



## Can we predict community-wide effects of herbicides from toxicity tests on macrophyte species?

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### ARTICLE INFO

#### Article history:

Received 4 March 2010

Received in revised form 26 August 2010

Accepted 28 August 2010

#### Keywords:

Pesticide mixture

Chemical stress

Community ecotoxicology

Water plant assemblages

### ABSTRACT

Macrophyte communities play an essential role in the way freshwater ecosystems function. It is thus of great concern to understand how environmental factors, especially anthropogenic ones, influence their composition and diversity. The aim of this study was to examine whether the effects of a herbicide mixture (50% atrazine, 35% isoproturon, 15% alachlor) on single macrophyte species can be used to predict its impact at a community level. In a first experiment we tested the sensitivity of six species (*Azolla filiculoides*, *Ceratophyllum demersum*, *Elodea canadensis*, *Lemna minor*, *Myriophyllum spicatum* and *Vallisneria spiralis*) grown separately and exposed to 0.6–600  $\mu\text{g L}^{-1}$  of the herbicide mixture. In a second experiment, conducted in microcosms, we tested the effects of herbicides on macrophyte assemblages composed of the same six species exposed to 0, 6 or 60  $\mu\text{g L}^{-1}$  of the herbicide mixture. Species grown separately exhibited growth inhibition at 60 and 600  $\mu\text{g L}^{-1}$ . At 600  $\mu\text{g L}^{-1}$  the sensitivity differed significantly between species. *V. spiralis* was the most resistant species, *C. demersum*, *M. spicatum* and *E. canadensis* exhibited intermediate sensitivities, and *A. filiculoides* and *L. minor* were the most sensitive species. In microcosms, community biomass and Shannon evenness index were reduced after 8 weeks at 60  $\mu\text{g L}^{-1}$ . Communities also exhibited changes in their composition: the relative and absolute abundance of *C. demersum* increased at 6  $\mu\text{g L}^{-1}$ , while the relative abundance of *V. spiralis* increased at 60  $\mu\text{g L}^{-1}$ . These results are in agreement with the individual responses of these species to the herbicides. It is therefore concluded that short-term effects of herbicides on simple macrophyte communities can be predicted from the sensitivity of individual species. However, further investigations are required to examine whether longer term effects can be predicted as well, especially in more complex communities.

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### 1. Introduction

Aquatic macrophytes are key elements of freshwater ecosystems. They provide food, shelter and substrate to various aquatic organisms (Carpenter and Lodge, 1986; Lodge, 1991). Moreover, rooted macrophytes influence flow, sediment stability and organic matter retention, and play an important role in oxygenating the water column and superficial sediment layers (Carpenter and Lodge, 1986; Sand-Jensen, 1998; Clarke, 2002). Macrophytes are also pivotal in sustaining water clarity of shallow standing waters, as they efficiently inhibit phytoplankton development via nutrient competition and release of allelopathic compounds (Scheffer et al., 1993). On the other hand, both native and exotic macrophyte species can proliferate in some instances, raising important ecological and economic concerns (Murphy, 1988; Peltre et al., 2002). For these different reasons, macrophytes are often of great importance

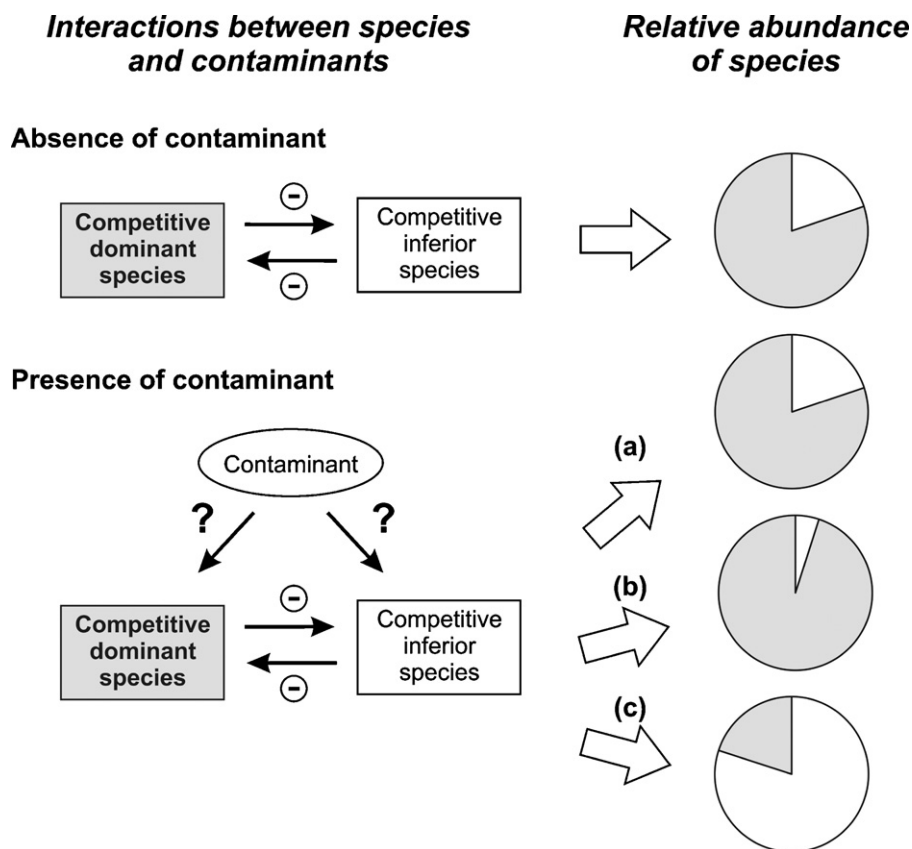
to environmental managers who aim to promote some species and control others, and are therefore eager to understand mechanisms of community structuring (Coops et al., 2002).

In natural conditions, the composition and diversity of freshwater macrophyte communities are ruled by various environmental factors, such as hydraulic disturbances, nutrient richness or light availability (Melzer, 1999; Amoros et al., 2000; Lacoul and Freedman, 2006). Biotic interactions, such as competition or herbivory, also play a role in structuring macrophyte communities (Barrat-Segretain, 2005; Elger et al., 2009). In addition, due to the importance of human impacts on freshwater ecosystems, predictive models of macrophyte community structure must take into account anthropogenic pressures such as river regulation, water eutrophication or contamination by xenobiotics. All these natural and anthropogenic determinants can be seen as environmental filters that select some species, based on their life-history traits, from among a local or a regional pool (Keddy, 1992; Clements and Newman, 2002).

Herbicides are the xenobiotics most likely to impact macrophyte communities. Even if we exclude direct applications of these sub-

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**Fig. 1.** Effects of a sublethal contaminant in the outcome of competitive interactions between two plant species. In the absence of contaminant we expect the competitively dominant species to be the most abundant in the community. In the presence of contaminant, several outcomes are possible depending on the relative sensitivity of species: (a) if both species are equally sensitive, their relative abundance should remain the same; (b) if the competitively dominant species is less sensitive, its abundance should further increase; (c) if the competitive inferior species is less sensitive, its abundance should increase and possibly lead to a shift in the relative abundance of the two-species (modified, after Rohr et al., 2006).

stances for controlling water plants (Hofstra and Clayton, 2001), herbicides frequently enter freshwater systems and can be present at non-negligible concentrations in water or sediments (Lerch and Blanchard, 2003; Muller et al., 2004). This is particularly the case for pre-emergent herbicides, used in agriculture at a period with low vegetation cover on arable lands, and easily leached to aquatic ecosystems through superficial run-off or drain-water supplies. For instance, in the mid-Garonne River and its tributaries, Devault et al. (2007) found herbicide concentrations above  $5 \mu\text{g g}^{-1}$  in sediments (in order of decreasing importance: chloro-s-triazines > substituted phenylureas, chloroacetanilides), and above  $2 \mu\text{g L}^{-1}$  in unfiltered water (mainly substituted phenylureas; chloro-s-triazines to a lesser extent). During flood events, herbicide concentrations in water can exceed  $10 \mu\text{g L}^{-1}$  (Debenest et al., 2009).

Early investigations have shown that these herbicide concentrations usually found in freshwater ecosystems only have a low inhibitory effect on the growth of common macrophyte species (Jones and Winchell, 1984; Jones et al., 1985). However, because herbicide sensitivity varies across macrophyte species and largely depends on the compound considered (Cedergreen et al., 2004a,b; Fairchild et al., 1998; Lambert et al., 2006), we cannot exclude important community-wide effects of sublethal herbicide concentrations on macrophyte assemblages, such as shifts in the dominance of some species or functional groups of macrophytes. Such shifts might be difficult to predict, as they can result from both direct effects of herbicides on individual species (and therefore be related to the intrinsic sensitivity of the species to the xenobiotics considered) and from indirect effects via the modulation of biotic

interactions involving macrophytes (Rohr et al., 2006). For instance, interspecific competition could be affected by herbicides, due to differential sensitivity among species (Relyea and Hoverman, 2006). This could enhance diversity, if the competitive dominant species is the most sensitive to the herbicides (direct detrimental effect on key species inducing positive indirect effect on other species, *sensu* Rohr et al., 2006). But other trajectories in community structure are also possible (Fig. 1). Despite the need to predict herbicide effects at a community scale, very few studies have assessed the toxicity of such compounds on freshwater plant communities (Coors et al., 2006). Moreover, most of these studies have not examined whether the observed effects could be predicted from individual responses of species to herbicides (but see McGregor et al., 2008), which is crucial information for risk-assessment approaches.

In the present article, we addressed this question through measured growth effects of a herbicide mixture (50% atrazine, 35% isoproturon and 15% alachlor) on six macrophyte species. The choice of compounds and their proportions in the mixture was made so as to represent the three major families of herbicides widely used in agriculture during the last 15 years (i.e. s-triazines, anilides and substituted ureas) (European Commission, 2007). In a first experiment, we tested the effects of a range of concentrations of the herbicide mixture on separately cultivated macrophytes, in order to assess their intrinsic sensitivity. In a second experiment, we studied the response of the species grown together and forming a simple macrophyte community, to the same herbicide mixture.

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