Evaluation of PFOA and PFOS for human health standards

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Today’s presentation

Basis for human health standards

Groundwater standard process

Recommended groundwater standard for PFOA

Recommended groundwater standard for PFOS
Standards are set to protect health of Wisconsin residents.
Most human health standards are based on toxicology studies conducted in research animals.
Dose response experiments are used to figure out **how much** of a chemical is needed to cause an effect.
Most effects have a threshold.

There is some level below which these effects are not expected to occur.
No Observable Adverse Effect Level (NOAEL)

Highest tested dose without a response
LOAEL
Lowest Observable Adverse Effect Level
Lowest tested dose with a response
Cancer effects are usually considered to not have a threshold.

Any level can increase the cancer risk.
Cancer slope factor = cancer risk per unit of dose

Dose
Most human health standards are based on toxicology studies conducted in research animals.

Toxicology studies called dose response experiments are used to figure out how much of a chemical is needed to cause an effect.

**NOAEL**  **LOAEL**  **Cancer slope factor**
Toxicology data are used to set standards that protect health of Wisconsin residents.
Groundwater
Two-thirds of Wisconsin residents use groundwater.
Wisconsin’s groundwater standards have 2 parts.

Enforcement Standard

Preventive Action Limit
Groundwater standard process

DNR
Identify substances of concern

DHS
Develop recommended standards

DNR
Promulgate standards
Groundwater standard process

DNR
Identify substances of concern

DHS
Develop recommended standards

DNR
Promulgate standards
The enforcement standard is established from available health information.
Enforcement standards can be based on:

- Federal number
- State drinking water standard
- EPA value
- Technical information
- Cancer risk
Enforcement standards can be based on:

- Federal number

**Concentration** of a chemical in drinking water that is established by the EPA.

Maximum contaminant level (MCL)
Maximum contaminant level (MCL)

The highest level of a contaminant that is allowed in drinking water.
Maximum contaminant level (MCL) is the highest level of a contaminant that is allowed in drinking water.

Maximum contaminant level goal (MCLG)
The level of a contaminant in drinking water below which there is no known or expected risk to human health.
Maximum contaminant level (MCL) is set as close to

Maximum contaminant level goal (MCLG) as feasible.
MCLG = 0

Most carcinogens
MCLG = Reference dose × Body weight × Relative source contribution

All other substances
Enforcement standards can be based on:

- Federal number
- Concentration of a chemical in drinking water that is established by the EPA.
- Maximum contaminant level (MCL)
- Health advisory
Health advisory

Level at which health effects are not anticipated to occur over a specified duration
Health advisories

- 1 day
- 10 day
- Lifetime
Health advisory

Reference dose

Body weight

Relative source contribution

Water consumption
Enforcement standards can be based on:

- Federal number

Concentration of a chemical in drinking water that is established by the EPA.

- Maximum contaminant level (MCL)
- Health advisory
- Concentration based on cancer risk level
Concentration based on cancer risk level

=  

Risk level \times \frac{\text{Cancer slope factor}}{\text{Body weight}} \times \frac{\text{Water consumption}}{\text{Water consumption}}
Enforcement standards can be based on:

- **State drinking water standard**

**Concentration** of a chemical in drinking water that is established by the DNR.

Maximum contaminant level in Wis. Admin Code NR 809
Enforcement standards can be based on:

**EPA value**

- **Amount** of a chemical a person can be exposed to every day without health effects.
- Oral reference dose
- **Acceptable daily intake (ADI)**
Oral reference dose = Toxicity value / Uncertainty factor
Enforcement standards can be based on:

- Technical information

DHS can establish an ADI from available scientific information when:

- There is no federal number or EPA value.
- The information was not considered when the federal number/EPA value was established.
Acceptable daily intake (ADI) = Toxicity value / Uncertainty factor
Enforcement standards can be based on:

Cancer risk

DHS must ensure the standard does not allow for unacceptable cancer risk.

More than 1 case in 1,000,000 people
When an enforcement standard is based on:

- Federal number
- State drinking water standard

Use the concentration as the standard.
When an enforcement standard is based on:

- EPA value
- Technical information
- Cancer risk

Calculate the appropriate standard
Enforcement standards based on

- EPA value
- Technical information

Set to protect a young child
Enforcement Standard

Acceptable daily intake

Body weight

Relative source contribution

Water consumption
Enforcement Standard

Acceptable daily intake

Body weight

Relative source contribution

Water consumption

 Specified in Statute

Enforcement Standard =

Acceptable daily intake × Body weight × Relative source contribution

10 kg

1 L/d

100%
Enforcement standards based on Cancer risk

Set to protect from a lifetime of exposure
Enforcement Standard = Risk level × Body weight

Risk level = Cancer slope factor × Water consumption
Enforcement Standard

\[
\text{Risk level} \times \text{Body weight} = \text{Cancer slope factor} \times \text{Water consumption}
\]

Specified in Statute

Recommended by EPA
The preventive action limit is set at a percentage of the enforcement standard.
Preventive action limit = 10% of the enforcement standard

Substances that cause carcinogenic, mutagenic, teratogenic, or interactive effects
Preventive action limit = 20% of the enforcement standard

All other substances
PFOA
Perfluorooctanoic acid
Available scientific information for PFOA:

<table>
<thead>
<tr>
<th>Federal number</th>
</tr>
</thead>
<tbody>
<tr>
<td>State drinking water standard</td>
</tr>
<tr>
<td>EPA value</td>
</tr>
<tr>
<td>Technical information</td>
</tr>
<tr>
<td>Cancer risk</td>
</tr>
</tbody>
</table>
Available scientific information for PFOA:

Federal number

Lifetime health advisory

70 ng/L for PFOA and PFOS
Established in 2016
Available scientific information for PFOA:

Federal number

Concentration based on cancer risk

500 ng/L for PFOA
1 in 1,000,000 risk
Established in 2016
Available scientific information for PFOA:

Oral reference dose

20 ng/kg-d

Established for use in setting the lifetime health advisory
Available scientific information for PFOA:

Intermediate minimum risk level (MRL)

3 ng/kg-d

Proposed by ATSDR in 2018

Exposure duration of 15 – 365 days

ATSDR = Agency for Toxic Substances and Disease Registry
Available scientific information for PFOA:

Technical information

Critical studies
- Toxicity studies
- Modeling studies
Available scientific information for PFOA:

Cancer slope factor

0.07 (mg/kg·d)^{-1}

Established by EPA to set the concentration based on cancer risk
Available scientific information for PFOA:

- Federal number
- State drinking water standard
- EPA value
- Technical information
- Cancer risk
In 2016, EPA established a combined health advisory of 70 ng/L for PFOA and PFOS.
200+ studies evaluating PFOA toxicity in research animals

Most common endpoints:

- Liver: 54
- Body weight: 48
- Development: 31
- Immune system: 18
- Reproduction: 18
- Neurology: 11

Adapted from Figure 2-1 (ATSDR PFAS Tox Profile)
Liver
Body weight
Development
Immune
Reproduction
Neurology

LOAEL ranges from non-acute studies
Development and reproduction are the most sensitive effects.

LOAEL ranges from non-acute studies.
Babies are most sensitive to the effects of PFOA.
EPA based their advisory on a reproductive study in rats.

Study selected was Lau et al., 2006
Key findings

Summarized from Lau et al., 2006

Increased percent of animals with full litter resorption

mg/kg-d PFOA

Reproduction
Key findings

Reduced percent survival based on number of implantations per animal

Summarized from Lau et al., 2006
Key findings

Decreased number of ossification sites in forelimb proximal phalanges

Bone formation

Reproduction

Offspring survival

No births

mg/kg-d PFOA

Summarized from Lau et al., 2006
Key findings

0 1 3 5 10 20 40 mg/kg-d PFOA

Reproduction
Offspring survival
Bone formation
Sexual development
No births

Altered preputial separation

Summarized from Lau et al., 2006
Key findings

mg/kg-d PFOA

0 1 3 5 10 20 40

Reproduction

Offspring survival

Bone formation

Sexual development

Summarized from Lau et al., 2006
We do not know how much PFAS has to be in our blood to cause health effects.
PFOA stays in people longer than animals.

- Half-life of PFOA in males: 840 days
- Half-life of PFOA in animals: 21 days
- Half-life of PFOA in people: 4 days
PFOA level in pregnant woman’s blood = PFOA level in mother rat’s blood at the dose that caused the critical effect in offspring.
EPA’s approach

PFOA

Estimate

Convert

Calculate
Estimate how much PFOA was in animal’s blood at the dose that caused the critical effect.
EPA’s approach

Convert to human dose using half-life and amount of blood in the body

Estimate

Convert

Calculate
EPA’s approach

Calculate the health advisory
Health advisory

= Reference dose \times \text{Body weight} \times \text{Relative source contribution}
We have learned more about PFOA since 2016.
PFOA can cross the placenta during pregnancy.
PFOA can pass through breastmilk.
How can we best protect unborn and breastfed babies?
Kieskamp et al. approach

PFOA level in offspring’s blood that caused the critical effect

PFOA level in baby’s blood

PFOA?
Kieskamp et al. approach

Estimate

Convert
Kieskamp et al. approach

Estimate how much PFOA was in the blood of the offspring that had the critical effect.

Convert
Kieskamp et al. approach

Converted to dose that would cause a baby to have the same level as the offspring – taking into effect half-life and breastfeeding duration.
DHS calculations for PFOA:

Acceptable daily intake (ADI) = Toxicity value / Uncertainty factor
DHS calculations for PFOA:

Acceptable daily intake (ADI) =

Toxicity value

540 ng/kg-d

Uncertainty factor

Human equivalent dose for breastfeeding duration of 12 months and half-life of 2.3 years
DHS calculations for PFOA:

Acceptable daily intake (ADI) = Toxicity value / Uncertainty factor

Accounts for differences between species, differences among people, and using a LOAEL.
DHS calculations for PFOA:

Acceptable daily intake (ADI)

\[
\frac{540 \text{ ng/kg-d}}{300} = 2 \text{ ng/kg-d}
\]
Body weight

Acceptable daily intake

10 kg

Relative source contribution

2 ng/kg-d

1 L/d

Enforcement Standard

Specified in Statute
DHS recommendation for PFOA

Enforcement Standard

= 20 ng/L
DHS recommendation for PFOA

Preventive action limit = 10% of the enforcement standard

PFOA has been shown to cause carcinogenic, teratogenic, or interactive effects.
DHS recommends a combined enforcement standard of 20 ng/L for PFOA and PFOS.
PFOS
Perfluorooctane sulfonate
Available scientific information for PFOS:

- Federal number
- State drinking water standard
- EPA value
- Technical information
- Cancer risk
Available scientific information for PFOA:

**Lifetime health advisory**
70 ng/L for PFOA and PFOS
Established in 2016
Available scientific information for PFOS:

**EPA value**

- **Oral reference dose**
  - 20 ng/kg-d
  - Established for use in setting the lifetime health advisory

160.01(3)
Available scientific information for PFOS:

Technical information

Intermediate minimum risk level (MRL)

2 ng/kg-d

Proposed by ATSDR in 2018

Exposure duration of 15 – 365 days

ATSDR = Agency for Toxic Substances and Disease Registry
Available scientific information for PFOS:

- Federal number
- State drinking water standard
- EPA value
- Technical information
- Cancer risk
In 2016, EPA established a combined health advisory of 70 ng/L for PFOA and PFOS.
Babies are most sensitive to the effects of PFOS.
EPA based their advisory on a 2-generation study in rats.

Study selected was Luekber et al., 2005b
Key findings

Reduced body weight in males and females at various timepoints during exposure - corresponding with reduced food consumption

Summarized from Luebker et al., 2005b
Key findings

Increased in number of pups found dead and decreased viability and lactation indices.

Summarized from Luebker et al., 2005b
**Key findings**

Summarized from Luebker et al., 2005b

Decreased weight per litter and reduced weight change per litter
Key findings

- Decreased weight per litter and reduced weight change per litter

Summarized from Luebker et al., 2005b
Key findings

Summarized from Luebker et al., 2005b

mg/kg-d PFOS

0 0.1 0.4 1.6 3.2

Body weight (F0)
Survival (F1)
Body weight (F1)
Body weight (F2)
EPA used the same modeling approach for PFOA and PFOS.
We have learned more PFOS since 2016.
PFOS can cross the placenta during pregnancy.
PFOS can pass through breastmilk.
PFOS may increase the risk for asthma, food allergies, and certain infectious diseases.
In 2018, ATSDR proposed a minimum risk level of 2 ng/kg-d for PFOS.

ATSDR = Agency for Toxic Substances and Disease Registry
ATSDR’s calculation for PFOS:

Minimum risk level = Toxicty value

= Uncertainty factor × Modifying factor
ATSDR’s calculation for PFOS:

Minimum risk level = Toxicity value × Modifying factor × Uncertainty factor

510 ng/kg-d

Human equivalent dose for pregnant women
ATSDR’s calculation for PFOS:

Minimum risk level = 30

Account for differences between species and differences among people

Uncertainty factor = 30

Toxicity value
ATSDR’s calculation for PFOS:

Minimum risk level

\[ \text{Toxicity value} = \frac{\text{Uncertainty factor} \times 10}{\text{Modifying factor}} \]

Account for potential for immune effects at low levels
ATSDR’s calculation for PFOS:

\[
\frac{510 \text{ ng/kg-d}}{30} \times \frac{10}{2} = 2 \text{ ng/kg-d}
\]
DHS recommends using ATSDR’s minimum risk level for PFOS.

This approach protects from potential immune effects and infant exposure.
Enforcement Standard

Acceptable daily intake: 2 ng/kg-d

Body weight: 10 kg

Relative source contribution: 100%

Water consumption: 1 L/d

Specified in Statute
DHS’ recommendation for PFOS

Enforcement Standard

= 20 ng/L
DHS’ recommendation for PFOS

PFOA has been shown to cause carcinogenic, teratogenic, or interactive effects

Preventive action limit = 10%
DHS recommends a combined enforcement standard of 20 ng/L for PFOA and PFOS.
Thanks!

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Wisconsin Department of Health Services

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Additional information can be found on DHS’ webpage: 
[dhs.wisconsin.gov\water\gws.htm](dhs.wisconsin.gov\water\gws.htm)

The full scientific support document for all of the Cycle 10 compounds is available here: 
[dhs.wisconsin.gov\publications\p02434v.pdf](dhs.wisconsin.gov\publications\p02434v.pdf).
Additional information
LOAELs from non-acute studies

Liver

Body weight

Development

Immune

Reproduction

Neurology

Data from Table 2-3 in ATSDR’s Toxicological Profile for Perfluoroalkyls
Lau et al., 2006 results (part 1)

<table>
<thead>
<tr>
<th>Effects observed in mothers</th>
<th>Dose (mg/kg-d)</th>
<th>1</th>
<th>3</th>
<th>5</th>
<th>10</th>
<th>20</th>
<th>40</th>
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<tr>
<td>Body weight</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reduced maternal weight gain</td>
<td></td>
<td>✓</td>
<td>✓</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Increased percent of dams with full litter resorption</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td>Reproduction</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reduced number of live fetuses</td>
<td></td>
<td>✓</td>
<td></td>
<td></td>
<td></td>
<td>N/A</td>
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<tr>
<td>Increased percent of prenatal loss</td>
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<td>✓</td>
<td></td>
<td></td>
<td></td>
<td>N/A</td>
<td></td>
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<tr>
<td>Increased time to parturition</td>
<td></td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
<td>N/A</td>
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</table>
### Lau et al., 2006 results (part 2)

<table>
<thead>
<tr>
<th>Effects observed in offspring</th>
<th>Dose (mg/kg-d)</th>
<th>1</th>
<th>3</th>
<th>5</th>
<th>10</th>
<th>20</th>
</tr>
</thead>
<tbody>
<tr>
<td>Survival</td>
<td>Reduced neonatal survival</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
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<tr>
<td>Body weight</td>
<td>Reduced fetal body weight</td>
<td></td>
<td></td>
<td></td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td>Bone development</td>
<td>Decreased number of ossification sites in sternebrae, caudal vertebrae, metacarpals, metatarsals</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Decreased number of ossification sites in forelimb proximal phalanges</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Decreased number of ossification sites in hindlimb proximal phalanges</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Reduced percent ossification in calvaria</td>
<td>✓</td>
<td></td>
<td></td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Reduced percent ossification in supraoccipital</td>
<td></td>
<td></td>
<td>✓</td>
<td>✓</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Reduced percent ossification in unossified hybrid</td>
<td>✓</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Increased number of enlarged fontanel</td>
<td>✓</td>
<td>✓</td>
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</tr>
<tr>
<td>Category</td>
<td>Examples</td>
<td>Number of Studies</td>
<td></td>
<td></td>
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<td>--------------------------------------------------------------------------</td>
<td>-------------------</td>
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<tr>
<td>Metabolic</td>
<td>Diabetes (type 1, 2, and gestational), glucose tolerance, insulin resistance, BMI, obesity/overweight, adiposity, cholesterol, triglycerides</td>
<td>41</td>
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<td></td>
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<tr>
<td>Birth outcomes</td>
<td>Birth size (weight, length, etc), gestation age, small for gestational age, fetal growth, anogenital distance at birth</td>
<td>25</td>
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<td></td>
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<tr>
<td>Neurological</td>
<td>Attention, impulse control, visual and spatial ability, cognitive development, executive function, autism spectrum disorder, intellectual disability</td>
<td>18</td>
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<td></td>
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<tr>
<td>Reproductive</td>
<td>Endometriosis, preeclampsia, reproductive hormones, time to pregnancy, fertility, semen characteristics, pregnancy loss, menopause, puberty onset</td>
<td>13</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Immune</td>
<td>Asthma, vaccine antibodies, allergic conditions, infectious disease incidence, atopic dermatitis</td>
<td>12</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Thyroid</td>
<td>Thyroid hormones, thyroid function</td>
<td>10</td>
<td></td>
<td></td>
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<tr>
<td>Cardiovascular</td>
<td>Heart attack, stroke, heart failure, arterial wall stiffness, coronary heart disease, blood pressure, hypertension</td>
<td>7</td>
<td></td>
<td></td>
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<tr>
<td>Kidney</td>
<td>Chronic kidney disease, kidney function, glomerular filtration</td>
<td>7</td>
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<tr>
<td>Other</td>
<td>Vitamin D, bone density, lung function, dental carries, gut bacteria and metabolites, mortality</td>
<td>6</td>
<td></td>
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<tr>
<td>DNA</td>
<td>Telomere length, DNA methylation</td>
<td>5</td>
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<tr>
<td>Liver</td>
<td>ALT (alanine aminotransferase), other liver function biomarkers</td>
<td>4</td>
<td></td>
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<tr>
<td>Cancer</td>
<td>Breast cancer</td>
<td>2</td>
<td></td>
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</tr>
</tbody>
</table>
### Lau et al., 2006 results (part 3)

<table>
<thead>
<tr>
<th>Effects observed in offspring</th>
<th>Dose (mg/kg-d)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>Birth defects</td>
<td></td>
</tr>
<tr>
<td>Increased percent of tail defects</td>
<td>✓</td>
</tr>
<tr>
<td>Increased percent of limb defects</td>
<td>✓</td>
</tr>
<tr>
<td>Heart</td>
<td></td>
</tr>
<tr>
<td>Increased percent of microcardia</td>
<td></td>
</tr>
<tr>
<td>Development</td>
<td></td>
</tr>
<tr>
<td>Delayed eye opening</td>
<td></td>
</tr>
<tr>
<td>Delayed vaginal opening</td>
<td>✓</td>
</tr>
<tr>
<td>Delayed first estrus</td>
<td>✓</td>
</tr>
<tr>
<td>Altered preputial separation</td>
<td>✓</td>
</tr>
</tbody>
</table>
### Leubker et al., 2005 results (part 1)

<table>
<thead>
<tr>
<th>Effects observed in F0 generation (males)</th>
<th>Dose (mg/kg-d)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.1</td>
</tr>
<tr>
<td>Body weight</td>
<td></td>
</tr>
<tr>
<td>Reduced body weight</td>
<td>✓ a</td>
</tr>
<tr>
<td>Food consumption</td>
<td></td>
</tr>
<tr>
<td>Reduced food consumption days 1-42</td>
<td>✓</td>
</tr>
<tr>
<td>Reduced food consumption days 56-63</td>
<td>✓</td>
</tr>
</tbody>
</table>

a. Days 56 through termination
b. Days 36 through termination
Leubker et al., 2005 results (part 2)

<table>
<thead>
<tr>
<th>Effects observed in F0 generation (females)</th>
<th>Dose (mg/kg-d)</th>
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<tbody>
<tr>
<td></td>
<td>0.1</td>
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<tr>
<td>Body weight</td>
<td></td>
</tr>
<tr>
<td>Reduced body weight during precohabitation</td>
<td>✓c</td>
</tr>
<tr>
<td>Reduced body weight during gestation</td>
<td></td>
</tr>
<tr>
<td>Reduced body weight during lactation</td>
<td></td>
</tr>
<tr>
<td>Food consumption</td>
<td></td>
</tr>
<tr>
<td>Reduced food consumption during premating and gestation</td>
<td>✓</td>
</tr>
<tr>
<td>Reduced food consumption during lactation</td>
<td></td>
</tr>
<tr>
<td>Reproduction</td>
<td></td>
</tr>
<tr>
<td>Reduced gestation duration</td>
<td>✓</td>
</tr>
<tr>
<td>Decreased implantation sites per delivered litter</td>
<td>✓</td>
</tr>
<tr>
<td>Increased percent of animals with stillborn pups</td>
<td>✓</td>
</tr>
<tr>
<td>Increased percent of animals with all pups dying (PND 1-4)</td>
<td>✓</td>
</tr>
</tbody>
</table>

c. Days 15-42
d. Gestation days 3-10
e. Gestation days 0-20; Lactation day 7
f. Lactation day 1; no results for days 4-21
### Leubker et al., 2005 results (part 3)

<table>
<thead>
<tr>
<th>Effects observed in F1 generation</th>
<th>Dose (mg/kg-d)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.1</td>
</tr>
<tr>
<td><strong>Survival</strong></td>
<td></td>
</tr>
<tr>
<td>Decreased liveborn</td>
<td>✓</td>
</tr>
<tr>
<td>Increased stillborn per litter</td>
<td></td>
</tr>
<tr>
<td>Increased percent of pups found dead</td>
<td>✓</td>
</tr>
<tr>
<td>Reduced viability index</td>
<td>✓</td>
</tr>
<tr>
<td>Reduced lactation index</td>
<td>✓</td>
</tr>
<tr>
<td><strong>Body weight</strong></td>
<td></td>
</tr>
<tr>
<td>Decreased weight per litter</td>
<td>✓</td>
</tr>
<tr>
<td>Reduced weight change per litter</td>
<td></td>
</tr>
</tbody>
</table>

- **g.** Postnatal days 2-4 and 5-7
- **h.** Postnatal day 1 and 2-4; not results for days 5-21
- **i.** Postnatal days 1-21
- **j.** Postnatal day 1; no results for days 2-21
- **k.** Postnatal days 1-4; 4-7; 7-14; 14-21
Leubker et al., 2005 results (part 3)

<table>
<thead>
<tr>
<th>Effects observed in F2 generation</th>
<th>Dose (mg/kg-d)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.1</td>
</tr>
<tr>
<td>Body weight</td>
<td></td>
</tr>
<tr>
<td>Reduced weight per litter</td>
<td></td>
</tr>
<tr>
<td>Reduced weight change per litter</td>
<td>✓l</td>
</tr>
</tbody>
</table>

l. Postnatal days 7 and 14
m. Postnatal days 4-7 and 7-14