Final Report

Efficacy and Non-Target Impacts of Imidacloprid Following Application to Control Burrowing Shrimp in Willapa Bay, Washington in 2012

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Executive Summary

Research was conducted in 2012 to collect data required by Washington's Department of Ecology to obtain an NPDES for the use of imidacloprid for burrowing shrimp control in Willapa Bay. Data were collected to a) assess and compare the efficacy of liquid and granular formulations of imidacloprid against burrowing shrimp on a commercial scale on plots of differing vegetation density and substrate, b) assess the impact of commercial-scale imidacloprid application on non-target megafauna, and c) map the non-target off-site impact of imidacloprid to benthic infauna. Sites, plots, treatment applications and data collection conformed to the specifications of the Sample and Analysis Plan, with any deviation from protocol noted. The only non-target impacts observed to megafauna were on Dungeness crab. There was an average of 2 affected crab/ ac. The highest count was 3.4 affected crab/ac for the Mallet site at Leadbetter 24 hours after treatment. No effects on fish or birds were observed. Mapping affected tidelands outside the treated area two weeks post-treatment indicated the effects were limited to a narrow band around the plot, with an average 15% increase in area beyond what was treated. Efficacy across all sites and treatments ranged from 65% to 84% burrow reduction.

Introduction

Research conducted by WSU from 2006 to 2011 has suggested that imidacloprid is a viable alternative to carbaryl for controlling burrowing shrimp. Research was conducted in 2012, under EPA Experiment Use Permit, to collect data required by Washington's Department of Ecology (Hart Crowser Inc. Sampling and Analysis Plan (SAP) for the Willapa Grays Harbor Oyster Growers Association, July 2012). This report contains application information, results on efficacy, non-target impacts to megafauna and indirect assessments of offsite benthic macro-invertebrates.

Objectives

- Assess and compare the efficacy of liquid and granular formulations of imidacloprid against burrowing shrimp on a commercial scale on plots of differing vegetation density and substrate.
- Assess the impact of commercial scale imidacloprid application on non-target megafauna.
- Map the non-target off-site impact of imidacloprid to benthic infauna.

Methods

In 2012, sites and plots were laid out to conform to the specifications of the SAP. Site locations were south of the Palix River and west of Leadbetter State Park. These were located on intertidal commercial shellfish beds not currently farmed. Detailed information about treatments, treatment sites, and applications are listed in Tables A1 to A5. Details on protocols for application, data collection and analysis are contained within the SAP. Variations in protocol are listed below. In general, efficacy (14 days after treatment) and impacts to megafauna (24 hours after treatment) were measured on all sites. Efficacy was assessed by counting shrimp burrows before and after treatment, and/or relative to an untreated control site. Whole bed density of epibenthic megafauna (Dungeness crab and fish) were assessed by making multiple closely spaced transects over the beds, counting all affected megafauna species on and within 150 feet of the site. Affected species were those exhibiting any signs of tetany, or were dead by any cause, directly or indirectly related to the treatment (e.g. bird predation of tetany crab). Raw data for non-target impacts and efficacy are provided in appendix tables.

<u>Deviation from protocol</u>

Randomization: Treatments were not assigned to their block by randomization. Due to very limited availability of sites that fulfilled the criteria, control sites were assigned to blocks not owned by growers (state land). Due to the need to apply Mallet to sites covered by shallow water at application, the Mallet sites were assigned to blocks that were lower in the tidal zone that the Nuprid sites.

Site size, shape and separation: Plot sizes for Nuprid were not 10 acres, but were 8.5 and 7.75 acres. This was due to limited site selection options with grower ownership. Sites selected at Leadbetter for treatment were very close to areas previously treated with carbaryl and imidacloprid. This confounded accurate inferences for offsite treatment impacts in all the cardinal directions. The Mallet site was boarded on the SE by two wedges of tidelands treated in 2010 by imidacloprid. The Nuprid site had two wedges of imidacloprid-affected ground to the

west. One of the wedges cut partially into the Nuprid site. Sites selected at Palix and Leadbetter for comparison between the control and treatments were not exactly similar due to the requirement for 500 m distance apart.

Vegetation: The Mallet site at Palix River had significantly less vegetation than the Nuprid site. The control site at Leadbetter had more vegetation than the Mallet or Nuprid sites, both of which only had a few thin patches of eelgrass.

Shrimp density and size: These were similar for all plots at Palix River. For the Leadbetter sites, the Mallet location had a good density of mature shrimp, the control site had moderate density of mature shrimp and the Nuprid site had moderate density of small shrimp in the 1-2 year age class.

Mapping off-site impacts -data collection: Counting of polychaete burrows on and off sites on the day preceding treatment and at 14 and 60 days was done, but was not a reliable data set. Differentiating what was a polychaete's burrow and what was not was too difficult and lacked consistency between counters. Also the thick vegetation totally obscured the holes making them all but impossible to count. Instead, affected acres of all treated sites were calculated based on GIS tracking of visual appearance of empty shells of commensal burrowing clam, *Cryptomya californica*, on the sediment surface. *C. californica* is a suspension feeder living within the burrows of *Neotrypaea californiensis*. *C. californica* shells migrate to the surface wherever *N. californiensis* die. The extent of off-target impacts can be easily traced by the presence or absence of these surface shells following imidacloprid treatment. This was true for all treated sites, except the Nuprid site at Leadbetter. The age-class population of burrowing shrimp there were too young to have any significant *C. californica* associated with them yet. Hence, the extent off-site impact at this site could not be obtained.

Results and Discussion

Non-target megafauna impacts: The number of affected Dungeness crab per acre varied by location and treatment (Tables 1 to 3). The average across all sites and treatments was 2 affected crab/ ac. The highest count was 3.4 affected crab/ac for the Mallet site at Leadbetter 24 hours after treatment. These results are similar to our 2011 data where the number of affected crab per acre ranged from 0.9 to 3.8. Bird predation of tetany crab was the main cause of cause of crab mortality (See appendix). However, crushing of crab with the ATV during application was also a significant cause of loss.

There were no significant effects on fish observed on any of the sites following any treatment (Tables1 to 3). Birds (seagulls) were observed foraging on and nearby the sites following treatments (Tables 1 to 3). These were either resting, or consuming affected burrowing shrimp or crab. The density varied from zero to 4.4 seagulls/ac over a 1 hour observation period. The majority of all seagulls present on the sites were resting during our observation, with very few exhibiting intensity foraging. No imidacloprid affected birds were observed.

Mapping affected tidelands outside the treated area two weeks post-treatment, based on the presence of dead commensal clam shells, indicated the pattern and range of significant off-site chemical movement (Table 4, Figure 1). For the most part these affected areas were confined to

a narrow band around the plot, with an average 15% increase in area beyond what was treated.

Efficacy: Efficacy across all sites and treatments ranged from 65% to 84% burrow reduction (Table 5). This was within the range of efficacy found in previous trials. Efficacy was reduced at sites with significant eelgrass coverage (Palix River). Areas immediately outside (3 and 30 m) the treated sites also exhibited some levels of burrowing shrimp reduction (zero to 72% reduction in burrow density). This is fairly similar to what was inferred from post-treatment non-target site mapping (Figure 1). Overall there were too few off-site efficacy measurements to account for the variability.

Conclusion

Large-scale applications of imidacloprid used on the tidelands of Willapa Bay to control burrowing shrimp provided reasonable efficacy of 65 to 84%. This level of efficacy is less than what would be expected for carbaryl, but adequate for managing this important pest to the shellfish industry. The only non-target impacts to megafauna observed from these treatments were on Dungeness crab. These impacts were fairly minimal (2 affected crab per acre). This is an order of two orders of magnitude less than what would be observed following a carbaryl application. Although there were off-site treatment effects observed on the sediment, this was confined to a narrow band around the treatment zone. This research was conducted with parallel studies by the University of Washington on the environmental fate and persistence of imidacloprid (Grue & Grassley 2013) and by the Pacific Shellfish Institute on the effect of imidacloprid on the sediment impact zone (Booth et al. 2013). Overall conclusions of this research project are addressed in a report by Hart Crowser, Inc. (2013).

Literature cited

Booth, S., K. Rasmussen, A. Suhrbier. 2013. Impact of imidacloprid on epibenthic and benthic invertebrates: 2012 studies to describe the Sediment Impact Zone (SIZ) related to imidacloprid treatments to manage burrowing shrimp. Pacific Shellfish Institute.

Grue, C. and J. Grassley. 2013. Environmental Fate and Persistence of Imidacloprid Following Experimental Applications to Control Burrowing Shrimp in Willapa Bay, Washington. Washington Cooperative Fish and Wildlife Research Unit, University of Washington, Seattle, WA. 91 pp

Hart Crowser, Inc. 2012. Sampling and Analysis Plan: Experimental Trials for Imidacloprid Use in Willapa Bay, Washington. Hart Crowser, Inc., Edmonds, Washington. 72 pp.

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Table 1. Summary of total affected megafauna within the treated area and the 0 to 50 m perimeter of the treated area at the Nuprid, Mallet and control sites at the Palix River sites in 2012.

		Crab status (#/ac)							
	Date	Alive, no noticeable treatment effects	Alive with tetany	Dead, unknown cause	Dead, predated by birds	Total affected crab	Dead or tetany fish, any size or species (#/ac)	Dead or affected birds, any size or species (#/ac)	Mean hourly seagull foraging count within area over 1 hr period* (#/ac)
Nuprid	8/1/12	0.2	0	0	0	0	0	0	0
Nuprid	8/2/12	0	0.3	0.6	0	0.9	0	0	0
Nuprid	8/3/12	0.4	0.4	0	0.7	1.1	0	0	1.5
Mallet	8/1/12	0.1	0	0	0	0	0	0	0
Mallet	8/2/12	0	0.1	0.1	0	0.2	0	0	0
Mallet	8/3/12	0.1	1.6	0.2	0	1.8	0.1	0	3.5
Control	8/1/12	0.1	0	0.1	0	0.1	0	0	
Control	8/3/12	0	0	0.1	0	0.1	0	0	0

*Sum of seagulls on or near the site counted every 20 minutes. Birds were mostly resting, foraging.

Table 2. Summary of total affected megafauna within the treated area and the 0 to 50 m perimeter of the treated area at the Nuprid, Mallet and control sites at Leadbetter in 2012.

		Crab status (#/ac)							
Treat-ment	Date	Alive, no noticeable treatment effects	Alive with tetany	Dead, unknown cause	Dead, predated by birds	Total affected crab	Dead or tetany fish, any size or species (#/ac)	Dead or affected birds, any size or species (#/ac)	Mean hours seagulls foraging within area over 1 hr period* (#/ac)
Nuprid	8/5/12	0	0	0	0	0	0	0	0
Nuprid	8/6/12	0.3	0	0	0.2	0.2	0	0	0
Mallet	8/5/12	0	0	0	0	0	0	0	0
Mallet	8/6/12	0	0.6	0.2	2.6	3.4	0	0	4.4
Control	8/6/12	0	0	0.6	0.1	0.7	0	0	0

*Sum of seagulls on or near the site counted every 20 minutes. Birds were mostly resting, foraging.

Table 3. Summary of total affected megafauna per treated acre within the treated area and the 0 to 50 m perimeter of the treated area at the Nuprid, Mallet and control sites at Palix River and Leadbetter sites in 2012.				
		Crab	Fish	Birds
Treatment	Location	Total affected #/treated acres		
Nuprid	Palix R	2.0	0	0
Mallet	Palix R	2.0	0.1	0
Nuprid	Leadbetter	0.2	0	0
Mallet	Leadbetter	3.4	0	0

Table 4. Total treated and total affected acres (see figures below)			
Treatment	Location	Acres treated	Acres affected based on GIS tracking of commensal clam shells on surface.
Nuprid	Palix R	8.5	9.8
Mallet	Palix R	8.5	11.0
Nuprid	Leadbetter	7.7	*
Mallet	Leadbetter	5.0	5.0
* The lack of commensal clams at the Nuprid Leadbetter site precluded data collection. The average increase in acres was 15%. There was an average 15 % increase of affected ground over treated ground.			

Figure 1. GPS tracing of affected tidelands outside of the treated area based on change in ghost shrimp burrow densities and empty shells of commensal burrowing clam, *Cryptomya californica*, on the sediment surface.

Table 5. Summary of on- and offsite efficacy				
Location	Palix River sites		Leadbetter sites	
	Mallet	Nuprid	Mallet	Nuprid
	% reduction	% reduction	% reduction	% reduction
Onsite Inside	78±5	65±5	84±5	78±5
Offsite 3 m outside	61±10	45±13	21±21	42±16
Offsite 10 m outside	64±9	72±8	-1±22	19±27
Mean of % reduction 14 DAT vs. 1 DBT ± standard error, n=36 inside, n=8 for offsite 3 and 30 m.				

Appendix tables.

Table A1. Application Notes – Mallet -Palix River August 2, 2012	
Treatment site size and location	8.5 acres, south of Bay Center, Latitude 46.595 N, Longitude 123.947 W
Site characterization	Ownership Bay Center Mariculture, Tidal height range -1.6 to +1.4 ft. Sandy sediment with moderate coverage of *Zostera japonica* ,
Date/ time of application	8/2/12 - 06:30 to 07:26
Treatment applied	Imidacloprid – Mallet @ 0.5 lbs ai/ac, lot code - k11179k113 16:48 est
Method of application	ATV with 12v spin broadcast spreader mounted on back. 20' between passes. Calibrated by adjusting speed to achieve 17 lbs of product per pass.
Application notes	• Spin spreader did not provide an equal distribution, with more product delivered in the direction of spin. This meant that there was less than equal uniformity of the granular across each 20' pass. The start of each ATV pass was at the beginning of the boundary line for each pass. Because the spreader tossed backwards ~ 10', this resulted in offsite application of ~ 10'by 20' at the start of each pass. Applicator – Chase Metgzer. • 29 sacks of Mallet (30 lb sacks) were delivered on 8.5 acres. This was within 2% of the target rate (28.2 sacks) and, when adjusted for the small offsite application at the beginning of each run, this was within 1% of the target rate.
Application conditions	• Sediment temp @ 8" depth – 65F @ 6:30, 65F@ 7:26 • Air temp 62F @ 6:30, 65F @ 7:26 • Surface water temp 63.5F @ 7:30 • Wind speed 0 mph @ 6:30, 0 mph @7:26 • Slight mist during application, no rain • Tides -1.6 @ 8:30, 9.1 @ 2.56 • Could cover 100% @ 6:30 am , 100 % @ 11 am • Time ebb tide moved offsite high side 7:20, low side 7:30 • Time flood tide moves onsite low side 10:40, high side 10:55 • Amount of standing water on plot during application- 2- 4 cm on top east end, and 4-12 cm on bottom west end . All but the very west end went dry after the ebb tide pulled off the site.
Flowing water on plots (levels/location/speed) after application	After the tide quickly pulled offsite (7:30) there was a slow movement of water in the lower part of the plot from the SE to the SW. Dye markers used on site @ 7:26 indicated that the water on the plots did not move beyond 50 m of the plots by 8:30
Other observations	Product dissolved well and there was mixed uniformity across plot even though application uniformity was not perfect. There was good shrimp burrow collapse following treatment across the entire plot, and several dozen shrimp were noticed on the sediment surface. Several small Dungeness crabs were smashed by the ATV during application. No birds were foraging post-treatment. No dead or affected fish were observed following treatment.

Table A2. Application Notes – Nuprid -Palix River August 2, 2012	
Treatment site size and location	8.5 acres, south of Bay Center, Latitude 46.591 N, Longitude 123.94 W
Site characterization	Ownership Coast Seafood, Tidal height range -0.6 to +2.8, Sandy sediment with moderate to thick coverage of *Zostera japonica*
Date/ time of application	8/2/12 – 08:25 to 09:20
Treatment applied	Imidacloprid – Nuprid 0.5 lbs ai/ac, lot code - lot t12039001
Method of application	ATV with Boom-buster flat nozzle mounted on back to 5 gpa 12v sprayer. 20' between passes. Calibrated - @ 16 sec to spray 3 quarts = 8 gpa. Used 1 pt of product in every 4 gallons of mix.
Application notes	• Applicator (Chase Metzger) speed was variable, resulting in a lower uniformity than desired. He went too fast during first 1/3 of the plot (East end). After the last pass was made (West end) there were 8 gallons of tank mix left over. This was applied uniformly over the East 1/3rd of the plot to make the over site treatment rate more uniform. .
Application conditions	• Sediment temp @ 8" depth – 65F @ 6:30, 65F@ 7:26 • Air temp 65F @ 6:30, 65F @ 7:26 • Surface water temp 65F @ 7:30 • Wind speed 0 mph @ 8:25, 0 mph @ 9:20 • Slight mist during application, no rain • Tides -1.6 @ 8:30, 9.1 @ 2.56 • Could cover 100% @8:25 am, 100% @ 11 am • Time ebb tide moved offsite high side 6:20, low side 7:30 • Time flood tide move onsite low side 10:45, high side 11:00 • Amount of standing water on plot during application- The west end had pooled water 20 cm deep at application with no flow offsite. The East end was fairly dry at applications.
Flowing water on plots (levels/location/speed) after application	Due to thick Zostera *japonica,* water did not drain off the low swales on the site, even at low tide. Dye markers used on site @ 9:20 indicated that the water on the plots did not move beyond 50 m of the plots by 10:30. Water flowed from the NE to the SW, but very slowly once the tide was off.
Other observations	Very thick eelgrass on site almost everywhere. There was no burrow collapse noticed post -treatment, but 25 shrimp were observed crawling on the sediment after treatment. Several small Dungeness crabs were smashed by the ATV during application. No birds were foraging post-treatment. No dead or affected fish were observed following treatment.

Table A3. Application Notes – Nuprid –Leadbetter August 5, 2012	
Treatment site size and location	7.75 acres, NE tip of the Long Beach Peninsula. Latitude 46.619 N Longitude 123.027 W
Site characterization	Ownership Long Island Oyster Co, Tidal height range -2.0 to +2.4, Sandy sediment with only patches of *Zostera japonica and Z. marina* along a swale draining north to south. The site's population of shrimp is mostly 1 to 2 years-old.
Date/ time of application	8/5/2012 – 09:01 to 9:45
Treatment applied	Imidacloprid – Nuprid 0.5 lbs ai/ac, lot code - 005 nu pb6 1
Method of application	ATV with Boom-buster flat nozzle mounted on back to 5 gpa 12v sprayer. 20' between passes. Calibrated - @73 sec a pass, 10 gpa, 1 quart of product in 10 gal. Applicator Nick Haldeman. First pass on east side by backpack, due to equipment problems.
Application notes	• 1 7/8 gallon was target rate. That amount was applied over site exactly. Product applied very uniformly, but would have preferred a 30 minute earlier start.
Application conditions	• Sediment temp @ 8" depth – 61.5F @ 9:00, 62F @ 9:45, • Air temp 59F @ 9:00, 60F @ 9:45 • Surface water temp 62F @ 9:30 • Wind speed 8-10 SW @ 9:00 to 9:45 • no rain • Tides -0.5 @ 10:15, 9.5 @ 4:30 pm • Could cover 100% @ 9:00 am to 11:00, sun burned off haze at 11:30-11:45 • Time ebb tide moved offsite low side 9:20 • Time flood tide moves onsite low side 11:45, high side ~ 12:10 pm
Flowing water on plots (levels/location/speed) after application	A shallow swale drained through the middle of the plot running north to south. It had a fast but shallow flow of water during the application, but went dry prior to tidal inundation. It carried product offsite and into a deep channel (60 m offsite) within 1 minute of applying the dye. The wind and tide pushed the dye north along the end of channel (6 m wide and ~ 100 m long). Another swale drained the western edge of the plot, but was not actually within the site. Evidence within this swale suggested that it had been treated before as there were no shrimp populations within it.
Other observations	Site dried fast, and had a dry time of 2.5 to 3 hrs. before flooding. Site had a hardpan at about 25 cm deep. This might affect infauna populations.

Table A4. Application Notes – Mallet –Leadbetter August 5, 2012	
Treatment site size and location	5 acres, NE tip of the Long Beach Peninsula. Latitude 46.619 N Longitude 123.038 W
Site characterization	Ownership Grassy Island Clam Farm LLC, Tidal height range +0.95 to +2.8, Sandy sediment with only a trace of *Zostera japonica* along the east edge of the plot.
Date/ time of application	8/5/2012 – 06:15 to 07:40
Treatment applied	Imidacloprid – Mallet 0.5 lbs ai/ac, lot code - k11179k113 16:48 est
Method of application	Belly grinder hand spreader – set on middle opening– 9' spread with 2' overlap between applicators. 3 applicators (Nick Haldeman, Kim Patten and Jacob Moore).
Application notes	• 12.5 lbs per pass was target rate. 12.5 lbs was loaded into each unit. Walking speed and opening were adjusted so that product could flow out at the end of each pass. One applicator was off-rate at the beginning of the plot and over-applied his section. The target rate for the site was 16 2/3 30# sacks. We used 16 2/3 sacks.
Application conditions	• Sediment temp @ 8" depth – 63F @ 6:30, 62F@ 7:26 • Air temp 59F @ 6:30, 59F @ 7:26 • Surface water temp 71F @ 10:30 • Wind speed 6.5 mph SW @ 7:00, 6.5 mph @9:20 SW • Slight mist during application, no rain • Tides -1.6 @ 8:30, 9.1 @ 2.56 • Could cover 100% @ 6:30 am , 100 % @ 12:00 noon, sun burned off haze at 12:30 pm • Time ebb tide moved offsite high side 6:30, low side 7:40, tide off middle section 6:40 • Time flood tide moved onsite low side 12:45, high side 1:15 • Amount of standing water on plot during application- none. Tide pulled off very fast during application. Went dry midway during treatment.
Flowing water on plots (levels/location/speed) after application	Product was applied in 6 to 20 cm of water during the first 20 minutes, after which the western half of the site went dry. Granular pellets did not dissolve on the west side until the incoming tide. This had an effect on efficacy.
Other observations	There was burrow collapse noticed post–treatment on the east end. A few seagulls were noted hanging around the site after treatment. No dead or affected fish were observed following treatment.

Table A5. Solar radiation data from WSU Agweather net weather station (Lat: 46.71 Lng: 123.97 Elevation: 10') Tokeland, WA during the application times for the SAP plots in 2012				
	2012-08-02		2012-08-05	
Time (PST)	Air Temp (F)	Solar Rad (W/m^2)	Air Temp (F)	Solar Rad (W/m^2)
05:00	57	0	57	0
05:15	57	3	57	2
05:30	57	5	57	6
05:45	57	13	57	9
06:00	57	24	57	15
06:15	58	30	57	24
06:30	58	44	57	29
06:45	58	68	57	31
07:00	57	79	56	35
07:15	57	114	56	42
07:30	58	111	56	61
07:45	58	131	57	76
08:00	58	125	57	85
08:15	58	118	57	91
08:30	58	153	57	101
08:45	58	168	57	122
09:00	58	215	57	136
09:15	58	268	58	170
09:30	58	247	58	227
09:45	58	323	59	320
10:00	58	273	59	354
10:15	58	235	60	539
10:30	59	292	60	619

Table A 6. Dungeness crab counts at the Palix River control site, within the treated area on 8/1/12 (day before treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	1	0	1	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 7. Dungeness crab counts at the Palix River control site, within the treated area on 8/3/12 (1 day before treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	1	0	0
2	0	0	0	0	0
3	0	0	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 8. Dungeness crab counts at the Palix River Mallet site, within the treated area on 8/1/12 (day before treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	1	0	0
2	0	0	0	0	0
3	0	0	0	1	0
4	0	0	0	1	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 9. Dungeness crab counts at the Palix River Mallet site, within 0 to 50m perimeter along the outside of the treated area on 8/2/12(day of treatment, immediately after application).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	1	0	1	0
3	0	0	0	1	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 10. Dungeness crab counts at the Palix River Mallet site, within the treated area on 8/3/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	2	1	0	0
2	0	1	0	0	0
3	0	5	1	3	0
4	0	6	0	0	0
5	1	0	0	1	0
6	0	0	0	0	0

Table A 11. Dungeness crab counts at the Palix River Mallet site, within 0 to 50m perimeter along the outside of the treated area, 8/3/1212 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	0	1	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 12. Dungeness crab counts at the Palix River Nuprid treatment site, within the treated area on 8/1/12 (day before treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	2	0	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 13. Dungeness crab counts at the Palix River Nuprid treatment site, within the treated area on 8/2/12 (day of treatment, immediately after application),					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	5	0	0
2	0	0	0	0	0
3	0	2	0	1	0
4	0	0	0	4	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 14. Dungeness crab counts at the Palix River Nuprid treatment site, within 0 to 50 m perimeter along the outside of the treated area, 8/2/12 (day of treatment, immediately after application).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	1	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 15. Dungeness crab counts at the Palix River Nuprid treatment site within 0 to 50 m perimeter along the outside of the treated area, 8/3/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	2	0	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 16. Dungeness crab counts at the Leadbetter control site, within the non-treated area on 8/6/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	0	1	0	1
4	0	0	0	0	0
5	0	0	1	0	0
6	0	0	0	0	0

Table A 17. Dungeness crab counts at the Leadbetter control site, within 0 to 50 m perimeter along the outside of the non-treated area on 8/6/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	0	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 17. Dungeness crab counts at the Leadbetter control site, within 0 to 50 m perimeter along the outside of the non-treated area on 8/6/12 (day after treatment.					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	1	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 18. Dungeness crab counts at the Leadbetter Mallet treatment site, within treated area on 8/5/12 (day of treatment, immediately after application).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	1	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 19. Dungeness crab counts at the Leadbetter Mallet treatment site, within 0 to 50 m perimeter along the outside of the treated area on 8/5/12 day of treatment, immediately after application).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	1	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 20. Dungeness crab counts at the Leadbetter Mallet treatment site, within the treated area on 8/6/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	1
3	0	0	0	0	2
4	0	1	0	0	4
5	0	0	0	0	1
6	0	1	0	0	0

Table A 21. Dungeness crab counts at the Leadbetter Mallet treatment site, within 0 to 50 m perimeter along the outside of the treated area on 8/6/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	0	1	0	0
4	0	1	0	0	3
5	0	0	0	0	2
6	0	0	0	0	0

Table A 22. Dungeness crab counts at the Leadbetter Nuprid treatment site, within the treated area on 8/5/12 (day of treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	1	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 23. Dungeness crab counts at the Leadbetter Nuprid treatment site, within 0 to 50 m perimeter along the outside of the treated area on 8/5/12 (day of treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	1	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 24. Dungeness crab counts at the Leadbetter Nuprid treatment site within the treated area on 8/6/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	0
3	0	1	0	0	0
4	0	0	0	0	0
5	0	0	0	0	0
6	0	0	0	0	0

Table A 26. Dungeness crab counts at the Leadbetter Nuprid treatment site, within 0 to 50 m perimeter along the outside of the treated area on 8/6/12 (day after treatment).					
Dungeness crab size (caprice diameter)	Crab status				
	Alive, no noticeable treatment effects	Alive with tetany	Dead, likely from treatment effect	Dead crushed by ATV	Dead, predated by birds
1	0	0	0	0	0
2	0	0	0	0	1
3	1	1	0	0	0
4	1	0	0	0	0
5	0	0	0	0	1
6	0	0	0	0	0

Table A 27. On-site treatment efficacy for Mallet and Nuprid treatments at the Palix and Leadbetter treatment sites in 2012.

	Palix River sites						Leadbetter sites					
	Mallet			Nuprid			Mallet			Nuprid		
	burrows/1/4m^2			burrows/1/4m^2			burrows/1/4m^2			burrows/1/4m^2		
Stake #	1 DBT	14 DAT	% reduction	1 DBT	14 DAT	% reduction	1 DBT	14 DAT	% reduction	1 DBT	14 DAT	% reduction
1	13	0	100	15	5	67	6	0	100	3	0	100
2	19	0	100	11	4	64			0	3	0	100
3	11	0	100	16	1	94	14	0	100	5	0	100
4	14	0	80	9	3	67	18	0	100	5	1	80
5	13	0	100	4		100	20	0	100	2	0	100
6	9	3	100	15	7	53	18	3	83	5	0	100
7	13	2	100	3	1	67	18	2	89	2	0	100
8	10	0	0	10	8	20	13	1	92	1	2	0
9	11	1	100	32	7	78	11	0	100	3	0	100
10	9	2	0	22	9	59	9	1	89	2	2	0
11	9	3	100	12	4	67	4	2	50	2	0	100
12	8	2	63	9	0	100	9	0	100	8	3	63
13	14	1	100	12	0	100	9	0	100	3	0	100
14	14	1	50	15	0	100	6	0	100	2	1	50
15	5		100	11	10	9	8	0	100	1	0	100
16	16	0	100	12	2	83	15	0	100	2	0	100
17	20		100	14	9	36	10	1	90	1	0	100
18	16	0	0	10	0	100	8	0	100	1	2	0
19	18	2	83	5	4	20			0	6	1	83
20	20	0	100	11	4	64	14	0	100	2	0	100
21	21	9	67	12	4	67	9	0	100	3	1	67
22	9	0	25	20	6	70	11	0	100	4	3	25
23	9	0	100	11	3	73	17	0	100	11	0	100
24	6	0	100	10	6	40	15	0	100	11	0	100
25	6	0	64	12	1	92	11		0	11	4	64
26	7	7	100	7	1	86	11		0	3	0	100
27	14	0	33	13	4	69	11	0	100	3	2	33
28	10	0	83	14	0	100	10	0	100	6	1	83
29	12	4	60	15	6	60	12	0	100	5	2	60
30	10	6	100	7	2	71	12	2	83	2	0	100
31	9	1	71	15	10	33	11	5	55	7	2	71
32	12	1	100	12	8	33	11	2	82	4	0	100
33	13	0	80	16	3	81	4	0	100	5	1	80
34	11	0	60	16	5	69	15	0	100	5	2	60
35	8	0	100	13	8	38	11	0	100	10	0	100
36	10	1	88	7	8	0	15	0	100	8	1	88
mean±SE			**78±5**			**65±5**			**84±5**			**78±5**

DBT- days before treatment, DAT days after treatment; counts taken at the Palix River Nuprid and Mallet sites 14 DAT were confounded by thick eelgrass.

Table A 28. Off-site efficacy subtending the treatment area for Mallet and Nuprid treatments at the Palix and Leadbetter sites in 2012.

Stake location outside of plot	Palix River sites						Leadbetter sites					
	Mallet			Nuprid			Mallet			Nuprid		
	burrows/1/4m^2			burrows/1/4m^2			burrows/1/4m^2			burrows/1/4m^2		
	1 DBT	14 DAT	% reduction	1 DBT	14 DAT	% reduction	1 DBT	14 DAT	% reduction	1 DBT	14 DAT	% reduction
3 m	8	6	25	6	6	0	4	4	0	2	2	0
3 m	12		100	10	4	60	12	14	-17	0	6	
3 m	12	5	58	7	1	86	19	0	100	2	2	0
3 m	9	3	67	6	1	83	4	4	0	5	1	80
3 m	13	0	100	8	2	75				3	1	67
3 m	15	8	47	11	9	18	12	16	-33	4	2	50
3 m	10	7	30	12	7	42	10	0	100	5	5	0
3 m	12	5	58	5	5	0	3	3	0	4		100
Mean±**SE**			**61±10**			**45±13**			**21±21**			**42±16**
10 m	7	4	43	5	2	60	8	8	0	3	8	-167
10 m	10	2	80	7	4	43	16	14	13	4	2	50
10 m	12	0	100	12	2	83	2		100	5	2	60
10 m	13	6	54	8	2	75	4	8	-100	4	3	25
10 m	17	0	100	6	0	100	11	11	0	7	4	43
10 m	5	3	40	7	1	86	16	20	-25	5	3	40
10 m	15	6	60	9	1	89				7	6	14
10 m	12	8	33	8	5	38	12	11	8	6	1	83
Mean±**SE**			**64±9**			**72±8**			**-1±22**			**19±27**

DBT- days before treatment, DAT days after treatment; counts taken at the Palix River Nuprid and Mallet sites 14 DAT were confounded by thick eelgrass. The lower numbers don't necessarily reflect a treatment effect, but a difficulty in accurately counting burrows under thick vegetation.