

## Research Article

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Florpyrauxifen-benzyl; yellow floating heart, *Nymphoides peltata* (S.G. Gmel.) Kuntze; variable-leaf watermilfoil, *Myriophyllum heterophyllum* (Michx.)

**Keywords:**

Aquatic weeds; spray technology; drone; remote sensing; pesticide application

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# Performance of unoccupied aerial application systems for aquatic weed management: Two novel case studies

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**Abstract**

Unoccupied aerial application systems (UAAS) are gaining popularity for weed management to increase applicator safety and to deliver herbicide treatments where treatment sites limit ground-based spray equipment. Several studies have documented UAAS application strategies and procedures for weed control in terrestrial settings, yet literature describing remote spray technology for use in aquatics remains limited. Currently, applicators seek guidance for UAAS deployment for aquatic weed management to overcome site access restrictions, deal with environmental limitations, and improve ground-based applicator safety in hazardous treatment scenarios. In the present case studies, we evaluate a consumer-available UAAS to deliver the herbicide, florpyrauxifen-benzyl, as both foliar and directed in-water spray applications. The first case study showed that the invasive floating-leaved plant, yellow floating heart, was controlled 80% to 99% by 6 wk after treatment (WAT) following UAAS foliar herbicide treatments. The second case study demonstrated that UAAS directed in-water herbicide application reduced variable-leaf watermilfoil visible plant material by 94% at 5 WAT. Likewise, directed in-water applications from UAAS eliminated the need to deploy watercraft, which improved overall operational efficiency. Data from both case studies indicate that UAAS can provide an effective and efficient treatment strategy for floating-leaved and submersed plant control among common herbicide treatment scenarios. Future integration of UAAS in aquatic weed control programs is encouraged, especially among smaller treatment sites ( $\leq 4$  ha) or where access limits traditional spray operations.

**Introduction**

Active management of aquatic invasive plants (AIP) like Eurasian watermilfoil (*Myriophyllum spicatum* L.) and exotic floating hearts (*Nymphoides* spp.) is required to sustain native species diversity, mitigate ecologically damaging effects from resource competition (Madsen and Sand-Jensen 1991; Zhang and Boyle 2010), and minimize habitat degradation (Houlahan and Findlay 2004; Madsen 2014). However, AIP regularly threaten ecosystem services, impede recreational opportunities, obstruct drainages and irrigation schedules, and hinder hydroelectric power generation (Langeland 1996; Wilcove et al. 1998). Although not as common, the management of native aquatic plants such as eelgrass (*Vallisneria americana* Michx.) and spatterdock [*Nuphar lutea* (Aiton) W.T. Aiton] is required, as some native species can quickly reach nuisance levels (Gettys 2019). Management typically integrates several weed control strategies (Gettys et al. 2020), though herbicides are widely used to selectively target AIP incursions (McFarland et al. 2004; Nelson et al. 2001). In the United States, more than \$100 million is allocated annually for aquatic plant control (Pimentel et al. 2005; Rockwell 2003).

Aquatic weed control programs are frequently paired with monitoring surveys to gauge plant presence and breadth prior to management activity. Similarly, plant surveys provide quantitative assessment of weed control efforts (Madsen and Wersal 2017). To date, monitoring remains the most effective method to combat deleterious AIP, especially when deploying early detection and rapid response tactics (Westbrooks 2004). Traditional assessment methods like point-intercept, destructive biomass harvests, or nondestructive hydroacoustic measurements (for submersed plants) are effective for estimating AIP abundance and distribution (Howell and Richardson 2019; Johnson and Newman 2011; Madsen 1999; Valley et al. 2015). However, these evaluations generally require boating access, skilled labor (e.g., species identification; boating



and diving proficiency), and substantial time to meet desired spatial coverage. Remote sensing data from satellite and airborne sensors have supported aquatic plant observations across a variety of species and landscapes to overcome traditional survey limitations (Ackleson and Klemas 1987; Nelson et al. 2006; Santos et al. 2016). Still, the limited spatiotemporal resolution of open-sourced satellites (e.g., Landsat 7 ETM+ offers 30-m<sup>2</sup> pixels; imagery every ~15 d) and the associated costs of airborne missions often confines the utility of these platforms within small water bodies. Recently, managers have deployed consumer available small unoccupied aerial systems (sUAS) to accompany in-field surveys to enhance mapping capability (Kislik et al. 2020). Image data from sUAS generate mapping elements that provide superior spatial resolution (<1- to 5-cm<sup>2</sup> pixels) at user-defined temporal resolutions (hours to days). Similarly, sUAS implementation is cost-effective (Lomax et al. 2005) and generally reduces survey time and labor requirements (Fitzpatrick 2015; Nowak et al. 2018). Several studies have revealed the convenience and accuracy of low-altitude (<122 m above ground level; AGL) true-color sUAS usage for emergent, floating, and submersed weed detection (Anderson et al. 2021; Chabot et al. 2016; Hill et al. 2017; Kislik et al. 2020).

In addition to providing a platform for optical imagers, some sUAS offer the opportunity to remotely deliver herbicide applications (Göktoğan et al. 2009; Hunter et al. 2020; Milling 2018; Rodriguez et al. 2022). These sUAS, commonly referred to as unoccupied aerial application systems (UAAS), continue to gain popularity where herbicide treatments are limited with ground-based spray equipment (Lan et al. 2017; Wang et al. 2019; Xue et al. 2016). Benefits of UAAS include improved safety of herbicide application (i.e., personnel have reduced exposure to spray solutions); moreover, the absence of an on-board pilot reduces the inherent risks associated with traditional aerial application techniques (Sheets 2018; Vu et al. 2019). Further, UAAS have the capability to navigate small (e.g., ≤4 ha) application sites that would be costly or prohibitive with a helicopter or plane (He 2018; Otto et al. 2018), and allow for targeted spray missions intended for site-specific weed management (e.g., spot treating) (Hunter et al. 2020; Pathak et al. 2020).

Positive results have ensued when deploying UAAS for site-specific weed management in terrestrial systems (Ahmad et al. 2020; Hunter et al. 2020; Martin et al. 2020), and there is an opportunity to adopt similar spray technology for use in aquatics. Because aquatic weed managers must design applications considering two- and three-dimensional spaces (i.e., foliar and in-water herbicide treatments, respectively), there is a niche for UAAS integration to meet management needs and increase applicator safety. In Australia, an autonomous rotary-wing UAAS used dye as an herbicide surrogate to simulate the spray of alligatorweed [*Alternanthera philoxeroides* (Mart.) Griseb.] and giant salvinia (*Salvinia molesta* D. Mitch) to demonstrate a proof-of-concept model (Göktoğan et al. 2009). However, no studies currently exist describing UAAS strategies and recommended spray application parameters designed for aquatic weed management with herbicides. Nevertheless, applicators express interest in UAAS to meet common environmental complexities (e.g., shallow water; stumps; lack of boat launch facilities) that frequently exist in aquatic herbicide applications. There remains a clear need to investigate the performance of herbicides delivered from UAAS to guide future aquatic weed management decisions.

Currently, there are 16 herbicides available for aquatic weed management in the United States (Schardt and Netherland 2020).

Although many of the available herbicides provide reliable control of AIP (Enloe et al. 2022), some may offer more suitable characteristics for UAAS applications than others. In 2018, the auxin herbicide florypyrauxifen-benzyl was registered as two different formulations (emulsifiable and soluble concentrates) for aquatic-site applications. Florypyrauxifen-benzyl is considered a reduced-risk herbicide for human and environmental exposures (USEPA 2017) and has a lower use rate (~100 times less) than the precursor registered aquatic auxin herbicides, 2,4-D and triclopyr. In mesocosm and field screenings, florypyrauxifen-benzyl has displayed high levels of efficacy on submersed, emergent, and floating-leaved AIP, including Eurasian watermilfoil, hydrilla [*Hydrilla verticillata* (L.f.) Royle], parrotfeather watermilfoil [*Myriophyllum aquaticum* (Vell.) Verdc.], and crested floating heart [*Nymphaoides cristata* (Roxb.) O. Ktze.] (Beets and Netherland 2018; Howell et al. 2022; Netherland and Richardson 2016; Richardson et al. 2016; Sperry et al. 2021). As commercially available UAAS have tank volumes typically ≤10 L to meet Federal Aviation Administration regulations (i.e., FAA 14 CFR Part 107), the low use rates of florypyrauxifen-benzyl make it an excellent candidate herbicide for preliminary UAAS evaluations.

In the present studies, we evaluate the performance of UAAS to remotely deliver florypyrauxifen-benzyl herbicide to floating-leaved and submersed vegetation for targeted weed control. We hypothesized that herbicide applications made with UAAS will provide effective control levels (>90%) and offer an additional spray strategy to manage aquatic weeds in sites having limited access. The objectives were to: (i) determine the feasibility of UAAS in common aquatic weed management scenarios, (ii) evaluate the efficacy of UAAS as an effective platform for the delivery of targeted weed control, and (iii) identify application constraints that exist with UAAS herbicide delivery technology in aquatics.

## Materials and Methods

### Case Study 1: Floating-Leaved Weed Control Using Foliar UAAS Techniques

In the summer of 2020, collaboration efforts with the North Carolina Department of Agriculture aimed to eradicate yellow floating heart from a residential dewatered pond in Lee County, NC (35.4317° N, 79.1987° W) using remotely delivered foliar herbicide applications. Following a series of tropical storms from 2016 to 2018 that led to dam failure, the watered surface of the pond was reduced 80% to 90% of the original area (0.78 ha). The only remaining water averaged 0.48 m (± 0.31 SD) in depth during initial site inspections. As a result of woody [e.g., sweetgum (*Liquidambar styraciflua* L.); willow (*Salix* spp.)], herbaceous [e.g., cattail (*Typha latifolia* L.) and bulrush (*Scirpus cyperinus* (L.) Kunth)] vegetation encroachment and limited site access, traditional ground-based herbicide application options were not feasible. Therefore, herbicide applications were designated using a remotely piloted UAAS to selectively treat the yellow floating heart infestation.

Prior to herbicide application, imagery was collected with a DJI Phantom 4 Advanced (DJI, Shenzhen, China) sUAS at 60 m AGL using the flight planning application, Pix4D Capture (Pix4D SA; Prilly, Switzerland), to locate and map yellow floating heart infestations within the pond. Individual images were stitched together with Agisoft Photoscan Metashape 1.5.1 (Agisoft LLC; St. Petersburg, Russia) to generate an orthomosaic GeoTIFF having

a ground sampling distance of 1.6 cm pixel<sup>-1</sup>. The georeferenced pretreatment orthomosaic was then imported into QGIS 3.14.16 under a projected coordinate system (EPSG: 32617) to visually reference areas within the pond requiring herbicide treatment (QGIS 2022). Digital polygons were generated around yellow floating heart populations that allowed aerial access for UAAS treatment (i.e., areas devoid of heavy tree cover). Georeferenced points from ground-based surveys were used to validate plant presence and cover within the aerial imagery. In total, four discrete yellow floating heart infestations ( $\mu = 0.031$  ha) were identified within the pond that totaled 0.125 ha (Figure 1; Table 1).

Herbicide applications were initiated on July 15, 2020 using a DJI Agras MG-1 octocopter UAAS (DJI, Shenzhen, China). Florypyrauxifen-benzyl (ProcCellaCOR SC; SePRO Corp., Carmel, IN) was applied at 58.3 g a.i. ha<sup>-1</sup> (i.e., 2 prescription dose units), and methylated seed oil was included at 1.0% v/v. Carrier volume was based on the minimum requirements of the herbicide label at the time of application (Anonymous 2020), and nozzle selection was determined from prior research evaluating spray deposition from the identical UAAS model deployed in the present study (Hunter et al. 2020). Specific flight and treatment parameters are provided in Table 1. Where treatment zones permitted, applications were conducted using the UAAS integrated autonomous spray function. Autonomous operations were arranged as 3.0-m spaced tracks following a serpentine spray pattern that worked off an A–B line set by the remote pilot. All other spray operations were manually piloted using comparable flight parameters to the autonomous spraying to achieve the desired application rate. All herbicide applications occurred when wind speeds were 3.2 to 12.6 km h<sup>-1</sup>. There was a 24-h rain-free period following the aerial herbicide application.

Evaluations of plant control were made at 0, 2, and 6 wk after treatment (WAT) using ground-based surveys coupled with aerial imaging missions from the sUAS using the same flight plans conducted for pretreatment measures. Aerial images from each evaluation time point were stitched to create a georeferenced orthomosaic using methods previously described. Imagery from the subsequent evaluation periods (i.e., post-treatment) were georectified to the pretreatment image using the QGIS Georeferencer toolbox and ground control points ( $N = 4$ ) to provide image alignment for spatiotemporal analyses. Orthomosaic images representing each evaluation were evaluated using 1.0-m<sup>2</sup> georeferenced digital quadrats ( $N = 4$  per treatment zone;  $N = 16$ ) to provide spatiotemporal visual density estimates of 0% (no plant cover) to 100% density (complete plant cover). When applicable, injury symptoms to nontarget species were recorded using qualitative methods. Collected data were subjected to ANOVA under the *agricolae* package in RStudio (R Core Team 2020). When significant effects among evaluation periods were detected, means were separated using Tukey's HSD test ( $\alpha = 0.05$ ).

### Case Study 2: Submersed Weed Control Using Directed in-Water UAAS Techniques

Field experiments were initiated in the summer of 2020 to evaluate the performance of directed in-water herbicide application techniques from UAAS for variable-leaf watermilfoil control within a farm pond (0.46 ha) in Moore County, NC (35.3184° N, 79.4010° W). Variable-leaf watermilfoil had become well established within the pond, severely limiting recreation and irrigation activity by the landowner. Tree cover around the pond and dense vegetation within the pond area restricted shoreline and watercraft

herbicide application, respectively. Prior herbicide treatments to control variable-leaf watermilfoil in the pond were reported to be ineffective because of poor accessibility, resulting in inadequate spray coverage from the shoreline. This study was developed to remotely deliver herbicide to a nuisance variable-leaf watermilfoil population to evaluate if adequate plant control could be achieved using UAAS techniques.

Using methods described in previous studies (Madsen 1999; Madsen and Wersal 2017; Valley et al. 2015), a point-intercept kayak survey ensued prior to herbicide treatment to gauge initial variable-leaf watermilfoil dynamics and determine treatment parameters needed to develop herbicide dilution rates. The survey additionally used a Lowrance Hook (Navico Inc., Tulsa, OK) echosounder and a 200-kHz transducer with a 20-deg beam angle (10 pings s<sup>-1</sup>) to passively record depth and submersed plant signatures. Acoustic measurements spanned the length of the pond in a serpentine pattern with tracks spaced approx. 5 m apart. Acoustic data were recorded to the echosounder using a 64-gigabit secure digital card. Logged data files (.sl2) from the completed survey were uploaded to BioBase 5.2 (Navico Inc., Egersund, Norway) cloud-processor to calculate the bathymetry of the pond, determine the spatial extent of submersed plants, and estimate plant biovolume (i.e., percentage of the water column occupied by plants).

Aerial imagery was collected at 46 m AGL, using analogous methods as described for case study 1, to locate and map surface level variable-leaf watermilfoil within the pond prior to treatment. Recorded images were stitched using procedures previously described to create an orthomosaic image having a ground sampling distance of 1.37 cm pixel<sup>-1</sup>. Pretreatment imagery was imported into QGIS under a projected coordinate system (EPSG: 32617) to identify the locations within the pond requiring herbicide treatment. Georeferenced point locations and biovolume estimates from the kayak survey were used to validate plant incidence observed from the aerial imagery. A digital polygon was created around a section of the primary variable-leaf watermilfoil population to serve as the designated treatment zone. The estimated surface coverage of the treatment zone was 0.13 ha (Figure 2; Table 1). An area approx. 50 m adjacent to the treatment zone, which also contained variable-leaf watermilfoil, served as a nontreated reference.

Directed in-water herbicide applications were initiated on July 15, 2020 using a DJI Agras MG-1 octocopter UAAS modified to dispense the spray solution through a single fertilizer nozzle (Table 1). This nozzle was selected for the reduced potential of spray drift compared to conventional nozzles commonly found on UAAS (Hunter et al. 2020), and the deposition of the solid stream was hypothesized to increase spray penetration through surface weed canopies. Florypyrauxifen-benzyl was aerially applied at a target concentration of 9.6  $\mu\text{g a.i. L}^{-1}$  (i.e., 1 prescription dose unit). The UAAS dispensed the herbicide solution to the water-column using a manual operation flight mode in a serpentine pattern within the selected treatment zone. Wind speed averaged 9.6 km h<sup>-1</sup> at the time of aerial herbicide application.

Plant assessments were made at treatment, and at 1, 2, and 5 WAT using kayak surveys coupled with aerial imaging missions from the sUAS using identical flight parameters conducted during pretreatment evaluations. Additionally, low-altitude (3 m AGL) sUAS point photographs were captured at each evaluation period to provide discrete geolocations for visually assessing plant symptomology to the herbicide treatment (e.g., epinasty; chlorosis and necrosis). Flight plan images from



**Figure 1.** Aerial image map of the yellow floating heart infestation in the Lee County pond and selected treatment zones for unoccupied aerial application system foliar floryrauxifen-benzyl applications. Treatment zones are shown in black, and the digital sampling quadrats used for pre- and post-treatment visual evaluations are displayed as red polygons ( $N = 16$ ). The inset image in the legend displays the typical density of yellow floating heart within a mixed community of cattail.

each evaluation time point were stitched to create a georeferenced orthomosaic using methods formerly defined. Orthomosaic images from each evaluation were then georectified to the pretreatment imagery using methods previously described, where ground control points ( $N = 4$ ) were used to align the image rasters for spatiotemporal analyses. Plant response to the herbicide treatment was evaluated using 3.0-m<sup>2</sup> georeferenced digital quadrats ( $N = 4$ ) to provide

spatiotemporal visual density estimates of 0% (no plants visible) to 100% density (complete plant surface coverage). When applicable, injury symptoms to nontarget species were recorded using qualitative methods. Collected data were subjected to ANOVA in RStudio to determine the main effect of the UAAS herbicide application on treated plant density. Where significant effects occurred, evaluation periods were separated from the nontreated control plot using Student's *t*-test ( $\alpha = 0.05$ ).

**Table 1.** Unoccupied aerial application system treatment parameters for Case Studies 1 and 2.

Parameter <sup>a</sup>	Case Study 1	Case Study 2
Plant target	Yellow floating heart	Variable-leaf watermilfoil
Treatment type	Foliar	Directed in-water
Nozzle type	AIXR 11002-VP	FERT SJ3-02-VP
Nozzle number	4	1
Spray angle (deg)	0 (vertically down)	0 (vertically down)
Swath width (m)	3	0.9
Release height (m)	3	3
Flow rate (single nozzle; ml min <sup>-1</sup> )	775	1,100
Application method	Manual/autonomous	Manual
Application speed (m s <sup>-1</sup> )	1	2.24
Carrier volume (L ha <sup>-1</sup> )	149.7	46.8
Application area (ha)	0.125	0.13
Treatment time (min)	13	6
Total application time (min)	150	20
Application efficiency (ha h <sup>-1</sup> ) <sup>b</sup>	~ 0.6	1.3

<sup>a</sup>For both case studies, a DJI Agras MG-1 UAAS was deployed for treatment.

<sup>b</sup>Application efficiency defined as the potential treatment area achievable in 1 h without battery exchanges or refilling the UAAS spray tank.

## Results and Discussion

Because of the inherent environmental complexities within the application sites for both case studies, only visual control data from sUAS imagery missions are reported for target plant response to the UAAS herbicide treatments. Data from both case studies are presented independently.

### Case Study 1: Floating-Leaved Weed Control Using Foliar UAAS Techniques

At treatment initiation, treatment zone quadrats had >92% yellow floating heart visual coverage (Figure 3). Any yellow floating heart plant populations in communities with emergent species (e.g., cattail) outside of the targeted treatment zones are not reported as a result of the inability for aerial image detection (i.e., not visible from the sUAS imagery because of surrounding erect canopy coverage). The most northern treatment zone (Figure 1), also contained the floating-leaved species white water-lily [*Nymphaea odorata* (Aiton)], which is commonly considered a native pest plant.

Foliar herbicide applications made from UAAS significantly reduced ( $P < 0.0001$ ) yellow floating heart density by  $\geq 99\%$  at 2 WAT (Figure 3). Rapid plant response to florypyrauxifen-benzyl was expected for yellow floating heart, as initial plant screenings showed the related species, crested floating heart, to be highly sensitive to the herbicide at concentrations ranging 1 to 3  $\mu\text{g L}^{-1}$  within days after treatment (Netherland and Richardson 2016). The nontarget species, white water-lily, was also injured, and remaining leaves displayed chlorosis and necrosis at 2 WAT, with complete absence observed at the 6-WAT evaluation (data not shown). At 6 WAT, varying levels of yellow floating heart regrowth were detected among three of the four treatment zones. Daughter plants (immature yellow floating heart) were detectable from aerial imagery at 6 WAT and covered <1% to 20% of the water's surface within sampling quadrats (Figure 3). As yellow floating heart reproduces both vegetatively and sexually (Markovich et al. 2020), it is likely that observed yellow floating heart plants within

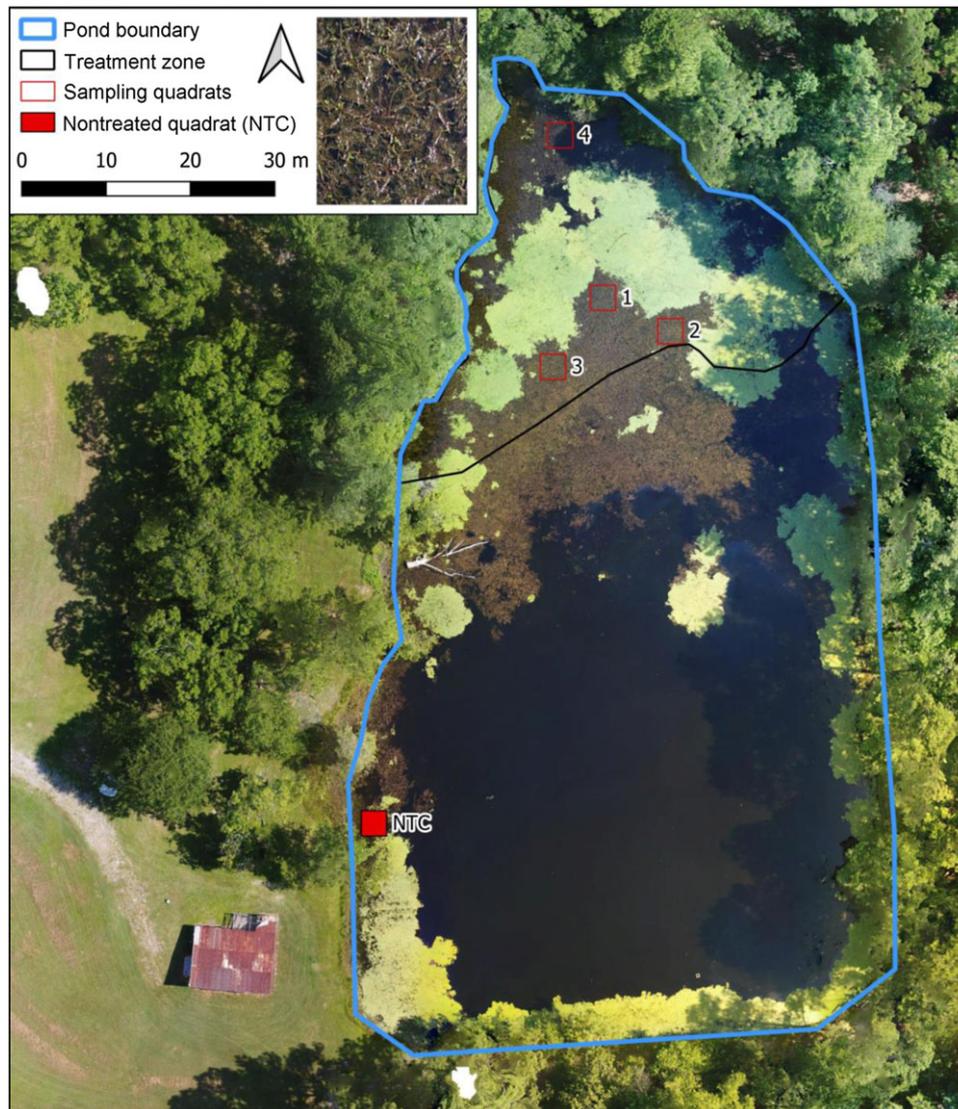
treatment zones where regrowth occurred from either stolon fragments or prior seed production rather than lack of control following herbicide application 6 wk prior. As such, a sequential treatment(s) would be necessary to fulfill eradication efforts in the current study. Future UAAS operations seeking yellow floating heart control should consider sequential applications as the herbicide label permits (Anonymous 2022).

Yellow floating heart foliar treatment response data from the present case study aligns well with former findings of florypyrauxifen-benzyl applications made in-water to crested floating heart. In a 28-d outdoor mesocosm study, crested floating heart above-ground biomass was completely controlled following static exposure of florypyrauxifen-benzyl at 24  $\mu\text{g L}^{-1}$  (Beets and Netherland 2018). Similarly, research evaluating the absorption and translocation of florypyrauxifen-benzyl within crested floating heart has shown high belowground concentrations of the herbicide in a <sup>14</sup>C study (Haug et al. 2021). However, it is likely that overspray from UAAS foliar applications that would provide in-water herbicide activity was limited by the shallow-water conditions within yellow floating heart treatment zones. Ultimately, environmental conditions probably reduced any residual in-water activity of florypyrauxifen-benzyl, as the herbicide rapidly degrades through photolysis (<1 d) and strongly binds to sediment (Heilman and Getsinger 2018; USEPA 2017). Future research investigating yellow floating heart control from foliar application techniques would benefit from evaluating herbicide fate in varying depth gradients and the influence on belowground vegetative structures (i.e., stolons) capable of successional growth.

Several treatment considerations that transpired will benefit future management action for floating-leaved aquatic weeds. Because of tall surrounding vegetation (e.g., cattail) within the infestation site, traditional backpack application techniques would have likely limited foliar application to yellow floating heart targets. Similarly, backpack applications from ground crew would have required more than one applicator to achieve effective and efficient herbicide treatments. Although access to the edge of the yellow floating heart pond was largely restricted by tree cover, UAAS takeoff and landing was not constrained by the complex environmental conditions, as an improvised landing zone was easily constructed by compressing bulrush in a circular configuration (1.5× the width of UAAS wingspan). The ability to remotely treat yellow floating heart under these environmental constraints further highlights the utility as another aquatic plant management application method. Further, the efficiency of UAAS demonstrated that a remote herbicide application could occur within expected timeframes of ground applications while also safely removing crew from environmental hazards (e.g., snakes) and contact with herbicide during application (Table 1). Notably, application efficiency could be improved upon as UAAS technologies advance, especially when integrating RTK UAAS platforms that allow for more precise autonomous spray missions (Zhang et al. 2020). In this case, herbicide applications would have minimal input from the remote pilot, as environmental parameters would be pre-mapped and uploaded to the UAAS platform, which would increase the overall accuracy and efficiency of the sprayer (Hunter et al. 2020).

### Case Study 2: Submersed Weed Control Using Directed in-Water UAAS Techniques

At the time of the UAAS directed in-water treatment, variable-leaf watermilfoil was topped-out (i.e., 100% coverage of the surface of

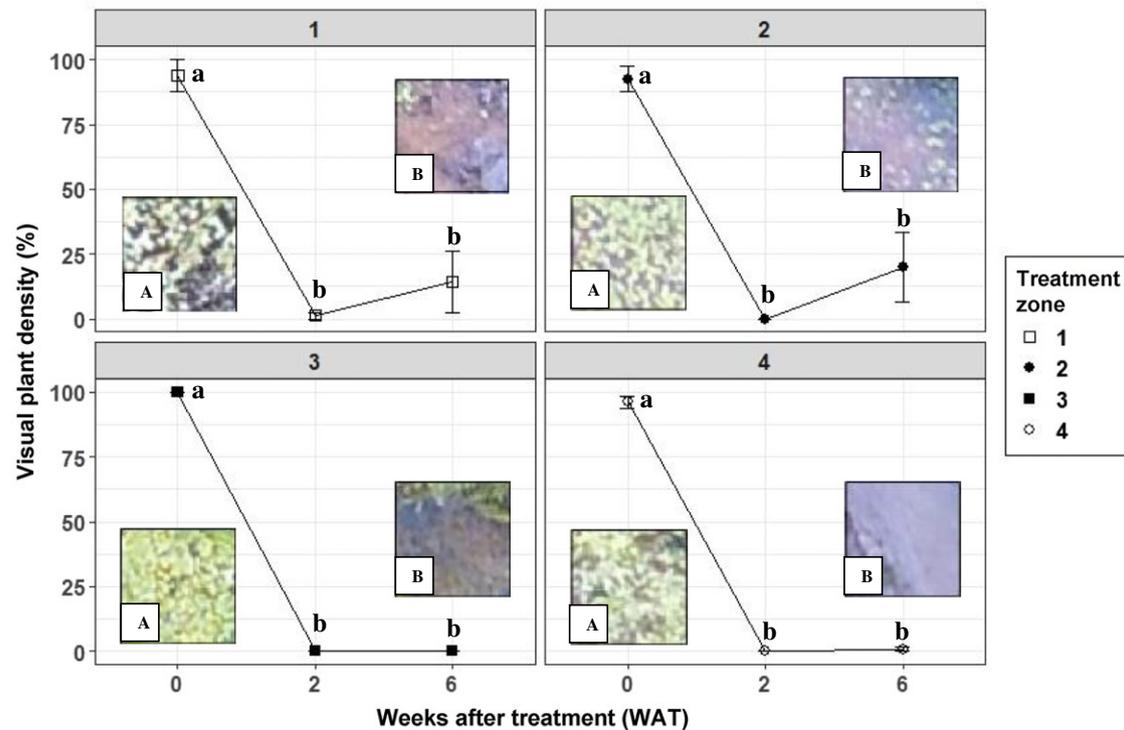


**Figure 2.** Aerial image map of the variable-leaf watermilfoil infestation in the Moore County pond and the selected treatment zone for unoccupied aerial application system-directed in-water florpyrauxifen-benzyl application. The treatment zone is in the upper section of the image, bounded by a black line, and the pond perimeter is outlined in blue. The digital sampling quadrats used for pre- and post-treatment visual evaluations are displayed as red polygons ( $N = 4$ ). The nontreated control quadrat (NTC) is displayed as the solid red polygon. Within the pond boundary, dark-brown submersed plant growth is variable-leaf watermilfoil, and the bright-green floating-leaved patches are watershield. The inset image in the legend displays the typical surface-level density of variable-leaf watermilfoil at pretreatment.

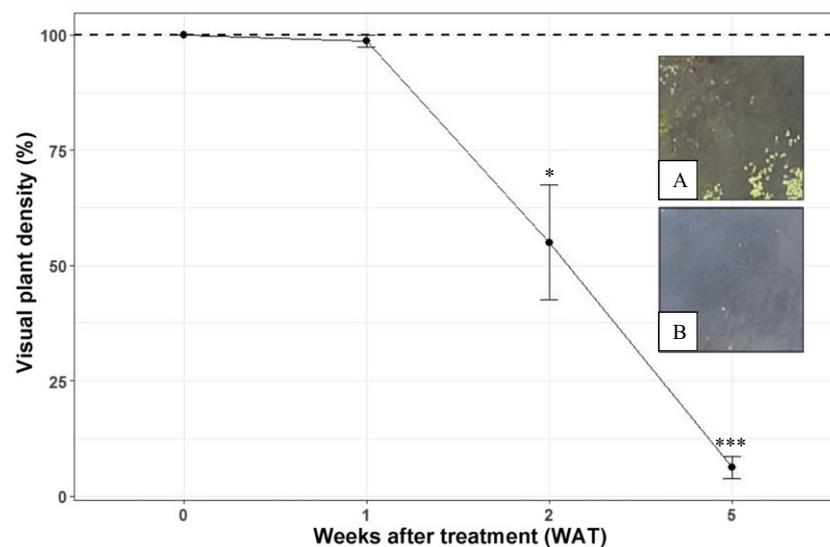
the water column) within the selected treatment zone. The native floating-leaved species, watershield [*Brasenia schreberi* (J.F. Gmel.)], covered approx. 50% of the treatment zone at the time of herbicide application. Though not specifically targeted, watershield injury was documented when appropriate, as the plant can be a nuisance within small ponds because of dense growth (Weldon et al. 1969). Plants within the nontreated control plot retained vigor with no signs of injury throughout the 5-wk study duration.

At 1 WAT, directed in-water florpyrauxifen-benzyl applications made from UAAS did not significantly ( $P = 0.391$ ) reduce visual surface level coverage of variable-leaf watermilfoil (Figure 4). However, injury was evident by 1 WAT, with terminal epinasty of plant shoots and inflorescences (data not shown). In greenhouse settings, variable-leaf watermilfoil treated with florpyrauxifen-benzyl at 0.3 to 81  $\mu\text{g L}^{-1}$  showed rapid stem growth and epinasty within 1 WAT (Richardson et al. 2016), a result that aligns well

with plant response in the present case study. Additionally, watershield within the treatment zone was chlorotic and displayed leaf curling at 1 WAT (data not shown). These observed injury symptoms at 1 WAT are common among plants sensitive to auxin herbicides like florpyrauxifen-benzyl (Grossmann 2010; Howell et al. 2021). By 2 WAT, a significant treatment effect was detected ( $P = 0.036$ ), where visual variable-leaf watermilfoil density was reduced by 45% compared to the pretreatment measures (Figure 4). Low-altitude image capture and kayak surveys visually indicated that variable-leaf watermilfoil had also begun fragmenting by 2 WAT (data not shown). Watershield response at 2 WAT showed an overall decline in plant vigor (e.g., chlorotic and necrotic leaves), and by 5 WAT the plant's coverage was reduced by approx. 80% (data not shown). These observations suggest that in-water exposures of florpyrauxifen-benzyl at 9  $\mu\text{g L}^{-1}$  readily controls watershield. At the final evaluation 5 WAT, variable-leaf watermilfoil imagery showed a significant decline in density



**Figure 3.** The effect of a single floryprauxifen-benzyl foliar treatment made with an unoccupied aerial application system for yellow floating heart eradication efforts within each respective treatment zone (1, 2, 3, and 4). Data describe the visual plant density response to the herbicide treatment within the digital sampling quadrats ( $n = 4$ ) placed within each treatment zone (mean visual density  $\pm$  SE;  $N = 16$ ). Means between evaluation periods with identical letters around the standard error bars are not different according to Tukey's HSD test ( $\alpha = 0.05$ ). The inset plot images provide a visual example of pretreatment plants (A) and plants treated with herbicide (B) at 6 wk after treatment.



**Figure 4.** The effect of a single directed in-water floryprauxifen-benzyl application made with an unoccupied aerial application system for variable-leaf watermilfoil control under static conditions. Data describe the visual plant density response to the herbicide treatment within the digital sampling quadrats (mean visual density  $\pm$  SE;  $N = 4$ ). The dashed trend line represents the nontreated control plot, whereas the solid line represents the trend in plant response to herbicide treatment. An asterisk above the standard error bars indicates the detection of a significant interaction between the treated and nontreated control plots according to Student's  $t$ -test at  $\alpha = 0.05$  (\*  $P < 0.05$ ; \*\*  $P < 0.0001$ ). The inset plot images provide a visual example of nontreated plants (A) and plants treated with herbicide (B) at 5 wk after treatment.

( $P < 0.0001$ ), as only 6% of the initial plant material remained detectable within the sampling quadrats (Figure 4). A former study evaluating variable-leaf watermilfoil control with floryprauxifen-benzyl at concentrations  $\geq 9 \mu\text{g L}^{-1}$  observed  $\geq 90\%$  plant biomass inhibition at 4 WAT (Richardson et al. 2016), which corroborates

the high level of plant suppression experienced in the present study.

Although additional evaluations did not occur beyond 5 WAT, variable-leaf watermilfoil response to the directed in-water treatments of floryprauxifen-benzyl from UAAS were considered

efficacious for targeted submersed-plant control. In a 4-wk mesocosm study, the lowest observed effect concentration for variable-leaf watermilfoil was  $9 \mu\text{g L}^{-1}$  of florpyrauxifen-benzyl (Richardson et al. 2016), which is analogous to the target herbicide concentration utilized in the present case study ( $9.6 \mu\text{g a.i. L}^{-1}$ ). Additional studies investigating florpyrauxifen-benzyl efficacy on invasive *Myriophyllum* spp. indicate that low herbicide concentrations ( $\leq 12 \mu\text{g L}^{-1}$ ) at static or short exposure periods (e.g., 3 h) reduce hybrid watermilfoil (*Myriophyllum spicatum* L.  $\times$  *M. sibiricum* Kom.) biomass  $\geq 98\%$  at 30 to 60 DAT (Beets et al. 2019; Mudge et al. 2021). Data from the present study and past studies suggest that high levels of *Myriophyllum* spp. control are achievable with florpyrauxifen-benzyl delivered using directed in-water UAAS techniques. Additionally, evidence suggests that similar aerial herbicide application methods could be implemented to manage native floating-leaved plant pests like watershield.

Treatment parameters from the current case study demonstrated that directed in-water aerial applications from UAAS provide an efficient and effective option to treat submersed plant targets when water access is restricted (Table 1). The total application time, which included UAAS setup, filling the spray tank, conducting treatment, and UAAS disassembly, occurred faster than would be expected for ground-based spray operations in similar scenarios. Additionally, the ease of application was notable, and the ability to treat submersed plants without the need to deploy watercraft improved operational efficiency. In general, application efficiency is highly dependent on the need to refill the spray tank with herbicide solution and exchange batteries for continued flight (UAAS flight duration is approx. 8 to 10 min per battery). Though current battery technology confines potential flight time (Ying et al. 2016), and federal aviation regulations dictate UAAS total weight capacity (i.e., FAA 14 CFR part 107 requires  $< 24.9 \text{ kg}$  at take-off), permitted modifications made to herbicide labels could improve the efficiency of remote spray operations. For example, the current herbicide label for florpyrauxifen-benzyl requires a minimum carrier volume of  $46.8 \text{ L ha}^{-1}$  for aerial applications (Anonymous 2022). In treatment zones  $> 1.0 \text{ ha}$ , application efficiency would likely increase if the minimum carrier volume requirements were reduced by half (e.g.,  $23.4 \text{ L ha}^{-1}$  volumes require three fewer tank fills for a 10-L capacity UAAS). Furthermore, directed in-water application techniques would be expected to achieve target herbicide concentrations regardless of carrier volume. In a submersed field application study evaluating florpyrauxifen-benzyl to suppress dense dioecious hydrilla, the herbicide was shown to rapidly dissipate within 6 h following treatment (Sperry et al. 2021). Research evaluating ultralow spray volumes with florpyrauxifen-benzyl to submersed plant targets would benefit future direction of directed in-water application strategies with UAAS.

This is the first published field data in the United States describing herbicide efficacy deployed from UAAS for targeted weed control in aquatics. Results from both case studies indicate that integrating UAAS for floating-leaved and submersed plant control can provide an efficient and effective alternative to traditional ground-based application strategies. Nevertheless, the efficacy and efficiency of foliar applications made at low-release altitudes ( $< 3 \text{ m AGL}$ ) will remain highly dependent on target plant distribution, UAAS spray deposition, and surrounding obstructions. The deployment of sUAS to monitor herbicide applications greatly improved the ability to estimate target plant control following herbicide application. Further, the use of sUAS imagery aided in the detection of yellow floating heart populations formerly

unidentified using ground-based assessment methods in the first case study and directly supports early detection and rapid response strategies. Although the potential exists to incorporate UAAS for aquatic plant management strategies, we recognize that current UAAS regulations and aquatic herbicide labels may restrict the broader integration of remote spray systems for applicators at present. Research investigating the effects of herbicide efficacy at ultralow carrier volumes would improve the utility of UAAS and provide further direction for UAAS in aquatics. Similarly, examining additional directed in-water application strategies (e.g., releasing herbicide in a grid pattern) could increase the overall efficiency of UAAS in larger submersed plant treatment scenarios.

## Practical Implications

Aquatic weed management with herbicides relies heavily upon site-specific control strategies. Nevertheless, the highly diverse nature of aquatic environments can restrict traditional ground-based spray operations in areas with limited access. Advances in unoccupied aerial application systems (UAAS) have shown the potential to deliver herbicide treatments in areas having limited access while reducing applicator exposure to the herbicide during the management activity. However, there is limited evidence on UAAS utilization in aquatics. Our studies indicated that aerial applications of florpyrauxifen-benzyl, as either a foliar or directed in-water spray, provides an effective and efficient treatment strategy to control floating-leaved and submersed weeds. Although current UAAS regulations and aquatic herbicide labels do have limitations for wide-scale use, future integration of UAAS in aquatic weed control programs is encouraged.

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## References

- Ackleson SG, Klemas V (1987) Remote sensing of submerged aquatic vegetation in lower Chesapeake Bay: a comparison of Landsat MSS to TM imagery. *Remote Sens Environ* 22:235–248
- Ahmad F, Qiu B, Dong X, Ma J, Huang X, Ahmed S, Chandio FA (2020) Effect of operational parameters of UAV sprayer on spray deposition pattern in target and off-target zones during outer field weed control application. *Comput Electron Agr* 172:1–10
- Anderson CJ, Heins D, Pelletier KC, Bohnen JL, Knight JF (2021) Mapping invasive *Phragmites australis* using unoccupied aircraft system imagery, canopy height models, and synthetic aperture radar. *Remote Sens* 13:1–27
- Anonymous (2020) ProcellaCOR® SC product label. Carmel, IN: SePRO Corp. <https://www.sepro.com/aquatics/procellacor>. Accessed: July 1, 2020
- Anonymous (2022) ProcellaCOR® SC product label. Carmel, IN: SePRO Corp. <https://www.sepro.com/aquatics/procellacor>. Accessed: September 23, 2022
- Beets J, Netherland MD (2018) Mesocosm response of crested floating heart, hydrilla, and two native emergent plants to florpyrauxifen-benzyl. *J Aquat Plant Manage* 56:57–62
- Beets J, Heilman M, Netherland MD (2019) Large-scale mesocosm evaluation of florpyrauxifen-benzyl, a novel arylpicolinate herbicide, on Eurasian and hybrid watermilfoil and seven native submersed plants. *J Aquat Plant Manage* 57:49–55
- Chabot D, Dillon C, Oumer A, Shemrock A (2016) Object-based analysis of UAS imagery to map emergent and submerged invasive aquatic vegetation: a case study. *J Unmanned Veh Syst* 5:27–33

- Enloe SF, Sperry B, Leary J (2022) Efficacy of herbicide active ingredients against aquatic weeds. SS-AGR-44. <https://edis.ifas.ufl.edu/publication/AG262>. Accessed: September 26, 2022
- Fitzpatrick BP (2015) Unmanned aerial systems for surveying and mapping: cost comparison of UAS versus traditional methods of data acquisition. PhD dissertation. University of Southern California. 49 p
- Gettys LA (2019) Breaking bad: native aquatic plants gone rogue and the invasive species that inspire them. *HortTechnology* 29:559–556
- Gettys LA, Haller WT, Petty DG, eds (2020) *Biology and Control of Aquatic Plants: A Best Management Practices Handbook*. 4th edn. Marietta, GA: Aquatic Ecosystem Restoration Foundation. 224 p
- Göktoğan AH, Sukkarieh S, Bryson M, Randle J, Lupton T, Hung C (2009) A rotary-wing unmanned air vehicle for aquatic weed surveillance and management. *J Intell Robot Syst* 57:467–484
- Grossmann K (2010) Auxin herbicides: current status of mechanism and mode of action. *Pest Manage Sci* 66:113–120
- Haug E, Ahmed K, Gannon T, Richardson R (2021) Absorption and translocation of florypyrauxifen-benzyl in ten aquatic plant species. *Weed Sci*. 69:624–630
- He X (2018) Rapid development of unmanned aerial vehicles (UAV) for plant protection and application technology in China. *Outlooks Pest Manag* 29:162–167
- Heilman MA, Getsinger K (2018) Early operational demonstration of selective invasive watermilfoil control with ProcellaCOR. [https://bugwoodcloud.org/mura/mipn/assets/File/UMISC-2018/Wednesday/PM/Heilman\\_EarlyOperationalDemo\\_ProcellaCOR.pdf](https://bugwoodcloud.org/mura/mipn/assets/File/UMISC-2018/Wednesday/PM/Heilman_EarlyOperationalDemo_ProcellaCOR.pdf). Accessed: April 18, 2022
- Hill DJ, Tarasoff C, Whitworth GE, Baron J, Bradshaw JL, Church JS (2017) Utility of unmanned aerial vehicles for mapping invasive plant species: a case study on yellow flag iris (*Iris pseudacorus* L.). *Int J Remote Sens* 38:1–23
- Houlahan J, Findlay C (2004) Effect of invasive plant species on temperate wetland plant diversity. *Conserv Biol* 18:1132–1138
- Howell AW, Richardson RJ (2019) Estimating standing biomass of exotic macrophytes using sUAS. Pages 1–17 in *Proceedings of Vol. 11008, Autonomous Air and Ground Sensing Systems for Agricultural Optimization and Phenotyping IV*, 110080Z-1:1–19. doi: [10.1117/12.2519199](https://doi.org/10.1117/12.2519199). Baltimore, MD: SPIE Defense + Commercial Sensing
- Howell AW, Hofstra DE, Heilman MA, Richardson RJ (2021) Evaluation of florypyrauxifen-benzyl to control three problematic submersed macrophytes in New Zealand. *J Aquat Plant Manage* 59:66–71
- Howell AW, Hofstra DE, Heilman MA, Richardson RJ (2022) Response of parrotfeather and alligatorweed to foliar florypyrauxifen-benzyl applications. *J Aquat Plant Manage* 60:53–61
- Hunter JE, Gannon TW, Richardson RJ, Yelverton FH, Leon RG (2020) Integration of remote-weed mapping and an autonomous spraying unmanned aerial vehicle for site-specific weed management. *Pest Manage Sci* 76:1386–1392
- Johnson JA, Newman RM (2011) A comparison of two methods for sampling biomass of aquatic plants. *J Aquat Plant Manage* 49:1–8
- Kislik C, Genzoli L, Lyons A, Kelly M (2020) Application of UAV imagery to detect and quantify submerged filamentous algae and rooted macrophytes in a non-wadeable river. *Remote Sens* 12:1–24
- Lan Y, Shengde C, Fritz B (2017) Current status and future trends of precision agricultural aviation technologies. *Int J Agric Biol Eng* 10:1–17
- Langeland KA (1996) *Hydrilla verticillata* (L.F.) Royle (Hydrocharitaceae), the perfect aquatic weed. *Castanea* 61:293–304
- Lomax A, Corso W, Etro J (2005) Employing unmanned aerial vehicles (UAVs) as an element of the Integrated Ocean Observing System. Pages 1–7 in *Proceedings of the Marine Technology Society (MTS)/Institute of Electrical and Electronics Engineers (IEEE) conference OCEANS 2005*, Washington, DC
- Madsen JD (1999) Point intercept and line intercept methods for aquatic plant management. APCRP Technical Notes Collection (TN APCRP-M1-02). Vicksburg, MS: US Army Engineer Research and Development Center. 17 p
- Madsen JD (2014) Impact of invasive aquatic plants on aquatic biology. Pages 1–11 in: Gettys LA, Haller WT, Petty DG, eds. *Biology and Control of Aquatic Plants: A Best Management Practices Handbook*. 3rd edn. Marietta, GA: Aquatic Ecosystem Restoration Foundation. 224 p
- Madsen TV, Sand-Jensen K (1991) Photosynthetic carbon assimilation in aquatic macrophytes. *Aquat Bot* 41:5–40
- Madsen JD, Wersal RM (2017) A review of aquatic plant monitoring and assessment methods. *J Aquat Plant Manage* 55:1–12
- Markovich IJ, Sigmon JW, Gettys LA (2020) Floating hearts. Pages 59–64 in Gettys LA, Haller WT, Petty DG, eds. *Biology and Control of Aquatic Plants: A Best Management Practices Handbook*. 4th edn. Marietta, GA: Aquatic Ecosystem Restoration Foundation
- Martin D, Singh V, Latheef MA, Bagavathiannan M (2020) Spray deposition on weeds (Palmer amaranth and morningglory) from a remotely piloted aerial application system and backpack sprayer. *Drones* 4:1–18.
- McFarland DG, Nelson LS, Grodowitz MJ, Smart RM, Owens CS (2004) *Salvinia molesta* D.S. Mitchell (giant salvinia) in the United States: a review of species ecology and approaches to management. ERDC/EL SR-04-2. Vicksburg, MS: US Army Engineer Research and Development Center. 41 p
- Milling P (2018) Unmanned aerial vehicles used to control giant reed (*Arundo donax* L.). Pages 177–180 in *Proceedings of the 21st Australasian Weeds Conference*. New South Wales, Australia
- Mudge CR, Sartain BT, Getsinger KD, Netherland MD (2021) Efficacy of florypyrauxifen-benzyl on dioecious hydrilla and hybrid water milfoil—concentration and exposure time requirements. Vicksburg, MS: US Army Corps of Engineers Research & Development Center ERDC/EL TR-21-8. 17 p
- Nelson LS, Skogerboe JG, Getsinger KD (2001) Herbicide evaluation against giant salvinia. *J Aquat Plant Manage* 39:48–53
- Nelson SAC, Cheruvilil KS, Soranno PA (2006) Satellite remote sensing of freshwater macrophytes and the influence of water clarity. *Aquat Bot* 85:289–298
- Netherland MD, Richardson RJ (2016) Evaluating sensitivity of five aquatic plants to a novel arylpicolinate herbicide utilizing an organization for economic cooperation and development protocol. *Weed Sci* 64:181–190
- Nowak MM, Dziob K, Bogawski P (2018) Unmanned aerial vehicles (UAVs) in environmental biology: a review. *Eur J Ecol* 4:56–74
- Otto A, Agatz N, Campbell J, Golden B, Pesch E (2018) Optimization approaches for civil applications of unmanned aerial vehicles (UAVs) or aerial drones: a survey. *Networks* 72:411–458
- Pathak H, Kumar GAK, Mohapatra SD, Gaikwad BB, Rane J (2020) Use of drones in agriculture: potentials, problems and policy needs. Publication no. 300, ICAR-NIASM. 17 p
- Pimentel D, Zuniga R, Morrison D (2005) Update on the environmental and economic costs associated with alien-invasive species in the United States. *Ecol Econ* 52:273–288
- QGIS (2022) QGIS Geographic Information System. QGIS Association. <http://www.qgis.org>. Accessed: May 31, 2023
- R Core Team (2020) R: A language and environment for statistical computing. Vienna: R Foundation for Statistical Computing. <https://www.R-project.org/>. Accessed: September 20, 2022
- Richardson RJ, Haug EJ, Netherland MD (2016) Response of seven aquatic plants to a new arylpicolinate herbicide. *J Aquat Plant Manage* 54:26–31
- Rockwell WH (2003) Summary of a survey of the literature on the economic impact of aquatic weeds. Report to the Aquatic Ecosystem Restoration Foundation. 18 p
- Rodriguez R, Leary JJK, Jenkins DM (2022) Herbicide ballistic technology for unmanned aircraft systems. *Robotics*. 11:1–15
- Santos MJ, Khanna S, Hestir EL, Greenberg JA, Ustin SL. (2016) Measuring landscape-scale spread and persistence of an invaded submersed plant community from airborne remote sensing. *Ecol Appl* 26:1733–1744
- Schardt J, Netherland MD (2020) Chemical control of aquatic weeds. Pages 163–178 in Gettys LA, Haller WT, Petty DG, eds. *Biology and Control of Aquatic Plants: A Best Management Practices Handbook*. 4th edn. Marietta, GA: Aquatic Ecosystem Restoration Foundation
- Sheets KD (2018) The Japanese impact on global drone policy and law: why a laggard United States and other nations should look to Japan in the context of drone usage. *Indiana J Glob Leg Stud* 25:513–537
- Sperry BP, Leary JK, Jones KD, Ferrell JA (2021) Observations of a submersed field application of florypyrauxifen-benzyl suppressing hydrilla in a small lake in central Florida. *J Aquat Plant Manage* 29:20–26

- [USEPA] United States Environmental Protection Agency (2017) Final registration decision on the new active ingredient flupyrauxifen-benzyl. 22 p
- Valley RD, Johnson MB, Dustin DL, Jones KD, Lauenstein MR, Nawrocki J (2015) Combining hydroacoustic and point-intercept survey methods to assess aquatic plant species abundance patterns and community dominance. *J Aquat Plant Manag* 53:121–129
- Vu NA, Dang DK, Dinh TL (2019) Electric propulsion system sizing methodology for an agriculture multicopter. *Aerosp Sci Technol* 90:314–326
- Wang G, Lan Y, Qi H, Chen P, Hewitt A, Han Y (2019) Field evaluation of an unmanned aerial vehicle (UAV) sprayer: effect of spray volume on deposition and the control of pests and disease in wheat. *Pest Manag Sci* 75:1546–1555
- Weldon LW, Blackburn RD, Harrison DS (1969) Common aquatic weeds. Agricultural Research Service, USDA Agriculture Handbook 352. 43 p
- Westbrooks RG (2004) New approaches for early detection and rapid response to invasive plants in the United States. *Weed Technol* 18:1468–1471
- Wilcove DS, Rothstein D, Dubow J, Phillips A, Losos E (1998) Quantifying threats to imperiled species in the United States. *BioScience* 48:607–615
- Xue X, Lan Y, Zhu S, Chun C, Hoffman WC (2016) Develop an unmanned aerial vehicle based automatic aerial spraying system. *Comput Electron Agr* 128:58–66
- Ying Z, Xiuyan G, Zhiyan Z, Xiwen L, Yu Z, Xingyuan Q, Wulan L, Chen Y (2016) Review of tensairity and its applications in agricultural aviation. *Int J Agric Biol Eng* 9:1–14
- Zhang C, Boyle K (2010) The effect of an aquatic invasive species (Eurasian watermilfoil) on lakefront property values. *Ecol Econ* 70:394–404
- Zhang XQ, Song XP, Liang YJ, Qin ZQ, Zhang BQ, Wei JJ, Li YR, Wu JM (2020) Effects of spray parameters of drone on the droplet deposition in sugarcane canopy. *Sugar Tech* 22:583–588