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Long-term ecological consequences of herbicide treatment to control the invasive grass, *Spartina anglica*, in an Australian saltmarsh

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ABSTRACT

Invasive plants acting as habitat modifiers in coastal wetlands can have extensive ecological impacts. Control of invasive plants often relies on herbicides, although little is known about subsequent environmental impacts. Studying effects of herbicides on non-target species and long-term cascading consequences may yield insights into the ecology of invasive species by revealing interactions with native species. We conducted a long-term field experiment measuring effects of treating the invasive saltmarsh grass, *Spartina anglica*, with the herbicide Fusilade Forte®. No changes in sedimentary macrofaunal abundances or species richness, diversity, or assemblages were detected 1–2 months after spraying, despite known toxicity of Fusilade Forte® to fauna. This lack of impact may have been due to low exposure, since the herbicide was taken up primarily by plant leaves, with the small amount that reached the sediment hydrolyzing rapidly. Six months after spraying, however, total macrofauna in treated plots was more than four times more abundant than in unsprayed control plots, due to a fifteen-fold increase in annelids. This population growth correlated with increased sedimentary organic matter in treated plots, likely due to decomposition of dead *S. anglica* leaves serving as food for annelids. After another year, no differences in macrofauna or organic matter remained between treatments. The indirect effect on annelid populations from herbicide treatment could benefit management efforts by providing greater food resources for wading birds, in addition to improving birds' access to sediments by reducing plant cover. This study shows that an invasive grass can have a significant impact on native fauna through food-web interactions, influenced by herbicide usage.

1. Introduction

Invasive plants functioning as ecosystem engineers in coastal habitats and wetlands can have extensive impacts on habitat complexity, floral and faunal assemblages, and food web dynamics (Grosholz and Ruiz, 2009; Kovalenko et al., 2012). Impacts of invasive species can also be difficult to predict (Branch and Steffani, 2004; Cutajar et al., 2012); therefore, eradication or control is of paramount importance. Programs to manage invasive plants rely mostly on herbicide treatment due to relatively low cost and ease of application.

Various herbicides are chosen for use, generally based on efficacy against the target species. For example, the high-profile invasive reed *Phragmites australis* and saltmarsh grasses *Spartina anglica* and *Spartina alterniflora* are commonly treated with glyphosate, imazapyr, dalapon, haloxyfop, or fluazifop (Marks et al., 1994; Palmer et al., 1995; Hammond and Cooper, 2002; Patten, 2002; Hedge et al., 2003; Mozdzer et al., 2008). Only in a few cases, however, is there information on any short-term effects of the herbicides on non-target species, particularly fauna, or on longer-term indirect consequences of plant die-back for the ecosystem. For example, experiments with glyphosate in microcosms and mesocosms have shown mixed results for toxicity to fauna (Chen et al., 2004; Relyea, 2005), but Warren et al. (2001) and Kulesza et al. (2008) found no impacts on macrofaunal assemblages over a year after treating *P. australis* stands in the field. On the other hand, die-back of treated *P. australis* can alter long-term hydrology and sediment geochemistry (Marks et al., 1994), and Back et al. (2012) found increased snail abundances one year after treatment

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of *P. australis* as an indirect consequence of canopy opening. Studying non-target impacts and long-term cascading consequences is crucial to protect native biodiversity and for informed management practices, and may yield insights into the ecology of the invasive species in the local environment by revealing direct and indirect interactions with native species.

Spartina anglica is an invasive weed in coastal areas of Europe, the U.K., North America, China, New Zealand, and Australia where it invades mudflats, saltmarshes, seagrass beds, and mangroves (Ranwell, 1964; Chung, 1983; Groenendijk, 1986; Kriwoken and Hedge, 2000; Nehring and Hesse, 2008). The species forms expansive swards with dense roots and rhizomes affecting sediment accretion, hydrology, and sediment characteristics such as grain size, pH, salinity, organic matter content, and oxygen (da Cunha Lana and Guiss, 1991; Bouma et al., 2005; Peralta et al., 2008; Cutajar et al., 2012; Sheehan and Ellison, 2014). There can be dramatic reductions in macrofaunal abundances, species richness, and diversity, although the impacts are highly variable (Bouma et al., 2009; Tang and Christensen, 2010; Cutajar et al., 2012). *S. anglica* is also considered a threat to populations of wading shorebirds by reducing foraging area and food abundances (Hammond and Cooper, 2002). Millard and Evans (1984) and Evans (1986) documented that several shorebirds, particularly flocking species, avoided *S. anglica* infestations. Some studies suggest, however, that *S. anglica*'s detritus contributes to estuarine ecosystem productivity (Kriwoken and Hedge, 2000; Hindell and Warry, 2010).

Spartina anglica infestations are difficult and expensive to eradicate. Small-scale control methods such as physical removal, smothering, or steam treatment are labor-intensive, and the most cost-effective control is considered to be herbicide treatment (Kriwoken and Hedge, 2000; Hedge et al., 2003; Roberts and Pullin, 2008). In Australia, the grass-specific herbicide Fusilade® was chosen for *S. anglica* control programs in the mid 1990s. The product is absorbed through leaves, after which the active ingredient, fluazifop-*P*-butyl, is translocated to growing parts of the plant (apical meristems and roots) where it inhibits lipid biosynthesis (Syngenta, 2010). The active ingredient is metabolized or hydrolyzed (in the plant or in the environment) to a breakdown product fluazifop-*P*-acid, which is also herbicidally active. The commercial product also includes ethoxylated oleic acid and several other non-hazardous compounds. The choice of this herbicide was based on field trials showing up to 92% reduction of live plant coverage over three years following a single application, and rapid hydrolysis of the active ingredient in the environment after application (Palmer et al., 1995; Pritchard, 2005). Although laboratory studies revealed toxicity to two indicator species (the amphipod, *Allorchestes compressa*, and the smallmouth hardyhead fish, *Atherinosoma microstoma*), field spraying of seagrass beds (*Zostera muelleri*) caused mortality in only 7 of the 74 invertebrate species sampled and no reduction of seagrass biomass after 4 weeks (Palmer et al., 1995).

These early studies, however, were of insufficient time to allow longer-term, possibly cascading effects to develop in the ecosystem, and the only study following macrofauna after herbicide spraying was done in seagrass beds rather than in *S. anglica* stands (Palmer et al., 1995). Furthermore, in 2005 Fusilade® was replaced with a newer formulation, Fusilade Forte®, which was not tested in field trials. The material safety data sheet for Fusilade Forte® (Syngenta, 2010) reports low toxicity to aquatic invertebrates (*Daphnia magna*) and to rainbow trout, which are not particularly relevant to *Spartina* application, yet the product is not registered for use in aquatic systems and requires an off-label permit for *Spartina* management (Tu et al., 2001). However, recent laboratory tests with Fusilade Forte® on invertebrates from our field site in Australia where *Spartina anglica* is managed showed moderate toxicity to two annelids (*Aglaophamus australiensis* and *Lumbriculus variegatus*) and the amphipod *Allorchestes compressa* (unpublished results, L. Kleinhenz, RMIT University).

We conducted a field experiment on effects of Fusilade Forte® application in a *Spartina anglica* meadow in temperate Australia. We asked the following questions. 1) How long do Fusilade Forte® and its breakdown product persist in various compartments of the environment (sediment, plant leaves, and roots)? 2) Are there any short-term (1–2 months) direct toxicity effects on macrofaunal populations in the field? 3) Are there any longer-term (up to 1.5 yr) indirect consequences for macrofaunal populations as dead plant biomass breaks down and alters sediment properties?

2. Methods

2.1. Field experiment

Experimental plots were established within a dense and mature meadow (ca. 1 ha) of *Spartina anglica* in the intertidal zone of Anderson Inlet (145° 49′ 31.3″ E, 38° 39′ 23.1″ S), southeast Australia, which had not previously been treated with herbicide. The coverage of *S. anglica* was 100%, with 2 kg m^{-2} (dry weight) of emergent biomass. Ten plots (35 m × 6 m, with the long axis oriented along the elevation gradient), separated by 12 m gaps, were marked with stakes at the corners. Five plots were randomly chosen for herbicide treatment, and the other five served as untreated controls.

Herbicide was applied to the treatment plots at low tide on 31 May (Australian autumn), 2012, from a helicopter with a boom sprayer. Application typically occurs in summer and autumn, when plants are growing and will incorporate the herbicide effectively (pers. comm. L. Leunig, Parks Victoria). The herbicide was sprayed at the start of a spring-tide low water on a day of calm wind, when the plants and sediment were fully exposed, allowing for maximum uptake of the herbicide before the next incoming tide. Fusilade Forte® was diluted with water to 16.5 L per 100 L and applied at 100 L ha^{-1}, equating to 2.112 kg ha^{-1} of the active ingredient fluazifop-*P*-butyl. This is the application rate applied by coastal management agencies and typically yields 98% control of the plant over one year, reducing to 92% after three years without reapplication (Pritchard, 2005).

Samples of sediment and *S. anglica* leaves and roots were collected and sent to Advanced Analytical Australia Pty Ltd for measurement of fluazifop-*P*-butyl and the breakdown product fluazifop-*P*-acid. Sediment for herbicide testing was collected from treatment and control plots with a 3-cm diameter corer to 1 cm depth at 2 months prior to herbicide application and immediately after spraying (before the ensuing tidal inundation). Further sediment samples were collected at later dates from treatment plots only (Table 1). Unwashed leaves and rinsed roots were collected for herbicide testing from treatment plots at various dates on or after the day of spraying (Table 1).

Samples for macrofauna, *S. anglica* root mass, and sediment properties (organic matter and chlorophyll *a*) were collected from all plots 2 months prior to herbicide application ("baseline" samples, Australian autumn), then at 1 and 2 months (winter), 6 months (summer), and 19 months (next summer) following herbicide application. All samples were collected at the same elevation within the plots. In each plot on each sampling date, a 15-cm diameter sediment core was taken to 10 cm depth to measure macrofauna and root mass. Above-ground *S. anglica* biomass was removed prior to core collection. The sediment was sieved over a 500 μm mesh screen, and the fauna and roots retained were preserved in 5% formalin in seawater. Within 1 m of each large