The Impact of Herbicides on Biota of the Intertidal Zone

Zostera muelleri

Gareth Lewis: A thesis submitted for admission to the degree of Master of Applied Science (Environmental Science), Soil & Land Systems, Adelaide University, Waite Campus, South Australia.
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by

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Soil & Land Systems

Faculty of Sciences

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**ABSTRACT**

Seagrasses provide an important habitat for gillfish, crustacea and migratory birds. Extensive losses of seagrass in the Northern Hemisphere have occurred since the 1930’s in what has been described as a ‘wasting disease’. More recently, point-source contamination by nutrient inflows, herbicides (anti-fouling agents used on commercial shipping), heavy metals and fresh water inflows have helped explain localised losses of seagrass amounting to 20 % in the case of Adelaide’s metropolitan coastline, South Australia. However, losses of seagrass acreage have also occurred in regions that are far removed from anthropogenic activity and these are less easily explained by point-source contamination.

Intertidal seagrasses, such as *Zostera muelleri*, are subjected to environmental pressures imposed on them by the marine and terrestrial environments. For the purpose of this thesis, the intertidal environment is regarded as a complex of several components or micro-environments, each imposing a selective pressure or stress upon seagrass. The many stress factors create a tolerance zone in which *Z. muelleri* can survive. *Zostera muelleri* has adapted its physiology and biochemistry to the selective pressures that operate within the intertidal region. *Zostera muelleri*’s internal leaf morphology has many gas storage compartments (lacunae) that extend from the leaves to the roots of the plants and its photosynthetic biochemistry has also adapted to the intertidal region enabling the sequestering of carbon under conditions of high irradiance and temperatures.

It is evident from the literature that the survival of intertidal seagrasses requires effective photosynthesis. It is also evident that events that interfere with the synthesis, translocation and release of photosynthesised oxygen from the roots of *Z. muelleri* will compromise seagrass survival.

The present study has revealed that herbicides, used in broad-acre farming, can be transported to the intertidal environment and negatively impact upon *Z. muelleri*. Extensive studies by others have shown that transport mechanisms, such as ‘spray drift’ and ‘run-off’, can move herbicides from their point of usage. However, ‘dust’ (wind-eroded soil) as a transport mechanism for herbicides to the intertidal environment is less well studied. This is surprising, inasmuch as there is a known rate of pedogenesis in Adelaide of five to ten tonnes per km$^2$ per annum from the accretion of dust.
Results of the present study suggest that farmed soils of the Yorke Peninsula have a range of potentials to form fine particulate matter (‘dust’) and this potential is likely determined by the soil type and farming practices. Soil surface applied herbicides, such as 2,4-D, are ‘lost’ from land at 5% of the applied rate while soil-incorporated herbicides, such as treflan (trifluralin), are lost at 1.5% of the applied rate. Indeed, such herbicides can be transported as dust for tens to thousands of kilometres.

Instrumental analytical techniques used in the present study have detected 2,4-D, trifluralin and sulfonylurea herbicides on whole soil. Additionally, 2,4-D-like chemicals have also been detected in whole soil and in dust obtained from whole soil. Bioassay techniques using *Z. muelleri* have shown that its photosynthetic pathways are negatively impacted upon by micromolar concentrations of 2,4-D that are similar to the known losses of this herbicide from land. It is concluded that, at these concentrations, 2,4-D acts as an auxin, up-regulating growth in affected plants. Such up-regulation is unlikely to be problematic in terrestrial plants since gas flows to the external environment are largely controlled by stomata. However, seagrasses lack stomata and the auxin-like activity of 2,4-D appears to have a negative impact on *Z. muelleri*. This is probably caused by an up-regulation in oxygen production and a subsequent oxygen-inhibition of a key enzyme (ribulose 1,5-bisphosphate carboxylase, RUBISCO) used in the carbon-sequestering photosynthetic process. The proposed inhibition of RUBISCO is then likely to cause a carbon deficit and a subsequent energy deficit within affected plants. One interpretation of the results presented is that *Z. muelleri* simply outgrows its intertidal environment after a transient exposure to an auxin-like concentration of 2,4-D.

With increasing use of auxin-like herbicides, and the associated increasing stress imposed on photosynthetic processes, it is likely that further negative impacts will occur on intertidal seagrass species. Continued depletion of seagrass acreage will further adversely affect fishing yields unless appropriate measures are not taken. Closer collaboration between regulators, farm managers and herbicide manufacturers is now necessary in order to minimise the negative impact of herbicides on intertidal species.
DECLARATION

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

I give consent to this copy of my thesis, when deposited in the University Library, being available for loan and photocopying.

Gareth Lewis.
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<td>GCMS</td>
<td>Gas chromatography with mass spectral detection</td>
</tr>
<tr>
<td>Sim mode</td>
<td>Gas chromatography with selective ion monitoring</td>
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<tr>
<td>GCECD</td>
<td>Gas chromatography with electron-capture detection</td>
</tr>
<tr>
<td>FTIR</td>
<td>Fourier transform infra-red spectroscopy</td>
</tr>
<tr>
<td>HPLC</td>
<td>High performance liquid chromatography</td>
</tr>
<tr>
<td>HPTLC</td>
<td>High performance thin layer chromatography</td>
</tr>
<tr>
<td>HIC</td>
<td>Hydrophobic interaction chromatography</td>
</tr>
<tr>
<td>BAZ</td>
<td>Buoyant Aquatic Zostera</td>
</tr>
<tr>
<td>ACC</td>
<td>1-aminocyclopropane-1-carboxylic acid synthase</td>
</tr>
<tr>
<td>RUBISCO</td>
<td>Ribulose 1,5-bisphosphate carboxylase</td>
</tr>
<tr>
<td>2,4-D</td>
<td>2,4-dichlorophenoxyacetic acid</td>
</tr>
<tr>
<td>2,4-D-asp</td>
<td>The amino acid (aspartate) conjugate of 2,4-D</td>
</tr>
<tr>
<td>2,4-D-glu</td>
<td>The amino acid (glutamate) conjugate of 2,4-D</td>
</tr>
<tr>
<td>ppb</td>
<td>Parts per billion ($10^9$)</td>
</tr>
<tr>
<td>ppt</td>
<td>Parts per thousand ($10^3$)</td>
</tr>
<tr>
<td>Mr</td>
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