

# American frogbit response to herbicides

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## ABSTRACT

*Limnobium spongia* (frogbit) is a free-floating aquatic plant that can produce extensive floating mats causing negative ecological, social, and economic impacts that can harm aquatic fauna (i.e., dissolved oxygen depletion) and restrict human uses of water. Literature describing effective control measures for frogbit is minimal. Efficacy of high and low doses of seven foliar-applied herbicides (2,4-D, floryprauxifen-benzyl, flumioxazin, glyphosate, imazamox, imazapyr, and triclopyr) were evaluated in a mesocosm setting in the summers of 2018, 2020, and 2021. Both emergent and submersed frogbit biomass were reduced at least 99% by imazamox (0.56 and 1.11 kg ai ha<sup>-1</sup>) and imazapyr (0.42 and 0.84 kg ae ha<sup>-1</sup>) 8 wk after treatment (WAT) compared with nontreated reference plants. Triclopyr (6.71 kg ai ha<sup>-1</sup>) reduced frogbit biomass 92% and flumioxazin (0.42 kg ai ha<sup>-1</sup>) reduced biomass 87 to 93% compared with reference plants. 2,4-D (2.12 and 4.24 kg ae ha<sup>-1</sup>), glyphosate (2.83 and 5.67 kg ai ha<sup>-1</sup>), triclopyr (3.36 kg ae ha<sup>-1</sup>), floryprauxifen-benzyl (0.02 and 0.05 kg ai ha<sup>-1</sup>), and flumioxazin (0.21 kg ai ha<sup>-1</sup>) did not reduce frogbit biomass 8 WAT compared with reference plants. Future research should consider the efficacy of different herbicide combinations to control frogbit, as well as the role of diluent volume per unit area, especially with imazamox and imazapyr. Field studies also will be useful in determining whether the results observed in this study will translate to management of frogbit in natural settings.

**Key words:** aquatic plant control, foliar herbicide application, *Limnobium spongia*, native nuisance species.

## INTRODUCTION

*Limnobium spongia* (Bosc) Rich. ex Steud. (American frogbit; hereafter frogbit) is a typically free-floating aquatic plant in the family Hydrocharitaceae, although it can occasionally be found growing as a rooted plant in moist soil environments (Cook and Urmi-König 1983; Les 2020). Frogbit is widely distributed throughout the southern United States and occasionally found in the northeastern United States (Mackenzie 1922; Wilder 1974; Les and

Mehrhoff 1999) growing in still or slow-flowing waterways such as ponds, lakes, and canals (Langeland et al. 1995) with a range of light and nutrient availabilities (Cook and Urmi-König 1983; USDA 2019). Frogbit foliage is very similar to that of the invasive water hyacinth (*Eichhornia crassipes* (Mart.) Solms) and identification can be difficult when the two species co-occur (Turnage, pers. comm.). Frogbit is consumed by waterfowl, insects, aquatic invertebrates, and reptiles and can serve as habitat for insects and invertebrates (Platt et al. 2013; Les 2020).

Frogbit can reproduce sexually and vegetatively through daughter plants (Cook and Urmi-König 1983; Les and Mehrhoff 1999) that can produce extensive floating mats that cause negative ecological, social, and economic impacts (Knight 1985; Bodle 1986; Madsen et al. 1998). Long-distance dispersal and range expansion of frogbit can occur through transport of seeds or plant fragments by waterfowl and human activities involved with the water garden industry and boating equipment (Les and Capers 1999; Les and Mehrhoff 1999; Bowles 2013; Gettys 2019).

Literature describing effective control measures for frogbit is minimal, in part because native plants are often considered of minor importance in aquatic plant management programs (Langeland et al. 1995). To date, there are no known biological controls for frogbit. Effective mechanical management would involve shredding, harvesting, digging, or cutting plants, which can be expensive, time consuming, and may cause further spread because of plant fragmentation (Clayton 1996; Madsen et al. 2017; Turnage et al. 2019). Physical control options (i.e., drawdown) are likely to be inefficient as frogbit can survive in moist soil environments (Cook and Urmi-König 1983; Howard and Wells 2009; Les 2020). Therefore, chemical control is the most common frogbit management strategy because of its cost effectiveness and ease of implementation; however, there are minimal data regarding reduction of frogbit by herbicide treatments (Langeland et al. 1995, Madsen et al. 1998). Langeland et al. (1995) assessed the efficacy and importance of the herbicides diquat and 2,4-D (alone and in combination) and glyphosate alone to control frogbit as a foliar spray. In a similar study, Madsen et al. (1998) tested the efficacy of foliar applications of the herbicides diquat, 2,4-D, glyphosate, and triclopyr to control frogbit. However, there are approximately 16 herbicides labeled for general aquatic use in the United States, many of which were not registered when previous frogbit control studies were conducted (Schardt and Netherland 2020).

Literature regarding frogbit control has focused on foliar applications from boat-based platforms (Langeland et al. 1995, Madsen et al. 1998). In addition to boat-based operations, foliar herbicide applications can also be applied from aerial platforms (e.g., helicopter or fixed wing). Boat-

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based application techniques usually dilute herbicide in more water (approximately 935 L ha<sup>-1</sup> [100-gal total solution per acre]), whereas aerial uses less water to reduce weight of the aircraft (< 280 L ha<sup>-1</sup> [ $< 30$  gal ac<sup>-1</sup>]). This results in better coverage (wetter leaves) from a boat but weaker spray droplets (less herbicide per droplet), whereas aerial applications have lower coverage (fewer droplets falling on leaves) but stronger droplets (more herbicide per droplet). Moreover, aerial application techniques can reach remote areas that are a challenge for ground access, allowing for the treatment of nursery frogbit populations in areas where boat access is limited.

Resource managers at the Sam D. Hamilton Noxubee National Wildlife Refuge (33°16'15.8016"N; 88°47'1.0926"W) near Starkville, MS have been attempting to control nuisance populations of American lotus (*Nelumbo lutea* Willd.), white waterlily (*Nymphaea odorata* Aiton), watershield (*Brasenia schreberi* J.F. Gmel.), and to a lesser extent frogbit in Bluff Lake (485 ha), Loakfoma Lake (242 ha), and Doyle Arm Lake (17 ha) over the last decade. In 2016, nuisance plant growth was estimated to cover more than 60% of Bluff Lake (291 ha), 85% of Loakfoma Lake (206 ha), and 50% of Doyle Arm Lake (9 ha; Steven Lewis, pers. comm.). Because of the areal extent of nuisance vegetation (515 ha [1,273 ac]), resource managers selected aerial applications via helicopter as the most cost-effective means of plant control. However, no information was available regarding the response of frogbit to herbicide residues in low-diluent volumes utilized in helicopter herbicide applications. Therefore, the purpose of this study was to explore the efficacy of high and low rates of seven herbicides applied at simulated aerial application carrier volumes for control of frogbit.

## MATERIALS AND METHODS

We conducted this study at the Aquatic Plant Research Facility at Mississippi State University's R.R. Foil Plant Research Center. Each year of the study (2018, 2020, and 2021), consisted of 48 outdoor mesocosms 242 L (64 gal) in size. Frogbit was inoculated into each mesocosm (five similarly sized rosettes per mesocosm) and given 2 mo to establish before herbicide applications were made in the summer.

Each experiment consisted of a nontreated reference and foliar applications of 14 herbicide treatments: 2,4-D<sup>1</sup> (2.12 and 4.24 kg ae ha<sup>-1</sup>), glyphosate<sup>2</sup> (2.83 and 5.67 kg ai ha<sup>-1</sup>), triclopyr<sup>3</sup> (3.36 and 6.71 kg ae ha<sup>-1</sup>), imazamox<sup>4</sup> (0.56 and 1.11 kg ai ha<sup>-1</sup>), imazapyr<sup>5</sup> (0.42 and 0.84 kg ae ha<sup>-1</sup>), florpyrauxifen-benzyl<sup>6</sup> (0.02 and 0.05 kg ai ha<sup>-1</sup>), and flumioxazin<sup>7</sup> (0.21 and 0.42 kg ai ha<sup>-1</sup>). These herbicides were chosen on the basis of those available to resource managers at the Noxubee National Wildlife Refuge when controlling American frogbit and other nuisance aquatic plant species (Steven Lewis, pers. comm.). Herbicide treatments represented half-maximum and maximum rates allowed by herbicide labels. A 1% v/v nonionic surfactant<sup>8</sup> was added to each treatment. Treatments (reference plus herbicide treatments) were randomly assigned to mesocosms, and each treatment was replicated three times (per

year) for a total of 45 treatment mesocosms. An additional three mesocosms were established for collection of pre-treatment specimens for a total of 48 mesocosms per experiment.

Before herbicide application, pretreatment plant specimens were harvested from three mesocosms by randomly placing two square floating polyvinyl chloride frames (0.1 m<sup>2</sup> in size) on top of the plant mat in mesocosms and harvesting all plant biomass within the frames to establish a baseline of plant growth. Harvested plant biomass was separated into emergent and submersed biomass, placed in labeled paper bags, and dried in a forced-air oven at 70 C for 3 d. After drying, plant biomass was weighed, and weights recorded.

Herbicide solutions were applied at a diluent rate of 280 L ha<sup>-1</sup> (30 gal ac<sup>-1</sup>) using a CO<sub>2</sub>-pressurized backpack sprayer calibrated to deliver herbicide treatments at 275.8 kPa (40 psi) with a TeeJet 8002EVS nozzle. At 8 wk after treatment (WAT), plants were harvested and processed from each mesocosm in the same manner as pretreatment specimens. Biomass data were subjected to mixed-model analysis of variance using treatment as a fixed effect and year (or trial replication) as a random effect. If differences existed in biomass, means were separated using the lsmeans function in R at the  $\alpha = 0.05$  significance level (R Core Team 2020).

## RESULTS AND DISCUSSION

Imazamox (0.56 and 1.11 kg ai ha<sup>-1</sup>) and imazapyr (0.42 and 0.84 kg ae ha<sup>-1</sup>) reduced emergent frogbit biomass by 99 to 100% at 8 WAT ( $P < 0.0001$ ) when compared with reference plants (Figure 1). Triclopyr (6.71 kg ae ha<sup>-1</sup>) and flumioxazin (0.42 kg ai ha<sup>-1</sup>) reduced emergent frogbit biomass 92 and 87% ( $P < 0.0001$ ), respectively, 8 WAT compared with reference plants (Figure 1). None of the other herbicide treatments reduced emergent frogbit biomass compared with reference plants 8 WAT (Figure 1); however, all other herbicide treatments except florpyrauxifen-benzyl (0.05 kg ai ha<sup>-1</sup>) had the same level of emergent biomass reduction as those treatments that were different from reference plants (Figure 1).

Imazamox (0.56 and 1.11 kg ai ha<sup>-1</sup>) and imazapyr (0.42 and 0.84 kg ae ha<sup>-1</sup>) also reduced submersed frogbit biomass by 99 to 100% 8 WAT ( $P < 0.0001$ ) when compared with reference plants (Figure 1). Triclopyr (6.71 kg ae ha<sup>-1</sup>) and flumioxazin (0.42 kg ai ha<sup>-1</sup>) reduced submersed frogbit biomass 92 and 93% ( $P < 0.0001$ ), respectively, 8 WAT compared with reference plants (Figure 1). All other herbicide treatments did not reduce submersed biomass 8 WAT compared with reference plants; however, all herbicide treatments except florpyrauxifen-benzyl treatments had the same level biomass reduction as those treatments that did reduce biomass compared with references (Figure 1).

Our work and others suggest that the auxinic herbicides 2,4-D and triclopyr may provide varying levels of frogbit control (Langeland et al. 1995; Madsen et al. 1998); however, the length of each study was different (4, 8, and 12 WAT). The longest study (12 WAT; Madsen et al. 1998) reported the

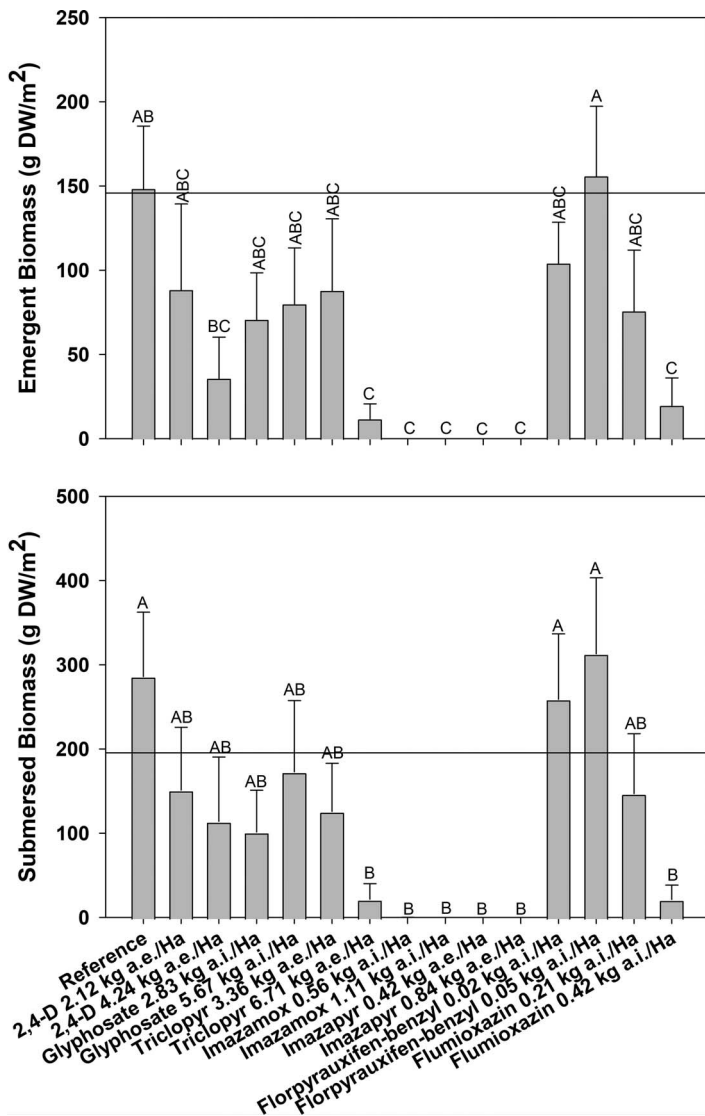


Figure 1. Emergent (top panel) and submersed frogbit biomass (bottom panel) 8 wk after foliar herbicide treatments; bars sharing the same letter are not different at  $\alpha = 0.05$  significance level ( $n = 9$ ); horizontal solid lines are pretreatment biomass; error bars are 1 standard error of the mean.

greatest levels of reduction, suggesting that the shorter studies may not have allowed enough time for 2,4-D symptomology to occur in treated plants. Langeland et al. (1995) found that 2,4-D (0.53, 1.06, 2.12, and 4.24 kg ae ha<sup>-1</sup>) did not reduce frogbit at 4 WAT, whereas Madsen et al. (1998) found that 2,4-D (1.08, 2.16, and 4.32 kg ae ha<sup>-1</sup>) significantly reduced frogbit biomass by 53 to 80% at 12 WAT. In our work, 2,4-D-treated plants had the same level of reduction as nontreated reference plants; however, 2,4-D-treated plants also had the same level of reduction as plants controlled 100% by imazapyr 8 WAT. At 12 WAT, Madsen et al. (1998) found that triclopyr (0.85, 1.69, 3.38 kg ae ha<sup>-1</sup>) gave excellent reduction of frogbit (87 to 95% biomass reduction), whereas our results (8 WAT) showed that plants treated with high rates of triclopyr (6.71 kg ae ha<sup>-1</sup>) were reduced 92% compared with reference plants, but frogbit treated with a lower triclopyr rate (3.36 kg ae ha<sup>-1</sup>) were not

affected. Our work, which was the first to investigate the use of florpyrauxifen-benzyl (0.02 and 0.05 kg ai ha<sup>-1</sup>) for frogbit control, did not detect any biomass reduction 8 WAT compared with reference plants (Figure 1). This suggests that different auxinic herbicide chemistries may exhibit vastly different effects on the same plant species.

To our knowledge, this is the first work to show that the acetolactate synthase (ALS)-inhibiting herbicides imazamox and imazapyr can control frogbit (Figure 1). Rates of imazamox (0.56 and 1.11 kg ai ha<sup>-1</sup>) and imazapyr (0.42 and 0.84 kg ae ha<sup>-1</sup>) used here are the half-maximum and maximum allowed by the herbicide labels (Anonymous 2020a,b) and provided 99 to 100% biomass reduction of frogbit. This is also the first work to investigate a protoporphyrinogen oxidase (PPO)-inhibiting herbicide (flumioxazin) for frogbit control. High rates of flumioxazin (0.42 kg ai ha<sup>-1</sup>) provided 87 to 93% reduction of frogbit 8 WAT (Figure 1), suggesting that flumioxazin may be useful for frogbit management. This is desirable because resource managers often want to use contact herbicides that provide rapid reduction of plant biomass rather than wait for slower-acting systemic herbicides to affect nuisance vegetation.

The diluent volume investigated here (280 L ha<sup>-1</sup>; 30 gal ac<sup>-1</sup>) is lower than those typically used in boat-based applications (> 700 L ha<sup>-1</sup> [ $> 50$  gal ac<sup>-1</sup>]). This may have reduced contact between herbicide droplets and frogbit foliage in mesocosms such that frogbit reduction was unlikely to occur. Langeland et al. (1995) found that increasing the total amount of herbicide solution per unit area containing the contact herbicide diquat (1.02 kg ai ha<sup>-1</sup>) from 935 to 1,870 L ha<sup>-1</sup> improved frogbit reduction from < 40 to > 90%, respectively, suggesting that greater diluent volume per unit area may increase frogbit reduction by attaining better coverage of plant foliage. However, Langeland et al. (1995) also found that doubling the rate of diquat from 1.02 to 2.04 kg ai ha<sup>-1</sup> provided the same level of frogbit control when mixed in less diluent (935 L ha<sup>-1</sup>) as the decreased herbicide rate (1.02 kg ai ha<sup>-1</sup>) in the higher diluent volume (1,870 L ha<sup>-1</sup>), suggesting that inefficient coverage of foliage can be overcome by increasing the amount of herbicide applied per unit area. Madsen et al. (1998) used similar 2,4-D and glyphosate rates as our work in higher volumes of diluent (935 L ha<sup>-1</sup>) but recorded biomass reduction compared with reference plants where we did not. Similar to Langeland et al. (1995), our work, taken with Madsen et al. (1998), suggests that higher diluent volume may provide better herbicide contact with frogbit foliage and thus greater biomass reduction. Therefore, the herbicides investigated here that did not reduce frogbit biomass should be investigated at higher diluent volumes to determine if better contact with frogbit foliage can improve biomass reduction by these herbicides.

Given the high level of biomass reduction observed by imazamox and imazapyr, other ALS-inhibiting herbicides labeled for aquatic use (penoxsulam and bispyribac-sodium) should be studied to determine if they will provide similar levels of control as observed here. Lower rates of imazamox and imazapyr should be investigated to determine if they can deliver the same level of frogbit control. Additionally,

tank mixes of systemic and contact herbicides as well as tank mixes of systemic herbicides with different modes of action (i.e., auxinic and ALS inhibitors) should also be investigated to determine if tank mixes can deliver better frogbit control than one herbicide alone. Because aerial applications can be conducted with one-third (93.5 L ha<sup>-1</sup> [10 gal ac<sup>-1</sup>]) or less of the diluent volume used here, future work should investigate the role of diluent volume per unit area in providing frogbit control from different herbicide application platforms (aerial vs. boat-based) so that resource managers will have suitable information to reference when making operational control decisions. Also, because surfactants can cost as much as herbicides and affect plant control outcomes, future work should also assess surfactant type and rate to give resource managers added information regarding the use of these products when planning plant control activities. Last, future work should investigate the timing of herbicide applications for reduction of frogbit biomass.

This work adds to the literature base regarding control of nuisance frogbit populations by providing evidence that frogbit is sensitive to ALS- (imazamox and imazapyr) and PPO (flumioxazin)-inhibiting herbicides. The lack of control by some herbicides tested here (glyphosate and florypyrauxifen-benzyl) may be of use to resource managers desiring to selectively release nonnuisance populations of frogbit from competition with invasive aquatic plants.

## SOURCES OF MATERIALS

<sup>1</sup>Weedar<sup>®</sup> 64 Broadleaf Herbicide, Nufarm Inc., 11901 S. Austin Ave., Alsip, IL 60803.

<sup>2</sup>Rodeo<sup>®</sup>, Dow AgroSciences LLC, 9330 Zionsville Rd., Indianapolis, IN 46268.

<sup>3</sup>Navitrol<sup>®</sup> Landscape and Aquatic Herbicide, Applied Biochemists, W175N11163 Stonewood Dr., Ste. 234, Germantown, WI 53022.

<sup>4</sup>Clearcast<sup>®</sup> Herbicide, SePRO Corporation, 11550 North Meridian St., Suite 600, Carmel, IN 46032.

<sup>5</sup>Habitat<sup>®</sup> Herbicide, SePRO Corporation, 11550 North Meridian St., Suite 600, Carmel, IN 46032.

<sup>6</sup>ProcellaCOR<sup>™</sup> SC, SePRO Corporation, 11550 North Meridian St., Suite 600, Carmel, IN 46032.

<sup>7</sup>Clipper<sup>®</sup> SC Aquatic Herbicide, Nufarm Inc., 11901 S. Austin Ave., Alsip, IL 60803.

<sup>8</sup>Top Surf<sup>®</sup>, Winfield Solutions, LLC, P.O. Box 64589 St. Paul, MN 55164.

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