DEVELOPMENTS IN LABORATORY & *IN-SITU* INVERTEBRATE BIOASSAYS FOR USE IN HIGHER TIER MESOCOSM TESTING

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INTRODUCTION & OBJECTIVES

Due to their closed system design, common problems associated with microcosm and mesocosm studies arise, including:

- Absence of biologically relevant recovery of taxa unable to re-colonise mesocosms by means other than reproduction: uni-voltine organisms and those lacking an aerial life stage (e.g. Asellidae and Crangonyctidae)
- Organisms with naturally low population densities may be overlooked (e.g. Odonata, Figure 1)
- Timescale flexibility may be required to address effect classifications (de Jong *et al*. 2008)

Laboratory based and *in-situ* invertebrate bioassays are established methods in mesocosm testing. Recent work carried out by CEA utilising these techniques for studies with numerous insecticides has attempted to address these issues.

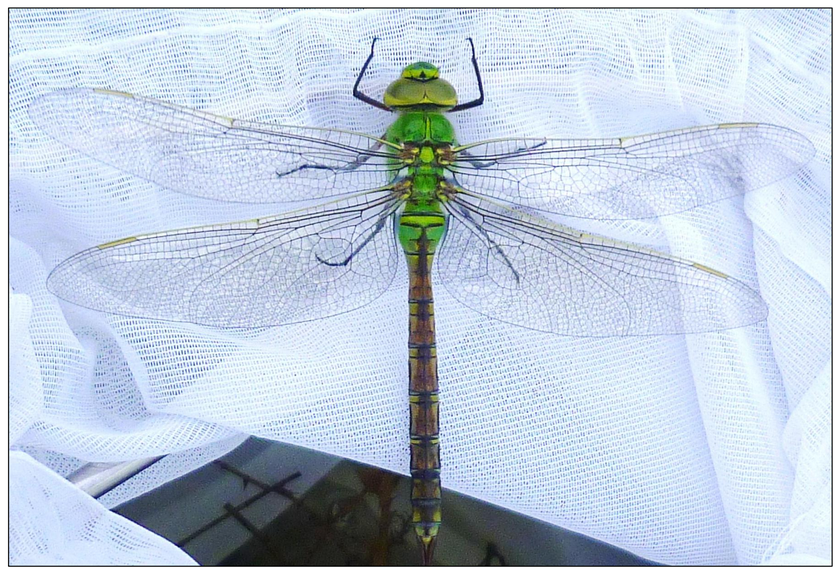

Figure 1: Emerged Aeshnidae from an *in-situ* bioassay

RESULTS & DISCUSSION

Low abundance species

Laboratory study – Aeshnidae:

- Test media = aged residues (water collected from the mesocosms 56 days after treatment and stored frozen)
- Each replicate contained one organism exposed to 600 mL of aged residue water (Figure 2)
- Monitored for lethal and sub-lethal effects over 7 days

Figure 2: Aeshnidae Laboratory bioassay.

***In-situ* bioassay** - Aeshnidae and Pleidae (Figures 3 and 4):

- During the study, organisms were periodically added to fine meshed cages situated in the mesocosms
- This facilitated exposure to previously treated mesocosms under the conditions of the test

Figure 3: Pleidae *in-situ* bioassay.

Results - Aeshnidae and Pleidae:

- Very low numbers of these organisms were found free-living in the mesocosm community
- Dose related residual toxicity was observed in *in-situ* bioassays on early assay occasions
- No effects were observed on later occasions from *in-situ* or laboratory bioassays
- Mean survival in the control group remained high for all organism types

Figure 4: Aeshnidae, Pleidae, Crangonyctidae and Asellidae *in-situ* bioassays.

Isolation & recovery

Laboratory study - *Daphnia magna* reproduction study (Figure 5):

- Test media = aged residues collected from treated mesocosms
- Each replicate contained one organism and 60 mL of test media
- Monitored for lethal and sub-lethal for 21 days

***In-situ* bioassay** - Crangonyctidae and Asellidae:

- Organisms were periodically added to fine meshed cages contained within the mesocosms

Results

Laboratory study - *Daphnia magna*:

- Results indicated some indirect effects on growth and reproduction however, *D. magna* exposed to aged residues were able to grow and reproduce normally

Figure 5: *D. magna* reproduction study.

***In-situ* bioassay** - Crangonyctidae and Asellidae:

- Free-living populations of Crangonyctidae and Asellidae were impacted at low doses during the study
- *In-situ* bioassays with *A. aquaticus* and *C. pseudogracilis* indicated no clear effect on the survival of caged organisms introduced following two overspray applications
- Evidence of reproduction was also observed on some occasions
- Clear evidence of the potential for recovery from up-stream refugia and un-impacted sites can be demonstrated

CONCLUSIONS & RECOMMENDATIONS

- Potentially impacted populations of invertebrates in real edge-of-field water bodies could recover through external recovery processes (e.g. re-colonisation from unexposed areas or refugia).
- *In-situ* bioassays are a better reflection of environmental realism however laboratory studies allow sub-lethal effects to be observed.
- Laboratory studies allow for timescale flexibility e.g. when organism's life histories do not coincide with the typical mesocosm timescale.
- Using a range of organisms including key-stone species in bioassays, ecotoxicological thresholds may be established for a range of organisms and this data may be extrapolated to understand potential effects on other organisms within taxonomic guilds or functional groups.

Reference

DE JONG, F.M.W., BROCK, T.C.M., FOEKEMA, E.M., & LEEUWANGH, P. (2008) Guidance for summarising and evaluating aquatic micro and mesocosm studies, RIVM Report 601506009/2008.