Differences of sex development with chromosomal mosaicism: histological characterization and immunohistochemistry markers in gonads during childhood

INTRODUCTION

Differences/disorders of sex development (DSD) comprise a group of congenital conditions, affecting human sex determination and/or differentiation. Patients with DSD are classified in: sex chromosome DSD, 46XY DSD and 46,XX DSD. Sex chromosome disorders, including sex chromosome mosaicism, result in a large clinical spectrum. There is scarce information about the histological pattern of gonads from these patients.

OBJECTIVE

The aim of this study was to characterize the histology and cell markers pattern in gonads of patients with chromosomal mosaicism.

CLINICAL MATERIAL AND METHODS

- Gonadal biopsies from 13 prepubertal patients with chromosomal mosaicism, including chromosome Y were studied.
- Six were rearing as male (M) and 7 as female (F).
- Patients were divided in two subgroups (G), according to external genitalia phenotype:
  - G1, with atypical genitalia: n=7, chronological age (CA) at biopsy, was 1.75, 0.25-12 years (y) expressed as median and range. Five patients were rearing as M and two as F.
  - G2, Turner syndrome: n=6, CA at biopsy was 13.8, 3.5-18.8 y, all patients were reared as females.
- H&E sections from gonadal biopsies were observed by two specialists.
- Immunohistochemical (IHC) analysis:
  - Sertoli cells (anti-SOX9 goat polyclonal 1:500, AF3075, R&D Systems).
  - Ovarian follicular cells (anti-FOXL2 goat polyclonal 1:250, ab5096, Abcam).
  - Pluripotent germ cells (anti-OCT3/4 mouse monoclonal 1:50, sc-5279, Santa Cruz Biotechnology).

RESULTS

Table 1. Histological characteristics of gonadal biopsies from prepubertal patients.

<table>
<thead>
<tr>
<th></th>
<th>Testis</th>
<th>Ovary</th>
<th>Müllerian remnants</th>
<th>Epididymis</th>
<th>Streak</th>
<th>Testis &amp; Müllerian remnants</th>
</tr>
</thead>
<tbody>
<tr>
<td>G1</td>
<td>6/7 (86%)</td>
<td>0/7 (0%)</td>
<td>3/7 (43%)</td>
<td>2/7 (29%)</td>
<td>3/7 (43%)</td>
<td>2/7 (29%)</td>
</tr>
<tr>
<td>G2</td>
<td>2/6 (33%)</td>
<td>1/6 (17%)</td>
<td>5/6 (83%)</td>
<td>1/6 (17%)</td>
<td>3/6 (50%)</td>
<td>1/6 (17%)</td>
</tr>
<tr>
<td>Total</td>
<td>8/13 (62%)</td>
<td>1/3 (8%)</td>
<td>8/13 (62%)</td>
<td>3/13 (23%)</td>
<td>6/13 (46%)</td>
<td>3/13 (23%)</td>
</tr>
</tbody>
</table>

In 4 samples of 2 patients of G2 gonadoblastoma, embryonic carcinoma and dysgerminoma were found. Of the samples that presented testicular parenchyma, 43% had structures compatible with Müllerian remnants.

Table 2. Immunohistochemical characterization of gonadal biopsies from prepubertal patients.

<table>
<thead>
<tr>
<th></th>
<th>OCT ¾ +</th>
<th>SOX9 +</th>
<th>FOXL2 +</th>
</tr>
</thead>
<tbody>
<tr>
<td>G1</td>
<td>57%</td>
<td>75%</td>
<td>33%</td>
</tr>
<tr>
<td>G2</td>
<td>67%</td>
<td>50%</td>
<td>100%</td>
</tr>
<tr>
<td>Total</td>
<td>60%</td>
<td>67%</td>
<td>60%</td>
</tr>
</tbody>
</table>

* OCT 3/4 expression in G1 corresponded to testicular parenchyma in all cases.
All the patients were older than 3 months.
=b SOX9 was present in the nucleus of Sertoli cells inside the seminiferous cords in G1.
© Positive expression of SOX9 was found in isolated nuclei of tissues of G2 without seminiferous cords.

CONCLUSIONS

- The complexity of the tissues corresponding to patients with chromosomal mosaicism requires a deep histological and immunohistochemical analysis that allow the characterization of cell types and cell cancer risk.
- Samples of G1 (2/7) showed testicular parenchyma and Müllerian remnants, which might indicate an early alteration in the function of the Sertoli cell.
- The expression of Sertoli cell marker SOX9 in streak tissues of G2, suggests an increased risk of gonadoblastoma development.