Vermont Department of Environmental Conservation
Lakes and Ponds Protection and Management Program

ProcellaCOR EC Aquatic Macrophyte Species Frequency of Occurrence Pre-and Post-Treatment Statistical Analysis April 2022

Abstract

In 2019, the first herbicide treatments in Vermont with the relatively new herbicide, active ingredient florpyrauxifen-benzyl (trade name ProcellaCOR EC) were conducted to control the aquatic invasive species Eurasian watermilfoil (*Myriophyllum spicatum*). ProcellaCOR EC is a registered herbicide with the U.S. Environmental Protection Agency (Registration Number 67690-80) and the Vermont Agency of Agriculture, Food and Markets. Treatments were conducted under the conditions of Aquatic Nuisance Control permits issued by the Vermont Department of Environmental Conservation (VTDEC), Lakes and Ponds Protection and Management Program. As a permit condition, permittees are required to conduct pre-and post-treatment point intercept plant surveys using scientifically valid protocols and methods that report on the Frequency of Occurrence (FOC) for all aquatic macrophyte species found.

To ensure permit compliance and to assess any unforeseen or unanticipated adverse impacts on non-target aquatic macrophyte species, VTDEC completed statistical analysis (Wilcoxon signed-rank test, 0.05 significance) of the FOC of both target and non-target aquatic macrophyte species using data from pre-and post-treatment plant surveys (first and most recent year) from eleven waterbodies treated with ProcellaCOR EC. For the purposes of this analysis, VTDEC considers the target species as being the species targeted for control, *Myriophyllum spicatum*, and non-target species as being all native aquatic macrophyte species and aquatic invasive species not targeted for control (i.e., *Potamogeton crispus*). While VTDEC considers all native aquatic macrophyte species as non-target species, there are native aquatic macrophyte species listed on the ProcellaCOR EC label as being controlled by this herbicide, which are included in this analysis (i.e., *Brasenia schreberi*, *Ceratophyllum demersum*).

Results from the first-year post-treatment (post-treatment 1) show that the average FOC of *Myriophyllum spicatum* decreased from 58.4 to 19.3 ($p = 0.005$) between pre-treatment and post-treatment. *Myriophyllum spicatum* also showed a decrease in both treatments from an average of 53.5 to 10.8 ($p=0.008$) in post-treatment 2. Post-treatment 1 *Potamogeton illinoensis* and *Vallisneria americana* both increased between pre- and post-treatment values ($p=0.008$, $p=0.037$, respectively). The most recent year post-treatment (post-treatment 2) *Potamogeton illinoensis* again showed an increase with the post-treatment 2 average increasing from 24.9 to 50.4 ($p=0.016$). *Ceratophyllum demersum* was the only non-target species to result in a statistically significant difference ($p = 0.039$) with an average decrease in post-treatment 2 results.

Future research into the effects of ProcellaCOR EC on Vermont lakes should closely monitor the effect of the herbicide on *Brasenia schreberi* and *Ceratophyllum demersum* populations as well as observe any statistical significance in changes of FOC to any other non-target species. Studies

on any potential significant increase in non-target species after the application of ProcellaCOR EC may add value to the research in this herbicide, as well as aid stakeholders in the decision to use this method for the management of *Myriophyllum spicatum*.

Methods

Lakes and Species Investigated

The data collected from eleven Vermont lakes was used for this study. The lakes included in this study were Lake St. Catherine (main basin), Lily Pond (St. Catherine), Little Lake (St. Catherine), Lake Morey, Burr Pond (Sudbury), Lake Hortonia, Sunrise Lake, Beebe Pond (Hubbardton), Lake Dunmore, Lake Iroquois, and Lake Pinneo. The species chosen for the statistical analysis included the target species controlled by ProcellaCOR EC, *Myriophyllum spicatum*, non-target species that are controlled by ProcellaCOR EC (i.e., *Brasenia schreberi*, *Ceratophyllum demersum*, and *Nymphoides* spp.), non-target species that are known to be sensitive (not controlled/sublethal) to ProcellaCOR EC (i.e., *Nymphaea odorata* and *Nuphar variegata*), and non-target species that have no anticipated impacts from ProcellaCOR EC treatments that include *Bidens beckii*, *Chara* sp., *Eleocharis* sp., *Elodea canadensis*, *Najas flexis*, *Potamogeton amplifolius*, *Potamogeton crispus*, *Potamogeton epihydrus*, *Potamogeton gramineus*, *Potamogeton illinoensis*, *Potamogeton natans*, *Potamogeton praelongus*, *Potamogeton pusillus*, *Potamogeton robbinsii*, *Potamogeton zosteriformis*, *Stuckenia pectinate*, *Utricularia gibba*, *Utricularia vulgaris*, *Vallisneria americana*, and *Zosterella dubia*. Although there are other species that have FOC data from pre and post ProcellaCOR EC treatment aquatic plant surveys, there was insufficient data (sample size was too small; less than five lakes) for the species to be considered in this analysis.

Statistical Analysis

The non-parametric Wilcoxon signed-rank test was used to determine statistical significance between pre- and post-treatment groups with the significance determination being a p-value of <0.05. Paired t-tests were also completed to provide a more statistically robust result. A normality test was not analyzed but may be in the future to better inform the statistical analysis. Statistical analysis was done on the application RStudio.

Results and Discussion

Post-treatment 1

Analysis of the results from the first post treatment survey show that:

- FOC of *Myriophyllum spicatum* (target species) decreased between pre-treatment and post treatment 1 ($p=0.005$).

- FOC of *Potamogeton illinoensis* and *Vallisneria americana* (non-target species) increased between pre-treatment and post-treatment 1 (p=0.008, p=0.037, respectively).
- FOC of *Elodea canadensis* demonstrated an increase on average with statistical significance from a t-test. However, our analysis couldn't validate this result as no normality test was completed and the non-parametric Wilcoxon signed-rank test indicated that the difference between pre-treatment and post-treatment 1 was not significant for *Elodea canadensis*.
- All other non-target native species that were analyzed demonstrated no statistically significant difference between pre-treatment and post-treatment 1.

Post-treatment 2

Analysis of the results from the second post treatment survey show that:

- FOC of *Myriophyllum spicatum* (target species) decreased between pre-treatment and post treatment 2 (p=0.008).
- FOC of *Potamogeton illinoensis* (non-target species) increased between pre-treatment and post-treatment 2 (p=0.016).
- FOC of *Ceratophyllum demersum* (non-target species) decreased between pre-treatment and post-treatment 2 (p=0.039).
- All other non-target native species that were analyzed demonstrated no statistically significant difference between pre-treatment and post-treatment 2.

Future Research

Future research into the effects of ProcellaCOR EC on non-target species should focus on *Brasenia schreberi* and *Ceratophyllum demersum* in all the lakes where the herbicide is utilized. It is possible that there may be other confounding factors that contributed to the significant change in *Ceratophyllum demersum* populations and more data would help to clarify the relationship between this non-target species and ProcellaCOR EC.

Further, there were non-target species that increased in FOC, and this difference was found to be statistically significant. This information could aid not only in our understanding on effects of ProcellaCOR EC, but also help stakeholder groups in the decision to use or not use the herbicide. If native aquatic macrophyte species have increased FOC after *Myriophyllum spicatum* populations decline, this may contradict the notion that there would be reduced submerged aquatic vegetation habitat following control of *Myriophyllum spicatum*. To address this, future research might involve comparing the exact FOC reduction of *Myriophyllum spicatum* against the FOC increase in other species of submerged aquatic vegetation within each waterbody.

This research may benefit from completing a power analysis to identify the minimum sample size needed to determine the effect of ProcellaCOR EC most accurately between pre- and post-treatment FOC.

Additional Resources

Beets, J., M. Heilman, and M. D. Netherland. 2019. *Large-scale mesocosm evaluation florpyrauxifen benzyl, a novel arylpicolinate herbicide on Eurasian and hybrid watermilfoil and seven native submersed plants.* Journal of Aquatic Plant Management 57:49-55.

Mudge, Christopher R., Sartain, Bradley T., Sperry, Benjamin P., and Getsinger, Kurt D. 2021. *Efficacy of Florpyrauxifen-benzyl for Eurasian Watermilfoil Control and Nontarget Illinois Pondweed, Elodea, and Coontail Response.* https://hdl.handle.net/11681/42063

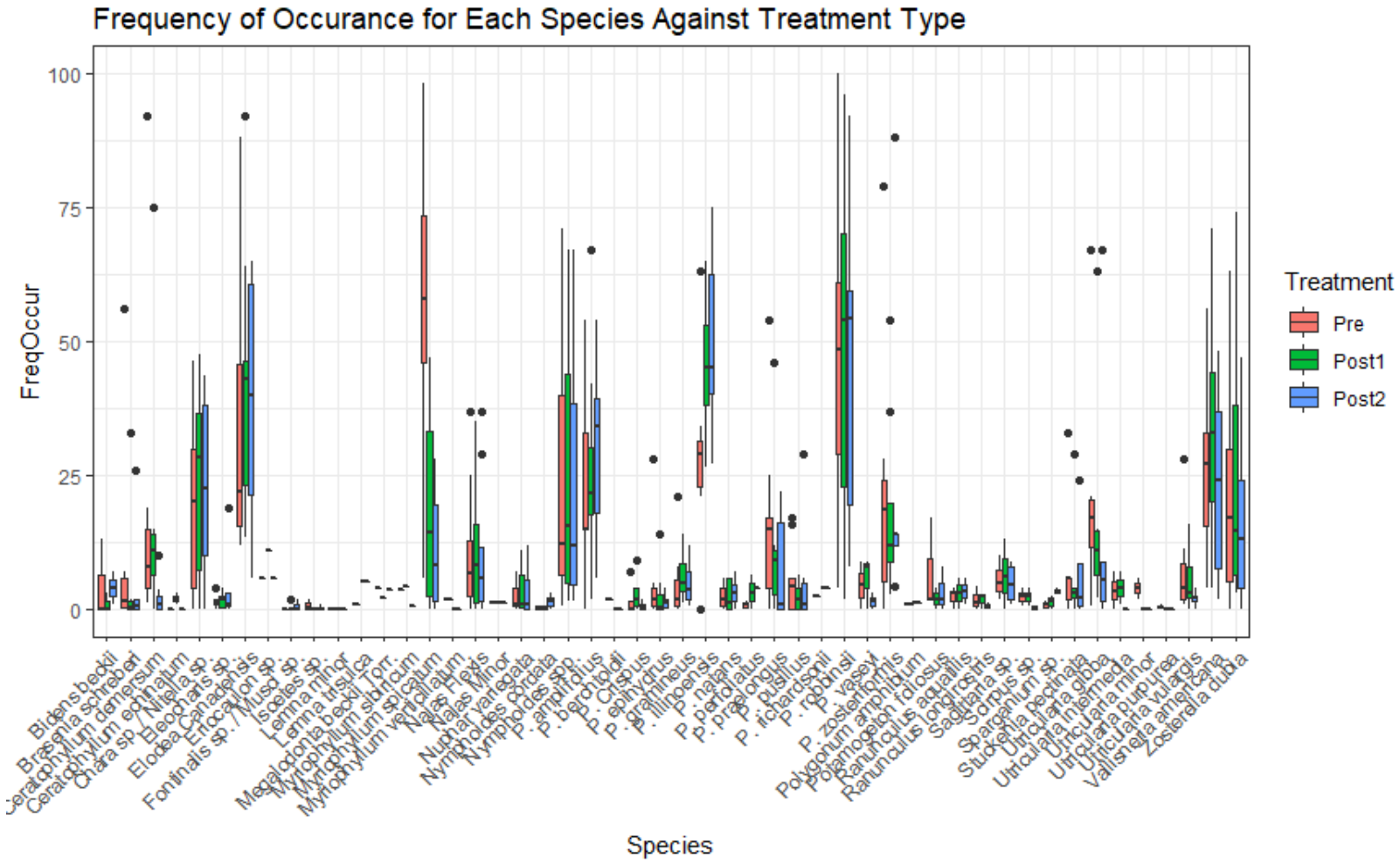

Figure 1: Frequency of occurrence (%) for species analyzed in this study with pre-treatment, post-treatment 1, and post-treatment 2. “P.” is shorthand for Potamogeton. Outliers are indicted as dots.

Species	n.pairs	meanPre	meanPost1	t.test.pvalue	t.test.sign	medPre	medPost1	wilcox.pvalue	wilcox.sign
Brasenia schreberi	6	11.6	6	0.18227	not significant	3.3	0.4	0.10035	not significant
Ceratophyllum demersu	9	19.3	15.8	0.19197	not significant	9	11	0.23395	not significant
Eleocharis sp.	6	1.6	1.7	0.91744	not significant	1.4	1.8	1	not significant
Elodea Canadensis	10	32.8	41.3	0.04904	SIGNIFICANT	22	43	0.05041	not significant
Myriophyllum spicatum	10	58.4	19.3	0.00062	SIGNIFICANT	60	14.2	0.00589	SIGNIFICANT
Najas Flexis	10	10.4	11.2	0.47864	not significant	6.7	8.2	0.77891	not significant
Nuphar variegata	7	2.4	3.7	0.25298	not significant	1	1	0.26929	not significant
Nymphoides spp.	9	24.2	24	0.91033	not significant	12.2	15.6	0.91016	not significant
P. amplifolius	10	23.5	26.2	0.31447	not significant	15	21.6	0.28462	not significant
P. epihydrus	8	5.6	2.7	0.15758	not significant	3	0.4	0.17552	not significant
P. gramineus	7	5.1	6.3	0.60124	not significant	2	5	0.68447	not significant
P. illinoensis	9	28.7	44.8	0.00828	SIGNIFICANT	29	45	0.00781	SIGNIFICANT
P. praelongus	9	15.7	10.7	0.50914	not significant	15	9.1	0.44065	not significant
P. pusillus	9	5.8	2.3	0.2149	not significant	4.3	2	0.30963	not significant
P. robbinsii	9	49.6	48.5	0.59098	not significant	48.4	53.9	0.59363	not significant
P. zosteriformis	8	21.9	18.6	0.4563	not significant	18.5	12	0.57493	not significant
Vallisneria americana	11	25.5	34	0.03302	SIGNIFICANT	27.1	33	0.03658	SIGNIFICANT
Zosterella dubia	10	20.3	24.3	0.39708	not significant	17.2	14.6	0.55664	not significant
Chara sp. / Nitella sp.	10	20.4	24.6	0.25045	not significant	20	28.2	0.25997	not significant
P. Crispus	7	1.4	2.9	0.36442	not significant	0	2	0.58071	not significant
Utricularia vulargis	7	7.7	5.6	0.35554	not significant	4	3	0.375	not significant
P. natans	5	2.5	2.7	0.9177	not significant	2	1.4	1	not significant
Utricularia gibba	6	22.2	18	0.03487	SIGNIFICANT	17	10.9	0.05848	not significant
Stuckenia pectinata	5	9.3	7.7	0.85213	not significant	5.7	3.5	0.78927	not significant

Table 1: Plant species analyzed in this study with number of pairs, mean pre-treatment FOC (%), mean post-treatment 1 FOC (%), t-test p-values and significance between pre-treatment and post-treatment 1, median pre-treatment 1 FOC (%), median post-treatment 1 FOC (%) and Wilcoxon signed-ranks test p-value and significance between pre-treatment and post-treatment 1. “P.” signifies Potamogeton species.

Species	n.pairs	meanPre	meanPost2	t.test.pvalue	t.test.sign	medPre	medPost2	wilcox.pvalue	wilcox.sign
Brasenia schreberi	**5**	12.5	5.8	0.31117	not significant	1.6	0.8	0.18145	not significant
Ceratophyllum demersu	**8**	18.2	2.2	0.18896	not significant	7	0.8	0.03906	SIGNIFICANT
Elodea Canadensis	**7**	33.4	39.2	0.51694	not significant	21	40	0.67499	not significant
Myriophyllum spicatum	**8**	53.5	10.8	0.00387	SIGNIFICANT	58.5	8.2	0.00781	SIGNIFICANT
Najas Flexis	**8**	10.8	10.7	0.91196	not significant	6.5	5.8	1	not significant
Nuphar variegata	**6**	1.6	3.5	0.22567	not significant	0.9	0.9	0.37109	not significant
Nymphoides spp.	**7**	23.6	23.8	0.91183	not significant	8	12	0.83348	not significant
P. amplifolius	**7**	25.6	29.8	0.26049	not significant	15	34	0.41637	not significant
P. epihydrus	**6**	2.4	1.4	0.10288	not significant	2.8	1.2	0.10035	not significant
P. gramineus	**5**	5.5	5.1	0.87125	not significant	2	3.8	0.85513	not significant
P. illinoensis	**7**	24.9	50.4	0.02164	SIGNIFICANT	29	45	0.01563	SIGNIFICANT
P. praelongus	**6**	19.6	7.5	0.23324	not significant	16	1	0.20716	not significant
P. pusillus	**6**	7.1	6.2	0.8085	not significant	4.8	1	1	not significant
P. robbinsii	**7**	46.1	44.6	0.48749	not significant	48.4	54.1	0.41849	not significant
P. zosteriformis	**5**	23.6	26.4	0.48607	not significant	14	14	0.46145	not significant
Vallisneria americana	**8**	20.9	23.2	0.51289	not significant	24.6	24.2	0.67405	not significant
Zosterella dubia	**7**	17.4	16.6	0.93881	not significant	6	13	0.8125	not significant
Chara sp. / Nitella sp.	**7**	23.7	23.2	0.79127	not significant	29.5	22.6	1	not significant
Utricularia vulgaris	**5**	22.5	16.6	0.12051	not significant	15	5.6	0.20124	not significant

Table 2: Plant species analyzed in this study with number of pairs, mean pre-treatment FOC (%), mean post-treatment 2 FOC (%), t-test p-values and significance between pre-treatment and post-treatment 2, median pre-treatment 2 FOC (%), median post-treatment 2 FOC (%) and Wilcoxon signed-ranks test p-value and significance between pre-treatment and post-treatment 2. “P.” signifies Potamogeton species.