**46,XY and 45,X/46,XY testicular dysgenesis: similar gonadal and genital phenotype, different prognosis**

**SUMMARY**

The objective of this study was to describe the change in diagnosis and prognosis of a child with testicular dysgenesis and 46,XY karyotype after detection of a 45,X cell line and to discuss the difficulties caused by the terms mixed gonadal dysgenesis (MGD) and XY partial gonadal dysgenesis (XYPGD). One case was reported including clinical and laboratory findings of a child of 41-day-old infant with 1.3-cm phallus, penoscrotal hypospadias and left prepubertal testis. Karyotype 46,XY (16 cells), normal hormone levels. Right streak gonad, epididymis and müllerian remnants were removed; initial diagnosis was XYPGD. Persistent growth retardation led to further cytogenetic analysis (50 cells) and detection of a 45,X cell line. Detection of a 45,X lineage changed both the diagnosis to MGD and also the prognosis. The number of cells analyzed in karyotyping is critical. Use of MGD and XYPGD to designate both a histological picture and a syndromic diagnosis, results in lack of emphasis on clinical differences between 46,XY and 45,X/46,XY subjects.

In 2005, the Chicago Consensus on Management of Intersex Disorders proposed the substitution of terms such as intersex, hermaphroditism and pseudo-hermaphroditism for the term disorders of sex development (DSD), as defined by congenital conditions in which development of chromosomal, gonadal, or anatomical sex is atypical. A classification was also proposed in which DSD associated with sex chromosome abnormalities (Sex Chromosome DSD) were separated from those with a normal chromosome complement (46,XX DSD and 46,XY DSD) (Table 1). Disorders of gonadal development, including ovotesticular DSD (proposed nomenclature for true hermaphrodite) and gonadal dysgenesis, can be found in all groups (1,2).

Gonadal dysgenesis is a defective embryonic development of the gonads. Failure in gonadal development may result in extremely hypoplastic and dysfunctioning gonads mainly composed of fibrous tissue (streak
Table 1. DSD classification proposed by the Chicago consensus (1,2)

| Sex chromosome DSD                  | Disorders of gonadal (testicular) development | Disorders of gonadal (ovarian) development |
|-------------------------------------|-----------------------------------------------|------------------------------------------|
| 45, X (Turner syndrome and variants) | Complete gonadal dysgenesis (Swyer syndrome), partial gonadal dysgenesis, testicular regression, ovotesticular DSD | Gonadal dysgenesis, testicular DSD*, ovotesticular DSD |
| 47,XY (Klinefelter syndrome and variants) | Androgen biosynthesis defect (e.g., 17-hydroxysteroid dehydrogenase deficiency, 5α-reductase 2 deficiency), defect in androgen action (androgen insensitivity syndromes), LH receptor deficiency, disorders of AMH and AMH receptor (persistent müllerian duct syndrome) | Androgen excess: fetal (e.g., 21-hydroxylase deficiency), fetoplacental (e.g., aromatase deficiency), maternal (e.g., luteoma, exogenous) |
| 45, X/46, XY (mixed gonadal dysgenesis, ovotesticular DSD) | Other: e.g., severe hypospadias, cloacal extrophy | Other: e.g., MURCS association, cloacal extrophy |
| 46, XX/46, XY (chimeric, ovotesticular DSD) | | |

* Proposed nomenclature for XX male.
AMH: anti-müllerian hormone; DSD: disorder of sex development; MURCS: müllerian duct aplasia, renal aplasia and cervicothoracic somite dysplasia.

gonad) or may lead to incomplete testicular development (dysegnetic testis). The finding of bilateral streak gonads in phenotypic females with a 46,XX or 46,XY karyotype characterizes pure gonadal dysgenesis.

Dysgenetic testes may be bilateral or associated with contralateral streak in subjects with a 46,XY or 45, X/46,XY karyotype. The histological picture of a dysgenetic testis ranges from a gonad with predominance of fibrous tissue and a few tubular structures to only a reduction in tubular size and reduced number of germ cells. As a consequence, the external genitalia of patients with dysgenetic testes ranges from predominantly male to predominantly female, including cases of striking genital ambiguity, and there is usually persistence of müllerian structures (3).

The term mixed gonadal dysgenesis (MGD) was initially used in a histological context, referring to the finding of a dysgenetic testis associated with a streak gonad. The same occurred with incomplete or partial gonadal dysgenesis (PGD), which designated the existence of bilateral dysgenetic testes (4,5). In subjects with 45, X/46,XY mosaicism, the histological picture of dysgenetic testis plus contralateral streak gonad (MGD) is more frequently observed than bilateral dysgenetic testes (PGD) (6,7), while among those with a 46,XY karyotype the frequencies are similar (6).

In clinical practice, however, as well as in the new classification proposed by the Chicago Consensus (1,2), the term MGD has been employed in cases of testicular dysgenesis with a 45, X/46,XY karyotype, and PGD in those with a 46,XY chromosome constitution, regardless of the histological picture.

In patients with a 45, X/46,XY karyotype, management includes not only issues related to sex assignment, gonadectomy and genitoplasty, but also those related to the clinical features of Turner syndrome derived from the 45,X cell line. We report on a child with sex ambiguity whose initial diagnosis was 46,XY PGD, and whose prognosis changed significantly after detection of a 45,X cell line.

CASE REPORT

Our patient, a 41-day-old infant, was referred to us before gender assignment due to sex ambiguity. The child was delivered by cesarean section after an uneventful 36-week pregnancy. Birth weight was 1,510 g, length 39 cm and head circumference 29 cm (all below the 3rd centile). He was the first child of young unrelated parents; according to them, two maternal cousins had micropenis and gynecomastia.

When first examined by us, weight was 2.360 g and length 45 cm (both below the 3rd centile). There was no dysmorphic picture, and genital examination revealed a 1.3 cm phallus with chordee, penoscrotal hypospadias, and scrotum with rugae and pigmentation. There was a right inguinal hernia containing a palpable gonad and the left gonad was in the scrotal fold, both measuring 1 cm at the greatest diameter. On pelvic sonography, the kidneys had normal size and location, with two small anechoic cortical cysts in the right one.

Hormonal evaluation at the age of 41 days revealed normal gonadotropin levels (FSH = 3.89 mU/mL, normal range (NR) = 1.5-12.4; LH = 1.89 mU/mL, NR = 1.7-8.6), and also normal levels of total testosterone (2.00 pg/mL; NR > 1.5), 17 OH progesterone (1.36 ng/mL, NR = 1.06-40.41), androstenedione (1.0 ng/mL, NR = 0.4-2.00) and SDHEA (< 15 µg/dL,
NR = 2-215). G-banding karyotype in 16 cells revealed a 46,XY karyotype.

After three injections of testosterone enanthate (50 mg over successive months), phallus size increased to 3 cm and a second urethral opening with urinary flow was detected on the glans. Biopsy of the left gonad revealed a prepubertal testis. After discussions with the parents, the child received a male sex assignment.

At the age of 8 months, laparoscopy showed a small right gonad which was removed together with an epididymus and with müllerian remnants. This gonad was found to be dysgenetic, with fibrovascular tissue and absence of germ cells.

During follow-up the child’s length remained below 3.5 standard deviations. Thus, another karyotype was performed to analyze a higher number of cells. As a result, 10/50 cells had a 45,X constitution, thus changing the karyotype to 45,X/46,XY. Clinical reevaluation in search for dysmorphic signs did not reveal significant abnormalities. Screening for specific conditions associated with Turner syndrome revealed no cardiovascular abnormalities. Screening for metabolic disorder, of a dysmorphic picture which could indicate a syndromic picture as well as careful evaluation of the external genitalia and palpation of the gonads. The chromosome complement, 46,XX or 46,XY, is shown in karyotype, which may also reveal numeric and structural chromosome abnormalities. Depending on further evaluations, particularly hormone measurements, surgical exploration and gonadal biopsy may be necessary to reach a specific diagnosis (1,2).

In this case, as the child had normal LH, FSH and testosterone levels, a testis in the scrotal fold and good response to testosterone injections, he had a male sex assignment. As surgical exploration revealed a streak gonad with ipsilateral müllerian and wolffian remnants, the diagnosis of XY partial gonadal dysgenesis was initially established. There are no large studies on the prognosis of XY partial gonadal dysgenesis regarding spontaneous puberty and fertility; on the other hand, there are also no known associations with other congenital anomalies.

Although the histological picture of testis and streak gonad may occur in individuals with a homogeneous 46,XY karyotype, the finding of pre- and postnatal growth retardation in this case led to chromosome analysis of a larger number of cells and detection of a 45,X cell line. This finding changed the diagnosis (from 46,XY DSD, partial gonadal dysgenesis, to sex chromosome DSD, mixed gonadal dysgenesis), the prognosis (short stature, risk of cardiovascular, renal and urinary anomalies, autoimmune thyroid disease, among others) and the follow-up of the child (which must be similar to those of Turner syndrome patients) (8), and also brought the possibility of hGH treatment to improve final height (9).

Results of G-banding karyotype from peripheral blood lymphocytes must be carefully evaluated, because the number of metaphases analyzed is a critical factor to detect low frequency cell lines. If 16 cells are counted, as is done routinely in most cytogenetic laboratories, an 18% mosaicism is excluded with 95% confidence. When 50 or 100 cells are analyzed, a 6% and 3% mosaicism is excluded with 95% confidence, respectively (10). In addition, karyotype analysis of both lymphocytes and gonadal fibroblasts has revealed different levels of mosaicism, and the phenotype of gonads and external genitalia is more consistent with the chromosome constitution of gonads than that of lymphocytes (11). In some instances, fluorescent in situ hybridization (FISH) analysis may be necessary for detecting low frequency mosaicism through analysis of a large number of cells.

The confusion brought about by the use of the terms mixed and partial gonadal dysgenesis, which in recent studies are still applied to histological rather than clinical pictures (4,12), results in lack of emphasis on significant differences in prognosis and clinical follow-up between 46,XY and 45,X/46,XY subjects. A wide use of the classification proposed by the Chicago Consensus is necessary to avoid misunderstanding both in research and clinical practice. Alternatively, a diagnostic label which included both the presence of testicular dysgenesis and the result of the karyotype (46,XY testicular dysgenesis and 45,X/46,XY testicular dysgenesis) could also be useful.

Finally, if one takes into account the limitations of routine karyotype analysis in diagnosing low frequency mosaicism, it is essential to have at least 50 metaphases counted before a diagnosis of 46,XY testicular dysgenesis is performed; FISH analysis should also be
performed when the patient with a 46,XY karyotype in 50 cells has characteristic features of Turner syndrome.

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