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Intriguing link between fetal intracranial hemorrhage and X-linked recessive chondrodysplasia punctata

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X-linked chondrodysplasia punctata 1 (CDPX1) is a rare congenital disorder that only affects males. It manifests as a non-severe skeletal dysplasia characterized by nasomaxillary hypoplasia, punctate cartilage calcifications, and brachytelephalangy[1]. This condition is due to constitutional defects in the ARSL gene (OMIM #302950). ARSL protein, an arylsulfatase, likely influences the delicate balance between osteoclast and osteoblast activity in cartilaginous tissue.[2]

Our study involves 7 male fetuses from 6 unrelated French families recruited retrospectively between 2011 and 2021. Antenatal findings, postnatal clinical observations, and genetic data are detailed in Table 1. Molecular diagnoses were established through a combination of array-CGH and Sanger sequencing, with comprehensive sequencing of all 11 exons and adjacent intronic regions of the *ARSL* gene. All subjects gave their informed consent before participating in the study.

Fetal demise within the 18th to 29th gestational weeks was observed in all cases of our cohort. Among them, 4 fetuses displayed anomalies in the second trimester, including 2 suspected intracranial hemorrhages. Autopsies were conducted in 5 cases, all of which confirmed the presence of intracranial hemorrhage. In 6 out of the 7 cases, pallor and/or anemia were observed, primarily attributed to the intracranial hemorrhage. This condition was verified in 4 cases through autopsy or CT scans, while 2 cases showed suspected intracranial hemorrhage, evident from specific indications of cephalic pole congestion and enlargement (Figure 1). Other clinical and radiological findings were consistent with those previously described in CDPX1 (Figure 2).

In 3 out of 6 families, CDPX1 was linked to hemizygous Xp22.3 deletions encompassing the entire *ARSL* gene (NM_000047.3). In the remaining three families, CDPX1 was associated with loss-of-function *ARSL* point mutations. Familial segregation analyses indicated that only one mutation was *de novo* while the other cases were maternally inherited. Concerning the 3 point mutations (p.G391R, p.G434S, p.G376R), they were mapped onto a 3D model of the ARSL protein, indicating their proximity to the catalytic center (Cys86) and the likelihood of causing destabilization (Figure S1). Two of these variants (p.G391R and p.G434S) were demonstrated to exhibit reduced functional activities compared to the wild-type enzyme[3]. The p.G376R variant, reported for the first time, is predicted to have an impact on splicing, likely abolishing the wild-type splice site and leading to a complete loss of enzymatic activity. Lastly, any substitution of G376 is projected to be pathogenic, as indicated by the AI AlphaMissense[4],

suggesting that G376 is an indispensable amino acid for the proper function of the ARSL protein.

This report signifies the initial documentation of major intracranial hemorrhage in CDPX1, expanding our insight into the condition's phenotype and emphasizing the critical nature of the second trimester of pregnancy for these fetuses, during which they appear to be more vulnerable to intracranial bleeding. Determining the precise rate of intracranial hemorrhage in CPDX1 remains an imperative task. As the pregnancy progresses, the risk may abate, offering the potential for a favorable outcome, as exemplified by the living CPDX1 patients.

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FIGURE LEGENDS

Figure 1. Postmortem findings suggestive of or confirming intracranial hemorrhage. Visible body pallor and pronounced cephalic congestion in Cases 4 (a) and 1A (c); hemorrhagic cerebral tissues post-formalin fixation in Case 4 (c); bilateral subdural hematoma with associated ventriculomegaly observed in the CT scan of Case 2 (d).

Figure 2. Classic prenatal and postmortem findings in CPDX1. Observations of the Binder facial dysmorphia (maxillonasal dysplasia) in Cases 3 (a) and 2 (e); punctate epiphyses evident in Cases 3 (b, d); and the presence of brachytelephalangy in Case 3 (c).

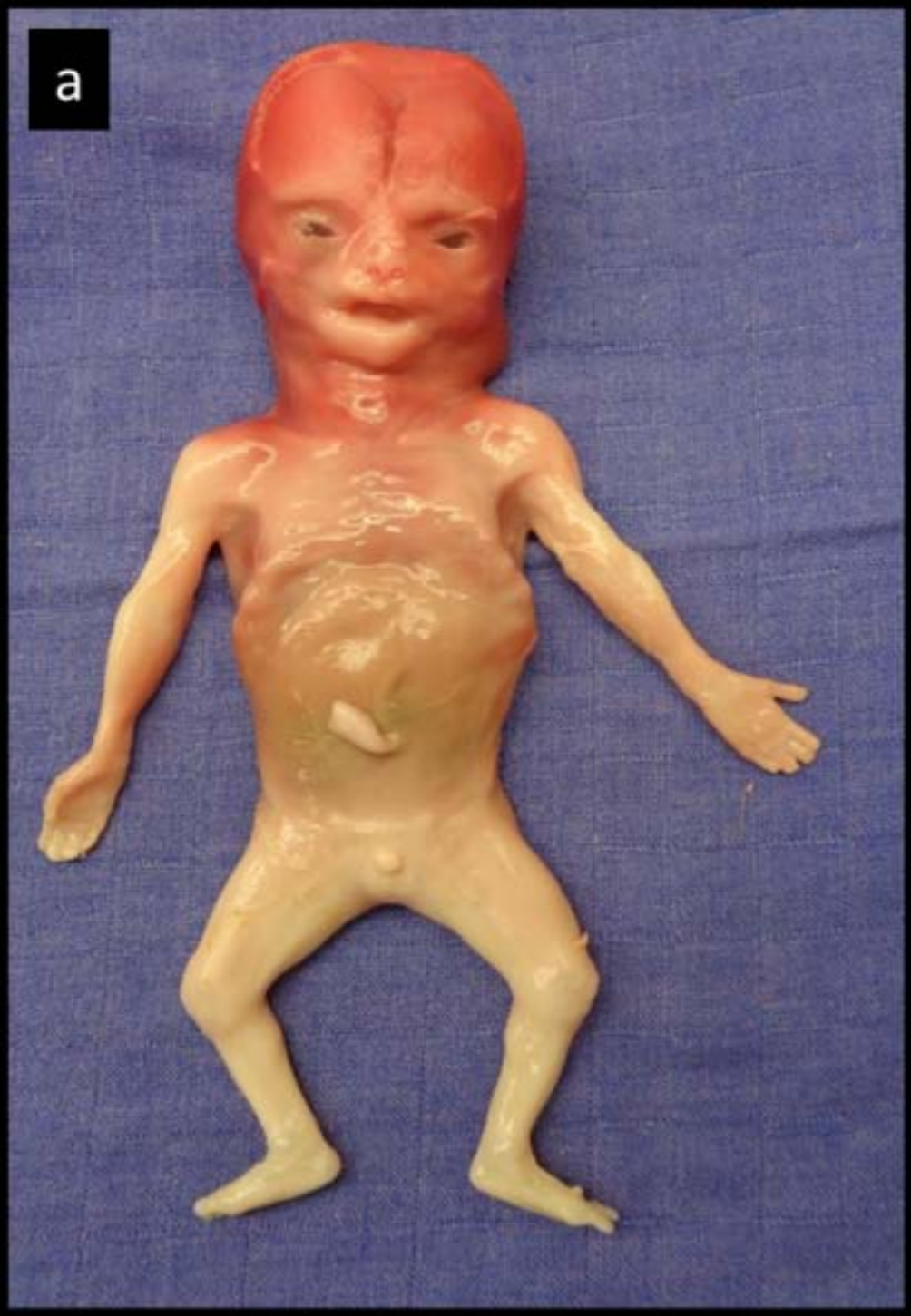
Table 1- Antenatal, postmortem and genetic data of 7 fetuses with CPDX1

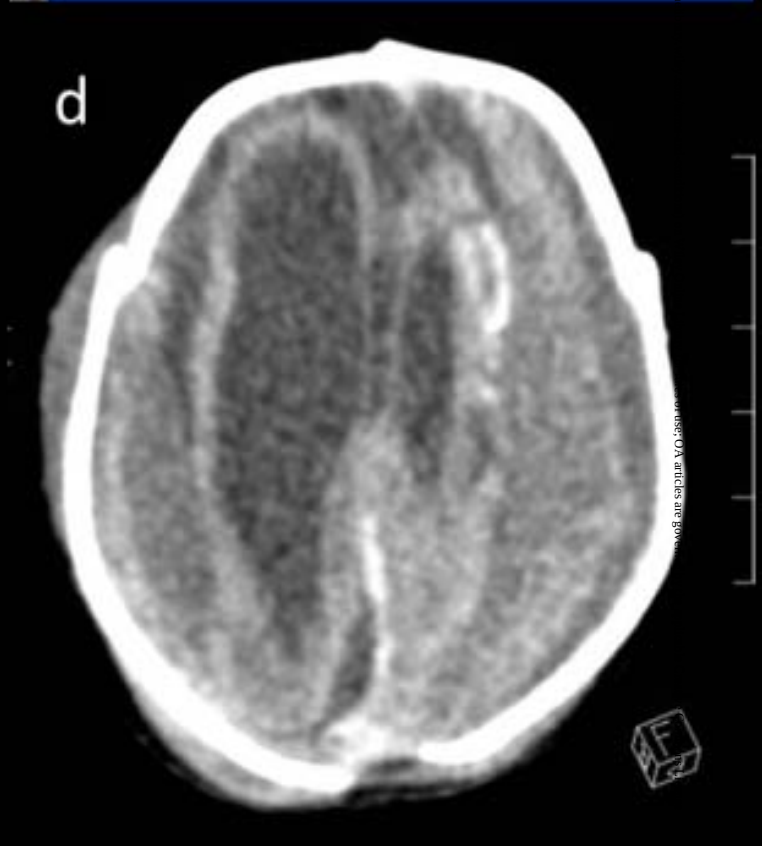
	Case 1A	**Case 1B**	**Case 2**	**Case 3**	**Case 4**	**Case 5**	**Case 6**
Gender	M	M brother of 1A	M	M	M	M	M
Pregnancy outcome	IUFD at 18 GW	TOP at 21 GW	neonatal death after PTD at 29 GW	IUFD at 23 GW	IUFD at 19 GW	IUFD at 22 GW	IUFD at 26 GW
Genetic alteration (CGCh37/hg19)	delXp22.33 (chrX:1314894-3248235) 1.6Mb maternally inherited		*ARSL* mutation c.1171G>A p.Gly391Arg de novo mutation	del Xp22.3 (chrX:2766830-7017221) 4,25 Mb maternally inherited	del Xp22.3 (chrX:61091-4473572) 4,41 Mb maternally inherited	*ARSL* mutation c.1300G>A p.Gly434Ser maternally inherited	*ARSL* mutation c.1126G>C p.Gly376Arg maternally inherited
Classification	Pathogenic loss		Reported pathogenic missense mutation	Pathogenic loss	Pathogenic loss	Reported pathogenic missense mutation	Splice site abolition; new pathogenic variant
Antenatal findings	none	intracranial hemorrhage suspicion	Binder phenotype, short long bones, intracranial hematoma and ventriculomegaly	Binder phenotype, femoral epiphyseal calcification, ventriculomegaly	none	none	Binder phenotype, short long bones
Postnatal findings							
Short long bones	-	+	+	-	+	-	+
Binder phenotype	-	-	+	+	+	+	+
Brachytelephalangy	-	-	+	+	+	NA	NA
Punctate epiphyses	-	-	+	+	+	+	NA

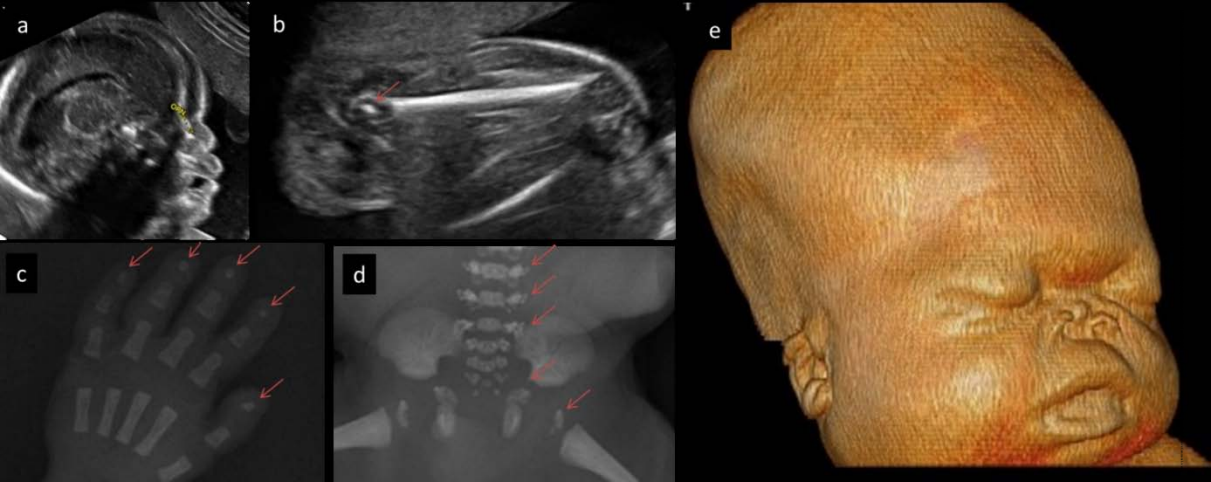
Body pallor or anemia	+	+	+	+	+	+	NA
Congestive cephalic pole	+	+	-	-	+	+	NA
Autopsy	yes	yes	no	yes	yes	yes	no
Intracranial hemorrhage	NA	+	+	+	+	NA	NA
Ventriculomegaly	NA	+	+	+	NA	NA	NA
Other	Enlarged fontanel	Enlarged fontanel, hepatomegaly	Bilateral subdural hematoma	none	none	macrocrania	none

GW, gestational weeks; NA, not available; PTD, preterm delivery; TOP, termination of pregnancy; IUFD, in utero fetal demise

Binder phenotype: maxillonasal dysplasia



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