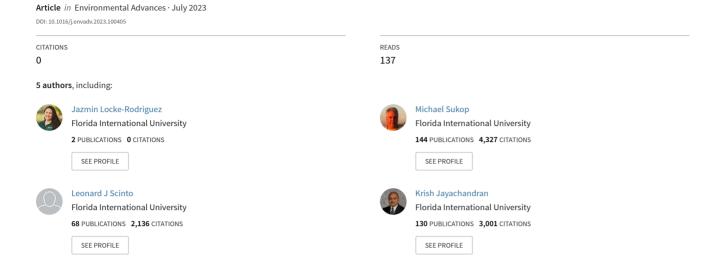
Floating Flowers: Screening Cut-Flower Species for Production and Phytoremediation on Floating Treatment Wetlands in South Florida



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Floating flowers: Screening cut-flower species for production and phytoremediation on floating treatment wetlands in South Florida

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ABSTRACT

Floating treatment wetlands (FTWs) are artificial ecosystems designed to mimic the nutrient removal capabilities of natural wetlands through the hydroponic cultivation of plants on floating rafts, thereby utilizing the process of phytoremediation. This approach provides plants with protection against submersion, creating an optimal environment for the growth of valuable hydroponic crops. To ensure the removal of nutrients absorbed and incorporated into plant biomass from aquatic systems, the implementation of routine plant harvesting serves as an effective management strategy. This practice prevents the decomposition and subsequent release of nutrients back into the water. Furthermore, the cultivation of crops for commercial purposes can serve as an incentive to enhance biomass harvesting and replanting efforts, which may be financially impractical otherwise. This study aimed to assess the growth success and nutrient remediation capacities of five cut-flower species on FTWs in controlled mesocosm systems at Florida International University in Miami, Florida. The surviving species were evaluated based on growth metrics, bloom count, and nutrient removal abilities, specifically for total phosphorus (TP) and total nitrogen (TN). Among the five species tested, only marigold (Tagetes erecta) survived throughout the 12-week trial on the FTWs. The marigold-planted treatment exhibited a significant enhancement in nutrient reduction efficiencies compared to the control treatment, removing 52% more TP and 33% more TN mass from the mesocosm system. This resulted in a nutrient removal rate of 0.062 g of TP \cdot m 2 \cdot day $^{-1}$ and 0.321 g of TN \cdot m 2 · day ⁻¹ in the marigold-treated mesocosm. Additionally, the marigold treatment yielded an average of 65 marketquality blooms per m^2 , with mean widths of 6.4 ± 1.8 cm and lengths of 27.6 ± 7.3 cm. Given the substantial nutrient removal and the production of marketable blooms, marigold (Tagetes erecta) shows promising potential as a commercially viable remediating crop cultivated on FTWs in South Florida.

Statement of Significance: This study demonstrated that marigolds grew successfully on an FTW, removed a comparable amount of nutrients from the water as other wetland plants tested on FTWs in previous studies, and produced a high number of blooms for market. This species is worth testing at a field scale to see how well they thrive in application. Future studies should increase the thickness of the FTW for roots to anchor better and help stabilize the growth of additional cut-flower species. Specifically, the production of cut-flowers on FTWs in South Florida could take advantage of the floral industry infrastructure in Miami, FL while also helping to address increasing water quality issues in the region.

1. Introduction

Excess nutrient inputs from both agricultural and urban runoff have degraded water quality across South Florida since the 1960s (Snyder and Davidson, 1994). Nitrogen (N) and phosphorus (P) fertilizer runoff from major agricultural operations such as sugarcane farming in the Everglades Agricultural Area (EAA) have severely impaired sensitive habitats

such as the Everglades (Lang et al., 2010). Stormwater flowing over city streets also carries excess nutrients derived from fertilizers, herbicides, detergents, plant debris, atmospheric deposition, and improperly functioning septic tanks (Peluso and Marshall, 2002). Due to Southeast Florida's shallow unconfined aquifer and intricately connected water systems, nutrient pollution can be a risk to Florida's most critical ecosystems including the Everglades, Florida Bay, and Biscayne Bay given

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their oligotrophic conditions (Perry, 2008). When combined with warm and stagnant water, these nutrients can fuel the growth of detrimental algal blooms leading to hypoxic water and poor water clarity that can kill fish, and important aquatic vegetation like seagrasses, while also degrading the ecosystems at large (Sharpley et al., 1994).

Floating Treatment Wetlands (FTW) are an affordable and adaptable form of green infrastructure that can be conveniently installed and removed from water bodies to address excessive nutrient pollution (Headley and Tanner, 2012). Unlike conventionally constructed wetlands, FTWs integrate their filtering capabilities into pre-existing retention features like stormwater ponds and canals, eliminating the need to alter their flow or depth characteristics (Winston et al., 2013). This makes FTWs particularly advantageous in regions like South Florida, where land availability is limited, as they can effectively utilize the existing water landscape to enhance water quality.

FTWs are artificial floating islands made from buoyant materials to support the growth of plants above water while their roots extend down into the water to uptake dissolved nutrients and other pollutants. The rafts protect the plants from submersion while allowing their root systems to grow unrestricted in open water (Chen et al., 2016). Direct contact with dissolved nutrients supports both increased uptake and plant growth compared to growth in soil. A larger root surface area promotes the colonization of microbial biofilm which further assists the plants in the uptake of nutrients through this microbial digestion (Urakawa et al., 2017). These key features of FTWs often make their nutrient removal rates more efficient than their constructed wetland counterparts whose roots are embedded in the sediment rather than in direct contact with the water (Bi et al., 2019; Headley and Tanner, 2012; Yeh et al., 2015). FTWs are a distinct opportunity to improve South Florida water quality through nutrient reduction while preserving a system's water supply or flood control capacity. Recent studies indicate examples of FTWs successfully removing nutrients from water bodies and canals across Florida (Brown et al., 2018; Chang et al., 2012; White, 2021).

The simple design of an FTW allows for plants to be removed from the raft as they reach maturity and can be replaced by new plants. Routine plant harvesting is a management strategy that can be used to ensure that nutrients absorbed and incorporated into plant biomass are removed completely from an aquatic system, (White, 2021; Xu et al., 2017). Removing a plant from an FTW at its peak maturity encourages the highest level of nutrient removal from the system while avoiding the risk of nutrients returning to the water if plants begin to die and decompose (Garcia Chance et al., 2019; Pavlineri, et al., 2017; Zhao et al., 2012). Plants that can be pruned to encourage new growth can be harvested throughout the season to promote additional nutrient removal (Bi et al., 2019).

Increased maintenance of green infrastructure, like frequent harvesting, requires labor and time which can be difficult to support without financial incentives (USEPA, 2013). Cultivating crops on an FTW whose harvests can be sold and/or consumed may incentivize and support the regular harvesting of these plants. This increased harvesting increases remediation capacities, while also developing a potential market opportunity. Various studies have explored this potential to grow plants for sale on FTWs. Ornamental plants, including Canna Lily, Iris, and perennial grasses, have been successfully cultivated on FTWs in freshwater and have shown promising potential for transplantation and sale as potted plants (Chen et al., 2009; Garcia Chance et al., 2022; Spangler et al., 2018). While these studies have demonstrated comparable nutrient reduction efficiencies between ornamental plants and traditionally used wetland plants, it is important to note that the ornamental plants were harvested at a relatively young stage for transplantation, which may have limited their overall nutrient removal capacities by preventing them from reaching full maturity. To optimize nutrient removal and promote biomass growth, the exploration of cut-flower crops on FTWs provides a promising opportunity. Regular harvesting of blooms for sale not only allows the plants to reach their maximum nutrient removal capacity but also stimulates additional biomass growth. This research aims to assess the viability and effectiveness of different cut-flower crops on FTWs in removing nutrients from polluted water bodies in South Florida.

Due to its strategic location, Miami receives 80% of the country's imported cut flowers and distributes them across the nation (Siegler, 2020). According to the USDA Floriculture Crops summary (USDA, 2021), Florida is also the highest producer of cut cultivated greens in the country, which are used as floral bouquet fillers. This well-established network can be leveraged to facilitate the delivery of Florida-grown flowers throughout the country as well. Growing cut-flowers on FTWs may also be a great way to increase public support for their implementation as they will add a beautification element where they are installed. Additionally, cut flower production can create revenue with FTWs that can incentivize its broader implementation and regular management. Hydroponic crop cultivation is increasing in its application across the world; this FTW system mimics a deep-water hydroponic operation by simply using the excessive nutrients already present and burdening our many aquatic ecosystems (Chen et al., 2016). This research study investigates the cultivation of cut-flowers on FTWs as a strategy to remove excess nutrients in stormwater while also creating a potential economic opportunity in South Florida. The objectives of this study aimed to screen potential cut-flower species suitable for cultivation in an FTW setting and evaluate the removal rates of total nitrogen (TN) and total phosphorus (TP) by surviving species, to assess their capacity to remediate simulated nutrient pollution in mesocosm tanks.

2. Materials and methods

2.1. Study site

The experiment was conducted over 12 weeks during the fall season of 2020. It was located adjacent to the Florida International University (FIU) Wertheim Conservatory (25° 45'32.4" N, 80°22'28.7" W) in Miami, Florida, USA, which is in the Southeastern region of the U.S and hardiness Zone 10. During the experimental period, the ambient temperature averaged 25.0 $^{\circ}$ C and a total of 51.2 cm of precipitation.

2.2. Mesocosms and experimental treatments

The controlled experiment took place in a series of cylindrical mesocosm tanks, measuring 0.914 m high with a 1.829 m diameter as represented graphically in Fig. 1. The tanks were filled with 2,350 L of municipal water to the height of 0.762m as recommended by Chen et al. (2016) to allow optimal root growth. A standing pipe with a hole at 0.762m height allowed water to flow out of the tank if the water levels reached above that height from precipitation. Six mesocosms were arranged in 2 rows of 3, side by side. The mesocosms were designed as closed systems, meaning that water loss from the system was limited to overflow or evaporation. As a result, the hydraulic retention time (HRT) for the entire study period was 12 weeks (84 days). To promote adequate water and oxygen circulation within each mesocosm, a Vivosun commercial aerator, operating at 32W and moving 3,596 liters per hour (L•h⁻¹), was installed in each tank. The aerator remained operational continuously to maintain a consistent and sufficient flow rate. Though this magnitude of aeration is not often available in a field application, the inclusion of commercial aerators alongside FTW installations has increased in practice because of its ability to significantly improve nutrient uptake in FTWs (Garcia Chance and White 2018, White 2021 and Yeh et al. 2015). The managed aeration also ensured optimal conditions for evaluating the hydroponic suitability of cut-flower species, preventing the rejection of any species solely based on low dissolved oxygen (DO) tolerance.

The FTW rafts used were supplied by Beemats LLC (New Smyrna Beach, FL). The Beemats system is made of 1.3 cm thick buoyant closed cell foam mats with pre-cut holes for specially designed aerator pots to hold plants. The mats were arranged in a 1.8m x 1.2m rectangle to



Fig. 1. Diagram illustrates mesocosm dimensions and design (left). The picture shows a floating treatment wetland in mesocosm with cut-flower planted treatment spaced at 9 plants per m^2 (right).

comfortably fit into each mesocosm, covering 84% of the water surface. The FTW floated on top of the water with the planting pots submerged for the entire study period. Each tank had an FTW planted with a different cut-flower species treatment. Each treatment was planted with 20 equally spaced replicates (9 plants/ m^2) as represented in the photo of Fig. 1. The control tank contained the same size FTW, but without plants.

The following species were used as planted treatments for 5 separate mesocosms: Jazzy Zinnias (*Zinnia haageana*), Giant Dahlia Zinnias (*Zinnia elegans*), Sonja Branching Sunflower (*Helianthus annuus*), Sunbright F1 Single Stem Sunflower (*Helianthus annuus*) and Giant Coco Gold F1 Marigold (*Tagetes erecta*). These species were chosen for their heat tolerance and vitality in a South Florida climate (IFAS, 2022). They have also been identified as low-maintenance cut-flower species that are sought after in the floral industry (Loyola et al., 2019). Sunflowers and marigolds have been shown to possess hyperaccumulating properties, making them effective in remediating heavy metals from soils due to their extensive root systems (Adesodun et al., 2010; Biswal et al., 2021). It is hypothesized that these same mechanisms may contribute to their ability to remove nutrients from the environment.

All seeds were purchased from Johnny's Selected Seeds (Waterville, Maine) and placed into $2.5~{\rm cm}^2$ rock wool starter blocks at 1-2 seeds per block. Seedlings were watered with the same nutrient concentrations they would be exposed to in the mesocosms. Seedlings were transplanted at 6 weeks old into 5 cm diameter net pots surrounded by clay pebbles. The rock wool transplants were filled with enough pebbles beneath them to ensure the crowns of the flowers would be held above the water line.

2.3. Simulation of nutrient-rich water

Tank solution concentrations of 0.5 mg L^{-1} of P and 3.5 mg L^{-1} of N were maintained in each tank. These concentrations were chosen as they fell within the range of runoff concentrations reported from both the Everglades Agricultural Areas (Pietro and Ivanoff, 2015) and South Florida urban areas (Harper and David, 2007). Nutrient solutions were created by dissolving two fertilizers: hydroponic plant nutrient (6-12-28) and calcium nitrate (15-0-0) from Verti-gro Inc. (Verti-gro Inc., Summerfield, Fl). Dry nutrients were dissolved into water as per the manufacturer's instructions to create a nutrient solution concentrate. On the first day of the trial, solutions equivalent to adding 1.17 g of P and 8.18 g of N were added into each 2,350 L tank. This combined to create solution concentrations of 0.5 mg $\rm L^{-1}$ of P and 3.5 mg $\rm L^{-1}$ of N in each tank. Every four days, half of the concentration loads were applied to represent regular runoff flow that may accumulate into local waterways over time. Over the entire course of the trial, tanks were loaded with a total of 13.73g of P and 90.65 g of N.

2.4. Water nutrient and physiochemical monitoring

Throughout the trial, water measurements and samples were taken every 4 days, both before and after adding additional fertilizer to each tank. Since the mesocosms were constructed as closed systems, these samples were collected over the entire HRT, which encompassed the full 84-day duration of the trial. The Apera PC60 Premium Multi-Parameter Tester (Apera, Columbus, Ohio) was used to record physicochemical water parameters such as Electrical Conductivity (EC, μ S· cm⁻¹), pH, and water temperature (°C), while the Orion Star Series A329 portable monitor (Thermo Scientific Inc., Waltham, Massachusetts) measured Dissolved Oxygen (DO, mg ${\it L}^{-1}$). These measurements were taken for each tank upon arrival along with two water samples collected in 20 ml scintillation vials at 30cm depth. Next, mesocosm tanks were spiked with dissolved fertilizer at concentrations of 0.25 mg L-1 of P and 1.75 mg L-1 of N (half of the target loads). After an hour of mixing, physicochemical parameters were measured again and two more water samples were collected to monitor the nutrient levels. In total, four water samples were collected per tank every 4 days, resulting in up to 84 samples collected from each tank over the 12-week trial. If a planted treatment died prematurely, plants were removed, and water sampling ceased for that tank. All water samples were kept on ice in a cooler until fieldwork was completed and then transported to the lab. To preserve the samples, 0.5 ml of 5N sulfuric acid was added, and they were stored in the refrigerator and analyzed within 28 days of receipt.

Water samples were analyzed for N and P values based on their plantavailable, dissolved forms. This analysis may be considered acceptable because the nutrients were added via commercial fertilizer to create the applied load in the solution. N values reported for water samples of this study are the sum of nitrate-N and nitrite-N (NOx- N), the primary N constituent in both agricultural and urban waters (Hugo et al., 2009). Due to the use of an aerator, ammonia levels were expected to remain low (Dunqiu et al., 2012). This made monitoring NO_x- N an appropriate method for assessing N within water samples, as supported by studies such as Shahid et al. (2019) and Yang et al. (2008). The P values reported reflect orthophosphate-P (OP-P), the plant-available component of total P. All water nutrient analyses were conducted by the AQ2 Discrete Auto Analyzer (SEAL Analytical, Mequon, Wisconsin). OP-P was measured by the AQ2 method USEPA-146-A Rev.0 and NO_x-N values were measured by the AQ2 method USEPA 114-A Rev. 9. To maintain quality assurance/quality control (QA/QC) protocol, duplicate samples were analyzed every 10 samples. All glassware was acid-rinsed followed by flushing in distilled water before analysis.

Weekly precipitation volume and air temperature were averaged during the study period (September 28, 2020- December 19, 2020) from 4 of the nearest publicly accessible weather stations: two stations from Weather Underground (WU, 2020) including Stations CHSS-KFLTOWNP2 (25.76 $^{\circ}$ N, 80.35 $^{\circ}$ W) and KFLMIAMI624 (25.77 $^{\circ}$ N, 80.35

°W) and two stations from South Florida Water Management Districts Environmental Database DBHydro (SFWMD DBHydro, 2021) including Station Miami.FS_R (25.49 °N, 80.20 °W) and Station NP-NESS20 (25.43 °N, 80.20 °W).

2.5. Plant growth, harvest, and biomass assessment

Every 8 days, plant height and flower count were recorded. Additionally, flowers were harvested and recorded for their length, bloom width, and individual mass. Light pruning was practiced to encourage bloom growth and help remove dead or damaged foliage. All pruned mass was recorded for wet weight. Prunings were air-dried throughout the trial and then measured for total dry weight at the end of the trial. Flowers were cut right above the lowest node to harvest the longest stems per cut. Stem lengths and bloom width were measured per flower.

At the end of the 12-week trial, all plants were removed from the FTWs and measured for their shoot and root length. If a planted treatment died prematurely, they were removed from the tank before 12 weeks. Roots and shoots were separated at the crown, divided by roots, shoots (stem/leaves), and flowers, and then weighed for their wet mass (g). The divided plant parts were oven dried for 3 days at 20 °C, after which dry weight was recorded, and then plant material was ground in the 8000M Mixer/Mill (SPEX Sample Prep, Metuchen, NJ). Biomass samples were analyzed for each plant part for total nitrogen (TN) and total phosphorus (TP) content of roots, shoots, and flowers. The TN was analyzed by dry combustion with a Truspec Carbon/Nitrogen Analyzer (LECO Corporation, St. Joseph, Michigan). The TP was assayed from plant biomass through wet acid digestion and then assessed with the SEAL Analytical AQ2 Discrete Auto Analyzer (Mequon, Wisconsin) utilizing method EPA-146-A Rev.0.

2.6 Calculation & statistical analysis

Total nutrient mass accumulation from planted treatments (M_p , g) was based on nutrient concentration analyses of plant biomass. The total mass accumulated was extrapolated by Equation 1.

$$M_p = \sum (B * C_b) \tag{1}$$

where the sum of nutrient mass totaled for all plant parts based on the mass of each plant part (B, g) was multiplied by the corresponding nutrient concentration of biomass (C_b, g) .

Cumulative nutrient removal (M_r, g) from each treatment tank was based on

$$M_r = (M_T - C_F V) \tag{2}$$

where M_T is the total mass (g) of nutrients added to the tank throughout the entire trial, C_F (g L^{-1}) is the nutrient concentration of the water on the final day of the trial and V (L) was the volume of water in the tank.

Removal efficiency (E, %) for all treatments was calculated using

$$E(\%) = M_r/M_T * 100 \tag{3}$$

Nutrient removal rates of both OP-P and NOx- N of planted treatments were determined by calculating the regressions extrapolated from water samples calculated for nutrients removed to date. Water samples were first converted to daily mass (g) loads using the following equation:

$$M_x = C_x * V (4)$$

Where M_x is the daily nutrient load in a tank (g), C_x is the daily nutrient concentration (g L^{-1}) of a tank, and (V) is tank volume (L). This product was then subtracted from the total accumulated nutrient mass (g) loaded (M_{AL}) of the corresponding date to calculate the amount of accumulated mass (g) removed (M_{AR}) to date.

$$M_{AR} = M_{AL} - M_x \tag{5}$$

The M_{AR} were then plotted, and a regression was assessed. The slope of this regression line represents the nutrient removal rate for each treatment over the trial. By dividing this slope by the total area (A) of the FTW (2.23 m²), the final nutrient removal rate (g · m² · d) of the treatment was determined.

Statistical analyses were performed using SAS JMP1 Pro 16.2.0 software (SAS Institute, Inc. Cary, North Carolina, USA). Statistical analyses were used to determine whether treatments differed from the control in terms of N or P removals or differed in other physiochemical properties. Normality assumptions were tested both visually using the histogram and suggested guidelines for skew and kurtosis were compared. Normally distributed data were analyzed using a One-way ANOVA and T-Test. Not normally distributed data utilized nonparametric Wilcoxon/Kruskall-Wallis test. Statistical tests were considered significant at the p<0.05 level.

3. Results and discussion

3.1. Plant survival, growth, and bloom production for planted treatments

This study tested five different cut flower species including *Zinnia haageana*, *Zinnia elegans*, *Helianthus annuus* (both single-stem and branching sunflowers), and *Tagetes erecta* (giant marigolds). All five of these species survived as seedlings in the hydroponic seed starting table until at least 5 cm tall and were then transplanted into net pots directly into FTW mats. At transplant, the seedlings were 6 weeks old.

Fig. 2 shows the mortality rates along with the plant heights reached for each planted treatment over the time of the trial. The branching sunflower treatment was terminated at week 3 as no further growth was observed after transplantation. Interestingly, the sunflowers began blooming immediately in week 3, indicating an accelerated flowering response. However, the presence of decaying leaves and brittle stems suggested senescence. This early blooming phenomenon, with plants reaching a maximum height of 10 cm, could be attributed to the stress of transplantation triggering an adaptive survival mechanism. Crop timing is a recognized concern for cut-flower producers cultivating sunflowers (Loyola et al., 2019). During the trial, single-stem sunflowers exhibited slow growth initially but started blooming by week 5, reaching an average height of only 44.5 cm instead of the expected 168 cm. Harvesting the blooms caused the termination of the treatment at week 5. Since harvesting blooms from single-stem sunflowers results in plant mortality, the treatment had to be terminated prematurely after the blooms were harvested at week 5. Direct seeding or transplanting before 3 weeks old is recommended when planting sunflowers (Schoellhorn et al., 2003). A sudden change in light intensity, temperature, or humidity could be the environmental stress that triggered these early blooms (Dyer et al.,1959). Future trials would benefit from direct seeding into net pots to test sunflowers again.

Neither zinnia species developed healthy plant systems. Raft thickness did not provide a sufficient above-water area for roots to stabilize. This prevented zinnias from developing the structural foundation for plants to grow upright and would instead fall over and into the water frequently. This would cause fungal infections to develop, increasing the plant's vulnerability to pests. Armyworm caterpillars were observed frequently and consumed large portions of individual plants. By week 5, nearly 70% of individuals had died in both zinnia treatment tanks (Fig. 2 Top). The remaining were heavily damaged from armyworm or covered in powdery mildew fungus. Little growth in height was seen between October 26 and November 6th (Fig. 2 Bottom). To avoid plants decaying into the water, both zinnia-planted treatments were terminated on November 6th (day 40).

The marigolds were the most successful of all the flowers, growing through to the end of the 12-week trial. The marigolds developed adventitious roots from their stems that grew out and over the net pots, which helped stabilize the foundation for the plant to grow (Fig. 3). When the plant became too heavy to support itself, the plant began to

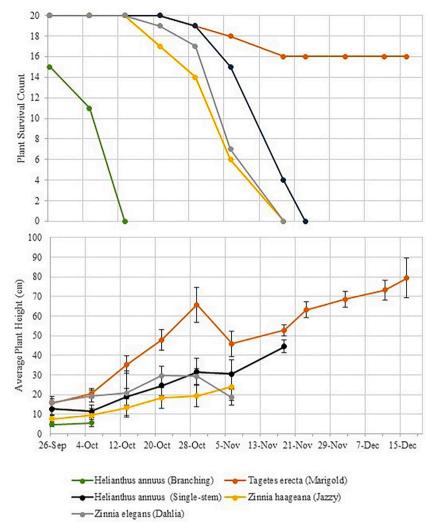


Fig. 2. These two graphs represent the mortality rates (top) and the average plant heights (cm) \pm standard deviation calculated every 8 days of each planted treatment over time (bottom).



Fig. 3. Pictures of marigold parts planted on FTW. Pictures show adventitious roots sprouting along stems of marigolds providing stability (A), extensive root development from beneath the FTW mat and into water (B), cut blooms (C), and above-ground biomass production (D).

grow laterally, and more adventitious roots developed from the stem to create an even stronger, horizontal foundation. Branches sprouted upright from this lateral growth and were able to reach heights of over 70 cm (Fig. 2). By the end of the trial, there was a 20% mortality rate of marigolds (Fig. 2). The individuals that had died were shaded out by surrounding plants, leading to the inference that the marigold would benefit from increased spacing for their lateral growth.

At the time of transplant, marigold seedlings were on average 14 ± 2 cm tall with root lengths an average of 23 ± 3 cm long. By the end of the trial, the marigold plants grew to an average height of 79 ± 10 cm with roots an average of 48 ± 9 cm long. To encourage blooms, excess leaf mass was pruned as needed along with blooms which reduced average plant heights. In general, the average plant height increased continuously throughout the entire experiment. Had they not been terminated at the end of week 12 they may have continued to grow, reaching their recorded peak heights of 92 cm (Gilman and Howe, 1999).

Marigold bloom production began on November 6^{th} , and increased over time, producing 63 blooms • m^2 over the course of the 12 weeks, or a total equivalent of 142 blooms. Bloom widths ranged from 2.5 -10.2 cm wide with an average of 6.4 \pm 1.8 cm. The average stem length ranged between 12.7 -50.8 with an average of 27.6 \pm 7.3 cm. With

improved pruning practices, the average stem lengths of each cut flower could likely be increased, which improves their market value (Society of American Florists, 1994).

To estimate potential revenue from marigold bloom sales, market research was performed utilizing United States Department of Agriculture USDA (2023). Though there was little data for marigold cut flower sales specifically, marigolds fall into the chrysanthemum family which includes variants sold on average \$1.00 per bloom. Based on the 142 blooms produced, this trial could have earned a revenue of \$142.00 on the $2.23~\text{m}^2$ FTW, equating to \$63.70 m² for the 12 weeks.

3.2. Water quality analyses

3.2.1. Physicochemical analysis

Water quality physicochemical parameters were measured for each tank until the day of their termination, as seen in Fig. 4. All samples collected in the first month (9/ 28- 10/26/20) of each treatment were statistically compared to each other to determine differences. Additionally, all samples collected in the last month (11/23 - 12/19/20) were also compared statistically to determine differences. Since only the marigold-planted treatment survived until the final month of the trial, it

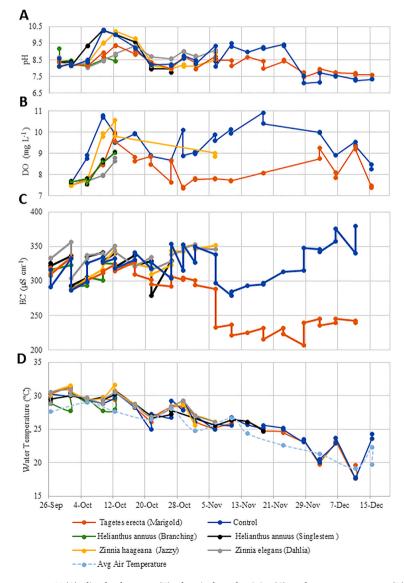


Fig. 4. Water physiochemical measurements pH (A), dissolved oxygen (B), electrical conductivity (C), and water temperature (D) recorded every 4 days throughout floating treatment wetland study conducted between September 2020 and December 2020.

was the only one compared to the control tank for that period.

Water pH levels amongst treatments fluctuated slightly but all remained within a range from 7-10.5 throughout the trial. No treatment had significantly different pH levels than another within the first nor the last month of the trial. The marigold-treated tank had an overall observed lower pH throughout the trial compared specifically to the control tank (Fig. 4A). This could be due to the increased removal of nutrients occurring in the marigold-treated tank. Natural processes occurring in the rhizosphere such as the release of root exudates and organic acids may also contribute to the difference in pH (Nye, 1981).

Dissolved Oxygen (DO) readings ranged between 7-11 mg L^{-1} for all treatments throughout the trial and were not significantly different from each other within the first nor the last month of the trial (Fig. 4 B.). DO in the control tank remained highest among all treatments for nearly the entire trial, reaching its maximum level at 10.9 mg L⁻¹ during month 2 before dropping back down to below 10 mg L⁻¹ in the last month. This may be due to algae growth that peaked and increased photosynthesis and DO injection at first until it began to die back in the last month. Both Headley and Tanner (2012) and Pavlineri et al. (2017) discuss how increased vegetative coverage from FTW can reduce DO concentrations by interrupting atmospheric oxygen diffusion. The additional shading from vegetation can prevent the growth of photosynthetic microbes and the oxygen demand from the floating plant matter may also impact this reduction. This would help explain why nearly all treatments had DO levels below that of the control throughout the trial. In contrast, these readings are also a result of the added aerator in both tanks which ensured DO levels were maintained at a minimum level of 7 mg•L⁻¹. Aeration studies on both mesocosm (Dunqiu et al., 2012; Garcia Chance and White, 2018) and field scales (White, 2021) demonstrate that using aerators alongside FTW systems can help ensure high DO levels which also improves nutrient removal. Though increased DO may prevent denitrification, it instead enhances ammonia and nitrogen oxide removal while also promoting increased phosphorus removal (Dunqiu et al., 2012). For this reason, FTWs are often utilized with commercial-grade aerators in practice to help optimize these systems.

Electrical conductivity (EC, μ S·cm⁻¹) can be used as a proxy parameter for nutrient accumulation or removal in water over time. In the initial month of the trial, the EC values were relatively similar among the treatment tanks, ranging from 275 to 350 μ S·cm⁻¹(Fig. 4C). Among these, Zinnia elegans exhibited the highest EC levels, significantly surpassing the marigold-treatment tank which displayed the lowest levels (p < 0.005). By employing linear regression analysis of EC readings, valuable insights can be gained regarding nutrient removal rate and efficiency.

The regression lines for all treatment EC values had positive slopes, except for the marigold treatment. This suggests that the other treatment tanks were accumulating nutrients over time. This observation aligns with the fact that treatments that were terminated early exhibited minimal growth during the initial establishment period and showed signs of degradation, which hindered their ability to effectively uptake nutrients. The positive EC slopes further confirm why these specific treatments were terminated early, as they demonstrated their unsuitability for the hydroponic conditions and inability to remove nutrients in their current state. Among the treatments evaluated, including the control, none exhibited a significant correlation between accumulation rate and time, with correlation coefficients (R²) below 0.55. However, the marigold treatment showed a distinct pattern with a negative slope of -1.42 (μ S • cm⁻¹ • day⁻¹) and a confidence level of 70% (R² = 0.70). This negative EC slope indicates a consistent uptake of nutrients by the growing marigold plants. By the end of the experiment, the surviving marigold treatment displayed significantly lower EC values compared to the control tank (p < 0.009), indicating a more effective nutrient removal capability.

Finally, water temperature (°C) naturally began to drop for all treatments as the trial progressed from the fall to winter season, reducing nearly 10 degrees Celsius from about $30^\circ C$ in October to as low

as 20°C in December. However, the water temperature in all the treatments remained comparable to each other over time (Fig. 4D). This demonstrates that planted treatments did not add a substantial amount of insulation or cooling to the water in their tank, considering the control tank still contained the same sized FTW mat on its surface. Using conductivity as a proxy for nutrient accumulation, there was no significant correlation between temperature and conductivity for any of the treatments ($R^2 < 0.40$). Dungiu et al. (2012) demonstrated that the nutrient removal efficiency of plants on FTWs are significantly impacted by cold temperatures, but that above 13°C, temperature no longer has a significant impact on plant uptake efficiency. Despite the trial spanning the last quarter of the year, South Florida's daily low temperatures remain around 18.3°C in winter, rarely falling below $10^{\circ}\text{C}.$ Interestingly, these mild temperatures make it the optimal growing season for the region's agricultural industry. As plants grew larger in the marigold treatment to the end of the trial, the conductivity in the tank decreased, indicating a continued uptake in removal. This is counterintuitive to our understanding of lower temperatures' impact on efficiency. That leads to support that temperature may not have as significant an impact above 13°C as demonstrated by Dunquiu et al. (2012). It has been shown that N and P removal rates were typically higher in warm climates, which is very promising for the South Florida region (Akratos and Tsihrintzis, 2007; Picard et al., 2005).

3.2.2. Water nutrient concentrations and nutrient removal rate

Zinnia and sunflower treatments were terminated early due to premature blooms, minimal biomass, or decay. Consequently, the remaining individuals had minimal impact on nutrient removal in their respective tanks, as indicated by EC measurements. As a result, the nutrient concentrations in the water samples and the biomass of these flowers were not analyzed. As marigolds survived through the entire trial, water nutrient concentrations were analyzed to calculate the nutrient removal rate of that treatment compared to the control tank. Fig. 5 plots the accumulated nutrient mass removed (MaR) of OP-P (Fig. 5. A) and NOx- N (Fig. 5.B) from both treatments over the course of the trial. Nutrient removal within the first month of the trial was similar for both treatments in terms of OP-P and NOx- N removal. This coincides with the 4-week plant establishment phase typically required for plants to adapt to FTW conditions (Headley and Tanner, 2012).

Into the second month of the trial, the marigold treatment began to remove more nutrients when compared to the control tank at the same sampling time. Although not to the same degree, it can be observed that the control treatment was also increasing its nutrient removal during the second month of the trial (Fig. 5). Precipitation accounted for part of this nutrient loss for both tanks. Algae growth, particularly in the control tank, was also observed, potentially consuming nutrients and contributing to the continued nutrient removal in the control treatment. This increased algae growth is typical of the eutrophic conditions being created in the tank especially as higher nutrients were accumulating in the control tank over time and available for algae consumption. The increased algae growth in the control treatment vs. the marigold-planted treatment can be observed in Fig. 6.

In the third month, the mass of nutrients removed for both NOx- N and OP-P in the marigold treatment continues to increase. The marigold-planted treatment removed the most nutrient mass this month as plants continued to grow and increase in bloom production. In contrast, the control treatment begins to plateau and drop. This could be in part due to the dying back of the algae over time and the return of nutrients to the water through decomposition.

The slopes determined in Fig. 5 from the regression lines represent the nutrient removal rates of each treatment from the water $(g \cdot day^{-1})$. As the marigolds continued to grow throughout the trial, the water samples supported linear regressions for NOx-N and OP-P removal over time. Alternatively in the control tank, the small algae bloom consumed nutrients but then returned nutrients as it died, better supporting quadratic regressions for nutrient removal. When these slopes are

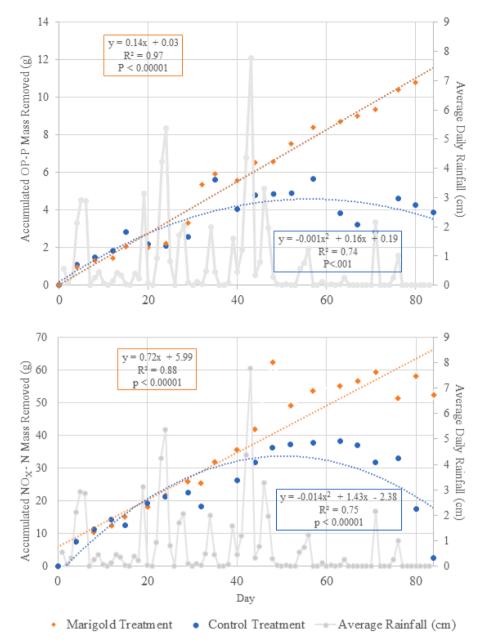


Fig. 5. Accumulated Mass Removed (M_{AR}) plotted for both marigold-planted treatment and control treatment over time. Nutrient removal rates ($g \cdot day^{-1}$) for both NOX - N and OP-P of both treatments are represented by regression coefficients. To determine the rate per area the slope must be divided by FTW area of 2.23 m².



Fig. 6. Week 8 water samples of control tank (two left bottles) presenting a green color due to algae growth compared to marigold-treated tank (two right bottle) with minimal color and algae growth.

divided by the area of the FTW (2.23 m²), removal rates for each nutrient per treatment can be determined. The removal rate of OP-P was 0.062 $\,{\rm g\cdot d^{-1}\cdot m^2}$ in the marigold-treated tank compared to -0.0006 $\,{\rm g\cdot d^{-1}\cdot m^2}$ of the control tank. The removal rate of NOx- N was 0.321 $\,{\rm g\cdot d^{-1}\cdot m^2}$ in the marigold tank versus -0.006 $\,{\rm g\cdot d^{-1}\cdot m^2}$ in the control tank. The significant difference of means for water nutrient concentrations of both NOx- N and OP-P (p<0.05) demonstrates that the marigold-planted treatment made a substantial impact on the water quality compared to the control treatment with no plants.

3.3. Biomass nutrient concentrations, mass balance, and nutrient removal efficiency

Planted treatments that were terminated early were excluded from the biomass nutrient analyses. At the end of the experiment, all surviving plants from the marigold-planted treatment were removed from the FTW mat and separated into plants parts, flowers, roots, and shoots (stem & leaf). They were individually measured for their wet weights as

Average \pm standard deviation wet and dry mass of marigold plant parts (n=13) collected from the entire 2.2 m² FTW at the end of the 12-week trial. Nutrient mass accumulation per plant part was provided based on plant part nutrient concentrations multiplied by the mass per plant part. Averaged \pm standard deviation of these sections is provided (n=13).

	Wet biomass per plant section (g)	Dry biomass per plant section (g)	TP mass per plant section (g)	TN mass per plant section (g)
Flower	55 ± 23	$\textbf{5.2} \pm \textbf{2.4}$	0.03 ± 0.02	0.13 ± 0.06
Root	228 ± 10	28.6 ± 20.7	0.21 ± 0.25	0.75 ± 1.45
Shoots	777 ± 143	87.1 ± 40.2	0.63 ± 0.13	3.16 ± 0.42
Whole plant	1056 ± 234	109.6 ± 63.3	0.86 ± 0.25	4.04 ± 1.52
Cumulative plants*	14281	1491.4	12.67	55.85

^{*} Sum of all plants.

summarized in Table 1. Most of the plant mass was contained in the shoots of the plant. The total wet weight of all plants combined was 14,281 g. After 3 days in the oven at 20° C, there remained 1491 g of dry weight, equating to a 10:1 wet-to-dry weight ratio.

The average nutrient concentrations of all harvested marigold plants were 6.59 ± 1.40 mg of TP per gram of dry plant weight and 29.2 \pm 7.05 mg of TN per gram of dry plant weight. The dry weight (g) of each plant part was multiplied by its respective nutrient concentration for an estimated sum of total nutrient accumulation per plant. Therefore, each surviving marigold plant contained an estimated total of 0.86 \pm 0.25 g of TP and 4.04 \pm 1.52 g of TN in their biomass (Table 1). The sum of all marigold plants equated to a total accumulated mass of 12.67g of P and 55.85g of N. These values account for the estimated nutrient mass (g) within all plants at the time of transplanting into the FTW.

Table 2 presents a nutrient mass balance for each tank based on the total mass of nutrients loaded to tanks as fertilizer over the trial compared to the total nutrient reduction calculated from both water samples and biomass. The load reduction represents all nutrients removed from the tank by the end of the trial based on Day 84 water samples (n=2). The load reduction of the marigold-treated tank (11.53 ± 0.01 g of TP and 56.62 ± 0.74 g of TN) compared to the control tank (3.33 ± 0.11 g of TP and 9.45 ± 8.96 g of TN) demonstrates the contribution that marigold plants made to the nutrient reduction in its tank. Using the load reduction mass over the total trial load mass, the percentage of nutrient removal efficiency can be calculated for each tank. The marigold treatment removed 86.15% and 64.31% of TP and TN whereas the control treatment was reduced by 34.27% and 31.35% of TP and TN respectively.

Factors that could have impacted the mass balance include precipitation and aeration. When precipitation escalated water levels above the 0.76 m level, water would drain from the standpipe, and nutrients were

Table 2
Total phosphorus (TP) and total nitrogen (TN) mass balance after a 12-week FTW study comparing non-planted control treatment and marigold-planted treatment.

	TP (g)	TN (g)
Total nutrient load	13.73	90.65
Marigold Treated Tank		
Total load after 84- day HRT1	1.90	32.35
Load Reduction ²	11.83 (86)	58.30 (64)
Plant Uptake	12.67	55.85
Other removal processes	-0.84	2.45
Control Tank		
Total load after 84- day HRT1	9.03 ± 0.09	62.23 ± 5.77
Load Reduction ²	4.71 ± 0.09 (34)	28.42 ± 5.77 (31)
Plant Uptake	-	-
Other removal processes	$\textbf{4.71} \pm \textbf{0.09}$	28.42 ± 5.77

 $^{^{1}\,}$ n=2 of water samples collected on last day of trial.

lost from the system. Due to evapotranspiration, water levels would occasionally fall below the standpipe. These levels were not regularly documented however so the mass of nutrients lost from precipitation could not be estimated exactly. It is fair to assume however that a portion of the control nutrient reduction efficiency came from displacement from precipitation and would have likely had a similar impact on the marigold treatment. Even with this contribution in mind, the marigold treatment nutrient efficiency surpassed the control tank with a difference of 52% more TP and 33% more TN removed.

The nutrient contents and removal rates of marigolds in this study are favorable compared to other floating wetland mesocosm studies. Wang et al. (2014) and Zhou et al. (2012) had similar plant spacing densities on their FTW, and in both cases, the removal rates of their chosen wetland species were lower than that of the marigolds in this study. Wang et al., (2014) did have lower water nutrient influent, which likely contributed to the lower nutrient removal rates by the *Schoenoplectus tabernaemontani* used in that study.

Alternatively, studies such as Spangler et al., (2018), Li et al., (2011), and Yang et al., (2008) conducted mesocosm studies with similar influent levels, but their plant densities were either higher or unreported. Typically, wetland plants are spaced with a much higher density than used in this study to maximize nutrient removal. The marigolds of this study had higher P and N removal rates than the *Lolium perenne* utilized in Li et al. (2011) and the *Oenanthe javanica* planted in Yang et al. (2008). Marigolds' removal rates fell within the cumulative range of the five wetland plants tested in Spangler et al. (2018), which ranged between 0.052-0.200 P (g·m⁻²·d⁻¹) and 0.147-0.738 N (g·m⁻²·d⁻¹).

The experimental setup of Spangler et al. (2019) resembled our study the most, with a plant density of 15 plants/m2 and nutrient solutions within the range presented in our trial. Marigolds performed similarly to Juncus effusus and Pontedaria cordata when exposed to low nutrient solutions but were outperformed when those plants were exposed to higher nutrient solutions.

In summary, marigolds demonstrate nutrient removal performances comparable to several commonly used wetland plants tested on FTWs in mesocosm studies. Although they may not be the highest performers, marigolds could be included in FTW planting arrangements to contribute. It is important to keep in mind that the inclusion of artificial aeration would have helped diffuse nutrients and make them more accessible for plant uptake. Therefore, the nutrient removal metrics evaluated in this study may be the upper limit of nutrient uptake capacity for marigolds and may perform differently in an unaerated scenario. Due to the increased application of commercial aerators with scaled FTW installations, it is reasonable to assume this performance is still viable in the field.

4. Conclusion

This study screened 5 cut-flowers for their ability to survive in the hydroponic conditions of an FTW and evaluated the viable species for their nutrient removal rates and efficiencies. The 5 species tested were Zinnia haageana, Zinnia elegans, Helianthus annuus (branching), Helianthus annuus (singlestem), and Tagetes erecta (marigold) which were chosen for their known resilience to South Florida's subtropical climate (Zone 10). All zinnia and sunflower-treated tanks were terminated early due to premature blooms, minimal biomass, or decay from disease and pests. Using EC as a proxy, it was evident that remaining individuals from these zinnia and sunflower treatments made minimal impact on nutrient removal to their respective tanks. Improved transplanting practices could help protect these plants from shock, and a more supportive FTW design could keep stems and leaves from dropping into the water. The absence of observed root rot suggests that these species may still succeed in hydroponic conditions with improved FTW support, such as a thicker mat to help roots anchor better and keep plants upright.

The marigold-planted treatment grew the most successfully on the FTW, thriving until the end of the 12-week trial. These marigolds

² Mean \pm SD reduction (Reduction efficiency %).

removed a comparable amount of nutrients from the water as other wetland plants tested on FTWs in previous studies, with nutrient removal rates of $0.062~g\cdot d^{-1}\cdot m^2$ of OP-P and of $0.321~g\cdot d^{-1}\cdot m^2$ of NOx-N. The marigold treatment showed a significant increase in nutrient reduction efficiencies compared to the control treatment, with 52% more total phosphorus (TP) and 33% more total nitrogen (TN) mass removed from their mesocosm system.

Over the course of the trial, the marigold-planted treatment produced 64.5 market-quality blooms per $\rm m^2$, and 6.45 kg per $\rm m^2$ of fresh biomass on the FTW. Using the USDA market value of \$1 per bloom of chrysanthemum as a reference from the same plant family, these marigolds could have been sold for \$64.50 per $\rm m^2$ from the FTW. For reference, material costs for all supplies totaled \$96.89 per $\rm m^2$ (this includes Beemat floating wetlands, net pots, clay pebbles, and seeds). Though various FTW products can range over \$300 per $\rm m^2$, this potential revenue shows promising opportunities to recover material costs within just a few succession plantings.

Future studies would benefit from trialing marigold-planted FTWs on a field scale in open water to investigate how well they thrive in a natural environment. Considering the potential impact of temperature, flow rate, and dissolved oxygen (DO) variations on flower production and nutrient uptake, it is crucial to conduct field trials spanning multiple seasons to gain a comprehensive understanding of the optimal implementation of marigolds on Floating Treatment Wetlands (FTWs).

Routine biomass harvesting from FTWs has been shown to optimize the nutrient removal impacts of FTWs but can be costly due to increased labor and maintenance expenditures. In areas with water scarcity, the adequacy of management measures for this resource is fundamental and shouldn't be limited by funding. Cultivating market crops that produce continuous yields like these cut-flowers could help both offset the regular costs of FTWs and incentivize regular biomass harvesting throughout the season. The production of cut flowers on FTWs specifically in South Florida can leverage the existing floral industry infrastructure that exists in Miami, FL while also helping to address critical water quality issues in the region.

CRediT authorship contribution statement

Jazmin Locke-Rodriguez: Conceptualization, Investigation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing. Tiffany Troxler: Conceptualization, Methodology, Resources, Investigation, Data curation, Writing – review & editing. Michael C. Sukop: Data curation, Writing – review & editing. Leonard Scinto: Methodology, Writing – review & editing. Krish Jayachandran: Funding acquisition, Resources, Project administration, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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