DATA REPORT

Syndromic disorder of sex development due to a novel hemizygous mutation in the carboxyl-terminal domain of ATRX

Masaki Takagi1,2,5, Hiroko Yagi3,5, Ryuji Fukuzawa2, Satoshi Narumi1,4 and Tomonobu Hasegawa1

Alpha-thalassemia/mental retardation syndrome X-linked (ATRX; OMIM #301040), which is caused by mutations in the ATRX gene, is characterized by alpha-thalassemia, distinct dysmorphic facies, psychomotor development delay and genital abnormalities. Here, we describe a neonatal case of syndromic disorder of sex development, harboring a novel hemizygous mutation, p.Asp2352fs*1 in the carboxyl-terminal domain of ATRX. Our study provides additional evidence that deletion of the carboxyl terminus of ATRX is associated with severe genital anomalies.

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1Department of Pediatrics, Keio University School of Medicine, Tokyo, Japan; 2Department of Pathology and Laboratory Medicine, Tokyo Metropolitan Children’s Medical Center, Tokyo, Japan; 3Department of Endocrinology and Metabolism, Tokyo Metropolitan Children’s Medical Center, Tokyo, Japan and 4Department of Molecular Endocrinology, National Research Institute for Child Health and Development, Tokyo, Japan.

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1These authors contributed equally to this work.
The ATRX protein belongs to the family of SWI/SNF DNA helicases, which are involved in chromatin remodeling. The p.Asp2352fs*1 mutation results in the lack of only the carboxyl-terminal domain, whereas other functional domains, such as the amino-terminal ATRX–DNMT3–DNMT3L (ADD) domain, which is a plant homeodomain-like zinc finger with an additional C2–C2 motif, and the helicase/adenosine triphosphatase (ATPase) domain, which comprises seven highly conserved collinear helicase motifs, remain intact. To date, more than 150 mutations of ATRX have been described. The majority of mutations reported so far in ATRX are missense mutations (about 100 missense mutations have been reported), and clustered in the two ADD and helicase domains. Mutations in the ADD domain are significantly associated with more severe psychomotor impairment than mutations in the helicase domain. The carboxyl terminus of ATRX encodes two additional domains of potential functional importance. The P-box is an element conserved among other SNF2-like family members involved in transcriptional regulation, and a stretch of glutamine residues (Q-box) represents a potential protein interaction domain. Of interest, it has been reported that in the absence of this carboxyl-terminal domain, severe urogenital abnormalities occur, such as female phenotype with streak gonads or ambiguous genitalia, as was observed in our case. It appears likely that this region plays a specific role in urogenital development.

Theoretically, the messenger RNA with c.7054delG seems to result in the nonsense-mediated messenger RNA decay, because this frameshift occurs in the third last exon of ATRX. However, previous studies have shown that some truncating mutations in ATRX with premature stop codon escape from nonsense-mediated messenger RNA decay, and truncated protein was seen in western blotting with protein extracts derived from cell lines carrying these mutations. Further, some full-length ATRX protein has been identified in protein extracts derived from cell lines carrying these truncating mutations. Therefore, the mutant Asp2352fs*1 ATRX protein could be expressed to some extent, which results in the ambiguous genitalia in our case.

Alpha-thalassemia is one of the defining elements of this syndrome (85% in cases) and is a useful reminder that a simple investigation can be used to establish a diagnosis. However, it is well known that there is considerable variation in the hematologic manifestations associated with ATRX mutations. A diagnosis of ATRX in a neonate without anemia essentially depends on facial

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**Table 1. Endocrinological findings in the patient**

| Stimulus                      | 4 months | Reference |
|-------------------------------|----------|-----------|
|                               | Basal    | Peak      | Basal | Peak               |
| LH (miU/ml)                   |          |           |       |                   |
| GnRH                          | 4.4      |           | 115.48| 13.11–25.15       |
| FSH (miU/ml)                  | 15.93    | →         | 81.1  | 2.12–5.24         |
| Testosterone (ng/ml)          |          |           |       |                   |
| hCG                           | 1.549    | →         | 3.52  | 2.0–7.5           |
| Dihydrotestosterone (ng/ml)   |          |           | 0.77  | 0.2–1.01          |
| TSH (miU/ml)                  | 3.37     |           | 0.77  | 11.0–149          |
| Free T4 (ng/dl)               | 0.9      |           |       | 1.01–1.95         |
| Free T3 (pg/ml)               | 2.12     |           |       | 2.23–5.30         |
| IGF1 (ng/ml)                  | 34.9     |           |       |                   |

Abbreviations: FSH, follicle-stimulating hormone; GnRH, gonadotropin-releasing hormone; hCG, human chorionic gonadotropin; IGF1, insulin-like growth factor 1; LH, luteinizing hormone; TSH, thyroid-stimulating hormone.

*Reference data of pubertal (Tanner stage II–III) Japanese boys.

**Figure 1.** Clinical features of the patient. (a) The patient showed ambiguous genitalia at 4 months of age. Micropenis (penile length was 1.9 cm) with penoscrotal hypospadias was observed. No masses were palpable bilaterally. There was no skin hyperpigmentation. (b) Photographs of the feet. The right foot had two additional toes.

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*Reference data of pubertal (Tanner stage II–III) Japanese boys.

1Reference data of Japanese adult male.

*Reference data of Japanese boys (0 years old).

The conversion factors to the SI unit are as follows: LH 1.0 (IU/l), FSH 1.0 (IU/l), testosterone 0.035 (nmol/l), TSH 1.0 (miU/l), free T4 12.87 (pmol/l), and free T3, 1.54 (pmol/l) and IGF-I 0.131 (nmol/l).
gestalt, DSD in males and psychomotor development delay combined with other nonspecific findings. Facial dysmorphism, such as telecanthus or widely spaced eyes, short nose, tented vermilion of the upper lip, and thick or everted vermilion of the lower lip, are too subtle to be easily identified, particularly at younger ages. Therefore, the diagnosis of ATRX without anemia based on clinical manifestation is not necessarily easy in a neonate. However, the early diagnosis of ATRX is important for medical intervention and follow-up, as many children with ATRX have gastrointestinal symptoms, such as profound gastroesophageal reflux, which may lead to aspiration pneumonia and is a known cause of death in this syndrome, difficulties in feeding manifested often by abdominal pain, and distention and the patient’s refusal for food, along with DSD and intellectual disability. Through whole-exome sequencing, we successfully identified a causative mutation in ATRX in this male patient, who did not exhibit anemia.

In summary, we described a patient with syndromic DSD harboring a novel hemizygous mutation in ATRX. Our study provides additional evidence that deletion of the carboxyl terminus of ATRX is associated with severe genital anomalies. Because whole-exome sequencing enables the screening of most protein-coding genes in an unbiased manner, this technique may help to identify atypical or previously unappreciated phenotypes associated with a known disease-associated gene.

HGV DATABASE

The relevant data from this Data Report are hosted at the Human Genome Variation Database at http://dx.doi.org/10.6084/m9.figshare.hgv.1345.

Supplementary Information for this article can be found on the Human Genome Variation website (http://www.nature.com/hgv)

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COMPETING INTERESTS

The authors declare no conflict of interest.

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