Review

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Review

Review and Prospects on Ultrafine Bubble (UFB) Water for Managing Crop Stresses in Plant Protection Practices

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Abstract

Plants are subjected to long-term biotic stresses and abiotic stresses which cause significant changes in complex crop ecosystems and have been often accompanied by the struggle against plant pests. Pests control methods relying heavily on chemical pesticides have resulted in numerous adverse effects. One of innovative methods involves using ultrafine bubble (UFB) waters may realize the pesticide reduction action for the plant pest control. The classification and six properties of UFBs were summarized, and the generation approaches of UFBs were introduced based on physical and chemical methods. The applications of UFBs and ozone UFB waters in plant protection practices were comprehensively reviewed, in which, UFB waters against the plant pest insects and the soilborne, airborne and waterborne diseases were analyzed, and the abiotic stresses of crops in salinity soil and contaminated soil were reviewed. Because UFB water is not omnipotent, several prospects were proposed aiming at pesticide reduction and replacement, for example, the mechanism of UFB water controlling plant pests and diseases, the molecular mechanism of UFB water affecting plant pest resistance, the plant growth in harsh polluted environments, the UFB behavior with hydrophobic and hydrophilic surfaces of crops and the building of integrated intelligent crop growth system.

Keywords: UFB; OUFBW; properties and generation; plant protection practices; biotic and abiotic stresses of crops; review and prospects

1. Introduction

During the growth process, plants are subjected to long-term biotic stresses of diseases, insects, weeds and so on, as well as abiotic stresses of droughts, floods, frost and freezing, and soil salinization, which differ in intensity, duration and severity and cause significant changes in the structure and function of complex crop ecosystems and often accompany secondary disasters. Over thousands of years, the history of human survival and development has been accompanied by the struggle against plant pests caused by the biotic stresses and abiotic stresses [1,2]. As FAO (Food and Agriculture Organization of the United Nations) mentions, global agriculture faces mounting challenges from pests that threaten food security, biodiversity, and farmers' livelihoods. Pest control methods relying heavily on chemical pesticides have resulted in numerous adverse effects, including soil degradation, water contamination, resistance development of pests and environmental degradation, serious health risks to humans and animals. FAO organized an event focused on a critical challenge: how to scale up biodiversity-friendly pest control practices as effective alternatives to highly hazardous pesticides which highlighted the importance of transitioning away from toxic chemical solutions in agriculture toward safer, sustainable and biodiversity-enhancing approaches like integrated pest management and biological control [3]. Therefore, it is necessary to

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carry out research on pesticide replacement and reduction action of plant protection methods and pesticide application equipment.

Reducing pesticide application has become a global issue for environmental sustainability and human health. As the demand for safer and more sustainable crop production grows, advanced and sustainable plant protection methods are emerging that could benefit the way we cultivate crops. One such innovative method involves the use of ultrafine bubble (UFB) water, a unique type of water that has shown promising results in enhancing crop growth and resilience which offers the potential alternative to reduce the reliance on chemical pesticides, improving both crop yield and quality in a natural way [4].

Nanotechnology can address most of the plant pest concerns and bring revolutionary changes in agricultural plant protection practices. Nano-pesticides are an emerging technological advance bringing a new possibility for managing plant pests and pathogens [5]. Nanobubbles are cavities filled with gas and exist in liquids. Ultrafine bubble water (UFW) offers sustainable practices by promoting photosynthesis, improving oxygenation, disrupting growth of harmful pests, and strengthening plant defensive abilities [6–8]. UFWs could be easily integrated into existing irrigation and pesticide spraying systems, providing farmers with a practical solution to reduce plant pest outbreaks, improve crop yields and quality while minimizing environmental impact and realizing sustainable development. Research data showed that UFWs have significant effects on sterilization and pest control. The application rate of chemical pesticides in vegetable greenhouses using UFB technology could be reduced by about 70%. This not only reduces the potential risk of pesticide residues on human health, but also reduces the environmental pollution caused by agricultural planting, which is conducive to achieving sustainable development [9][10]. Research showed that ozone ultrafine bubble water (OUFBW) spraying can effectively control various bacterial plant diseases, effectively prevent and control common diseases such as crop powdery mildew and gray mold, improve crop disease resistance and reduce crop incidence [11]. Obviously, the use of OUFBW for plant protection can reduce the application of chemical pesticides and promote the development of green agriculture.

A few crop protection practices of UFWs in agriculture applications have been conducted around the world [12] [13] [14,15], but no comprehensive reviewing article introduces the UFWs in crop protection practices. The objectives of this paper were to review the UFW properties and generation approaches, mainly introduce the research on UFB waters applied in crop protection practices even though the research is in an infant stage. Finally, several research prospects were proposed aiming at reducing pesticide consumption and increasing pesticide application efficiency from the source.

2. Properties and Generation of UFW

The generation and property analysis of UFWs is an important part of the practices to realize pollution-free and residue-free plant protection using the UFWs as broad-spectrum pesticides.

2.1. Bubble Classification and Definition

Generally, the gas aggregating and remaining in a medium enclosed by an interface is called a bubble. When the gas is affected by shear force in the liquid, bubbles of different sizes and shapes will be formed. International Standard Organization (ISO) defines the bubbles with a volume equivalent diameter of less than 100 μm as fine bubble, the fine bubble with a volume equivalent diameter in the range from equal or greater than 1 μm to less than 100 μm as microbubble, and the fine bubble with a volume equivalent diameter of less than 1 μm as ultrafine bubble [16]. However, the bubble classification name and size range recommended by different literature sources are not very strict and not quite consistent. Through analyzing the classification names and definition of ISO and the size ranges recommended by relevant literatures, we summarize the bubbles with the classification, typical names and size ranges of different bubbles shown in Table 1.

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According to the order from large to small, the bubbles are divided into macrobubble (MaB), microbubble (MiB) and micro-nano bubble (MNB) [16–18]. The macrobubble (MaB) is further divided into centimeter bubbles (CMB) with diameters of >10 mm and millimeter bubbles (MMB) with diameters ranging from 100 μm to 10mm. Microbubbles (MiB) typically refer to gaseous cavities in aqueous medium with diameters ranging from 1 μm to 100 μm, in which bubbles with diameters ranging from 1 μm to 10 μm are called sub-microbubbles (SMB). The micro-nano bubbles (MNB) refer to bubbles with diameters ranging from hundreds of nanometers to about 10 μm when the bubble occurs. The sub-microbubbles (SMB) with diameters ranging from 1 μm to 10 μm can be included into micro-nano bubbles (MNB) either. The ultrafine bubbles (UFB) with diameters of less than 1 μm are also included into the micro-nano bubbles (MNB), which is the focus of this paper. Among them, ultrafine bubbles with diameters below 200 nm are classified as nanobubbles (NB). The typical corresponding reference objects for different size bubbles are as follows: CMB corresponds to grape, MMB corresponds to most raindrops, MB corresponds to ordinary hair, SMB corresponds to erythrocyte, UFB corresponds to cigarette smoke, NB corresponds to viruses.

Table 1. The classification, typical names and size ranges of different bubbles.

Classification	**Typical Names**	**Size ranges**	**Typical objects** [1]
Macro-bubbles (MaB)* (>100μm)	Centimeter Bubble (CMB)	>10mm	Grape
	Millimeter Bubble (MMB)	100μm-10mm	Most raindrops
Microbubbles (MiB) (1 μm-100 μm)	Micron bubbles (MB)	<100μm	Ordinary hair
	Sub-microbubbles (SMB)	1-10μm	Erythrocyte
Micro-nano bubbles (MNB) (<10 μm)	Ultrafine bubbles (UFB)	<1μm	Cigarette smoke
	Nano bubbles (NB)	<200nm	Viruses

[1] Typical corresponding reference objects.

2.2. Ultrafine Bubble Water (UFW) Properties

The UFBs endow them with unique physicochemical characteristics that the conventional bubble does not have due to their small micro-nano-scale sizes, including large specific surface area, slow rising velocity in water, easy self-pressurization and high gas dissolution rate, strong mass transfer efficiency, strong interfacial zeta potential, generating much of free hydroxyl radicals. The special physicochemical properties of UFB waters are summarized in Table 2 [7,17–26]. Figure 1 shows the fundamental physiochemical properties of nanobubbles [17].

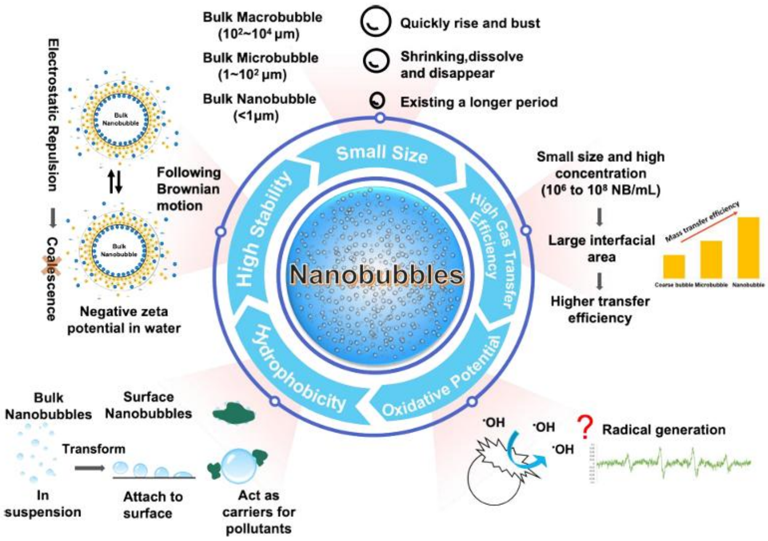

Figure 1. Fundamental physiochemical properties of nanobubbles [17].

Table 2. The physicochemical properties of UFB waters.

	Physicochemical properties *	Phenomenon	Theoretical foundation	References
1	Large specific surface area	The large specific surface area of a UFB leads to high surface energy, making the bubble a natural tendency to coalesce or dissolve to reduce the surface energy.	The specific surface area of a bubble refers to the ratio of its total surface area to the volume of the gas it contains.	[18]
2	Slow rising velocity in water	The UFBs can remain suspended in water for extended periods and have a relatively long residence time in the water.	Using Stokes' law, where the rising velocity of a bubble is directly proportional to its size and inversely proportional to the viscosity of the surrounding liquid.	[18,25]
3	Easy self-pressurization and high gas dissolution rate	During the self-pressurization process, the gas inside the bubble continuously dissolves into the liquid resulting in the shrinkage and disappearance of the nanobubbles.	Based on the Young–Laplace equation, the pressure is directly proportional to the gas-liquid interface tension and inversely proportional to the bubble diameter.	[19,24,26]
4	Strong mass transfer efficiency	The mass transfer efficiency of UFBs is significantly higher than that of conventional bubbles to make the UFBs spread over a larger region and reach confined spaces more easily.	Based on mass transfer coefficient formula and mass transfer flux formula, the larger contact surface area, lower surface tension, massive quantities and long-term interaction with the liquid result in higher mass transfer efficiency.	[7,18,27]
5	Strong interfacial	When the Zeta potential is high, the electrostatic	According to the electrostatic laws and Poisson-Boltzmann	[7,18,24,28]

	Zeta potential	repulsion between the UFBs is strong, which can prevent the bubbles from approaching and coalescing, thereby improving the stability of the UFB water.	equation, bubbles with the charge interfaces generate an electrical field that preferentially attracts the opposite charge ions distributed in solutions.	
6	Generating hydroxyl radicals with strong oxidation	Hydroxyl radicals can oxidize the surface-active substances on the surface of the UFBs, reducing the surface activity and stability of the UFBs.	Oxidation-reduction potential measures the ability of an aqueous solution to oxidize or reduce another substance, and it changes linearly with the logarithmic change in the O_2 concentration.	[20,29]

* All the physicochemical properties of UFBs are related to the small size of UFBs.

2.2.1. Large Specific Surface Area [18]

The specific surface area of a bubble refers to the ratio of its total surface area to the volume of the gas it contains, which describes the dispersion degree of the bubble in the liquid and the size of the bubble. Under the same bubble volume, the specific surface area is inversely proportional to the bubble diameter. This large specific surface area-to-volume ratio of a UFB leads to high surface energy, causing the bubbles to have a natural tendency to coalesce or dissolve to reduce the surface energy. When there are substances in the solution that can adsorb on the surface of the nanobubbles, they can reduce the surface tension and increase the stability of the nanobubbles.

2.2.2. Slow Rising Velocity in Water [18,25]

The bubble shape and rising velocity can be determined based on fluid mechanics. The rising velocity is a function of bubble diameter. When the bubbles are too small, the inertial force is much smaller than the surface tension or the viscous force and thus the bubbles are spherical. The small size of UFBs keeps Brownian motion in liquid with negligible buoyant force and slows down their rising velocity, making it difficult for them to quickly float to the surface because the rising velocity of UFBs can be expressed using Stokes' law, where the rising velocity of the bubble is directly proportional to its size and inversely proportional to the viscosity of the surrounding liquid. Therefore, the UFBs can remain suspended in water for extended periods and have a relatively long residence time in the water. For example, the rising velocity of a nanobubble with a diameter of 100 nm is 2.7 nm/s, indicating a longer residence time and slower rising velocity.

2.2.3. Easy Self-Pressurization and High Gas Dissolution Rate [19,26]

The gas-dissolving capability (solubility of gas) is affected by factors such as temperature, pressure and bubble size. There is interfacial tension at the gas-liquid interface, which compresses the gas in the bubble to dissolve the gas into liquid. When the bubble size decreases, the tensile stress experienced by the liquid increases. Based on the Young–Laplace equation, the pressure is directly proportional to the gas-liquid interface tension and inversely proportional to the bubble diameter. The internal pressure of UFBs increases sharply with the decrease in bubble size, making it difficult for UFBs to remain stable under normal pressure. The self-pressurization and dissolution characteristic of UFBs refers to the process where the bubble wall gradually thins and eventually dissolves due to the internal pressure being higher than the external environmental pressure. Because the self-pressurization process breaks the assumption of supersaturated gas dissolution conditions, the gas inside the bubble continuously dissolves into the water, increasing the solubility of gas in the liquid and resulting in the gradual shrinkage and disappearance of the nanobubbles.

2.2.4. Strong Mass Transfer Efficiency [7,18]

The mass transfer efficiency of a bubble refers to its ability to facilitate the transport of substances in the liquid. The solubility of gas in liquid has a significant impact on the stability of UFBs. If the solubility is low, the gas in the UFBs is less likely to dissolve, which is beneficial to the stability of the UFBs. When the shrinkage of the UFB reaches a certain limit value, the internal pressure of the bubble will tend to be infinite. This self-pressurization effect will make the UFB dissolve in liquid or break away at the liquid surface, so that the gas solubility in liquid reaches a supersaturated state, and better gas-liquid mass transfer efficiency is achieved. Due to their larger contact surface area and long-term interaction with the liquid, the mass transfer efficiency of UFBs is significantly higher than that of conventional bubbles. Moreover, the lower surface tension of UFBs promotes their close contact and fusion with the liquid, shortening the mass transfer distance and thereby enhancing the mass transfer efficiency. UFBs in massive quantities are also expected to spread over a larger region and reach confined spaces more easily.

2.2.5. Strong Interfacial Zeta Potential [7,18,24,28]

The Zeta (ζ) potential reflects the surface charge state of bubbles in a dispersed system. It represents the potential difference at the sliding interface between the bubble and the surrounding solution and is affected by the solution pH, ionic strength, electrolyte type, and surface properties. Pure aqueous solution is composed of water molecules and a small amount of ionization generated H+ and OH-. According to the electrostatic laws and Poisson-Boltzmann equation, bubbles with the charge interfaces generate an electrical field that preferentially attracts the opposite charge ions distributed in solutions. Generally, cations are easier to leave the gas-liquid interface than anions, making the interface negatively charged with a negative zeta potential value. When UFBs shrink, charged ions are rapidly concentrated and enriched, making the interface ζ potential significantly increased. When the Zeta potential is high, the electrostatic repulsion between the UFBs is strong, which can prevent the bubbles from approaching and coalescing, thereby improving the stability of the UFB water. But if the Zeta potential is low, the electrostatic repulsion is weak, and the UFBs are more likely to aggregate and collapse.

2.2.6. Generating Hydroxyl Radicals with Strong Oxidation [20,29]

Oxidation-reduction potential measures the ability of an aqueous solution to oxidize or reduce another substance, and it changes linearly with the logarithmic change in the O_2 concentration. At the moment of UFB explosion, the high concentration ions on the interface will release the accumulated chemical energy instantly and stimulate the generation of a large amount of hydroxyl radicals with ultra-high oxidation-reduction potential and strong oxidation. UFBs rich in O_2 molecules favor the generation of hydroxyl radicals compared to gases without O_2. Hydroxyl radicals are highly reactive species. They can react with the substances on the surface of the UFBs or in the surrounding solution, changing the surface properties of the UFBs or the composition of the solution, and thus affecting the stability of the UFBs. For example, hydroxyl radicals can oxidize the surface-active substances on the surface of the UFBs, reducing the surface activity and stability of the UFBs.

These properties endow UFBs with great potential and broad application prospects in various fields, including water treatment, enhanced oil recovery, mineral flotation, medical drug delivery and health care, and agriculture production. UFB technology is highly efficient, energy-saving, and environmentally friendly. In agriculture production, UFB can promote crop nutrient absorption, crop quality, and yield enhancement. These unique physicochemical characteristics of UFB waters allow them to penetrate biofilms, oxidize harmful pathogens, and enhance plant resilience. UFBs can be infused with oxygen, ozone, or other gases to target specific agricultural challenges [29,30].

2.3. *Generation of UFB Water and Ozone UFB Water*

According to the different distribution states, UFBs can be divided into surface UFBs and bulk UFBs. Surface UFBs are non-spherical interface bubbles, which are located on the liquid and solid interface. Bulk UFBs are spherical bubbles suspended in liquid. Besides the phase transition process of forming gas phase through the crystal nucleus in metastable liquid phase, the formation of bulk UFBs is usually considered as a static and/or quasi-static to dynamic processes through coalescence or break-up. The bubble coalescence refers to the aggregation of small bubbles in the liquid to form large bubbles, and the bubble break-up refers to the micro pore shrinkage and rupture of large bubbles to form small bubbles. The generation of bulk UFB water (UFW) and ozone UFB water (OUFBW) are reviewed in this paper.

2.3.1. UFB Water Generation

There are several ways to divide the generation approaches which can produce UFBs under certain conditions [7,29–32]. Generally, UFBs can mainly be generated through both physical and chemical methods, depending on the formation mechanism, as shown in Table 3. The physical generation approaches of UFBs only change the state or form of materials, without producing new substances, including cavitation (either using hydrodynamic cavitation or acoustic cavitation), gas dispersion（including mechanical agitation，micro-porous dispersion and microfluidic device）, solvent exchange, temperature alteration, electrohydrodynamic effect, and pressurized gas dissolution. While the chemical generation approaches of UFBs are accompanied by generating new substances, including electrolysis and photocatalysis technology.

Table 3. Methods of generating UFBs.

Methods	Generation approaches		Mechanism	References
Physical	Cavitation	Hydrodynamic	Using a localized low-pressure region to draw in gases and form UFBs	[31]
		Acoustic	Using ultrasonic waves to cause gas nuclei in liquid for generating UFBs	[31]
	Gas dispersion	Mechanical agitation	Using a rotating disc device to stir gas–liquid mixture in high speed	[33–35]
		Microporous structure	Applying micro-porous structures to disperse gas into UFBs when gas passes porous pipe	[30,31]
		Microfluidic device	Narrower main channel width and increased shear gradient reduce bubble size	[31]
	Solvent exchange		Replacing one fluid with high gas solubility by another fluid with low gas solubility	[31,32]
	Temperature alteration		Altering temperature suddenly to provide sufficient energy forming bubble nucleus	[31]
	Electrohydrodynamic effect		Weakening gas–liquid interface tension and leading to breakup of gas phase	[31]
	Pressurized gas dissolution		Utilizing changes in gas–liquid pressure to dissolve and release gases	[18,30]

Chemical	Electrolysis	Dissolving hydrogen in water through electrochemical reactions on electrode surface	[31,36]
	Photocatalysis technology	Catalyzing decomposition of hydrogen peroxide solution to produce UFBs	[37]

(1) Physical methods

Cavitation approaches

Cavitation is known as the creation of new gas/liquid interfaces and generates localized low-pressure areas in the liquid, usually occurs after an energy input causes a local pressure fluctuation (including acoustic cavitation) or an increase in flow velocity from hydraulic devices (including hydrodynamic cavitation), which causes the local pressure to drop below the vapor pressure [18,31].

Hydraulic cavitation uses the localized low-pressure region generated in high-speed flowing or strongly turbulent liquids to draw in gases and form UFBs, such as venturi tube devices [38], jet-type bubble generators [39], and swirl-type bubble generators [40], as shown in Figure 2 (a) (b) and (c).

Acoustic cavitation uses the cavitation effect produced by ultrasonic waves propagating in the liquid to break the gas in the liquid and form UFBs. The ultrasonic generator uses ultrasonic waves to create alternating positive and negative pressure zones in the liquid, causing gas nuclei to expand into cavitation bubbles in the negative pressure zone and then collapse in the positive pressure zone to generate UFBs. As shown in Figure 2(d), an additional hollow ultrasonic horn attached to the tip of a standard hollow ultrasonic horn which are ultrasonic amplifiers with internal channels for gas supply, and the ultrasonic oscillation is amplified as the cross-sectional area decreases. The gas is supplied from the horn tip inserted into the liquid and ultrasonic oscillation is simultaneously applied to disturb the gas-liquid interface and to form surface waves in the test section. UFBs are generated by the separation of the gas phase from these surface waves [18,41,42].

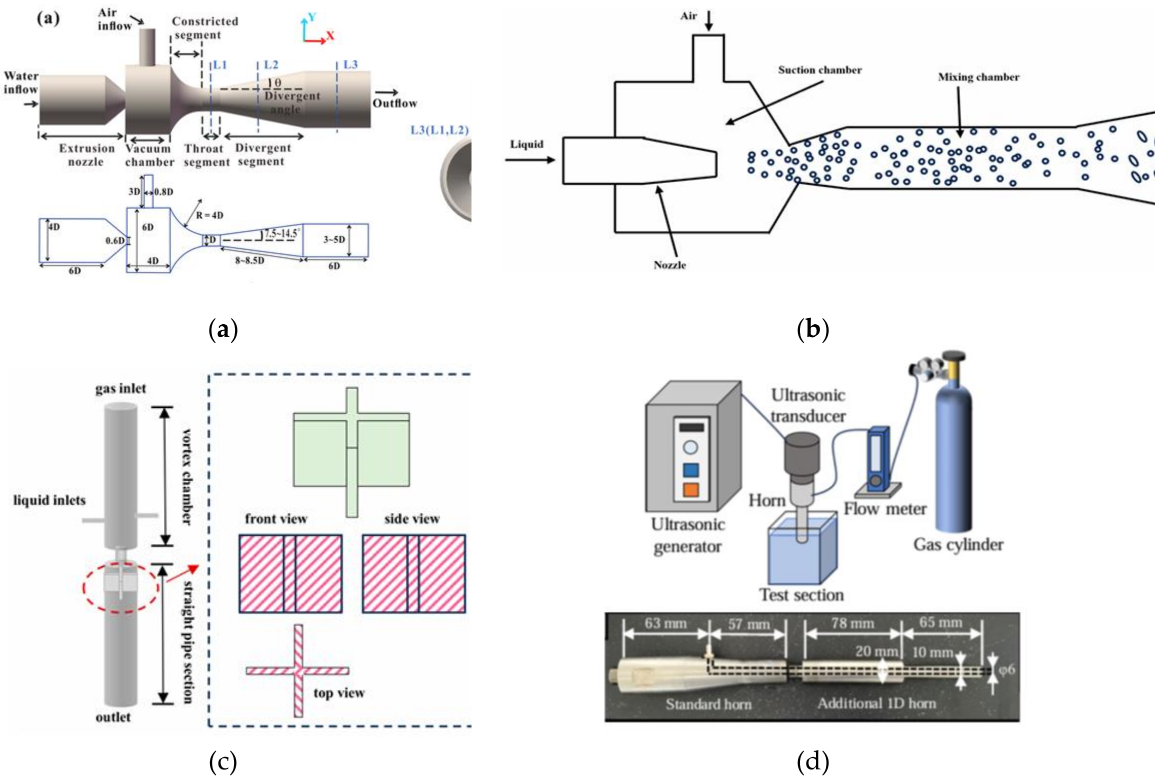

Figure 2. Schematic diagram of UFB generation principle of cavitation approaches: (**a**) Venturi tube nozzle-type bubble generator; (**b**) Jet-type micro-nano bubble generator; (**c**) Swirl-type micro-nano bubble generator with baffles; (**d**) Hollow Ultrasonic Horn.

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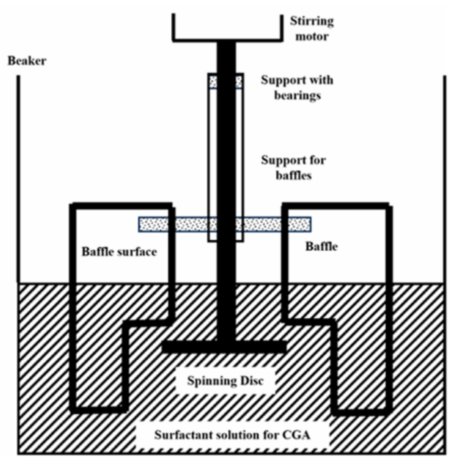
Gas dispersion approaches

In generating UFBs through gas dispersion, there is no sharp pressure drop during gas dispersion, but using mechanical agitation (mechanical shearing), microporous structure (membrane filtration) and microfluidic devices etc., to break the gas phase into smaller size, forming UFBs [31].

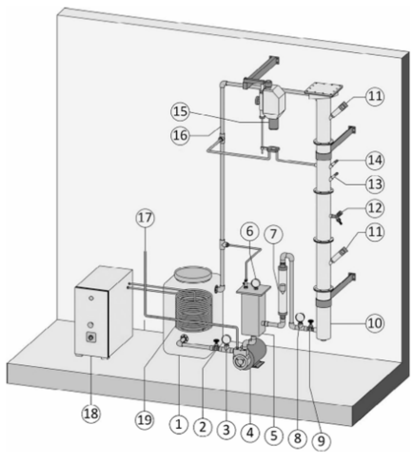
The mechanical agitation approach is a simple and easy approach for generating UFBs, featuring a straightforward apparatus and low manufacturing. Mechanical shearing methods generate bubbles by shearing the gas with mechanical force. A typical device is the mechanical stirring type [18]. The early mechanical stirring method used a rotating disc device to prepare microbubbles through high-speed shearing, as shown in Figure 3 (a) [34]. Then a centrifugal multiphase pump impellers and a recycle column were introduced to generate the nanobubbles (150–200 nm) aqueous solutions, coupling with hydrodynamic cavitation effects, and at various operating pressures and air/liquid surface tension throughout several bubble generation cycles, as shown in Figure 3 (b) [35].

The micro-porous structures were applied to disperse the gas into UFBs when gas passes through the porous pipe and stably exists in water, as shown in Figure 3 (c)-(e) [18,43]. In Figure 3(d), a mechanical high-speed mixing device was introduced to UFB preparation. An air inlet connects with high-pressure gas cylinder that the compressed gas is injected into the UFB generator through the cylinder to introduce different gas sources to regulate the atmosphere environment. A propeller is provided with ultra-high rotation speed by high-speed motor, and the generated bubbles are discharged from an air outlet, and the nano bubble nozzle of the generator is immersed in liquid in the bubble containing vessel. The microporous membrane of the generator is composed of 50 microporous filaments in the nozzle outlet [33].

The microfluidic technology for generating UFBs relies on precise hydrodynamic control within microscale channels, utilizing shear forces, surface tension, and gas–liquid interfacial interactions to fragment and stabilize gas into homogeneous micro-nanoscale bubbles [18]. The bulk nanobubbles were generated in the glycine solution using porous membrane. The membrane was dipped, and the gas was sparged into the glycine solution. The bulk nanobubble solution in pure water was characterized by nanoparticle tracking analysis and dynamic light scattering analysis in terms of bubble size, concentration, and zeta potential, respectively[44]. Figure 3(f) is the schematic representation of the three main microfluidic geometries used for the formation of droplets and bubbles, the core device architectures include T-junction, flow-focusing, and co-flow configurations—enable precise bubble size control through regulation of gas/liquid flow ratios, microchannel geometry, and fluid properties. The dispersed phase is injected (a) in a cross-flowing stream through a T-shaped junction, (b) in a co-flowing stream, and (c) in a focused stream imposed by the continuous phase [45].



(**a**)



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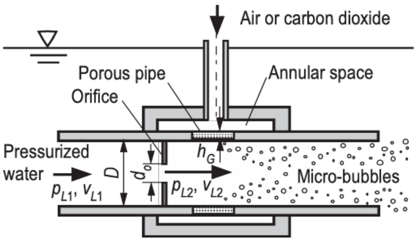

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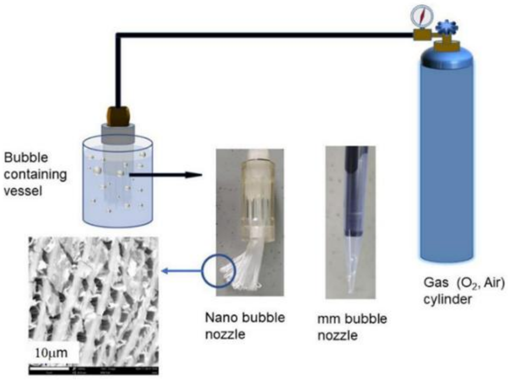

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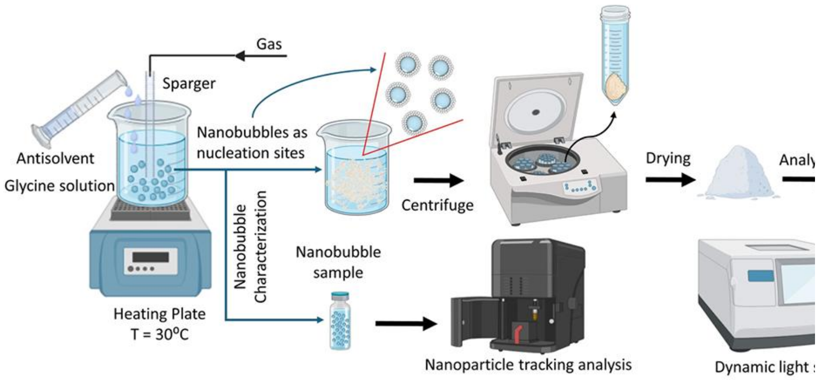

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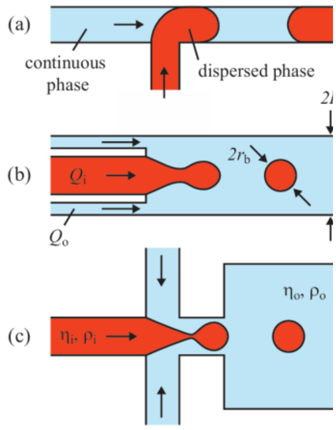

(f)

Figure 3. Schematic diagram of UFB generation principle of gas dispersion approaches: (**a**) Spinning-disc MBD generator; (**b**) Generator with multiphase pump: (1) Feed water tank; (2) Ball valve; (3) Vacuum meter; (4) Centrifugal multiphase pump; (5) Pressure tank; (6) Pressure gauge; (7) Flowmeter; (8) Pressure gauge; (9) Needle valve; (10) Column; (11) Pressure sensors; (12) Sampler; (13) Temperature sensor; (14) pH meter; (15) LTM B Sizer; (16) Recycling hose; (17) Atmospheric air; (18) Cooler; and (19) Heat exchanger; (**c**) Multi-fluid mixer with orifice and porous tube[18,43]; (**d**) Nano bubble nozzle with microporous filaments [33]; (**e**) nanobubbles generated using porous membrane as active nucleation sites [44]; (**f**) Three main microfluidic geometries: (a) in a cross-flowing stream through a T-shaped junction, (b) in a co-flowing stream, and (c) in a focused stream imposed by the continuous phase.

Solvent exchange approach

The solvent exchange approach requires two mutually miscible fluids with different gas solubility. When one fluid with high gas solubility is replaced by another fluid with low gas solubility, excess gas will be released to form UFBs which shows great potential in preparing microscopic, functional materials. As shown in Figure 4, firstly the ethanol was injected into liquid cell by a glass syringe. Then pure water was slowly added to the liquid cell which had been filled with ethanol using a glass syringe. Injecting rate was controlled by a syringe pump. During the ethanol-water exchange to produce interfacial nanobubbles, surface nanobubbles could be pinned on the substrate [31,46,47].

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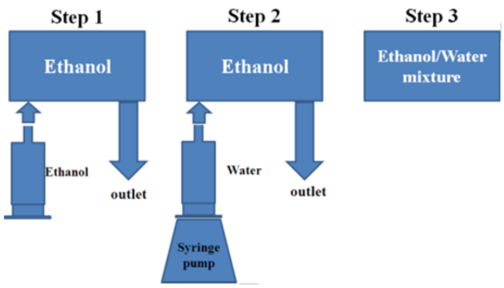

Figure 4. Schematic diagram of ethanol-water exchange.

Temperature alteration approach

The gas solubility is easily saturated in a liquid at a lower temperature. Sudden alteration in temperature offsets the solubility equilibrium and results in nucleus formation of gas bubbles with creation of cavities in the liquid. Temperature alteration approach provides sufficient energy for developing new gas–liquid interface. As shown in Figure 5, the generation of air bulk nanobubbles and oxygen bulk nanobubbles in Jet A-1 was performed. Instantaneous mixing of liquids at different temperatures results in an excessive quantity of gas in the mixture relative to its equilibrium value at a specific temperature and pressure. Bulk nanobubbles were generated in Jet A-1 (commercially used aviation fuel) by the hot and cold solvent mixing method [31,48].

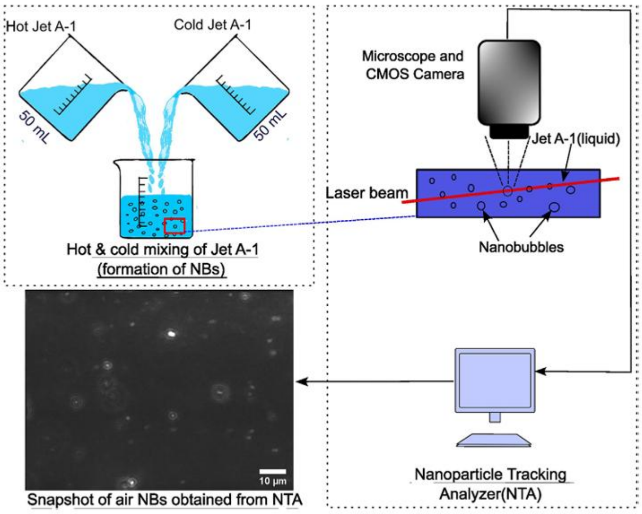

Figure 5. Schematic diagram of hot & cold mixing in Jet A-1.

Electrohydrodynamic effect approach

An electric field can weaken the surface tension at the gas–liquid interface. The electrohydrodynamic (EHD) effect facilitates the breakup of the dispersed phase (gas phase), leading to the formation of UFBs [31]. As shown in Figure 6, the syringe pump is used to supply gas and control the gas flow rate in the metal capillary precisely. A copper ring is set at a height of 20 mm from the capillary orifice. The high-voltage direct current power supply is applied between the capillary electrode and ring electrode to create a non-uniform electric field. The interactions between bubbles under the electric field are usually featured by the applied field strength and the superficial gas velocity. The force balance ratios are the electric force to the liquid surface tension and gas inertial force to the liquid surface tension. The electric field strength induces a change in the coalescence regime. The volume electric force acting on a fluid is composed of different types of physical mechanisms, the force acting on a charged fluid element and the gradient of the field. The jump of the dielectric constant across the gas–liquid interface is prominent. The negative electrical free charges generated at the bubble bottom significantly exceed the positive electrical free charges

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generated at the bubble top. The resultant Coulomb force acting on the bubble surface is opposite to the electric field direction which can drive the liquid motion, forming the classical EHD flow, which accelerates the evolution of bubbles in the streamline direction, the bubble will be further accelerated by the dielectrophoresis force [49].

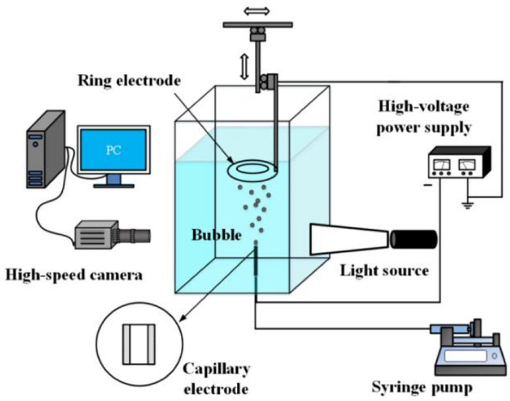

Figure 6. Schematic diagram of liquid–gas electrostatic dispersion system.

Pressurized gas dissolution approach

Pressurized gas dissolution methods generate UFBs by utilizing changes in gas–liquid pressure to dissolve and release gases. Figure 7 shows the operational principle of the self-suction MNB generator which dissolves gas in a pressurized dissolved gas tank and then releases the gas through a depressurization device (throttling nozzle), causing the gas to dissolve in the water as a result of the increased flow velocity and sudden decrease in pressure, forming the cluster MNBs [18,30,50].

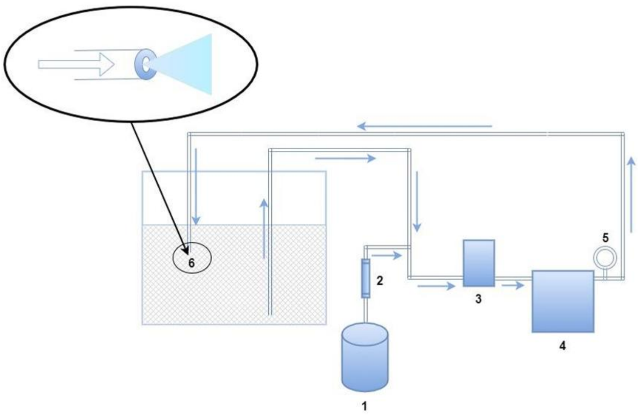

Figure 7. Schematic diagram of self-suction MNB generator: (1) gas supply (CO_2 cylinders), (2) gas flow meter, (3) diaphragm pump, (4) dissolved gas tank, (5) hydraulic pressure gauge, (6) throttling nozzle.

(2) Chemical methods

Photocatalysis technology

Photocatalysis technology refers to that when a certain wavelength of light is irradiated on the photocatalysis material, the electrons in the photocatalysis material will undergo a transition, and the electrons will precipitate from the surface of the material as the conditions required by thermodynamics to release gas. Nanoscale motors are ubiquitous in biology and operate by enzymatic catalysis of spontaneous reactions. Three-striped (Au/Pt/Au) rods, which the dimensions of the rods and their speeds are similar to those of multiflagellar bacteria, were successfully used to catalyze the decomposition of hydrogen peroxide solution to produce H_2 and O_2 nanobubbles through Pt electrode [37].

Electrochemical (electrolysis) approach.

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Electrochemical approach generates UFBs via gas production through electrochemical reactions on the electrode surface. Water electrolysis is a convenient means to dissolve hydrogen in the water because hydrogen is supersaturated in the vicinity of the electrode. As shown in Figure 8, electrolysis applies voltage to the electrode, and water molecules are electrolyzed to produce H_2 and O_2 gases. These gases nucleate on the electrode surface and gradually detach to form fine bubbles which the bubble size can be precisely controlled by adjusting electrolysis parameters [18,36].

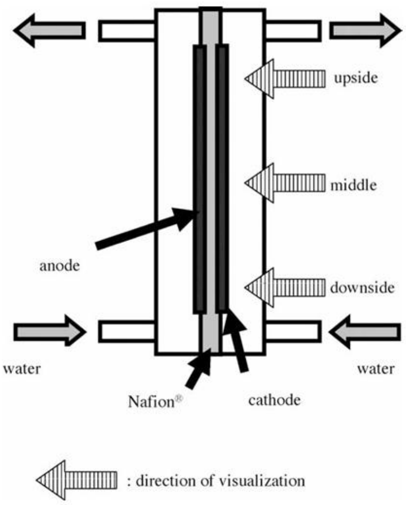

Figure 8. Schematic diagram of microbubble generator by electrolysis.

2.3.2. Ozone UFB Water Generation

Ozone is a powerful oxidizing agent that is applied in aqueous form for sanitation and is an effective alternative to chemical pesticides for soil treatment and pest control[51] [52,53]. Combining ozone with other materials such as water-mist produces a very reactive intermediate, hydroxyl radicals(•OH) which are stronger oxidizing agents than ozone itself. Ozone ultrafine bubble water (OUFBW) exhibits bactericidal activity against pathogenic bacteria. Ozone micro-bubbles were generated by a ceramic ultrafiltration membrane to enhance efficient ozone gas–liquid mass transfer [54,55].

Figure 9 is the schematic diagram of an ozone UFB water generator. OUFBW was generated by micro blender and re-circulated in the polyvinyl chloride water tank. Generation of UFBs needed more than 0.2 MPa of pump pressure, a cooling tank was assembled to keep water temperature 10˚C or less, which generates OUFBW containing high concentration (4–6 ppm) of ozone [56].

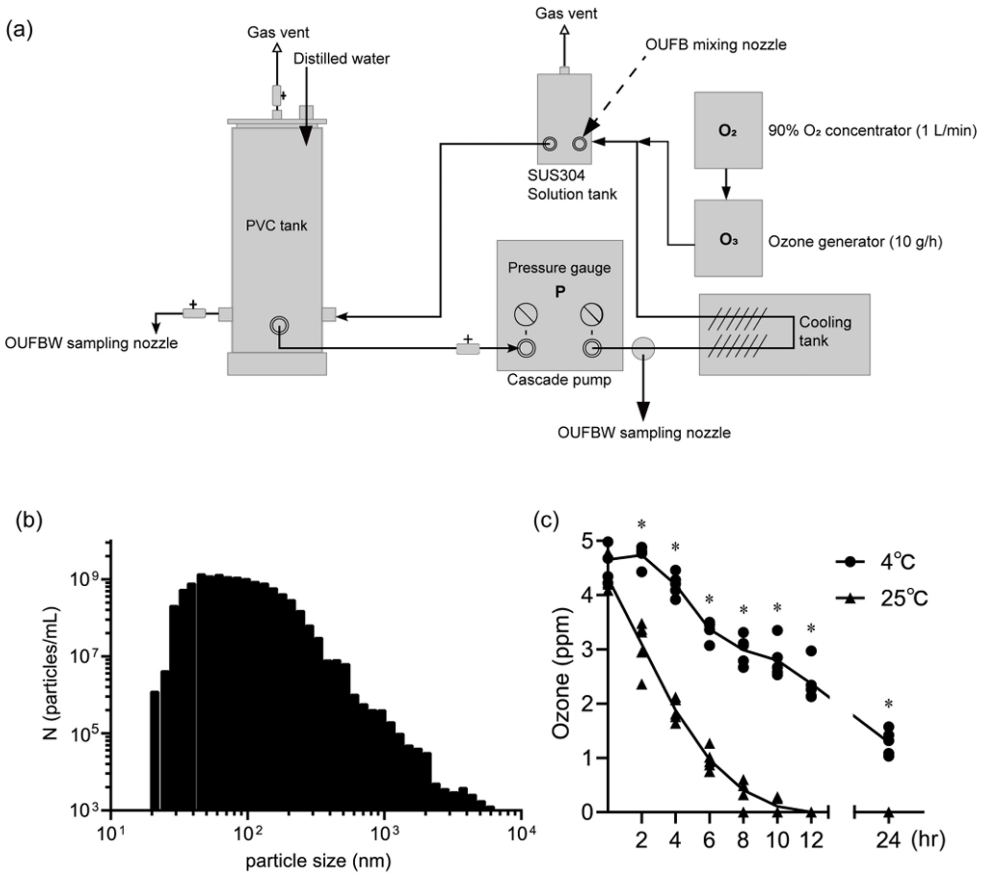

Figure 9. Generation of ozone UFB water (OUFBW). (a) Schematic diagram of OUFBW generator, (b) Particle size distribution of OUFBWs, (c) Changes of ozone concentration in OUFBW stored at 4°C or 25°C.

3. Controlling Biotic Stresses of Crop Pests using UFW

Traditional pest and disease management relies on synthetic chemicals that can harm beneficial insects, soil health, and water sources. Nanobubble technology provides an eco-friendly alternative by enhancing natural biological control mechanisms. This reduces the need for pesticides while maintaining crop protection [10].

3.1. Plant pest Insects Control Using UFW

There have been a few documented cases of about the effect of UFW on agricultural insect pests.

Melon farming has traditionally relied heavily on pesticides to manage pests such as aphids, thrips, and whiteflies, which can severely damage crops and reduce yield. However, the excessive use of pesticides poses significant environmental and health risks. To investigate whether it was H_2 or O_2 that helped to deter insect infestation and improve melon yield and quality, as shown in Figure 10, the melon seedlings were planted in a plastic greenhouse and irrigated separately with UFW enrichment of hydrogen (UF+H_2) and oxygen (UF+O_2). Cryo-scanning electron microscope (cryo-SEM) observation results indicated that both UF+H_2 and UF+O_2 can increase the density of trichomes in melon leaves and petioles. RT-qPCR showed that UF+H_2 significantly increased the gene expression level of the trichome-related gene GLABRA2 (GL2). The melon irrigated with UFW containing H_2 and O_2 produced more root hairs, increased shoot height, and produced more flowers than the control irrigated with reverse osmosis (RO) water, especially enhanced trichome development and reduced significantly aphid infestation [4].

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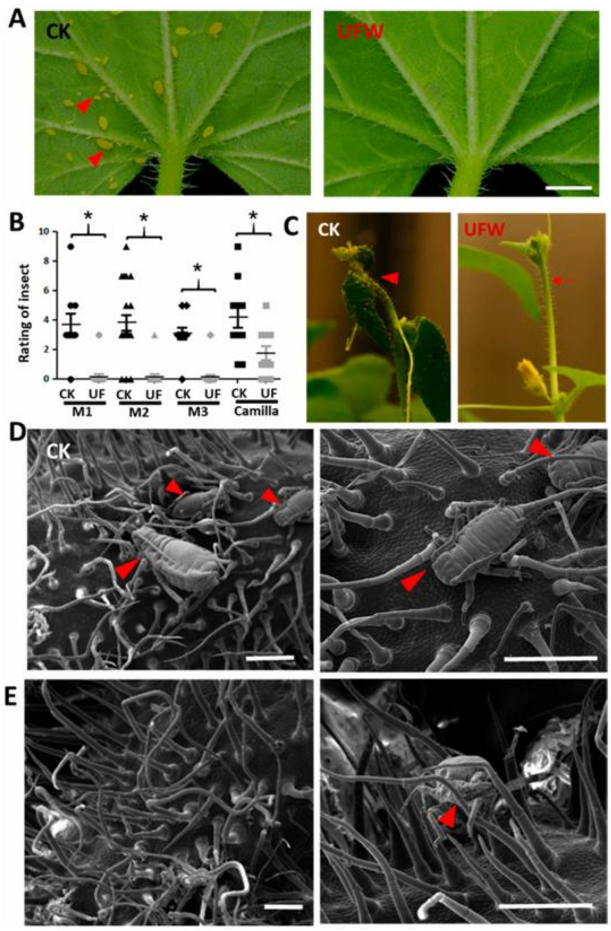

Figure 10. UFW irrigation affected aphid infestation on melon seedlings. (A) Phenotype of melon leaves attacked by aphids 14 days after transplantation. (B) Scatter plot of aphid infestation rating. (C) Aphids attacked the young flower buds of melon (arrowhead). Trichomes development after UFW treatment (arrow). (D) Aphid infestation on flower buds of CK (D) and UFW (E). The arrowheads point to the aphids (D, E).

Figure 11 is the outline of an ozone nano-mist disinfection of insect pests and spraying system which is composed of an ozone nano-mist generator and an automatic spraying system [54]. The highly dense ozone generated by dielectric barrier discharges on surface electrodes is injected into water nano-mist flow ejected from an ultrasonic oscillator. The spraying system is operated by signals from the Raspberry Pi microcomputer board which communicates remotely with the main computer using Wi-Fi. The water nano-mist reacts immediately with the flow originating in the ozone generator to form the ozone nano-mist which is composed of residual ozone, reactive hydroxyl radical(•OH) and other radicals. Six species of insect pests (aphid, moth, beetle, fly, whitefly and ant) were selected to study control performance of the disinfection spraying on these pests and biological damages on plants in the greenhouse. The formed ozone nano-mist of the size in the range 200 nm−1250 nm produced by nozzles is sprayed on the detected pests and directly taken into the pest body through spiracles, travel via the tracheae and reach the cells. The ozone nano-mist provides specified behavior at the spiracles of the insect pests, thereby can enhance the disinfection effect on the pests. Oxidizing agents including ozone and the radicals react many macromolecules in cell such as proteins, DNA and RNA to collapse their structure. The experimental results show the ozone nano-mist spraying during 30 s almost killed winged insect pests and small larva of aphids.

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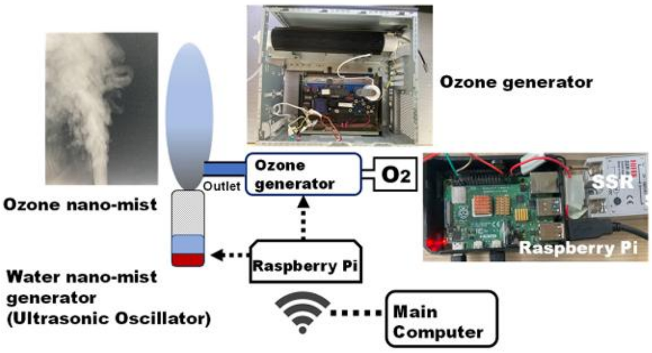

Figure 11. Outline of automatic ozone-mist spraying disinfection system.

3.2. Plant Diseases Control Using UFW

3.2.1. Function of UFB Water for Controlling Crop Diseases

UFB water offers potential practices to eliminate harmful pathogens, break down biofilms, strengthen plant immunity and reduce pest infestations for minimizing chemical pesticide dependency. Ozone UFB water exhibits bactericidal activity against pathogenic bacteria in the oral cavity and upper airway and disinfects contaminated healthcare equipment [10] [56].

(1) Reducing and eliminating harmful pathogens.

Many plant diseases, including root rot and fungal infections, thrive in stagnant water and oxygen-deficient conditions. Ozone, even in low concentrations, can effectively react with microbes in two different ways, directly and indirectly, due to its high oxidation potential against bacteria. The direct reaction involves the ozone molecules, and the indirect reaction involves the hydroxyl radicals produced by the ozone decomposition in water and are more reactive and less selective than ozone. Nanobubbles increase dissolved oxygen levels in irrigation water, creating an environment that disrupts anaerobic pathogens. Nanobubbles can prevent the spread of waterborne pathogens in the production of hydroponic and aquaponic plants. Ozone-infused nanobubbles act as a natural disinfectant against waterborne pathogens, breaking down bacteria, viruses and fungal spores without leaving harmful residues. Oxygen-enriched nanobubbles improve water circulation and continuous oxygenation, improving soil health and reducing pathogen load in the root zone, making the environment less hospitable for pest larvae. Therefore, the ozone-infused nanobubbles can also disrupt insect eggs and larvae, reducing pest populations naturally [10] [57].

(2) Breaking down biofilms.

Biofilms formed by bacteria and fungi provide a protective shield for pathogens, making them resistant to traditional treatments. Ozone attacks glycoproteins and glycolipids in the cell membrane that the direct oxidation/destruction of the cell wall resulting in rupture of the cell with leakage of cellular constituents outside of the cell. Nanobubbles can penetrate and break down the biofilms, exposing harmful microbes to oxygen and oxidative stress. This weakens their structure and makes them more susceptible to natural plant defences [10] [57].

(3) Strengthening plant immunity and health.

Improved healthy plants can foster the growth of beneficial microbes that bolster plant immunity and are more resistant to pests and diseases. Nanobubbles enhance nutrient uptake, improve root oxygenation, and promote beneficial microbial activity in the soil. This leads to stronger plants with better immune responses, reducing the need for chemical pesticides. Soaking seeds in nanobubble water enhances germination rates and reduces early-stage disease risks [10].

(4) Reducing pest infestations.

Pests thrive in stagnant water and oxygen-deficient conditions. Nanobubbles improve water circulation and oxygenation, making the environment less hospitable for pest larvae. Ozone-infused nanobubbles can also disrupt insect eggs and larvae, reducing pest populations naturally

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[10]. Controlled environment agriculture offers a protected system for plants cultivation, but remains vulnerable to diseases, particularly root diseases such as Pythium root rot and Fusarium wilt. Plant-beneficial microbes can help mitigate these harmful diseases which produce natural antibiotics and promote induced systemic resistance for enhancing nutrient uptake, stress tolerance, and disease resistance. Insufficient levels of dissolved oxygen can hinder microbial activity, lead to the accumulation of harmful compounds, and cause stress to the plants. Figure 12 shows the effects of oxygenated nanobubbles versus ordinary oxygenated bubbles on plant growth and rhizosphere microbial communities [58]. Comparing (A)(C) and (B)(D), oxygenated nanobubble technology can provide better oxygen distribution in the root zone, where many root-zone diseases can emerge and cause significant yield losses, and promote a more diverse and balanced rhizosphere microbial community, favoring beneficial microbes, enhanced root development, improved plant vigor and reduced pest infestations.

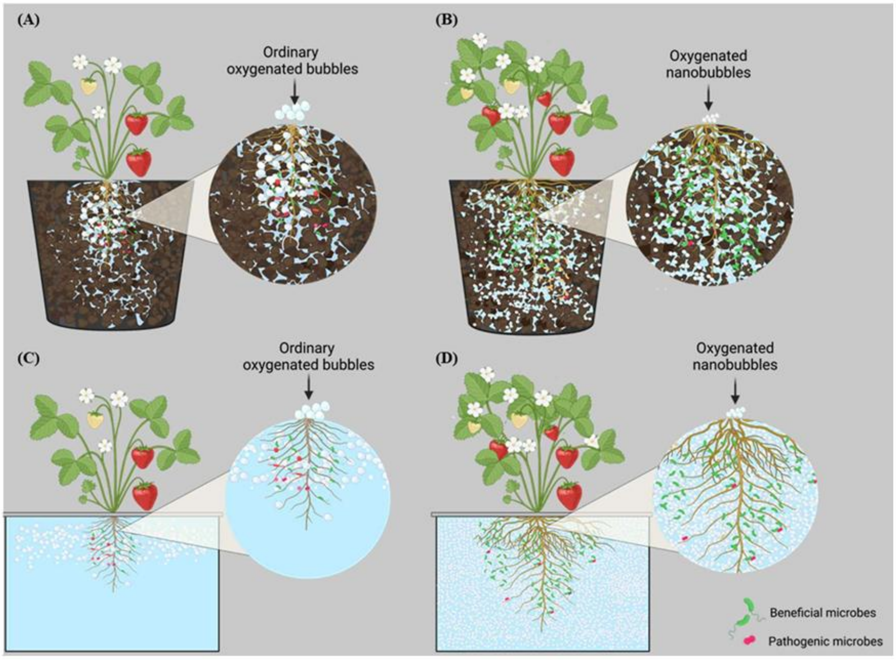

Figure 12. The effects of oxygenated nanobubbles versus ordinary oxygenated bubbles on plant growth and rhizosphere microbial communities.

3.2.2. UFB water to Control Crop Diseases

Ozone has high oxidation potential, but low ozone gas–liquid mass transfer efficiency. The remarkable properties of ultrafine bubbles, the ozone delivery by UFBs has been found to improve the disinfection capacity which has significantly increased the ozone mass transfer efficiency while reducing ozone dose. The attribute of UFBs to ozonation has stimulated widespread interest, and hence, a growing body of literature has investigated the effect of combined micro- and nanobubbles technology and ozonation in many fields [57]. The ozone ultrafine bubble water (OUFBW) exhibits bactericidal activity against pathogenic bacteria. Consequently, ozone micro-bubble aeration caused severe membrane damage to bacterial cells and fragmented the bacterial DNA, which caused a rapid decrease in the bacterial metabolic activity (> 80 %) [55,56]. UFB/OUFBW can kill soil borne pathogens and control soil borne diseases in the circulating nutrient solution, control airborne diseases, etc., as well as solve water borne diseases of nutrient deficient and rotten roots of hydroponics vegetables, and deal with pesticide residues in fruits and vegetables.

(1) Controlling soilborne diseases.

In the vegetables cultivated in protected areas, there are more than 50 kinds of diseases that often occur or cause serious damage. The initial infection of most of these diseases is almost all from the soil. Soilborne plant pathogens persist in the soil matrix or in residues over the soil surface which are distributed widely in soil, however few species exhibit localized distribution patterns. Once established, these pathogens accumulate through synergistic associations and cause greater economic losses that are difficult to control as they have wide host range and can survive for long periods on soil organic matter and plant debris, as free-living organisms or by producing resistant structures. The soilborne diseases remain unnoticed until the above ground plant parts exhibit symptoms such as chlorosis, stunting, wilting and finally death. Soilborne diseases, caused by fungi, bacteria, nematodes, oomycetes, protozoa and viruses, are considered vital in realization of potential yield and listed as the top five difficult diseases to control in the field of plant diseases due to their extensive and serious damage. Soilborne pathogens are significant contributors of plant yield loss globally [59]. The control of soilborne diseases has gone through a long process, from the application of biocidal soil disinfectants such as cobalt chloride, methyl bromide and a variety of fungicides to biological control, but the spread and harm of soilborne diseases are still rampant.

Studies on the regulation role of related genes have found that a certain ozone concentration can raise the anthocyanin and carotenoid of plants and revealed the molecular mechanisms underlying the anthocyanin and carotenoid. When plants were exposed to elevated ozone, the ozone affected the utilization of light energy in plants [60]. Ozone concentration in nutrient solution can effectively kill the pathogens of epiphyte and bacteria in the nutrient solution causing soilborne diseases, such as Fusarium Wilt and mustard family soft rot pathogens, and mosaic virus causing viral diseases [61]. Ozonated water soil drenches significantly reduced nematode (*Meloidogyne incognita* Kofoid and White) infection rate in tomatoes grown in a growth chamber by 23% compared to controls [51].

However, under normal temperature and pressure, the solubility of ozone in water is about 13 times higher than that of oxygen and 25 times higher than that of air. Ozone in water is easy to dissipate. The retention, persistence and self-pressurized dissolution of UFBs in water just make up for the defect of the ozone. With irrigation water as the sterilization medium, micro-nano aeration technology is used to produce high concentration ozone water to jointly sterilize and disinfect the soil and air in the protected area, and integrate physical, chemical and biological technologies to effectively alleviate the invasion of soilborne diseases.

The MNB generator was combined with the ozone generation system to prolong the retention of ozone in water. The ozone MNB water with a concentration of 2.2~8.0 mg/L was prepared by adjusting the air inflow and water inflow, was applied to the disinfection and sterilization of soil and substrate. After a series of tests found that the microbial killing rates of bacteria, fungi and actinomycetes in soil and substrate could reach more than 50% after three times of disinfection with 6.0mg/l ozone MNB water, and the sterilization effect of substrate was better than that of the soil [62]. The properties of micro/nano bubble ozone water and its disinfection efficacy in the inactivation of soilborne pathogens and soil microbes were investigated. Ozone was bubbled into the samples by micro/nano bubble aeration system and the irrigation of micro/nano bubble ozone water to greenhouse soil samples once a week was conducted to investigate the germicidal efficacy of the target bacteria. The results showed that micro/nano bubble water could promote the ozone solubility and enhance the stability of the ozone water and confirm the high germicidal efficacy of micro/ nano bubbles ozone water on *F. oxysporum* f. sp. *lycopeersici* in soil [63].

(2) Controlling airborne diseases.

Airborne diseases, caused by plant pathogens such as spores, bacteria, and viruses that spread or disperse by wind, rain splash, insects, or human activities, often travelling far rapidly over wide areas and infecting plants at a distance from the original sources, pose a significant threat to plant health, agricultural productivity, and ecosystem stability. Many pathogenic fungi are remarkably well adapted to airborne spread. Some plant pathogens can travel thousands of kilometers by air while maintaining their viability and ability to cause new epidemics [64][65].

Ozonated water applied by foliar treatment spray in growth chamber tomato led to a reduction in the airborne tomato spotted wilt virus severity, as treated plants displayed 20% less disease incidence and severity [52]. In vegetable greenhouse, ozone micro/nano bubble water has significant control effect on vegetable powdery mildew, gray mold and other common diseases. The feasibility of spraying micro/nano ozonated water to prevent and control tomato airborne diseases was studied, the schematic diagram of experimental setup is shown in Figure 13. The experiments included the dissolution and attenuation characteristics of micro/nano bubble ozonated water, the prevention effect of micro/nano bubble ozonated water on the pathogens of early blight and leaf mold, and the effect of spraying different concentrations of micro/nano bubble ozone water on tomato growth. The results showed that micro/nano bubble ozonated water is effective on both pathogen conidia in vitro, and the germicidal efficacy. Meanwhile, praying ozonated water in a certain concentration range (0.6–1.8 mg/L) had no significant negative effects on tomato growth [66][67].

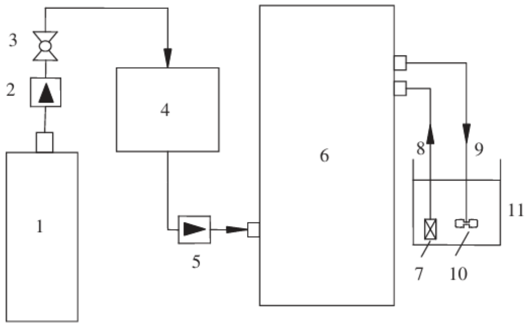

Figure 13. Schematic of micro/nano bubbles ozonated water generator. 1. Oxygen bottle; 2. Flow meter; 3. Regulating valve; 4. Ozone generator; 5. Flow meter; 6. Micro/nano bubble generator; 7. Inlet valve; 8. Inlet; 9. Outlet; 10. Aerator; 11. Plastic cylinder.

(3) Controlling waterborne diseases and solving root rot of hydroponic vegetables.

Soilless and hydroponic systems, which do not use natural soil as cultivation medium, cultivate crops in a controlled amount of water and nutrients or on a cultivation bed formed by sand gravel, vermiculite, perlite, rice husk fumigation, coal cinder, rock wool and other soilless substrates. Soilless and hydroponic systems allow greater control of plant health regardless of soil quality and environment. But in soilless culture, especially hydroponics, the lack of nutrient solution often leads to plant nutrient deficiency and even root rot. The long-term recycling of nutrient solutions will breed a large number of microorganisms, and waterborne pathogens such as fungi, bacteria, and algae can spread faster through water than in soil. The hydroponic system is like a little piece of heaven that provides moisture, nutrients, and the right temperatures needed to thrive where they have billions of years of survival expertise at their disposal.

The change from soil-based production to hydroponic systems could lead to a significant risk of the occurrence of other pathogens especially the waterborne diseases adapted to aquatic environments in hydroponic crops, especially those caused by some species such as *Fusarium*, *Pythium* and *Phytophthora*. To keep the hydroponic system free of pathogens, it's important to clean the water and maintain a correct PH value, use water filters and UV light, control diseases using biological control agents and balance fertilizers, monitor the temperature and provide oxygen [68][69]. The best way to manage waterborne diseases in a hydroponic system is to prevent them in the first place.

Therefore, the nutrient solution needs to be oxygenated and disinfected in the actual production process to meet the needs of crop growth. Through micro/nano aeration technology, air and oxygen are dissolved into the nutrient solution, and the supernormal solubility of micro/nano bubbles suspended in water is used to rapidly improve the dissolved oxygen value of the nutrient solution to promote buds, strengthen roots and increase yield and reduce the occurrence of hypoxia and root rot in the hydroponic system. Using micro/nano aeration technology, the low concentration of ozone can be wrapped into micro/nano bubbles, and the ozone can be slowly and gently released into the nutrient solution. Using the charge on the surface of micro/nano bubbles to adsorb microorganisms in the water, it can instantly kill pathogenic microorganisms and inhibit effectively the spread of

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diseases in the nutrient solution. The waste nutrient solution produced by soilless cultivation system is sterilized with micro/nano bubble ozone water, which can be recycled or used as liquid fertilizer in cultivated land to avoid environmental problems such as groundwater pollution caused by direct discharge [14,70,71] [72].

(4) Treatment of pesticide residues in fruits and vegetables.

It is an emerging and promising technology for removing pesticides in the aqueous environment and degrading the residual pesticides from the fruits and vegetables surfaces as the intensive use of pesticides, such as ozonation, microwaves/ultrasonication and advanced oxidation process. The micro-nanobubble ozonation was used to reduce/degrade residual pesticides from fruits and vegetables. Figure 14 shows the major processes, reactions, and mechanisms involved for the effective degradation of pesticides in aqueous medium by micro-nano bubbles [73].

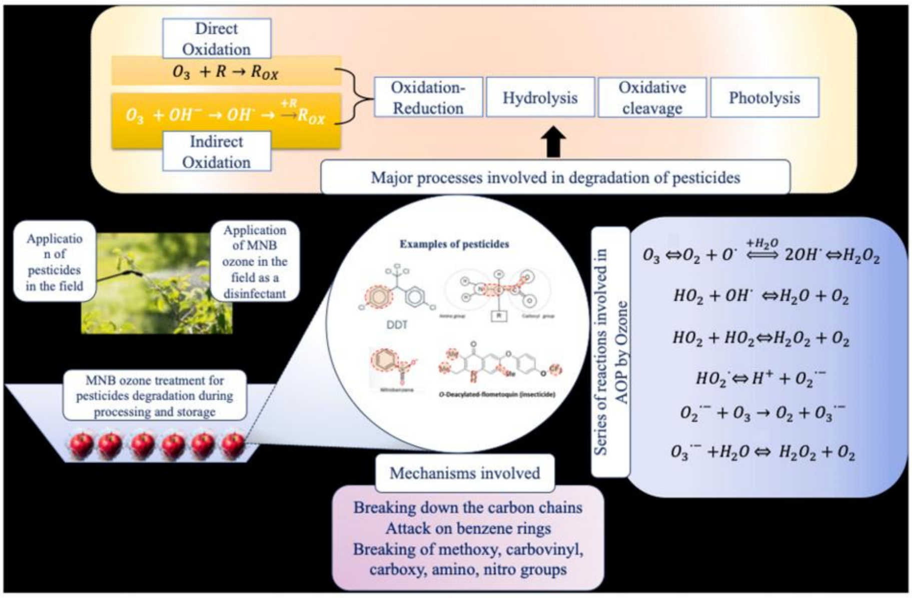

Figure 14. Effective pesticide degradation in aqueous medium by micro-nanobubbles.

Using ozone micro-nano bubble water to clean and disinfect fruits and vegetables, because of the ultrafine bubbles, it can go deeper into the cracks of fruits and vegetables, kill bacteria and decompose residual pesticides. In the cleaning process, mechanical damage can be avoided to the greatest extent, so that the original quality of fruits and vegetables can be better maintained, and better product appearance can be obtained while avoiding the loss of nutrients. By using ozone micro-nano bubble water, the cleaning rate of pesticide residues in cabbage, cucumber and leek can reach about 90% [74].

When acidic electrolyzed water containing ozone ultrafine bubbles and strong mechanical action combined to wash fresh vegetables, the lowest viable bacterial count was recorded among other treatments including sodium hypochlorite which shows that ozone-rich microbubbles exhibited higher disinfection activity and efficiency against pathogens than the millibubbles over the same period of applications [57].

4. UFB water Against Abiotic Stresses of Crops

Climate change, triggered by anthropogenic activities and other inexorable factors, has led to a surge in abiotic stresses, such as drought stress, salinity stress, heavy metal stress, which significantly impair crop yields, underscoring the need for innovative solutions [75]. Positive effects of UFBs on plant growth have been controversial. But micro/nano bubble water could be applied to improve abiotic stress tolerance for addressing the pressing challenges.

4.1 UFW on Plant Growth Under Salt Stress.

Soil salinization is becoming a severe environmental issue limiting the growth and yield of crops worldwide. Subsurface drip irrigation with micro-nano bubble hydrogen water (SDH) is an innovative way to realize the role of hydrogen gas in improving plant resistance to salt stress in practical agricultural productions. Figure 15 shows an irrigation system which was composed of two main parts, a micro-nano bubble hydrogen water generation unit and a water supply unit using the subsurface irrigation system. To investigate the effect of micro-nano bubble hydrogen water on lettuce physiological processes with and without the salt stress, four treatments were arranged, SDH, NaCl, NaCl+SDH treatments and CK. The results show the positive effects of SDH on various aspects of physiology in plants. SDH significantly enhanced the lettuce growth performance, including fresh weight, leaf number, photosynthesis, and root development under salt stress [76].

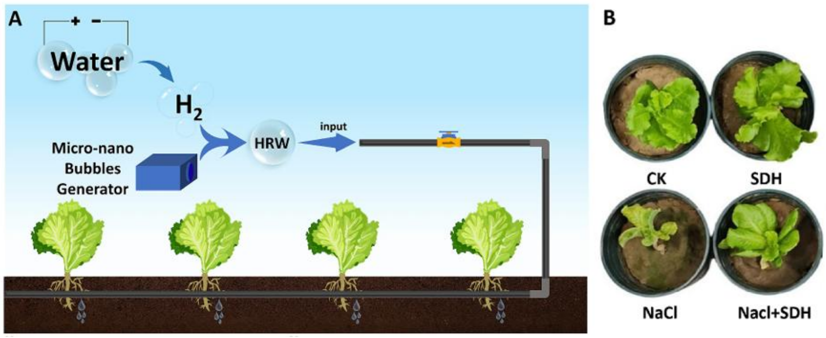

Figure 15. Phenotype observation and shoot-root growth parameters. (A) Schematic layout of irrigation system. (B) Effects of SDH, NaCl, and NaCl+SDH treatments on lettuce growth using CK as a control.

As shown in Figure 16, to explore the effect of micro/nano bubble water oxygenation on the growth of rice seedlings under salt stress, the rice variety 9311 (model species) and JX99 (relatively salt-tolerant) seedlings were used as test materials, rice seedlings were cultivated with Yoshida nutrient solution + distilled water and Yoshida nutrient solution + micro/nano bubble water, and then were treated with 0.6% NaCl salt stress and no salt stress to determine the growth characters and physiological and biochemical indexes of rice seedlings. Results showed that under salt free stress, the root length, stem fresh weight and stem base width of 9311 rice seedlings decreased significantly by 24.87%， 11.20% and 12.50%, the root length and leaf number of JX99 increased significantly by 31.76%，21.88%，the malondialdehyde content of the two varieties decreased significantly by 41.19%，15.85%, and the proline content of the two varieties did not change significantly. Under 0.6% salt stress, the root length, stem fresh weight and root fresh weight of 9311 rice seedlings decreased significantly by 45.16%, 46.81% and 38.64%. The stem length, root length, stem fresh weight and root fresh weight of JX99 increased significantly by 28.09%， 35.67%， 67.97% and 55.43%, the malondialdehyde contents of the two varieties increased significantly by 46.25% and 36.11%, the proline content of the two varieties increased significantly by 85.63% and 131.64%． Under salt free stress, micro/nano bubble water slowed down the degree of membrane peroxidation and reduced the damage of reactive oxygen species. Under 0.6% salt stress， micro/nano bubble water exacerbated the degree of membrane peroxidation and the damage of reactive oxygen species and osmotic stress [77].

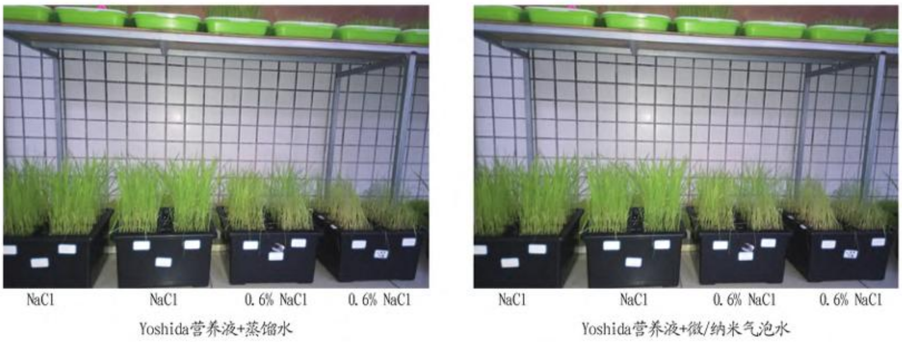

Figure 16. Comparison of the growth status of rice seedlings under micro /nano bubble water and salt stress. Note: The left row of pot was variety 9311, and the right row of pot was JX99.

4.2 UFW Mitigates Plant Growth in Damaged Soil

To specify the UFB water act on microorganisms, the UFB water promoting plant growth by mitigating the damage of the soil and the effect of UFB water on pathogens causing the deterioration of the soil were focused on. The results showed that water containing ultrafine/nano bubbles (UFBs) promoted the growth of tomato (*Solanum lycopersicum*) in soil damaged by cultivation of tomato in the previous year or bacterial wilt-like disease, and also promoted the growth of lettuce (*Lactuca sativa*) when lettuce was grown in the soil damaged by repeated cultivation of lettuce. The growth of lettuce was not affected by UFB water treatment in the soil damaged by the cultivation of tomato. UFB water partly suppressed the growth of the pathogen of bacteria wilt disease, *Ralstonia solanacearum in vitro*. The data suggest that UFB water is effective to recover the plant growth from soil damage. The effects of UFBs on the growth of *S. lycoper sicum* germinating in the “disease” soil close to *S. lycopersicum* showing the symptom of bacterial wilt-like disease was examined. *S. lycopersicum* in bacterial wilt-like disease was detected with probability more than 5% every year. The growth of the plants developing the disease was severely retarded, eventually the whole plants were completely wilted. UFB water somehow affects damaged soil due to bacterial infection. In tomato cultivation in a greenhouse, bacterial wilt disease commonly occurs. The soil was prepared by mixing the portions of the root zone and rhizosphere of the diseased plants or healthy plants as a control. Tap water was used as raw water taking practical use into consideration. Figure 17 showed that effects of UFB water on the growth of *L. sativa* in the soil damaged by repeated cultivation of *L. sativa*. Variety Falcon showed abnormal germination in several cases. UFB water significantly improved the root growth of plants grown in the diseased soil while it showed no effect on the growth of the one grown in the healthy soil. No effect of UFB water in the soil recovered from the plant with either bacteria wilt disease-like or healthy were observed [78].

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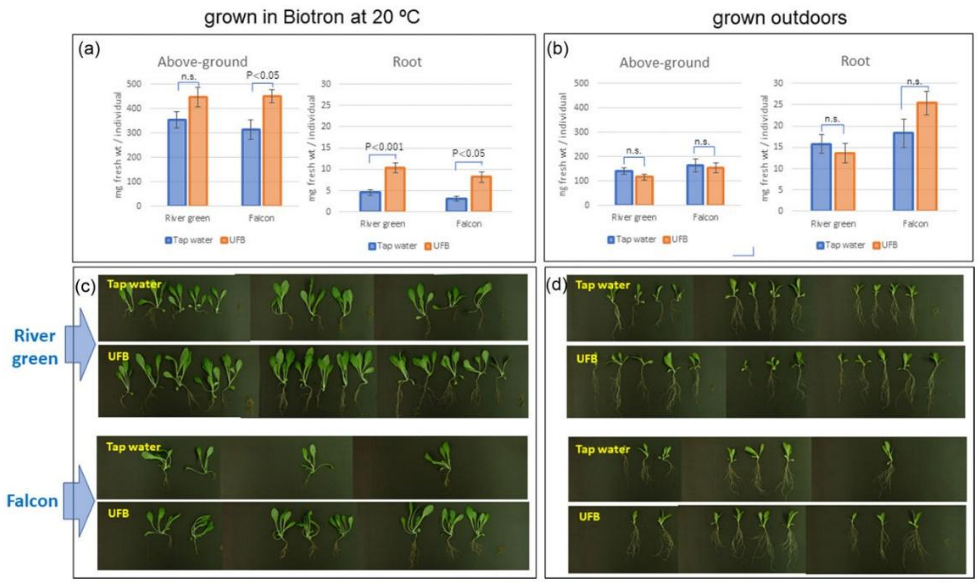

Figure 17. Effects of UFB water on the growth of *L. sativa* in the soil damaged by repeated cultivation of *L. sativa*. Two varieties, River green and Falcon, were sown in 3pots each, and with 5 seeds each pot. They were grown at 20°C in a biotron (a, c) or outdoors (b, d). Variety Falcon showed abnormal germination in several cases. Such abnormal individuals were eliminated for the succeeding growth test. n.s. means the effect of UFB water is not significant.

5. Summary, Conflict and Prospects

The properties and generation of UFB water, controlling biotic stresses of crop pests via UFW, and UFW against abiotic stresses of crops were reviewed, but obviously, UFB water is not omnipotent. When carrying out plant protection practices using UFB water, its efficiency must be investigated because the results might conflict with some studies.

Willis T. Spratling et al. assessed the impacts on turfgrass health from repeated applications of oxygenated nanobubble water and ozonated nanobubble water treatments. Two different application methods, soil drench versus foliar spray, were compared for all treatments. The effectiveness of these treatments was evaluated and investigated against dollar spot (caused by *Clarireedia* spp., one of the most detrimental diseases of turfgrasses) development in seashore paspalum and for turf quality in both field and growth chamber-controlled environment experiments. The results show that the application methods had no effect on treatment efficacy, and the treatments did not cause any noticeable phytotoxic damage to seashore paspalum tissues but failed to mitigate dollar spot severity in any capacity. The contrasting plant protection results about the different mechanisms of pathogen spread and/or infection in different oxygen conditions or anaerobic environments were discussed. The lack of efficacy from nanobubble water treatments in their trial may be attributed to inherent infection processes of dollar spot pathogens and could have been due to dissolved oxygen loss when spraying was directed after passing through a nozzle and substantial off-gassing because of a high-pressure spray nozzle into the lower pressured atmospheric environment. But they still maintain that oxygenated and ozonated water treatments did not negatively impact turfgrass health and nanobubble aeration is a valuable and viable approach for producing oxygenated or ozonated water [53].

Therefore, the key directions for future research could focus on in-depth exploration of generation mechanisms, development of advanced materials, and expansion of application fields for improving the UFW treatments, including increasing bubble stability, enhancing generation efficiency, reducing energy consumption, optimizing device design and promoting large-scale application remain. A series of fundamental researches could be carried out in plant protection practices, including the mechanism of UFW controlling plant pests and diseases, the molecular

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mechanism of UFW affecting plant pest resistance, the plant growth in harsh polluted environments like salinity soil and contaminated soil, the UFBs behavior with hydrophobic and hydrophilic surfaces of crops and the integrated intelligent crop growth system.

5.1 Mechanism of UFW Controlling Plant Pests and Diseases

The nanobubble waters, as carrier of active antimicrobial agents, may enhance the solubility, wettability, dispersion and bioavailability of pesticides and further improved pesticide application technologies could be studied.

5.1.1 Insecticidal Mechanism of UFW Combined with Pesticides

The respiration of insects is generally realized through gas exchange between stomata and the outside body. Therefore, to take advantage of the slow rising properties of UFBs and more evenly distributed in pesticides, it is possible to apply the UFB water spraying of mixing pesticides on the targeted plants suffering from insect pests, the UFBs of mixed pesticides can enter the insect body through the insect respiratory stomata. It is necessary to study the poisoning mechanism of pesticides and the unique performance advantages of UFW, as well as the mechanism of the combination of UFBs and pesticides to destroy the internal organizational structure of insects, so as to achieve the purpose of insecticidal effect without any negative impacts on crops. As we know, ozone may have broad-spectrum plant pest control capability, we can conduct the integration of different ozone concentrations and UFW to seek the optimal ozone concentration which is safe for crop growth and has control effect. Combining ozone with UFB technology, that is, injecting ozone into pesticides in the form of micro-nano bubbles through a special device, can not only increase the contact area between ozone and pesticides, improve the solubility and utilization of ozone, but also take advantage of UFBs to extend the retention time of ozone in pesticides. Using the respiration system of insects, the application of ozone UFW spraying can make high concentration of ozone UFWs enter the insect body, destroy its internal tissue structure, and achieve the purpose of killing insects.

5.1.2 Plant Disease Control Mechanism of OUFBW

Ozone has a strong inactivation effect on almost all bacteria, viruses, molds, fungi, protozoa and oocysts. Oxygen atoms can oxidize the bacterial cell membrane until they penetrate the unsaturated bond between the cell membrane and their bodies and immediately control bacterial life. The oxidation of ozone can directly destroy the ribonucleic acid DNA material of the virus, showing a bacteriolytic bactericidal process. However, ozonated water is unstable and has a short half-life, and excessive ozone concentration may destroy the photosynthetic system of plant leaves. Then the ozone UFB water may exert potent bactericidal activities which are effective against bacteria. If serving the OUFBW as a disinfectant against crop diseases, further studies are required to explore optimal application methods and machinery. The microbiocidal effect of ozone confined to UFB mainly depends on the action of ozone. UFB might be used as a device holding ozone in water for a prolonged period.

5.2 Molecular Mechanisms Underlying UFW's Effects on Pest Resistance

It was reported that non-glandular trichomes play a role in the mechanical defense against insects, while glandular trichomes can secrete secondary metabolites including mono- and sesquiterpenes that contribute to host plant resistance against pests. The glandular trichome derived terpenes in cultivated and wild tomato species produce metabolites that have repellent and toxic activity against multiple biting-chewing or piercing-sucking herbivores such as aphids [79]. Further research can be carried out into the molecular mechanisms underlying UFW's effects on plant growth and pest resistance. The mode of action of the effective sesquiterpenes may be unraveled, the potential performance, feeding and choice behavior of specific targets in aphids may be characterized underlying the UFW's involvement.

5.3 Hydrophilic Nanopatterned Surfaces of Targeted Crops

A rough surface can influence the apparent contact angle at the boundary between a liquid and the surface, and many examples have been given of the difficulty of wetting rough surfaces because of their large apparent contact angles. If the water is under zero hydrostatic pressure it will come to rest on a porous surface in some position [80]. The hydrophilic surfaces possess high surface energy that draw water and enable the surface wetting which the water disperses evenly on the surface [6]. It is desirable that the pesticide droplets adhere to the crop leaf surface, called hydrophilic surface which possesses high surface energy that draws droplets and enables surface wet, and the pesticide droplets can deposit evenly on the surface at the contact angle of less than 90°. In some cases, pesticide droplets move away from the leaf surface, called hydrophobic surfaces. There has been an accumulation of evidence for the existence of nanobubbles on hydrophobic surfaces in water [81]. The formation of interfacial nanobubbles on substrates with different hydrophobicity and the effect of interfacial nanobubbles on the interaction of inter-particles were explored [82]. Due to their unique physiochemical properties, the distinct behavior of nano-bubbles on hydrophobic and hydrophilic surfaces could be studied. The effects of different plant leaves on the deposition characteristics of UFB water and ordinary water can be compared to improve the deposition performance of droplets on the plant target. A hydrophilic nanopatterned leaf surface might be generated by combining the UFB with pesticides through hydrodynamics of mechanical shearing, pressurized gas dissolution, microfluidic technology, micro-porous dispersion, electrochemical and cavitation effect methods.

5.4 Application of Activated UFB Water in Plant Protection Practices

A large number of activated waters have been studied for plant disease and pests control, such as the application of physical methods to increase the dissolved oxygen concentration in ordinary water to form aerated water, the magnetized water formed by making ordinary water vertically pass through a fixed magnetic field at a certain velocity, the deionized activated water that can significantly change the physical and chemical properties of water and increase the activity of water molecules, and the plasma active water, the electrolyzed water, the electricity generating functional water, and so on [83–88]. The significant effectiveness of antimicrobial activity of electrolyzed water against pathogenic bacteria has been reported. Ultrasound and mild heating can enhance the bactericidal efficiency of electrolyzed water through reducing the dip time, the synergistic effect of slightly acidic electrolyzed water combined with ultrasound and mild heat in *L. monocytogenes* and *S. Typhimurium* adherent on surface of fresh-cut bell pepper was evaluated [84]. However, electrolyzed water is relatively unstable and vulnerable to external environmental factors such as light, temperature, sealing degree and other conditions. Therefore, the best application scheme for integrating UFBs to improve the performance of various activated UFB waters should be proposed according to different environments, crops and plant diseases through. Further study the application of activated UFB water in plant protection, improve plant growth in saline alkali land, contaminated soil fields and other harsh environments, promote plant roots, and optimize breeding. In addition, whether the high oxygen environment provided by UFB water will also promote the growth and development of harmful microorganisms in soil, we need to further study the bactericidal mechanism of activated UFB water whether it will affect plant roots or soil environment.

5.5 Integrated Intelligent Plant Cultivation System

Most of the present studies on UFB water applications in plant protection practices are performed with relatively small scales. It would be helpful to examine upscaling the UFB water applications in plant protection practices and consider the cost/benefit analysis. As a smart sprayer can spot-spray the most pertinent selective herbicides onto the susceptible weeds based on precision weed mapping [89], UFW may be used alongside integrated pest management strategies. UFB technology can also be combined with the Internet of Things and intelligent pesticide prescription spraying [90] to realize intelligent applications, such as installing sensors in the greenhouse to

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monitor environmental parameters such as temperature, humidity, carbon dioxide concentration and plant growth status in real time. When plant diseases and pests occur in orchard environments, the multi-dimensional prescription map and variable-rate spraying operations [91] can automatically adjust the concentration composition, spraying time frequency and spraying amount of UFWs or OUFBW, effectively sterilize and kill pests, and further integrate the intelligent tree growth system with multiple functions such as aeration, disinfection, fertilization, irrigation and plant protection, so as to realize unmanned precision cultivation, and remote management using mobile devices.

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Abbreviations: The following abbreviations are used in this manuscript:

UFB	Ultrafine Bubble
UFW	Ultrafine Bubble Water
OUFBW	Ozone Ultrafine Bubble Water
MNB	Micro-nano Bubble
NB	Nanobubble

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