Dioxins in the Sediments of Sydney Harbour: Potential for Bioremediation

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Project Team and Funding

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**NSW Government**

Environmental Trust grant programs  
This project has been assisted by the New South Wales Government through its Environmental Trust.
Presentation Overview

Site History

Results
1. Concentration and composition of dioxins in sediments
2. Bioinformatic analysis
3. Testing for reductive dechlorination of dioxins

Summary
Site History: Homebush Bay and Rhodes

Table. Chemical products from Union Carbide (Birch et al., 2007)

<table>
<thead>
<tr>
<th>Years</th>
<th>Chemical Products</th>
</tr>
</thead>
<tbody>
<tr>
<td>1928-1986</td>
<td>Timber products (includes <strong>Pentachlorophenol</strong>)</td>
</tr>
<tr>
<td>1932-1985</td>
<td>Xanthates</td>
</tr>
<tr>
<td>1940-1961</td>
<td>Aniline and nireobenzene</td>
</tr>
<tr>
<td>1942-1971</td>
<td>Phenol</td>
</tr>
<tr>
<td>1947-1976</td>
<td>Chlorophenol and chlorobenzenes</td>
</tr>
<tr>
<td>1949-1976</td>
<td>2,4,5-T and 2,4-D</td>
</tr>
<tr>
<td>1952-1976</td>
<td>Chlorine gas</td>
</tr>
<tr>
<td>1955-1968</td>
<td>DDT and DDD</td>
</tr>
<tr>
<td>1960-1976</td>
<td>DPP</td>
</tr>
<tr>
<td>1964-1976</td>
<td>Phenol-formaldehyd resines and moulding component</td>
</tr>
</tbody>
</table>
Site History: Homebush Bay, Sydney

• Pentachlorophenol (PCP)
  • widely utilized in timber preservative and in pesticides,
  • typically contaminated with highly chlorinated dioxins congeners: Octa-chlorodibenzo-p-dioxin or -furans

\[
\begin{align*}
\text{PCP} & \rightarrow \text{OCDD} \\
\text{PCP} & \rightarrow \text{OCDF}
\end{align*}
\]

• Agent Orange
  • a mixture of 2,4-dichlorophenoxyacetic acid (2,4-D) and 2,4,5-trichlorophenoxyacetic acid (2,4,5-T)

\[
\begin{align*}
2,4,5\text{-trichlorophenol} & + \text{2,4-D} \rightarrow 2,3,7,8\text{-tetrachlorodibenzo-p-dioxin} + 2\text{HCl} \\
& \text{(2,3,7,8-TCDD)}
\end{align*}
\]
Sampling locations in Sydney Harbour

At each location: 50 cm sediment core samples were taken and then divided into 25 cm upper and lower fractions.

Each sample:

1. quantify the concentration and composition of dioxins;

2. DNA extraction, sequencing and bioinformatic analysis; and

3. inoculation into laboratory microcosms, incubation and monitoring for reductive dechlorination of dioxins.
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Summary
Concentration and composition of dioxins in sediments

Upper layer:
- OCDD: 0.0003
- TCDD: 1.0

Lower layer:
- OCDD
- 2,3,7,8-TCDD

Dioxin congener | TEF (WHO 2005)
---|---
OCDD | 0.0003
TCDD | 1.0
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1. Concentration and composition of dioxins in sediments
2. **Bioinformatic analysis**
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Summary
Microbial community in sediment

High cell numbers
• contained an average of $5.6 \times 10^8$ total bacterial cells per gram of wet weight sediment (cells/gram) across the 20 samples.

Microbial diversity
• high level of microbial diversity spread across approximately 500 bacterial genera.
Bacteria of the genera *Dehalococcoides*, *Dehalogenimonas* and *Dehalobacter* are capable of respiring organohalide compounds.

Quantitative PCR of sediment cores for 16S rRNA gene

*Dehalococcoides* genus

- Average $7 \pm 2 \times 10^6$ cells/gram
- $\sim 1\%$ of total microbial population
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Summary
Anaerobic Enrichment Cultures

1) Enrichment for organohalide respiring bacteria (ORBs)
2) Subculture enriched cultures of ORBs - dioxins
Anaerobic Enrichment: Chloroethenes

Sediment microbial community capable of reductive dechlorination of PCE
Anaerobic Enrichment: Chlorobenzenes

Sediment microbial community capable of reductive dechlorination of TCBs
Anaerobic Biotransformation: Dioxins

Enrichment cultures

- Subculture from enrichment cultures (PCE, TCBs) + TCDD or OCDD

After over 6 months none of the cultures showed dioxin degrading activity

Microcosm experiments established with sediments

- Anaerobic cultures of HBB sediment + TCDD or OCDD

After ~ 2 years none of the cultures showed dioxin degrading activity
Bioaugmentation: *Dehalococcoides mccartyi* CBDB1

PeCDD dechlorination by *Dhc mccartyi* CBDB1 via TCDD (Bunge et al, 2003)

Set up microcosm experiments – Sediments spiked with TCDD plus:

- *Dehalococcoides mccartyi* strain CBDB1
- Homebush bay enrichment culture on TCBs

![Chemical structures]
Bioaugmentation: *Dehalococcoides mccartyi* CBDB1

CBDB1

Analysis by GC-TQMS after 1 month

Homebush Bay (HBB) enrichment culture on TCBs
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Summary
Summary - Key Points

1. Presence of known organohalide respiring bacteria in the sediments

2. Metabolic activity confirmed through the dechlorination of PCE and TCBs

3. Sediment microbial community does not appear capable of reductive dechlorination of dioxins (2,3,7,8- TCDD or OCDD)

4. Bioaugmentation with known dioxin- degrading bacterial isolate (*Dhc mccartyi* CBDB1) shows initial promising results and is being investigated further.
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