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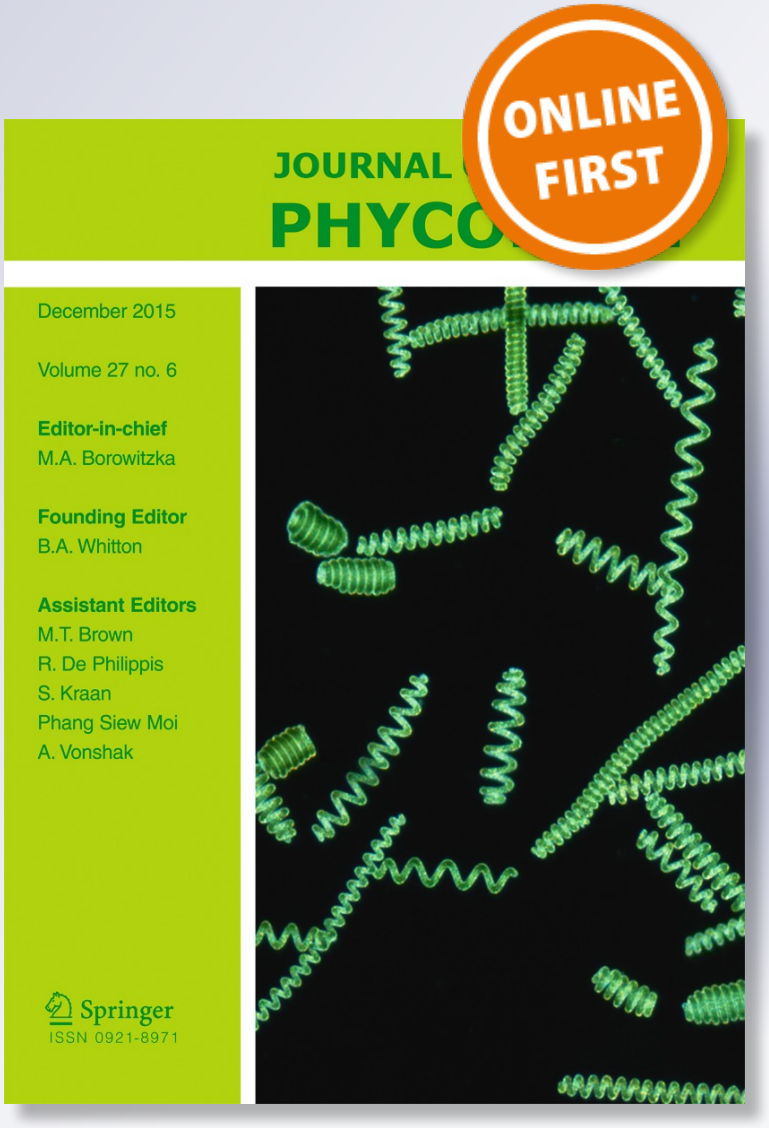

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Glyphosate herbicide toxicity to native Hawaiian macroalgal and seagrass species

Ronald Paul Kittle III[1] · **Karla J. McDermid**[1]

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Abstract Glyphosate-based herbicides are commonly used to combat weeds and unwanted grasses in many habitats in the Hawaiian Islands, including near freshwater, marine, and anchialine pond shorelines. Glyphosate is reported to degrade within a few days of application and to break down rapidly in soil, which suggests that it is safe for use near aquatic environments. However, glyphosate can be transported to coastal waters, especially during run-off events. Five native macroalgael and seagrass species and one introduced aquatic vascular plant found in coastal anchialine ponds or in the adjacent intertidal zone were exposed to freshly mixed solutions of a glyphosate-based herbicide in lab experiments. Chlorophyll absorbance and photosystem II (PSII) efficiency were measured after 5 to 7 days of incubation. At herbicide concentrations (0.225 to 1.8 g L^{-1} glyphosate) below the manufacturer's lowest recommended concentration (3.6 g L^{-1} glyphosate), chlorophyll absorbance and PSII efficiency differed significantly from the control (0.0 g L^{-1} glyphosate). Native macroalgae and seagrasses in marine and anchialine aquatic habitats may be negatively affected by use of glyphosate herbicides to control shoreline weeds.

Keywords Glyphosate · Herbicide toxicity · Macroalgae · Seagrass · Hawaii · Pulse amplitude-modulated (PAM) fluorometry · Chlorophyll · Photosystem II

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Introduction

Within the last century, agricultural methods to control noxious weeds shifted from active tillage, alteration of the soil pH, or increase of soil salinity, to a greater reliance on chemical herbicides (Franz et al. 1997). Glyphosate or N-(phosphonomethyl) glycine ($C_3H_8NO_5P$, molecular weight 169.07 g $mole^{-1}$) was developed in 1974, and is utilized in a wide range of applications including weed control in croplands, vineyards, olive groves, fruit orchards, grass pastures, forests, parks, gardens, urban sidewalks and streets, railroad tracks, and underwater in rivers and lakes. As of 2013, glyphosate was the most commonly used herbicide in the world. In the USA, over 750 products with glyphosate as the active ingredient are available (Newton 2013). In 2011, worldwide usage of glyphosate products was 650,000 t (CCM International Ltd 2012); the 2012 global market for the active ingredient glyphosate was 718,600 t just for agricultural applications (Transparency Market Research 2013). Glyphosate usage is predicted to rise to 1.35 million t by 2017 (Global Industry Analyst 2011).

Glyphosate is absorbed through foliage and translocated to the growing points of plants, where it inhibits a key enzyme, 5-enolpyruvylshikimate-3-phosphate (EPSP) synthase, within the shikimate pathway. This is a seven-step metabolic pathway in plants, and some bacteria and fungi for the biosynthesis of aromatic amino acids (phenylalanine, tyrosine, and tryptophan) that are essential for protein synthesis (Franz et al. 1997). The death of the plants exposed to glyphosate involves growth inhibition, wilting, and then necrosis of the tissues (Franz et al. 1997). There have been reports of glyphosate resistance in plants and microorganisms due to the high usage of glyphosate for weed control (Franz et al. 1997; López-Rodas et al. 2007; Mamy et al. 2010). As a result, many agricultural facilities are using higher concentrations of

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glyphosate, in combination with other herbicides, such as 2,4-dichlorophenoxyacetic acid (2-4D), to combat the resistant species (Franz et al. 1997).

Glyphosate can be adsorbed onto sediment particles and inactivated (Andréa et al. 2003). Glyphosate, not bound to soil, can be broken down by microbes into aminomethylphosphate (AMPA) and CO_2 (Balthazor and Hallas 1986; Franz et al. 1997; Schuette 1998). The half-life of glyphosate in soil ranges from 2 to 197 days, and its half-life in freshwater is 3 to 91 days (U.S.EPA 1993). In flask experiments, the half-life for glyphosate in seawater with marine bacterial populations was 47 days (25 C in low-light) up to 310 days (31 °C in the dark) (Mercurio et al. 2014), leading to the conclusion that "little degradation would be expected during flood plumes in the tropics, which could potentially deliver dissolved and sediment-bound glyphosate" (p. 385) into the marine environment. Low levels of glyphosate were detected in Marennes–Oléron Bay along the Atlantic coast of France, during an 11-day period (maximum concentration = 1.2 μg L^{-1}) of a run-off event in the spring 2004 (Burgeot et al. 2008), indicating that measurable amounts of glyphosate can occur in coastal waters. Glyphosate and AMPA were also detected in water samples from ten estuaries in the Baltic Sea in 2012 (Skeff et al. 2015). These studies show that glyphosate and its primary metabolite can be transported to coastal environments where marine species may be exposed to these contaminants.

Studies have tested the effects of glyphosate-based herbicides on non-target aquatic species. Freshwater microalgae (Hernando et al. 1989; Sáez et al. 1997; Ma 2002; Pechlaner 2002; Pérez et al. 2007; Kyriakopoulou et al. 2009; Vendrell et al. 2009; Lipok et al. 2010; Saxton et al. 2011) and freshwater macrophytes (Perkins 1997; Cedergreen and Streibig 2005) are sensitive to glyphosate-based herbicides. The concentrations of glyphosate-based RoundUp® that caused 50 % mortality (LC_{50}) in submerged vascular freshwater plants, such as *Myriophyllum aquaticum* (Turgut and Fomin 2002) and *Myriophyllum spicatum* (Lewis 1995), were similar to those of the floating freshwater plant, duckweed (*Lemna minor*). Stachowski-Haberkorn et al. (2008) found that low concentrations (~1 μg L^{-1}) of glyphosate reduced species diversity of coastal marine microbial communities. Although animals lack the shikimate pathway, toxicology studies on amphibians (Clements et al. 1997; Relyea 2012), on freshwater and marine invertebrates (Folmar et al. 1979; Mottier et al. 2015), and on freshwater and marine fish (Mitchell et al. 1987; Guilherme et al. 2012; Yusof et al. 2014) have reported anatomical deformities and DNA damage after short-term exposure to glyphosate-based herbicides.

Studies on glyphosate-based herbicide toxicity to marine flora are few. Castro et al. (2015) reported significant changes in biomass, leaf growth, and chlorophyll content of a brackish water seagrass, *Ruppia maritima* Linnaeus, after exposure to glyphosate-based herbicide. The impact of varying concentrations of glyphosate-based RoundUp® on kelp (marine brown seaweeds in the order Laminariales) has been assessed by measuring cell viability in gametophytes (R.J. Lewis, personal communication). The LC_{50} (the concentration of a substance that kills 50 % of cells exposed to it) ranged from 2×10^{-5} to 3.7×10^{-4} M RoundUp® which is equivalent to 4.56 to 84.4 mg L^{-1} glyphosate. Pang et al. (2012) soaked two red seaweeds in 0–4 g L^{-1} glyphosate solutions for 1 to 5 min and reported "great changes" in chl *a* fluorescence curves, reduced PS II efficiency, pigment loss, withering, and rhizoid detachment in the filamentous, epiphytic species, *Neosiphonia savatieri* (Hariot) M.S. Kim & I.K. Lee (order Ceramiales). In contrast, in the large, fleshy carrageenophyte, *Kappaphycus alvarezii* (Doty) Doty ex P.C. Silva (order Gigartinales), only "negligible" changes in chl *a* fluorescence curves and "no visible harm" to thalli were observed. Based on their results, Pang et al. (2012) recommended the use of glyphosate to control epiphytic filamentous algae in *Kappaphycus* mariculture farms.

In early 2012, the County of Hawaii proposed to spray AquaPro® around the anchialine ponds (brackish water ponds with no surface connection with the sea that fluctuate in depth and salinity during the tidal cycle) at Richardson Ocean Park, Hilo, HI (B. Wilkins, personal communication). AquaPro®, manufactured for SePRO Corporation by DowAgro Sciences LLC is a glyphosate-based systemic herbicide for broad-spectrum non-selective shoreline weed and grass control. The active ingredient formulation of AquaPro® consists of 53.8 % glyphosate: N-(phosphonomethyl) glycine, isopropylamine salt, which is the same concentration as RoundUp® (SePRO 2001). SePRO advertises that AquaPro® is specially formulated for use around aquatic environments and will not harm fish. The AquaPro® material safety data sheet lists the effective concentration for 50 % growth inhibition (EC_{50}) for duckweed as 24.4 mg L^{-1} AquaPro®, and the EC_{50} for the freshwater green alga, *Selenastrum capricornutum*, as 127 mg L^{-1} AquaPro® (SePRO 2001).

Anchialine ponds are an important habitat in the Hawaiian Islands for many native species, including macroalgae and one species of seagrass (Stock et al. 1986). Anchialine ponds and the adjacent nearshore waters may be exposed to glyphosate through direct or indirect herbicide spraying, as well as run-off during storm events. This study focused on five native species: *Gayralia oxysperma* (Kützing) K.L. Vinogradova ex Scagel et al. (Chlorophyta, Ulothricales), *Rhizoclonium riparium* (Roth) Harvey (Chlorophyta, Cladophorales), *Ulva intestinalis* Linnaeus (Chlorophyta, Ulvales), and *R. maritima* Linnaeus (Tracheophyta, Alismatales), which are found in the anchialine ponds, and *Pterocladiella capillacea* (Gmelin) Santelices & Hommersand (Rhodophyta, Gelidiales), which is found along the shore on intertidal rock, adjacent the anchialine ponds. *Myriophyllum aquaticum* (Vell.) Verdc. (Tracheophyta, Saxifragales) or water parrotfeather milfoil, an invasive

submerged aquatic plant found in the ponds, was also included in this study. Our objective was to evaluate the sensitivity of native marine macroalgal and seagrass species to glyphosate herbicide by using two indicators of cell health and plant stress: chlorophyll absorbance and photosystem II efficiency.

Methods

Study site

Richardson Ocean Park, also known as Wai'uli, located near Hilo, Hawai'i, USA (19.734722°N, 155.013611°W) on the windward side of Hawai'i Island, is composed of diverse habitats: shallow water, rocky basalt terrain, black sand pocket beaches, strong wave action, and cold freshwater seeps. Near the landward entrance to the site are many brackish anchialine ponds, whose water level and salinity fluctuate with the tidal cycle due to subterranean connections with the ocean. This area receives a mean annual rainfall of 3303 mm, experiences approximately 275 days year^{-1} of precipitation (Giambelluca et al. 2013), and is subject to seasonal flash flooding and run-off. Approximately 138,000 visitors use Richardson Ocean Park annually (Kearns 2008).

Six aquatic species were collected in the anchialine ponds or in the intertidal zone on the shoreline at Richardson Ocean Park: *G. oxysperma*, *R. riparium*, *U. intestinalis*, *P. capillacea*, *R. maritima*, and *M. aquaticum*. Samples were transported in an insulated container to the laboratory in the Marine Science Building at the University of Hawaii-Hilo within 60 min of collection, and were quickly rinsed with filtered ambient water (33 ppt for *P. capillacea* and 5 ppt for all other species) three times to remove epiphytes, debris, and organisms. Specimens were identified, based on their morphology, using taxonomic references (Wagner et al. 1990; Abbott 1999; Abbott and Huisman 2004; Huisman et al. 2007; Imada 2012). Voucher specimens were prepared as microslides and on herbarium paper for future reference and deposited at B.P. Bishop Museum (BISH) in Honolulu. Samples were blotted with paper towels for 6 s and weighed into 1 g portions just prior to the experiments.

Initial experiment

Samples (1 g wet weight) of *R. riparium* and *R. maritima* were placed in individual sterilized glass containers containing one of five different concentrations (0.9, 1.8, 3.6, 7.2, and 36 g L^{-1}) of glyphosate in 250 mL of filtered brackish (5 ppt) water and the control (0.0 g L^{-1} glyphosate) of 250 mL brackish water. These concentrations were dilutions or amplifications based on the manufacturer's lowest recommended concentration (0.75 % AquaPro® = 3.6 g L^{-1} glyphosate) for non-woody weed control (AquaPro® label). No surfactant was added. Samples were incubated for 7 days with aeration, at 25 °C under fluorescent lights (24.98 μmol photons m^{-2} s^{-1}) on a 12:12-day length cycle. Three containers of each of the two species were used for each concentration and the control. Positions of the containers in the incubation area were randomized using a random number generator.

At the start of the experiment, an additional 1 g wet weight of each species was immersed in 10 mL 90 % methanol solution for 48 h in the dark. The liquid extract was refrigerated at 4 °C. A 5-mL portion of the extract was analyzed for chlorophyll absorbance. Absorbance was read at 664 nm in a Beckman DU-600 spectrophotometer. At the end of the experiment, chlorophyll was extracted from each sample exactly as from the pre-experiment samples, and chlorophyll absorbances were compared to the initial values. The purpose was

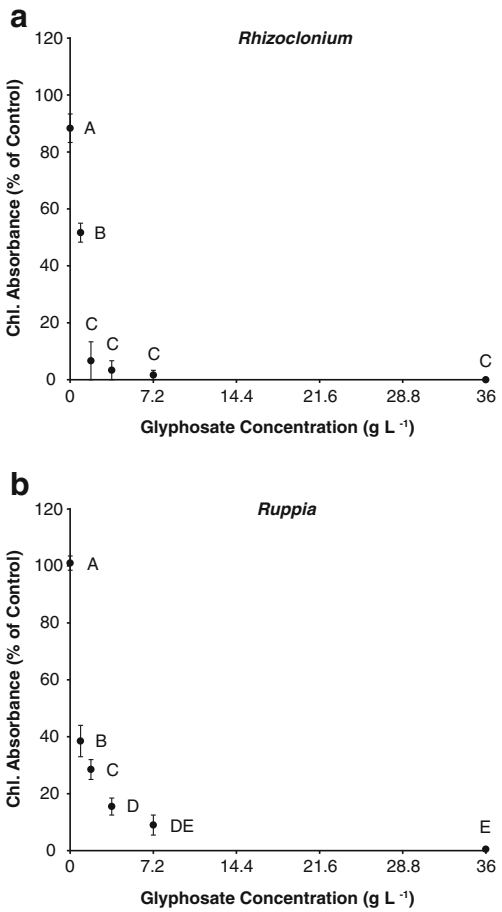

Fig. 1 **a** Mean chlorophyll absorbance for *Rhizoclonium riparium* after 7 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 323.03$, df = 17). **b** Mean chlorophyll absorbance for *Ruppia maritima* after 7 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 361.64$, df = 17)

not to determine total chlorophyll content, but to measure relative changes in chlorophyll content.

Low concentrations experiment

Samples (1 g wet weight) of all six species were placed in individual sterile glass containers containing one of five different concentrations (0.225, 0.45, 0.9, 1.8, and 3.6 g L^{-1}) of glyphosate in 250 mL of either filtered brackish water (5 ppt) for Chlorophyta and Tracheophyta species, or filtered seawater (33 ppt) for *Pterocladiella*. The control samples of all six species were placed in individual sterile glass containers with 0.0 g L^{-1} glyphosate in 250 mL of filtered brackish water or seawater. Three containers of each of the six species were used for each concentration and the control. Positions of the containers in the incubation area were randomized using a random number generator. The concentrations were chosen based on the results from the initial experiment and on the manufacturer's lowest recommended concentration for non-woody weed control (AquaPro® label). No surfactant was added. Samples were incubated for 5 days with aeration, at 25 °C under fluorescent lights (24.98 μmol photons m^{-2} s^{-1}) on a 12:12–day length cycle. After incubation, chlorophyll absorbance was assessed as described in the previous experiment.

Photosynthetic yield experiment

Prior to collection, photosynthetic yield (F_v'/F_m') (Cosgrove and Borowitzka 2010) of thalli of the six species was measured in situ in triplicate using a submersible photosynthesis

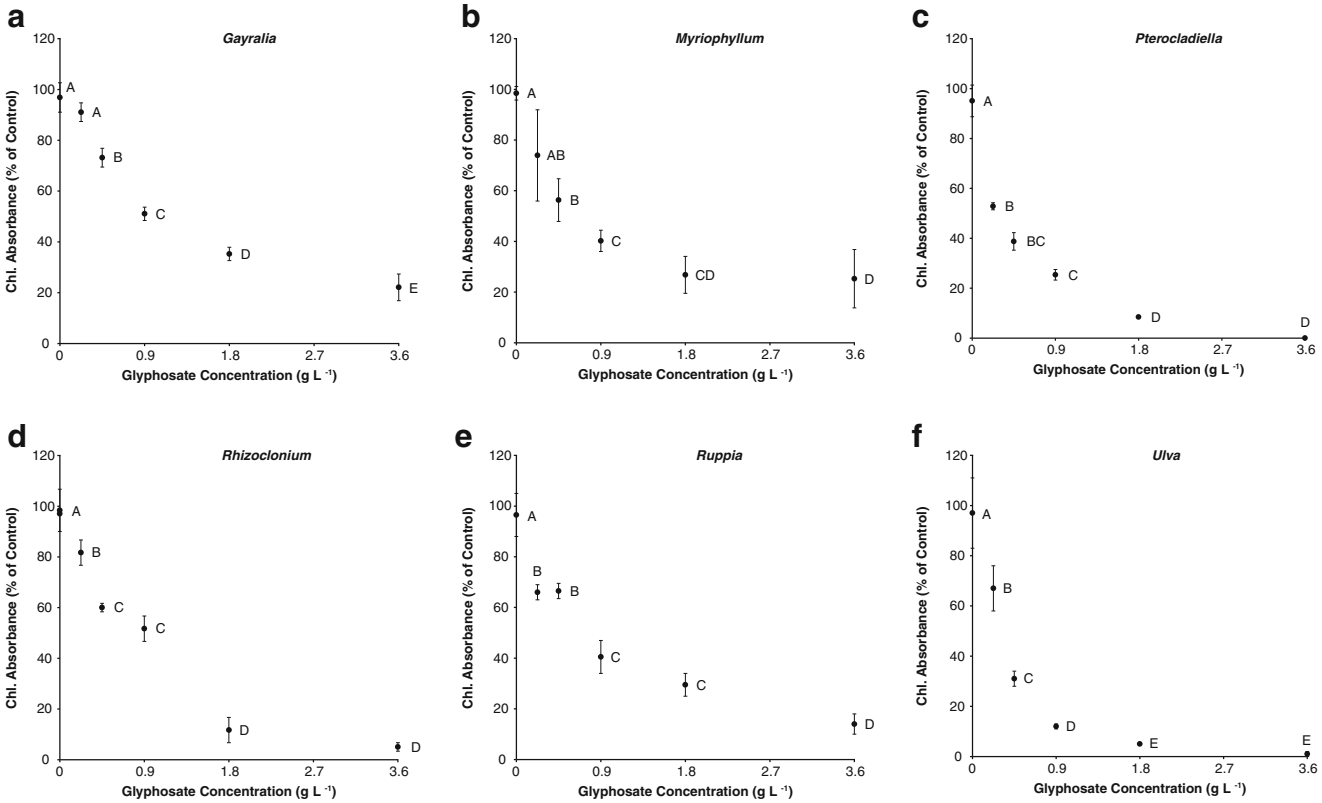

Fig. 2 **a** Mean chlorophyll absorbance for *Gayralia oxysperma* after 5 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 160.64$, df = 17). **b** Mean chlorophyll absorbance for *Myriophyllum aquaticum* after 5 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 25.03$, df = 17). **c** Mean chlorophyll absorbance for *Pterocladiella capillacea* after 5 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 25.41$, df = 17). **d** Mean chlorophyll absorbance for *Rhizoclonium riparium* after 5 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 165.01$, df = 17). **e** Mean chlorophyll absorbance for *Ruppia maritima* after 5 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 96.23$, df = 17). **f** Mean chlorophyll absorbance for *Ulva intestinalis* after 5 days incubation with glyphosate herbicide expressed as percentage of initial pre-incubation value. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 47.23$, df = 17)

yield analyzer, pulse amplitude-modulated (PAM) fluorometer (Heinz Walz). F_v'/F_m' was again measured in triplicate in the lab within 1 h of collection before exposure to glyphosate. Samples (1 g wet weight) of each of the six species were placed in individual sterilized glass containers containing one of five different concentrations (0.225, 0.45, 0.9, 1.8, and 3.6 g L^{-1}) of glyphosate in 250 mL of either filtered brackish (5 ppt) water or seawater and the control (0.0 g L^{-1} glyphosate) of 250 mL filtered brackish water or seawater. No surfactant was added. One container of each of the six species was used for each concentration and the control. Positions of the containers in the incubation area were randomized using a random number generator. After 5 days of incubation under the same conditions as previous experiments, photosynthetic yield measurements were re-assessed in triplicate. The control used the same PAM settings for all experiments to maintain consistency. PSII inhibition was expressed as percentage of the control using similar methods as Sjollema et al. (2014).

Statistical analyses

Results of all experiments were subjected to analysis of variance test (ANOVA) and Tukey tests with an a priori significance level of $\alpha = 0.05$.

Results

Initial experiment

In both species, *R. riparium* and *R. maritima*, chlorophyll absorbance in samples incubated with glyphosate was

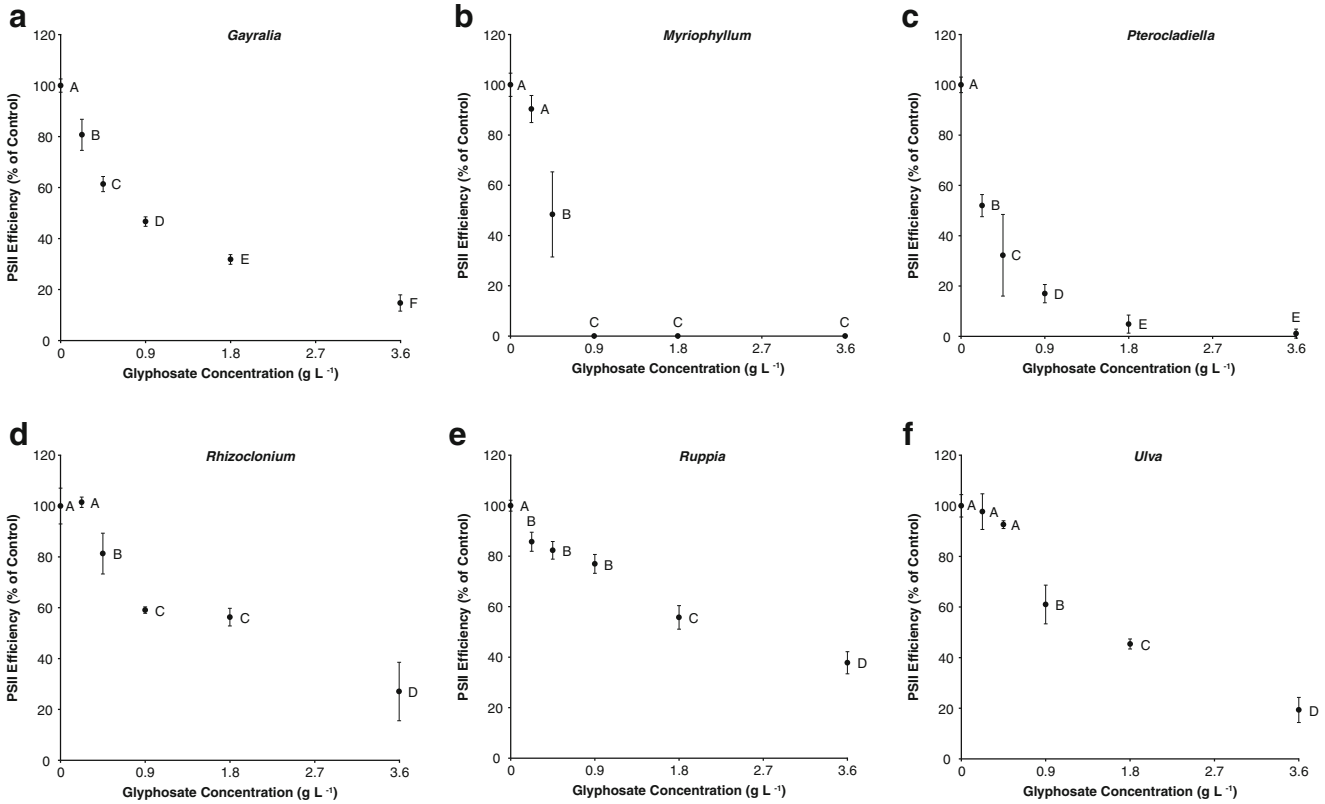

Fig. 3 **a** Dose-response relationships for glyphosate on the photosynthetic yield (% of control, $\bar{x} = 0.663$) of *Gayralia oxysperma* after 5 days of incubation. Error bars represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 269.32$, df = 17). **b** Dose-response relationships for glyphosate on the photosynthetic yield (% of control, $\bar{x} = 0.580$) of *Myriophyllum aquaticum* after 5 days of incubation. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 113.62$, df = 17). **c** Dose-response relationships for glyphosate on the photosynthetic yield (% of control, $\bar{x} = 0.608$) of *Pterocladiella capillacea* after 5 days of incubation. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 215.16$, df = 17). **d** Dose-response relationships for glyphosate on the photosynthetic yield (% of control, $\bar{x} = 0.511$) of *Rhizoclonium riparium* after 5 days of incubation. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 63.51$, df = 17). **e** Dose-response relationships for glyphosate on the photosynthetic yield (% of control, $\bar{x} = 0.630$) of *Ruppia maritima* after 5 days of incubation. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 112.77$, df = 17). **f** Dose-response relationships for glyphosate on the photosynthetic yield (% of control, $\bar{x} = 0.544$) of *Ulva intestinalis* after 5 days of incubation. *Error bars* represent standard deviation. $n = 3$ for each treatment. *Shared letters* denote no significant difference between means at $p < 0.005$ (Tukey test $F = 212.88$, df = 17)

significantly lower than initial values ($p < 0.05$) (Fig. 1a, b). Even at the lowest concentration of glyphosate (0.9 g L^{-1}), chlorophyll absorbance was reduced by approximately 50 % of the initial pre-incubation value. Concentrations above the manufacturer's lowest recommended concentration of 3.6 g L^{-1} caused loss of thallus integrity; thus, for the following experiments, lower concentrations were used to assess responses.

Low-concentrations experiment

Chlorophyll absorbance in all species was significantly lower in samples incubated with 0.45 g L^{-1} glyphosate than initial values ($p < 0.05$) (Fig. 2a–f). Chlorophyll absorbances of controls (0 g L^{-1} glyphosate) incubated for 5 days were not significantly different from the chlorophyll absorbances measured before incubation. However, *Rhizoclonium*, *Ulva*, and *Pterocladiella* showed significantly reduced chlorophyll absorbance at 3.6 g L^{-1} glyphosate. All species had at least a 50 % reduction in chlorophyll absorbance at 0.9 g L^{-1} glyphosate.

Photosynthetic yield experiment

Photosynthetic yield (F_v'/F_m') values measured before collection of the thalli did not differ significantly from values measured an hour later in the laboratory just prior to the start of the incubation (*t* test, $p > 0.97$). Dose-response relationships (see Seefeldt et al. 1995) of the effect of glyphosate on photosynthetic yield of all six species were evident (Fig. 3a–f). Even when incubated with 0.9 g L^{-1} glyphosate, all species showed photosynthetic yields that were significantly lower than the initial pre-incubation values. Controls (0.0 g L^{-1} glyphosate) incubated 5 days had photosynthetic yields that were not significantly different from the F_v'/F_m' values recorded prior to incubation (*t* test, $p > 0.97$).

Discussion

Although the sensitivity to glyphosate varied among the six species tested, we observed a reduction in chlorophyll for all species even at glyphosate concentrations as low as 0.45 g L^{-1}. In our study, PAM fluorometry proved to be a sensitive tool for measuring subtle effects of a glyphosate-based herbicide on macroalgae and seagrass (Enriquez and Borowitzka 2010), similar to other PAM fluorometry studies on microalgal responses to herbicides (Magnusson et al. 2008; Sjollema et al. 2014). However, unlike the red macroalga, *K. alvarezii* (Pang et al. 2012), all six Hawaiian species tested, *G. oxysperma*, *R. riparium*, *U. intestinalis*, *P. capillacea*, *R. maritima*, and *M. aquaticum*, showed photosynthetic yields that were significantly lower than the initial pre-incubation values at concentrations well below the manufacturer's lowest recommended concentration for non-woody weed control (0.75 % AquaPro® = 3.6 g L^{-1} glyphosate). Based on changes in chlorophyll content and photosynthetic efficiency in response to glyphosate exposure, the six species were ranked in terms of their sensitivity to glyphosate. *Pterocladiella capillacea* had the greatest sensitivity ranking, while *G. oxysperma* was least sensitive (Fig. 4).

Seaweeds and seagrasses in the Hawaiian Islands provide critical habitat and food resources to many marine organisms, including sea urchins, gastropods, and herbivorous fish (Hoover 2012, 2014). The diet of green turtles (*Chelonia mydas* Linneaus), the largest marine herbivore in the Hawaiian Archipelago, includes more than 275 species of macroalgae and two species of seagrasses (Russell and Balazs 2000). *Pterocladiella capillacea*, which showed measurable responses to glyphosate exposure, is one of the ten macroalgal species that dominate the diet of the green turtles in the Hawaiian Islands (Balazs 1980; Arthur and Balazs 2008).

Although Solomon and Thompson (2003) viewed overwater uses of glyphosate as low risk to aquatic organisms, Vera et al. (2010) documented ecological shifts to cyanobacteria-favored mesocosms after 42 days of exposure to glyphosate. Similarly, concentrations of glyphosate as low as 1 μg L^{-1} (equivalent to levels measured in coastal waters) reduced species richness, lowered species diversity, and significantly changed species composition of natural marine planktonic microbial communities (Stachowski-Haberkorn et al. 2008). Long-term spraying of herbicides near seagrass and macroalgal populations in Japanese lagoons, resulted in alteration of water quality and phase-shifts from macroalgae-rich habitats to cyanobacteria- and diatom-dominated areas (Yamamuro 2012). In addition, studies have reported glyphosate

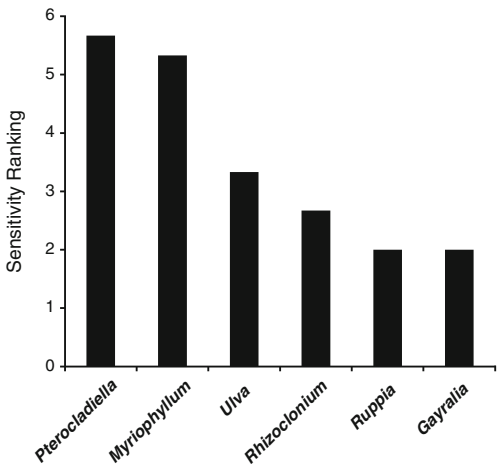

Fig. 4 Glyphosate sensitivity for six Hawaiian marine plant species, ranked from most to least sensitive, based on changes in chlorophyll content and PSII efficiency in response to exposure to glyphosate-based herbicide

persistence in estuarine ponds (Tsui and Chu 2008), and in seawater (up to 315 days) (Mercurio et al. 2014). The ecosystem-level effects of glyphosate herbicide use in and around anchialine ponds and nearshore habitats in the Hawaiian Islands are not known. However, this study shows that native macroalgae and seagrasses in marine and anchialine aquatic habitats may be negatively affected by use of glyphosate herbicides to control shoreline weeds. Based on these data, we strongly recommend that alternative methods of weed control be used around anchialine ponds and shorelines, especially in areas subject to flooding and run-off.

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