US EPA ARCHIVE DOCUMENT
Data Evaluation Report on the acute toxicity of Penoxsulam metabolite to aquatic vascular plants *Lehna gibba*

PMRA Submission #: 1

Data Requirement:

- PMRA Data Code: 
- EPA DP Barcode: D288160
- OECD Data Point: 
- EPA MRID: 45831109
- EPA Guideline: 122-2

Test material: Penoxsulam

Purity: 99% (TEA salt) and 72% (Acid)

Common name: XDE-638 metabolite TPSA

Chemical name: IUPAC: 5,8-dimethoxy[1,2,4]triazolo[1,5-C]pyrimidin-2-yl) sulfamic acid

CAS name: Not reported

CAS No.: Not reported

Synonyms: Not reported

Primary Reviewer: Rebecca Bryan
Staff Scientist, Dynamac Corporation

Signature: 
Date: 11/21/03

QC Reviewer: Dana Worcester
Staff Scientist, Dynamac Corporation

Signature: 
Date: 11/21/03

Primary Reviewer: Bill Erickson

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[Date: 11/21/03]

Secondary Reviewer(s): 

[Primary Reviewer: Bill Erickson]

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Company Code: [For PMRA]
Active Code: [For PMRA]
EPA PC Code: 119031

Date Evaluation Completed: 
dd-mmm-yyyy

EXECUTIVE SUMMARY:
Acute toxicity of Penoxsulam metabolite to aquatic vascular plants- *Lemna gibba* MRID 45831109

In a 14-day acute toxicity study, freshwater aquatic vascular plants Duckweed, *Lemna gibba* G3, were exposed to Penoxsulam at a single, nominal concentration of 1.4 mg a.i./L under static conditions. The mean frond numbers, dry weights, areas under the growth curve, and growth rates were not affected in the 1.4 mg a.i./L treatment group compared to the control. The NOAEC was 1.4 mg/L. The EC$_{05}$ was not determined. The frond number EC$_{50}$ was >1.4 mg a.i./L. This toxicity study is scientifically sound, but it does not satisfy the U.S. EPA Guideline Subdivision J, §122-2 because the single nominal test concentration, which greatly exceeded the expected environmental concentration (14 µg/L), was not analytically determined. As a result, this study is classified as SUPPLEMENTAL, but it need not be repeated.

Results Synopsis

Test Organism: *Lemna gibba* G3  
Test Type: Static

**Number of fronds:**  
NOAEC: 1.4 mg/L.  
EC$_{05}$: ND  
EC$_{50}$/IC$_{50}$: >1.4 mg/L  
95% C.I.: N/A

**Growth rates:**  
NOAEC: 1.4 mg/L.  
EC$_{05}$: ND  
EC$_{50}$/IC$_{50}$: >1.4 mg/L  
95% C.I.: N/A

**Plant biomass (area under the growth curve):**  
NOAEC: 1.4 mg/L.  
EC$_{05}$: ND  
EC$_{50}$/IC$_{50}$: >1.4 mg/L  
95% C.I.: N/A

**Dry Weights:**  
NOAEC: 1.4 mg/L.  
EC$_{05}$: ND  
EC$_{50}$/IC$_{50}$: >1.4 mg/L  
95% C.I.: N/A

Endpoint(s) Affected: None
I. MATERIALS AND METHODS


1. The study was conducted with one test concentration (limit test), which was not analytically determined. As a result, it does not satisfy the Tier I §122-2 guidelines.

2. The pretest health of the test organism was not reported.

3. The dilution water characteristics were not reported.

4. The number of plants (3) was less than recommended, so the number of fronds (12) was less than the recommended 15 fronds per replicate.

5. The stability of the test substance was not determined.

These deviations were not considered to have affected the results of the study, but the failure to analyze the single test concentration affected the acceptability of the study.

COMPLIANCE: Signed and dated GLP, Quality Assurance and No Data Confidentiality statements were provided. The test was conducted according to the OECD Principles of Good Laboratory Practice (GLP, 1998).

A. MATERIALS:

1. Test Material

   Penoxsulam metabolite (TPSA)

   Description: White-beige powder

   Lot No./Batch No.: F-380-205a

   Purity: 99% (TEA salt) and 72% (Acid)

   Stability of Compound

   Under Test Conditions: The test concentration was not measured during the study, therefore, stability was not determined. OECD requirements were not reported.

   (OECD requires water solubility, stability in water and light, pKa, Pow, vapor pressure of test compound)

   Storage conditions of test chemicals: Stored in a freezer.

2. Test organism:
Name: Duckweed, *Lemna gibba* L.  
*(EPA requires a vascular species: *Lemna gibba*.)

Strain, if provided: G3 1913

Source: Laboratory cultures (original supplier: UTCC #310)

Age of inoculum: 10-14 days old (p. 16)

Method of cultivation: 20X AAP Medium

**B. STUDY DESIGN:**

a) Range-finding Study: No range-finding study was conducted.

b) Definitive Study

**Table 1. Experimental Parameters**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Details</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acclimation period:</td>
<td>At least 3 weeks</td>
<td>EPA expects the test concentrations to be renewed every 3 to 4 days</td>
</tr>
<tr>
<td>culturing media and conditions: (same as test or not)</td>
<td>20X AAP Medium; same as test.</td>
<td>(one renewal for the 7 day test, 3-4 renewals for the 14 day test).</td>
</tr>
<tr>
<td>health: (any toxicity observed)</td>
<td>Not reported</td>
<td></td>
</tr>
<tr>
<td>Test system static/static renewal/renewal rate for static renewal:</td>
<td>Static</td>
<td>EPA requires a duration of 14 days. Seven day studies will be accepted for review by the Agency.</td>
</tr>
<tr>
<td>Incubation facility</td>
<td>Growth chamber</td>
<td></td>
</tr>
<tr>
<td>Duration of the test</td>
<td>14 days</td>
<td>Test vessels covered with Jaece® non-toxic foam plugs.</td>
</tr>
<tr>
<td>Test vessel material: (glass/polystyrene)</td>
<td>Glass Erlenmeyer flasks</td>
<td></td>
</tr>
<tr>
<td>size; fill volume:</td>
<td>500 mL, 200 mL</td>
<td></td>
</tr>
<tr>
<td>Details of growth medium name:</td>
<td>Modified 20X AAP Medium</td>
<td>EPA recommend the following culture media: Modified hoagland's E+ or 20X-AAP.</td>
</tr>
<tr>
<td>pH at test initiation:</td>
<td>8.16-8.31, 9.43-9.85</td>
<td></td>
</tr>
<tr>
<td>pH at test termination:</td>
<td>Not reported, Not reported</td>
<td></td>
</tr>
<tr>
<td>Parameter</td>
<td>Details</td>
<td>Remarks</td>
</tr>
<tr>
<td>--------------------------------------------------------------------------</td>
<td>-------------------------------------------------------------------------</td>
<td>-------------------------------------------------------------------------</td>
</tr>
<tr>
<td>If non-standard nutrient medium was used, detailed composition provided (Yes/No)</td>
<td>Not applicable</td>
<td>The dilution water characteristics were not reported.</td>
</tr>
<tr>
<td>Dilution water source/type:</td>
<td>20X AAP Medium</td>
<td>EPA recommends a pH of ~5.0. A solution pH of 7.5 is acceptable if type 20X-AAP nutrient media is used.</td>
</tr>
<tr>
<td>pH:</td>
<td>7.5 ± 0.1</td>
<td></td>
</tr>
<tr>
<td>water pretreatment (if any):</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>Total Organic Carbon:</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>particulate matter:</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>metals:</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>pesticides:</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>chlorine:</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>Indicate how the test material is added to the medium (added directly or used stock solution)</td>
<td>Stock solution</td>
<td></td>
</tr>
<tr>
<td>Aeration or agitation</td>
<td>Not reported.</td>
<td></td>
</tr>
<tr>
<td>Sediment used (for rooted aquatic vascular plants)</td>
<td>Not applicable</td>
<td></td>
</tr>
<tr>
<td>origin:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>textural classification (% sand, silt and clay):</td>
<td></td>
<td></td>
</tr>
<tr>
<td>organic carbon (%):</td>
<td></td>
<td></td>
</tr>
<tr>
<td>geographic location:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of replicates control:</td>
<td>6</td>
<td>EPA requires 5 plants.</td>
</tr>
<tr>
<td>solvent control:</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>treatments:</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Number of plants/replicate</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>Number of fronds/plant</td>
<td>4 fronds per plant (12 total fronds per replicate)</td>
<td>EPA requires 3 fronds per plant.</td>
</tr>
<tr>
<td>Test concentrations nominal:</td>
<td>0 (negative control) and 1.4 mg/L.</td>
<td>The measured test concentration was not determined.</td>
</tr>
<tr>
<td>measured:</td>
<td>Not determined</td>
<td>EPA requires at least 5 test concentrations with a dose range of 2X or 3X progression.</td>
</tr>
<tr>
<td>Solvent (type, percentage, if used)</td>
<td>N/A</td>
<td></td>
</tr>
<tr>
<td>Method and interval of analytical</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
2. Observations:

Table 2: Observation parameters

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Details</th>
<th>Remarks/Criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parameters measured (eg: number of fronds, plant dry weight or other toxicity symptoms)</td>
<td>Number of fronds (number of colonies), dry weights, and toxicity symptoms.</td>
<td></td>
</tr>
<tr>
<td>Measurement technique for frond number and other end points</td>
<td>Direct counts using a hand-magnifying lens.</td>
<td></td>
</tr>
<tr>
<td>Observation intervals</td>
<td>0, 3, 5, 7, 10, and 14 days.</td>
<td>3 replicates harvested on day 7 and 3 replicates harvested on day 14.</td>
</tr>
<tr>
<td>Other observations, if any</td>
<td>Area under the growth curve and growth rates.</td>
<td></td>
</tr>
<tr>
<td>Indicate whether there was an exponential growth in the control</td>
<td>Yes</td>
<td></td>
</tr>
<tr>
<td>Were raw data included?</td>
<td>Replicate data provided.</td>
<td></td>
</tr>
</tbody>
</table>

II. RESULTS and DISCUSSION:

A. INHIBITORY EFFECTS:

The mean frond numbers, dry weights, areas under the growth curve, and growth rates were not affected in the 1.4 mg/L treatment group compared to the control.
Acute toxicity of Penoxsulam metabolite to aquatic vascular plants - *Lemna gibba*  MRID 45831109

**Table 3: Effect of Penoxsulam metabolite on frond number and dry weight of Duckweed, *Lemna gibba***

<table>
<thead>
<tr>
<th>Treatment nominal concentration, mg/L</th>
<th>Initial frond number/test solution</th>
<th>Mean frond number at 3 days</th>
<th>Mean frond number at 7 days</th>
<th>Mean frond number at 14 days</th>
<th>% inhibition at 14 days</th>
<th>Mean dry weight (mg)</th>
<th>Mean Growth Rate</th>
<th>Mean Area Under the Growth Curve</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negative control (dilution water)</td>
<td>12</td>
<td>30.8</td>
<td>152.3</td>
<td>700.7</td>
<td>---</td>
<td>65.14</td>
<td>0.01214</td>
<td>66,688.54</td>
</tr>
<tr>
<td>1.4</td>
<td>12</td>
<td>34.8</td>
<td>164.0</td>
<td>730.3</td>
<td>-4.2</td>
<td>66.64</td>
<td>0.01226</td>
<td>72,911.58</td>
</tr>
<tr>
<td>Reference chemical (if used)</td>
<td>Not applicable</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Negative percent inhibition represents positive growth.
NR-Not reported

**Table 4: Statistical endpoint values.**

<table>
<thead>
<tr>
<th>Statistical Endpoint*</th>
<th>frond No.</th>
<th>Dry weight</th>
<th>growth rate</th>
<th>area under the growth curve</th>
</tr>
</thead>
<tbody>
<tr>
<td>NOAEC(mg/L)</td>
<td>1.4</td>
<td>1.4</td>
<td>1.4</td>
<td>1.4</td>
</tr>
<tr>
<td>EC$_{05}$</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>LOAEC (mg/L)</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
</tr>
<tr>
<td>EC$_{50}$ (mg/L) (95% C.I.)</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
</tr>
<tr>
<td>EC$_{25}$ (mg/L) (95% C.I.)</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
<td>&gt;1.4</td>
</tr>
<tr>
<td>Reference chemical NOAEC</td>
<td>Not applicable</td>
<td>Not applicable</td>
<td>Not applicable</td>
<td>Not reported</td>
</tr>
<tr>
<td>IC$<em>{50}$/EC$</em>{50}$</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Statistical data based on nominal test concentrations.

**B. REPORTED STATISTICS:** Percent inhibition was determined for all endpoints. The 14 day NOAEC, and EC$_{50}$ values were estimated using the significance data for all endpoints. All statistical calculations were performed using the nominal concentration.

**C. VERIFICATION OF STATISTICAL RESULTS:**

Statistical analyses were not required to verify the results of this study, as there were no reductions in the treatment group for any endpoint.

**Number of fronds:**
NOAEC: 1.4 mg/L.
EC$_{05}$: ND
Acute toxicity of Penoxsulam metabolite to aquatic vascular plants - *Lemna gibba*  

**EC₅₀/IC₅₀:** >1.4 mg/L  
95% C.I.: N/A

**Growth rates:**  
NOAEC: 1.4 mg/L.  
EC₅₀: ND  
EC₅₀/IC₅₀: >1.4 mg/L  
95% C.I.: N/A

**Plant biomass (area under the growth curve):**  
NOAEC: 1.4 mg/L.  
EC₅₀: ND  
EC₅₀/IC₅₀: >1.4 mg/L  
95% C.I.: N/A

**Dry Weights:**  
NOAEC: 1.4 mg/L.  
EC₅₀: ND  
EC₅₀/IC₅₀: >1.4 mg/L  
95% C.I.: N/A

**Endpoint(s) Affected:** None

**D. STUDY DEFICIENCIES:**

While no toxicity was shown in this study and the nominal concentration greatly exceeded the expected environmental concentration (14 μg/L), this test level was not analytically determined. As a result, this study does not satisfy the U.S. EPA Guideline Subdivision J, §122-2 for a limit test with a metabolite.

**E. REVIEWER’S COMMENTS:**

The reviewer’s conclusions agreed with the study author’s; there was no toxicity of XDE-638 Metabolite TPSA (TSN 103594) to duckweed. While no toxicity was detected in this study and a concentration which greatly exceeded the expected environmental concentration (14 μg/L) was tested, this study cannot be classified as Core because the test concentration was not analytically verified.

The projected environmental concentration (PEC) for TPSA is 14 μg/L.

The doubling time of the control was used as validity criteria (OECD). An increase of approximately 8-fold in seven days is required.

The colony number percent inhibition was -9.1 in the 1.4 mg/L treatment group after 14 days (no effect).

**F. CONCLUSIONS:** This toxicity study is scientifically sound, but it does not satisfy the U.S. EPA Guideline Subdivision J, §122-2 because the single nominal test concentration, which greatly exceeded the expected environmental concentration (14 μg/L), was not analytically determined. As a result, this study is classified as SUPPLEMENTAL, but it need not be repeated.

**Number of fronds:**  
NOAEC: 1.4 mg/L.  
EC₅₀: ND  
EC₅₀/IC₅₀: >1.4 mg/L  
95% C.I.: N/A
Growth rates:
NOAEC: 1.4 mg/L.
EC_{90}: ND
EC_{50}/IC_{50} > 1.4 mg/L 95% C.I.: N/A

Plant biomass (area under the growth curve):
NOAEC: 1.4 mg/L.
EC_{90}: ND
EC_{50}/IC_{50} > 1.4 mg/L 95% C.I.: N/A

Dry Weights:
NOAEC: 1.4 mg/L.
EC_{90}: ND
EC_{50}/IC_{50} > 1.4 mg/L 95% C.I.: N/A

Endpoint(s) Affected: None

III. REFERENCES:


