Non-target Impacts to Eelgrass from Treatments to Control Spartina in Willapa Bay, Washington

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ABSTRACT

Four methods to control the smooth cordgrass Spartina (*Spartina alterniflora*) and the footwear worn by treatment personnel at several sites in Willapa Bay, Washington were evaluated to determine the non-target impacts to eelgrass (*Zostera japonica*). Clone-sized infestations of Spartina were treated by mowing or a single hand-spray application of Rodeo® formulated at 480 g L^{-1} acid equivalence (ae) of the isopropylamine salt of glyphosate (Monsanto Agricultural Co., St. Louis, MO; currently Dow AgroSciences, Indianapolis, IN) with the non-ionic surfactant LI 700® (2% v/v) or a combination of mowing and hand spraying. An aerial application of Rodeo® with X-77 Spreader® (0.13% v/v) to a 2-ha meadow was also investigated. Monitoring consisted of measuring eelgrass shoot densities and percent cover pre-treatment and 1-yr post-treatment. Impacts to eelgrass adjacent to treated clones were determined 1 m from the clones and compared to a control 5-m away. Impacts from footwear were assessed at 5 equidistant intervals along a 10-m transect on mudflat and an untreated control transect at each of the three clone treatment sites. Impacts from the aerial application were determined by comparing shoot densities and percent cover 1, 3 and 10 m from the edge of the treated Spartina meadow to that at comparable distances from an untreated meadow. Methods utilized to control Spartina clones did not impact surrounding eelgrass at two of three sites. Decreases in shoot densities observed at the third site were consistent across treatments. Most impacts to eelgrass from the footwear worn by treatment personnel were negligible and those that were significant were limited to soft mud substrate. The aerial application of the herbicide was associated with reductions in eelgrass (shoot density and percent cover) at two of the three sampling distances, but reductions on the control plot were greater. We conclude that the unchecked spread of Spartina is a far greater threat to the survival and health of eelgrass than that from any of the control measures we studied. The basis for evaluating control measures for Spartina should be efficacy and logistical constraints and not impacts to eelgrass.

Key words: estuary, glyphosate, *Zostera eelgrass, Spartina alterniflora,* Rodeo®, non-target.

INTRODUCTION

Seagrasses are recognized as important components of coastal and estuarine ecosystems (Hemminga and Duarte 2000). In the Pacific Northwest and on Canada's Pacific and

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Atlantic coasts, the native eelgrass marina (*Zostera marina)* provides protection or food for many crustacea, fish and birds (Harrison and Bigley 1982). Included are species of commercial importance such as Dungeness crabs (*Cancer magister*) and Pacific herring (*Clupea harengus pallasi*) (Spencer 1932; Taylor 1964). However, seagrasses are declining worldwide from anthropogenic activities (Short and Wyllie-Echeverria 1996). These activities are resulting in deterioration of light and sediment conditions, physiological stresses, or physical uprooting.

The eelgrass japonica (*Zostera japonica)*, most likely introduced with shipments of Japanese oysters *(Crassostrea gigas)* to Willapa Bay and northern Puget Sound, was first recorded on the Washington State coast in 1957 (Hitchcock et al. 1969). Japonica quickly colonized inter-tidal mudflats along the Oregon and Washington coasts (Harrison and Bigley 1982) and has since naturalized, does not compete with the native marina due to a difference in tidal zone preference (Harrison 1979), and is believed to have contributed positively to both the health and biodiversity of the Bay (Posey 1988).

In a recent study to determine the efficacy of different treatments to control the non-native, invasive cordgrass Spartina (*Spartina alterniflora)* in Willapa Bay, Washington (Major et al. 2003a), data were simultaneously collected to assess off-target impacts to japonica. Because japonica and Spartina overlap extensively in their intertidal ranges, efforts to control Spartina may impact this eelgrass. While it is known that Spartina, if left uncontrolled, effectively out competes eelgrass over time, it is still important to consider the immediate impacts to japonica of control measures in developing a Bay-wide strategy for Spartina control. Herein, we compare the impacts on japonica (hereafter eelgrass) of four techniques to control Spartina in the Bay, mowing, hand spraying with the herbicide glyphosate, and a mow/spray combination for Spartina clones; and aerial application of glyphosate for Spartina meadows.

METHODS

One study site within each of three different substrate types (mud, sand, mixed sand/mud) was chosen in order to assess non-target impacts to eelgrass of treatments to control Spartina clones. Spartina clones are circular patches greater than 1 m in diameter and generally assumed to have arisen from a single shoot and spread rhizomotously. Clones may grow indefinitely in size until they coalesce with surrounding clones to form a single, larger meadow. Control strategies at each site were chosen based upon the mandates of the participating management agencies, degree of Spartina infestation, substrate type and location within the Bay (Major et al. 2003a). The Lewis Unit was located at the southernmost part of the Bay on the Willapa National Wildlife Refuge (WNWR) at ca. 46,20 N-124,00 W and was characterized by a deep, soft muddy substrate. Eelgrass shoot density (shoots 0.25 m$^{2-1}$) and percent cover of eelgrass within the treatment area averaged 50.7 (SE = 16.2) and 0.7 (SE = 0.2), respectively. The Nemah Beach site was located mid Bay at approximately 46,35 N-123,55 W and was characterized by a hard packed, sand substrate with an underlying clay layer at higher tidal elevations. The study site extended ca. 1.2 km north and south along the beach.

Shoot density and percent cover of eelgrass averaged 255.4 (SE = 26.2) and 3.3 (SE = 0.3), respectively. The North River site was located at the north end of the Bay at 46,45 N-124,00 W and ca. 0.5 km SW of the confluence of Smith Creek and the North River. The substrate was a mixture of sand and mud. There were a few small channels draining the site, but none more than 0.5 m deep. Shoot density and percent cover of eelgrass averaged 91.9 (SE = 12.1) and 1.4 (SE = 0.2), respectively.

Kaffee Meadow, located on the northeast side of Long Island at approximately 46,30 N-123,55 W between Kaffee and Lewis Sloughs, was selected to study the non-target impacts from aerial application of the herbicide. The substrate was soft mud and a number of deep (1 to 2 m) channels drained the site. The study site consisted of two, 2-ha plots (treatment and control) within the meadow including the mudflat (eastern) edge of the meadow where the eelgrass shared space with a natural set of Pacific oysters (*Crassostrea gigas*). Shoot density and percent cover of eelgrass averaged 284.5 (SE = 74.9) and 14.8 (SE = 3.5) and 525.9 (SE = 63.8) and 28.5 (SE = 3.1) on the treatment and control sites, respectively.

Sampling Design

We were able to select 7 to 31 clones per treatment at the three sites ranging in size from 5 to 15 m. Each clone was marked with a stake at the center of the clone that was numbered and color-coded to treatment type.

The surrounding mudflat was divided by four equidistant transects running from the center of each clone onto the mudflat. Sampling points for shoot density and percent cover were located 1 m from the clone's perimeter. A control sampling point was located along the same transect 5 m from the clone perimeter, beyond the range of treatment effects. These points were located during each sampling period using a pre-marked cord that was stretched in a straight line from the center pole, across a fixed point within the clone and out onto the adjacent mudflat. This allowed sampling points to remain 'blind' during treatment to avoid any bias by control personnel. We used a 0.25 m^2 frame centered over the spot marked on the cord to determine shoot density and percent cover. This frame contained a grid of 25, 100-cm^2 squares. Shoot density was calculated by counting all shoots originating within the 25 squares. Percent cover was determined by counting the number of times eelgrass shoots intersected the 36 corners within the frame's grid. Due to logistical constraints, shoot density was recorded at each distance from the clone along only one randomly chosen transect. Percent cover was determined at both sampling points on all four transects with the 1 m and control points averaged separately for each clone.

We monitored the non-target effects of the aerial application of glyphosate at Kaffee Meadow within the adjacent mudflat along one length of the treated plot (Bay side). The mudflat contained 5, 10-m transects spaced at equal distances with sampling points along each transect marked with polyvinyl chloride (PVC) poles at 1, 3, and 10 m running perpendicular from the edge of the treated meadow. A control plot was established 400 m north of the treatment plot. The control plot design was identical to that of the treatment

plot. Measurements of shoot density and percent cover were similar to those described for clones with the exception that shoot density was measured at all points (1, 3 and 10 m) along each transect. In addition to eelgrass measurements, we used glass fiber filter papers (Whatman, Inc., Clifton, NJ) to monitor non-target deposition of the herbicide from the aerial spray. Immediately prior to spray, we affixed the circular (9-cm diameter) glass fiber filter papers to the top of the PVC poles marking the sampling points at each distance along each transect (n = 15). Filter paper collection and analyses are described in Major et al. 2003a.

Treatments

Clones. Mowing of clones was carried out by management personnel using various hand-held brush cutters. Depending on the substrate and density of Spartina, a variety of cutting, attachments were used including steel and plastic blades, and heavy-duty plastic line. All mowing was to within 10 cm of the substrate. Each clone at Nemah and North River was mowed three times, once in June, July and August. However, due to logistical constraints, the clones on the Lewis Unit were mowed only twice (June and August 1995, Table 1).

At the Lewis Unit, herbicide was applied using a hovercraft equipped with a Model 60-Spotlyte® agricultural sprayer (C.P.I. Equipment Ltd., Saanichton, BC) with a hand-held wand and adjustable brass nozzle. The Nemah Beach, and North River sites were sprayed using Solo®, 15-L backpack sprayers (Solo, Kleinmotoren GmbH, Germany). Both types of hand spray application used glyphosate at 20.2 kg ae ha^{-1} in 842 L of water ha^{-1} with the non-ionic surfactant LI 700® (Loveland Industries, Inc., Greeley, CO) at 2.0% v/v following the label directions for hand-held and high volume equipment of "spray to wet". This application rate represented the maximum 5 percent solution of Rodeo® recommended by the label. However, we increased the volume of LI 700® in the tank mix from the maximum label recommendation (0.5%) because the higher volume was associated with an increase in efficacy of the herbicide to control Spartina on small experimental plots in the Bay (Norman and Patten 1996). The combination treatment of mowing and spraying utilized the two techniques described above, except clones were mowed only once. Clones were first mowed, then allowed to recover for approximately 6 wks before being treated once with glyphosate in July (Table 1). All chemical treatments to Spartina were made at low tides allowing 5 to 6 h of drying time before inundation of 50% of the plant. Eelgrass beds were normally lying dry upon the mudflat, though an occasional pool (≤2 cm deep) was associated with the sample locations. Weather conditions were optimal with air temperatures ranging between 19 and 29 C and wind speeds of 0 to 8 km h^{-1} with occasional gusts to 16 km h^{-1} at the Lewis Unit.

Meadow. A Soloy Bell® (Soloy Corporation, Olympia, WA) helicopter with a 9.1-m toe-mounted boom applied glyphosate to the meadow at 0915 on 13 August 1995. The tank mix included glyphosate at 4.2 kg ae ha^{-1} in 93 L of water ha^{-1} with X-77 Spreader® (Loveland Industries, Inc., Greeley, CO) at 0.13% v/v. Application occurred 1 h before low tide, allowing for ≥6 h of exposure time post-treatment before inundation of 50% of the plant. Weather conditions for the spray were optimal with winds ranging from 0 to 8 km h^{-1} from the south, and an ambient air temperature of 14.5 C.

Footwear Comparisons

Because the Spartina control measures we studied for clone infestations are labor intensive and require extensive movement on the intertidal mud flat, damage to eelgrass beds through trampling by treatment personnel is a probable outcome. To investigate this, we chose areas of homogenous eelgrass cover (visual interpretation) at the three clone sites previously described. Ten meter transects (treatment and control) marked at each end with a PVC pole were established parallel to shore within the same tidal range as the Spartina clones scheduled for treatment. Individual sampling points were located at the 1, 3, 5, 7 and 9 m distances along each transect using a removable, non-stretch cord graduated in meters. Transects were established in June, July and August to test for possible seasonal differences in eelgrass response to disturbance. Treatments included the placement of a footprint at the center of each sample point using three different types of footwear, all employed by treatment personnel. These included: rubber boots, Mudders™ (Amark, Inc., Merrimack, NH) and Mudlucks (USFWS, Vancouver, WA). All footprints were created by the same individual: weight = 68 kg, height = 170 cm, shoe size = men's 9).

TABLE 1. DATES OF SAMPLING AND TREATMENT OF SPARTINA CLONES. MOWING FOR THE MOW + HAND SPRAY COMBINATION OCCURRED AT THE TIME OF THE FIRST MOWING FOR THE MOW TREATMENT. SIMILARLY, SPRAYING FOR THE COMBINATION TREATMENT COINCIDED WITH THE HAND SPRAY ONLY TREATMENT AT EACH SITE. SAMPLING FOR THE MOW + HAND SPRAY COMBINATION COINCIDED WITH THAT OF THE MOW TREATMENT.

Site	Mow			Spray[1]		
	Pre-treatment (1995)	Treatment (1995)	Post-treatment (1996)	Pre-treatment (1995)	Treatment (1995)	Post-treatment (1996)
Lewis	May 18, 19	June 2, 9, 12 Aug 8, 9	May 25, 26	July 2, 3	July 19, 27, 28	June 20
Nemah	May 23-25	June 8 July 24 Aug 24	May 27-29	July 5, 6	July 18, 19	June 22
North River	June 7, 8	June 2, 9, 12	June 30, 31	July 7	July 20	June 24

[1]Glyphosate applied at 20.2 kg ae ha^{-1} in 842 L water ha^{-1} with the non-ionic surfactant, LI 700® at 2.0% v/v following label directions of "spray to wet".

Only footwear types appropriate to a particular substrate were tested on that substrate; thus, not all sites had the same combination of treatments. Pre-treatment shoot density was measured within 10 randomly selected 100 cm² subunits of the sampling frame on treatment and control transects. On treatment transects, only those subunits directly impacted by footwear within the frame were used for post-treatment measurements of shoot density and subsequent analyses. Post-treatment controls were counted on the same 10 subunits chosen randomly for pre-treatment measurements.

Non-target Deposition from Aerial Spray

APT Labs, Inc. (Wyomissing, PA) analyzed the filter papers for glyphosate using the methodology described in Kilbride et al. (1995) with a detection limit of 0.05 µg. Because of the high percent recovery of glyphosate from the filter papers (99.8%, SD = 3.2%), sample residues were not corrected for percent recovery. Deposition of the ae glyphosate on the filter papers was reported as µg dry weight and converted to a percentage of the expected deposition of ae glyphosate based on the nominal application rate. For calculation of mean deposition (geometric) for specific distances away from the treated meadow, detection limits (0.02 µg) were used for values reported as non-detected.

Statistical Analyses

We used a parametric approach to assess differences in shoot density and percent cover among treatments within sites. Assumptions of normality and homogeneity of variance were tested first, and the former was met in most cases. Data were log transformed and the differences between the log values at each of the two sampling times of pre and 1 yr post-treatment were used as the response variable. This is equivalent to using the log of the ratio, with 1.0 added to the original response to accommodate zero values. We used ANOVA for comparing response variables at individual locations between the aerially sprayed and control plots and a paired sample t-test for testing differences between control and 1 m locations at individual clones across treatments at each site. When variances were unequal, we used a t-test (Welch's) or ANOVA accommodating variance heterogeneity (Zar 1999). When differences were detected between treatment and control, the degree of impact among treatments was separated using the Tukey-Kramer Honestly Significant Difference test. Footwear comparisons between treatments and controls were assessed using ANCOVA with boot type, month and substrate as factors. Only measurements of shoot density in the subunits directly impacted by footwear were included in the analyses. Differences were considered statistically significant if the probability associated with the test statistic was ≤0.05.

RESULTS AND DISCUSSION

Initial eelgrass shoot densities and percent cover for the clone treatments at Lewis, Nemah and North River varied. Averages over both distances (1 and 5 m) indicate the Nemah site had the greatest initial shoot density and percent cover of eelgrass of the three sites followed by the North River and Lewis Unit, respectively.

In most cases, treatments of clones resulted in no differences in eelgrass shoot density or percent cover between the 1 m and control locations (Table 2). No differences were detected on the Lewis Unit. Only the mow spray treatment resulted in reductions (t = -2.37, DF = 11, P = 0.036) in shoot

TABLE 2. AVERAGE CHANGES (REPRESENTED AS A PERCENT) IN SHOOT DENSITY AND PERCENT COVER (PRE-TREATMENT VS. 1 YR POST-TREATMENT) OF EELGRASS AT 1 AND 5 M (CONTROL) FROM SPARTINA CLONE EDGE FOLLOWING ONE OF THREE TREATMENTS: MOWING[1], HAND SPRAYING[2], OR A MOW + HAND SPRAY[1,2] COMBINATION. MEAN (±SE) PRE AND POST-TREATMENT SHOOT DENSITY (SHOOTS 0.25 M²[-1]) AND PERCENT COVER ARE GIVEN IN PARENTHESES. P-VALUES AND DEGREES OF FREEDOM ASSOCIATED WITH STATISTICAL TESTS FOLLOW THE 1 M DATA. CHANGES WERE CONSIDERED SIGNIFICANT IF P ≤ 0.05.

		Mow		Spray		Mow + Spray	
		Shoot density	Percent cover	Shoot density	Percent cover	Shoot density	Percent cover
Lewis	1 m	-84	-83	-86	-88	-81	-75
		(20 ± 11, 3 ± 1)	(0.6 ± 0.2, 0.1 ± 0.0)	(57 ± 25, 8 ± 2)	(2.7 ± 0.8, 0.3 ± 0.1)	(25 ± 19, 5 ± 2)	(0.8 ± 0.4, 0.2 ± 0.1)
		0.945, 14	0.806, 14	0.353, 15	0.622, 16	0.956, 7	0.198, 7
	5 m	-71	-85	-91	-88	-75	-87
		(18 ± 9, 5 ± 2)	(0.7 ± 0.3, 0.1 ± 0.0)	(137 ± 55, 11 ± 4)	(4.5 ± 1.5, 0.5 ± 0.2)	(13 ± 6, 3 ± 1)	(1.6 ± 0.5, 0.2 ± 0.1)
Nemah	1 m	-39	-45	-36	10	-66	-54
		(303 ± 41, 184 ± 37)	(10.4 ± 1.4, 5.7 ± 0.9)	(249 ± 52, 158 ± 40)	(6.8 ± 1.0, 7.5 ± 1.2)	(229 ± 75, 76 ± 28)	(8.1 ± 1.9, 3.7 ± 0.8)
		0.586, 26	0.338, 30	0.698, 28	0.622, 29	0.036, 11	0.462, 12
	5 m	-49	-53	-25	6	-29	-39
		(292 ± 41, 146 ± 29)	(11.5 ± 1.4, 5.3 ± 0.8)	(213 ± 42, 158 ± 43)	(7.9 ± 1.3, 8.4 ± 1.6)	194 ± 59, 136 ± 34)	(7.9 ± 1.7, 4.8 ± 1.2)
North River	1 m	-63[a]	-83[a]	-32[a]	68[b]	-53[a]	-65
		(81 ± 21, 30 ± 17)	(3.6 ± 0.9, 0.6 ± 0.2)	(66 ± 24, 45 ± 26)	(2.5 ± 1.0, 4.2 ± 1.4)	(49 ± 16, 23 ± 15)	(2.0 ± 0.5, 0.7 ± 0.2)
		0.001, 15	0.005, 15	0.037, 15	0.008, 15	0.024, 6	0.618, 6
	5 m	30	-39	35	205	27	-37
		(127 ± 22, 166 ± 27)	(5.8 ± 1.1, 3.5 ± 0.7)	(116 ± 31, 157 ± 46)	(3.4 ± 1.3, 10.4 ± 2.5)	(82 ± 15.7, 104 ± 36)	(4.3 ± 1.4, 2.7 ± 0.9)

[1]Number of mowings: Lewis = 2; Nemah and North River = 3.
[2]Glyphosate applied at 20.2 kg ae ha^{-1} in 842 L water ha^{-1} with the non-ionic surfactant, LI 700® at 2.0% v/v following label directions of "spray to wet".
[a,b]Different superscripts denote significant differences among treatments in response variables.

density at Nemah. In contrast, at North River, reductions in shoot density were detected for all three treatments (mow: t = -3.96, DF = 15, P = 0.001: spray: t -2.28, DF = 15, P = 0.037; mow-spray: t = -2.99, DF = 6, P = 0.024) and in percent cover for all treatments except the mow-spray combination (mow: t = -3.29, DF = 15, P = 0.005: spray: t = -3.08, DF = 15, P = 0.008). Because North River was the only site to have reductions in the response variables for more than one treatment, it was possible to make comparisons among treatments. Based on the Tukey's HSD for multiple comparisons, there was no separation between mean differences (pre-treatment vs. 1-yr post-treatment) in shoot density for the three treatments. However, there were greater reductions (mean difference = -4.70; 95%CI = ±2.6; P ≤ 0.001) in percent cover associated with mowing than hand spraying.

Initial eelgrass shoot density and percent cover on the mudflat adjacent to the aerially sprayed and control meadows differed slightly at each distance (1, 3 and 10 m) with the control plot having a greater average density of eelgrass (Table 3). Shoot density and percent cover were reduced on both plots at each distance 1 yr after treatment. However, decreases on the control plot were greater than that on the treated plot.

Impacts to eelgrass by the different footwear were non-significant for nearly all comparisons. Only one treatment (Mudluck, P = 0.004) in July at a mud site resulted in a decrease in shoot density (Table 4).

Based on a comparison of the control techniques for the clone-sized infestations we monitored, non-target impacts to adjacent eelgrass were not significant at two out of three sites, with the exception of Nemah Beach for the mow-spray treatment. At the North River site, shoot density was reduced across all three treatments and percent cover was reduced on the mow and spray treatments. Why effects were primarily restricted to only one of our sites is not clear, although differences in substrate may have been more influential in determining impacts to eelgrass than any given Spartina treatment. As indicated by the footwear study, eelgrass in the soft, muddy substrate of the Lewis Unit and North River sites appeared to incur more physical damage from the movement of personnel and equipment around clones than at the

hard packed sand present at the Nemah Beach site. Because the turion nodes and rhizome structure of eelgrass lie at a depth of 0 to 3 cm, they are likely damaged or destroyed by footwear penetrations exceeding this depth. However, it was only at the North River, not the Lewis Unit with the softest substrate where we saw significant impacts to eelgrass. This could be a result of treatment personnel limiting their movement at Lewis to areas of Spartina coverage during control efforts due to the extreme difficulty of working on soft mud at this site. This was certainly the case for the authors when gathering data for this study. At North River, the substrate did not necessitate special footwear or demand as much effort to move about, but was still soft enough to allow foot penetration to a depth which could damage eelgrass. Therefore, it is quite possible that treatment personnel spent more time on the adjacent mudflat at North River than at the Lewis Unit.

Samples of sediment collected at 1 m from the clone edge after treatment at all three sites suggest the adjacent eelgrass was exposed to the herbicide, but at two out of three sites we observed no impact from the spray treatment 1 yr later. Concentrations of glyphosate in sediment samples collected 5 m from sprayed clones were below detection limits (Major et al. 2003b). Some tolerance to glyphosate by seagrasses has been reported. Ralph (2000) found that exposure to glyphosate produced no photosynthetic stress response by the seagrass *Halophila ovalis* as measured by it's ability to fluoresce chlorophyll *a*. In our study, the small amounts of water remaining on the surface of the mudflat may also have diluted the chemical, rendering it less toxic to the eelgrass.

Non-target deposition from the aerial application of the herbicide resulted in no damage to the adjacent eelgrass even though there was measurable herbicide in the mudflat at all three sampling distances of 0, 3 and 10 m (Major et al. 2003b). These results support the conclusion that impacts seen at the hand-spray treatment at North River were the result of physical disturbance by treatment personnel and not by contact with glyphosate.

Assessing the impacts to eelgrass was an important step in determining if any of the techniques employed to control Spartina would pose a significant or disproportionate threat

TABLE 3. MEAN (± SE) SHOOT DENSITY (SHOOTS 0.25 M^{2-1}) AND PERCENT COVER OF EELGRASS 1, 3 AND 10 M FROM SPARTINA MEADOW BEFORE AND 1 YR AFTER AERIAL APPLICATION OF RODEO®[1] AND AN UNSPRAYED CONTROL MEADOW. CHANGES EXPRESSED AS PERCENT.

Location & Plot	Pre spray		Post-spray		Change	
	Shoot density	Percent cover	Shoot density	Percent cover	Shoot density	Percent cover
1 m						
Treatment	409 ± 149	17.6 ± 6.4	186 ± 112	8.8 ± 5.3	-54 [a]	-50 [a]
Control	604 ± 70	31.6 ± 3.2	290 ± 86	10.4 ± 3.6	-52 [a]	-67 [b]
3 m						
Treatment	179 ± 85	13.2 ± 5.0	133 ± 75	9.4 ± 5.7	-25 [a]	-28 [a]
Control	656 ± 40	35.8 ± 0.2	371 ± 96	18.0 ± 5.4	-43 [b]	-49 [a]
10 m						
Treatment	266 ± 151	13.6 ± 7.5	101 ± 50	4.6 ± 2.9	-62 [a]	-66 [a]
Control	318 ± 141	18.0 ± 6.6	122 ± 81	6.0 ± 3.5	-61 [a]	-66 [a]

[1]Glyphosate applied at 4.2 kg ae ha^{-1} in 93 L water ha^{-1} with X-77® Spreader at 0.13% v/v.
[a,b]Superscripts denote significant differences (P ≤ 0.05) in response variable (at individual distances) between treatment and control.

TABLE 4. AVERAGE CHANGE (REPRESENTED AS A PERCENT) IN SHOOT DENSITY OF EELGRASS FOLLOWING USE OF THREE TYPES OF FOOTWEAR: MUDDERS™, MUDLUCKS OR BOOTS. MEAN (±SE) SHOOT DENSITY (SHOOTS 100 CM$^{2-1}$) PRE AND POST-TREATMENT FOLLOW THE PERCENT CHANGE.

	Mud			Sand		Mixed		
	Mudder	Mudluck	Control	Boot	Control	Mudder	Boot	Control
June	-8.4	-33.9[a]	9.0	-19.8	-32.0	-90.6	-88.0	-87.5
	16.7 ± 0.4,	23.0 ± 0.4,	17.2 ± 0.1,	23.7 ± 0.6,	28.7 ± 0.4,	24.5 ± 0.7,	20.0 ± 0.1,	19.2 ± 0.3,
	15.3 ± 0.9	15.2 ± 0.9	18.9 ± 1.4	19.0 ± 1.1	19.5 ± 1.7	2.3 ± 0.2	2.4 ± 0.2	2.4 ± 0.4
July	-55.5	-64.5	-67.0	8.2	5.0	-86.6	-87.2	-90.3
	25.2 ± 0.2,	35.8 ± 0.2,	34.2 ± 0.3,	23.1 ± 0.5,	22.0 ± 0.3,	26.5 ± 0.5,	23.5 ± 0.3,	24.8 ± 0.3,
	11.2 ± 0.6	12.7 ± 0.6	11.3 ± 0.8	21.2 ± 0.7	23.1 ± 1.0	3.5 ± 0.2	3.0 ± 0.2	2.4 ± 0.3
August	-48.0	-57.3	-43.6	-5.4	-6.8	-62.5	-64.7	-66.9
	25.6 ± 0.2,	26.7 ± 0.2,	25.9 ± 0.3,	25.9 ± 0.6,	30.7 ± 0.3,	11.2 ± 0.2,	13.3 ± 0.3,	14.5 ± 0.2,
	13.3 ± 0.6	11.4 ± 0.6	14.6 ± 1.1	24.5 ± 0.6	28.6 ± 1.2	4.2 ± 0.3	4.7 ± 0.3	4.8 ± 0.6

[a]Denotes significant difference from control (P ≤ 0.05).

to eelgrass. Although some damage appears to be caused by the physical movement of treatment personnel, especially in softer substrates, it is our opinion that the single most deleterious phenomena to the annual eelgrass is the spread of Spartina. Due to the effects of shading and sediment entrapment, Spartina out competes eelgrass during the cordgrass' initial growth stage and for the duration of it's life cycle. With an expansion rate in the diameter of clones of ca. 0.8 to 1.5 m yr^{-1} (Riggs 1992; Simenstad and Thom 1995; Feist and Simenstad 2000) and a rate of sediment entrapment of ca. 2 to 7 mm yr^{-1} (Gleason et al. 1979; Thom 1992; Simenstad and Thom 1995), Spartina has the potential to convert sparsely vegetated mud flats to higher marsh elevation within a relatively short time.

Although there may be some initial set back and loss of eelgrass during Spartina control operations, impacts appear to be localized, short-lived and more related to substrate than type of mechanical or chemical treatment. Even if control techniques are affected over larger areas and multiple years, eelgrass should recover. It's resilience as an annual (over-wintering as buried seeds which germinate in the spring), its ability to disperse locally into new locations as uprooted vegetative mats, and the inadvertent transport of the seagrass throughout the Bay by animal and human activities (Harrison and Bigley 1982) should provide sufficient means of recovery in most situations. Impacts to eelgrass associated with Spartina control operations appear to be minor compared to what might occur should the spread of Spartina go unchecked in the Bay (Willapa Bay Spartina Management Task Force 2001; WDNR 2000). We conclude that the unchecked spread of Spartina is a far greater threat to the survival and health of eelgrass than that from any of the control measures we studied. The basis for evaluating control measures for Spartina should be efficacy and logistical constraints and not impacts to eelgrass.

ACKNOWLEDGMENTS

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