Use of Higher-Tier Laboratory Testing in Mesocosm Studies

Jenna Ashford, Frances Pickering, Hanna Schuster
Cambridge Environmental Assessments, Boxworth, UK; Corresponding author: jenna.ashford@cea-res.co.uk

Introduction

- Outdoor mesocosms are an excellent resource for producing aquatic community endpoints for Plant Protection Products (PPP). However, regulators occasionally require more information on a particular species and this can be addressed through higher-tier laboratory testing (Fig. 1, 3 and Fig. 5).
- Here we present two examples of higher-tier laboratory studies, showing the setup of the tests using several non-standard aquatic invertebrates to provide additional information for clients and produce endpoints to support mesocosm data.

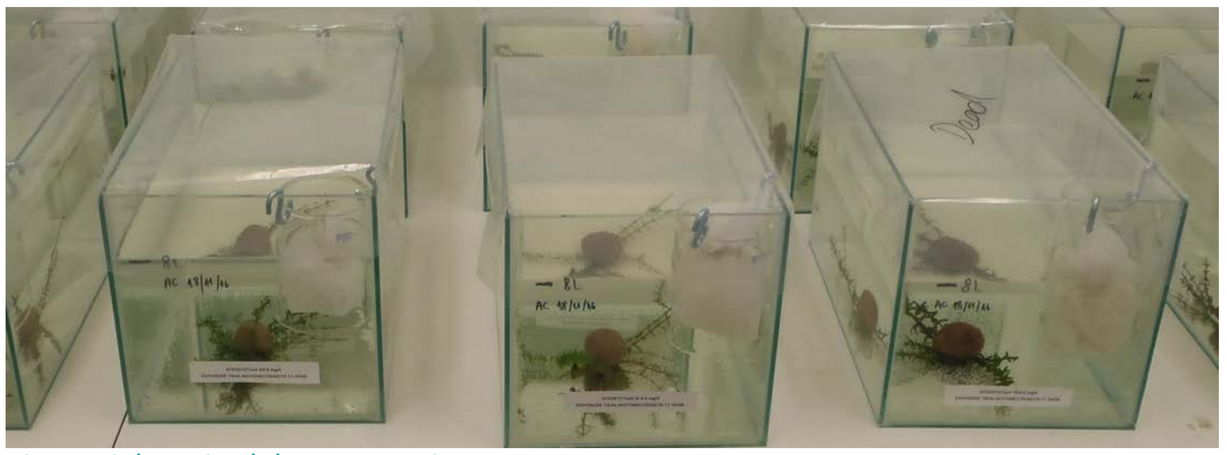

Fig. 1 Higher-tier laboratory microcosms.

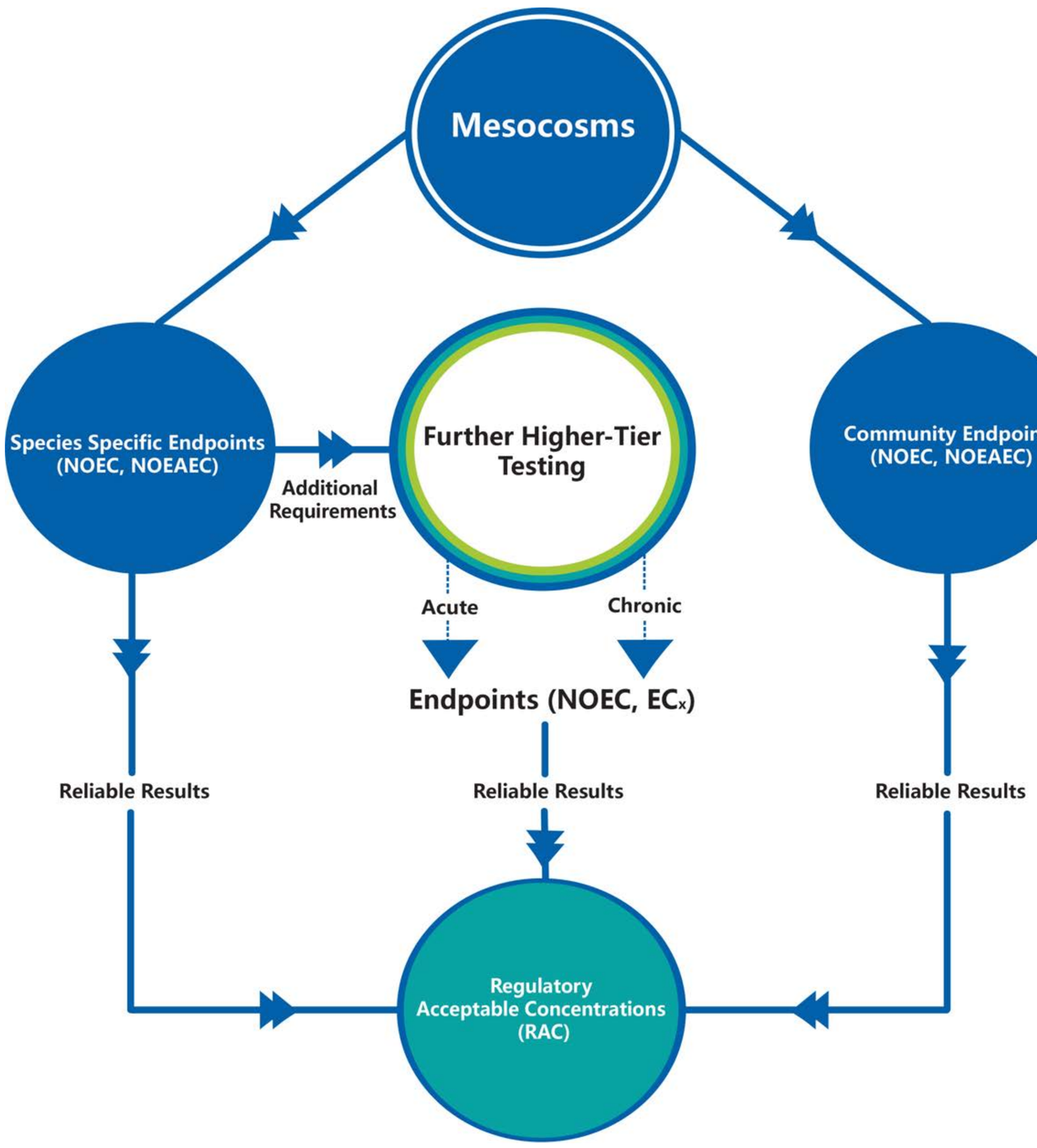

Fig. 2 Use of acute and chronic higher-tier laboratory testing to support species specific endpoints to derive reliable Regulatory Acceptable Concentrations.

Case Study 1: Mortality or Migration?

- A previous mesocosm study showed significant reductions of a flying macroinvertebrate in response to test item application.
- It remained unclear if the test item caused mortality or if the organisms migrated away from the treated systems and would survive application.

Study and setup

- 2 week exposure to the test substance to investigate the survival and well being of the test organism.
- 16 tanks each covered by a plastic lid held 8 L filtered (45 µm) pond water with elodea, rocks (Fig. 1).
- Water temp. 18-22°C and 16 h light: 8 h dark photoperiod.
- Test substance was applied once via air gun to replicate the chemical application in the environment (Fig 5).

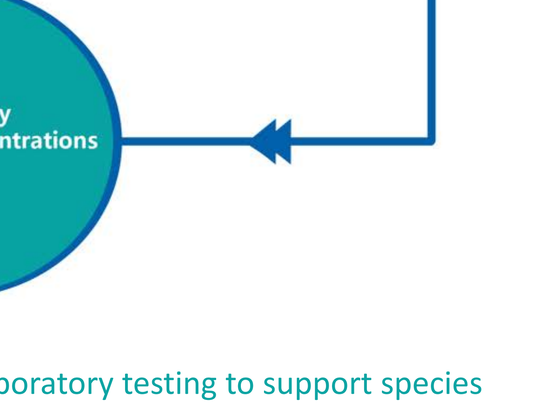

Fig. 3 Application of test substance via air gun.

Results

- No difference between control and treatment was detected.
- The additional laboratory experiment showed that the ability of the organism to migrate away from the treated water body resulted in a lower endpoint compared to the laboratory experiment where migration was prevented.

Case Study 2: Comparative Sensitivity

- A series of tests was conducted on three different aquatic vulnerable macroinvertebrates (stonefly, alderfly and caddisfly larvae) and two species with a known sensitivity to the test substance (Chaoborus and Crangonyx) to determine relative sensitivity.

Collection and culturing

- Macroinvertebrates from different environments in the UK were collected using various sampling techniques (Fig. 3).
- *Chaborous* and *Crangonyx* were collected from the mesocosm facility at CEA.
- All organisms were kept in cultures with vegetation prior to study start, between 16 - 20°C, 16 h light and dissolved oxygen >60 % ASV.

Fig. 4 Collection of test organisms

Study and setup

- Microcosms were tailored specifically for each species to reduce stress while keeping the test systems and the exposure period similar (Fig. 4) so that the results were comparable.
- A single dose of the test substance was applied to the water column, in the presence of sediment in order to simulate the exposure-profile of a static edge-of-field ditch and the previous mesocosm study.

Results

- In comparison to *Chaoborus* and *Crangonyx*, the stonefly, alderfly and caddisfly larvae were less sensitive to the test item.
- The mesocosm endpoint was shown to be protective over taxa not present in the study.
- Reduced uncertainty over the effects on aquatic insect larvae, of which stonefly larvae are amongst the most sensitive to pesticide exposure.

Fig. 5 Microcosms

Conclusion

Open questions that can be highlighted by regulators as a response to a mesocosm study or the re-analysis of old mesocosm studies (MDD analysis; EFSA 2013) can be addressed by customised higher tier laboratory studies:

- More realistic than standard laboratory studies as they recreate an outdoor mesocosm's environment in the laboratory.
- Provide the potential to assess the sensitivity to contaminants of species that cannot be found in mesocosms.
- Focus on potential data gaps of test item effects on individual organisms.
- Specific questions (e.g. mortality or migration) can be more easily addressed in a laboratory setting.
- Producing reliable results and supporting data for risk assessments.

→ Overall, these laboratory microcosms can be used in conjunction with standard laboratory and outdoor higher-tier mesocosm studies and thereby increase certainty in the risk assessment approach.

References

- EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues) 2013 Guidance on tiered risk assessment for plant protection products for aquatic organisms in edge-of-field surface waters. EFSA Journal 2013;11(7):3290