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CHROMOSOMAL MICROARRAY IN CHILDREN BORN SMALL FOR GESTATIONAL AGE – SINGLE CENTER EXPERIENCE

Perović D[1], Barzegar P[2], Damnjanović T[1], Jekić B[1], Grk M[1], Dušanović Pjević M[1], Cvetković D[3], Đuranović Uklein A[1], Stojanovski N[1], Rašić M[1], Novaković I[1], Elhayani B[2], Maksimović N[1*]

[*]**Corresponding Author:** Nela Maksimovic, PhD, University of Belgrade Faculty of Medicine, Institute of Human Genetics, Visegradska 26a, 11000 Belgrade, Serbia, Tel: +381113607052; Email: nela.maksimovic@med.bg.ac.rs

ABSTRACT

The association between small for gestational age birth and chromosomal abnormalities identified through karyotyping is well-established. Notably, advancements in cytogenetic techniques have shifted from routine karyotyping to the recommended use of microarray technology. This transition allows higher resolution and the detection of sub-microscopic copy number variants (CNVs).

Our study included 49 patients born small for gestational age, 27 males and 22 females. Clinical data were gathered from reports by clinical genetic specialists, and a questionnaire was included in the referral list to our laboratory. All participants were of pediatric age, ranging from neonatal to 12 years old. Chromosomal microarray testing was conducted by the Agilent SurePrint G3 Human CGH Microarray 8×60K.

The application of molecular karyotyping yielded clinically significant results in 16 cases (32.65%), which included 13 deletions and 6 duplications. Three patients presented with two clinically significant CNVs (csCNVs). In ten cases, we identified recurrent microdeletion or microduplication syndromes well-documented in the literature: Williams syndrome as the most commonly identified (three patients), and others like Koolen de Vries, Prader-Willi, Miller-Dieker, Dryer, DiGeorge syndrome, 7q11.23 microduplication, 16p13.11 microdeletion, and 1q21.1 microdeletion syndrome. Six patients had rare non-recurrent pathological CNVs. There was no statistically significant difference between patients with csCNVs and those without regarding the presence of intellectual disabilities, central nervous system, cardiac or skeletal malformations.

Chromosomal microarray proves to be a useful diagnostic tool in the etiology diagnosis of children born small for gestational age.

Keywords: chromosomal microarray, CNVs, small for gestational age

INTRODUCTION

Fetal growth restriction (FGR), or intrauterine growth restriction, refers to a condition where a fetus fails to reach its full growth potential [1]. Small for gestational age (SGA) is a term usually used to describe newborns (or fetuses) who weigh less than the 10th percentile of their population or customized growth charts based on gestational age [2, 3]. It is estimated that FGR impacts up to 10% of pregnancies while SGA is seen in at least 11% of newborns. It is important to note that around 40% of fetuses diagnosed as SGA do not have any underlying pathology and are simply constitutionally small in contrast to FGR where pathological mechanisms are frequently described. Therefore, SGA fetuses are not always growth-restricted and some fetuses with FGR could be appropriate for their gestational age but have not reached their maximum growth potential [2]. While there is considerable overlap between the two terms and despite existing inconsistencies in definition, most specialists use the term SGA to describe newborn size, which may or may not be linked to an underlying pathological cause. In contrast, FGR is generally caused by an antenatal pathologic disease [4].

FGR/SGA may have significant prenatal and postnatal consequences, such as increased risk of perinatal death, neurodevelopmental abnormalities, metabolic syndrome, and cardiovascular disease [5, 6]. Although the etiology and pathophysiological mechanisms can overlap, utero-placental

[1] Institute of Human Genetics, Faculty of Medicine, University of Belgrade, Serbia.

[2] Faculty of Medicine, University of Belgrade, Serbia.

[3] Faculty of Biology, University of Belgrade, Serbia.

dysfunction is the cause in the vast majority of cases of FGR [2]. However, multiple gestation, maternal disease, and structural and genetic fetal abnormalities are all possible causes [7]. Among these factors, fetal genetic defects, particularly chromosomal abnormalities, emerge as significant contributors.

The association between fetal growth impairment and chromosomal abnormalities identified through karyotyping is well-established. However, the strength of this association is significantly influenced by the gestational age at which growth impairment is identified [8], and the presence of structural fetal anomalies [9].

Over the past decades, the landscape of prenatal and postnatal screening has undergone a transformative shift, marked by advancements in technology and methodology. The introduction of the first-trimester combined test, along with other ultrasound exams during early pregnancy, has revolutionized the ability to screen for both structural and genetic abnormalities in fetuses [10]. The enhanced quality of imaging and expertise in ultrasound further contribute to the precision of assessing fetal phenotypes. Additionally, genetic testing has evolved from routine karyotyping to the recommended use of chromosomal microarray technology, enabling higher resolution and the detection of submicroscopic copy number variants (CNVs) [11]. CNVs are usually 1 kb to several Mb in length, include both duplications and deletions, and can affect single exons, one or several genes as well as regulatory sequences [12].

Through the postnatal application of CNV microarray technology, this research aims to clarify the complexities associated with small-for-gestational-age infants. It explores their phenotypic and genotypic spectrum, enhancing our knowledge of prenatal growth failure and paving the way for informed clinical decision-making and parental counseling.

MATERIAL AND METHODS

Patients

Our retrospective study included 49 patients born small for gestational age (27 males and 22 females). All patients were of pediatric age, ranging from newborn to 12 years. Their measured birth weights were below the 10th percentile for gestational age. Each patient was examined by clinical genetic specialists who provided detailed phenotypic reports. Clinical data were collected based on specialists' reports and the questionnaire included in the laboratory referral list. All guardians of the patients provided informed consent. The study was approved by the Ethics Committee Faculty of Medicine, University of Belgrade (1322/VII-4).

CNV detection and interpretation

Patients' DNA was isolated from 3-5ml of peripheral blood by the standard salting-out method [13]. The array-CGH method was performed using Agilent microarray oligonucleotide slides (SurePrint G3 Human CGH Microarray 8 × 60K) (Agilent Technologies, Santa Clara, CA, USA) according to manufacturer's protocol. Microarray slides were scanned with a DNA Microarray Scanner and data were obtained by Cytogenomic software (Agilent Technologies). Genomic positions were based on human genome reference sequence GRCh 37/hg19.

All identified copy number variations (CNVs) were analyzed and classified according to the most recent guidelines from the American College of Medical Genetics and Genomics (ACMG) [14]. The significance of these variants was evaluated based on several factors, including type (gain or loss), size, gene content (particularly dosage sensitivity), and inheritance pattern, all considered to the patient's clinical phenotype. To ensure proper classification of detected CNVs, a comprehensive review of relevant peer-reviewed literature and accessible databases such as PubMed, the Database of Genomic Variants (DGV), DECIPHER, ClinGen, and Online Mendelian Inheritance in Man (OMIM) has been performed. Benign CNVs were not reported. Pathogenic and likely pathogenic CNVs are considered clinically significant (csCNV). The diagnostic yield in our study was determined by detecting at least one csCNV in a patient.

Statistical analysis

Statistical analysis was performed by Pearson's chi-squared (χ^2) or Fisher's exact test using SPSS v.20.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

The clinical characteristics of 49 patients who were born small for their gestational age are summarized in Table 1.

Molecular karyotyping yielded clinically significant results in 16 cases, resulting in a detection rate of 32.65%. We identified 13 deletions, ranging in size from 442 kb to 15480 kb, and six duplications, 404 kb to 64280 kb in size. Additionally, three patients had two csCNVs. In ten cases, we identified CNVs linked to well-known syndromes (see Table 2). The most common was Williams syndrome,

Table 1. Overview of the phenotypic characteristics of the patient group.

Feature	Patients n=49, n (%)
male/female	27 (55.1)/22 (44.9)
DD/ID	46 (93.9)
facial dysmorphism	40 (81.6)
microcephaly	14 (28.6)
cardiac anomalies	13 (26.5)
skeletal malformations	11 (22.4)
urogenital tract anomalies	8 (16.3)

DD – developmental delay; ID – intellectual disability

Table 2. Description of the genomic imbalances classified as pathogenic/likely pathogenic

Case	Gender	Age	GW	Birth weight (g)	Region involved	Boundaries	Size (kb)	MD syndromes, OMIM #	Additional phenotype
1	f	21	≥37	2500	1q21.1-q21.2	chr1:(146564743-147786706)x1	1200	1q21.1 microdeletion syndrome, #612474	failure to thrive, microcephaly, DD
2	m	19	38	2080	2p25.3	chr2:(1842071-2246200)x3	404		Microcephaly, DD
3	f	1	38	1860	3q22.1-q29	chr3:(133562250-197840339)x3	64280		congenital heart anomaly, cleft palate
4	m	1	≥37		7p15.3-p14.3	chr7:(20993642-30739239)x1	9750		severe FGR, renal hypoplasia, ASD, shortened long bones, facial dysmorphism
5	m	32	≥37	2350	7q11.23	chr7:(72726578-74139390)x1	1412	Williams, #194050	DD, facial dysmorphism
6	m	29	≥37	2360	7q11.23	chr7:(72726578-74139390)x1	1412	Williams, #194050	DD, gastroesophageal reflux disease, pulmonary artery stenosis
7	f	2	≥37		7q11.23	chr7:(72726578-74139390)x1	1412	Williams, #194050	pulmonary artery stenosis
8	m	24	38	2700	7q11.23 22q11.21	chr7:(72726578-74139390)x3 chr22:(18919942-21440514)x1	1412 2520	7q11.23 microduplication, #609757 DiGeorge, #188400	DD, palatoschisis, hydronephrosis
9	m	1	39	2230	7q35-q36.3 16q24.1-q24.3	chr7:(143425418-158909738)x1 chr16:(86743412-90111263)x3	15480 3370		Microcephaly, hypotropia, facial dysmorphism
10	m	1	≥37		9p24.3-p22.3 19q13.33-q13.43	chr9:(271257-14956477)x1 chr19:(50380618-59092570)x3	14685 8710		facial dysmorphism, shortened long bones,cryptorchidisam CRYPTORCHIDISM hypospadias
11	f	29	31/32	1150	15q26.2-q26.3	chr15:(94447479-102383473)x1	7940	Drayer sy, #612626	DD, ASD, VSD, VUR, short stature
12	m	1	≥37	1850	16p13.11	chr16:(14910205-16525348)x1	1600	16p13.11 microdeletion syndrome	Hypotrophy, hypotonia, facial dysmorphism
13	f	121	≥37	2650	15q11.2 - q13.1	chr15:(22765628-29085896)x1	6300	Prader-Willi, #176270	DD/ID, obesity, brachydactyly
14	f	1	39	2020	17p13.3-p13.2	chr17:(51885-3882130)x1	3830	Miller-Dieker, #247200	VSD, aberrant brain MRI findings
15	m	11	38	2080	17q21.31	chr17:(43717703-44159862)x1	442	Koolen de Vries, #610443	DD, neonatal hypothroidism microcephaly, colon perforation
16	f	132	≥37		19p13.2-p13.12	chr19:(12474346-14485846)x3	2010	;	microcephaly, ASD, short stature, learning difficulties

GW – gestational week; MD – microdeletion/microduplication, DD – developmental delay; FGR – fetal growth restriction; ASD – atrial septal defect; VSD – ventricular septal defect; VUR – vesicoureteral reflux; ID – intellectual disability; MRI – magnetic resonance imaging

diagnosed in three patients (18.7%). Other syndromes previously associated with intrauterine growth restriction include Koolen de Vries syndrome, Prader-Willi syndrome, Miller-Dieker syndrome, Dryer syndrome, and 1q21.1 microdeletion syndrome. In one patient CNVs typical for DiGeorge and 7q11.23 microduplication were detected. However, in patients with either syndrome, SGA is not one of the phenotypic characteristics. Another detected microdeletion that typically does not include SGA is the 16p13.11 microdeletion. We identified unique CNVs in six cases. The main phenotypic characteristics and microarray findings of all patients with csCNVs are summarized in Table 2. Most of the patients were in neonatal age (6 of them) and besides SGA they had additional clinical features. Older children had mild to moderate developmental delay/intellectual disabilities (DD/ID) and other comorbidities.

To determine if additional specific phenotypic characteristics could predict the detection of csCNV, we analyzed the frequency of these phenotypes in two groups: those with positive molecular karyotype findings and those without. All phenotypes except skeletal malformations were more common in a group with pathogenic CNVs compared to the group with normal molecular karyotype. It was pronounced for urogenital anomalies and microcephaly, although those differences did not reach statistical significance (Table 3).

Table 3. Differences in phenotypic characteristics between patients with pathogenic CNVs and those with normal molecular karyotype

Features	Pathogenic CNVs n=16	Normal molecular karyotype n=33	p-value
Facial dysmorphism	15 (93.7)	25 (75.7)	0.238
Microcephaly	7 (37.5)	7 (18.2)	0.176
Cardiac anomalies	6 (37.5)	7 (21.2)	0.304
Skeletal malformations	2 (12.5)	9 (27.3)	0.300
Urogenital tract anomalies	4 (25.0)	4 (12.1)	0.132

DISCUSSION

The purpose of our study was to enhance the understanding of SGA in the context of chromosomal abnormalities, encompassing advancements in diagnostic methodologies with a specific focus on SGA infants. The nature of growth disturbances is highly heterogeneous making it crucial to comprehend the complex relationship between fetal growth restrictions and genetic abnormalities. Accurate diagnostic testing is vital, as a genetic diagnosis significantly influences prognosis.

The majority of our patients presented with complex forms of SGA with a lot of comorbidities, which may explain the high detection rate of positive findings: 36.4%. The detection rate is significantly higher than the 16% found in other cases tested in our laboratory. These different cases involved individuals with DD/ID, congenital anomalies, autism, and epilepsy, but without intrauterine growth restriction. It highlights the importance of SGA as a key predictive phenotype in the diagnostic yield of molecular karyotyping. Chen et al. reported a chromosomal structural copy number variation (csCNV) detection rate of 33.3% in cases of FGR associated with structural anomalies, which aligns closely with our findings [15]. In contrast, FGR cases without structural malformations, which correspond to small for gestational age (SGA) infants without additional complications, have a lower incidence of genetic abnormalities. Wu et al. conducted an analysis of 488 fetuses diagnosed with FGR but without structural malformations. They found that the diagnostic yield for classic and molecular karyotypes was 3.9% [16]. Additionally, one meta-analysis indicated a 4% increased yield of chromosomal microarray analysis (CMA) compared to classic karyotyping in non-malformed growth-restricted fetuses. Furthermore, the incremental yield of CMA in cases of FGR associated with fetal malformations was 10% [17].

In our patient group, we most frequently observed Williams syndrome (P5, P6, P7), caused by a microdeletion on chromosome 7q11.23. This syndrome is characterized by a unique combination of clinical features, including distinctive facial characteristics, cardiovascular anomalies, intellectual disability, and a remarkably sociable personality [18]. The association between Williams syndrome and intrauterine or postnatal growth failure has been well documented, highlighting its importance among different types of fetal growth restrictions. At least 82% of fetuses with typical 7q11.23 deletion have intrauterine growth retardation [19]. Our study supports these findings, emphasizing that this deletion should be considered during prenatal assessments of FGR and in cases of SGA birth.

Our study unveiled several other genetic syndromes previously associated with FGR and SGA. In a two-year-old girl (P1) born small for gestational age, with microcephaly, failure to thrive, and facial dysmorphism, we

detected a clinically significant microdeletion of 1.2 Mb in 1q21.1-q21.2 region, as well as 5p14.1-p13.3 microdeletion of 3.1 Mb, classified as a variant of unknown significance (VUS). A microdeletion detected on chromosome 1 is the recurrent deletion of distal region 1q21.1 located between breakpoints BP3-BP4 and includes the *GJA5* gene. Liu et al. summarized prenatal phenotypes characteristic for 1q21.1 microdeletions and observed IUGR in 26.7% of the cases [20]. This microdeletion has low penetrance and variable expressivity. In many cases, it is inherited from healthy parents. The second CNV, a deletion in the region 5p14.1-p13.3, encompasses ten genes, none of which are protein-coding. To our knowledge, there are no reports on the phenotype of patients with similar deletions.

One well-known syndrome associated with prenatal and postnatal growth failure is Drayer syndrome (MIM #612626), caused by a deletion in the 15q26-qter region. Patient P11 exhibited a 5.6 Mb deletion in this region (15q26.2-q26.3) and presented with developmental delay, mild facial dysmorphia, short stature, and skeletal dysplasia. Microcephaly, congenital heart disease, epilepsy, diaphragmatic hernia, renal anomalies, neonatal lymphedema, and aplasia cutis congenita could be additional characteristics of this syndrome [21]. Haploinsufficiency of the insulin-like growth factor-1 receptor (IGF1R) gene, located in this region, has been previously associated with the growth pathway and linked to impaired prenatal and postnatal growth [22]. More proximally on chromosome 15 is a region frequently linked to benign but also pathogenic CNVs, 15q11.2, which contains imprinted genes. Deletion of paternal copy of *SNRPN* and the *NDN* genes in this region cause Prader-Willi syndrome (PWS; MIM #176270). P13, from our cohort, is a 12-year-old girl with DD/ID, obesity, brachydactyly, and a 6 Mb deletion characteristic of PWS. Her obstetric history includes intrauterine growth restriction beginning in the third trimester. During her first year of life, she experienced failure to thrive but subsequently became overweight, which is typical for individuals with PWS. This syndrome is rarely diagnosed prenatally due to the lack of well-defined fetal phenotypes, which would warrant prenatal molecular genetic testing [23]. In the study by Dudley et al. focusing on prenatal, perinatal, and postnatal complications in PWS, it was observed that 29.4% (10 out of 34) of patients with PWS caused by uniparental disomy (UPD) and 42.3% (22 out of 52) of PWS patients resulting from deletion were classified as small for their gestational age [24]. The reasons for SGA in these PWS cases remain unexplained.

We identified other recurrent syndromes linked to fetal and postnatal growth restriction. This includes a 3.8 Mb microdeletion in the 17p13.3-p13.2 region, causing Miller-Dieker syndrome, and a 425 kb microdeletion in the 17q21.31 region, associated with Koolen-de Vries syndrome. Haploinsufficiency of the *PAFAH1B1* and *YWHAE* genes in Miller-Dieker syndrome is believed to cause intractable seizures, severe developmental delays, lissencephaly, facial dysmorphisms, intrauterine growth restriction, and involvement of other organ systems. Growth restriction often persists during the postnatal period [25]. In our sample, this syndrome was diagnosed in a one-month-old infant with FGR identified from the 20th week of gestation, along with facial dysmorphia and abnormal neuroimaging findings observed after birth (P14). In the case of an 11-month-old boy (P15) born SGA, with DD, microcephaly, congenital hypothyroidism, and surgically corrected colon perforation postnatally, diagnosis of Koolen-de Vries syndrome was established by CMA. This condition is multisystemic and characterized by DD/ID, epilepsy, distinct facial features, and congenital malformations affecting multiple organ systems. Research conducted by Koolen et al. on a cohort of 45 children with this syndrome revealed that 26% of cases experienced intrauterine growth retardation, 30.4% presented with low birth weight, and 41.7% also had proportionate short stature postnatally [26].

A rare and interesting example of microduplication of the 19p13.2-p13.12 region associated with impaired growth was detected in a 12-year-old girl born SGA (P16), later followed by short stature treated with growth hormone therapy. She also had microcephaly, atrial septal defect, mild ID with learning difficulties, and autistic features. Previously, in Trimouille et al., ten patients with 19p13 duplications were reported. Common clinical features included short stature, small head circumference, delayed bone age, and ID (mild to severe). Unfortunately, birth parameters were unknown for six patients, and only one had a birth weight below the 10th percentile. The research indicates that the phenotype linked to 19p13 duplication may, in some respects, be regarded as the reciprocal phenotype of Malan syndrome (MIM # 614753, previously known as Sotos syndrome-2), which is caused by heterozygous mutations, including deletions of the *NFIX* gene [27, 28]. This syndrome is characterized by DD/ID, overgrowth, macrocephaly, prominent forehead, high anterior hairline, up-slanted palpebral fissures, and prominent chin. The observed phenotype in all patients with 19p13 microduplications that include *NFIX* indicates that the phenotypes associated with both 19p13 microdeletions and microduplications may result from the contrasting effects of *NFIX* haploinsufficiency and overexpression.

Patient P3, a 1-month-old infant, has a 64 Mb duplication of the region 3q22.1-q29. The patient presented with cleft lip, and a congenital heart anomaly. In most cases, this condition is diagnosed only after birth. Individuals with this syndrome may exhibit a range of defects, including

abnormalities of the central nervous system, facial dysmorphia, congenital heart defects, urogenital tract anomalies, intellectual disabilities, and growth disturbances [29].

Our cohort revealed some recurrent or non-recurrent syndromes not previously linked to SGA. In the first group it is interesting to mention a two-year-old boy (P8) with mild DD, palatoschisis and hydronephrosis, with two recurrent CNVs: 22q11.2 deletion characteristic for DiGeorge syndrome, and 7q11.23 microduplication, reciprocal to Williams syndrome chromosomal region. In patients with DiGeorge syndrome, FGR/SGA has been noted at a rate close to the population incidence [30]. There is insufficient prenatal information available regarding 7q11.23 microduplication. In the study conducted by Wang et al., fetuses diagnosed with 7q11.23 microduplication syndrome presented with several intrauterine phenotypes, including low-lying conus medullaris, dilated ascending aorta, cleft palate, anencephaly, hydronephrosis, and other renal anomalies [31]. While the other characteristics of the phenotype could be accounted for by the presence of the two different CNVs, associated with two well-described syndromes, intrauterine growth restriction is not typically observed in these cases.

Similarly, a one-month-old infant with SGA (P12) exhibited a 1.6 Mb microdeletion in the 16p13.11 region. This CNV has been previously described and reported as a predisposition to neurodevelopmental disorders and different congenital anomalies. Short stature has been observed in several cases [32]. This deletion can occur *de novo*; however, due to its incomplete penetrance and variable expressivity, it is often inherited from a parent who is either mildly affected or presents with a completely normal phenotype. In a study by Cai et al. on the 16p13.11 microdeletion/microduplication, it was found that fetuses with CNVs in this region typically do not exhibit any characteristic features on intrauterine ultrasound and are generally healthy after birth. [33]. Therefore, the SGA observed in our case cannot be attributed to the detected CNV.

Additionally, our study identifies some csCNVs in children with SGA that have not been linked to growth restriction before, either prenatal or postnatal. A 19-month-old boy (P2), who experienced FGR, developmental delay, and microcephaly, exhibited a microduplication at chromosome 2p25.3. This microduplication includes part of the *MYT1L* gene, previously associated with neurodevelopmental disorders. *MYT1L* acts as a transcriptional repressor in neuronal progenitor cells, inhibiting Notch signaling and promoting neuronal differentiation [34]. However, there is no possible explanation for FGR in patients with deletion of this gene. In the study by Coursimaults et al., which investigated 40 children with pathogenic variants of *MYT1L* and 22 previously published patients, FGR was not frequently observed (6-8% of the patients) [35].

Patient P4 was referred for CMA due to severe FGR, born small for gestational age, with atrial septal defect, renal hypoplasia, and shortened long bones. The analysis revealed a deletion of 9.75 Mb in the region 7p15.3-p14.3, which includes 60 protein-coding genes, 16 linked to various human diseases. Crippa et al described a patient with *de novo* deletion of 7.5 Mb in the same region. This patient experienced both prenatal and postnatal growth restriction and was part of a cohort exhibiting features consistent with Silver-Russell syndrome. The authors suggested that the growth restriction might be attributed to the insulin-growth factor 2 mRNA binding protein 3 (IGF2BP3) gene (OMIM*608259) deletion and confirmed its decreased expression. This gene looks like a promising candidate for FGR since it regulates the amounts of IGF2 transcripts by encoding an RNA-binding factor unique to the 5′UTR of IGF2 mRNA [36].

We had three patients with two pathogenic CNVs on different chromosomes. In these cases, it is challenging to determine the impact of a single region or specific gene because the phenotype often results from the interaction between the two variants from different genomic regions. A newborn (P9) was found to have a 15.5 Mb deletion in the region 7q35-q36.3. This presented with microcephaly, SGA, and a progeroid appearance characterized by a "bird-like face." Fan et al. summarized the phenotypes of 17 previously reported patients with terminal deletions in the 7q35-q36.3 region, noting that 16 of these patients experienced growth restriction. Additionally, multiple congenital malformations were observed, including abnormalities in brain and facial structures, DD/ID, limb and sacral anomalies [37]. The patient in our cohort also had a 3.37 Mb microduplication in the 16q24.1-q24.3 region, which has previously been linked to mild-to-moderate ID, speech delay, and slight dysmorphic features [38]. However, there are no reports connecting this microduplication with FGR or SGA. It appears that terminal deletion on chromosome 7q has a more significant impact on growth restriction.

A newborn patient (P10) was born small for gestational age and presented with facial dysmorphism, cryptorchidism, and hypospadias. Genetic analysis revealed a terminal deletion of 14.7 Mb in the region 9p24.3-p22.3 and a terminal duplication of 8.71 Mb in the region 19q13.33-q13.43. The deleted region contains 44 protein-coding genes and 14 disease-associated. The phenotype of patients with a distal deletion of chromosome 9 includes trigonocephaly, DD/ID, and genitourinary malformations (MIM# 158170). The duplicated region on chromosome 19 contains 290 protein-coding genes, with 29 of these genes associated with diseases. Distal duplications of chromosome 19 are linked to various conditions, including low birth weight, short stature, craniofacial dysmorphia, and psychomotor delay. Additionally,

individuals may exhibit hypotonia, epilepsy, congenital heart defects, as well as urogenital and gastrointestinal malformations [39]. In this case, the expression of both pathogenic CNVs overlaps; however, the distal duplication of 19q may better explain the observed features related to SGA.

Identifying these syndromes has significant clinical implications beyond merely confirming a diagnosis. It includes early interventions to improve long-term outcomes for individuals affected by these conditions. Key management strategies for individuals with genetic syndromes include cardiac and other organ systems examination and monitoring, developmental support, and behavioral interventions. This underscores the importance of early detection and timely intervention in their care.

Most patients born SGA experience catch-up growth until they are 2 to 4 years old; however, 10% to 15% of them do not. Six of our patients with pathogenic CNVs were at neonatal age when the CMA was performed. At the same time, only five were older than two years. As a result, we have limited information regarding the postnatal growth of our patients. Among the five children older than two, short stature was documented only in two: one girl with Dryer syndrome (P11) and another with duplication at 19p13.2-p13.12 (P16). The latter patient was already receiving recombinant human growth hormone therapy (rhGH). For the past twenty years, rhGH has been approved for use in children SGA and short stature aged 2 in the USA and 4 in Europe. The response to this treatment is not always optimal, and it depends on the underlying cause of the SGA, which highlights the importance of genetic testing before starting the therapy [40].

In cases with detected csCNVs but without a clear association with SGA, it is possible that SGA may have been caused by different external environmental factors. The limitation of our study is that we did not have information about possible factors present during pregnancy that could lead to SGA. Other significant limitations are the small number of patients that were available for our research since this was a single-center study and the lack of follow-up data on the growth in the case of the children who were tested as newborns. Increasing the size of the study group and collecting information regarding the growth of children born small for gestational age would be of great importance for the following research.

In conclusion, integrated genetic testing that combines chromosomal microarray analysis with a thorough assessment of phenotypes provides valuable insights into the genetic causes of growth restriction. This method allows for the identification of clinically significant copy number CNVs and supports the development of personalized management strategies tailored to the specific needs of individuals affected by this condition.

Ethics Committee approval

The study was approved by the Ethics Committee Faculty of Medicine, University of Belgrade (1322/VII-4).

Conflict of interest

The authors declare no conflict of interest.

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