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Combining Endothall with Other Herbicides for Improved Control of Hydrilla — A Field Demonstration

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PURPOSE: This technical note describes the results of a field demonstration to examine the effectiveness of combining endothall applied as Aquathol® K with other aquatic herbicides for control of hydrilla. Concentrations and combinations were based on previous greenhouse trials conducted at the Lewisville Aquatic Ecosystem Research Facility (LAERF) in Lewisville, Texas, using Aquathol® K, Reward® (diquat), Hydrothol® 191 (endothall) and Cutrine®-Plus (chelated copper).

BACKGROUND: The use of herbicide combinations is a widespread and documented practice in the agricultural community but is a less defined practice in aquatic vegetation management. Herbicide combinations may offer significant advantages over the use of a single material, including:

- Improved and extended weed control.
- Reduced herbicide rates and application costs.
- Shorter contact times for improved results in flowing water.
- Less stringent use restrictions.
- Improved selectivity.

While interest in the use of herbicide combinations is increasing with field applicators, little research has been conducted to evaluate effectiveness of these combinations. Identifying appropriate herbicide rates and combinations is essential for efficient and effective aquatic weed control.

Previous research has shown that combining diquat [6,7-dihydrodipyrido (1,2-α;2',1'-c) pyrazinediium dibromide] and copper increased control (Mackenzie and Hall 1967; Sutton et al. 1970, 1972) of target plants and showed a "quickened knockdown" of hydrilla (*Hydrilla verticillata* (L.f.) Royle). Additionally these studies suggest that combining the two herbicides increased the uptake of each.

Concentration exposure time (CET) studies (Netherland et al. 1991) showed that the dipotassium salt of endothall (7-oxabicyclo [2,2,1] heptane-2,3-dicarboxylic acid), hereafter referred to as endothall AQ (applied as Aquathol® K), is an effective herbicide for controlling hydrilla. Additional studies showed that several exotic aquatic plants, including hydrilla and Eurasian watermilfoil (*Myriophyllum spicatum* L.) can be selectively controlled by applying endothall AQ at lower rates for longer periods of contact time, with minimal to no visible damage to many native aquatic plant species (Skogerboe and Getsinger 2001, 2002).

A small-scale greenhouse study (Pennington et al. 2001) was conducted to evaluate the potential of using endothall AQ in combination with other herbicides including diquat applied as the Reward® formulation, the amine of endothall applied as Hydrothol® 191 (endothall HY), and chelated copper applied as Cutrine®-Plus (copper) to control hydrilla. Treatments included three rates of endothall AQ (1, 2, and 3 mg/L active ingredient [ai]) applied alone or in combination with either copper (0.5 mg/L ai), endothall HY (0.2 mg/L ai), or diquat (0.5 mg/L ai salt). Results of this study showed low rates of endothall AQ (1 mg/L ai) combined with either copper (0.5 mg/L ai) or diquat (0.5 mg/L ai) resulted in as good or better control than much higher rates of endothall (3 mg/L ai) when used alone. Endothall HY used in combination with endothall AQ also increased hydrilla control compared to endothall AQ alone but not as well as the higher rates of endothall AQ.

Using reduced rates of herbicides in combination allows for greater efficiency and may make the herbicides more valuable and desirable as tools for aquatic plant managers. To demonstrate the effectiveness of herbicide combinations under more rigorous conditions, a field demonstration was conducted at Toledo Bend Reservoir, Louisiana, in the summer and fall of 1999.

MATERIALS AND METHODS: Seven plots were established in two coves on Toledo Bend Reservoir, Louisiana, a 74,867-ha (185,000-acre) impoundment on the Texas and Louisiana border. Five rectangular plots were established in Diamond Cove and two in Yokum Bay, marked using buoys at the corners of each plot. All plots bordered the shoreline and were characterized by a high density of hydrilla, growing at or just below the water surface. Each 2-ha (5-acre) plot was measured using a Bushnell Yardage Pro 800 laser range finder (Bushnell Corporation, Overland Park, KS).

Herbicides were applied from an airboat using a dual-tank sprayer with submersed injectors that kept products separated to prevent unwanted interaction resulting from mixing of herbicide concentrates. Application rates for each herbicide and plot are shown in Table 1.

Table 1
Herbicide Combination Application Rates for Plots of Hydrilla on Toledo Bend Reservoir, LA, September 1999[1]

Plot No.	Treatment
1	Endothall AQ, 1 mg/L ai + endothall HY, 0.2 mg/L ai
2	Endothall AQ, 1 mg/L ai + copper, 0.5 mg/L ai
3	Endothall AQ 1.5 mg/L ai + diquat, 0.21 mg/L ai salt
4	Endothall AQ, 3 mg/L ai + diquat, 0.21 mg/L ai salt
5	Endothall AQ, 3 mg/L ai + copper, 0.5 mg/L ai
6	Endothall AQ, 3 mg/L ai
7	Untreated reference

[1] Endothall AQ applied as Aquathol K, endothall HY applied as Hydrothol 191.

An inert fluorescent dye, rhodamine WT, was injected into the plots at the time of herbicide application to quantify the water exchange in the treatment plots. The dye is used to estimate herbicide dissipation resulting from water movement and does not account for loss of herbicide resulting from photo or biological degradation. Water samples were collected at 1, 3, and 6 hr after treatment (HAT) and 1, 2, 3, 4, and 5 days after treatment (DAT). These samples were analyzed for dye concentrations using a Turner Designs 10AU fluorometer (Turner Designs, Inc, Sunnyvale, CA). The dissipation half-life was calculated from an equation determined by a linear regression where the dependent variable was log [dye concentration (ug/L)], and the independent variable was time (hours). Half-lives were calculated for each plot using 0 to 72 HAT and 0 to 120 HAT.

Shoot biomass evaluations were conducted pre-treatment and 3, 8, and 12 weeks after treatment (WAT), and 12 months after treatment (MAT). Biomass samples were collected by lowering a 35-cm-wide garden rake attached to a 3-m pole vertically to the lake bottom. The rake was slowly turned and simultaneously lifted up to the boat to collect plant material in the sample area cylinder. Plants were returned to the laboratory and dried to a constant weight at 80°C. Percent control was calculated by dividing a post-treatment biomass mean value by the pretreatment mean biomass value. Biomass means were compared between sample periods for each plot using Analysis of Variance (ANOVA).

RESULTS: Dye dissipation data are summarized in Table 2. For comparison, the simulated water exchange, half-life used in the previously conducted small-scale studies was approximately 24 hr (Pennington et al. 2001). All hydrilla biomass data are summarized in Table 3, and percent control data are summarized in Table 4.

Table 2
Rhodamine WT Half Lives (hours) Calculated for 72 and 120 hr After Treatment (HAT)[1]

Plot	Treatment	72 HAT		120 HAT	
		Half-Life	R-sq	Half-Life	R-sq
1	Endothall AQ, 1 mg/L ai + endothall HY, 0.2 mg/L ai	135[2]	0.18	90	0.50
2	Endothall AQ, 1 mg/L ai + copper, 0.5 mg/L ai	65	0.55	95	0.58
3	Endothall AQ, 1.5 mg/L ai + diquat, 0.21 mg/L ai salt	45	0.59	42	0.42
4	Endothall AQ, 3 mg/L ai + diquat, 0.21 mg/L ai salt	83	0.45	35	0.69
5	Endothall AQ, 3 mg/L ai + copper, 0.5 mg/L ai	27	0.35	22	0.61
6	Endothall AQ, 3 mg/L ai	23	0.49	23	0.67
7	Untreated Reference	ND[3]	ND	ND	ND

[1] Endothall AQ applied as Aquathol K, endothall HY applied as Hydrothol 191.
[2] Model was not significant ($p < 0.05$).
[3] ND - no data.

Table 3
Mean Hydrilla Biomass (g dry weight) Collected 3, 8, and 12 Weeks After Treatment (WAT), and 12 Months After Treatment (MAT)[1,2]

Plot No.	Treatment	Pretreatment	3 WAT	8 WAT	12 WAT	12 MAT
1	Endothall AQ, 1 mg/L ai endothall HY, 0.2 mg/L ai	52.7 ±11.1 A[3]	6.5 ± 5.1 B	4.9 ± 2.8 B	2.9 ± 1.9 B	0.9 ± 0.6 B
2	Endothall AQ, 1 mg/L ai copper, 0.5 mg/L ai	65.0 ± 9.1 A	2.8 ± 0.7 B	3.6 ± 1.3 B	4.7 ± 1.1 B	6.9 ± 4.2 B
3	Endothall AQ, 1.5 mg/L ai diquat, 0.21 mg/L ai salt	37.5 ± 3.1 A	24.9 ± 3.6 A	7.7 ± 3.5 B	3.0 ± 1.2 B	50.0 ± 21.8 A
4	Endothall AQ, 3 mg/L ai diquat, 0.21 mg/L ai salt	33.2 ± 5.4 A	7.9 ± 2.3 B	8.0 ± 1.5 B	11.0 ± 5.7 B	55.3 ± 6.3 A
5	Endothall AQ, 3 mg/L ai copper, 0.5 mg/L ai	78.6 ± 13.4 A	13.9 ± 2.5 B	19.2 ± 3.8 B	10.5 ± 3.5 B	95.8 ± 8.3 A
6	Endothall AQ, 3 mg/L ai	38.2 ± 8.6 B	5.6 ± 3.8 D	14.3 ± 3.5 C	7.0 ± 1.1 CD	155.6 ± 19.9 A
7	Untreated Reference	71.6 ± 15.3 B	62.1 ± 11.4 B	56.6 ± 13.2 B	215.8 ± 29.6 A	185.6 ± 28.2 A

[1] Endothall AQ applied as Aquathol K, endothall HY applied as Hydrothol 191.
[2] Means followed by same letter are not significantly different between sample periods for each plot.
[3] ± standard error.

Table 4
Hydrilla Control Based on Biomass Data at 3, 8, and 12 Weeks After Treatment (WAT): and 12 Months After Treatment (MAT)[1]

Plot No.	Treatment	3 WAT	8 WAT	12 WAT	12 MAT
1	Endothall AQ, 1mg/L ai Endothall HY, 0.2 mg/L ai	88 percent	91 percent	94 percent	98 percent
2	Endothall AQ, 1 mg/L ai Copper, 0.5 mg/L ai	96 percent	94 percent	99 percent	89 percent
3	Endothall AQ, 1.5 mg/L ai Diquat, 0.21 mg/L ai salt	34 percent	79 percent	92 percent	-33 percent
4	Endothall AQ, 3 mg/L ai Diquat, 0.21 mg/L ai salt	76 percent	76 percent	67 percent	-67 percent
5	Endothall AQ, 3 mg/L ai Copper, 0.5 mg/L ai	82 percent	76 percent	87 percent	-22 percent
6	Endothall AQ, 3 mg/L ai	85 percent	63 percent	82 percent	-307 percent
7	Untreated Reference	13 percent	21 percent	-201 percent	-159 percent

[1] Endothall AQ applied as Aquathol K, endothall HY applied as Hydrothol 191.

Plot 1: Endothall AQ, 1 mg/L ai + endothall HY, 0.2 mg/L ai. Dye dissipation half-life in Plot 1 was 135 hr based on the 72 HAT data and 90 hr when calculated using 120 HAT data. The linear regression model used to calculate the half-life from the 72 HAT data was not significant ($P < 0.05$), which suggests that little water exchange occurred in Plot 1 during the first 72 HAT. Plant biomass from Plot 1 was significantly less 3 WAT compared to pretreatment biomass from the same plot. Based on visual observations, most plants dropped from the water column 5 to 6 days after treatment (DAT). By 3 WAT, 88 percent control of hydrilla had been achieved and 94 percent control had occurred by 12 WAT. However, there were no significant differences in biomass among the 3, 8, and 12 WAT. By 12 MAT, recovery of hydrilla still had not occurred with 98 percent control measured.

Plot 2: Endothall AQ, 1 mg/L ai + copper, 0.5 mg/L ai. Dye dissipation half-life was 65 hr based on the 72 HAT data and 97 hr based on the 120 HAT data. Plant biomass from Plot 2 was significantly less 3 WAT compared to pretreatment biomass from the same plot. Based on visual observations, most plants dropped from the water column 5 DAT. By 3 WAT, 96 percent control of hydrilla had been achieved, 94 percent at 8 WAT, and 93 percent at 12 WAT. There were no significant differences in biomass among the 3, 8, and 12 WAT; and 12 MAT. Little recovery of hydrilla was observed by 12 MAT, with 90 percent control measured.

Plot 3: Endothall AQ, 1.5 mg/L ai + diquat, 0.21 mg/L ai salt. The dye half-life calculated using the 72 HAT data was 45 hr, and 42 hr based on the 120 HAT data. Plant biomass from Plot 3 was significantly less 3 WAT compared to pretreatment biomass from the same plot. Based on visual observations, most plants dropped from the water column 5 to 6 DAT. By 3 WAT, only 33-percent control of hydrilla had been achieved, but increased to 79 percent at 8 WAT and 92 percent at 12 WAT. Biomass at 8 WAT and 12 WAT was significantly less than pre-treatment biomass. By 12 MAT, the mean biomass was 33 percent greater than the pretreatment biomass; however, this was not a statistical difference. Visual observations at 12 MAT showed that large portions of the eastern half of the plot had not recovered, while the western half showed very dense hydrilla growth. The western half of the plot was adjacent to a boat channel where rapid re-infestation most likely occurred from hydrilla fragments introduced by boat traffic.

Plot 4: Endothall AQ, 3 mg/L ai + diquat, 0.21 mg/L ai salt. The dye dissipation half-life was 83 hr based on 72 HAT dye data and 35 hr based on 120 HAT dye data. Plant biomass from Plot 4 was significantly less 3 WAT compared to pretreatment biomass from the same plot. Based on visual observations, most plants dropped from the water column 5 to 6 DAT. By 3 WAT, 76 percent control had been achieved, 76 percent at 8 WAT and 67 percent at 12 WAT. There were no significant differences in biomass between 3, 8, and 12 WAT. Complete recovery of hydrilla occurred by 12 MAT, and biomass was not significantly different than pretreatment levels.

Plot 5: Endothall AQ, 3 mg/L ai + copper, 0.5 mg/L ai. The dye dissipation half-life was 27 hr based on the 72 HAT dye data and 22 hr based on the 120 HAT dye data. Plant biomass from Plot 5 was significantly less at the 3 WAT evaluation compared to pretreatment biomass from the same plot. Based on visual observations most plants dropped from the water column about 5 to 6 DAT. By 3 WAT, 82 percent control had been achieved, 76 percent at 8 WAT and 87 percent at 12 WAT. There were no significant differences in biomass between 3, 8, and 12 WAT. Complete recovery of hydrilla occurred by 12 MAT, and biomass was not significantly different than pretreatment levels.

Plot 6: Endothall AQ, 3 mg/Lai. The dye dissipation half-life was 23 hr based on both the 72 HAT dye data and 120 HAT data. Plant biomass from Plot 6 was significantly less 3 WAT compared to pretreatment biomass from that plot. Based on visual observations, most plants dropped from the water column 5 to 6 DAT. By 3 WAT, 85-percent control had been achieved, 63 percent at 8 WAT and 82 percent at 12 WAT. There were no significant differences in biomass at 3, 8, and 12 WAT. Complete recovery of hydrilla occurred by 12 MAT; however unlike other plots, biomass was significantly greater than pretreatment levels.

Plot 7: Untreated reference. There were no significant changes in hydrilla biomass in the untreated reference, Plot 7, until 12 WAT and 12 MAT when biomass increased by 200 percent and 159 percent, respectively.

DISCUSSION: Previous small-scale, controlled experiments (Pennington et al. 2001) have illustrated the usefulness of combining endothall AQ with copper, diquat, or endothall HY for effective control of hydrilla using lower rates of herbicides. Additional small-scale CET studies (Netherland et al. 1991) showed that herbicide exposure time was as important to efficacy as herbicide application rates. Results of this field demonstration support conclusions from the small-scale studies where lower concentrations of herbicide combinations control hydrilla equal to or better than higher concentrations of herbicides used alone. Plots were selected to be as similar to each other as possible; however, dye dissipation data (water exchange) suggest that herbicide exposure time varied greatly, and may have had as great an effect on efficacy as application rates. These field results emphasize the importance and need for controlled, replicated small-scale studies to develop valid recommendations for operational control of hydrilla.

Plots treated with the lower rates of endothall AQ (1 to 1.5 mg/L) combined with other herbicides tended to have increased hydrilla control and slower biomass recovery, but these plots (1, 2, and 3) also had longer dye dissipation rates (i.e. less water exchange). Plot 4, however, was treated with a high rate of endothall AQ (3 mg/L) combined with diquat and had a dye dissipation rate similar to that of the plots treated with low rates of endothall AQ and either endothall HY, copper, or diquat. Biomass data showed that hydrilla control on Plot 4 was less than 70 percent at 12 WAT compared to greater than 90 percent for Plots 1, 2, and 3. Biomass data also showed that hydrilla had completely recovered 12 MAT in Plot 4 but not in Plots 1 and 2. These data suggest that higher rates of endothall AQ may quickly burn off plant material without killing all of the plant. Lower rates of endothall AQ may allow more time for endothall uptake, resulting in improved efficacy and slower recovery of hydrilla.

FUTURE WORK: Future research will focus on evaluating other herbicide combinations and timing of treatments to improve the management of invasive aquatic plant species such as Eurasian watermilfoil and curly leaf pondweed (*Potamogeton crispus* L.). Initially, these evaluations will be conducted in controlled, small-scale systems by the U.S. Army Engineer Research and Development Center (ERDC). The most promising results will then be verified in selected field sites across the United States.

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