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Research article

Herbicide effectiveness in controlling invasive plants under elevated CO_2: Sufficient evidence to rethink weeds management

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ABSTRACT

Previous studies have reported that chemical weed control will be less effective for some weed species under future atmospheric CO_2 concentrations. Such reductions in plant sensitivity to herbicides under elevated CO_2 may be due to greater biomass accumulation and differences among growth types. However, these studies have been limited to few growth types (herbaceous and grass species) and to a single herbicide (glyphosate).

This study tested a more extensive range of weed species (both in number and growth form) and herbicides to assess general patterns of plant response. We grew 14 environmental weed species representing four different growth forms (grasses, herbs, shrubs and vines), that are commonly found in south-eastern Australia, under ambient (380 ppm) and elevated (550 ppm) CO_2 concentrations. We then applied the recommended and double-recommended concentrations of two herbicides: glyphosate and fluroxypyr-meptyl. We found that responses of the weed species to herbicide under elevated CO_2 were species-specific. However, the C_3 grasses tended to be the most sensitive to herbicide application followed by the herbs and C_4 grasses while shrubs and vines demonstrated the highest resistance. Our results highlight the need for broader testing to determine the species most likely to exhibit increased tolerance to herbicide in the future in order to improve management options beforehand and thus offset a future liability.

1. Introduction

It has been predicted that ongoing climate change may advantage many weedy plant species due to their broad environmental tolerances and capacity for rapid dispersal and colonisation (Bajwa et al., 2018; Carboni et al., 2015; Mgidi et al., 2007; Seebens et al., 2015); although this may not be true for all weed species (O'Donnell et al., 2012; Roger et al., 2015). Furthermore, rising atmospheric CO_2 concentration and associated changes in disturbance regimes (*e.g.*, increased frequency and intensity of flood, drought and fire events), are likely to increase the invasion success of many weed species (Diez et al., 2012; Fernandino et al., 2018; Manea et al., 2016; Sorte et al., 2013; Sutherst, 2000). Therefore, weed management under future climates will need to incorporate measures to identify species likely to become problematic due to climate change, in addition to assessing and accounting for the associated risks (Downey et al., 2010a). Weed management must also put in place measures to mitigate against any potential adverse effects that may impact on the way by which weed species are currently managed *i.e.,* through physical, biological and chemical means, to ensure weed management adapts to the challenges associated with future climate change (Leishman and Gallagher, 2015). Given that chemical control using herbicides is the most widely used weed management technique worldwide (Fernando et al., 2016), there has been surprisingly little attention given to how the chemical control of weeds may be altered under future climate conditions (Dukes, 2000; Hellmann et al., 2008; Sutherland et al., 2017; Sutherst, 2000).

Rising atmospheric CO_2 concentration is one of the best documented global changes of the past half century (Jiménez et al., 2018; Prentice et al., 2001). As a result, plant responses to elevated CO_2 have been extensively studied over the past four decades (Ainsworth and Long, 2005; Amthor, 1995; Leakey et al., 2009; Morison and Gifford, 1984). These responses include a reduction in stomatal conductance and transpiration, improved water and nitrogen use efficiency and higher rates of C_3 photosynthesis (Leakey et al., 2009). Despite this, there have been very few studies that have attempted to link these biochemical, physiological, metabolic, and morphological shifts under elevated CO_2 to changes in herbicide efficacy on weed species (Fernando et al., 2016; Varanasi et al., 2016). Early studies that

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addressed this question (Archambault et al., 2001; Manea et al., 2011; Ziska et al., 2004; Ziska and Teasdale, 2000) reported that weedy grass and herbaceous species tended to display increased tolerance to the herbicide glyphosate when grown under elevated CO_2 (Table S1). However, subsequent studies (e.g., Jabran, 2016; Marble et al., 2015; Zhang et al., 2015) examining a different set of weedy grass, herbaceous and sedge species provided limited evidence to support these initial studies (Table S1). In addition to this, the mechanism by which herbicide efficacy may be altered under elevated CO_2 remains unclear. Perhaps the simplest explanation is that an increase in biomass under elevated CO_2, particularly in belowground biomass, may have a dilution effect on herbicides (Ziska et al., 1999; Ziska and Teasdale, 2000). However, Fernando et al. (2016) have subsequently argued that such morphological changes alone cannot explain the underlying mechanisms of herbicide resistance and that biochemical and physiological changes must also be examined. For example, altered enzymatic activity, pigment production, increased starch levels in C_3 plants and decreased protein levels have been shown to interfere with a range of herbicides (Patterson, 1995). This lack of consensus highlights the need for further research to determine the underlying mechanism determining herbicide tolerance under elevated CO_2. This may then enable us to shed some light on the species-specific responses we have observed to date. In a broader context, it is essential to address this knowledge gap as weed management under climate change not only has to account for changes in plant growth and performance but also potential changes in herbicide effectiveness, given that any increase in herbicide tolerance would have significant implications for future weed management options (Ziska and Dukes, 2011).

Of the thousands of weed species globally, we are aware of only 21 weed species that have been examined for changes in herbicide tolerance under elevated CO_2 (Table S1). Furthermore, these 21 weeds are confined to either grasses or herbaceous species which are weedy mostly in agricultural systems; a focus which has flowed through to a restricted discussion on the outcomes, focusing on crop-weed management (Naidu, 2015; Varanasi et al., 2016; Ziska, 2016; Ziska and McConnell, 2015). It is not feasible to investigate every weed species and herbicide-specific responses to elevated CO_2 and consequently broader approaches to understanding the likely outcomes are required. Thus, the aims of this study were two-fold. Firstly, to extend the existing knowledge on herbicide efficacy under elevated CO_2 to a larger number of weed species by assessing growth and survival of 14 common environmental weed species of south-eastern Australia treated with recommended herbicides under ambient and elevated CO_2 conditions. These 14 weed species span a wide range of growth forms (C_3 and C_4 grasses, herbs, vines and shrubs) to provide a more representative assessment of weed flora. Secondly, to investigate if a higher dose of herbicide would be needed in the future to offset the effects of elevated CO_2 on herbicide tolerance. We hypothesized that herbicide efficacy under elevated CO_2 is likely to decrease.

2. Materials and methods

2.1. Species selection

The 14 selected weed species encompassed a range of plant families and growth forms (Table 1). *A. cordifolia*, *A.s aethiopicus* and *L. camara* are listed as Weeds of National Significance in Australia while *A. adenophora* is a declared weed in some areas of Australia. All of the other species are considered environmental weeds (Downey et al., 2010b; Skarratt et al., 1994). Two of these 14 plant species (*C. gayana* and C. *clandestinum*) are C_4 species and the remainder are C_3 species. The 14 environmental weed species were represented by two vine, three herb, four shrub and five grass species (Table 1).

2.2. Plant preparation

Plants were propagated from seed, or cuttings or tubers when seeds were not available, which were all collected in the greater Sydney region, Australia (Table 1). Seeds, sampled from multiple plants, were germinated on moist filter paper at room temperature. Seedlings at the stage of cotyledon emergence were transplanted into 700 mL pots filled with commercial potting mix (Australian Native Landscapes, Terrey Hills, NSW, Australia) and 10 g of slow release fertilizer (16:3.9:10 N:P:K; Osmocote, Gordon, NSW, Australia). The pots were lined with newspaper to prevent soil loss through the drainage holes. Species propagated vegetatively were planted directly into pots (Table 1). *A. cordifolia* was grown from tubers while *L. camara*, *T. fluminensis,* and *I. indica* were propagated from stem fragments approximately 100 mm long. All plants were grown between December 2011 and July 2012.

2.3. Experimental design

The experiment followed a randomised fully factorial design, with the factors being CO_2 concentration (ambient or elevated) and herbicide treatment (recommended and double recommended label rate). Four glasshouses were used: two at the ambient and two at the elevated CO_2 concentration.

Ten replicates of each weed species for each CO_2 × herbicide treatment combination were grown. These were evenly split between the treatment glasshouses. Additionally, six replicates of each weed species were grown under each CO_2 treatment to assess the biomass allocation of each species at the time of herbicide application. This could not be done after herbicide treatment due to plant mortality. These plants were harvested into their above- and belowground components on the day of herbicide application and oven-dried at 60 °C until the weight remained constant (48–72 h) before being weighed.

Pots were randomly rearranged within the glasshouses each fortnight to minimise any within-glasshouse effects. All pots were evenly spaced to minimise shading from neighbouring plants. As *L. camara* and *I. indica* were propagated from cuttings, they were re-potted into 2.8 L pots after eight weeks and six weeks respectively to allow them ample space for root development. The vine species *A. cordifolia* and *I. indica* were trained onto stakes. Pots were mist watered for one minute four times daily.

The elevated CO_2 treatment was maintained by a dosing and monitoring system (Canary Company Pty Ltd, Lane Cove, NSW, Australia) at 550 ppm, from 6 am to 6 pm, with air continuously circulated within each glasshouse. The elevated CO_2 treatment represents the predicted atmospheric CO_2 concentration by 2030 under most emissions scenarios (IPCC, 2001). The ambient CO_2 treatment was 380 ppm. The glasshouse temperature was set to 17 °C at night and 24 °C during the day.

2.4. Herbicide application

We reviewed the weed control literature and herbicide manufacturer guidelines for recommended application rates of glyphosate and other commonly used herbicides for each of the 14 weed species to determine the recommended or label rate and double rate. Half of the replicates for each of the 14 weed species were sprayed individually with glyphosate (Accensi Pty, Narangba, QLD, Australia) at the recommended concentration or label rate for that species (Table 1) and the other half of the replicates were treated with double the recommended rate (n = 10). In each case, 3 mL of the surfactant LI-700 (Nufarm, Hunter Valley, NSW, Australia) was added per litre of herbicide mixture. Three species (*i.e., T. fluminensis, L. camara* and *A. cordifolia*) were also treated with fluroxypyr-meptyl (Starane Advanced, Dow Agro Sciences, Frenchs Forest, NSW, Australia), at the recommended and double recommended rate (n = 10), as these species