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CLINICAL REPORT

# Further characterization of the 9q31 microdeletion phenotype; delineation of a common region of overlap containing *ZNF462*

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## Abstract

**Background:** Loss of function variants and whole gene deletions of ZNF462 has been associated with a novel phenotype of developmental delay/intellectual disability and distinctive facial features. Over two dozen cases have been reported to date and the condition is now known as Weiss-Kruszka syndrome (OMIM# 618619). There are several older reports in the literature and DECIPER detailing individuals with interstitial deletions of 9q31 involving the ZNF462 gene. Many of the characteristic facial features described in these microdeletion cases are similar to those who have been diagnosed with Weiss-Kruszka syndrome.

**Methods:** We describe three additional patients with overlapping 9q31 deletions and compare the phenotypes of the microdeletion cases reported in the literature to Weiss-Kruszka syndrome.

**Results:** Phenotypic overlap was observed between patients with 9q31 deletions and Weiss-Kruszka syndrome. Several additional features were noted in 9q31 deletion patients, including hearing loss, small head circumference, palate abnormalities and short stature.

**Conclusions:** The common region of overlap of microdeletion cases implicates ZNF462 as the main driver of the recognizable 9q31 microdeletion phenotype. The observation of additional features in patients with 9q31 microdeletions that are not reported in Weiss-Kruszka syndrome further suggests that other genes from the 9q31 region likely act synergistically with ZNF462 to affect phenotypic expression.

KEYWORDS

9q31 microdeletion syndrome, Weiss-Kruszka, ZNF462

## 1 | INTRODUCTION

Developmental delay is a common phenotype that affects 1–3% of all children (Shevell et al., 2003). Published guidelines for testing have emphasized using chromosomal microarray as a first-tier genetic test for patients with neurodevelopmental phenotypes, especially when seen in conjunction with dysmorphic features and/or congenital anomalies (Bélanger & Caron, 2018; Miller et al., 2010; Mithyantha et al., 2017; Moeschler et al., 2014; O'Byrne et al., 2016). The enhanced resolution of microarray for the detection of submicroscopic deletions or duplications allows for a significantly improved diagnostic yield (10–15%) compared to more traditional cytogenetic methods (2–3%) (Miller et al., 2010). However, with improved resolution comes increased detection of rare copy number variants (CNVs) for which we do not fully understand the clinical relevance. Sharing information about CNVs and patient phenotypes can lead to the identification and characterization of new microdeletion and microduplication syndromes.

Terminal deletions of 9q have been well characterized in the literature (Kleefstra et al., 2009; Stewart & Kleefstra, 2007), yet the significance of many interstitial 9q microdeletions remains poorly understood. Emerging information about one of these interstitial microdeletions (9q31) suggests that the haploinsufficiency of this region may cause a recognizable microdeletion syndrome. To date there have been several cases reported in the literature (Cao et al., 2015; Chien et al., 2010; Dugan et al., 2018; Gamerdinger et al., 2008; Iivonen et al., 2021; Mucciolo et al., 2014; Ramineni et al., 2019) and over a dozen additional cases recorded in DECIPHER (Firth et al., 2009). These cases share many phenotypic similarities such as developmental delay/intellectual disability, hearing loss, short stature/growth retardation, microcephaly (<10th percentile) and distinctive facial features of ptosis, arched eyebrows, and ear malformations. We report here three additional patients with interstitial microdeletions of 9q31.

One gene in the common region of overlap (CRO) of these cases, *ZNF462*, has recently been implicated in a syndrome called Weiss-Kruszka syndrome (Cosemans et al., 2018; Kruszka et al., 2019; Ramocki et al., 2003; Talisetti et al., 2003; Weiss et al., 2017). Individuals with Weiss-Kruszka syndrome have distinctive facial features which can include ptosis, arched eyebrows, downslanting palpebral fissures, hypertelorism, a short upturned nose with a bulbous tip, an abnormal philtrum, and an exaggerated Cupid's bow. Global developmental delays/intellectual disability, autism spectrum disorder, ear malformations, hearing loss, craniosynostosis/metopic ridging, and congenital heart defects are also common.

The distinctive facies and clinical features of those with 9q31 microdeletion syndrome and Weiss-Kruszka syndrome are very similar. This appears to implicate haploinsufficiency of *ZNF462* as the main cause of the phenotype seen in 9q31.1q31.3 microdeletion cases, although the contribution of other genes to phenotypes in patients with a 9q31.1q31.3 deletion cannot be entirely ruled out.

## 2 | CLINICAL REPORT

Informed consent for genetic testing and publication has been obtained from each patient or their legal guardian.

### 2.1 | Patient 1

The patient was conceived naturally to non-consanguineous parents of East Indian descent. Prenatally, there was a history of decreased fetal movements during the last trimester. The patient was born by spontaneous vaginal delivery at 40 weeks of gestation with no postnatal complications. Birth weight was 6 pounds (10th percentile), and head circumference was 32 cm (5th percentile). Family history was non-contributory with respect to multiple congenital anomalies, intellectual disability, autism, seizures, neurologic disorders, metabolic disorders, recurrent pregnancy loss, or infertility. The patient has two older siblings with no developmental concerns.

Early development included a delay in speech and language, originally thought to be related to recurrent chest and ear infections beginning at 3 months of age. He achieved his first word at 18 months. His last evaluation at 4 years and 4 months of age found that the patient was able to speak in three-word sentences but was not able to tell a story or maintain a conversation. His receptive language skills appeared intact. The patient had no delays with respect to his gross and fine motor milestones; he first sat at 7 months, crawled by 9 months, and walked by 12 months. He developed a pincer grasp by 9 months with right-hand preference evident by 24 months. At 4 years of age he weighed 13.4 kg (5th percentile), height was 95.6 cm (5th percentile), and head circumference was 48.5 cm (10th percentile). He was sociable and able to maintain good eye contact but was noted to be a bit hyperactive with a short attention span. He had bilateral ptosis that was reported to be present since birth with normal extra ocular eye movements. Other facial dysmorphisms included thick and tented eyebrows, a low hairline (anterior and posterior), an upturned nose, low-set ears, depressed nasal bridge, long philtrum, and a high-arched palate with prominent tonsils. No other family members were reported to have similar facial dysmorphisms. He

also had a very small build with a slightly prominent anteroposterior (AP) diameter. His neurological examination including motor examination, reflexes, sensory, gait, and coordination was normal and his skin did not show any abnormal pigmentation.

Chromosomal microarray analysis detected a copy number loss at 9q31.1q31.3 that was approximately 5849 kb in size (arr[hg19] 9q31.1q31.3(106,635,504_112,484,507) × 1) (Figure 1A). The deletion was further confirmed by G-banding and metaphase FISH (Figure 1A: a,b). Maternal studies using the same FISH probe showed an abnormal signal pattern suggesting the insertion of the 9q31.1q31.3 region into the short arm of chromosome 9. An apparently balanced intrachromosomal insertion of 9q31.1q31.3 material into cytoband 9p13 was further confirmed by G-band analysis in the maternal sample (Figure 1A: c,d). Based on the FISH and G-band results, the abnormal chromosome 9 in Patient 1 is interpreted as a recombinant chromosome from maternal intrachromosomal insertion resulting in loss of the 9q31.1q31.3 segment without the concomitant insertion into 9p.

## 2.2 | Patient 2

Patient 2 is a 33-year-old female with cognitive impairment and dysmorphic features. She was born by cesarean section with a birth weight of 7.25 pounds (50th percentile). She was admitted to the hospital at 4 months of age because of an umbilical hernia. At admission, she was noted to have generalized hypotonia with poor head control when pulled from a supine to a sitting position. Chest, skull, and T-L spine radiographs identified 13 ribs and a brachycephalic skull. Dysmorphic facial features were also noted including bilateral ptosis and micrognathia. Assessment at two and a half years of age found a normal height and weight with a head circumference of 46 cm (10th percentile). Facial features included bilateral ptosis, a short upper lip, a high-arched palate, and an elongated upper middle incisor. The other upper middle incisor had been lost.

Clinical history was significant for surgeries to correct the umbilical hernia and bilateral ptosis. Bilateral ear canal tubes were inserted at 2 years of age and follow-up audiogram testing showed normal hearing in at least the right ear though reports of 'mild conductive hearing loss' were also noted in the patient record.

Neurodevelopmental examination showed that she functioned at 15–24 months as measured by the revised Yale schedule and expressive and receptive abilities did not seem to exceed the 15–18 month level. She had been enrolled in an integrated program in grade school and could read but had limited math skills. She was overly friendly and spoke with short, individual words. Attention deficit and impulsivity were also noted. She had episodes of petit mal seizures. There is no information available regarding the age of puberty onset.

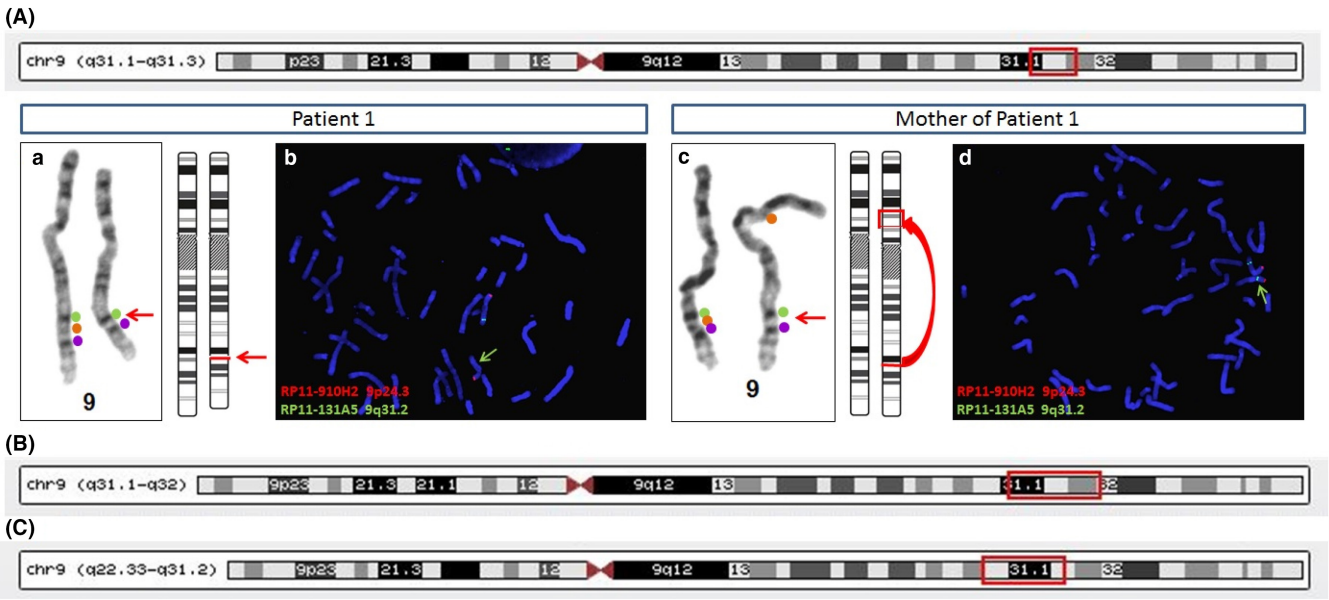



FIGURE 1 Interstitial 9q deletions spanning the 9q31 region of chromosome 9 in patients 1, 2, and 3. A: Region of chromosome 9 material loss (9q31.1q31.3) in patient 1. (A) a,b; confirmation of 9q31.1q31.3 deletion in patient 1 by G-banding and metaphase FISH (red and green arrows). (A) c,d; maternal G-banding and metaphase FISH showing a balanced intra-chromosomal insertion of the 9q31.1q31.3 region into the short arm of chromosome 9. Metaphase FISH probes are labeled red (chromosome region 9p24.3) and green (chromosome region 9q31.2) at the bottom-left of each FISH panel. (B) Region of chromosome 9 material loss (9q31.1q32) in patient 2. (C) Region of chromosome 9 material loss (9q22.33q31.2) in patient 3

At 33 years of age, her height and weight were measured at the 5th and greater than the 95th percentile, respectively. Her head circumference was 53.5 cm (20th percentile). Physical examination was significant for a short neck and normal hands with tapered fingers. Her feet appeared normal but she had clinodactyly of the 4th and 5th toes. Hearing aids were prescribed but were not often being worn. Facial features included a redundant chin, normal lips, prominent nose, short philtrum, a low narrow forehead, and hypotelorism.

Chromosomal microarray analysis detected a loss of material from chromosome region 9q31.1q32 (arr[hg19] 9q31.1q32(103,669,815_115,515,202) × 1) that was approximately 11,845 kb in size and is predicted to result in deletion of greater than 70 RefSeq genes (Figure 1B). The 9q31.1q32 deletion was further confirmed by karyotype analysis of G-banded metaphase chromosomes. Parental karyotype testing was normal suggesting that this deletion occurred de novo in this individual.

### 2.3 | Patient 3

Patient 3 is a 13-year-old boy born at term by spontaneous vaginal delivery. His birthweight was 7.3 pounds (<50th percentile). He was observed in the neonatal intensive care unit for 4 days following delivery because of possible sepsis. An echocardiogram at the time revealed an atrioventricular septal defect with a large atrial component, which has since been corrected surgically.

Failure to thrive, feeding difficulties, gastrointestinal reflux, and hepatomegaly were noted at 4 months of age, and hypothyroidism was more recently diagnosed. Head circumference and weight were at the 10th percentile but length remained stable at the 50th percentile. At the age of nine, he was enrolled in grade 4 but was functioning at a senior kindergarten/grade 1 level. Central obesity was noted with a weight at >95th percentile, height at the 70–95th percentile, and head circumference at +1 standard deviation. He had hypoplastic toenails, small calves, a broad forehead, downslanting palpebral fissures, hypotelorism, a long columella extending below the alae nasi, and normal lips.

At his most recent examination at the age of 12 years, his height and weight were at the 97th percentile and his head circumference remained at +1 standard deviation. A normal head shape was noted. Downslanting eyes and hypotelorism were noted again, with a normal nose and philtrum. He had slender hands and fingers and tapered toes with hypoplastic nails on the 3-5th toes, particularly the 5th toe.

Cytogenetic investigation of G-banded metaphases detected a large interstitial deletion from the long arm of chromosome 9 (46,XY,del(9)(q22.32q31.3)). Chromosomal microarray analysis detected a loss of material from chromosome region 9q22.33q31.22 that is approximately 10,727 kb in size (arr[hg19] 9q2 2.33q31.2(99,302,837_110,030,098) × 1) (Figure 1C). Parental karyotypes were normal suggesting that the deletion had occurred de novo.

## 3 | METHODS

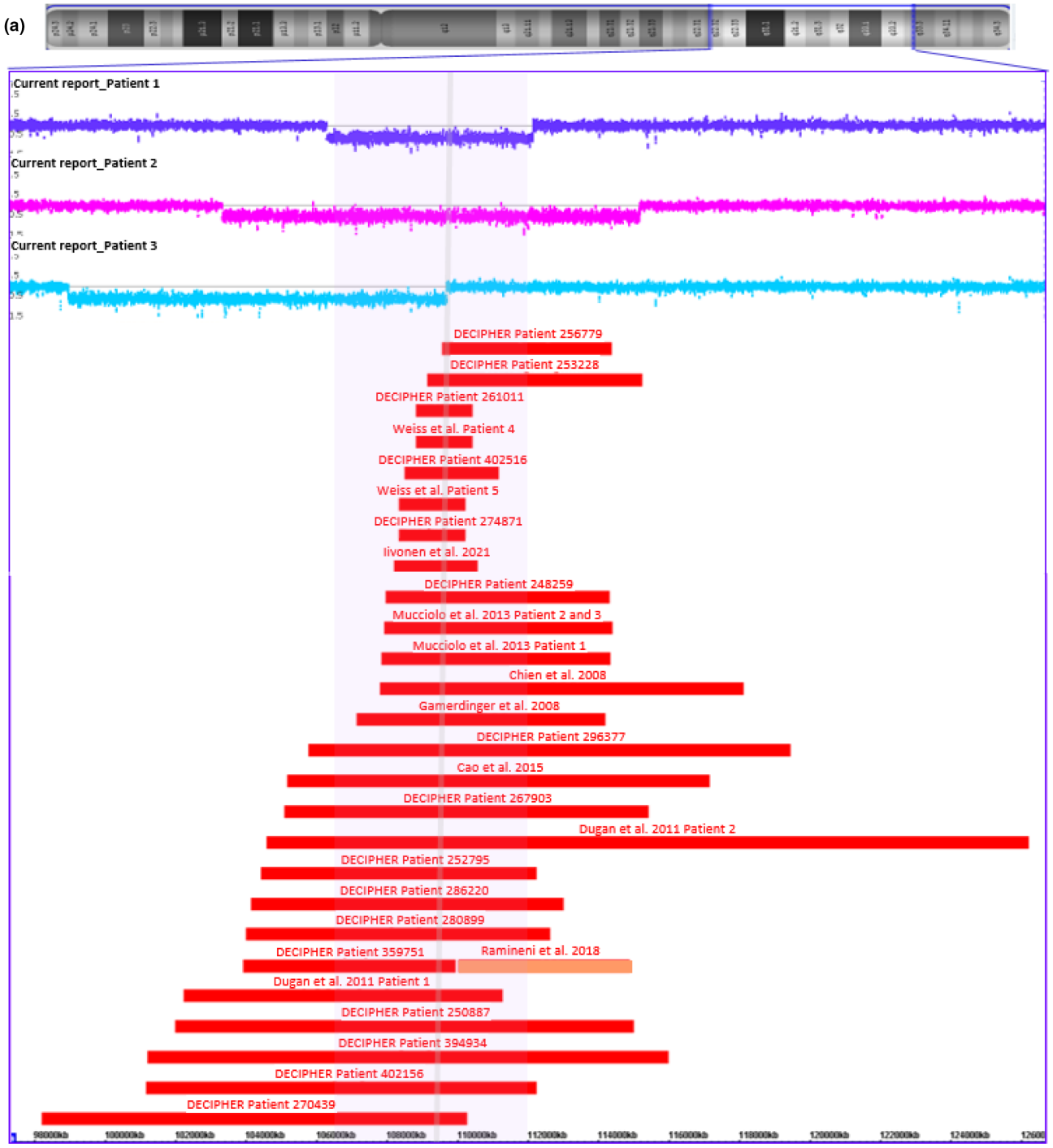
### 3.1 | Chromosomal microarray

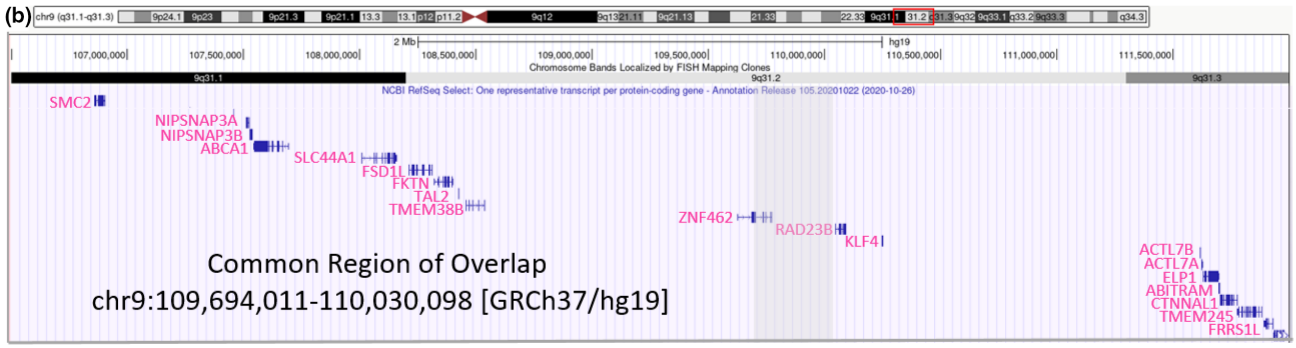
Genomic DNA was extracted from peripheral blood using the QIAamp DNA Purification Kit and the QIAcube automated extractor (QIAGEN, Hilden, Germany) as per the manufacturer's instructions. Extracted DNA was subsequently assessed for copy number variation using the Cytoscan HD Cytogenetics Solution platform (ThermoFisher Scientific, Waltham, MA, USA) as per the manufacturer's recommendations. Array findings were assessed using Chromosome Analysis Software, version 2.0.0 (ChAS), and reported at a resolution of 200 kb for copy number loss and 500 kb for copy number gains across the genome. Regions known to be clinically significant were analyzed at an increased resolution (at least 200 kb gains; 50 kb losses). Detected variants below these size limits were also assessed assuming sufficient data quality and probe density across the variant region. Additionally, results were assessed for long contiguous stretches of homozygosity of >3 Mb. The analysis was based on the human genome build 19 (GRCh37/hg19, February 2009).

### 3.2 | Karyotype and fluorescence In-Situ hybridization

Phytohemagglutinin (PHA)-stimulated peripheral blood cultures were synchronized by treatment with excess

**FIGURE 2** Comparing chromosome 9 deleted region of patients 1, 2, and 3 to that of published reports and DECIPHER cases. (a) Chromosomal microarrays showing copy number loss of the 9q31 region in patients 1, 2, and 3 are demonstrated in green, orange, and blue logarithm (Log2) fluorescent ratios, respectively (top of panel). Other reported and DECIPHER cases are aligned at the bottom (red rectangles). Comparison narrows down the common region of overlap (CRO) to approximately 345 kb (chr9: 109,694,706 - 110,039,595) (gray bar). One reported 9q31 cases that do not include the CRO but have a proximal breakpoint that neighbors the CRO is highlighted in orange. A view of the genes from the chromosomal segment highlighted in a light purple box is shown in Figure 2b. (b) Zoom-in of the CRO region showing *ZNF462* and neighboring protein-coding genes. The CRO is highlighted in a gray box


(a)
Current report_Patient 1
Current report_Patient 2
Current report_Patient 3
DECIPHER Patient 256779
DECIPHER Patient 253228
DECIPHER Patient 261011
Weiss et al. Patient 4
DECIPHER Patient 402516
Weiss et al. Patient 5
DECIPHER Patient 274871
Iivonen et al. 2021
DECIPHER Patient 248259
Mucciolo et al. 2013 Patient 2 and 3
Mucciolo et al. 2013 Patient 1
Chien et al. 2008
Gamerdinger et al. 2008
DECIPHER Patient 296377
Cao et al. 2015
DECIPHER Patient 267903
Dugan et al. 2011 Patient 2
DECIPHER Patient 252795
DECIPHER Patient 286220
DECIPHER Patient 280899
DECIPHER Patient 359751
Ramineni et al. 2018
Dugan et al. 2011 Patient 1
DECIPHER Patient 250887
DECIPHER Patient 394934
DECIPHER Patient 402156
DECIPHER Patient 270439


(b)
SMC2
NIPSNAP3A
NIPSNAP3B
ABCA1
SLC44A1
FSD1L
FKTN
TAL2
TMEM38B
ZNF462
RAD23B
KLF4
ACTL7B
ACTL7A
ELP1
ABITRAM
CTNNAL1
TMEM245
FRRS1L
Common Region of Overlap
chr9:109,694,011-110,030,098 [GRCh37/hg19]

thymidine overnight before washing the cultures. Cultured cells were further incubated with ethidium bromide before adding colcemid to block mitotic cells in metaphase. Cell pellets were treated with potassium chloride and fixed using 3:1 methanol: glacial acetic acid. Metaphase spreads were treated with trypsin and banded with Giemsa stain using routine cytogenetic protocols.

Cultured lymphocytes from Patient 1 and her mother were also assessed by fluorescence in-situ hybridization using a BAC probe that maps to 9q31.2 (BAC RP11-131A5* The Centre for Applied Genomic, Toronto, ON, probe coordinates—chr9:110,001,016-110,163,524 hg19 genome build). Hybridization to the correct chromosomal location was established by the use of an internal probe and by assessment of metaphase under reverse-DAPI imaging.

## 4 | DISCUSSION

We present three patients with intellectual disability/global developmental delay and craniofacial dysmorphisms with interstitial deletions of the 9q31 chromosome region. Deletions in this region are relatively uncommon and there have been less than three dozen cases reported in the literature (Cao et al., 2015; Chien et al., 2010; Dugan et al., 2018; Gamerdinger et al., 2008; Iivonen et al., 2021; Mucciolo et al., 2014; Ramineni et al., 2019; Xu et al., 2013) and DECIPHER database (Firth et al., 2009) (DECIPHER ID: 270439, 250,887, 261,011, 253,228, 256,779, 286,220, 280,899, 252,795, 267,903, 296,377, 248,259, 274,871, 359,751, 394,934, 402,156, 402,516) (Figure 2). Common craniofacial dysmorphisms in this cohort include bilateral ptosis, arched eyebrows, a broad nasal root, low forehead, low set ears, anteverted nares, palate abnormalities, and a long philtrum. Many patients are also reported to have developmental delays/intellectual disability, short stature, congenital cardiac anomalies, minor limb anomalies, and hearing loss (Table 1 and Supplementary Table S1). Most 9q31 microdeletions are reported to have occurred de novo or are derived from a balanced chromosome rearrangement in either parent. These similarities suggest that microdeletions of the 9q31chromosome region cause a recognizable microdeletion syndrome.

In 2015, Cao et al. (2015) recognized several common features between patients with these 9q31 deletions and Cornelia de Lange Syndrome, notably developmental delay, distinctive facial features, growth and cognitive retardation, and limb anomalies. Based on these similarities they suggested two possible candidate genes, *SMC2* and *WHRN*, as the main drivers of the 9q31 microdeletion phenotype. *SMC2* was a particularly attractive candidate as it encodes a protein belonging to the cohesion complex. Pathogenic variants in several other cohesion complex genes are associated with Cornelia de Lange syndrome, including *NIPBL, SMC1A, SMC3, HDACB, BRD4*, and *RAD21* (Deardorff, Bando, et al., 2012; Deardorff, Wilde, et al., 2012; Gil-Rodríguez et al., 2015; Kline et al., 2018; Musio et al., 2006; Olley et al., 2018). However, when compared to a large cohort of 9q31 patients, both *SMC2* and *WHRN* mapped outside the common region of overlap (CRO) in many patients (Figure 2).

Recently, additional information about another gene within the CRO has become available. The *ZNF462* gene encodes for a zinc finger protein that appears to play an important role in embryonic development and chromatin remodeling (Eberl et al., 2013). Several publications have reported that loss of function variants, deletions, and chromosome translocations disrupting *ZNF462* cause a recognizable syndrome with developmental delay, autism spectrum disorder, feeding difficulties, hypotonia, and craniosynostosis. Characteristic facial features include ptosis, arched eyebrows, abnormal philtrum, short upturned nose with bulbous tip, and downslanting palpebral fissures (Cosemans et al., 2018; Deardorff, Bando, et al., 2012; Kruszka et al., 2019; Talisetti et al., 2003; Weiss et al., 2017). This condition is now known as Weiss-Kruszka syndrome.

A comparison of 9q31 microdeletion cases to Weiss-Kruszka syndrome shows many overlapping similarities in phenotype (Table 1). In comparing the frequencies of these phenotypes it is important to note that since many of the 9q31 microdeletion cases were collected from DECIPHER the clinical information may not be complete and has not been validated. Despite these limitations, similar craniofacial features include ptosis, arched eyebrows, abnormal philtrum/lips, craniosynostosis, hypertelorism, short upturned nose with a bulbous tip, global developmental delay/intellectual disability, cardiac abnormalities, autism spectrum disorder, and hypotonia. The striking similarities in phenotypes of Weiss-Kruzka syndrome and 9q31 microdeletion cases, and the observation that *ZNF462* is in the CRO of almost all published/DECIPHER cases (Figure 2), strongly implicates *ZNF462* as the main driver of the 9q31 microdeletion phenotype.

Despite the remarkable similarities between the features of patients with 9q31 deletions and Weiss-Kruzka syndrome, there are some differences, and it is presently unclear if the cases with 9q31 microdeletions should be diagnosed with Weiss-Kruszka syndrome or if they should be classified as a separate microdeletion syndrome. For example, though dysmorphic/low-set ears appear to be seen in about half of 9q31 microdeletion and Weiss-Kruszka patients, hearing loss appears to be significantly more common in the 9q31 microdeletion

**TABLE 1** Summary of clinical features in Weiss-Kruszka and 9q31 microdeletion syndrome

| | Weiss-Kruszka syndrome | 9q31 microdeletion syndrome | Present case 1 | Present case 2 | Present case 3 |
|---|---|---|---|---|---|
| **Region** | | | 9q31.1q31.3 | 9q31.1-q32 | 9q22.33-q31.22 |
| **Deletion Size (Mb)** | | | 5.85 | 11.85 | 10.7 |
| **Deletion Coordinates** | | | chr9:106,635,504-112,484,507 | chr9:103,669,815-115,515,202 | chr9:99,302,837-110,030,098 |
| **Inheritance** | | | Familial ins (9;9) | De novo | De novo |
| **Phenotype** | | | | | |
| Hypotonia | 50% | 19% | − | + | |
| Autism Spectrum Disorder | 33% | 11% | − | − | − |
| Developmental delay/Intellectual Disability | 79% | 78% | + | + | + |
| Cardiac abnormalities | 21% | 44% | | | + |
| Feeding Problems | 50% | 15% | − | | + |
| Abnormal Brain MRI | 25% | 15% | | | |
| Hearing Loss | 14% | 41% | + | + | |
| Minor Limb Anomalies | 25% | 41% | − | + | + |
| Postnatal Growth Retardation/Short Stature | Not Reported | 48% | + | + | − |
| Other | | | | third pair of ribs, seizures | gastrointestinal reflex, hepatomegaly, hypothyroidism |
| **Craniofacial Features** | | | | | |
| Abnormal Philtrum/Lips | 54% | 30% | + | + | − |
| Arched Eyebrows | 50% | 22% | + | | |
| Ptosis | 83% | 52% | + | + | |
| Craniosynostosis/Abnormal Skull Shape | 33% | 19% | | + | − |
| Hypertelorism | 25% | 11% | | − | |
| Dysmorphic/Low Set Ears | 50% | 37% | + | | |
| Short upturned nose with bulbous tip | 46% | 26% | + | + | |
| Broad/ flattened nasal bridge | Not Reported | 30% | + | | |
| Small Head Circumference (<10th percentile) | Not Reported | 26% | + | + | + |
| High arched palate/cleft palate | Not Reported | 19% | + | + | |

*Note*: Common features reported in Weiss-Kruszka syndrome and 27 patients with 9q31microdeletion overlapping ZNF462 (Cao et al., 2015; Chien et al., 2010; Dugan et al., 2018; Firth et al., 2009; Gamerdinger et al., 2008; Iivonen et al., 2021; Mucciolo et al., 2014; Ramineni et al., 2019; Xu et al., 2013). More detailed information on clinical features previously reported in 9q31 microdeletion patients is available in supplemental Table S1. Although included in the calculations for 9q31 microdeletion syndrome, clinical features observed in the current case are also provided for comparison.

group (Table 1). Small head circumference (<10th percentile), palate abnormalities, and postnatal growth retardation/proportionate short stature were seen in the microdeletion cases that involved *ZNF462* but were not reported in patients with Weiss-Kruzka syndrome (Cosemans et al., 2018; Kruszka et al., 2019; Talisetti et al., 2003; Weiss et al., 2017) (Table 1). Of potential interest, seizures were reported in Patient 2 from this cohort but have not been reported in others with 9q31 deletions or Weiss-Kruzka syndrome so it remains unclear whether this is an unrelated finding or a rare phenotype. The observation of these different clinical features in patients with 9q31 deletions in comparison to Weiss-Kruszka syndrome could suggest that there may be additional genes in the 9q31 region that act synergistically with *ZNF462* to affect phenotypic expression.

The hypothesis that 9q31 deletions could represent a contiguous gene disorder may be supported by a family of individuals with a 4.8 Mb deletion (9q31.2-9q32) that does not appear to include *ZNF462* (Ramineni et al., 2019). While this family does not appear to have the same cognitive delays or intellectual disability that is common to both the cohorts of patients with a 9q31 deletion that involves *ZNF462* and Weiss-Kruzka syndrome there are still some overlapping features such as cardiac abnormalities, sensorineural hearing loss, and postnatal growth retardation/proportionate short stature (Ramineni et al., 2019). However, the reported 9q31 deletion breakpoints in this family are close to the *ZNF462* region and therefore disruption of gene expression cannot be completely ruled out without further investigation (Figure 2). Characterization of additional patients with 9q31 deletions that do not include *ZNF462* is needed to better understand how this and neighboring genes contribute to the phenotypic spectrum in these patients.

In summary, there have been 28 cases with interstitial deletions of the 9q31chromosome region reported in the literature and DECIPHER database. These cases share many phenotypic similarities to Weiss-Kruszka syndrome, caused by loss of function variants in *ZNF462. ZNF462* is in the CRO of all but one of these cases, suggesting that it is the main driver of the phenotype seen in 9q31 microdeletion cases. Unlike Weiss-Kruszka syndrome, patients with 9q31 microdeletions were reported to have short stature, small head circumference, and palatal abnormalities. Other phenotypes, such as hearing loss, also appear to be more common in the 9q31 microdeletion cohort. It is presently unclear if this is influenced by other genes in the 9q31 region. The observation of derivative or recombinant chromosomes involving the 9q31 region, such as the recombinant chromosome observed in Patient 1 and other reported cases (Chien et al., 2010; Cosemans et al., 2018) further highlights the importance of cytogenetic methods (FISH or G-banding) to identify parental chromosome rearrangements that may increase the recurrence risk for microdeletion/microduplications of this region.

## AUTHOR CONTRIBUTION

Conceptualization, Lauren Brady and Elizabeth McCready; Formal analysis, Lauren Brady, Mark Ballantyne, John Duck, Thomas Fisker, Ryan Kleefman, Chumei Li, Lee-Anne Schultz; Data curation, Lauren Brady and Elizabeth McCready; Writing - original draft preparation, Lauren Brady and Landry Nfonsam; Writing - review and editing, Lauren Brady, Landry Nfonsam, Lee-Ann Schultz, Chumei Li, Mark Tarnopolsky, Elizabeth McCready.

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## CONFLICTS OF INTEREST

The authors declare not conflict of interest.

## ETHICS STATEMENT

Informed consent was obtained from all subjects involved in the study.

## DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available in DECIPHER at https://www.deciphergenomics.org/. These data were derived from the following resources available in the public domain: DECIPHER, https://www.deciphergenomics.org/.

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## SUPPORTING INFORMATION

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