Evidence on the Male Reproductive Toxicity of BPA in Humans and Laboratory Animals

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Development of the Male Reproductive System

- Testis, epididymis, vas deferens, seminal vesicles, prostate, penis
- Milestones in testicular development: GD10-12
- Milestones in prostate development: PND 0, 35

<table>
<thead>
<tr>
<th>GD</th>
<th>0</th>
<th>12</th>
<th>21</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(birth)</td>
<td></td>
<td>(weaning)</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>21</td>
<td>40-45</td>
</tr>
<tr>
<td></td>
<td>(puberty)</td>
<td></td>
<td>(adult)</td>
</tr>
<tr>
<td></td>
<td>100-110</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

- anogenital distance
- reproductive performance
- nipple retention (pnd13)
- preputial separation
- Gross pathology
- Histopathology
- Immuno-histochemistry

### Occupational Studies of Male Reproductive Outcomes

<table>
<thead>
<tr>
<th>Study/Design</th>
<th>Population/Exposure Measure</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Hanaoka et al., 2002</strong></td>
<td>42 non-exposed workers, 42 exposed workers to bisphenol A diglycidyl ether (BADGE) Urinary BPA</td>
<td>↓ follicle stimulating hormone No association with luteinizing hormone or free testosterone</td>
</tr>
<tr>
<td>Cross-sectional</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Cha et al., 2008</strong></td>
<td>25 controls, 25 exposed workers Urinary BPA</td>
<td>↑ luteinizing hormone No association with testosterone or follicle stimulating hormone</td>
</tr>
<tr>
<td>Cross-sectional</td>
<td></td>
<td></td>
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</table>
# Male Reproductive Effects in Laboratory Animals

## Number of Studies *in vivo*

<table>
<thead>
<tr>
<th>Exposure Period</th>
<th>Pre</th>
<th>Neo</th>
<th>Peri</th>
<th>Pub</th>
<th>Adult</th>
<th>RACB</th>
<th>Multi</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Repro. Performance</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td>3</td>
<td>3</td>
<td></td>
<td>8</td>
</tr>
<tr>
<td>Testis</td>
<td>14</td>
<td>9</td>
<td>13</td>
<td>14</td>
<td>10</td>
<td>1</td>
<td>3</td>
<td>64</td>
</tr>
<tr>
<td>Epididymis</td>
<td>10</td>
<td>3</td>
<td>5</td>
<td>12</td>
<td>3</td>
<td>2</td>
<td>3</td>
<td>38</td>
</tr>
<tr>
<td>Seminal vesicles</td>
<td>6</td>
<td>1</td>
<td>6</td>
<td>9</td>
<td>5</td>
<td>2</td>
<td>3</td>
<td>32</td>
</tr>
<tr>
<td>Prostate</td>
<td>8</td>
<td>2</td>
<td>9</td>
<td>7</td>
<td>3</td>
<td>1</td>
<td>3</td>
<td>33</td>
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<tr>
<td>Sexual Maturation</td>
<td>7</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td></td>
<td></td>
<td>3</td>
<td>15</td>
</tr>
<tr>
<td>Hormones</td>
<td>5</td>
<td>3</td>
<td>5</td>
<td>7</td>
<td>3</td>
<td>2</td>
<td></td>
<td>22</td>
</tr>
</tbody>
</table>

RACB: Reproductive Assessment through Continuous Breeding; Multi: Multi-gen Reproductive Toxicity Studies

## Number of Studies *in vitro*

- Sertoli cells: 4
- Leydig cells: 5
- Prostate tissue or cells: 6

Office of Environmental Health Hazard Assessment
Effects on Reproductive Performance

Neonatal studies in rats: 2 studies
• s.c. inj., 2 µg/kg-d – 300 mg/kg-d. No effect.

RACB studies in mice: 3 studies
• Dietary (2), s.c. implants (1).
• Reduced number of live pups per litter at ≥ 437 mg/kg-d.

Two-generation studies: 2 studies
• Dietary, mice: apparent increase in still birth index (≥0.003 mg/kg-d).
• Gavage, rats: no overall effect. No data on live birth per litter.

Three-generation studies: 1 dietary study in rats
• Reduced number of live pups per litter in F1, F3, but not F2 generation at ≥ 0.22 mg/kg-d; significant at ≥ 434 mg/kg-d.
Effects on the Testis: In Vivo

42 in rats, 22 in mice
oral, s.c. injection, implant
0.002 – 1823 mg/kg-d

• Organ weight: 48 studies, 9 reported reduced weight.
  – LOELs: 0.002 - 600 mg/kg-d

• Histopathology (routine): 28 studies, 1 reported changes
  – LOEL: 600 mg/kg-d

• Sperm parameters: 26 studies, 10 reported reduction in sperm count or motility.
  – LOELs: 0.02 – 875 mg/kg-d
Effects on the Testis: In Vivo (continued)

- **Histopathology (quantitative or ultrastructural):**
  12 studies, 10 reported abnormal changes
  - LOELs: 0.1 – 20 mg/kg-d

- **Histopathology (immunostaining):**
  6 studies, 5 reported changes
  - LOELs: 0.024 – 0.1 mg/kg-d

- **Biochemistry (enzyme, gene expression, etc.):**
  7 studies, 6 reported abnormal changes
  - LOELs: 0.02 mg/kg-d
BPA Effects on the Testis: In Vitro

Sertoli Cells
• Primary cell cultures: 2 studies
  – BPA treatment: 10 – 300 µM for 24-48 hrs
  – Effects: increased apoptosis, alterations in functions.
• Cell lines: 2 studies
  – BPA treatment: 50 – 400 µM for 24-48 hrs

Leydig Cells
• Primary cell cultures: 2 studies
  – BPA treatment: 0.01 – 1000 nM for 18-24 hrs
  – Effects: varied, depending on the BPA concentration, strain/age of the rats used as cell donors.
• Cell lines: 3 studies
  – BPA treatment: 0.01 pM – 100 µM for 30 min - 48 hrs
  – Effects: alterations in functions and/or gene expression.
BPA Effects on the Prostate

24 in rats, 9 in mice
oral, s.c. injection, implant
0.002 – 1750 mg/kg-d

Parameters for prostate effects:
• Weight: whole prostate, lobe-specific
• Histopathology: routine, quantitative, immunostaining
**Effects on the Prostate: In Vivo**

Weights: 31 studies
- Prenatal studies: 1 of 6 studies reported increased prostate weights at doses ≥ 2.0 µg/kg-d.
- None of 11 neonatal or perinatal studies reported changes in the prostate weight.
- 4 of 13 adult, RACB, or multi-gen studies reported reduced prostate weights following exposure to BPA at mg/kg-d levels.

Histopathology (routine): 7 studies
- None of them reported abnormal changes.

Histopathology (quantitative or immunostaining): 5 studies
- All 4 prenatal or neonatal studies reported changes indicative of increased proliferation at doses ≥ 10 µg/kg-d.
- 1 study in adult mice reported abnormal differentiation.
**BPA Effects on the Prostate: In Vitro**

**Primary tissues or cells: 2 studies**
- BPA treatment: 0.01 – 1000 nM for 3-6 days
- Effects: alterations in gene expression, abnormal proliferation and differentiation

**Cell lines: 4 studies**
- BPA treatment: 0.1 – 100 nM for 1-8 days
- Effects: Increased proliferation, alterations in cellular response to androgens, and/or alterations in gene expression
Effects on the Epididymis, Seminal Vesicles, Sexual Maturation, and Hormones

Epididymis & Seminal Vesicles: 38 studies
• Most studies only reported organ weights with no histopathological data

Sexual Maturation: 15 studies
• Most studies used AGD, age of preputial separation, and/or nipple retention incidence

Hormonal Effects: 22 studies
• Most studies measured T levels at different ages after exposure

Most studies that found effects on these endpoints also reported effects on the testis or prostate in the same studies.
Male Reproductive Effects of BPA
Summary

• Very limited data in humans; numerous studies in lab animals.
• Reproductive performance:
  - Reduced number of live pups per litter.
• Testis:
  - Varied results on the testis weight, sperm parameters, and routine histopathology.
  - Consistently reported BPA effects on the testicular tissue when evaluated with quantitative pathology or molecular approaches.
  - Consistently reported BPA effects in cultured Sertoli or Leydig cells.
Male Reproductive Effects of BPA
Summary (continued)

- **Prostate:**
  - Varied results on the prostate weight.
  - No morphological changes using the routine histopathological method.
  - Consistently reported BPA effects on the prostate tissue when evaluated with quantitative pathology or immunostaining approaches.
  - Consistently reported BPA effects in cultured prostate tissues or cells.

- **Other organs/endpoints:**
  - Results from different studies varied.
  - Effects observed are often associated with testicular or prostate effects in the same study.