Elucidating mechanisms of toxic action of dissolved organic chemicals in oil sands process-affected water (OSPW)

Garrett Morandi, 1,2 Steve Wiseman, 3,4 Guan Miao, 5,6 Xiaowei Zhang, 4,7 Jonathan W. Martin and 1,3,6,7 John P. Giesy 1Toxicology Centre, University of Saskatchewan, Saskatoon, Canada. 2Department of Biological Sciences and Water Institute for Sustainable Environments, University of Lethbridge, Lethbridge, Canada. 3State Key Laboratory of Pollution Control and Resource Reuse, School of the Environment, Nanjing University, Nanjing, China. 4Division of Analytical and Environmental Toxicology, University of Alberta, Edmonton, Canada. 5Department of Environmental Sciences and Analytical Chemistry, Stockholm University, Stockholm, Sweden. 6Department of Veterinary Biomedical Sciences, University of Saskatchewan, Saskatoon, Canada. 7Zoology Department, Center for Integrative Toxicology, Michigan State University, East Lansing, USA.

INTRODUCTION

Oil Sands Process Affected Waters (OSPW)
OSPW is a by-product of the extraction and separation of bitumen in the Alberta Oil Sands,(1) is restricted to a zero-discharge policy and requires remediation before release.(2,3)
OSPW has acute and chronic toxicity to a range of species.(4,5)
Dissolved organic fraction is responsible for most toxicity.
Previous work has identified acutely toxic extracts of dissolved organic chemicals in OSPW.
Lack of information as to mechanisms of toxicity of the dissolved organic fraction of OSPW.

Escherichia coli K-12 MG-16553 gene reporter system
The Live Cell Array (LCA) is an open-format approach for investigating the adverse effects of chemicals.(10)
System facilitates the measurement of promoter activity (1820 genes) using transcriptionally fused fast-folding fluorescent proteins.(10,11)

PURPOSE/ METHODS

1. Utilize previously produced extracts of OSPW (Morandi et al., 2015) to identify molecular mechanisms of toxicity by use of the LCA system.
   i. 5 acutely toxic, 1 ‘non-toxic’ extract.
   ii. 1820 genes monitored over 4 hours.
2. Identify differentially expressed genes (>1.5-fold).
4. Perform Gene Ontology and KEGG pathway analysis by use of Cytoscape (3.2).
5. Principle component analysis (PCA) to identify relationships among gene expression, pathways of effect, uHMRs data and toxicity.

RESULTS

Table 1. Numbers of genes differentially expressed ≥1.5- and 2-fold in E.coli exposed to extracts of OSPW relative to E.coli exposed to a solvent control.

<table>
<thead>
<tr>
<th>Sample</th>
<th>1.5-fold</th>
<th>Total</th>
<th>2-fold</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1-Pool</td>
<td>2</td>
<td>121</td>
<td>123</td>
<td>27</td>
</tr>
<tr>
<td>F1-NE</td>
<td>20</td>
<td>98</td>
<td>118</td>
<td>23</td>
</tr>
<tr>
<td>F2-NE1</td>
<td>14</td>
<td>78</td>
<td>92</td>
<td>29</td>
</tr>
<tr>
<td>F2-NE2a</td>
<td>32</td>
<td>88</td>
<td>120</td>
<td>45</td>
</tr>
<tr>
<td>F2-NE2b</td>
<td>6</td>
<td>121</td>
<td>121</td>
<td>37</td>
</tr>
</tbody>
</table>

Table 2. Genes differentially expressed in E. coli exposed to each of the acutely toxic fractions of OSPW.

<table>
<thead>
<tr>
<th>Gene</th>
<th>Description</th>
<th>Biological function</th>
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<tbody>
<tr>
<td>yceP</td>
<td>Transcription repressor</td>
<td>Biofilm formation</td>
</tr>
<tr>
<td>chpA</td>
<td>Transcription repressor</td>
<td>Chaperone protein</td>
</tr>
<tr>
<td>ddAB</td>
<td>Enzyme</td>
<td>Outer-membrane phospholipase</td>
</tr>
<tr>
<td>adlB</td>
<td>Enzyme</td>
<td>Alcohol dehydrogenase</td>
</tr>
<tr>
<td>NOP</td>
<td>Putative surface protein</td>
<td>L-lysine transmembrane transporter</td>
</tr>
</tbody>
</table>

Figure 1. Total ion count for chemical classes detected in extracts of OSPW in ESI+/− (L/R).

Figure 2. Percent inhibition of growth (%) of E. coli cells exposed to extracts of OSPW.

Figure 3. Venn diagram comparing genes expressed in E. coli exposed to acutely toxic fractions of extracts of OSPW.

Figure 4. Individual factor map of extracts of OSPW, clustered by use of identified BP.

CONCLUSIONS

• Six genes were uniquely responsive to acutely toxic extracts of OSPW.
• Differentially expressed genes (Table 2) were indicative of general stress, protein damage and DNA damage.
• Acutely toxic fractions demonstrated a general down-regulation of catabolic and anabolic processes.
  • Indicative of general non-specific toxicity.
• GO analysis identified BP KEGG pathways related to changes to the redox state of the cell, response to oxidative stress and are consistent with previous results across a range of extracts and species.
• Sulphur and nitrogen containing chemical classes contribute to toxic potencies of extracts of OSPW.

REFERENCES


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