ELSEVIER

FEMS Microbiology Ecology 54 (2005) 157–165

FEMS MICROBIOLOGY Ecology

www.fems-microbiology.org

Response of water column microbial communities to sudden exposure to deltamethrin in aquatic mesocosms

Charles W. Knapp [a], Thierry Caquet [b], Mark L. Hanson [b,c], Laurent Lagadic [b], David W. Graham [a,*]

[a] *Department of Civil, Environmental and Architectural Engineering, 4112 Learned Hall, 1530 W, 15th Street, University of Kansas, Lawrence, KS 66045, USA*

[b] *UMR 985 INRA-Agrocampus Ecobiologie et Qualité des Hydrosystèmes Continentaux, Equipe Ecotoxicologie et Qualité des Milieux Aquatiques, F-35042 Rennes, France*

[c] *Department of Environment and Geography, 213 Isbister Building, University of Manitoba, Winnipeg, Man., Canada R3T 2N2*

Received 1 December 2004; received in revised form 4 February 2005; accepted 16 March 2005

First published online 1 April 2005

Abstract

Sudden exposure of an aquatic system to an insecticide can have significant effects on populations other than susceptible organisms. Although this is intuitively obvious, little is actually known about how such exposure might affect bacterial communities and their relative metabolic activity in ecosystems. Here, we assessed small sub-unit (ssu)-RNA levels in open and shaded 9 m^3 aquatic mesocosms (16 units – 2×2 factorial design in quadruplicate) to examine the effects of sudden addition of deltamethrin to the units. When deltamethrin was added, a cascade of bacterial then phytoplankton "blooms" occurred over time. The bacterial bloom, which most likely included organisms from the plastid/cyanobacterial phylogenetic guild, was almost immediate (within hours), whereas the phytoplankton (algal) bloom lagged by about 4 days. This sequential response can be explained by an apparent sudden release of nutrients consequent to arthropod death that triggered a series of responses in the microbial loop. Interestingly, bacterial blooms were noted in both open and shaded mesocosms, whereas the algal bloom was only seen in open units, suggesting that both deltamethrin addition (and presumptive nutrient release) and an adequate light supply was required for the phytoplankton response. Overall, this work shows that microbial activities as reflected by ssu-rRNA levels can respond dramatically via apparently indirect effects following insecticide application.

Keywords: Deltamethrin; Pesticide; Aquatic; Ssu-rRNA; Bacteria; Phytoplankton; Indirect effects

1. Introduction

Deltamethrin (3-(2,2-dibromoethenyl)-2,2-dimethylcyclopropanecarboxylic acid cyano(3-phenoxyphenyl) methyl ester) is a pyrethroid ester insecticide that is often used for the control of mosquitoes and other nuisance arthropods in or close to aquatic systems. Typically, it is applied to an infested area over a short timeframe effectively killing the majority of the target insect(s) and many other co-existing invertebrates at the site [1]. However, Caquet et al. [2] showed that, although insecticide applications directly impact arthropods, they can also indirectly impact other organisms in the food web through both bottom-up and top-down mechanisms. For example, the elimination of herbivorous arthropods reduces grazing pressure that might result in increased levels of primary producers, such as periphyton [3] and phytoplankton [4,5]. Alternately, die-off of the target

* Corresponding author. Tel.: +1 785 864 2945; fax: +1 785 864 5379.
E-mail address: dwgraham@ku.edu (D.W. Graham).

0168-6496/$22.00
doi:10.1016/j.femsec.2005.03.004

organisms might release secondary nutrients that become available for other microbial activity. As a result, ancillary ecological and chemical responses may cause microbial blooms or other ecosystem modifications not directly intended by the insecticide application [6,7].

Although such effects might be intuitive, little documentation exists on the exact nature of microbial community responses, especially at the bacterial level. Here we specifically assess microbial responses within the context of a much larger study assessing whole ecosystem recovery after exposure to deltamethrin [8]. In the larger study, the insecticide was added to a set of sixteen 9-m^3 aquatic field mesocosms, with and without screen lids (and appropriate controls), as a single dose targeted at 2 $\mu g\, l^{-1}$ nominal concentration to assess the short-term impact of insecticide addition and the effect of mesocosm covers on re-colonization after exposure. Although detectable deltamethrin was retained in the water column for less than 4 days, it had its desired effect on invertebrates with a loss of over 90% of emergent insects and a major reduction in zooplankton and benthic arthropods (e.g., insect larvae) over the week following insecticide addition [8]. Thus, conditions in the units (in conjunction with controls) were created to allow the study of ''secondary'' microbial community responses in the same systems, which was the primary goal of this work.

The key to successfully assessing microbial responses in the systems was to choose a technique that would detect both culturable and non-culturable species and also have the potential to target individual bacterial groups at higher taxonomic resolution. Therefore, ssu-rRNA (small sub-unit ribosomal RNA) hybridization techniques were adopted, which have advantages over other microbiological techniques in that they allow the detection of in situ microbial activities and not just abundance [9]. For example, ssu-rRNA methods provide a generally good representation of metabolically active organisms [10–12] because cell ribosome content tends to be proportional to growth rate [13–15], although some deviation is sometimes noted during non-steady-state growth experiments [16]. Further, sss-rRNA methods do not require microbial culturing that skews communities towards species that can be grown on plates. Finally, Pace and Cole [17] showed that increased bacterial activity (as suggested by ssu-rRNA levels) often precedes population growth and can be important when assessing a possibly rapid microbial response, which may be the case here. Although ssu-rRNA methods were primarily employed, direct microscopic counts were also performed to compare our molecular data with whole-organism data generated by others in this project.

The primary goal in this work, therefore, was to assess the effect of deltamethrin addition on microorganisms in aquatic systems. An ancillary goal of the work was to assess the influence of shading on the nature and extent of the observed bacterial and phytoplankton responses. It was hypothesized that the pulse addition of deltamethrin would result in increased bacterial and phytoplankton activities either by the release of nutrients from decaying arthropods or due to reduced grazing pressures. Our results demonstrate a rarely seen ''top-down effect'' in natural microbial communities resulting from insecticide exposure only previously seen in soil studies [18].

2. Materials and methods

2.1. Mesocosm experimental design

The experiment assessing the response among microbial communities to deltamethrin addition used aquatic mesocosms and was performed during Spring 2003 at the Rennes site of the Experimental Unit of Aquatic Ecology and Ecotoxicology of the Institut National de la Recherche Agronomique (INRA), France. It was a sub-component of a large two-year study that assessed aquatic arthropod recovery after sudden deltamethrin exposure as influenced by aerial re-colonization (or not) as controlled by screened lids on selected units. The experiment used a 2×2 factorial design with deltamethrin addition being one factor and the inclusion of white fiberglass 1-mm mesh screen lids ($\sim$37% reduction of Photosynthetically Active Radiation – PAR) being the other. The four treatments were (1) open control, (2) open deltamethrin, (3) covered control, and (4) covered deltamethrin. All treatments were maintained in quadruplicate. Further details of the experimental design are provided elsewhere [8].

The large experiment commenced in Spring 2002, although detailed molecular and microbial monitoring was only performed between mid March 2003 ($\sim$1 month prior to deltamethrin addition) and August 2003. Deltamethrin was added on 22 April 2003 by subsurface spraying, and monitoring was performed weekly with the exception of a two-week window of more intensive sampling immediately before and after deltamethrin addition.

2.2. Monitoring program

The physical, chemical and biological conditions in the 16 mesocosms were monitored by the collection of water samples for laboratory analysis and the real-time measurement of various parameters using field probes. Samples for water chemistry and molecular microbiological analysis were collected using rinsed, pre-sterilized PVC tube samplers (one sampler per mesocosm) that had a screen-covered one-way valve at the bottom for easy withdrawal to sample storage bottles (similar to Graham et al. [19]). Typically, individual volumes were