Timing and Rates of Two Products Using Hydrogen Peroxide (H₂O₂) to Control Algae in Ebb and Flow Hydroponic Systems

Dharti Thakulla and Bruce L. Dunn

Department of Horticulture and Landscape Architecture, Oklahoma State University, 358 Ag Hall, Stillwater, OK 74078

Carla Goad

Department of Statistics, Oklahoma State University, 301F MSCS Building, Stillwater, OK 74078

Bizhen Hu

Department of Horticulture and Landscape Architecture, Oklahoma State University, 358 Ag Hall, Stillwater, OK 74078

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Abstract. Algae is not desirable in hydroponics and creates problems such as reduced yield and decreased dissolved oxygen, and affects the physiology of plants and, thus, needs to be controlled. An experiment was conducted in Ebb and Flow hydroponic systems to investigate the application timing and rates of two hydrogen peroxide products (Zerotol and PERpose Plus). Treatments included 35 mL weekly, 35 mL biweekly, 70 mL weekly, 70 mL biweekly, and a control with no application of hydrogen peroxide using a 40-gallon reservoir of water. Pepper ‘Early Jalapeno’ and ‘Lunchbox Red’ and tomato ‘Geronimo’ and ‘Little Sicily’ were used. The study was conducted in a split-plot design with two replications over time. Plant growth parameters, including plant height, flower number, net CO₂ assimilation, fresh weight, and dry weight were recorded. Algae data, including dry weight, algae cell counts, and chl *a* were also measured. Results indicated that with increasing rate and timing of either product decreased algae counts, dry weight, and chl *a* values. However, weekly and biweekly application of 70 mL of both products were not different for algae quantification. In pepper, plant height, shoot fresh and dry weight, and root fresh and dry weight were found to be significantly greater with Zerotol 35 mL biweekly, Zerotol 70 mL weekly, PERpose Plus 35 mL biweekly, and PERpose Plus 70 mL weekly compared with the control. ‘Lunchbox Red’ was significantly greater than ‘Early Jalapeno’ in all growth parameters, except soil plant analysis development (SPAD). ‘Lunchbox Red’ had the greatest flower number, with weekly application of 70 mL PERpose Plus. In tomato, greatest flower number and SPAD were observed in ‘Geronimo’ with a weekly application of 70 mL PERpose Plus and 70 mL Zerotol, respectively. Greater shoot and root fresh and dry weight for both tomato cultivars were recorded with 35 mL biweekly or 70 mL weekly application with either product. The results from both plants as well as algae analysis suggest that weekly application of 70 mL of either Zerotol or PERpose Plus produced the best results in terms of controlling algae and improving the growth of pepper and tomato plants.

Hydroponics is the method of growing plants under soilless conditions with nutrients, water, and an inert medium (Savvas, 2003). Systems can be circulating, in which the nutrient solution is recirculated and nutrient levels are manipulated, or noncirculating, in which the nutrient solution is not recirculated and

flows through the system only once (Singh, 2017). Although there are different types of hydroponic systems, every system must deliver water, nutrients, and oxygen to achieve success for plant production (Aires, 2018). Algae needs water, nutrients, and light to grow and because a hydroponic system provides it all, algae growth is often seen in hydroponics (Morgan, 2017), especially those with recirculating nutrient solution (Schwarz and Gross, 2004).

Algae enters the system through irrigation water and is commonly present in the physical structures that make up the hydroponic system. Under uncontrolled conditions, algae can cause organic loading and clogging of

pipes (Supraja et al., 2020). Algae poses several threats to hydroponic crops by competing for the nutrients and releasing harmful toxins that might inhibit or even stop crop growth (Borowitzka, 1995; Schwarz and Gross, 2004). It is generally accepted that algae growth can affect the water quality parameters, such as pH, dissolved oxygen, and nutrients in the water, and may compete with the target vegetables (Abdel-Raouf et al., 2012). Thus, it is critical that algal levels are kept to a minimum in hydroponic systems (Tesoriero et al., 2010).

It is a common practice in hydroponics to clean the system manually by basic cleaning or using black plastic sheets to reduce algal growth (Vänninen and Koskula, 1998), but time for application and high use of labor adds to costs (Caixeta et al., 2018). Alternatively, chemical algacides are also used occasionally (Nonomura et al., 2001). In recent years, interest in environmental-friendly chemicals has been on the rise. One such chemical is hydrogen peroxide (H₂O₂), which decomposes rapidly into harmless products water and oxygen (Randhawa et al., 2012). Hydrogen peroxide is generated via superoxide, presumably in a noncontrolled manner, during electron transport processes such as photosynthesis and mitochondrial respiration, and increases in response to environmental stresses such as excess excitation (light) energy, drought, and cold (Dat et al., 2000). Many studies have reported application of H₂O₂ resulted in effective and selective inhibition of algal growth with low or no effects on other phytoplankton groups in natural waters (Barrington and Ghadouani, 2008). In plants, H₂O₂ plays an important physiological role as a reactive oxygen species (ROS), which is produced under both biotic and abiotic conditions and plays a key role against O₂-derived cell toxicity (Almeida et al., 2005). Exogenous application of H₂O₂ was found to increase plant growth, physiological activities, and biochemical properties of wax apple (*Syzygium samarangense* L. var. Jambu Madu) fruits (Khandaker et al., 2012). Similarly, Orabi et al. (2015) reported that a low level of H₂O₂ can have a significant positive effect on plant growth, endogenous growth regulators, antioxidant enzyme activity, fruit yield, and quality of tomato (*Solanum lycopersicum* L.). Many studies have demonstrated that seed priming by H₂O₂ can enhance abiotic stress tolerance by modulating ROS detoxification and by regulating multiple stress-responsive pathways and gene expression (Mohammad et al., 2015). Alternatively, high concentration of H₂O₂ can promote stomatal closure in plants, reducing net photosynthesis and growth (Xia et al., 2014), and produce carcinogenic effects and induce cell death (Nurnaeimah et al., 2020). At higher levels, H₂O₂ has also been recognized as a toxic molecule that can cause damage at different levels of cell organization and thus losses in cell viability (Gechev et al., 2005). Among ROS, H₂O₂ is recognized as comparatively long-lived ROS that can diffuse easily through membranes and reach targets far from production sites; however, H₂O₂ has strong oxidizing capacities that render it capable of interacting

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B.L.D. is the corresponding author. E-mail: bruce.dunn@okstate.edu.

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with most biomolecules resulting in oxidative stress (Wojtyla et al., 2016).

Although H₂O₂-based products have been used widely as aquatic herbicide and algacide in wastewater treatment, its application in hydroponic crop production is still limited (Caixeta et al., 2018). The signaling role of H₂O₂ in plants is well established, particularly with reference to plant processes like stress acclimation, cell wall cross-linking, stomatal behavior, phytoalexin production, regulation of the cell cycle, and photosynthesis. However, if not applied within the optimum range, H₂O₂ can result in decreased net photosynthesis and plant dry mass (Khan et al., 2016). So, the toxicity or danger associated with H₂O₂ on one hand and signaling cascades on other makes it a versatile chemical whose rate and concentration need to be tightly controlled in plants during the application (Petrov and Breusegem, 2012). The objective of this study was to find the optimum amount and timing of application of two broad-spectrum algacide-labeled H₂O₂ products to reduce algae while limiting effects on growth, including plant height, flower number, net CO₂ assimilation, SPAD, and fresh and dry weight of pepper (*Capsicum annuum* L.) and tomato plants using an Ebb and Flow hydroponic system.

Materials and Methods

Plant materials and growth conditions.

The experiment was conducted at the Department of Horticulture and Landscape Architecture Research Greenhouses in Stillwater, OK. Seeds of tomato 'Geronimo' and 'Little Sicily' and pepper 'Early Jalapeno' and 'Lunchbox Red' were obtained from Johnny's Selected Seeds (Winslow, MN). Pepper seeds were sown in oasis Horticubes (Harris Seeds, Rochester, NY) of size 1.5 cm³ on 22 Apr. 2020 and kept on the mist bench. Tomatoes were planted 3 weeks later on 13 May 2020 in oasis Horticubes of the same size. Nutrients were applied once when the seedlings were 3 weeks old. Nutrient solution was made by mixing 4.9 g 5N-5.2P-21.6K (Jack's; J.R. Peters, Allentown, PA) and 3.2 g of calcium nitrate (American Plant Products, Oklahoma City, OK) diluted in 10 L of water and watered over the seedlings. Both peppers and tomatoes were transplanted to an Ebb and Flow table [Gro Master, Maple Park (Virgil), IL] on 17 June 2020. A Styrofoam sheet containing 5-cm-diameter slots spaced 28 cm apart was placed on each table to support the plants. A 5-cm net pot was placed on each slot of the Styrofoam and one plant was placed into each net pot. Each Ebb and Flow bench was supplied with a 40-gallon-capacity tank. Tanks were filled with tap water and 147.41 g of 5N-5.2P-21.6K listed previously along with 97.52 g of calcium nitrate listed previously was added initially as recommended by Singh et al. (2019). The pH and electrical conductivity (EC) of the solution were checked every day to maintain the pH between 5.5 and 6.5 and the EC at 1.5 to 2.5 mS·cm⁻¹.

Treatments and data collection. Both Zeroto (Biosafe Systems, East Hartford, CT) and

PERpose Plus (Bioworks, Victor, NY) had four different combinations of rate and timing, 35 mL weekly and twice weekly (biweekly), 70 mL weekly and biweekly, and a control without any H₂O₂ application. Zeroto active ingredient includes hydrogen peroxide and peroxyacetic acid, whereas PERpose Plus uses hydrogen peroxide and hydrogen dioxide. Each plant on the table was scanned using a chlorophyll meter (SPAD-502; Konica Minolta, Tokyo, Japan) 63 d after transplanting. SPAD readings were taken from each plant from top, middle, and bottom leaves and then averaged to determine the chlorophyll concentration. Data on photosynthesis rate was taken using a LI-COR 6400 (LI-COR, Lincoln, NE). The LI-COR with 6400-02B LED light source chamber was used by keeping the reference CO₂ at 400 ppm. The block temperature was set at 28 °C and the light level was set at 1000 μmol·m⁻²·s⁻¹. The third leaf from the top was selected per plant and was used as a nondestructive sample for the LI-COR measurement. Plants were harvested 70 d after transplanting and data were collected on number of flowers (fully opened), plant height, and root and shoot fresh weight and dry weight (oven dried for 2 d at 53.9 °C). Dissolved oxygen (D.O.) level of the solution was measured every day with a HI9146 D.O. meter (Hanna Instruments, Woonsocket, RI).

Quantification of algae. After harvesting plants, 300 mL of solution was collected from five tables (control, Zeroto and PERpose Plus 35 mL weekly, Zeroto and PERpose Plus 70 mL biweekly) and sent to EnviroScience Laboratory (Stow, OH) for quantitative algae analysis. Total suspended solids method was used to measure the dry weight of algae. A 100-mL solution was collected per table and thoroughly mixed by shaking each bottle before vacuum filtering it through a filter paper (Whatman GF/A Glass Microfiber; Cytiva, Marlborough, MA) of known weight. The suspended algae in the filter paper was then oven dried for 24 h at 53.9 °C. The dry weight of algae along with the filter paper was measured and the dry weight of algae (mg·L⁻¹) was calculated by using the following equation as reported by Michaud (1994).

$$[\text{Weight of filter} + \text{Dried residue (mg)}]$$

$$- \text{Weight of filter (mg)]}$$

$$\times 1000/\text{volume used}$$

[1]

A hemocytometer (Hausser Scientific, Horsham, PA) was used to count the number of algae cells. A 100-μL water sample was collected from each table and 100 μL of trypan blue dye was added to make the solution for the slide before 1 μL homogeneous solution was added to the hemocytometer slide. The slide was examined under a compound microscope (Olympus, Waltham, MA) at ×40 and the average number of viable algae cells were counted. The average cell count was multiplied by 10,000X the dilution factor (2) to calculate the algae concentration (viable cells/mL) according to LeGresley and McDermott (2010).

A water sample from each treatment was collected to measure the chlorophyll (chl)-a of algae with a spectrophotometer (GENESYS 30; The Laboratory Depot, Dawsonville, GA) to measure the absorbance of the samples at various wavelengths (750 nm, 665 nm, 647 nm, and 630 nm) as reported by Kumar and Saramma (2013).

Experimental design and statistics. Eight plants per cultivar per species were randomly planted in tables and treatments were arranged in a split-plot design with two replications over time. The experiment was replicated by planting another set of pepper seeds on 28 Aug. 2020 and tomatoes on 18 Sept. 2020 adopting the similar methods listed previously. Statistical analysis was performed using SAS/STAT software (version 9.4; SAS Institute, Cary, NC). Tests of significance were reported at the 0.05, 0.001, and 0.0001 level. The data were analyzed using generalized linear mixed models methods. Tukey multiple comparison methods were used to separate the means.

Results

Quantification of algae. Dry weight of algae, chl *a*, and algal cell counts were significantly affected by the rate and application timing of H₂O₂ products (Table 1). As H₂O₂ rate increased, dry weight, algal cells, and chl *a* were found to decrease. Lowest algae dry weight was recorded with 70 mL biweekly application of either product; however, this was not different from 70 mL weekly treatment, and 35 mL biweekly treatment. A similar trend was observed in algal cell counts as well, with the lowest cell counts in 70 mL biweekly treatment of either product. Lowest chl *a* was recorded in samples collected from 70 mL biweekly treatment of either product, which was similar to all other treatments except control and 35 mL weekly treatment for either product. Algae species were still found in treatments that were sent for analysis with *Chlamydomonas* spp., *Gleocystis vesiculosa*, and *Scenedesmus acutus* being recorded in the greatest numbers collectively from all treatments (Table 2). *Chlamydomonas* spp. was found in all treatments, *Gleocystis vesiculosa* was found in all treatments except Zeroto 70 mL biweekly treatment, and *Scenedesmus* spp. was found in all treatments except PERpose Plus 70 mL biweekly treatment. Average cells/mL of water for *Scenedesmus acutus* decreased 99.9%, 99.8%, and 94.8% in Zeroto 35 mL weekly, PERpose Plus 35 mL weekly, and Zeroto 70 mL biweekly treatment, respectively, compared with the control. Average cells/mL of pennate diatom species decreased 99.8%, 98%, 66.3%, and 52.1% in Zeroto 70 mL biweekly, Zeroto 35 mL weekly, PERpose Plus 70 mL biweekly, and PERpose Plus 35 mL weekly treatment, respectively compared with the control. Average cells/mL of centric diatom species was reduced 17.5%, 62.1%, and 99.2% compared with the control in Zeroto 35 mL weekly, PERpose Plus 35 mL weekly, and Zeroto 70 mL biweekly treatment, respectively. Average cells/mL of

Table 1. Least square means for rate and application timing of two hydrogen peroxide products on algae samples from Ebb and Flow hydroponic systems in Stillwater, OK.

Product	Rate (mL)	Timing of application	Dry wt (mg·L ⁻¹)	Algal cells (10 ⁵)	Chl <i>a</i> (μg·L ⁻¹)
Control	0	None	1082.50 a ²	37.51 a	2556.75 a
Zerotol	35	Weekly	361.30 bc	27.91 ab	1949.30 a
Zerotol	35	Biweekly	113.20 cd	11.77 bc	1409.85 ab
Zerotol	70	Weekly	105.65 d	8.79 cd	1262.85 ab
Zerotol	70	Biweekly	60.05 d	3.53 d	626.65 b
PERpose Plus	35	Weekly	444.70 ab	27.76 ab	1929.55 a
PERpose Plus	35	Biweekly	154.75 cd	13.94 abc	1297.85 ab
PERpose Plus	70	Weekly	149.50 cd	11.46 cd	1323.30 ab
PERpose Plus	70	Biweekly	67.30 d	3.84 d	593.35 b

²Means (n = 20) within a column followed by same lowercase letter are not significantly different by pairwise comparison in mixed model ($P \leq 0.05$).

Leptolyngbya spp. was recorded to be 2016 in the control treatment but was not found in any other treatments.

Plant growth of pepper. There was a significant cultivar × H₂O₂ interaction for flower number and CO₂ assimilation (Table 3). Greatest CO₂ assimilation was observed in ‘Early Jalapeno’ with Zerotol 35 mL biweekly treatment, which was not different from Zerotol 35 mL weekly and Zerotol 70 mL weekly treatments. Within cultivars, application of Zerotol 35 mL biweekly and 70 mL weekly resulted in greatest value for CO₂ assimilation in ‘Lunchbox Red’. Between cultivars for CO₂ assimilation, ‘Early Jalapeno’ was found to have greater CO₂ assimilation in comparison with ‘Lunchbox Red’. Greatest flower number was recorded in ‘Lunchbox Red’ with PERpose Plus 70 mL weekly treatment (Table 4). SPAD, plant height, and fresh and dry root

and shoot weight showed significant main effects for both cultivar and H₂O₂ (Table 3). Within H₂O₂ treatments, Zerotol 35 mL weekly treatment had the greatest SPAD reading and was found to be similar to all other treatments except the control and Zerotol 70 mL biweekly treatments (Table 5). For plant height, greatest value was recorded with Zerotol 70 mL weekly treatment but was not different from all other treatments except control, Zerotol 35 mL weekly, and Zerotol 70 mL biweekly treatments (Table 5). Greatest root fresh weight was recorded with 70 mL weekly treatment of either products and Zerotol 35 mL biweekly treatment and were different from the control treatment only. Whereas, biweekly application of 35 mL Zerotol produced plants with the greatest root dry weights but was similar to all other treatments except the control and 70 mL biweekly treatments of either product. Weekly

application of 70 mL PERpose Plus resulted in the greatest shoot fresh weight. However, 70 mL weekly application of Zerotol and 35

Table 3. Tests of effects for pepper ‘Early Jalapeno’ and ‘Lunchbox Red’ and rate and application timing of Zerotol and PERpose Plus, both hydrogen peroxide (H₂O₂) products, in Ebb and Flow hydroponic systems at Oklahoma State University research greenhouses in Stillwater, OK.

	Cultivar	H ₂ O ₂	Cultivar × H ₂ O ₂
No. of flowers	***	***	*
SPAD	***	***	NS
Height	***	***	NS
CO ₂ assimilation	***	***	*
Root fresh wt	***	***	NS
Shoot fresh wt	***	***	NS
Root dry wt	***	***	NS
Shoot dry wt	***	***	NS

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.

Table 2. Different rates and application timing of two hydrogen peroxide products on taxonomic counts of algae present in Ebb and Flow hydroponic systems in Oklahoma State University research greenhouses, Stillwater, OK.

Product	Rate (mL)	Time of application	Scientific name	Avg (cells/mL ²)	Avg (natural units/mL ²)
Control	0		<i>Scenedesmus acutus</i>	183,477	144,989
			<i>Scenedesmus ecornis</i>	127,991	94,733
			Pennate diatom spp.	22,452	22,452
			<i>Gloeocystis vesiculosa</i>	16,410	1,675
			Centric diatom spp.	12,810	12,810
			<i>Chlamydomonas</i> spp.	2,066	2,066
			<i>Leptolyngbya</i> spp.	2,016	2,016
Zerotol	35	Weekly	<i>Gloeocystis vesiculosa</i>	331,969	331,969
			<i>Chlamydomonas</i> spp.	163,278	163,278
			Centric diatom spp.	10,567	8,884
			Pennate diatom spp.	443	443
			<i>Scenedesmus acutus</i>	266	89
			<i>Chlamydomonas</i> spp.	1,568	1,568
			<i>Scenedesmus acutus</i>	9,676	4,718
Zerotol	70	Biweekly	<i>Chlorella minutissima</i>	309	309
			Centric diatom spp.	103	103
			<i>Chlorella ellipsoidea</i>	103	103
			<i>Monoraphidium pusillum</i>	82	82
			<i>Tetrastrum komarekii</i>	41	10
			Pennate diatom spp.	34	34
			<i>Gloeocystis vesiculosa</i>	57,388	46,309
			<i>Chlamydomonas</i> spp.	602,261	602,261
			Pennate diatom spp.	10,744	10,744
			Centric diatom spp.	4,855	4,855
PERpose Plus	35	Weekly	<i>Scenedesmus acutus</i>	413	207
			Pennate diatom spp.	7,556	7,556
			<i>Gloeocystis vesiculosa</i>	3,142	399
			<i>Aphanocapsa nubilum</i>	1,771	59
			<i>Chlamydomonas</i> spp.	1,391	1391
			<i>Oocystis parva</i>	266	59
			<i>Chroococcus dispersus</i>	142	14
PERpose Plus	70	Biweekly	<i>Cyclotella</i> spp.	30	30

²Derived from a 100-mL solution.

Table 4. Least squares means for 'Early Jalapeno' and 'Lunchbox Red' and different rate and application timing of two hydrogen peroxide products for flower number and net CO₂ assimilation of peppers grown in Ebb and Flow hydroponic systems at Oklahoma State University research greenhouses in Stillwater, OK.

Cultivars	Product	Rate (mL)	Timing of application	No. of flowers	CO ₂ assimilation (μmol·m ⁻² ·s ⁻¹)
Early Jalapeno	Control	0	None	2.1 h ^z	21.9 cdefgh
	Zerotol	35	Weekly	4.9 g	24.4 abc
	Zerotol	35	Biweekly	5.9 g	26.8 a
	Zerotol	70	Weekly	4.7 g	26.0 ab
	Zerotol	70	Biweekly	3.6 gh	20.7 efghi
	PERpose Plus	35	Weekly	4.9 g	22.7 cdefg
	PERpose Plus	35	Biweekly	4.8 g	22.8 cdefg
	PERpose Plus	70	Weekly	5.3 g	23.3 bcdef
	PERpose Plus	70	Biweekly	4.0 gh	20.8 efghi
Lunchbox Red	Control	0	None	11.5 f	19.3 hi
	Zerotol	35	Weekly	15.7 fe	20.6 fghi
	Zerotol	35	Biweekly	23.2 bcd	23.6 bcde
	Zerotol	70	Weekly	26.7 b	23.8 bcd
	Zerotol	70	Biweekly	18.8 cde	20.2 ghi
	PERpose Plus	35	Weekly	26.3 b	20.7 efghi
	PERpose Plus	35	Biweekly	24.0 bc	22.0 cdefgh
	PERpose Plus	70	Weekly	33.9 a	22.5 cdefg
	PERpose Plus	70	Biweekly	17.8 de	21.0 defghi

^zMeans (n = 16) within a column followed by same lowercase letter are not significantly different by pairwise comparison in mixed model ($P \leq 0.05$).

mL biweekly application of either product also resulted in plants with greater shoot fresh weights. Whereas, greatest shoot dry weight was recorded with Zeritol 35 mL biweekly and PERpose Plus 70 mL weekly treatments but were similar to all other treatments except control and 70 mL biweekly treatments of either product (Table 5). 'Lunchbox Red' performed better in terms of all the growth parameters except SPAD, which was greater in 'Early Jalapeno' (Table 6).

Plant growth of tomato. There was a significant cultivar × H₂O₂ interaction for all growth parameters except for net CO₂ assimilation, whereas CO₂ assimilation was found to be affected by H₂O₂ product only (Table 7). Zeritol application at the rate of 35 mL biweekly and 70 mL weekly resulted in the greatest CO₂ assimilation compared with any other treatments (Table 8). Greatest flower number was observed in 'Geronimo' with a weekly application of 70 mL PERpose Plus,

which was not different from Zeritol 35 mL biweekly and PERpose Plus 35 mL weekly treatments as well as Zeritol 35 mL weekly and 35 mL biweekly treatment of either product in 'Little Sicily'. Greatest SPAD value was recorded in 'Geronimo' within Zeritol 70 mL weekly treatment, which was also similar to PERpose Plus 70 mL weekly treatment and 35 mL biweekly treatment of either product. In 'Lunchbox Red', both weekly and biweekly application of 35 mL Zeritol resulted in the greatest SPAD and were similar to all other treatments except the control and 70 mL biweekly treatments of either product (Table 9). In 'Little Sicily', Zeritol 35 mL biweekly application resulted in the greatest plant height and was similar to all the treatments except Zeritol 70 mL biweekly treatment, whereas 'Geronimo' had the greatest plant height with PERpose Plus 70 mL biweekly treatment and was similar to all the treatments except Zeritol 35 mL biweekly

and PERpose Plus 70 mL biweekly treatments. When compared across the cultivars, 'Little Sicily' had greater plant height than 'Geronimo' (Table 9). Greater fresh and dry root weight were observed in 'Little Sicily' in comparison with 'Geronimo'. In 'Little Sicily', greatest root fresh weight was observed with biweekly application of 35 mL Zeritol and was similar to all other treatments except control and 70 mL biweekly treatments of either product. The greatest root dry weight was observed in PERpose Plus 35 mL biweekly treatment, which was similar to 35 mL weekly and biweekly treatments of Zeritol, as well as 35 mL and 70 mL weekly treatments of PERpose Plus. In 'Geronimo', 70 mL weekly application of either product resulted in the greatest fresh weight of roots and was similar to all the treatments except the control, Zeritol 35 mL weekly, and PERpose Plus 70 mL biweekly treatments. However, greater root dry weight was recorded in PERpose Plus 70 mL weekly treatment compared with any other treatments (Table 9). 'Little Sicily' had the greatest shoot fresh weight with PERpose Plus 70 mL weekly treatment, which was similar to all other treatments except the control and Zeritol 70 mL biweekly treatments. The shoot dry weight was found to be greatest with PERpose Plus 70 mL biweekly treatment but was only different from the control treatment. In 'Geronimo', PERpose Plus 35 mL biweekly and 70 mL weekly treatments resulted in greatest shoot fresh weight but was not different from PERpose Plus 35 mL weekly, Zeritol 35 mL biweekly, and Zeritol 70 mL weekly treatments. For shoot dry weight, PERpose Plus 70 mL weekly had the greatest value but was similar to PERpose Plus 35 mL biweekly treatment (Table 9).

Dissolved oxygen. D.O. levels varied from 5.2 to 6.6, 6.2 to 7.5, 6.2 to 7.9, and 6.8 to 8.2 mg·L⁻¹ for PERpose Plus treatments of 35 mL weekly, 35 mL biweekly, 70 mL weekly, and 70 mL biweekly, respectively (Fig. 1). Average D.O. levels were 6.0, 6.6, 7.0, and 7.7 mg·L⁻¹ for PERpose Plus treatments of 35 mL weekly, 35 mL biweekly, 70 mL weekly, and 70 mL biweekly, respectively.

Table 5. Least square means for rate and application timing of two hydrogen peroxide products on growth of pepper 'Early Jalapeno' and 'Lunchbox Red' grown in Ebb and Flow hydroponic systems at Oklahoma State University research greenhouses in Stillwater, OK.

Product	Rate (mL)	Time of application	SPAD	Ht (cm)	Shoot wt (g)	Root wt (g)	Shoot dry wt (g)	Root dry wt (g)
Control	0	None	44.9 c ^z	31.1 cd	36.4 ef	23.7 c	8.0 cd	2.6 d
Zerotol	35	Weekly	49.4 a	33.2 bcd	46.3 de	37.5 ab	9.9 abc	3.1 ab
Zerotol	35	Biweekly	47.8 abc	37.9 ab	68.7 abc	42.3 a	13.4 a	3.6 a
Zerotol	70	Weekly	47.9 abc	39.4 a	71.4 ab	43.4 a	12.6 ab	3.0 abc
Zerotol	70	Biweekly	45.3 bc	30.5 d	38.8 ef	31.5 b	9.2 bcd	2.6 bcd
PERpose Plus	35	Weekly	46.6 abc	37.0 abc	55.1 bcd	37.9 ab	10.1 abc	3.1 ab
PERpose Plus	35	Biweekly	48.7 ab	34.2 abcd	62.3 abcd	41.1 ab	11.4 ab	3.1 ab
PERpose Plus	70	Weekly	47.5 abc	36.4 abcd	78.9 a	45.4 a	13.1 a	3.3 ab
PERpose Plus	70	Biweekly	46.8 abc	34.4 abcd	50.1 cde	35.3 ab	8.0 cd	2.3 cd

^zMeans (n = 16) within a column followed by same lowercase letter are not significantly different by pairwise comparison in mixed model ($P \leq 0.05$).

Table 6. Least square means for 'Early Jalapeno' compared with 'Lunchbox Red' for growth of peppers grown in Ebb and Flow hydroponic systems at Oklahoma State University research greenhouses in Stillwater, OK.

Cultivar	SPAD	Ht (cm)	Shoot fresh wt (g)	Root fresh wt (g)	Shoot dry wt (g)	Root dry wt (g)
Early Jalapeno	50.4 a ^z	29.0 b	28.0 b	24.1 b	4.7 b	2.4 b
Lunchbox Red	43.4 b	40.7 a	96.5 a	51.6 a	21.6 a	3.5 a

^zMeans (n = 16) within a column followed by same lowercase letter are not significantly different by pairwise comparison in mixed model ($P \leq 0.05$).

Table 7. Tests of effects for tomato ‘Little Sicily’ and ‘Geronimo’ and rate and application timing of Zeritol and PERpose Plus, both hydrogen peroxide (H₂O₂) products, in Ebb and Flow hydroponic systems at Oklahoma State University research greenhouses in Stillwater, OK.

	Cultivar	H ₂ O ₂	Cultivar × H ₂ O ₂
No. of flowers	*	***	**
SPAD	***	***	*
Height	***	***	**
CO ₂ assimilation	NS	***	NS
Root wt	***	***	***
Shoot wt	***	***	*
Root dry wt	***	***	***
Shoot dry wt	***	***	*

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.001, or 0.0001, respectively.

D.O. levels varied from 5.4 to 6.4, 6.0 to 7.6, 6.1 to 7.8, and 6.1 to 8.1 mg·L⁻¹ for Zeritol treatments of 35 mL weekly, 35 mL biweekly, 70 mL weekly, and 70 mL biweekly, respectively (Fig. 1). Average D.O. levels were 5.9, 6.6, 6.9, and 7.6 mg·L⁻¹ for Zeritol treatments of 35 mL weekly, 35 mL biweekly, 70 mL weekly, and 70 mL biweekly, respectively, whereas the control levels varied from 5.1 to 5.7 mg·L⁻¹ and averaged 5.5 mg·L⁻¹. Spikes in D.O. levels were observed on days in which H₂O₂ was added to the tanks, then levels decreased within a couple of days until the next application. The trend showed that with increasing dose and application of H₂O₂, there was an increase in D.O. level.

Discussion

In the present study, dry weight of algae, chl *a*, and algal cell counts were found to decrease with increasing rate and application timing of H₂O₂ products. Hydrogen peroxide is a strong oxidant that has been used in industrial application and water treatment processes (Ismail et al., 2015). When catalyzed in water, H₂O₂ can generate a wide variety of free radicals and other reactive species that are capable

Table 8. Least square means for rate and application timing of two hydrogen peroxide products on net CO₂ assimilation of tomato ‘Little Sicily’ and ‘Geronimo’ grown in Ebb and Flow hydroponic systems at Oklahoma State University research greenhouses in Stillwater, OK.

Product	Rate (mL)	Time of application	CO ₂ assimilation (μmol·m ⁻² ·s ⁻¹)
Control	0	None	20.7 de ^z
Zeritol	35	Weekly	23.4 b
Zeritol	35	Biweekly	25.9 a
Zeritol	70	Weekly	25.5 a
Zeritol	70	Biweekly	20.2 e
PERpose Plus	35	Weekly	22.7 bc
PERpose Plus	35	Biweekly	23.1 b
PERpose Plus	70	Weekly	23.1 b
PERpose Plus	70	Biweekly	21.6 cd

^zMeans (n = 16) within a column followed by same lowercase letter are not significantly different by pairwise comparison in mixed model ($P \leq 0.05$).

of transforming or decomposing organic chemicals (Petri et al., 2003). Hydrogen peroxide-based products have been used as aquatic herbicides and microbiocide to manage algae, cyanobacteria, fungi, and microorganisms in water (Randhawa et al., 2012). However, the efficiency of H₂O₂ can be influenced by abiotic factors such as light intensity and nutrient availability, as well as biotic factors like phytoplankton and bacterioplankton composition (Santos et al., 2021). According to a study conducted by Vänninen and Koskula (1998), a single application of 1 dL of 125 ppm H₂O₂ or daily applications over 3 weeks of 1 dL of 100 ppm peroxide reduced algal growth by 40% to 60%, which was similar to our findings. However, our results showed that both Zeritol and PERpose Plus reduced algae by as much as 95% even at lower rates. Exposure of *Planktothrix agardhii* to H₂O₂ resulted in the degeneration of cells and filaments. Swollen cell walls and some degree of cytoplasmic alteration were observed after 24 h of treatment with 0.83 mg·L⁻¹ H₂O₂, whereas, after 48 h under the highest concentration (3.33 mg·L⁻¹) resulted in cell wall and plasmatic membrane disruption, disorganized and degraded thylakoids, and changes in the cytoplasmic inclusions (Bauzá et al., 2014). The same study also reported that the chl *a* decreased 40%, 86%, 86%, 78%, and

79%, respectively, with application of 0.17, 0.33, 0.83, 1.67, and 3.33 mg·L⁻¹ H₂O₂, which corresponds to our finding that chl *a* decreases with increasing rate of H₂O₂. Santos et al. (2021) evaluated the effects of a single application of H₂O₂ (10 mg·L⁻¹) over 120 h in mesocosms introduced in a reservoir and reported that H₂O₂ efficiently decreased the biomass of cyanobacteria, green algae, and diatoms over 72 h along with the chlorophyll concentration, leading to an increase in transparency. However, after 120 h, subsequent dominance of green algae was observed. In some cases, suppression of one algae species might benefit the growth of another species, which could be the reason why some of the treatments in our experiment had new species that were not recorded in the control treatment. Sinha et al. (2018) observed that a reduction in cyanobacteria biomass was followed by an increase in the abundance of eukaryotic diatom *Synedra* sp. and green algae *Cladophora* sp., suggesting that these species benefited from the collapse of cyanobacteria and used the available nutrients. In another study conducted by Wang et al. (2019), in which a *Microcystis aeruginosa* bloom was suppressed with the application of H₂O₂, growth of *Chlamydomonas* spp. was found to be promoted.

Our results indicated that either a 70-mL weekly or 35-mL biweekly application of

Table 9. Least square means for tomato ‘Little Sicily’ and ‘Geronimo’ and different rate and application timing of two hydrogen peroxide products for growth of tomatoes grown in Ebb and Flow hydroponic system at Oklahoma State University research greenhouses in Stillwater, OK.

Cultivars	Product	Rate (mL)	Timing of application	No. of flowers	SPAD	Ht (cm)	Shoot fresh wt (g)	Root fresh wt (g)	Shoot dry wt (g)	Root dry wt (g)
Little Sicily	Control	0	None	17.6 efg ^z	47.2 f	97.0 ab	276.3 def	70.1 efg	55.8 bcdef	6.6 cdefg
	Zeritol	35	Weekly	25.6 abc	55.5 bcd	101.6 ab	527.0 abc	110.6 abcd	105.2 ab	7.7 abcde
	Zeritol	35	Biweekly	26.5 abc	56.1 bcd	102.2 a	500.6 abcd	142.8 a	97.6 ab	9.0 abc
	Zeritol	70	Weekly	22.9 bcde	55.2 bcde	101.0 ab	453.3 abcd	111.4 abcd	85.6 abc	7.3 bcde
	Zeritol	70	Biweekly	15.3 g	50.3 ef	72.5 c	357.9 bcdef	89.6 cdef	69.0 abcd	5.6 efgh
	PERpose Plus	35	Weekly	23.7 bcd	51.5 def	100.0 ab	528.3 abc	103.5 abcd	84.5 abc	7.7 abcde
	PERpose Plus	35	Biweekly	25.9 abc	52.0 def	95.3 ab	635.4 ab	133.3 ab	91.9 ab	10.5 a
	PERpose Plus	70	Weekly	24.0 bc	55.2 bcde	97.2 ab	680.6 a	124.2 abc	88.0 ab	9.4 ab
	PERpose Plus	70	Biweekly	21.9 defg	50.5 ef	90.6 abc	506.7 abcd	93.4 bcde	119.6 a	6.9 bcdef
	PERpose Plus	70	Biweekly	21.7 cdef	51.4 def	45.8 def	214.7 f	52.9 g	27.9 g	4.9 ghi
Geronimo	Control	0	None	21.7 cdef	51.4 def	45.8 def	214.7 f	52.9 g	27.9 g	4.9 ghi
	Zeritol	35	Weekly	22.6 cde	56.6 bcd	46.0 def	223.9 f	67.1 efg	44.3 def	5.3 fgh
	Zeritol	35	Biweekly	25.3 abc	58.2 abc	42.6 efg	391.8 bcd	82.9 def	39.6 efg	5.9 efgh
	Zeritol	70	Weekly	24.6 bc	62.9 a	45.2 def	366.1 bcde	103.8 abcd	40.7 def	5.7 efgh
	Zeritol	70	Biweekly	15.1 g	56.5 cde	46.3 def	251.7 ef	90.6 cde	37.5 fg	5.8 efgh
	PERpose Plus	35	Weekly	29.2 ab	55.1 bcde	50.6 de	284.5 def	82.8 def	33.9 fg	5.9 efgh
	PERpose Plus	35	Biweekly	24.1 bc	58.8 ab	48.7 de	415.8 abcd	90.5 cde	48.2 def	6.2 defgh
	PERpose Plus	70	Weekly	31.3 a	59.7 ab	52.0 d	442.3 abcd	112.8 abcd	51.0 cdef	8.0 abcd
	PERpose Plus	70	Biweekly	22.0 cde	52.7 bcd	39.5 fg	211.5 f	67.3 efg	33.7 fg	3.9 i

^zMeans (n = 16) within a column followed by same lowercase letter are not significantly different by pairwise comparison in mixed model ($P \leq 0.05$).

Fig. 1. Effects of rate and application timing of two H_2O_2 products Zerotel (Zer) and PERpose Plus (PP) (35 mL weekly and biweekly, 70 mL weekly and biweekly, and control) on dissolved oxygen (D.O.) levels of nutrient solution in Ebb and Flow hydroponic systems at Oklahoma State University research greenhouses in Stillwater, OK.

either H_2O_2 product resulted in improved growth performance of both pepper and tomato plants. This may be due to the positive role of H_2O_2 in plant growth and development. H_2O_2 is said to be the most stable ROS, and therefore plays a crucial role as a signaling molecule in various physiological processes, including photosynthesis, respiration, translocation, and transpiration (Slesak et al., 2007). Growth (root and shoot length; fresh and dry weight) and photosynthetic performance of cowpea (*Vigna unguiculata* L.) was found to increase by foliar application of H_2O_2 at 0.5 to 1.0 mM solution (Hasan et al., 2016). Similarly, root dipping treatment of H_2O_2 to tomato plants under copper stress and stress-free conditions improved growth, including shoot and root length, shoot and root fresh weight, shoot and root dry weight, and leaf area (Nazir et al., 2019). Foliar application of diluted H_2O_2 (at 20 mM) has been shown to increase the dry weight of leaves, and increase fruit set, biomass, and quality of fruit, as well as decreasing bud drop in wax apple ('Jambu Madu') crops (Khandaker et al., 2012). Uchida et al. (2002) reported that H_2O_2 application induces the activity of sucrose phosphate synthase, which is an enzyme important in the formation of sucrose from triose phosphates during and after photosynthesis and positively regulates the metabolic and antioxidant enzyme activities in favor of plant growth and development. Furthermore, application of H_2O_2 has been found to increase the shoot and root length in two wheat (*Triticum aestivum* L.) cultivars by

increasing the activity of starch hydrolyzing enzymes, which resulted in better root carbohydrate. As the root length increases, absorption of water and nutrients usually increases, leading to increased growth parameters and prevention against the harmful effects of stress (Latef et al., 2019). On the other hand, H_2O_2 functions by decomposing into an unstable free radical oxygen molecule that can destroy biotic cell tissue and has the potential to indiscriminately damage healthy living root tissue, consequently reducing the fresh weight as well (Lau and Mattson, 2021). Lower root and shoot weight at 70 mL biweekly treatments in our study may be the result of this phytotoxicity.

Khan et al. (2018) studied the effects on roots of two varieties (S-22 and PKM-1) of tomato using three different concentrations of H_2O_2 (0.01, 0.1, and 0.5 mM). The 0.1 mM application of H_2O_2 for 4 h proved best and resulted in improved growth and photosynthetic attributes, enhanced activity of various antioxidant enzymes, and greater accumulation of proline. However, the effects were more prominent in 'S-22' compared with 'PKM-1'. The induced activity of antioxidant enzymes and proline content by H_2O_2 was recorded to be more in 'S-22' than 'PKM-1', which helped in protecting photosynthesis and maintaining high plant fresh and dry mass than in PKM-1. They concluded that both the concentration of H_2O_2 as well as cultivars influenced the effects of H_2O_2 on growth of tomato, which supports our findings.

Physiological effects of exogenous application of H_2O_2 and calcium (Ca) were studied on sweet and hot pepper under heat stress and a significant change in biochemical, physiological, and fatty acid contents in both cultivars was reported; however, sweet peppers absorbed greater amounts of Ca in comparison with the hot peppers (Motamedi et al., 2019). Ca is an essential macronutrient in plants and provides structural stability to cell walls and membranes and acts as a secondary messenger in cellular signaling (White and Broadley, 2003). This could be a reason for greater height and shoot and root weights in 'Lunchbox Red' compared with 'Early Jalapeno'.

Hydrogen peroxide treatment also improved photosynthesis as a result of improved gas exchange parameters, chlorophyll content, and net photosynthesis along with improved scavenging of ROS and recovery of photosynthetic efficiency in tomato (Nazir et al., 2019). In addition, H_2O_2 treatment also improved chlorophyll content in mung bean (*Vigna radiata* L.) and tomato by modulating endogenous plant hormones (Fariduddin et al., 2014; Orabi et al., 2015). Root dipping treatment with H_2O_2 significantly increased photosynthesis of tomato plants because of increase in the activity of Rubisco and PS II resulting in increased stomatal conductance (g_s) and intercellular CO_2 concentration (Khan et al., 2018). In a similar study conducted by Nurnaeimah et al. (2020), plant height, leaf area, chlorophyll content, net photosynthetic rate, g_s , and quantum yield of

mistletoe fig (*Ficus deltoidea* L.) increased after treatment with 16 and 30 mM H₂O₂. They concluded that the application of H₂O₂ increased stomatal opening, CO₂ concentration, and accumulated photosynthetic pigments, which ultimately resulted in the increased photosynthetic rate. However, increasing the concentration of H₂O₂ to 60 mM led to a decrease in growth parameters and increased the accumulation of arsenic, iron, and sodium content in the leaves of mistletoe fig. These results can be supported by the fact that H₂O₂ at low concentration acts as a regulator of some major processes, such as assimilation, photosynthesis, respiration, g_s, cell cycle, growth and development, and plant response to biotic and abiotic pressure; however, it can increase the oxidative damage, and ultimately cause cell death above a certain threshold (Latef et al., 2019). The study of Xia et al. (2014) also reported that a low concentration of H₂O₂ promotes stomatal opening, whereas a high concentration of H₂O₂ promotes stomatal closure in tomatoes. This might be a reason why a decrease in growth parameters of pepper and tomato was observed in our study with 70 mL biweekly application of either H₂O₂ products. In addition, H₂O₂ has also been reported to promote reproductive growth in fruits by inhibiting the growth of rudimentary leaves as well as by promoting the expression of the flower-related gene, LcLFY and reducing bud drop (Ismail et al., 2015).

An increase in D.O. levels with increased rate and application timing of H₂O₂ was observed. In hydroponic nutrient solutions, decomposition of H₂O₂ gives rise to byproducts H₂O and O₂, and this released O₂ can increase the D.O. concentration in the root zone (Lau and Mattson, 2021). Hydrogen peroxide, when added into nutrient solution, breaks down, readily releasing a molecule of water and a reactive O-molecule, which can bind with another O-molecule. This O-molecule can also react with organic compounds, which often results in degradation of the compound (Fredrickson, 2014). In addition, reduced amounts of algae in water can result in greater D.O. level as the result of increased permeability of light in water (Lauguico et al., 2020). Plant growth of hydroponic crops has been shown to increase when irrigated with additional oxygen in the solution (Soffer et al., 1990). Butcher (2016) reported that the treatment with high-frequency application of H₂O₂ alone produced the greatest level of D.O., whereas combination of high-frequency application (every 3 d) of H₂O₂, vortex oxygenation, and air-pump injection treatment yielded the greatest production of chlorophyll in peppermint geranium (*Pelargonium tomentosum* L.). The increased production of chlorophyll was likely due to healthy root growth and respiration within a nutrient solution where high levels of D.O. and turbulence were facilitated. According to Chérif et al. (1997), tomato plant roots were susceptible to *Pythium* infection when the oxygen in the root zone dropped below 2.8 mg·L⁻¹. Poor root and plant performance as

well as an increase in the incidence of disease were reported when the root zone was oxygen deficient (Chérif et al., 1997). Marfà et al. (2005) reported greater yield of pepper plants grown in perlite with nutrient solution enriched with oxygen (16 mg·L⁻¹) compared with unenriched solution (6 mg·L⁻¹). In contrast to our result, Ouyang et al. (2021) reported a significant increase in growth, photosynthesis, yield, and quality of tomato when the D.O. of irrigation water was increased (9 mg·L⁻¹) compared with the control (4 mg·L⁻¹). This could be because they used aeration as a method of increasing D.O., whereas the high level of H₂O₂ in our study might have caused phytotoxicity and resulted in poor plant growth.

Conclusion

With increasing rate and timing of either H₂O₂ product, algae counts, dry weight, and chl *a* values decreased. However, weekly and biweekly application of 70 mL of the products were not significantly different from each other for controlling algae growth, so weekly would be recommended. Surprisingly, Zeritol 35 mL biweekly, Zeritol 70 weekly, PERpose Plus 35 mL biweekly, and PERpose Plus 70 mL weekly resulted in better growth in both tomato and pepper. Our results also indicated that 70 mL biweekly application of either product caused phytotoxicity and resulted in decreased growth. Results from both plants as well as algae analysis suggested that weekly application of 70 mL of either Zeritol or PERpose Plus produced the best results in terms of controlling algae and improving the growth of pepper and tomato plants. Future research should investigate other plants using these two products and rates, effects on fruiting, and overall nutrition of fruits.

Literature Cited

- Abdel-Raouf, N., A.A. Al-Homaidan, and I.B.M. Ibraheem. 2012. Microalgae and wastewater treatment. *Saudi J. Biol. Sci.* 19:257–275, <https://doi.org/10.1016/j.sjbs.2012.04.005>.
- Aires, A. 2018. Hydroponic production systems: Impact on nutritional status and bioactive compounds of fresh vegetables. *IntechOpen* 55–66, <https://doi.org/10.5772/intechopen.73011>.
- Almeida, J.M., F. Fidalgo, A. Confraria, A. Santos, H. Pires, and I.E. Santos. 2005. Effect of hydrogen peroxide on catalase gene expression, isoform activities and levels in leaves of potato sprayed with homobrassinolide and ultrastructural changes in mesophyll cells. *Funct. Plant Biol.* 32:707–720, <https://doi.org/10.1071/FP04235>.
- Barrington, D.J. and A. Ghadouani. 2008. Application of hydrogen peroxide for the removal of toxic cyanobacteria and other phytoplankton from wastewater. *Environ. Sci. Technol.* 42: 8916–8921, <https://doi.org/10.1021/es801717y>.
- Bauzá, L., A. Aguilera, R. Echenique, D. Andrinolo, and L. Giannuzzi. 2014. Application of hydrogen peroxide to the control of eutrophic lake systems in laboratory assays. *Toxins (Basel)* (6): 2657–2675, <https://doi.org/10.3390/toxins6092657>.
- Borowitzka, M.A. 1995. Micro-algae as sources of pharmaceutical and other biologically active

- compounds. *J. Appl. Phycol.* 7:3–15, <https://doi.org/10.1007/BF00003544>.
- Butcher, J.D. 2016. A comparative study of oxygenation techniques in the hydroponic cultivation of *Pelargonium tomentosum*. Cape Peninsula University of Technology, Cape Town, South Africa, Master's Thesis.
- Caixeta, V., A. Mata, C. Curvelo, W. Tavares, L. Ferreira, and A. Pereira. 2018. Hydrogen peroxide for insect and algae control in a lettuce hydroponic environment. *J. Agr. Sci.* 10(8):221, <https://doi.org/10.5539/jas.v10n8p221>.
- Chérif, M., Y. Tirilly, and R.R. Bélanger. 1997. Effect of oxygen concentration on plant growth, lipid peroxidation, and receptivity of tomato roots to *Pythium F* under hydroponic conditions. *Eur. J. Plant Pathol.* 103:255–264, <https://doi.org/10.1023/A:1008691226213>.
- Dat, J., S. Vandenabeele, E. Vranová, M.V. Montagu, D. Inzé, and F.V. Breusegem. 2000. Dual action of the active oxygen species during plant stress responses. *Cell. Mol. Life Sci.* 57(5):779–795, <https://doi.org/10.1007/s000180050041>.
- Fariduddin, Q., T.A. Khan, and M. Yusuf. 2014. Hydrogen peroxide mediated tolerance to copper stress in the presence of 28-homobrassinolide in *Vigna radiata*. *Acta Physiol. Plant.* 36:2767–2778, <https://doi.org/10.1007/s11738-014-1647-0>.
- Fredrickson, B. 2014. Hydrogen peroxide and horticulture. *Quick Grow* 5 Nov. 2021. <<https://www.quickgrow.com/hydrogen-peroxide-horticulture/>>.
- Gechev, T.S., I.N. Minkov, and J. Hille. 2005. Hydrogen peroxide-induced cell death in Arabidopsis: Transcriptional and mutant analysis reveals a role of an oxoglutarate-dependent dioxygenase gene in the cell death process. *IUBMB* 57:181–188, <https://doi.org/10.1080/15216540500090793>.
- Hasan, S.A., M. Irfan, Y.S. Masrahi, M.A. Khalaf, and S. Hayat. 2016. Growth, photosynthesis, and antioxidant responses of *Vigna unguiculata* L. treated with hydrogen peroxide. *Cogent Food Agr.* 2(1):1–13, <https://doi.org/10.1080/23311932.2016.1155331>.
- Ismail, S.Z., M.K. Mohammad, M. Nashriyah, and N.B. Amru. 2015. Effects of hydrogen peroxide on growth, development and quality of fruits: A review. *J. Agron.* 14:331–336, <https://doi.org/10.3923/ja.2015.331.336>.
- Khan, M.I.R., N.A. Khan, M. Asim, T.S. Per, and M. Asgher. 2016. Hydrogen peroxide alleviates nickel-inhibited photosynthetic responses through increase in use-efficiency of nitrogen and sulfur, and glutathione production in mustard. *Front. Plant Sci.* 7:44, <https://doi.org/10.3389/fpls.2016.00044>.
- Khan, T.A., Q. Fariduddin, and M. Yusuf. 2018. Effect of exogenously sourced hydrogen peroxide treatments on growth, photosynthesis and antioxidant traits in two contrasting cultivars of tomato: A model and concentration dependent study. *AJAR* 6(1):019–029, <https://doi.org/10.15413/ajar.2018.1201>.
- Khandaker, M.M., A.N. Boyce, and N. Osman. 2012. The influence of hydrogen peroxide on the growth, development and quality of wax apple (*Syzygium samarangense*, [Blume] Merrill & L.M. Perry var. jambu madu) fruits. *Plant Physiol. Biochem.* 53:101–110, <https://doi.org/10.1016/j.plaphy.2012.01.016>.
- Kumar, S. and A.V. Saramma. 2013. A revised method for pigment extraction from marine nannoplanktonic algal cultures. *J. Algal Biomass Util.* 4(2):47–52.
- Latef, A.A., M. Kordrostami, A. Zakir, H. Zaki, and O.M. Saleh. 2019. Eustress with H₂O₂ facilitates plant growth by improving tolerance

- to salt stress in two wheat cultivars. *Plants* 8(9):303, <https://doi.org/10.3390/plants8090303>.
- Lau, V. and N. Mattson. 2021. Effects of hydrogen peroxide on organically fertilized hydroponic Lettuce (*Lactuca sativa* L.). *Horticulturae* 7:106, <https://doi.org/10.3390/horticulturae7050106>.
- Lauguico, S., R. Concepcion, R.R. Tobias, J. Alejandrino, D. Macasaet, and E. Dadios. 2020. Indirect measurement of dissolved oxygen based on algae growth factors using machine learning models. *IEEE 8th R10 Humanitarian Technology Conference (R10-HTC)* 1-6, <https://doi.org/10.1109/R10-HTC49770.2020.9357014>.
- LeGresley, M. and G. McDermott. 2010. Counting chamber methods for quantitative phytoplankton analysis—haemocytometer, Palmer-Maloney cell and Sedgewick-Rafter cell. *UNESCO (IOC Manuals and Guides)* 25-30.
- Marfa, O., R. Cáceres, and S. Guri. 2005. Oxygenation: A new technique for soilless culture under Mediterranean conditions. *Acta Hort.* 697:65–72, <https://doi.org/10.17660/ActaHortic.2005.697.6>.
- Michaud, J.P. 1994. Measuring total suspended solids and turbidity in lakes and streams. *A Citizen's Guide to Understanding and Monitoring Lakes and Streams*. Washington State Dept. of Ecology, Publications Office, Olympia, WA.
- Mohammad, A.H., S. Bhattacharjee, S.M. Armin, P. Qian, W. Xin, H. Li, D.J. Burritt, M. Fujita, and L.P. Tran. 2015. Hydrogen peroxide priming modulates abiotic oxidative stress tolerance: Insights from ROS detoxification and scavenging. *Front. Plant Sci.* (New Haven) 6:420, <https://doi.org/10.3389/fpls.2015.00420>.
- Morgan, L. 2017. Algae growth in your hydroponic system: Friend or foe? *Maximum Yield* 5 Nov. 2021. <<https://www.maximumyield.com/algae-friend-or-foe/21180>>.
- Motamedi, M., M. Haghighi, and A. Goli. 2019. Physiological changes of sweet and hot peppers in vegetative and reproductive growth stages treated by Ca and H₂O₂ under unforeseen heat stresses. *Scientia Hort.* 249:306–313, <https://doi.org/10.1016/j.scienta.2019.01.040>.
- Nazir, F., A. Hussain, and Q. Fariduddin. 2019. Hydrogen peroxide modulate photosynthesis and antioxidant systems in tomato (*Solanum lycopersicum* L.) plants under copper stress. *Chemosphere* 230:544–558, <https://doi.org/10.1016/j.chemosphere.2019.05.001>.
- Nonomura, T., Y. Matsuda, M. Bingo, M. Onishi, K. Matsuda, S. Harada, and H. Toyoda. 2001. Algicidal effect of 3-(3-indolyl) butanoic acid, a control agent of the bacterial wilt pathogen, *Ralstonia solanacearum*. *Crop Prot.* 20:935–939, [https://doi.org/10.1016/S0261-2194\(01\)00048-5](https://doi.org/10.1016/S0261-2194(01)00048-5).
- Nurnaimah, N., M. Nashriyah, S.M. Khamsah, A.B. Noor, Y. Norasuha, H.S. Mohammad, M. Khairil Mahmud, A.F. Ahmad, and M.K. Mohammad. 2020. The effects of hydrogen peroxide on plant growth, mineral accumulation, as well as biological and chemical properties of *Ficus deltoidea*. *Agronomy* (Basel) 10(599):1–19, <https://doi.org/10.3390/agronomy10040599>.
- Orabi, S.A., M.G. Dawood, and S.R. Salman. 2015. Comparative study between the physiological role of hydrogen peroxide and salicylic acid in alleviating the harmful effect of low temperature on tomato plants grown under sand-ponic culture. *Sci. Agr.* 9:49–59, <https://doi.org/10.15192/pscp.sa.2015.1.9.4959>.
- Ouyang, Z., J. Tian, X. Yan, and H. Shen. 2021. Effects of different concentrations of dissolved oxygen on the growth, photosynthesis, yield and quality of greenhouse tomatoes and changes in soil microorganisms. *Agr. Water Manage.* 245:106579, <https://doi.org/10.1016/j.agwat.2020.106579>.
- Petri, B.G., R.J. Watts, A.L. Teel, S.G. Huling, and R.A. Brown. 2003. Fundamentals of ISCO using hydrogen peroxide, p. 33–88. In: R.L. Siegrist, M. Crimi, and T.J. Simpkin (eds.). *In situ chemical oxidation for groundwater remediation*. Springer, New York.
- Petrov, V.D. and F.V. Breusegem. 2012. Hydrogen peroxide—a central hub for information flow in plant cells. *AoB Plants* 2012:pls014, <https://doi.org/10.1093/aobpla/pls014>.
- Randhawa, V., M. Thakkar, and L. Wei. 2012. Applicability of hydrogen peroxide in brown tide control – culture and microcosm studies. *PLoS One* 7(10):E47844, <https://doi.org/10.1371/journal.pone.0047844>.
- Santos, A.A., D.O. Guedes, M.U.G. Barros, S. Oliveira, A.B. Pacheco, S.M. Azevedo, V.F. Magalhães, C.J. Pestana, C. Edwards, L.A. Lawton, and J. Capelo-Neto. 2021. Effect of hydrogen peroxide on natural phytoplankton and bacterioplankton in a drinking water reservoir: Mesocosm-scale study. *Water Res.* 197:117069, <https://doi.org/10.1016/j.watres.2021.117069>.
- Savvas, D. 2003. Hydroponics: A modern technology supporting the application of integrated crop management in greenhouse. *Food Agr. Environ.* 1:80–86.
- Schwarz, D. and W. Gross. 2004. Algae affecting lettuce growth in hydroponic systems. *J. Hort. Sci. Biotechnol.* 79(4):554–559, <https://doi.org/10.1080/14620316.2004.11511804>.
- Singh, H. 2017. Fertilizer and cultivar selection of different vegetable crops and evaluation of different pH buffers in hydroponics. *Oklahoma State Univ., Stillwater, MS Thesis*.
- Singh, H., B. Dunn, M. Payton, and L. Brandenberger. 2019. Fertilizer and cultivar selection of lettuce, basil, and swiss chard for hydroponic production. *HortTechnology* 29:50–56, <https://doi.org/10.21273/HORTTECH04178-18>.
- Sinha, A.K., M.A. Eggleton, and R.T. Lochmann. 2018. An environmentally friendly approach for mitigating cyanobacterial bloom and their toxins in hypereutrophic ponds: Potentiality of a newly developed granular hydrogen peroxide-based compound. *Sci. Total Environ.* 637:524–537, <https://doi.org/10.1016/j.scitotenv.2018.05.023>.
- Slesak, I., M. Libik, B. Karpinska, S. Karpinski, and Z. Misalski. 2007. The role of hydrogen peroxide in regulation of plant metabolism and cellular signaling in response to environmental stresses. *Acta Biochim. Pol.* 54:39–50, https://doi.org/10.18388/abp.2007_3267.
- Soffer, H., D.W. Burger, and J.H. Lieth. 1990. Plant growth and development of *Chrysanthemum* and *Ficus* in aero-hydroponics: Response to low dissolved oxygen concentrations. *Scientia Hort.* 45:287–294, [https://doi.org/10.1016/0304-4238\(91\)90074-9](https://doi.org/10.1016/0304-4238(91)90074-9).
- Supraja, K.V., B. Bunushree, and P. Balasubramanian. 2020. Performance evaluation of hydroponic system for co-cultivation of microalgae and tomato plant. *J. Clean. Prod.* 2722:122823, <https://doi.org/10.1016/j.jclepro.2020.122823>.
- Tesoriero, L., S. Jelinek, and L. Forsyth. 2010. On farm hygiene and sanitation for greenhouse horticulture. *PRIMEFACT* 1-5.
- Uchida, A., A.T. Jagendorf, T. Hibino, T. Takabe, and T. Takabe. 2002. Effects of hydrogen peroxide and nitric oxide on both salt and heat stress tolerance in rice. *Plant Sci.* 163:515–523, [https://doi.org/10.1016/S0168-9452\(02\)00159-0](https://doi.org/10.1016/S0168-9452(02)00159-0).
- Vänninen, I. and H. Koskula. 1998. Effect of hydrogen peroxide on algal growth, cucumber seedlings and the reproduction of shore flies (*Scatella stagnalis*) in rockwool. *Crop Prot.* 17:547–553, [https://doi.org/10.1016/S0261-2194\(98\)00060-X](https://doi.org/10.1016/S0261-2194(98)00060-X).
- Wang, B., S. Qingyang, L. Jijian, S. Gao, M. Wujuan, and B. Yonghong. 2019. Optimization method for *Microcystis* bloom mitigation by hydrogen peroxide and its stimulative effects on growth of chlorophytes. *Chemosphere* 228: 503–512, <https://doi.org/10.1016/j.chemosphere.2019.04.138>.
- White, P.J. and M.R. Broadley. 2003. Calcium in plants. *Ann. Bot.* 92:487–511, <https://doi.org/10.1093/aob/mcg164>.
- Wojtyla, L., K. Lechowaska, K. Szymon, and G. Małgorzata. 2016. Different modes of hydrogen peroxide action during seed germination. *Front. Plant Sci.* 7:66, <https://doi.org/10.3389/fpls.2016.00066>.
- Xia, X., C. Gao, L. Song, Y. Zhou, K. Shi, and J. Yu. 2014. Role of H₂O₂ dynamics in brassinosteroid-induced stomatal closure and opening in *Solanum lycopersicum*. *Plant Cell Environ.* 37(9): 2036–2050, <https://doi.org/10.1111/pce.12275>.