Degradation of the pesticide isoproturon is associated with the proliferation of diverse Sphingomonas spp.

Shengjing Shi and Gary D. Bending
Warwick HRI, University of Warwick, Wellesbourne, Warwick, CV35 9EF UK

Introduction

- The phenyl-urea herbicide isoproturon (IPU) is one of the most widely used pesticides in Europe, and is a major contributor to surface- and ground-water contamination of agricultural catchments.
- Earlier work at Deep Slade field in Wellesbourne, Warwickshire, UK, indicated that within field spatial variation of IPU degradation rate resulted from interactions between IPU catabolising Sphingomonas spp. and soil pH.
- The aim of this work was to investigate the diversity of Sphingomonas communities contributing to spatial variability in IPU catabolism using Sphingomonas genus primers.

Materials & Methods

- Soil samples (labelled as B, C, D, E, F) were taken at 20m intervals along a high pH (7-7.5) area known to give rapid degradation of IPU (transect 1), and a low pH area (6-6.5) known to give slow degradation (transect 2), which were separated by 50m (Bending et al., 2003).
- An aqueous suspension of IPU was added to the soils to give a concentration of 15 mg kg⁻¹ soil. Parallel control samples were set up using distilled H₂O in place of IPU. At intervals over 65 days, IPU residues were extracted from sub-samples of soil using acetonitrile, and concentrations were measured by HPLC.
- DNA was extracted from IPU treated and untreated soil samples at the point of 90% degradation (DT90).
- The Sphingomonas spp. community was amplified using the 16S rDNA primers Sphingo 108f/GC-40 and Sphingo 420r (Leys et al., 2004) and subsequently analysed by denaturing gradient gel electrophoresis (DGGE).

Results

- Time to 50% degradation (DT50) ranged from 6.1 to 6.7 days in transect 1, and 8.4 to 25.7 days in transect 2 (Table 1).
- In samples from transect 1 and samples D, E and F from transect 2, degradation followed growth-linked kinetics with an exponential phase of degradation following a lag phase.
- In transect 1 and one sample from transect 2 in which degradation rate was rapid (E), IPU degradation was associated with appearance of up to 12 new bands, 7 of which matched those from isolate Sphingomonas sp. SRS2, which was isolated from Deep Slade field in an earlier study (Sørensen et al., 2001).
- In transect 2, growth-linked degradation was associated with the appearance of a band showing homology with Sphingomonas mali, which did not occur in samples from transect 1.
- There was no change to the DGGE profile in samples B and C from transect 2, which showed slow linear degradation kinetics indicative of cometabolism.

Table 1 IPU degradation data

<table>
<thead>
<tr>
<th>Transect 1</th>
<th>Transect 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Site</td>
<td>B C D E F</td>
</tr>
<tr>
<td>DT50 (days)</td>
<td>6.1 6.7 6.2 6.1 6.2</td>
</tr>
<tr>
<td>Degradation mode</td>
<td>GL GL GL GL GL</td>
</tr>
</tbody>
</table>

GL: Growth-linked metabolism; CM: Cometabolism.

Conclusions

- The specific strains of Sphingomonas spp. adapted to degrade IPU varied over a small area within a single field.
- In a high pH area, degradation was rapid, and was associated with proliferation of a variety of Sphingomonas spp., including a strain previously isolated from the field using enrichment techniques.
- In a low pH area, degradation rate was slow, and associated with proliferation of a lower number of Sphingomonas spp., including a strain which did not occur in transect 1.

References


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