The localization of cells with XX and XY in gonadal tissues associated with ovoestrous disorder of sexual development with a 46, XX/46, XY karyotype

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Background

Individuals with a mixed 46, XX and XY karyotype have gonads with either an ovary in one side and a testis in the other side or an ovoestrous in both which both ovarian and testicular tissues exist in the same gonadal tissue. Such conditions are categorized as the ovoestrous disorder of sexual development (OvDS). The question arises as to how cells with 46, XX and 46, XY distribute in ovoestrous and separated bisexual gonads in 46, XX and 46, XY OvDS.

Objective

The aim of this study was to investigate the distribution of sex chromosomes (XX and XY) in testicular and ovarian tissues in gonads associated with 46, XX and 46, XY.

Methods

Six gonadal tissues from three patients with a 46, XX/46, XY karyotype were available for immunofluorescence in situ hybridization (FISH), immunohistochemistry (IHC) and immunofluorescence (IF).

1) We reviewed histopathological features of the gonads and performed FISH for X and Y chromosomes to examine the distribution of sex chromosomes in the testicular and ovarian structures.

2) The localization of SOX9 (testicular marker, BA03757, R&AD SYSTEMS, goat polyclonal) in Sertoli cells and FOXL2 (ovarian marker, IMG-128, IMGENEX, goat polyclonal) in ovarian follicular epithelium2 was ascertained by IHC and IF.

Results

1. The clinical, pathological, and cytogenetic data of three patients are summarized in Table 1.

2. Histopathology of the gonads (Fig. 1)

Case 1 had an ovoestrous in the left gonad (A, B) and an ovary in the right gonad (C). Case 2 had a testis in the left (D) and an ovary in the other gonad (E).

Case 3 had a dysgenetic testis in the left gonad (F) and a normal appearing testis in the right gonad (G). No ovarian tissue was evident because the gonadal examination was performed in infancy stages which occasionally contain too little gonadal parenchyma to reliably have any apparent ovarian tissue.

3. FISH analysis for X and Y chromosomes in the gonads (Fig. 2)

Sertoli cells with XX signals were scattered in seminiferous tubules of the ovotestis (Case 1: Fig. 2C), testes (Case 2: 3: Fig. 2D, F) where most Sertoli cells showed XY signals. Conversely, a small amount of ovarian epithelial cells with XY signals were unequivocally present in ovarian follicles in the ovotestis (Case 1: Fig. 2A, B) and ovary (Case 2: Fig. 2E) where the majority of cells had XX signals (Fig. 2A, B, E).

4. IHC and IF for SOX9 and FOXL2 in the ovotestis (Fig. 3)

Since XX cells were observed in the seminiferous tubules and XY cells were seen in the ovarian follicles, we examined whether or not a testicular lineage marker (SOX9) is expressed in the ovotestis and an ovarian marker (FOXL2) is expressed in the testicular tissue. SOX9 was exclusively expressed in the nucleus of Sertoli cells and FOXL2 was also exclusively expressed in the nucleus of ovarian follicular epithelial cells in Case 1 (Fig. 3A-D). Case 2 and 3 showed the same results as Case 1.

Discussion

• It is indicated that precursor sex cord cells with an XX karyotype incorporated in testicular tissues might be able to alter its lineage and differentiate into Sertoli cells because all of the epithelial cells in the seminiferous tubules solely expressed SOX9. Similarly, precursor sex cord cells with an XY karyotype distributed in ovarian tissues might be able to differentiate into ovarian follicular epithelium because all of ovarian follicular epithelial cells solely expressed FOXL2 (Table 2).

• It is suggested that the destiny of individual gonadal epithelial cells is influenced by local environmental factors rather than by the sex chromosomal type.

Table 1. Clinical, pathological, and cytogenetic data

<table>
<thead>
<tr>
<th>Case</th>
<th>Gender</th>
<th>Age of onset</th>
<th>External genitalia</th>
<th>Karyotype (Peripheral blood)</th>
<th>Gonads Left/Right</th>
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<tbody>
<tr>
<td>1</td>
<td>Male</td>
<td>3 years old (lt. gonad, rt. hypo)</td>
<td>microphene, lt. cryptorchism, hypoplasia</td>
<td>46, XX [81]/46, XY [19]</td>
<td>Ovoestrous</td>
</tr>
<tr>
<td>2</td>
<td>Male</td>
<td>10 months old (lt. gonad, rt. hypo)</td>
<td>microphene, lt. cryptorchism, hypoplasia</td>
<td>46, XX [11]/46, XY [7]</td>
<td>Hypoplastic testis</td>
</tr>
<tr>
<td>3</td>
<td>Male</td>
<td>13 months old (lt. and rt. hypo)</td>
<td>microphene, hypoplasia</td>
<td>46, XX [21]/46, XY [5]</td>
<td>Dysgenetic testis</td>
</tr>
</tbody>
</table>

Fig. 1. Histopathology of the gonads of 46, XX/46, XY patients

Fig. 2. FISH analysis for X and Y chromosomes in the gonads

Fig. 3. IHC (A, B) and IF (C, D) for SOX9 and FOXL2 in the ovotestis

Table 2. Relationship between sex chromosomes and sex lineage makers

<table>
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<tr>
<th>Cell type</th>
<th>SOX9</th>
<th>FOXL2</th>
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<tbody>
<tr>
<td>Sertoli cells</td>
<td>XY</td>
<td>+</td>
</tr>
<tr>
<td>XX</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Ovarian follicular epithelial cells</td>
<td>XX</td>
<td>-</td>
</tr>
<tr>
<td>XY</td>
<td>-</td>
<td>+</td>
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Reference