A New Mutation Identified by Whole Exome Sequencing in a Cornelia de Lange Syndrome Newborn

Hua Zhang, Li-Ming Yang, Lu Yuan, Xin Tan, Fu-Qing Zhang

Department of Eugenics and Genetics, Women and Infants Hospital of Zhengzhou, Zhengzhou, Henan 450000, China

To the Editor: Cornelia de Lange syndrome (CdLS, OMIM #122470), also called Brachmann-de Lange syndrome, is a serious genetic disease with unknown exact incidence. The estimated prevalence varies with a reported 1:10,000 individuals in the United States and 1:50,000 in Denmark. It may be underestimated because of underdiagnosis of mild phenotyope. CdLS patients have a distinguishing facial appearance, namely synophrys joining at the midline and with an arched appearance of the eyebrows, thick, and long eyelashes, extending down to the bridge of the nose, long prominent philtrum with down-turned lip corners short and flattened nose, hirsute forehead, and cutis marmorata. Besides, prenatal/postpartum growth retardation, cognitive impairment, psychomotor delay, and behavioral problems such as repetitive and self-injurious behaviors are also remarkable in CdLS patients. Moreover, malformations of other organs have also been reported including the genitourinary system, the cardiovascular system, and the skeletal system.

CdLS is a rare multisystem genetic disease mainly caused by the mutations of genes code for cohesion system including SMC1A, SMC3, RAD21, NIPBL, and HDAC8.[1] Cohesion plays a key role in the maintenance of genomic stability, as it is involved in the regulation of gene expression. DNA repair, sister chromatid cohesion, chromatin remodeling, and long-range gene interactions. Mutations in NIPBL (cohesion regulator) are presented in approximately 60% of classical CdLS patients.

Only a few Chinese cases have been reported, regardless of the many CdLS cases reported worldwide. Herein, we presented a 2-day-old newborn who was suspected to be a CdLS patient. The pathogenic mutation was searched using whole exome sequencing (WES) and copy number variation sequencing (CNV-seq).

The girl was the first child of the first pregnancy of healthy, nonconsanguineous parents without any familiar diseases or genetic defects. The child’s birth length was 39 cm; birth weight was 1800 g; and head circumference was 30.5 cm. Physical examination revealed that she was smaller than gestational age with birth length 39 cm; birth weight 1800 g; and head circumference was 30.5 cm. Peripheral blood of the patient was collected for karyotype analysis and gene tests including CNV-seq and WES. The platforms of CNV-seq and WES were Illumina NextSeq CN 500 and Illumina Novaseq, respectively. In addition, peripheral blood was also collected from the parents for verification. Karyotype analysis revealed a normal female karyotype. CNV-seq test did not indicate any changes in gene copy number exceed 100 kb. Results of WES identified a missense heterozygous mutation in the exome 29 of NIPBL (RefSeq NM-133433), namely c.5567G>T. The mutation was confirmed by Sanger sequencing; however, it was not found from the patient’s parents, indicating that it was a de novo mutation. Such mutation resulted in the replacement of Arg by Ile at codon 1856. The c.5567G>T mutation in NIPBL has never been reported in the gnomAD database, ExAC03 database, ShenZhou Genomic database or 1000G database previously. The NIPBL gene is the human homolog of the Drosophila Nipped-B gene which locates on chromosome 5p13. It encodes protein delangin, which is involved in chromatin cohesion processes and enhancer–promoter communications. NIPBL is required for cohesion loading on chromatin, a function that is conserved across evolution. In our case, the missense mutation occurs in the Armadillo-like helical functional domain, which is a highly conservative site similar to a previously reported mutation c.6272G>T.[2] Therefore, it is considered as the pathogenic gene mutation of the patient. In addition, this variant is predicted be “damaging” by PolyPhen and SIFT software, which further demonstrate the pathogenicity of the mutation. Our genetic diagnosis can not only help the patient to identify the disease but also has certain guiding significance for the parents in reproduction. Gillis et al.[3] reported that the recurrence risk of a subsequent affected child from healthy parents with a previously affected offspring was estimated to be approximately 60% of classical CdLS patients. Moreover, both her fingers and toes were short, with one knuckle lacking in each finger. Furthermore, she was diagnosed with patent foramen ovale, hearing impairment, and feeding difficulty.

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1%. Thus, prenatal diagnosis is strongly recommended during the next pregnancy.

There is no clear indication that CdLS is ethnically diverse, but cases of CdLS in China are rarely reported, suggesting that many cases might have been missed. On the one hand, our pediatrician or genetic experts do not know this disease very well. On the other hand, patients are usually limited to detect the major five disease-causing genes due to technical constraints. However, other genes such as KMT2, SETD5, or chromosome deletions such as 9q31.1-q32 deletion and 8p23.1 deletion can also lead to CdLS phenotype. In this study, the WES, which relies on high throughput and comprehensive detection, has a great advantage in the diagnosis of single-gene disease. Besides, CNV-seq can detect chromosome deletions. Thus, the combination of these two methods might contribute to discovering all possible pathogenic gene mutations or deletions at one time, which might improve the detection rate and save time.

Gillis et al. showed significant differences between patients with and without NIPBL mutations by genotype-phenotype correlation study. They found that patients with NIPBL mutations had more severe phenotype for growth retardation and development delay compared with other gene mutations. The exact function of NIPLB is unknown; however, it is strongly expressed in heart, skeletal muscle, and thymus and is essential for the proper development in the growing embryo. Mannini et al. reviewed the mutation spectrum that occurred in the five genes underlying CdLS and found that the association between phenotypes caused by the mutations in five different genes were NIPBL, HDAC8, RAD21, SMC1A, and SMC3, from severe to mild. Among various mutation types of NIPBL, nonsense, splice site, and frameshift mutations that lead to a truncated and presumably nonfunctional NIPBL protein are associated with a more severe phenotype. Missense mutations, in general, are associated with a milder phenotype characterized by absent limb abnormalities and less severe developmental and growth involvement. However, this is not absolute. Missense mutations, involving the HEAT domain, in particular H2-H4 repeat, have been identified in probands with severe phenotype. In our case, though the patient was confirmed to be a carrier of missense mutation in NIPBL, but she had obvious intrauterine growth delay: low weight (1800 g), low height (39 cm), as well as low head circumference (30.5 cm) at birth. What’s more, her lower extremities and upper extremities were all short, and all fingers have only two knuckles, which could be ascribed the influence of environment and other genetic factors in the expressivity of CdLS. Thus, the relationship of genotype-phenotype is complicated, which needs further study.

In conclusion, a novel missense mutation has been identified in NIPBL by WES. To the best of our knowledge, this mutation site has not been reported previously, which will broaden the mutation spectrum and might provide new insights into the diagnosis of CdLS.

**Declaration of patient consent**

The authors certify that they have obtained patient consent form. In the form, the parents have given their consent for the patient’s images and other clinical information to be reported in the journal. They understand that the patient’s name and initials will not be published and due efforts will be made to conceal her identity, but anonymity cannot be guaranteed.

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**Conflicts of interest**

There are no conflicts of interest.

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