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Another Case of Imprinting Defect in a Girl with Angelman Syndrome Who Was Conceived by Intracytoplasmic Sperm Injection

To the Editor:

Intracytoplasmic sperm injection (ICSI) has been established as an efficient treatment for male infertility and also as a supplement to in vitro fertilization (IVF) without obvious male infertility. ICSI is now regarded as a procedure that is safe overall, and no increase in developmental delay was found in a follow-up of 221 ICSI-conceived children in the 2nd year of life (Sutcliffe et al. 2001). However, the possibility of an increased risk of imprinting defects has been raised (Manning et al. 2000). Two children conceived by ICSI who had Angelman syndrome (AS [MIM 105830]) due to a presumably sporadic imprinting defect have recently been reported (Cox et al. 2002).

We here report a 3.5-year-old girl with AS due to a sporadic imprinting defect, born of a pregnancy that was also the result of ICSI. The girl was the first child of a 35-year-old mother and a 36-year-old father. The father has a healthy daughter by another partner, and sperm analysis was normal on three different occasions. The mother had one spontaneous abortion and two extrauterine pregnancies before treatment with IVF. Traditional IVF did not result in fertilized eggs, and ICSI was therefore performed in spite of the normal sperm analysis of the father. The first ICSI pregnancy resulted in another spontaneous abortion, whereas the second ICSI procedure resulted in a normal pregnancy. Birth was at term, birth weight was 3,760 g, length was 54 cm, and head circumference was 36 cm (75th percentile). Development was considered normal for the first 3–4 mo, after which she started to have infections. She walked at age 2 years. She had no epilepsy but had an abnormal electroencephalogram with large-amplitude slow-spike waves. There was no language development. Chromosomes, including subtelomeres, were normal. At age 3 years, her height and weight were at the 50th percentile, whereas her head circumference was 1 cm below the 2.5th percentile. She was mentally retarded and atactic. She was dysmorphic, with a square face, deep-set eyes, and a protruding tongue.

FISH analysis using the *SNRPN* probe (MIM 182279), as well as microsatellite studies, revealed normal chromosomes 15 of biparental origin. A common large deletion of 15q11-q13 and uniparental paternal disomy could therefore be excluded. Methylation-specific Southern blot analysis and methylation-specific PCR (Zeschnigk et al. 1997) for the *SNRPN* locus showed the presence of a normal unmethylated paternal band and the complete ab-

sence of a methylated maternal band, indicating that the patient had an imprinting defect. Quantitative Southern blot analysis of the critical AS imprinting center (IC) region (Buiting et al. 1999) showed a normal dosage; therefore, an IC deletion was unlikely. This result was confirmed by sequence analysis of the 880-bp AS-IC element, where the patient was heterozygous for three different SNPs. Both parents had normal chromosomes and a normal methylation pattern. These findings suggest that the patient belongs to the group of patients with a sporadic imprinting defect (Buiting et al. 1998).

Both patients reported by Cox et al. (2002) had fathers with sperm abnormalities, and the possibility that the imprinting defect could be related to male infertility was discussed. The father of the child reported here had normal sperm, and a relationship to male infertility is therefore unlikely. However, similar to the mother, the maternal grandmother also had a history of reproductive difficulties. In addition to three healthy children, she had four spontaneous abortions and one daughter who was stillborn at term. Since both the maternal grandmother and the mother had reproductive difficulties, a maternal oogenesis defect cannot be excluded.

A sporadic imprinting defect is a very rare cause of AS, and Cox et al. (2002) therefore considered a relationship to the ICSI procedure to be likely. The report of a third patient with this rare disorder further supports the assumption that ICSI can lead to an increased risk for imprinting defects.

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Electronic-Database Information

Accession numbers and the URL for data presented herein are as follows:

Online Mendelian Inheritance in Man (OMIM), http://www.ncbi.nlm.nih.gov/Omim/ (for the *AS* gene [MIM 105830] and the *SNRPN* probe [MIM 182279])

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