP array

Fetus 2



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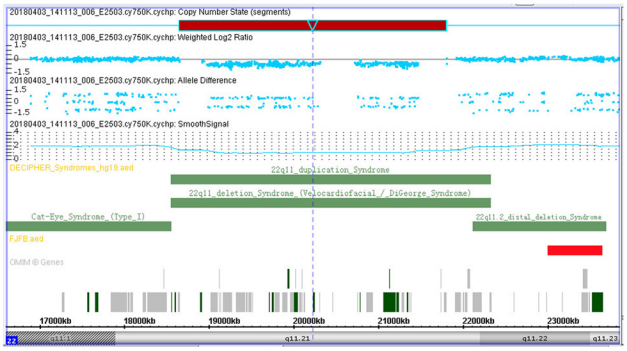
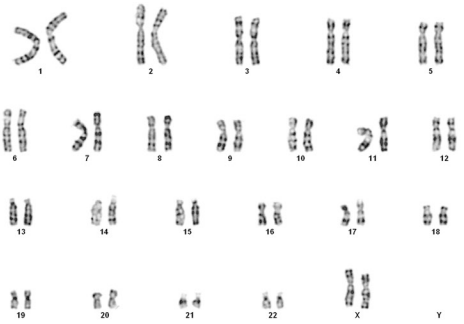


SNP array

Fetus 3



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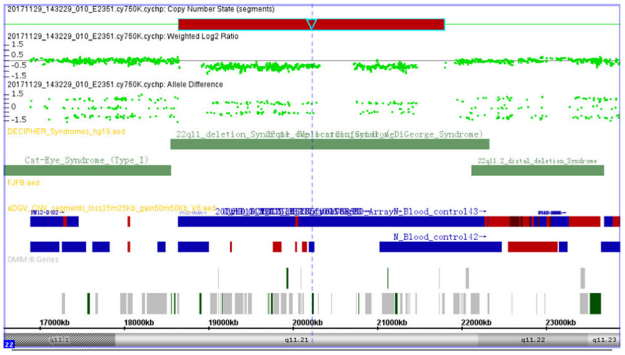
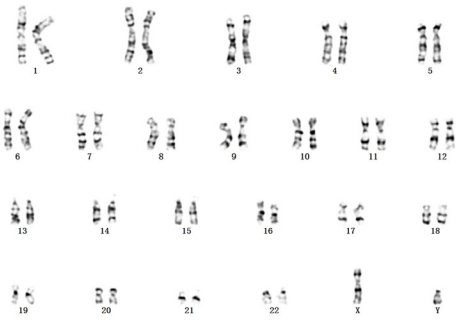


SNP array

Fetus 4



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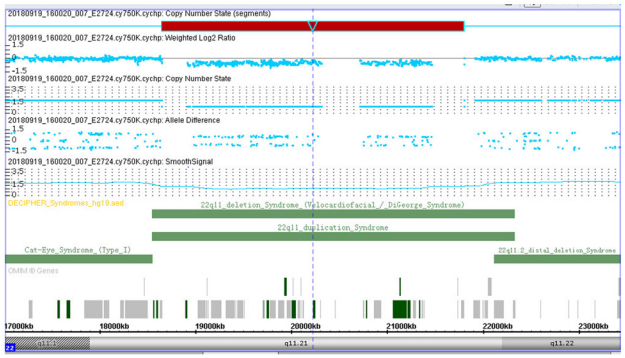
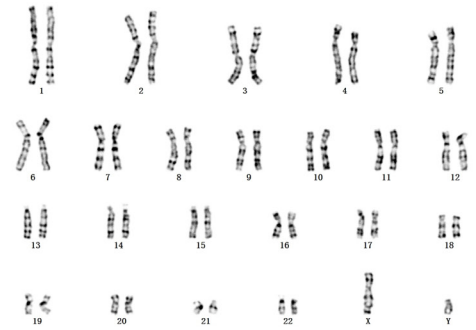


SNP array

Fetus 5



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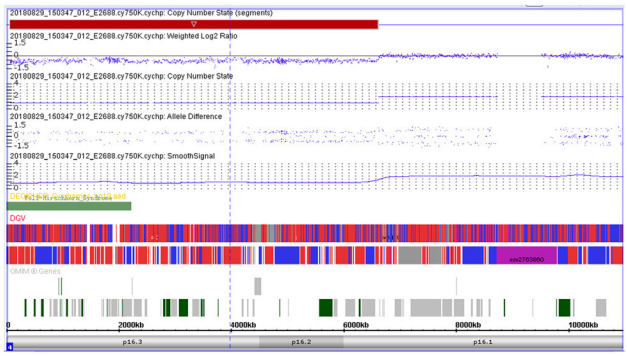
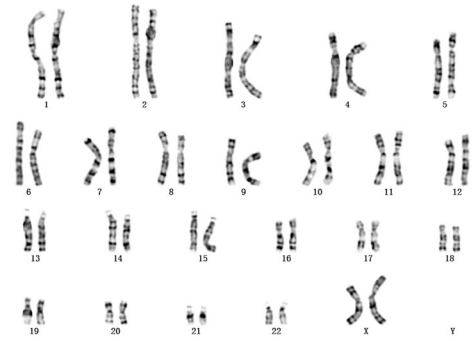
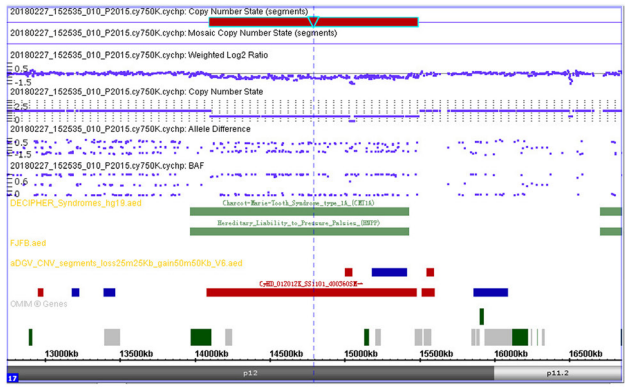


SNP array

Fetus 6

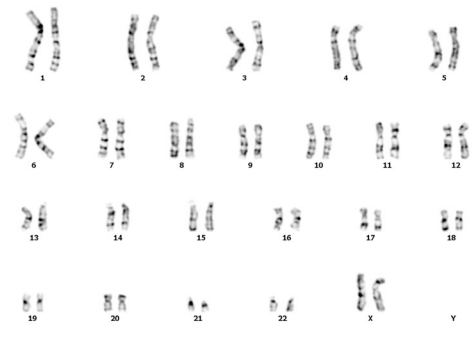


Karyotype



SNP array

Fetus 7



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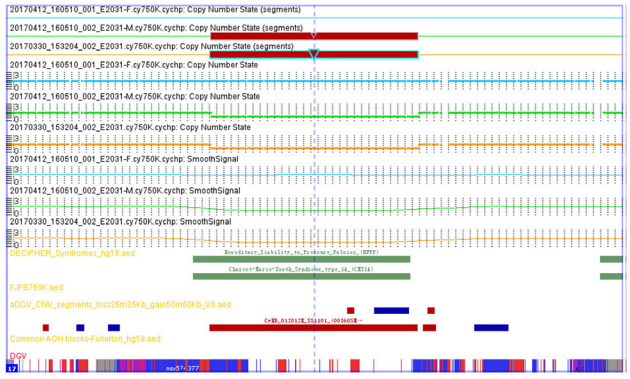
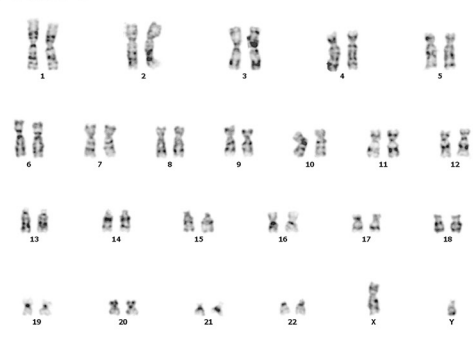


SNP array

Fetus 8



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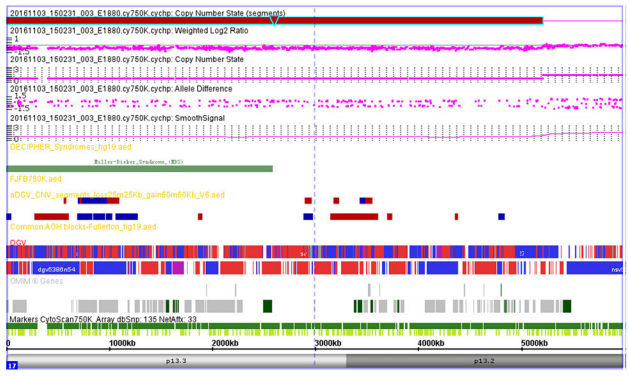
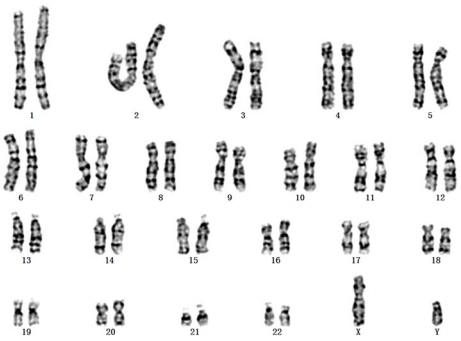


SNP array

Fetus 9



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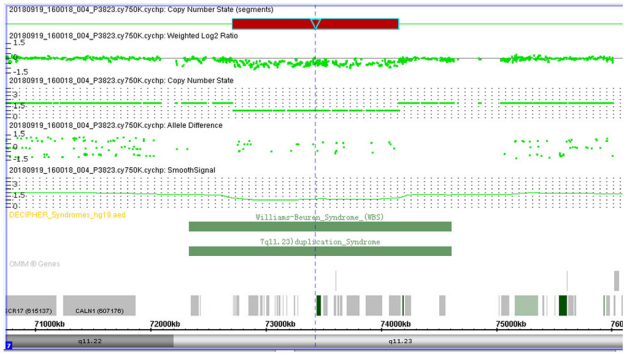
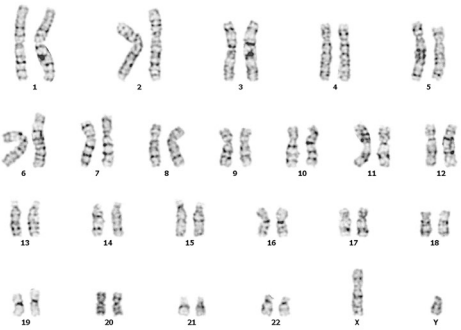


SNP array

Fetus 10



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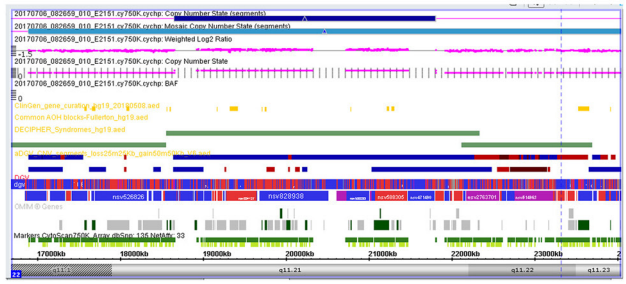
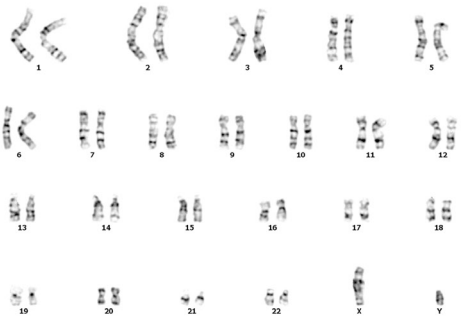


SNP array

Fetus 11



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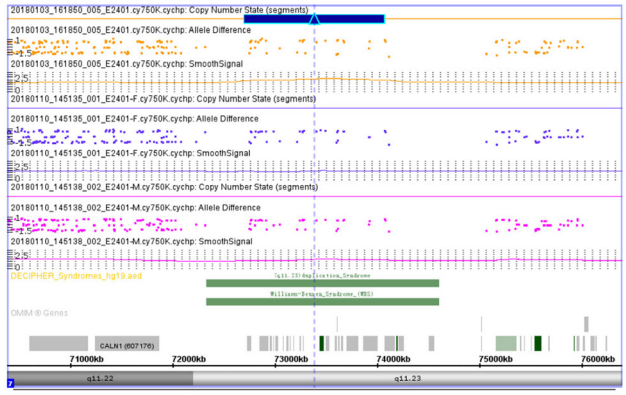
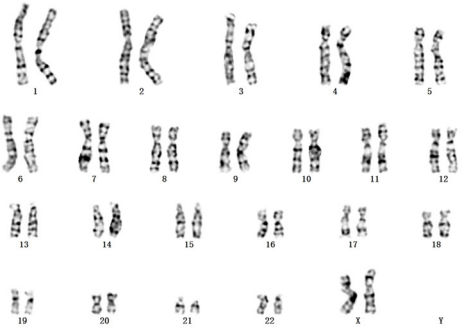


SNP array

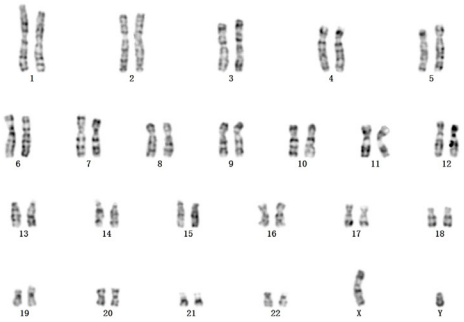
Fetus 12



Karyotype

SNP array

Fetus 13



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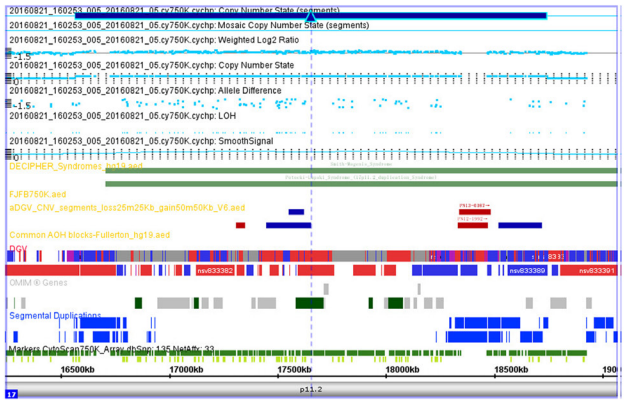
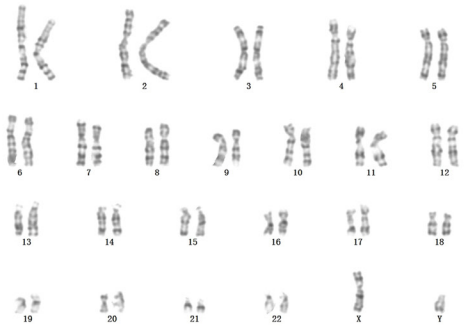


SNP array

Fetus 14



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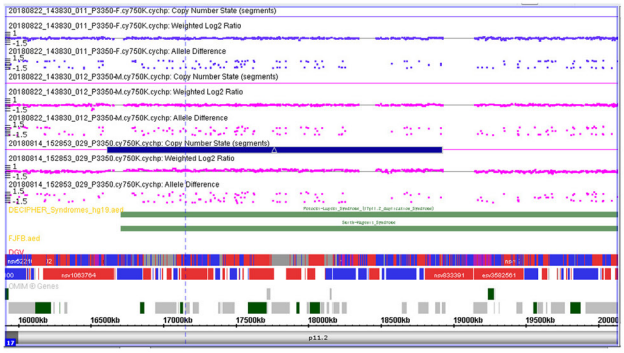
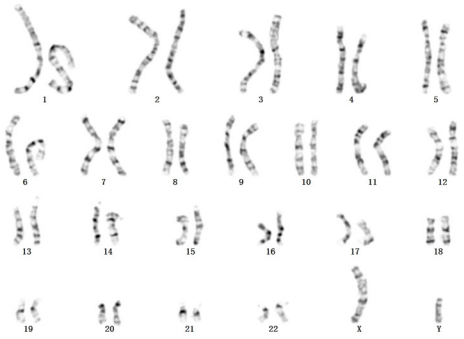


SNP array

Fetus 15



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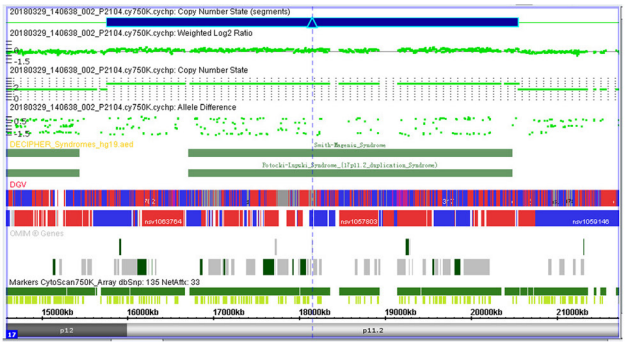
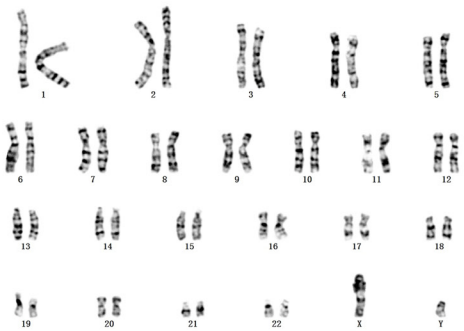


SNP array

Fetus 16



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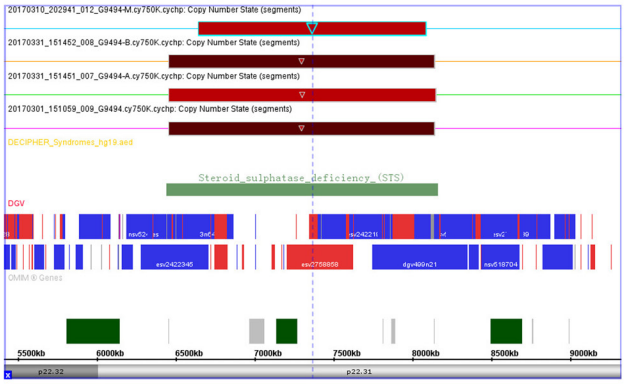

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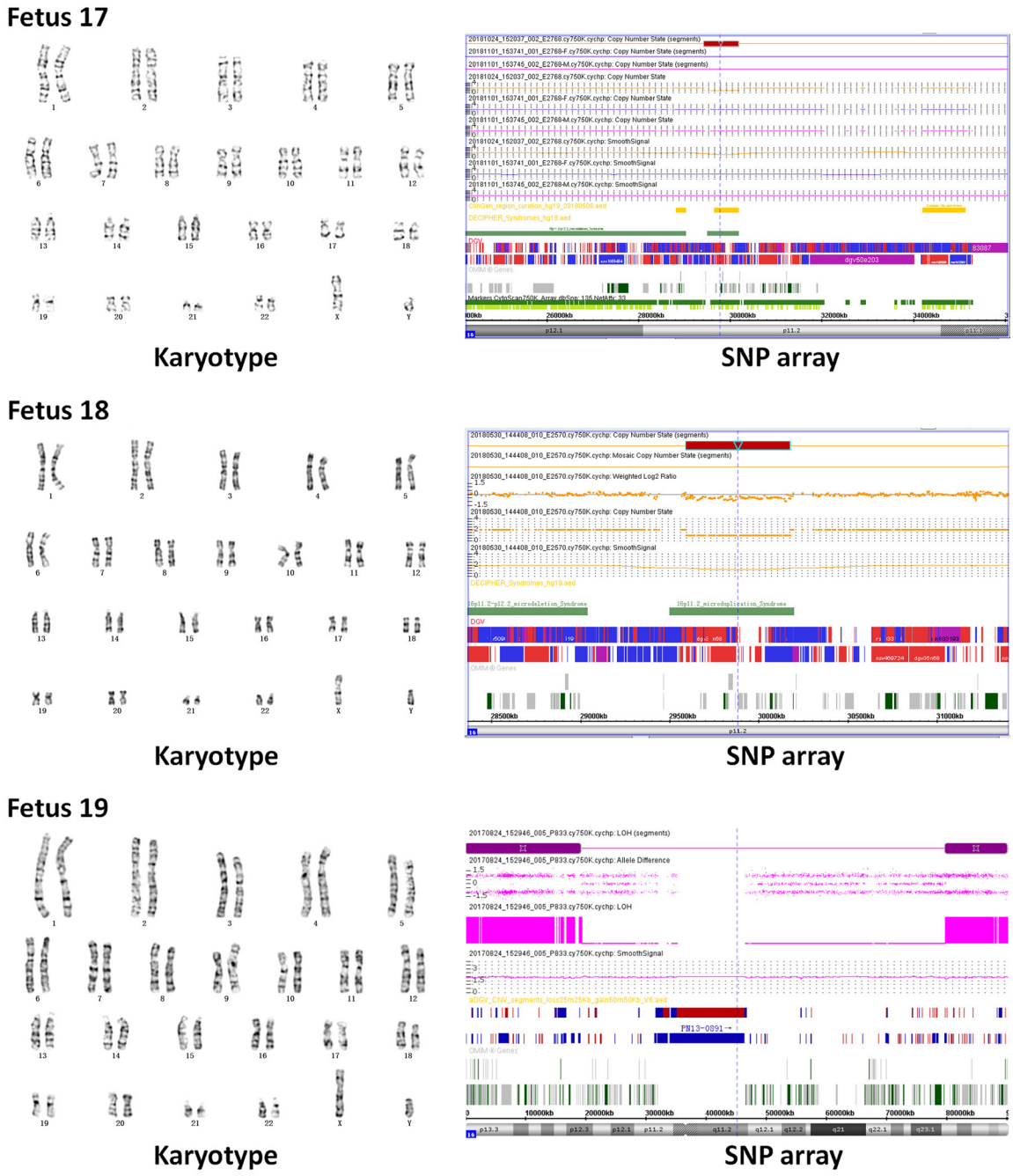
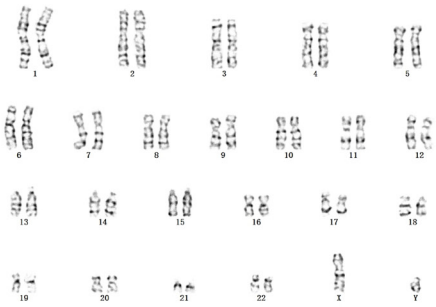


Figure S1. Karyotyping and SNP array results of 19 fetuses with pathogenic CNVs.

Sample 1



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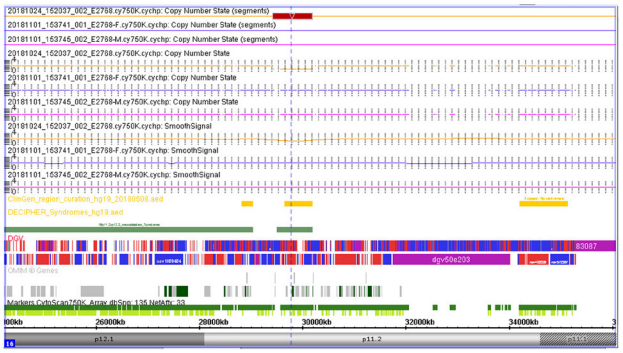
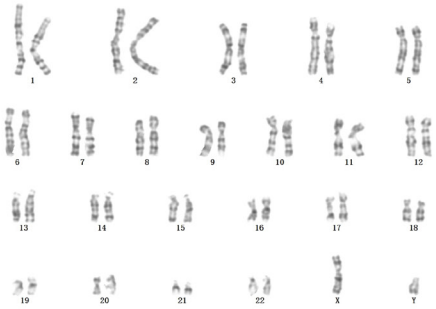
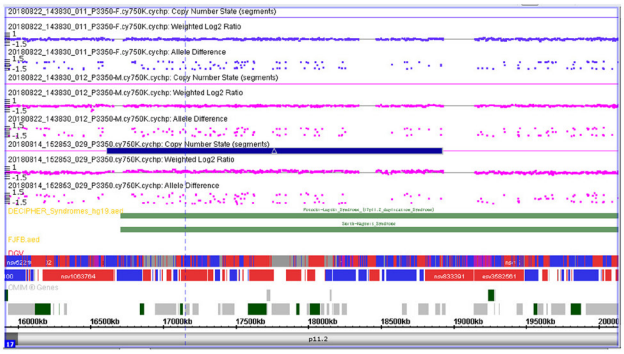


SNP array

Sample 2

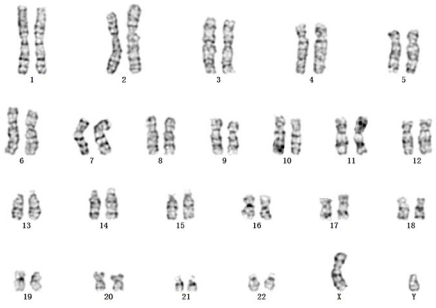


Karyotype



SNP array

Sample 3



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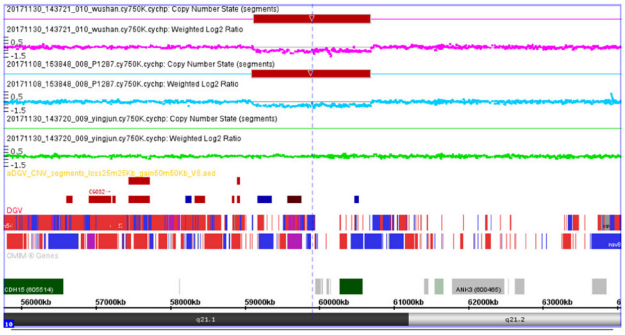
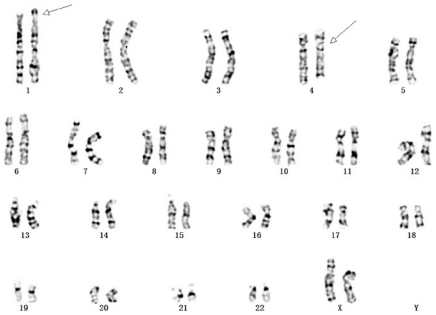


SNP array

Sample 4



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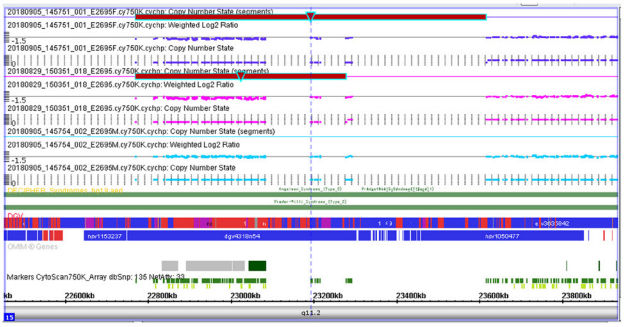
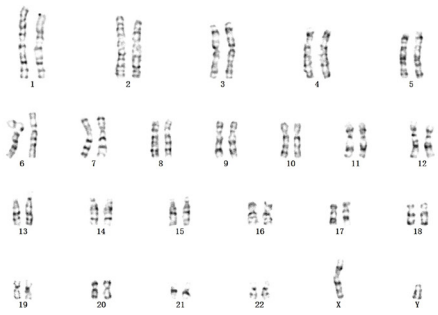


SNP array

Sample 5



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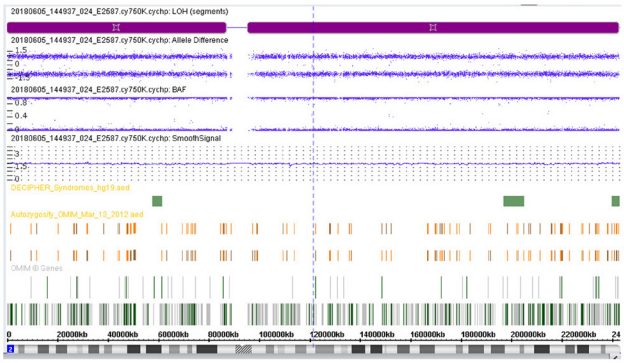
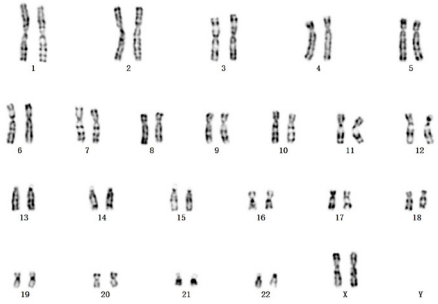
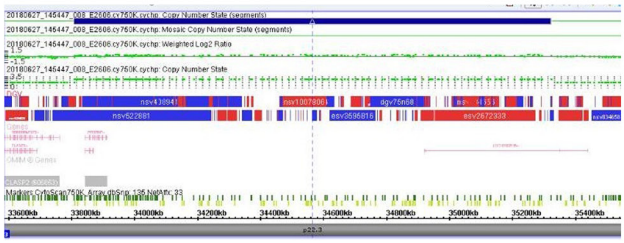


SNP array

Sample 6

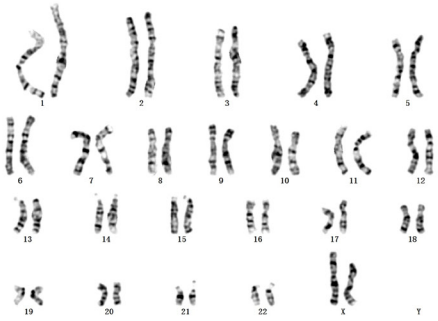


Karyotype



SNP array

Sample 7



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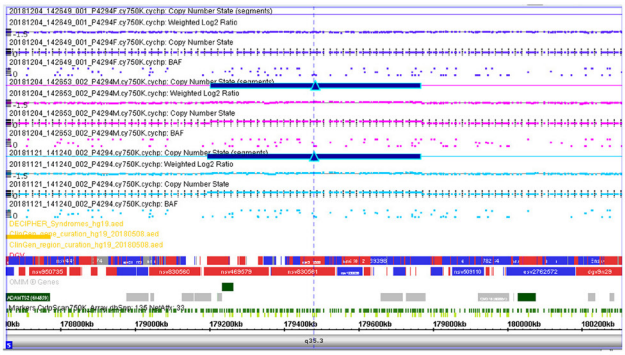
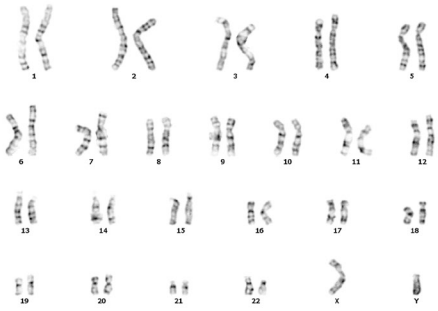
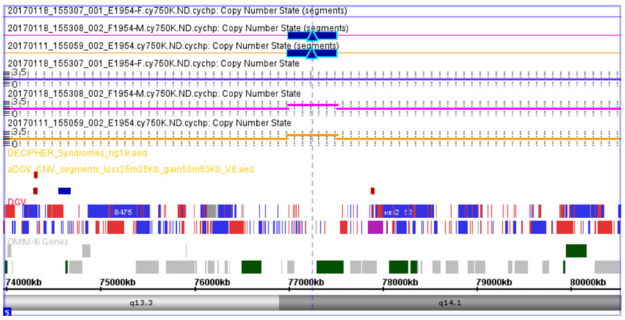


SNP array

Sample 8

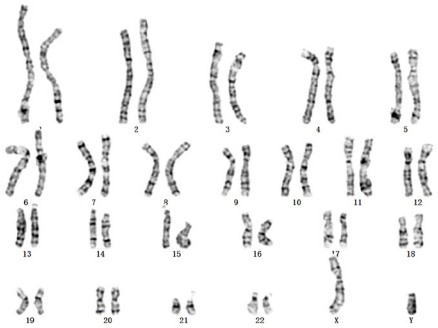


Karyotype

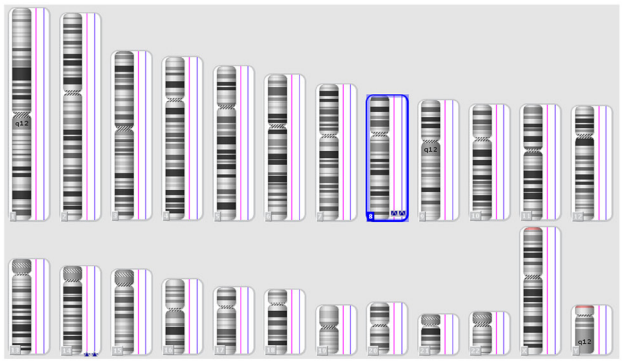


SNP array

Sample 9

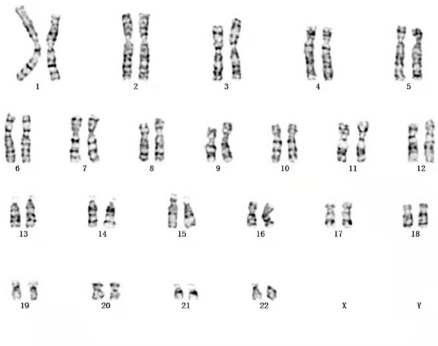


Karyotype

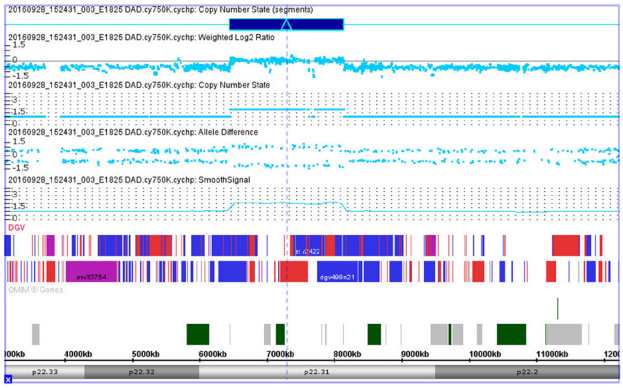


SNP array

Sample 10

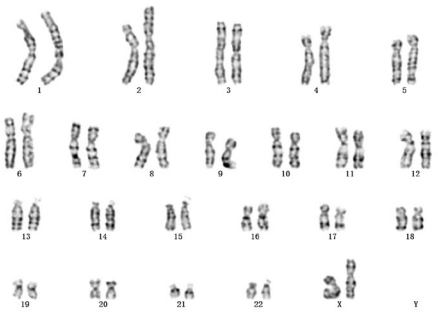


Karyotype



SNP array

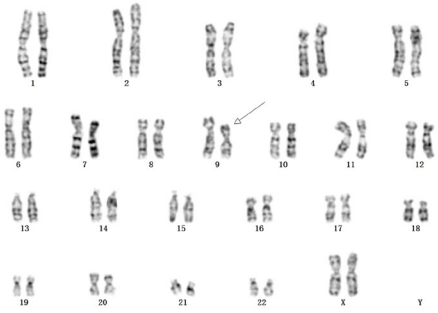
Sample 11



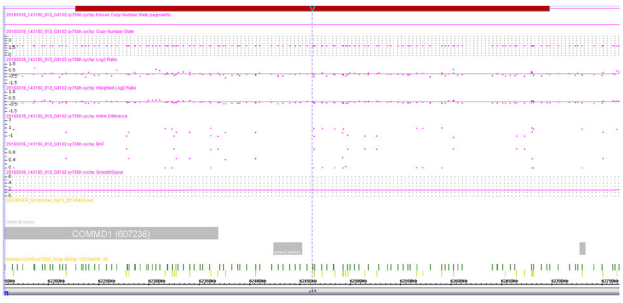
Karyotype

SNP array

Sample 12



Karyotype



SNP array

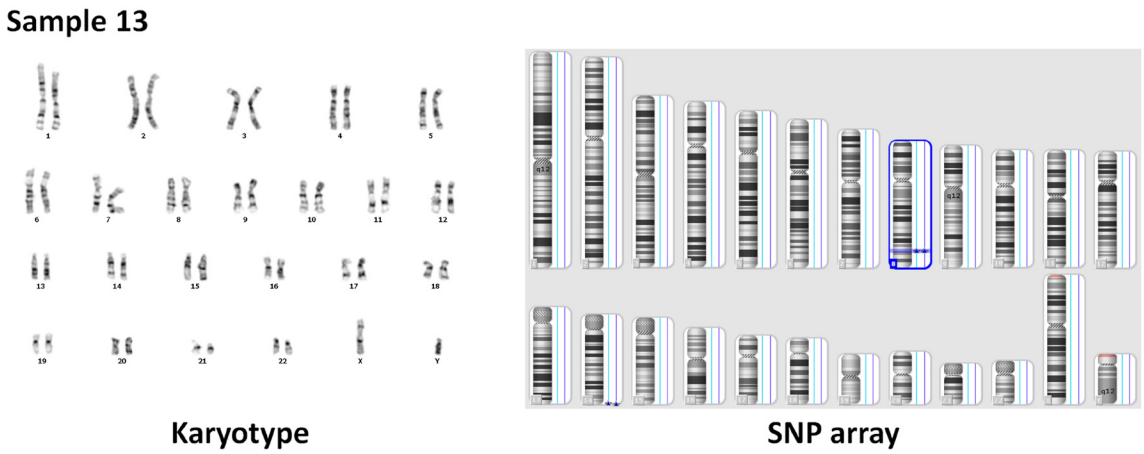


Figure S2. Karyotyping and SNP array results of 13 parental samples.