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Case Report

Three pediatric patients with dual rare genetic diagnoses: genetic and clinical findings

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Abstract: In certain clinical scenarios, a single diagnosis may be insufficient or even inadequate to fully explain complex or atypical phenotypes. Herein, we present three pediatric cases diagnosed with dual rare genetic disorders and analyze their medical histories and diagnostic trajectories. A total of nine gene mutations were detected, among which seven were novel, including c.[791T>C];[695G>A] in *DNAH1*, loss2(EXON:3-5) in *SGCB,* c.[1A>G] (reported);[1024A>G] (reported) in *RARS2*, c.[962-1G>T];[592A>T] in *KIAA0586*, and c.358C>T in *IRF2BPL*, c.2714C>T in *KDM6A*.

Keywords: Dual diagnoses, dual rare genetic disorders, whole-exome sequencing

Introduction

When clinical assessment or standard testing can not provide an accurate diagnosis, genetic testing can provide it, and as a result, more accurate clinical care. Whole-exome sequencing (WES), the most up-to-date genetic sequencing, has become one of the most important advances in genetics in the past 1-2 decades in the medical profession [1]. Dual genetic diagnoses are common, but have been underreported: a recent prospective study showed 4.9% of patients diagnosed by WES had multiple genetic diagnoses [2]. In particular, a comprehensive molecular work-up in cases of dual diagnoses can not only improve clinical management of the patients, but also provide prognostic insight and inform individualized treatment. We present detailed clinical evaluations of three pediatric patients with dual genetic diagnoses.

Whole-exome sequencing found six diseases associated with these patients: Primary ciliary dyskinesia-37 and Limb-girdle muscular dystrophy-4 in case 1, Pontocerebellar hypoplasia-6 and Joubert syndrome-23 in case 2, and Neurodevelopmental disorder with regression, abnormal movements, loss of speech, and seizures (NEDAMSS) and Kabuki syndrome-2 in case 3. A total of nine gene mutations were detected, including c.[791T>C];[695G>A] in *DNAH1*, loss2(EXON:3-5) in *SGCB*, c.[1A>G]; [1024A>G] in *RARS2*, c.[962-1G>T];[592A>T] in *KIAA0586*, and c.358C>T in *IRF2BPL*, c.2714C>T in *KDM6A*.

The study demonstrated the essential aspect of carrying out genetic counseling in prenatal and neonatal screening programs. When a diagnosis is unclear or the analysed primary pathogenesis does not fully explain the phenotype in a pediatric patient, then whole-exome sequencing should be used as a first-tier diagnostic method to address the underlying etiology and stimulate further rehabilitative and healthcare practices.

Case

Case 1: Primary ciliary dyskinesia type 37 and limb-girdle muscular dystrophy type 2E

A 5-month-old male infant was initially admitted to our hospital due to "diarrhea for over one month and hematochezia for 5 days". The infant presented with mucus-flecked and bloody stools, suggestive of intestinal inflammation.

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Stool culture was positive for pathogenic Escherichia coli and Clostridioides difficile, and rotavirus antigen testing was also positive. Following treatment with cefazolin for infection and rehydration therapy, and a switch to an extensively hydrolyzed formula, the symptoms of diarrhea and hematochezia improved. However, besides diarrhea, the infant also presented with symptoms including cough, wheezing, and hearing impairment. Respiratory pathogen testing (nucleic acid detection) was negative. Brainstem auditory evoked potentials (BAEP) identified mild left-sided conductive hearing loss. Peripheral blood liver enzymes were elevated (Alanine Aminotransferase 142 U/L, Aspartate Aminotransferase 164 U/L), and creatine kinase was markedly elevated (5086 U/L, normal range: 50-310 U/L). Electromyography (EMG) showed mild myogenic damage with electrophysiological changes, which were more pronounced in proximal muscle groups. The infant's cough was alleviated after combined nebulization therapy with budesonide, ipratropium bromide, and terbutaline. However, the liver enzymes and creatine kinase levels remained abnormal (**Table 1**).

Given the infant's multi-system and multi-organ involvement, his parents opted for whole-exome sequencing (WES). WES identified compound heterozygous variants in DNAH1 c.[791T>C];[6958G>A], p.[Met264Thr];[Gly2320Ser] (**Figure 1A**). The presence of mild conductive hearing loss and cough with wheezing corroborated the diagnosis of primary ciliary dyskinesia type 37 [3]. Additionally, a homozygous exonic deletion (exons 3-5) was detected in SGCB, which was inherited from both parents who were carriers of this variant (**Figure 1B**). Elevated liver enzymes, creatine kinase levels, and abnormal electromyography findings supported the clinical diagnosis of Limb-Girdle Muscular Dystrophy Type 2E [4] (**Table 2**, Supplementary Table 1).

At present, no disease-modifying therapies are available for either of these two genetic conditions. Under medical supervision, the parents adopted a structured regimen of enhanced supportive care and rehabilitative exercises. In Primary Ciliary Dyskinesia Type 37, management centers on airway clearance strategies aimed at preventing respiratory infections and controlling cough and wheezing. For Limb-Girdle Muscular Dystrophy Type 2E, essential measures include tailored nutritional support and muscle-focused rehabilitation. Both conditions contribute to ineffective airway clearance, through ciliary dysfunction and respiratory muscle weakness, respectively making comprehensive respiratory management the top priority. Due to early detection and timely intervention, the infant's condition has not progressed to date.

Case 2: Pontocerebellar hypoplasia type 6 and Joubert syndrome-23

Case 2 was a female infant who presented to our hospital at 1 month of age. Her clinical manifestations included emesis, poor sucking and swallowing ability, and diminished mental responsiveness accompanied by lethargy. During the course of the disease, she experienced transient apnea and limb tremors (suspected seizures). Video-electroencephalography (EEG) revealed a severely abnormal neonatal EEG pattern, characterized by marked delays in electrographic development and widespread multifocal sharp waves predominantly over the Rolandic regions during sleep. Treatment with a combination of phenobarbital and topiramate was initiated for seizure control, with suboptimal effect. By 3 months of age, the infant exhibited epileptic seizures (manifested as nodding and embracing - like movements, stiffness of the extremities, staring, smiling, and fixed gaze of both eyes, each episode lasting 2-10 minutes). Antiseizure medications, including levetiracetam, vigabatrin, and lacosamide, were sequentially administered. Seizure frequency subsequently improved following combination therapy with topiramate and lacosamide, although breakthrough events still occurred occasionally. At 3 months of age, the child had unstable head - lifting and was unable to track visual and auditory stimuli. The body weight was 4.2 kg, which was at the third percentile of the weight for the same gender and age, suggesting severe growth and development retardation. At 11 months of age, the child still had feeding difficulties and poor appetite, and nasogastric and gastric tube feeding were initiated. Physical examination showed low muscle tone in the extremities. Cranial magnetic resonance imaging (MRI) demonstrated atrophic changes in both cerebral and cerebellar hemispheres, including widened and deepened cerebral sulci, reduced cerebellar volume, enlarged cisterna magna

Table 1. Clinical profiles of three patients

Clinical feature	Patient 1	Patient 2	Patient 3
Gender	M	F	F
Age at diagnosis	5 months	1 month	3 days
Genetic disease	primary ciliary dyskinesia type 37	pontocerebellar hypoplasia type 6	Neurodevelopmental disorder with regression, abnormal movements, loss of speech, and seizures
	limb-girdle muscular dystrophy type 2E	Joubert syndrome-23	Kabuki syndrome-2
Neurology	no relevant symptoms	epileptic seizures, General developmental delay, hypotonia	poor responsiveness, hypotonia
Digestive	liver dysfunction, diarrhea, hematochezia	feeding difficulties, poor appetite, Gastric volvulus	no relevant symptoms
Other features	cough, wheezing, hearing impairment	cranial MRI demonstrated bilateral cerebral and cerebellar hemisphere atrophy, and enlarged posterior cranial fossa cisterns	pleural effusion

Table 2. Genotype profiles of three patients

Patient	Gene	Nucleotide substitution	Amino acid substitution	Parental origin	Novel/Reported	ACMG criteria
Case 1	DNAH1	c.791T>C	p.Met264Thr	Father	Novel	VUS: PM2_Supporting+PP3
	DNAH1	c.6958G>A	p.Gly2320Ser	Mother	Novel	VUS:
	SGCB	loss2(EXON:3-5)	-	Father & Mother	Novel	LP: PVS1+PM2_Supporting
Case 2	RARS2	c.1A>G	p.Met1?	Father	Reported	P: PVS1+PM2_Supporting+PM3_Strong+PP1
	RARS2	c.1024A>G	p.Met342Val	Mother	Reported	VUS: PM2_Supporting+PM3+PP3
	KIAA0586	c.962-1G>T	-	Father	Novel	LP: PVS1+PM2_Supporting
	KIAA0586	c.592A>T	p.Ser198Cys	Mother	Novel	VUS: PM2_Supporting+PM3+PP3
Case 3	IRF2BPL	c.358C>T	p.Gln120Ter,677	NA	Novel	LP: PVS1+PM2_Supporting
	KDM6A	c.2714C>T	p.Thr905Ile	NA	Novel	VUS: PM2_Supporting+PP3

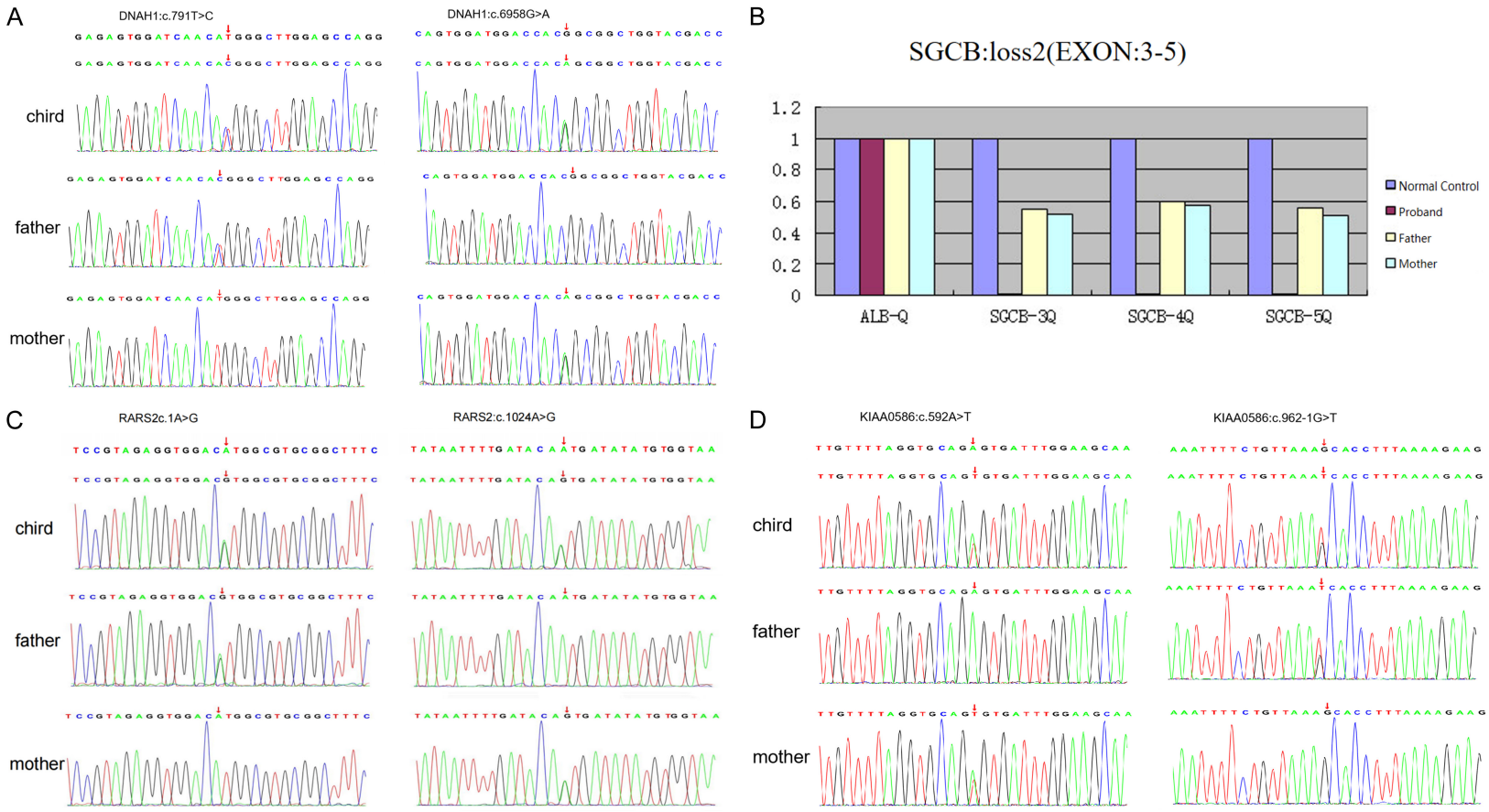

Figure 1. Results of whole-exome sequencing. A: WES detection results of c.[791T>C] and [695G>A] in *DNAH1*. B: The verification quantitative results of the parental carrier about loss2(EXON:3-5) in *SGCB*. The normal control samples, the proband and the family samples were subjected to the same group of fluorescence quantitative PCR by using the ALB gene as the internal reference gene. The ratio in the proband to that of the normal control was approximately 0, suggesting that there was a homozygous deletion in the 3-5 exons of the SGCB gene. The ratio in the parents to that of the normal control was approximately 0.5, suggesting that there was a heterozygous deletion in the 3-5 exons of the SGCB gene. The X-axis represents the ratio of exon copy number of the normal control samples, the proband and the family samples to that of normal control. C: WES detection results of c.[1A>G] and [1024A>G] in *RARS2*. D. WES detection results of c.[962-1G>T] and [592A>T] in *KIAA0586*.

and retrocerebellar spaces, and deepened cerebellar folia (**Table 1**).

Given the multisystem involvement observed at 1 month of age, the parents opted for whole-exome sequencing. WES identified compound heterozygous variants in the *RARS2* gene c.[1A>G];[1024A>G] (**Figure 1C**), supporting the diagnosis of pontocerebellar hypoplasia type 6 [5]. Additionally, compound heterozygous variants in the *KIAA0586* gene c.[962-1G>T];[592A>T] were detected (**Figure 1D**), confirming the second diagnosis of Joubert syndrome type 23 [6] (**Table 2**, Supplementary Table 1).

Pontocerebellar hypoplasia type 6 most commonly presents with refractory epilepsy, feeding difficulties, and severe developmental delay, while Joubert syndrome type 23 is mainly characterized by central nervous system developmental delay and cerebellar dysplasia. Based on the overall clinical presentation, we consider pontocerebellar hypoplasia type 6 to be the primary condition contributing to the patient's phenotype, with Joubert syndrome type 23 likely exacerbating the severity of refractory epilepsy and developmental delay. Following diagnostic confirmation, antiseizure medications were continuously adjusted. By around 8 months of age, a final antiepileptic regimen was established, leading to relative control of seizure symptoms. Timely initiation of nasogastric feeding also contributed to some mitigation of the severity of her developmental delay.

Case 3: Neurodevelopmental disorder with regression, abnormal movements, loss of speech, and seizures (NEDAMSS) and Kabuki syndrome-2

Case 3, a neonate, presented with poor responsiveness, tachypnea, and weak crying shortly after birth. Physical examination revealed generalized cyanosis, cephalic edema, mild retractions, and hypotonia of the extremities. Prenatal ultrasound at 28 weeks of gestation identified bilateral fetal pleural effusion, necessitating thoracoamniotic shunting to manage fetal hydrothorax. Prenatal ultrasound at 28 weeks identified fetal bilateral pleural effusion, necessitating fetal thoraco-amniotic shunt placement. Fetal echocardiography demonstrated pulmonary hypertension. Laboratory investigations revealed nocturnal hyperglycemia (peak value 17.7 mmol/L, considered to be associated with a stress response) and mild coagulopathy. Unfortunately, the parents elected to withdraw life-sustaining interventions shortly after admission, and the infant passed away on the day of discharge (**Table 1**).

Whole-exome sequencing identified a heterozygous pathogenic variant in the *IRF2BPL* gene [c.358C>T], associated with neurodevelopmental disorder with regression, abnormal movements, loss of speech, and seizures [7]. Concurrently, a *KDM6A* gene variant [c.2714C>T] was detected, consistent with molecular findings for Kabuki syndrome type 2 [8] (**Table 2**, Supplementary Table 1).

Discussion

The rapid advancement of whole-exome sequencing (WES) has led to the proliferation of genetic diagnostics in the clinical context, where clinicians frequently assign the clinical manifestations seen in children and parents to a single disorder [1]. However, the occurrence of dual and even triple genetic diagnoses and is not uncommon, as Posey et al. previously reported them in 4.9% (101/7,374) of patients undergoing WES had multiple genetic diagnoses [2]. Once monogenic disease is firmly established as a diagnosis with clinical findings difficult to explain, additional diagnoses cannot be excluded. This does not permit full diagnostic accuracy, since a vexing aspect of phenotypic expansion of the monogenic diagnosis may be that the abnormal manifestations are simply the clinical manifestations of a second diagnosis.

The occurrence of more than one disorder in a patient is referred to as comorbidity [9]. Co-occurrence of two unrelated genetic disorders allows for a classification of separate comorbidity patterns [10]. This report outlined three anecdotal cases of unrelated patients who exemplified multimorbidity of genetic conditions. Case 1 received a primary diagnosis of limb-girdle muscular dystrophy type 2E by genetic testing and appearance of the clinical phenotype. Hematochezia and diarrhea were attributed to concurrent viral and bacterial enteric infection. Mild hearing loss and chronic cough supported a co-diagnosis of primary ciliary dyskinesia type 37. Genetic testing resulted

in two novel variants of DNAH1 that were not previously reported, nor present in population databases. Following the ACMG guidelines for variant classification, the two variants were assigned an "Uncertain Significance" classification and will require long-term clinical follow-up to further establish pathogenicity.

Case 2 presented two diagnoses of pontocerebellar hypoplasia (PCH) type 6 and Joubert syndrome (JBTS) type 23 which may have accounted for the unexpectedly severe phenotype exhibited in the patient: refractory epilepsy, global developmental delay, and profound feeding difficulties requiring long-term nasogastric tube support. Case 3 was unable to be confirmed due to lack of observation by care providers. Prior to our evaluation, genetic testing for amniocentesis was unremarkable (non-WES testing at an external center). Due to the intervention discussed by the obstetric team, therapeutic paracentesis was pursued, and further perinatal interventions in the most aggressive format were considered reasonable to improve viability of the neonate after birth, though the clinical course eventually worsened and care was withdrawn by family. Parenthetically, while the costs for WES have sharply declined, many families are still unable to afford testing. The development of more cost-efficient precision diagnostics is an immediate problem, and incremental innovation in genetic counseling and prenatal testing methods will be required to address it.

For pediatric patients, families, and healthcare providers, establishing an accurate diagnosis at the earliest possible stage is critically imperative, as it enables comprehensive understanding of the disease course across multiple prognostic dimensions and facilitates personalized therapeutic interventions [11]. Furthermore, when the clinical manifestations of the primary diagnosis appear atypical or demonstrate greater severity than anticipated, clinicians must promptly consider the possibility of dual diagnoses. Patients with dual diagnoses exhibit substantially heightened clinical complexity compared to those with single-gene disorders, resulting in exacerbated symptom severity, intricate clinical management requirements, and elevated healthcare cost.

Conclusions

We described three children with dual diagnoses established by WES. We identified seven previously unreported pathogenic variants associated with rare diseases, thereby broadening the mutational spectrum of the relevant genes and providing case-based evidence for the diagnosis of the corresponding genetic disorders. Definitive diagnoses were important for optimizing clinical management and prognostic evaluation. The case studies provide empirical evidence to support that when there is diagnostic uncertainty or where genetic testing does not adequately explain the phenotype complexity, investigation for dual diagnoses should be performed to achieve complete diagnostic resolution. The key to optimizing the diagnosis of dual rare diseases lies in a closed-loop process of "High Clinical Suspicion → Rational Use of WES → Multidisciplinary Collaborative Interpretation and Management". This requires a shift in clinical thinking from pursuing a "single answer" to accepting "multiple comorbidities", and implementing a systematic, collaborative care model to achieve precise diagnosis and integrated management, ultimately improving patient prognosis.

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Disclosure of conflict of interest

None.

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References

[1] Boycott KM and Innes AM. When one diagnosis is not enough. N Engl J Med 2017; 376: 83-85.

[2] Posey JE, Harel T, Liu P, Rosenfeld JA, James RA, Coban Akdemir ZH, Walkiewicz M, Bi W, Xiao R, Ding Y, Xia F, Beaudet AL, Muzny DM, Gibbs RA, Boerwinkle E, Eng CM, Sutton VR, Shaw CA, Plon SE, Yang Y and Lupski JR. Resolution of disease phenotypes resulting from multilocus genomic variation. N Engl J Med 2017; 376: 21-31.

[3] Guan Y, Yang H, Yao X, Xu H, Liu H, Tang X, Hao C, Zhang X, Zhao S, Ge W and Ni X. Clinical and genetic spectrum of children with primary ciliary dyskinesia in China. Chest 2021; 159: 1768-1781.

[4] Straub V, Murphy A and Udd B. 229th ENMC international workshop: limb girdle muscular dystrophies - Nomenclature and reformed classification Naarden, the Netherlands, 17-19 March 2017. Neuromuscul Disord 2018; 28: 702-710.

[5] Rankin J, Brown R, Dobyns WB, Harington J, Patel J, Quinn M and Brown G. Pontocerebellar hypoplasia type 6: a british case with PEHO-like features. Am J Med Genet A 2010; 152A: 2079-2084.

[6] Bachmann-Gagescu R, Phelps IG, Dempsey JC, Sharma VA, Ishak GE, Boyle EA, Wilson M, Marques Lourenço C, Arslan M, Shendure J and Doherty D. KIAA0586 is mutated in joubert syndrome. Hum Mutat 2015; 36: 831-835.

[7] Tran Mau-Them F, Guibaud L, Duplomb L, Keren B, Lindstrom K, Marey I, Mochel F, van den Boogaard MJ, Oegema R, Nava C, Masurel A, Jouan T, Jansen FE, Au M, Chen AH, Cho M, Duffourd Y, Lozier E, Konovalov F, Sharkov A, Korostelev S, Urteaga B, Dickson P, Vera M, Martínez-Agosto JA, Begemann A, Zweier M, Schmitt-Mechelke T, Rauch A, Philippe C, van Gassen K, Nelson S, Graham JM Jr, Friedman J, Faivre L, Lin HJ, Thauvin-Robinet C and Vitobello A. De novo truncating variants in the intronless IRF2BPL are responsible for developmental epileptic encephalopathy. Genet Med 2019; 21: 1008-1014.

[8] Adam MP, Banka S, Bjornsson HT, Bodamer O, Chudley AE, Harris J, Kawame H, Lanpher BC, Lindsley AW, Merla G, Miyake N, Okamoto N, Stumpel CT and Niikawa N. Kabuki syndrome: international consensus diagnostic criteria. J Med Genet 2019; 56: 89-95.

[9] van den Akker M, Buntinx F, Metsemakers JF, Roos S and Knottnerus JA. Multimorbidity in general practice: prevalence, incidence, and determinants of co-occurring chronic and recurrent diseases. J Clin Epidemiol 1998; 51: 367-375.

[10] Capra AP, La Rosa MA, Briguori S, Civa R, Passarelli C, Agolini E, Novelli A and Briuglia S. Co-existence of genetic diseases is a new clinical challenge: three unrelated cases of dual diagnosis. Genes (Basel) 2023; 14: 484.

[11] Liu Y, Ma X, Chen Z, He R, Zhang Y, Dong H, Ma Y, Wu T, Wang Q, Ding Y, Li X, Li D, Song J, Li M, Jin Y, Qin J and Yang Y. Dual rare genetic diseases in five pediatric patients: insights from next-generation diagnostic methods. Orphanet J Rare Dis 2024; 19: 159.

Supplementary Table 1. Results of variability hazards predicted by the software

Patient	Gene	Nucleotide substitution	Amino acid substitution	Provean	SIFT	mutationtaster	M-CAP	CADD
Case 1	DNAH1	c.791T>C	p.Met264Thr	deleterious (-3.13)	damaging (0.003)	polymorphism (0.33)	Tolerated (0.008482)	Deleterious (23.2)
	DNAH1	c.6958G>A	p.Gly2320Ser	neutral (-1.89)	tolerated (0.231)	polymorphism (0.17)	Tolerated (0.006262)	Deleterious (22.8)
	SGCB	loss2(EXON:3-5)	NA	NA	NA	NA	NA	NA
Case 2	RARS2	c.1A>G	p.Met1?	neutral (-0.43)	damaging (0.0)	disease_causing_automatic (0.98)	damaging (0.387409)	Deleterious (20.4)
	RARS2	c.1024A>G	p.Met342Val	deleterious (-3.66)	damaging (0.0)	disease_causing_automatic (0.7)	damaging (0.030527)	Deleterious (24.4)
	KIAA0586	c.962-1G>T	-	NA	NA	disease_causing (0.68)	NA	Deleterious (33.0)
	KIAA0586	c.592A>T	p.Ser198Cys	deleterious (-2.57)	damaging (0.001)	polymorphism (0.2)	damaging (0.051847)	Deleterious (25.6)
Case 3	IRF2BPL	c.358C>T	p.Gln120Ter,677	NA	NA	disease_causing_automatic (0.96)	NA	Deleterious (33.0)
	KDM6A	c.2714C>T	p.Thr905Ile	deleterious (-3.73)	damaging (0.002)	polymorphism (0.11)	damaging (0.096805)	Deleterious (23.1)