Copy number variants and placental abnormalities in stillborn fetuses: a secondary analysis of the Stillbirth Collaborative Research Network study

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February 12, 2022

Abstract

Objective To examine the association of DNA copy number variants (CNVs) with pathologic placental lesions (PPLs) in stillborn fetuses. Design A secondary analysis of stillbirth cases in the Stillbirth Collaborative Research Network case-control study. Setting Multicenter, 59 hospitals in 5 geographic regions in the USA. Population 387 stillbirth cases (2006-2008). Methods Using standard definitions, PPLs were categorized by type including maternal and fetal vascular, inflammatory and immune/idiopathic lesions. Using single-nucleotide polymorphism array, CNVs of at least 500 kb were detected. CNVs were classified into two groups: normal, defined as no CNVs>500 kb or benign CNVs, and abnormal, defined as pathogenic or variants of unknown clinical significance. Main outcome measures The proportions of abnormal CNVs and normal CNVs were compared between stillbirth cases with and without PPLs using the Wald Chi-squared test. Results Of 387 stillborn fetuses, 327 (84.5%) had maternal vascular PPLs and 60 (15.6%) had abnormal CNVs. Maternal vascular PPLs were more common in stillborn fetuses with abnormal CNVs compared with those with normal CNVs (81.7% vs. 64.2%; p=0.008). The proportions of fetal vascular, maternal/fetal inflammatory, and immune/idiopathic PPLs were similar among stillborn fetuses with abnormal CNVs compared to those with normal CNVs. Pathogenic CNVs in stillborn fetuses with maternal vascular PPLs spanned several genes with known relevant mechanisms. Conclusions Abnormal placental/fetal CNVs were associated with maternal vascular PPLs in stillborn fetuses. Findings may provide insight on the mechanisms of specific genetic abnormalities associated with placental dysfunction and stillbirth.

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This work was supported by grant funding from the *Eunice Kennedy Shriver* National Institute of Child Health and Human Development: U10-HD045953 Brown University, Rhode Island; U10-HD045925 Emory University, Georgia; U10-HD045952 University of Texas Medical Branch at Galveston, Texas; U10-HDO45955 University of Texas Health Sciences Center at San Antonio, Texas; U10-HD045944 University of Utah Health Sciences Center, Utah; and U01-HD045954 RTI International, RTP. Secondary analysis of the primary research was supported in part by the National Center for Advancing Translational Sciences of the National Institutes of Health under Award Number 1UL01TR002538. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

Presented in part at the 35th annual scientific meeting of The Society for Pediatric and Perinatal Epidemiologic Research, June 8, 2021, Boston, Massachusetts.

Abstract

Objective

To examine the association of DNA copy number variants (CNVs) with pathologic placental lesions (PPLs) in stillborn fetuses.

Design

A secondary analysis of stillbirth cases in the Stillbirth Collaborative Research Network case-control study.

Setting

Multicenter, 59 hospitals in 5 geographic regions in the USA.

Population

387 stillbirth cases (2006-2008).

Methods

Using standard definitions, PPLs categorized by type including maternal and fetal vascular, inflammatory and immune/idiopathic lesions. Using single-nucleotide polymorphism array, CNVs of at least 500kb detected. CNVs classified into two groups: normal, defined as no CNVs>500kb or benign CNVs, and abnormal, defined as pathogenic or variants of unknown clinical significance.

Main outcome measures

The proportions of abnormal CNVs and normal CNVs compared between stillbirth cases with and without PPLs using the Wald Chi-squared test.

Results

Of 387 stillborn fetuses, 327 (84.5%) had maternal vascular PPLs and 60 (15.6%) had abnormal CNVs. Maternal vascular PPLs were more common in stillborn fetuses with abnormal CNVs compared with those with normal CNVs (81.7% vs. 64.2%; p=0.008). The proportions of fetal vascular, maternal/fetal inflammatory, and immune/idiopathic PPLs were similar among stillborn fetuses with abnormal CNVs compared to those with normal CNVs. Pathogenic CNVs in stillborn fetuses with maternal vascular PPLs spanned several known genes.

Conclusions

Abnormal placental/fetal CNVs were associated with maternal vascular PPLs in stillborn fetuses. Findings may provide insight on the mechanisms of specific genetic abnormalities associated with placental dysfunction and stillbirth.

Keywords

Copy number variants; placenta; stillbirth; pathologic lesions

Running title

Copy number variants and placental abnormalities.

Tweetable abstract

Abnormal copy number changes in stillborn placental and fetal DNA are associated with maternal vascular placental lesions.

Introduction

Stillbirth, most commonly defined in the U.S. as fetal death at [?]20 weeks' gestation, occurs at an estimated rate of 5.7 per 1000 births in the United States.[1] The causes of stillbirth are multi-factorial, and histologic examination of the placenta, cord, or membranes can identify a cause of death in 11-65% stillbirths.[2, 3] Maternal and fetal vascular lesions in the placenta are commonly associated with stillbirth.[4-6] Proper function of the placenta requires uninterrupted flow of adequately oxygenated maternal and fetal blood, which is critical for fetal survival.[7]Macroscopic abnormalities of the placenta, such as aberrations in shape and size, and microscopic variation in villous morphology, often reflect placental function.[8, 9] Determining factors influencing placental pathological lesions will help facilitate understanding the pathogenesis, diagnosis, and treatment of stillbirth with an underlying placental etiology.[10]

Chromosomal assessment of the placenta and fetus with the use of single-nucleotide polymorphism (SNP) oligonucleotide microarray analysis is useful in determining causes of death in stillbirth and structural malformations in live births.[11-13] Some forms of chromosomal structural abnormalities such as trisomy and monosomy are known to be associated with stillbirth. However, the relevance of other types of genetic aberrations in stillbirth cases is not well characterized, and the mechanism by which they contribute to stillbirth is not understood. Therefore, our objective was to evaluate the associations of placental and fetal CNVs with placental pathological lesions in a well characterized study of stillbirth. In addition, we discuss and highlight specific CNV deletions and duplications in genes associated with placental pathological lesions.

Methods

This study was a secondary analysis of the Stillbirth Collaborative Research Network (SCRN) study. Briefly, the SCRN study was a racially, ethnically, and geographically diverse, multicenter case-control study of stillbirth and selected live births with enrollment at the time of delivery. Recruitment occurred in 59 hospitals in 5 geographic regions throughout the US. Details about the participating hospitals and study population have been described previously.[11, 14] The study was approved by the institutional review board at each clinical site and the data coordinating center. An advisory board reviewed the progress and safety of the study and written informed consent was obtained from each participant.

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Postmortem examinations and placental histologic examinations were performed by perinatal pathologists who underwent centralized training using a standardized format. Details of the placental pathologic evaluation have been previously reported.[9] The INCODE cause of death classification tool was used across sites to best identify cases where a fetal or placental condition significantly contributed to the fetal death.[15, 16] In the present analysis, we included singleton stillbirth deliveries with chromosomal microarray and postmortem examinations of the fetus and placenta (n=387). Of note, 98.6% of stillbirth cases in the SCRN study had placental examination completed.[9].

A consensus-determined protocol was implemented to define placental pathological lesions types as maternal vascular, fetal vascular, maternal inflammatory, fetal inflammatory and immune/idiopathic lesions.[17, 18] The specific pathologic placental lesions included in each category are listed in **Table S1** .

Biospecimens collected as part of the SCRN protocol included placental tissue, fetal liver, muscle, and cord blood. Sizes of placental biopsies varied, but they were as large as 1 cm^3 and were stored at – 20^0C from 2 – 5 years prior to DNA extraction. Microarray analysis was performed at a single laboratory (Columbia University Medical Center) in 2012. Samples were analyzed using the Affymetrix Genome Wide Human SNP Array 6.0 and the Chromosome Analysis Suite, version 1.0.1, and the NetAffx annotation database, version 28 with data aligned to the Human Genome release 18. CNVs of [?]500 kb in size were detected using the SNP array. Analysis of the array data was conducted to determine aneuploidy, potential maternal-fetal contamination, and sex discordance. Classification of CNVs was based on the American College of Medical Genetics (ACMG) standards and guidelines for interpretation and reporting, with modifications as described previously.[19] Due to improving resolution for determination of pathogenicity of CNVs, the number of novel structural variants is constantly increasing.[20] Therefore, we implemented the latest ACMG guidelines[21] in high-throughput CNV analysis to classify and update pathogenicity of CNVs previously categorized as variants of unknown clinical significance (VOUS) by using ClassifyCNV tool.[22] Since VOUS CNVs should not be considered benign, we classified CNVs into two groups: abnormal CNVs (abnormal CNVs), defined as pathogenic CNVs (including aneuploidy) or VOUS, and normal CNVs (normal CNVs), defined as no CNVs $>$ 500 kb or benign CNVs.[23-25]. As such, the abnormal CNVs and normal CNVs groups were compared in statistical analysis. In addition, study characteristics were described separately for pathogenic, VOUS and normal CNVs. We discussed the implications of the findings based on only pathogenic CNVs (excluding trisomy, monosomy, sex-chromosome and VOUS CNVs).

To test for associations of placental pathological lesions with CNV categorization, frequency and percentages were calculated within category and compared using chi-square. Other categorical measures were similarly compared by CNV category. To compare continuous measures, ANOVA was used. Data were analyzed with the use of statistical software programs: SAS version 9.4 (SAS Institute Inc), R and STATA version 15.0 (StataCorp), and ClassifyCNV tool.[22]

Results

Among stillbirth fetuses included in this study (n=387), there were 60 (15.6%) with abnormal CNVs (40 [10.3%] with pathogenic, and 20 [5.2%] with VOUS CNVs) and 327 (84.5%) with normal CNVs. Comparisons of proportions showed that stillborn fetuses with abnormal CNVs tended to be born to older women, of Hispanic ethnicity, and anomalous in comparison to those with normal CNVs (**Table 1**). Stillborn fetuses with abnormal CNVs did not differ from those with normal CNVs in regard to other socio-economic factors, parity, fetal sex, maternal chronic hypertension, preeclampsia, diabetes and gestational diabetes.

The proportion of stillborn fetuses with maternal vascular pathological lesions was higher among those with abnormal CNVs in comparison to those with normal CNVs (81.7% vs. 64.2%; p=0.008; **Figure 1** ;**Table S2**). However, the proportions of stillborn fetuses with fetal or any (i.e. fetal or maternal) vascular pathological lesions among those with abnormal CNVs were similar in comparison to those with normal CNVs (78.3% vs. 77.1%; p=0.8, 95.0% vs. 90.8%; p=0.3, respectively). Furthermore, the proportions of stillborn fetuses with maternal inflammatory, fetal inflammatory, any inflammatory or immune/idiopathic placental pathological lesions among those with abnormal CNVs were similar in comparison to those with normal CNVs (25.0% vs.

33.3%; p=0.2, 11.7% vs. 15.6%; p=0.4, 28.3% vs. 36.2%; p=0.2, and 15.8% vs. 10.4; p=0.3, respectively). Pathogenic deletion CNVs (n=4) in three stillborn fetuses and pathogenic duplication CNVs (n=4) in five stillborn fetuses with maternal vascular pathological lesions were identified (**Table 2**). Two stillborn fetuses with maternal vascular pathological lesions also had VOUS deletions CNVs (n=3), and thirteen stillborn fetuses with maternal vascular pathological lesions had VOUS duplications CNVs (n=14) (**Table S3**). Three stillborn fetuses with maternal vascular pathological lesions had multiple abnormal CNVs. Specifically, CNV 22q11.21 deletion, 16p13.11 duplication, and 4q32.3q35.2 and 17p13.3 were identified in stillborn fetuses with maternal vascular pathological lesions. The deletions and duplications each involved several genes, including the*MYH11* (myosin heavy chain 11) and *HNF1B* (hepatocyte nuclear factor 1B) genes.

Discussion

Main Findings

Our data suggest that abnormal CNVs are associated with maternal vascular lesions in placenta of stillborn fetuses. Several pathogenic CNV deletions and duplications were identified in eight-stillborn fetuses with maternal vascular lesions, involving several genes.

Interpretation

In 88% of stillborn fetuses, a direct cause or a major contributor to death was found by histopathologic examination of the placenta, showing placental abnormalities as the most common causes of death.[3, 4, 9] Specifically, maternal vascular supply abnormalities were more common in preterm stillbirth cases, while fetal vascular supply abnormalities were more common among term stillbirth cases.[6, 9] Moreover, a review of 120 autopsy reports of stillborn fetuses and placentas showed maternal vascular supply abnormalities in 54 (51%), fetal vascular supply abnormalities in 28 (26%), and inflammatory lesions in 13 (12%) cases as direct or major contributors of death.[4, 9] Yet, many deaths in stillbirth cases remain unexplained,[26-28] and our understanding of the mechanisms of placental dysfunction and stillbirth is limited.

In our study, 22q11.21 deletion, 16p13.11 duplication, and 4q32.3q35.2 and 17p13.3 deletion CNVs were identified in stillborn fetuses with maternal vascular pathological lesions. These CNVs were previously detected among stillborn fetuses in an analysis of SCRN cases,[11] but not described in the context of placental pathological lesions. The deletion of 22q11.21 is pathogenic for DiGeorge/velocardiofacial syndrome.[29] Pathologic placental lesions are found in DiGeorge sequence, characterized by hypoplasia in the umbilical cord arteries and widespread calcification of microthrombi in the arteries of the second and third order villous branches.[30] A case study showed a copy gain of the distal region of chromosome 4 at segment 4q32.3q35.2 in a pregnant patient that presented with fetal edema and subsequent fetal loss.[31] Furthermore, the duplication of 16p13.11 is implicated in multiple congenital anomalies in pediatric patients (n=1645).[32] Mutations in *MYH11* (myosin heavy chain 11) gene, among several genes in the 16p13.11 region, cause thoracic aortic aneurysms and/or dissections.[33, 34]Lastly, the 17q12 recurrent deletion syndrome is characterized by structural or functional abnormalities of the kidney.[35] *HNF1B* (Hepatocyte nuclear factor 1B) gene, among several genes in the 17q12 region, is implicated in renal cysts and diabetes syndrome.[36]

In light of prior data, our findings suggest that 22q11.21, 4q32.3q35.2, and 16p13.11 CNVs may contribute to fetal death through placental abnormalities, both directly, as well as indirectly, by contributing to cardiac abnormalities. Interestingly, maternal/fetal vascular and inflammatory placental pathological lesions were common in pregnancies complicated by congenital heart defects [37] and placental insufficiency is associated with congenital heart defects in animals and humans.[38-40] Further, the placental genome plays a role in mediating fetal and maternal health.[41-43] As such, 16p13.11 and 17q12 CNVs described in the context of congenital anomalies and kidney function in children and adults also suggest that genetic abnormalities in the placenta may underlie mechanisms of the developmental origins of health complications in later life.[38, 42] Together, these mechanisms may provide answers to causes of death in stillbirth, a basis for estimating recurrence risk, and offer insight into the developmental origins of diseases in later life.

Strengths and Limitations

Our study has several limitations. In the present analysis, we only reported pathogenic deletions and duplications spanning several genes in stillborn fetuses with maternal vascular placental pathological lesions. While only maternal vascular placental pathological lesions were associated with CNV types in our study, the small sample size in the abnormal CNVs group may have limited power to detect associations of CNV types with other placental pathological lesions. However, to date, our study included the largest sample size of stillbirths with CNVs that had placental pathological exam completed. Another limitation is that pathologists were not blinded to stillbirth or live-birth status in the SCRN study because of the need to perform both clinical and research placental examination. Furthermore, our ability to determine pathogenicity of CNVs is limited. This will improve with increasingly larger databases of normal and abnormal phenotypes. Additionally, there are limitations to microarray-based analyses, which include inability to detect truly balanced rearrangements. Microarray-based analyses also report large chromosomal regions that span several genes, such as the ones reported in our study, making it difficult to target specific mutations in genes for clinical application. Lastly, due to lack of paternal DNA, we were not able to distinguish inherited from newly occurring CNVs in the placenta or fetus. Using higher resolution technology, such as next generation sequencing of DNA from families, future studies will be able to identify genetic mutations causing placental dysfunction and stillbirth.

One of the strengths of our study is that our cohort included a geographically, racially, and ethnically diverse study population with stillbirth. In addition, participants had a complete evaluation, including fetal postmortem examination, placental pathological analysis conducted by perinatal pathologists, and maternal-fetal testing.[44] These study design features provided careful phenotyping of stillbirth included in our study and maximized the validity of the present analysis.

Conclusion

Our report provides additional support for the utility of high-density microarray analysis in the detection of CNVs associated with placental pathological lesions in stillbirth cases. Further clarification of the relationship between chromosomal aberrations and placental abnormalities will be important in order to better understand the specific mechanisms leading to placental dysfunction and stillbirth. As such, these mechanisms may provide answers to causes of death in stillbirth and the basis for estimating recurrence risk.

Disclosure of interests

Eunice Kennedy Shriver National Institute of Child Health and Human Development grants were received by the institutions listed. No other conflicts of interest are reported by authors. Completed disclosure of interests forms are available to view online as supporting information.

Contribution to authorship

RMS, UMR, HP and RLG had critical roles in the conception, planning and carrying out of the study. VT, AA and TW played a critical role in data management and secondary data analysis. TW, RMS, UMR, HP, RLG, SD, AA, AZC, JMP, NRB and VT each played critical roles in conception, interpretation of the secondary data analysis and writing the manuscript. The University of Utah Institutional Review Board found this secondary analysis qualified for IRB exemption 5/9/2019 IRB#122488 due to the de-identified nature of the data.

Ethical approval

The study was approved by the institutional review board at each clinical site and the data coordinating center. An advisory board reviewed the progress and safety of the study and written informed consent was obtained from each participant.

Funding/support

This work was supported by grant funding from the *Eunice Kennedy Shriver* National Institute of Child Health and Human Development: U10-HD045953 Brown University, Rhode Island; U10-HD045925

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Data availability statement

The data that support the findings of this study are avail- able from the corresponding author upon reasonable request.

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Table 1 . Maternal and Stillbirth Demographic and Clinical Characteristics

Characteristic	**Pathogenic CNV**	**VOUS CNV**	**Normal CNV**	**P-value**
N	40	20	327	
Maternal age at delivery, yr	28.9 (8.0)	23.6 (6.1)	27.4 (6.4)	0.012
Maternal age at delivery, categories				
<34 years	29 (72.5)	18 (90.0)	277 (84.7)	0.105
[?]35 years	11 (27.5)	2 (10.0)	50 (15.3)	
Maternal Body-mass-index (BMI), kg/m^2	28.5 (8.5)	24.9 (4.5)	27.4 (6.6)	0.152
Maternal BMI, categories				

Characteristic	Pathogenic CNV	VOUS CNV	Normal CNV	P-value
<24.9 (under/normal weight)	15 (40.5)	12 (60.0)	142 (45.4)	0.68
25-29.9 (overweight)	9 (24.3)	4 (20.0)	78 (24.9)	
[?]30-34 (obese)	13 (35.1)	4 (20.0)	93 (29.7)	
Maternal race/ethnicity				
white non-Hispanic	12 (30.0)	8 (40.0)	132 (40.5)	0.019
black, non-Hispanic	2 (5.0)	4 (20.0)	68 (20.9)	
Hispanic	23 (57.5)	5 (25.0)	108 (33.1)	
Other	3 (7.5)	3 (15.0)	18 (5.5)	
Maternal education, yr	12.7 (3.6)	12.5 (2.5)	13.0 (3.0)	0.678
Maternal education, categories				
Less than college	21 (60.0)	12 (60.0)	149 (49.0)	0.325
College or more	14 (40.0)	8 (40.0)	155 (51.0)	
Insurance/method of payment				
any assistance	22 (55.0)	13 (65.0)	185 (56.6)	0.738
commercial/HMO/Military	18 (45.0)	7 (35.0)	142 (43.4)	
Smoked 3 months prior to pregnancy	6 (17.1)	3 (15.0)	58 (19.1)	0.876
Parity				
Nulliparous	15 (37.5)	10 (50.0)	147 (45.1)	0.582
Multiparous	25 (62.5)	10 (50.0)	179 (54.9)	
Clinical history: Hypertension	4 (10.0)	3 (15.0)	32 (9.8)	0.754
Preeclampsia/Hypertension	3 (7.7)	2 (10.5)	33 (10.5)	0.862
Clinical history: Diabetes (type 1 or 2)	3 (7.5)	2 (10.0)	17 (5.2)	0.581
Gestational Diabetes	4 (10.0)	1 (5.0)	13 (4.0)	0.232
Preterm birth	38 (95.0)	16 (80.0)	269 (82.3)	0.112
Fetal sex: male	24 (60.0)	12 (60.0)	164 (50.3)	0.385
Any fetal structural anomaly	23 (67.6)	3 (15.8)	56 (18.8)	<.001

For categorical comparisons, we report frequency, percentage, and p-value from Wald chi-square test.

For comparisons of continuous measures, we report mean, standard deviation (SD), and p-value form ANOVA.

Table 2. Specific pathogenic CNVs (excluding trisomy, monosomy, sex-chromosome and variants of unknown significance CNVs) with maternal vascular lesions in placenta of stillbirth fetuses

Gestational age	ISCN Nomenclature [a]	Chromosomal position [b]	Copy number change [c]	Genes
27	arr 4q32.3q35.2 x1	chr4:164762764-171448183	DEL	AADAT, ANXA
	arr 17p13.3 x3	chr17:150721-2961603	DEL	YWHAE, PAFA
23	arr 16p13.11p12.3	chr16:15388064-NA	DUP	ABCC1, ABCC
34				
20	arr 17q12 x3;	chr17:37084992-38747513	DUP	AATF, ACACA
27	arr 18p11.21 x3	chr18:13584400-14770947	DUP	ANKRD30B, C
37	arr 22q11.21 x1	chr22:19941772-22169126	DEL	ARVCF, C22or
33	arr 22q11.21 x3	chr22:18760914-20280182	DUP	C22orf39, CDC
36	arr 22q11.21q11.23 x1	chr22:19941772-22169126	DEL	AIFM3, ARVCI

[a] The International System for Human Cytogenetic Nomenclature for array chromosomal region

[b] Build 38 hg38 chromosomal position (chromosome number: position start-position end)

[c] Copy number changes (CNVs) identified as pathogenic deletion (DEL) or duplication (DUP) CNVs

Table S1. Placental pathological lesions grouped by overarching categories.

Placental Pathological Lesion Categories	Associated Lesions
Maternal vascular malperfusion	1. Retroplacental hematoma/hemorrhage 2. Focal, multifocal, or diffuse parenchymal infarction 3. Intraparenchymal thrombus 4. Accelerated villous maturity 5. Terminal villous hypoplasia (diffuse) 6. Increased syncytial knots (Tenney-Parker changes) 7. Decidual vasculopathy
Fetal vascular malperfusion	1. Marginal insertion of umbilical cord 2. Velamentous insertion of umbilical cord 3. Furcate insertion of umbilical cord 4. Single umbilical artery 5. True knot (one or more) 6. Increased cord coiling (>3 coils/10 cm) 7. Decreased cord coiling ([?] 3 coils/10 cm) 8. Avascular villi, focal, multifocal, or diffuse 9. Villi with stromal-vascular karyorrhexis 10. Chorangiomas 11. Chorangiomatosis 12. Chorangiosis 13. Edema (hydrops)
Inflammatory lesions involving maternal compartment	1. Acute chorioamnionitis, placental membranes 2. Acute chorioamnionitis, chronic plate
Inflammatory lesions involving fetal compartment	1. Umbilical cord acute arteritis (1 vessels) 2. Umbilical cord acute arteritis ([?]2 vessels) 3. Umbilical cord acute phlebitis present

Placental Pathological Lesion Categories	Associated Lesions
	4. Umbilical cord acute vasculitis (all vessels) present 5. Acute funisitis present 6. Chorionic plate acute vasculitis 7. Villitis 8. Acute villitis
Immune/Idiopathic	Perivillous fibrin deposition Intervillous fibrin deposition Chronic intervillositis

Table S2. Associations of CNVs with placental pathological lesions in stillbirth

Placental Pathological Lesion Type	Pathogenic and VOUS CNV*	Normal CNV*	P-value*
Total stillbirth cases, N (%)	**60 (15.5)**	**327 (84.5)**	
Maternal Vascular	49 (81.7)	210 (64.2)	0.008
Fetal Vascular	47 (78.3)	252 (77.1)	0.829
Any Vascular	57 (95.0)	297 (90.8)	0.287
Maternal Inflammatory	15 (25.0)	109 (33.3)	0.204
Fetal Inflammatory	7 (11.7)	51 (15.6)	0.428
Any Inflammatory	17 (28.3)	118 (36.2)	0.241
Immune/Idiopathic	9 (15.8)	33 (10.5)	0.251

*Values are frequency, percentage, and p-value from Wald chi-square test

Table S3 . Specific pathogenic and VOUS CNVs with maternal vascular lesions in placenta of stillbirth fetuses

Gestational age	ISCN Nomenclature [a]	Chromosomal position [b]	Total size (kilo base pairs)	Copy num
27	arr 1p35.3 x1	chr1:28245805-28753654	507850	DEL
23	arr 2p16.3 x1	chr2:398808-1287541	881512	DUP
24	arr 3p21.31 x3	chr3:45789949-46439467	649517	DUP
23	arr 5p15.2 x3	chr5:10855222-11406627	551405	DUP
24	arr 6p25.1p24.3 x3	chr6:6898829-7511849	613020	DUP
23	arr 7q11.23 x1	chr7:74194985-74701057	506072	DEL
35	arr 7p12.3 x3	chr7:48088308-48659125	650946	DUP
20	arr 7p21.1 x3	chr7:18999800-19353210	353410	DUP
24	arr 10q23.3 x3	chr10:88908456-89458227	549771	DUP
23	arr 10q22.1 x3	chr10:70693170-71216306	523137	DEL
24	arr 11p13 x3	chr11:33026980-33613990	587010	DUP
27	arr 4q32.3q35.2 x1	chr4:164762764-171448183	6685419	DEL
	arr 17p13.3 x3	chr17:150721-2961603	2810882	DEL
23	arr 16p13.11p12.3	chr16:15388064-18463342	3075278	DUP
34				
20	arr 17q12 x3;	chr17:37084992-38747513	1662521	DUP
27	arr 18p11.21 x3	chr18:13584400-14770947	1186547	DUP
28	arr 19q13.12 x3	chr19:52003118-52527157	524039	DUP
31	arr 19q13.12 x3	chr19:36779213-37304888	525675	DUP

Gestational age	ISCN Nomenclature [a]	Chromosomal position [b]	Total size (kilo base pairs)	Copy num
29	arr 19p13.3 x3	chr19:422237-1310137	887899	DUP
40		chr19:490414-1310137	819722	
21	arr 21q21.3 x3	chr21:26867843-28045871	1178028	DUP
37	arr 22q11.21 x1	chr22:19941772-22169126	2227354	DEL
33	arr 22q11.21 x3	chr22:18760914-20280182	1519268	DUP
36	arr 22q11.21q11.23 x1	chr22:19941772-22169126	2227354	DEL

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