Heliyon 11 (2025) e41591

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Case report

Unveiling the prenatal features of HADDS: A case report and literature review

Lina Hu, Dongzhi Li *, Li Zhen, Yanan Wang

Guangzhou Women and Children Medical Center, Affiliated to Guangzhou Medical University, China

ARTICLE INFO

Keywords:
EBF3 gene
HADDS
Neurogenic bladder
Hypotonia
Prenatal features

ABSTRACT

Hypotonia, Ataxia, and Delayed Development Syndrome (HADDS), triggered by EBF3 mutations, is a neurodevelopmental disorder syndrome characterized by hypotonia, ataxia, and developmental delay. The affected individuals often are unable to care for themselves, which has a significant impact on society and families. Hence, prenatal screening and diagnosis are particularly important. However, symptoms and signs of HADDS caused by mutations in EBF3 have not been studied until now. Herein, we report the case of a 1-year-old boy carrying a heterozygous point mutation in the EBF3 gene (c.271 del, p. Asp91Thrfs*41), who had typical signs and symptoms of mental retardation, hypotonia, developmental delay, neurogenic bladder, constipation, and Pectus excavatum, in addition to atypical facial features. HADDS was diagnosed by Whole Exome Sequencing on a family trio (Trio-WES) for recurrent urinary tract infection with dysuria at 6 months of age, with a normal karyotype and chromosomal microarray analysis (CMA). The variant is a *de novo* shifted code mutation, which expands the pathogenic gene spectrum of EBF3. Furthermore, we did a retrospective analysis of HADDS patients with a history of pregnancy and childbirth. We emphasized that reduced fetal movement, systematic ultrasound scanning, and fetal MRI might add evidence for prenatal diagnosis. The study is the first to explore prenatal screening for this EBF3 gene-related HADDS and is of great relevance.

1. Introduction

Early B cell factor 3 (EBF3), located on chromosome 10q26.3, is a highly conserved member of transcription factor (TF) superfamily, which involved in cellular development and differentiation across species. Mutations in EBF3 is linked to hypotonia, ataxia, and delayed development syndrome (HADDS). In 2017, three research groups confirmed this gene-disease association [1–3]. The disorder is characterized by hypotonia, ataxia, and intellectual disability (ID), and distinct facial features such as a long face, prominent forehead, high nasal bridge, deep philtrum, straight eyebrows, short wide chin, mild dysmorphic ears, and strabismus; Rare, congenital malformations may affect the genitourinary system [4,5], the gastrointestinal tract, or musculoskeletal system. Normal

Abbreviations: EBF3, Early B cell factor 3; TF, transcription factor; HADDS, hypotonia ataxia and delayed development syndrome; ID, intellectual disability; PE, Pectus excavatum; CVS, chorionic villus sampling; CMA, chromosomal microarray analysis; WES, whole-exome sequencing; NT, nuchal translucency; UAlb, urine microalbumin; Cr, creatinine; uNAG, urine N-acetyl-β-D-glucosaminidase; MRI, magnetic resonance imaging; Trio-WES, Whole Exome Sequencing on a family trio; DBD, DNA-binding domain; ZNF, zinc finger domains; DA, dopaminergic; GH, growth hormone; DSD, detrusor sphincter dyssynergia; EEG, electroencephalogram.

* Corresponding author.
E-mail address: lidongzhi2013@aliyun.com (D. Li).

https://doi.org/10.1016/j.heliyon.2024.e41591
Received 6 May 2024; Received in revised form 24 September 2024; Accepted 30 December 2024
Available online 10 January 2025

intelligence has also been reported, but with neurodevelopmental disorders [4]. In 2021, the first pathogenic mutation linked to HADDS was identified in a Chinese patient, who, besides the typical symptoms, showed new symptoms like hemangiomas, mild hearing abnormalities, and tracheomalacia, broadening the syndrome's known characteristics [6]. To date, approximately nearly 100 patients have been reported [7].

HADDS is a monogenic genetic disorder causing severe motor and cognitive deficits, with no cure. Prenatal signs like congenital hypotonia, ataxia or ID are often very subtle and easily missed. A 2023 study by T Weissbach et al. found that non-specific signs such as polyhydramnios, persistent breech presentation, intrauterine growth restriction and reduced fetal movement are significantly more common in patients with congenital hypotonia. Common prenatal signs of hypotonia are structural abnormalities, decreased fetal movement, joint contractures, and testicular descent at or beyond 30 weeks [8]. However, to date, no studies have specifically explored the prenatal specific signs of HADDS caused by EBF3 mutations. Here we present a case of HADDS owing to EBF3 mutation (c.271 del, p. Asp91Thrfs*41). Prenatally, the patient showed only a nuchal cystic hygroma but later presented with urinary symptoms, delayed motor, speech development and PE. This *de novo* mutation, confirmed by sanger sequencing and absent in the general population, is classified as pathogenic according to ACMG. Our study expanded the mutational spectrum of causative gene. Furthermore, we retrospectively analyzed the reported cases of HADDS caused by EBF3 mutations, with a focus on prenatal symptoms. We aim to identify specific and nonspecific prenatal indicators for early diagnosis. The research covers prenatal signs, recent advances, management, and genetic counseling for HADDS caused by EBF3 mutations.

2. Case presentation

The patient, a 1-year-old male with local parents, had a 28-year-old mother who reported no family or personal history of hereditary or congenital anomalies. Early pregnancy ultrasound showed a CRL of 56.6 mm, consistent with gestational week and revealed a single live fetus with a nuchal cystic hygroma (Fig. 1). The thickness of the nuchal translucency (NT) was 6.5mm. Chorionic villus sampling revealed a normal karyotype with chromosomal microarray analysis (CMA). Routine ultrasound scanning at 18 weeks showed normal fetal morphology and nuchal fold thickness (5.6mm). Fetal echocardiography was also normal. Subsequent ultrasound examinations at 24, 28 and 32 weeks respectively showed normal fetal structure, meridian and amniotic fluid volume. The proband was born at 39 weeks via vaginal delivery, weighting 3700g and appeared free of malformations. At 2 months, he began experiencing recurrent urinary tract infections, slow weight again and constipation. He was breastfed until 5 months and then switched to formula feeding due to diarrhea.

At 6 months old, the proband was referred to Guangzhou Women and Children's Medical Center for recurrent abnormal urinalysis and subsequent dysuria. Upon admission, the physical examination: He had normal development and nutrition. Both the anterior and posterior fontanels were closed. There were no abnormal in the eye, ear, nose, and mouth. Neck was soft, no resistance; sternal depression was seen (Fig. 2b). No abnormal cardiopulmonary findings were noted. Abdomen and external genitalia showed no obvious abnormality. Laboratory tests: Blood routine and liver function were normal; Urine biochemical indexes was approximately normal (cystatin C 1.09mg/L↑, creatinine 13μmol/L). But renal tubulointerstitial has damaged (urine microalbumin (UAlb)/creatinine (Cr) 9.7μg/μmol↑, urine N-acetyl-β-D-glucosaminidase (uNAG)/creatinine (Cr) 2.1U/mmol↑); Urinary ultrasound showed bilateral ureteral distension, mild bilateral hydronephrosis, and multiple stones in both kidneys (Fig. 3). Cystography showed regular bladder morphology, trabecular formation, and multiple diverticula; The left renal pelvis and ureter were dilated, and no reflux was observed on the right side; the urethra was patent during the voiding phase, and no reflux of contrast into the ureter was visible (Fig. 4), suggesting neurogenic bladder and left vesicoureteral reflux. Cardiac ultrasound was unremarkable except for the patent foramen ovale. MRI scan of lumbosacral spine showed no abnormality (Fig. 5). After the above examinations, neurogenic bladder and abnormal bladder function (bladder neck elevation) were considered, and cystoscopy was performed, after which the patient was discharged with a urinary catheter.

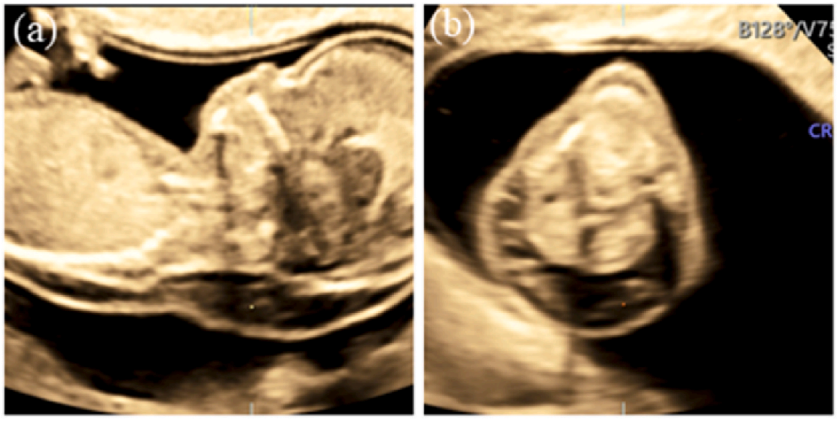

Fig. 1. Prenatal ultrasound showed Nuchal cystic hygroma. (a) Sagittal view of the fetus showed a cystic mass on the back of the neck with visible reticular division; (b) Transverse section of fetal head and neck showed the same result.

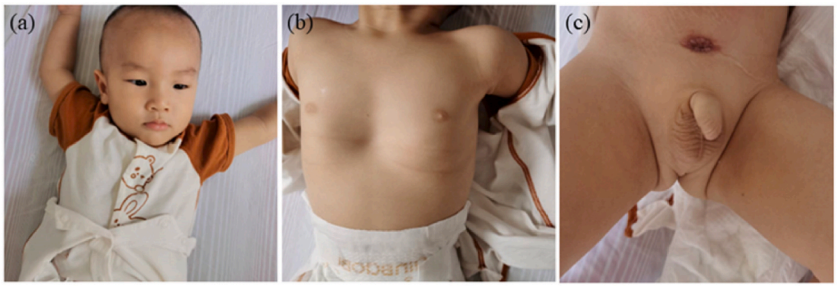

Fig. 2. Pictures of the proband. (a) Atypical facial features are visible. (b) Chest examination of the proband showed pectus excavatum. (c) After cystostomy, the incision healed well.

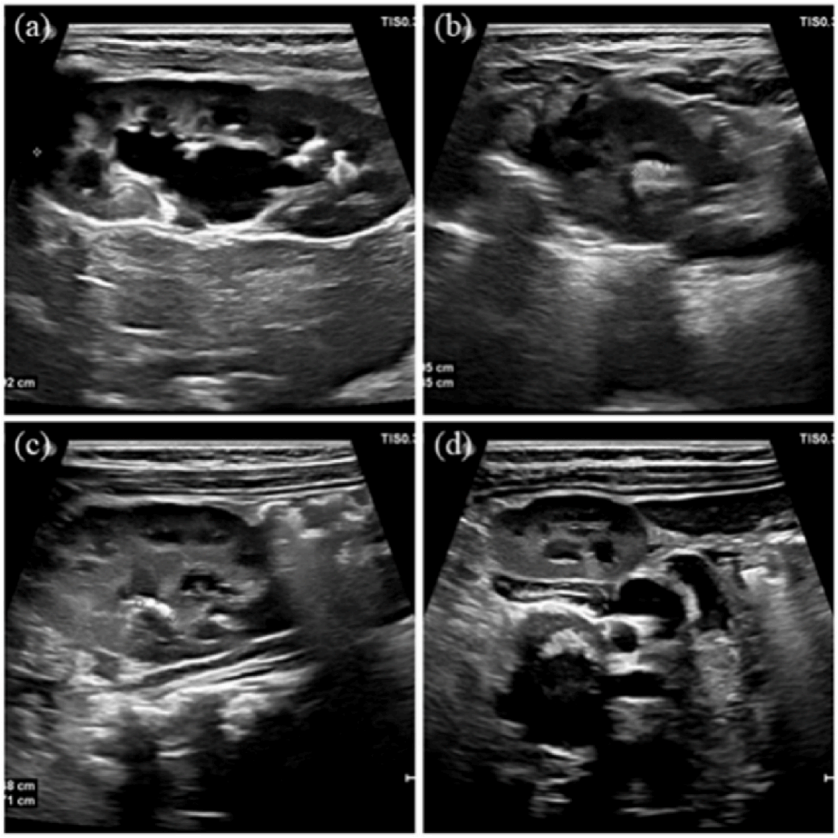

Fig. 3. Ultrasound images from bilateral kidneys and ureters of proband. (a) and (b) Ultrasound of the left kidney showed renal stones with hydronephrosis; (c) and (d) Ultrasound of the right kidney is similar to that of the left.

After discharge, the patient underwent whole-exome sequencing (Trio-WES) at an external institution, revealing a *de novo* c.271del (p. Asp91Thrfs*41) mutation in the EBF3 gene (Fig. 6). His parents had the wild-type allele. PS2 evidence was available. The mutation likely caused a frameshift and premature stop codon, indicating a loss of function. PVS1 evidence was available. The variant was not found in the population frequency databases, PM2_Supporting evidence was available. According to the ACMG guidelines, PVS1, one PS2 and one PM2_Supporting is sufficient evidence for a "pathogenic" classification for this variant. The mutation is linked to hypotonia, ataxia, and delayed development syndrome (HADDS), inherited autosomal dominant. The proband underwent cystostomy at 7 months at Guangzhou Women and Children's Medical Center and recovered well. However, he was still constipated. Barium enema revealed a somewhat narrowed lower rectum and a slightly dilated distal bowel. Anal manometry was grossly normal, and no rectal stenosis was detected. At 10 months, the patient referred to our department for developmental delay. He had atypical facial features (Fig. 2a), developmental delay and intellectual disability. In the physical movement examination, he had no head control and was unable to sit, roll, stand, or speak. As in addition, he had short attention span, decreased sensitivity to pain and stereotyped movements. All of these phenotypes were matched well with HADDS.

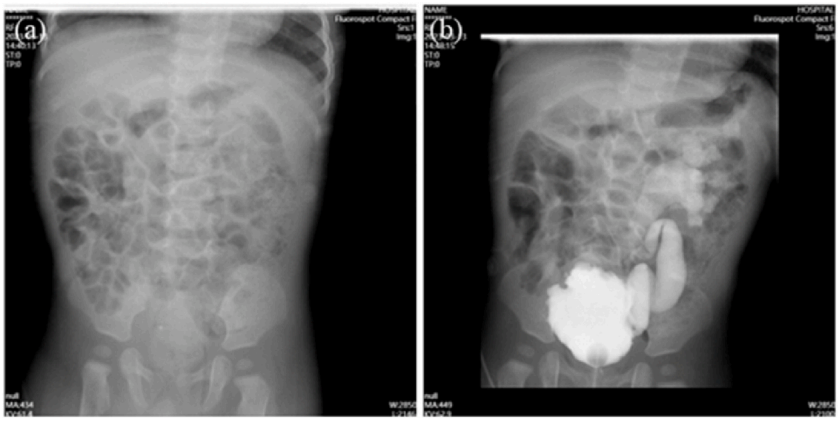

Fig. 4. Cystography examination the proband. Preoperative (a) and Postoperative (b) X-ray reveals Left ureter and renal pelvis dilated.

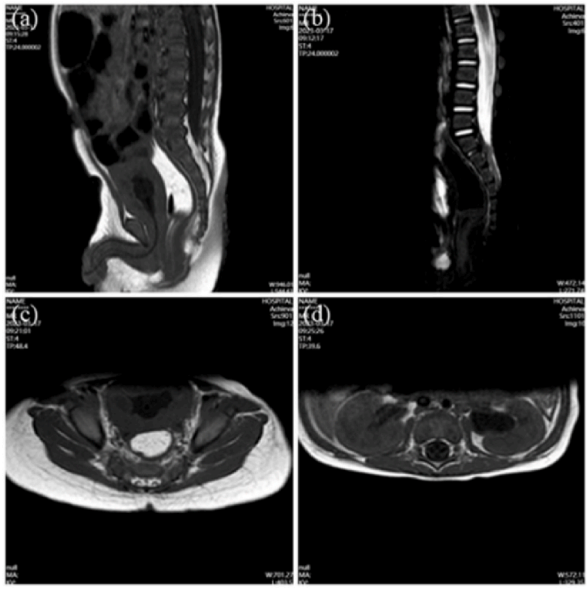

Fig. 5. Magnetic resonance imaging of the lumbosacral spine from the proband. (a) and (b) Sagittal Lumbosacral MRI showed there is no abnormality in the distribution of spinal cord, conus medullaris and cauda equina; (c) and (d) Lumbosacral magnetic resonance cross section showed bilateral pyelectasis and thickened bladder wall.

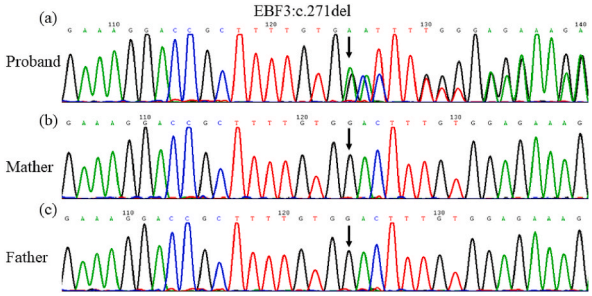

Fig. 6. Sanger sequencing results of the proband and his parents. (a) The proband was identified to be *de novo* heterozygous; (b) and (c) Figures showed his parents were wild type. Black arrows indicate the mutation sites.

3. Literature review

A systematic search of publications on EBF3 was performed in PubMed. The retrieval time was up to April 2024. The following keywords were used: EBF3. A total of 121 full articles were initially retrieved. After careful review, only 5 studies involving 7 patients with antenatal signs were deemed suitable [2,5,6,9,10]. Including our patient, the total was 8. Information on age, sex, mutation site and mutation type, prenatal ultrasound abnormalities indicators, mode of delivery and MRI was extracted, collated and analyzed, as shown in Table 1. The minimum age was 10 months, and the maximum age was 11 years. The male to female ratio was 3:5. 6 cases were point mutation and 2 cases were deletion. Of these point mutations, 5 cases were missense mutation, 1 case induced frameshift mutation. We discovered that the prenatal features appeared to be relatively specific. There were three patients with growth retardation, two patients with decreased fetal movement, and one patient each of oligohydramnios, breech presentation, distended bladder, and undescended testes; Case 5 was born with a markedly distended abdomen and abnormal urination. Urethral stenosis with atonic bladder was later discovered. In addition to respiratory distress and hypotonia, case 6 presented at birth with left abdominal cryptorchidism and hypermobility of the right testis as well as mild hydrocele. And yet the dystonia and testicular problems, which were not reported during pregnancy, were discovered after birth. Some screens are not indicated routinely during pregnancy, such as testicular position, which could be highly susceptible to a missed diagnosis. Some patients may present with more than one non-specific manifestation, and only the case we report showed nuchal cystic hygroma in the fetal period. Except for our patient who did not undergo MRI, five of the seven patients had MRI abnormalities. The mode of delivery was determined by usual obstetric or fetal indications, and no specific pattern was found.

4. Discussion

HADDS can be caused by EBF3 mutations, terminal 10q deletions [9], small deletions (<1 mb) [10], and ring chromosome 10 [r(10)] [11]. Haploinsufficiency is crucial for the observed phenotype in patients with these genetic alterations. Our results and the cases reviewed in the literature were all with intellectual disabilities, further supporting this view. Pathogenic variants affecting the DNA-binding domain (DBD), or paralogous residues [5], could cause changes in subcellular protein localization or distribution patterns, which may ultimately result in significantly reduced EBF3 transcriptional activity [7]. Case 6 is caused by a missense mutation in the important region, which can interact with DNA. By constructing the plasmid and transfecting HEK293T cells, EBF3-N197D mutant showed impaired activation of luciferase reporter expression of the p21 promoter, and the mutant affected its entry into the nucleus [6]. The c.488 site missense mutations affected a single conserved residue in a zinc finger motif crucial for DNA binding and are deleterious in a fly model [2]. Activation of reporter gene expression in HEK293 cells showed EBF3 p. Arg163Leu variant partial loss of transcriptional activation. And EBF3 p. Arg163Gln variant complete loss of transcriptional activity [2]. It is reported that motor milestones and language development are delayed in all mutated patients. Head control was achieved after 5 months. The patients have trunk control around 1 year of age and walked independently around 4 years of age. But the cases we described all presented with global developmental delay. Only case 7 could walk independently at 30 months of age. Expressive language was affected to a greater degree than receptive language. All children followed simple verbal requests well, but the development of the first word was delayed. In the 2 patients with deletions, there was no significant delay in developmental, but they had neurodevelopmental disorders. All the reported cases had language impairments regardless of type of mutation. Moreover, most patients had abnormal MRI, which may show cerebellar vermis hypoplasia and/or specific foliation anomalies. While cerebellar lobes with a radial distribution (dandelion sign) were thought to possibly represent a distinct and recognizable sigh [1,2,7,12]. In 2020, a study proposed that EBF3 gene is critical for neuronal migration during corticogenesis and its mutation results in a specific cerebellar lobule appearance [13]. Cerebellar signs have

Table 1
Clinical characteristics of this case and previously reported patients including age, sex, variant site and mutation type, prenatal ultrasound signs, mode of delivery, MRI, and references. MOD = mode of delivery; VD = vaginal delivery; CS = caesarean section; M = month; Y = year; ND = not done; DCDA: dichorionic diamniotic; FGR: fetal growth restriction; MRI: magnetic resonance imaging.

Age (case number)	Sex	Variant Sites	Mutation Type (Source of variation)	Prenatal Ultrasound Signs	Mode Of Delivery (gestation/birth weight)	MRI	References
1Y	male	c.271del(p. Asp91Thrfs*41)	Frameshift (*de novo*)	nuchal cystic hygroma	VD (39w/3.7kg)	ND	This case
7Y (case 1)	male	c.488G>A (p. Arg163Gln)	Missense (*de novo*)	decreased fetal movements	CS (38w/3.4kg)	abnormal	[2]
5Y (case 2)	female	c.488G>A (p. Arg163Gln)	Missense (*de novo*)	decreased fetal movements, oligohydramnios	VD (40w/3.35kg)	abnormal	[2]
3Y (case 3)	female	c.488G>T (p. Arg163Leu)	Missense (*de novo*)	breech position	CS (39w/2.7kg)	normal	[2]
1Y+11M (case 4)	female	c.487C>T; (p. Arg163Trp)	Missense (*de novo*)	distended bladder	VD (40w/3.49kg)	abnormal	[5]
1Y+6M (case 5)	male	c.589A>G (p. Asn197Asp)	Missense (*de novo*)	global developmental delay, Undescended testicles	CS (39w/3.2kg)	abnormal	[6]
11Y (case 6)	female	10q26.3	Deletion (*de novo*)	DCDA, developmental delay	VD (35w/1.83kg)	normal	[9]
3Y+10M (case 7)	female	10q26.13-q26.3	Deletion (*de novo*)	FGR	CS (35w/1.61kg)	abnormal	[10]

been underestimated in many articles about HADDS associated with EBF3 gene [12]. The MRI of case 4 was normal, which may be related to the extent of the effect on transcription activity.

It was shown that disruption of EBF3-mediated transcriptional regulation leads to ID and developmental delay [14]. The severity of patient symptoms is correlated with missense variants of EBF3 that interfere with ZNF, which is a key protein structural domain to stabilize the interaction between EBF3 and target DNA sequences [15]. Among 8 variants, 4 of 5 missense variants cause disease through interference with ZFN (Table 1). The development of the neocortex is vital for neuronal migration. EBF3 is a key regulator of neuronal multipolar to bipolar migration. EBF3-knockdown cells show severe deficits in the formation of leading processes and an inhibited shift to the locomotion mode [16]. In this study, it was again demonstrated the important role of EBF3 in neural development. As is well known, Parkinson's disease is linked with degenerative loss of midbrain dopaminergic (DA) neurons. The lowered EBF3 expression could decreased TH + DA neuron number. Although EBF3 is only transiently expressed during an early postmitotic stage, perhaps contributing to ataxia in patients with HADDS [17,18]. The patient with a duplication/triplication mosaicism of a region encompassing EBF3 has a rather mild phenotype compared to the core signs, which may reflect the importance of EBF3 gene-dosage for neurodevelopment [19]. There are reports suggesting that patients with10q26 deletion seem to lack cognitive impairment, whereas patients with an EBF3 point mutation always have cognitive impairment [12]. Mental retardation was present in our cases, both deletion and point mutation. Although ataxia tended to wan with age, cognitive impairment may become more severe. Therefore, systematic neuropsychological follow-up is recommended for patients with EBF3-HADDS [20]. For profound ID or global developmental delay, it is suggested that professional evaluation by a specialized neurologist and pediatrician is essential, which should be taken into full consideration in the management of the program, as the patient will benefit greatly from a rehabilitation program, including the exercise of gross and fine motor skills [12]. In cases of markedly reduced growth rate or growth hormone (GH) deficiency, GH therapy can be administered, generally starting at age 3 years. According to the literature, the disease is usually diagnosed after 6 months of age. Yet, the patients were in good general health. Life expectancy is not affected by the syndrome [12].

The urologic manifestations of HADDS include bladder dysfunction secondary to detrusor sphincter dyssynergia (DSD), vesicoureteral reflux, urinary tract infections, and cryptorchidism. Neurogenic bladder is diagnosed in infancy (median 6.5 months) and almost always requires cystostomy [19]. Case 5 had a distended abdomen at birth, revealing urethral stenosis and an atonic bladder. Unexplained neurogenic bladder presenting in infancy without spinal cord abnormalities is crucial for diagnosing HADDS (Fig. 5). If not properly diagnosed and managed, these patients are at risk for renal deterioration [21]. The proband had mild renal impairment, which may be associated with previous urological infection, hydronephrosis and reflux renal impairment. After cystostomy, urinary and renal function should be rechecked periodically; Analysis of composition was available for multiple stones (Fig. 3), while laboratory tests for blood PTH, calcium, magnesium, and phosphorus may be helpful in determining stone origin. EBF3 is a transcription factor marking tenocytes and connective tissue cells in skeletal muscle of embryos. A knockout of EBF3 in mice had no effect on chondrogenesis, but it led to sternum ossification defects due to diminished $Runx2^{+}$ preosteoblast generation [22]. In addition to our patient, we also found two patients with missense mutations had PE [5,6]. Reinforcement of nutrition was advised, and sucker therapy if necessary. EBF3 is a regulator of muscle cell-specific transcription. Under the absence of Ebf3, the expression of the Ca (2+) pump Serca1 (Atp2a1) is downregulated, which leads to impaired muscle function [23]. Probiotics and digestive drugs are expected to confer benefits on patients with constipation. The child should be regularly followed up to monitor growth.

In fact, the patient in our case underwent prenatal diagnosis without detection of the causative mutation. Because he performed only CMA without WES analysis, and EBF3-associated HADDS is a monogenic disease. It is an issue of the indications and limitations of the current genetic testing techniques. If CMA is negative, Trio-WES is routinely recommended for nuchal cystic hygroma. Unfortunately, his parents refused, maybe out of the high price or maybe just out of the fluke. There are only sporadic reports on prenatal-specific symptoms of the disease, but no specific pattern. However, Case-based review of the literature showed that the common prenatal features of HADDS caused by EBF3 are growth retardation, decreased fetal movement, oligohydramnios, breech presentation, bladder distention and undescended testes. But clearly, the symptoms are consistent with the previously reported literature. Nuchal cystic hygroma may be by chance but are more likely related to the HADDS. Nevertheless, it is important that they should be accumulated in the literature for inclusion in future analysis. MRI abnormalities need to be special attention. If an MRI abnormality is found in combination with a nonspecific feature, even one, then WES can be performed directly. And as a monogenic disease, an omission would most likely not occur. However, Prenatal diagnosis of MRI is difficult and requires experienced radiologists. Here, our study presents possible prenatal signs of HADDS to facilitate early recognition and diagnosis. Nevertheless, these results are based on a small sample size, and more studies is needed to provide more evidence. The limitation of our case is that MRI and electroencephalogram (EEG) were not performed in proband. But both tests did not affect the diagnosis and his parents did not mention a history of seizures. It has been shown that patients had abnormal EEG without epilepsy [12]. In some cases, ophthalmologic disorders such as macular hypoplasia may be present in combination. So, it is recommended that the patient complete the fundus examination. The mutation locus is *de novo*. Although the probability of bearing a patient again is very low. The mother of the patient is still advised to perform prenatal diagnosis for her next (second) pregnancy.

5. Conclusion

In this study, we described one male patient carrying a novel heterozygous EBF3 (c.271 del, p. Asp91Thrfs*41) variant. In addition to atypical facial features, the clinical features of the individual in our case overlaps with previously reported patients. Importantly, for the first time, we described nuchal cystic hygroma in patients with HADDS caused by EBF3 mutation. Clinical data from 8 individuals were retrospectively analyzed. We identified a number of prenatal features that may be specific, including growth retardation, decreased fetal movement, hypospadias, breech position, bladder dilatation, and undescended testes; The majority of them had

abnormal MRI. Therefore, we believe that a systematic scan should be performed by a sonographer with a background in prenatal diagnosis or fetal medicine, when atypical changes are present. Fetal MRI might also increase more phenotypes. It would provide more evidence for further prenatal diagnosis and WES detection. These findings may be useful in early identification of the disorder and prompted genetic testing. However, we have reported only a few cases, and additional studies are needed to fully elucidate genotype-phenotype interactions in HADDS caused by EBF3 gene.

CRediT authorship contribution statement

Lina Hu: Writing – review & editing, Writing – original draft, Visualization, Conceptualization. **Dongzhi Li:** Visualization, Supervision, Project administration, Conceptualization. **Li Zhen:** Supervision. **Yanan Wang:** Supervision.

Data availability statement

Data will be made available on request. Data associated with this study has not been deposited into a publicly available repository, which will be available upon reasonable request after the approval of the corresponding author (Dongzhi Li, Email: lidongzhi2013@aliyun.com).

Funding

None.

Declaration of competing interest

The corresponding author declare no potential competing or non-financial interests on behalf of all authors of the manuscript.

Acknowledgments

We thank the patient and his family for participating in this study.

References

[1] H. Sleven, S.J. Welsh, J. Yu, Mair, C.F. Wright, A. Henderson, R. Horvath, J. Rankin, J. Vogt, A. Magee, V. McConnell, A.J. Green, M.D. King, H. Cox, L. Armstrong, A. Lehman, T.N. Nelson, J. Williams, P. Clouston, J. Hagman, De novo mutations in EBF3 cause a neurodevelopmental syndrome, Am. J. Hum. Genet. 100 (2017) 138–150, https://doi.org/10.1016/j.ajhg.2016.11.020.

[2] H.T. Chao, M. Davids, E. Burke, J.G. Pappas, J.A. Rosenfeld, A.J. McCarty, T. Davis, L. Wolfe, C. Toro, C. Tifft, F. Xia, N. Stong, T.K. Johnson, C.G. Warr, S. Yamamoto, D.R. Adams, T.C. Markello, W.A. Gahl, H.J. Bellen, M.F. Wangler, A syndromic neurodevelopmental disorder caused by de novo variants in EBF3, Am. J. Hum. Genet. 100 (2017) 128–137, https://doi.org/10.1016/j.ajhg.2016.11.018.

[3] A. Tanaka, M.T. Cho, R. Willaert, K. Retterer, Y.A. Zarate, K. Bosanko, V. Stefans, K. Oishi, A. Williamson, G.N. Wilson, A. Basinger, T. Barbaro-Dieber, L. Ortega, S. Sorrentino, M. Gabriel, I.J. Anderson, M.J. Guillen, R.E. Schnur, W.K. Chung, De Novo Variants in EBF3 Are Associated with Hypotonia, Developmental Delay, Intellectual Disability, and Autism, vol. 3, 2017 a002097, https://doi.org/10.1101/mcs.a002097 a002097.

[4] Mar, A. Jiménez, P. Tirado, B. Calleja-Pérez, L.A. Alcaraz, S. Álvarez, J. Williams, J.R. Hagman, A.H. Németh, A. Fernández-Jaén, Neuroimaging findings in patients with EBF3 mutations: report of two cases, Mol. Syndromol. 12 (2021) 186–193, https://doi.org/10.1159/000513583.

[5] P.R. Blackburn, S.S. Barnett, M.T. Zimmermann, M.A. Cousin, C. Kaiwar, F. Pinto e Vairo, Z. Niu, M.J. Ferber, R.A. Urrutia, D. Selcen, E.W. Klee, P.N. Pichurin, Novel de novo variant in EBF3 is likely to impact DNA binding in a patient with a neurodevelopmental disorder and expanded phenotypes: patient report, in silico functional assessment, and review of published cases, Mol. Case Stud. 3 (2017) a001743, https://doi.org/10.1101/mcs.a001743.

[6] Y. Huang, L. Mei, Y. Wang, H. Ye, X. Ma, J. Zhang, M. Cai, P. Li, Y. Ge, Y. Zhou, A Novel de novo Mutation in EBF3 Associated with Hypotonia, Ataxia, and Delayed Development Syndrome in a Chinese Boy, Front. Genet. 12 (2021), https://doi.org/10.3389/fgene.2021.676832.

[7] J. Zhu, W. Li, S. Yu, W. Lu, Q. Xu, S. Wang, Y. Qian, Q. Guo, S. Xu, Y. Wang, P. Zhang, X. Zhao, Q. Ni, R. Liu, X. Li, B. Wu, S. Zhou, H. Wang, Further delineation of EBF3-related syndromic neurodevelopmental disorder in twelve Chinese patients, Front. Pediatr. 11 (2023), https://doi.org/10.3389/fped.2023.1091532.

[8] T. Weissbach, Z. Moran Hausman-Kedem, Yanay, R. Meyer, O. Bar-Yosef, L. Leibovitch, M. Berkenstadt, O. Chorin, H.A. Shani, R. Massarwa, Achiron, B. Weisz, R.S. Sharon, Mazaki Tovi, Eran Kassif, Congenital hypotonia: systematic approach for prenatal detection, Ultrasound Obstet. Gynecol. 62 (2023) 94–105, https://doi.org/10.1002/uog.26178.

[9] F. Lopes, G. Soares, M. Gonçalves-Rocha, J. Pinto-Basto, P. Maciel, Whole gene deletion of EBF3 supporting haploinsufficiency of this gene as a mechanism of neurodevelopmental disease, Front. Genet. 8 (2017), https://doi.org/10.3389/fgene.2017.00143.

[10] J. Pruccoli, C. Graziano, C. Locatelli, L. Maltoni, H.A. Sheikh Maye, D.M. Cordelli, Expanding the neurological phenotype of ring chromosome 10 syndrome: a case report and review of the literature, Genes 12 (2021) 1513, https://doi.org/10.3390/genes12101513.

[11] Á.C. Faria, E. Rabbi-Bortolini, M.R.G.O. Rebouças, A.L.A. de S Thiago Pereira, M.G.T. Frasson, R. Atique, N.C.V. Lourenço, C. Rosenberg, G.S. Kobayashi, M. R. Passos-Bueno, F.I.V. Errera, Craniosynostosis in 10q26 deletion patients: a consequence of brain underdevelopment or altered suture biology? Am. J. Med. Genet. Part A. 170A (2016) 403–409, https://doi.org/10.1002/ajmg.a.37448.

[12] C. Ciaccio, C. Pantaleoni, M. Moscatelli, L. Chiapparini, V. Nigro, E.M. Valente, F. Sciacca, L. Canafoglia, S. Bulgheroni, S. D'Arrigo, Neurologic, neuropsychologic, and neuroradiologic features of EBF3-related syndrome, Neurol. Genet. 9 (2023) e200049, https://doi.org/10.1212/NXG.0000000000200049.

[13] S. D'Arrigo, M. Moscatelli, C. Ciaccio, C. Pantaleoni, R. Castello, L. Chiapparini, TUDP Study Group: abnormal cerebellar foliation in EBF3 mutation, Neurology 94 (2020) 933–935, https://doi.org/10.1212/WNL.0000000000009486.

[14] F.L. Harms, K.M. Girisha, A.A. Hardigan, F. Kortüm, A. Shukla, M. Alawi, A. Dalal, L. Brady, M.A. Tarnopolsky, L.M. Bird, S. Ceulemans, E. Martina Bebin, K. M. Bowling, S.M. Hiatt, E.J. Lose, M. Primiano, W.K. Chung, J. Juusola, Zeynep Coban Akdemir, M.N. Bainbridge, Mutations in EBF3 disturb transcriptional profiles and cause intellectual disability, ataxia, and facial dysmorphism, Am. J. Hum. Genet. 100 (2017) 117–127, https://doi.org/10.1016/j.ajhg.2016.11.012.

[15] C.A. Deisseroth, V.C. Lerma, C.L. Magyar, Jessica Mae Pfliger, A. Nayak, N.D. Bliss, A.W. LeMaire, V. Narayanan, C. Balak, G. Zanni, Enza Maria Valente, E. Bertini, P.J. Benke, M.F. Wangler, H.T. Chao, An integrated phenotypic and genotypic approach reveals a high-risk subtype association for EBF3 missense variants affecting the zinc finger domain, Ann. Neurol. 92 (2022) 138–153, https://doi.org/10.1002/ana.26359.
[16] R. Iwai, H. Tabata, M. Inoue, K.I. Nomura, T. Okamoto, M. Ichihashi, K.I. Nagata, K.I. Mizutani, A Prdm8 target gene Ebf3 regulates multipolar-to-bipolar transition in migrating neocortical cells, Biochem. Biophys. Res. Commun. 495 (2018) 388–394, https://doi.org/10.1016/j.bbrc.2017.11.021.
[17] S. Baek, H. Choi, J. Kim, Ebf3-miR218 regulation is involved in the development of dopaminergic neurons, Brain Res. 1587 (2014) 23–32, https://doi.org/10.1016/j.brainres.2014.08.059.
[18] S. Thuret, L. Bhatt, D.D.M. O'Leary, H.H. Simon, Identification and developmental analysis of genes expressed by dopaminergic neurons of the substantia nigra pars compacta, Mol. Cell. Neurosci. 25 (2004) 394–405, https://doi.org/10.1016/j.mcn.2003.11.004.
[19] E. Ignatius, R. Puosi, M. Palomäki, N. Forsbom, M. Pohjanpelto, T. Alitalo, A.K. Anttonen, K. Avela, L. Haataja, C.J. Carroll, T. Lönnqvist, P. Isohanni, Duplication/triplication mosaicism of EBF3 and expansion of the EBF3 neurodevelopmental disorder phenotype, Eur. J. Paediatr. Neurol. 37 (2022) 1–7, https://doi.org/10.1016/j.ejpn.2021.12.012.
[20] E. Nishi, T. Uehara, K. Yanagi, Y. Hasegawa, K. Ueda, T. Kaname, T. Yamamoto, K. Kosaki, N. Okamoto, Clinical spectrum of individuals with de novo EBF3 variants or deletions, Am. J. Med. Genet. 185 (2021) 2913–2921, https://doi.org/10.1002/ajmg.a.62369.
[21] S.F. Batie, C.Q. Ashbrook, C.T. Coco, L.A. Baker, I. Stanasel, Urologic manifestations of hypotonia, ataxia, and delayed development syndrome (HADDS), a rare neurodevelopmental disorder, J. Pediatr. Urol. 19 (2023) 751.e1–751.e9, https://doi.org/10.1016/j.jpurol.2023.08.023.
[22] M. Kuriki, F. Sato, H.N. Arai, M. Sogabe, M. Kaneko, H. Kiyonari, K. Kawakami, Y. Yoshimoto, C. Shukunami, A. Sehara-Fujisawa, Transient and lineage-restricted requirement of Ebf3 for sternum ossification, Development (Cambridge, U. K.) 147 (2020) dev186239, https://doi.org/10.1242/dev.186239.
[23] S. Jin, J. Kim, T. Willert, T. Klein-Rodewald, M. Garcia-Dominguez, M. Mosqueira, R. Fink, I. Esposito, L.C. Hofbauer, P. Charnay, M. Kieslinger, Ebf factors and MyoD cooperate to regulate muscle relaxation via Atp2a1, Nat. Commun. 5 (2014) 3793, https://doi.org/10.1038/ncomms4793.