


Disinfection options for irrigation water: Reducing the risk of fresh produce contamination with human pathogens

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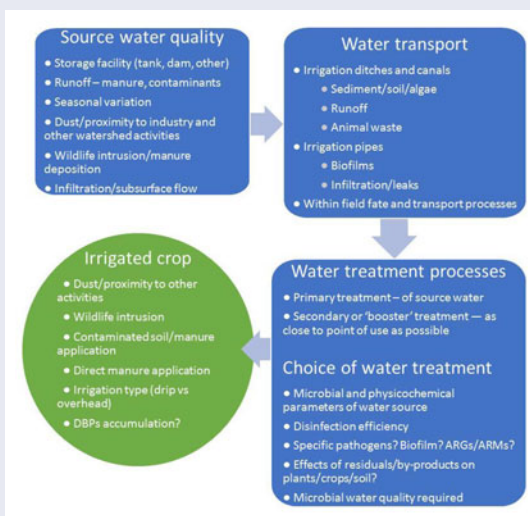
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ABSTRACT

The growing health and economic burden posed by food-borne pathogens has stimulated global interest in the development of safe, affordable, effective and environmentally-sustainable irrigation water treatment technologies. This review critically compares the potential of existing and emerging methods for disinfection of irrigation water to reduce pathogenic microbial loads on high-risk vegetables and minimally processed fresh produce. We explore electrochemical disinfection and electrolyzed oxidizing water as alternatives to traditional chlorination, and identify hydrodynamic cavitation as an emerging disinfection strategy


worthy of further investigation in this context. In addition, we assess the state of the knowledge regarding the impact of current water sanitation strategies on the ecological dynamics of plant and soil microbes and the potential induction of viable but non-culturable cells. Increased research in these areas could translate into substantial improvement in the overall quality and value of fresh produce, while maintaining environmentally-sustainable irrigation water usage.



KEYWORDS Foodborne pathogens; irrigation water disinfection; viable-but-non-culturable

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1. Introduction

Crop agro-ecosystems are at the heart of the food–energy–water nexus, accounting for ~70% of total freshwater withdrawal in the world (Food and Agriculture Organization of the United Nations (FAO), 2015). As an example, irrigated agriculture accounted for 58% of all water use in Australia in 2015–16 (ABS, 2017), and projected agricultural water demand is set to increase by 50% by 2050 (AWA, 2017). The increasing demand for water to support food production is a global trend that is significantly exacerbating pressure on water resources. Thus, alternative irrigation water sources are increasingly sought, and the quality and safety of those supplies must be ensured to safeguard future water and food safety.

1.1. Microbiological contamination of irrigation water and pathogen transfer to food

Irrigation water can be obtained from a range of water sources and the potential for microbiological contamination needs to be carefully considered. Table 1 lists the range of available water sources for irrigation and their relative risk of microbial contamination. In the case of irrigated food crops, particularly minimally processed foods such as lettuce, spinach, parsley and other leafy greens, opportunistic and human pathogens are of particular concern. Despite increasing efforts to improve sanitation, outbreaks linked to microbial contamination of minimally processed foods continue to occur around the world. In some instances, these outbreaks have been associated with pathogens that are uncommon in these foods, for example, *Salmonella* spp. and *Listeria monocytogenes* in cantaloupes, prepacked lettuce, and baby spinach leaves (FSANZ, 2016; Zhu, Gooneratne, & Hussain, 2017). Pre-harvest water supplies (i.e., irrigation water) and postharvest water (i.e., washing water) have previously been identified as the main sources of contamination in produce associated with illness (FSANZ, 2011), and the growing use of whole genome sequencing in outbreak investigations is providing increasing evidence for the role of contaminated irrigation water in pathogen outbreaks (Hoelzer, Switt, Wiedmann, & Boor, 2018). It is clear that contaminated irrigation water can transfer pathogens to edible produce (Jongman & Korsten, 2017; Markland, Ingram, Kniel, & Sharma, 2017) and leafy greens are especially vulnerable to contamination with opportunistic human pathogens because they have large surface areas, are often grown in close proximity to soil, are irrigated intensively, and are mostly consumed raw (De Keuckelaere et al., 2015).

Given the above, it is evident that in some settings effective sanitation of irrigation water is paramount in ensuring the safety of edible produce. Guideline values for pathogens in irrigation water have historically been

Table 1. Irrigation water sources and their potential for microbial contamination.

Water source	Potential for microbial contamination	References
Municipal/potable water	Low risk (treated to potable use standards), but cost and volumes required might be prohibitive	Uyttendaele et al., 2015
Surface water (incl. rivers, streams, creeks, lakes, dams and reservoirs)	High risk, with many potential sources of contamination, incl. wildlife or stock intrusion and fecal deposition, sewage or septic discharges and industrial effluents; high turbidity from suspended solids	Jones, Worobo, & Smart, 2014; Steele & Odumeru, 2004
Groundwater	Generally considered low risk, but overextraction of groundwater and contamination of shallow aquifers contributes to higher potential risk	Bradford & Harvey, 2017; Van Haute et al., 2015
Harvested rainwater	High risk of contamination by animal feces and organic debris; volumes required might be prohibitive except for small-scale applications	Dobrowsky, De Kwaadsteniet, Cloete, & Khan, 2014
Recycled wastewater	High initial microbial content but generally low risk with sufficient and appropriate treatment	Allende & Monaghan, 2015
Untreated wastewater/indirect wastewater reuse	High risk; prevalent because of insufficient wastewater treatment infrastructure in expanding urban areas	Thebo, Drechsel, Lambin, & Nelson, 2017

framed around fecal contamination and associated indicators (i.e., fecal coliforms), with the WHO guideline value of $\leq 1,000$ colony forming units of fecal coliforms per 100 ml in wastewater for irrigation (World Health Organization (WHO), 1989). Other guidelines might be more specific and restrictive, specifying *E. coli* rather than coliforms (i.e., < 1 *E. coli* per 100 ml of recycled wastewater; E.P.H.C., 2006) or targeting other pathogens (absence of *Salmonella* required in 100% of samples in recent EU legislation; European Commission, 2019). The impetus or trigger for irrigation water treatment should be derived from relevant local guideline values and microbial risk assessment of the potential pathogen exposure from contaminated crops (Uyttendaele et al., 2015).

The intention of this review is to critically assess the literature relating to existing methods for disinfection of irrigation water for food crops. The information presented is mainly focused on bacterial pathogens, whilst acknowledging that there are substantial disease burdens associated with other pathogens such as viruses, protozoa and helminths (Ramírez-Castillo et al., 2015). Where there is limited information on the application of treatments specifically to irrigation water, we have drawn on literature assessing the application of sanitation technologies in other scenarios and their potential for adoption for irrigation water treatment.

1.2. Human health effects of contaminated fresh produce and mechanisms of pathogen contamination

There are substantial human health effects of contaminated fresh produce—for instance, between 2004 and 2013, over one third of foodborne illnesses in the USA were from the consumption of contaminated fresh produce (Fischer, Bourne, & Plunkett, 2015). Considering that the World Health Organization (WHO) reported a global burden of 600 million cases of foodborne illness in 2010 (420,000 resulting in death), the importance of water sanitation during the pre- and postproduction of fresh produce should not be ignored (World Health Organization (WHO), 2015b).

Pathogen survival on plant surfaces has been clearly demonstrated, especially in biofilms, as has the internalization of pathogens into plant tissues – i.e., endophytes (Berg, Eberl, & Hartmann, 2005; Berg et al., 2013; Hardoim et al., 2015; Lim, Lee, & Heu, 2014). In fact, many opportunistic human pathogens colonizing fresh produce have an endophytic lifestyle, using vegetables as an alternative host to survive in the environment and as a vehicle to colonize human and animal hosts once ingested (Mendes, Garbeva, & Raaijmakers, 2013). Critically, the endophytic interaction leads to difficulties for postharvest decontamination of fresh produce (Berger et al., 2010). Therefore, while treatment of irrigation water might be

effective in reducing the incidence of pathogen contamination through direct transfer of pathogens from irrigation water to plant surfaces and soil, changes in agricultural management practices might also be required to reduce the potential for endophytic pathogen colonization from contaminated soil and/or manure-based fertilizers.

In addition to the general risks to human health associated with pathogen contamination in food, the heightened risks posed by antimicrobial resistant microorganisms and antimicrobial resistance genes, particularly when associated with pathogenic microorganisms, is also of key relevance (Thanner, Drissner, & Walsh, 2016). Antimicrobial resistance is a major concern worldwide and is recognized by the WHO as a “global health security emergency”, prompting the World Health Assembly to develop a Global Action Plan on antimicrobial resistance (World Health Organization (WHO), 2015a). A number of areas specifically highlighted as antimicrobial resistance research needs have been documented and many of them are directly relevant to food irrigation water supply (Wuijts et al., 2017), e.g., the identification of treatment technologies that can remove antibiotics and other antimicrobial agents, their metabolites, antimicrobial resistant microorganisms and antimicrobial resistance genes in water.

1.3. Strategies to reduce contamination of fresh produce

To reduce the potential for pathogen contamination of fresh produce, selection of an appropriate water source and/or pretreatment of irrigation water is critical (De Keuckelaere et al., 2015). Irrigation practices and distribution networks must be maintained to the highest possible standards to ensure that the potential for contamination is minimized. As with drinking water treatment, a multiple barrier approach is recommended to ensure that irrigation water quality remains high even in the event of failure or suboptimal performance of individual treatment modules (NHMRC & NRMCC, 2011). On-site treatment of irrigation water could represent an important component of a multiple barrier approach, especially in the context of irrigation with recycled water.

2. Treatment technologies for irrigation water

Water treatment for potable use and wastewater treatment for reuse or discharge draw on a range of different treatment technologies, many of which are potentially applicable to irrigation water treatment. A multitude of factors can affect the choice of irrigation water treatment technology (Figure 1). Selection criteria for treatment technologies can generally be broken down into three categories—technological, managerial, and sustainability

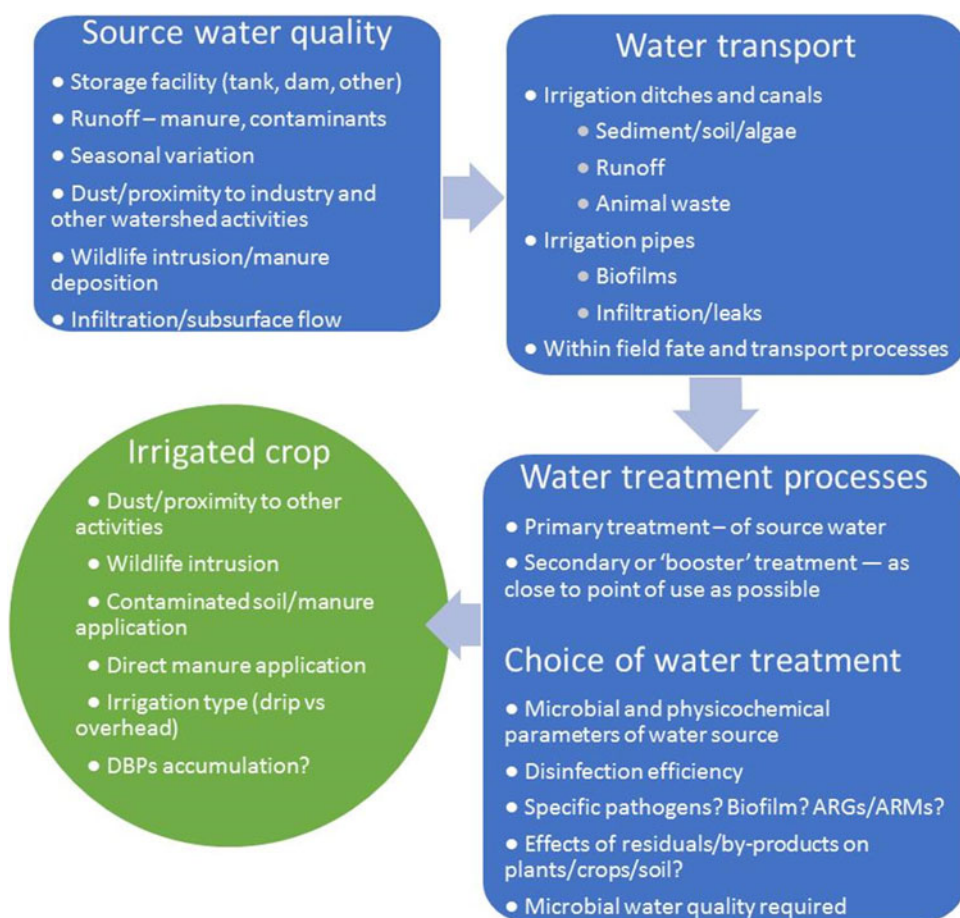


Figure 1. Factors affecting irrigation water quality and selection of water treatment processes to improve the microbial safety of fresh produce. ARGs: antimicrobial resistance genes; ARMs: antimicrobial resistant microorganisms; DBPs: disinfection byproducts.

related (Van Haute, Sampers, Jacxsens, & Uyttendaele, 2015). Technological criteria include the quality of the water source (e.g., microbiological load, temperature, pH, turbidity, suspended solids, organic matter content), distribution system characteristics, required water quality (in terms of physical and microbiological parameters), and water treatment parameters (i.e., treatment time and dose). Managerial criteria include the upfront and operational costs, complexity of operation, monitoring, and safety issues (in terms of chemical handling, storage, production of disinfection by-products (DBPs) and DBP accumulation in plants). Sustainability criteria cover maintenance, monitoring, environmental considerations and associated costs.

Generally, treatment approaches can be separated into clarification and disinfection processes. Clarification processes can be classified as follows: physical/mechanical methods, like screening, slow sand filters and

membrane filtration treatment; biological methods, such as biofilters; and chemical methods, such as coagulation and flocculation. Disinfection processes can involve the application of chemicals, such as chlorine, ozone (O_3), peroxyacetic acid (PAA), or hydrogen peroxide (H_2O_2), or might be based on non-chemical disinfection methods like ultraviolet (UV) irradiation.

Traditional treatment technologies and potential but largely untested treatment technologies for irrigation water are outlined below and the advantages and disadvantages of each process are summarized in [Tables 2](#) and [3](#). As the scientific literature about on-site disinfection of irrigation water is rather limited and generally targeted toward plant pathogens rather than human pathogens (Raudales, Parke, Guy, & Fisher, 2014), this review also draws on parallel literature and examples from other applications such as potable water and wastewater treatment when necessary.

2.1. Traditional water treatment technologies

The advantages and disadvantages of traditional water treatment technologies are summarized in [Table 2](#) and there have been several recent reviews covering many of these technologies in detail (Chahal et al., 2016; Hai, Riley, Shawkat, Magram, & Yamamoto, 2014; Hoslett et al., 2018; Jhaveri & Murthy, 2016; Kitis, 2004; Majsztrik et al., 2017; Martínez, Pérez-Parra, & Suay, 2011; Raudales et al., 2014; Scarlett et al., 2016; Yang, Li, Huang, Yang, & Li, 2016). Chlorination and UV irradiation are widely applied, mostly because of their low relative cost and convenient application.

Chlorination can be applied in gaseous form (Cl_2) or as hypochlorite (OCl^-) in either liquid or tablet form; it is well characterized, economical and effective against a broad range of pathogens. Optimum treatment conditions occur at pH 6, where the active form of undissociated hypochlorous acid is most prevalent. Hypochlorite treatment is relatively easy to implement for irrigation systems and has been widely applied in large-scale irrigation water treatment (Allende & Monaghan, 2015; Gil et al., 2015; Suslow, 2010). The disadvantages of chlorine treatment are mostly associated with the formation of DBPs, whose formation could be greater in irrigation water with high organic matter content, which would also have a high chlorine demand.

UV treatment efficacy can be substantially affected by water quality, turbidity and flow rate. Turbidity can reduce the penetration of UV irradiation, thus prefiltration or the use of thin films is required. Because of the lack of residual, there is significant potential for regrowth of pathogens after UV treatment, via photoreactivation mechanisms. UV is certified for



Table 2. Traditional water treatment technologies.

Treatment type	Mechanism	Target organism(s)/contaminants	Advantages	Disadvantages
Physical/Mechanical treatment Coagulation/flocculation	<p>Inorganic metal coagulants (i.e., aluminum sulfate or ferric chloride)</p> <p>PolymERIC coagulants</p> <p>Organic polyelectrolytes</p> <p>Composite inorganic-organic coagulants</p> <p>Natural compounds (i.e., chitosan)</p>	<p>Turbidity/suspended solids</p> <p>Organic matter</p> <p>Phosphorus</p> <p>Cryptosporidium</p> <p>Giardia</p> <p>Viruses</p> <p>Bacteria</p>	<p>Targets larger pathogens that are not targeted by or susceptible to many other disinfection processes</p> <p>Nutrient removal reduces nutrient source for pathogen growth/survival</p>	<p>Must be optimized for specific water source in terms of pH, dosage, temperature, ionic strength and treatment and settling times</p> <p>Insufficient log removal of pathogens for unrestricted irrigation</p> <p>Substantial expertise required</p> <p>Generates sludge that must be disposed of appropriately</p> <p>No residual</p> <p>Large footprint</p> <p>Clogging requires regular maintenance and filter downtime</p> <p>Insufficient log removal of pathogens for unrestricted irrigation</p> <p>May not provide sufficient throughput for large scale irrigation</p>
Slow bed sand filtration	<p>Pore size and depth of sand bed (straining and adsorption)</p> <p>Biofilm (or <i>Schmutzdecke</i>) (predation, starvation, lysis, reactive oxygen species)</p>	<p>Turbidity/suspended solids</p> <p>Bacteria</p> <p>Fungi (spores)</p>	<p>Low cost, low technology approach to provide substantial improvement in water quality</p> <p>Limited expertise required to implement and maintain</p>	<p>No residual</p> <p>Large footprint</p> <p>Clogging requires regular maintenance and filter downtime</p> <p>Insufficient log removal of pathogens for unrestricted irrigation</p> <p>May not provide sufficient throughput for large scale irrigation</p>
Membrane filtration	<p>Membrane pore size</p> <p>Microfiltration (MF: 0.1–1.0 µm)</p> <p>Ultrafiltration (UF: 5–100 nm)</p> <p>Nanofiltration (NF: 1–10 nm)</p> <p>Reverse osmosis (RO: ~0.1 nm)</p> <p>Pressure</p>	<p>Particles (MF/UF)</p> <p>Dissolved contaminants (NF/RO)</p> <p>Protozoa (MF)</p> <p>Bacteria (MF/UF)</p> <p>Viruses (UF/NF/RO)</p> <p>ARGS? (NF/RO?)</p>	<p>Removal of all contaminants possible</p> <p>Potential for contaminant-specific applications</p>	<p>No residual</p> <p>High expertise required to run and maintain</p> <p>Fouling/clogging requires backwashing, regular maintenance and membrane replacement</p> <p>Membrane failure can be catastrophic and hard to detect</p> <p>Costly to install and maintain</p> <p>No residual</p> <p>Potential for pathogen regrowth, limiting opportunity to store treated water</p> <p>Effectiveness limited in turbid water</p>
Ultraviolet (UV) irradiation	<p>Broad spectrum (i.e., UV-C 200–280 nm) or specific wavelength (i.e., 254 nm)</p> <p>DNA/RNA adsorption, production of thymine dimers, inhibition of replication, protein damage</p>	<p>Bacteria</p> <p>Viruses</p> <p>Protozoa</p> <p>Fungi</p>	<p>Widely used in drinking and wastewater treatment</p> <p>Effective against a wide range of pathogens</p> <p>Easy to install and maintain</p> <p>No DBPs</p> <p>UV-LED technology should lead to treatment innovation</p> <p>Can be combined with advanced oxidation processes</p>	<p>Costly to install and maintain</p> <p>No residual</p> <p>Potential for pathogen regrowth, limiting opportunity to store treated water</p> <p>Effectiveness limited in turbid water</p>

Disinfection processes Ozone (O_3)	Ozone (O_3) Free radicals	Bacteria Viruses Protozoa?	Generally regarded as safe for food industry use High oxidizing potential Decomposes upon exposure to oxygen	Can generate DBPs in reaction with organic matter (non-halogenated organic products, bromate) High cost of installation Residual O_3 must be removed from excess water and disposed of safely Potential phytotoxicity Potential microbial regrowth from higher organic content in effluent (acetic acid in PAA formulation and as breakdown product) High concentrations required to inactivate bacterial spores and protozoan cysts High initial purchase cost due to limited production capacity Formation of iodo-DBPs possible, which are highly cyto- and genotoxic Limited research into pre-harvest applications
Peroxyacetic acid (PAA) ($C_2H_4O_3$)	Acetic acid (CH_3COOH) Hydrogen peroxide (H_2O_2)	Bacteria Biofilms Fungi Spores Viruses Protozoan cysts	Easy to use and common for post-harvest treatment Broad-spectrum of activity and effective at low concentrations Relatively insensitive to organic loading, total suspended solids, ammonia, nitrite and phosphates No quenching (dechlorination) requirement Low formation of DBPs such as THMs and HAAs relative to chlorination Small dependence on pH; effective under a wide temperature range Strong oxidizing properties; requires short contact time	
Chlorine dioxide (ClO_2)	Chloride (Cl^-) Chlorite (ClO_2^-) Chlorate (ClO_3^-)	Multidrug-resistant bacteria Mycobacteria Protozoa Biofilms Fungi Bacterial and fungal spores Viruses	Potent across a wide pH range High oxidation capacity Does not generate DBPs such as THMs, HAAs, dioxins, furans, but can generate chlorates Does not leave odor or taste nuisance	Concerns around worker safety Efficacy is affected by high organic load and inorganic water content Concerns over transport of precursor chemicals (risk of explosion and instability) Unstable during on-site generation Sensitive to light and high temperatures Chlorate DBP generation Formation of DBPs (THMs, HAAs) in reaction with organic matter or during production (chlorate) Conc. solution (12%–15%) hazardous to workers – high pH, corrosive, burns Unstable On-site generation can produce hydrogen, an explosion hazard Potential phytotoxicity Inactivation in the presence of high organic matter
Chlorination/Hypochlorite	HOCl ClO ⁻	Bacteria Biofilms Fungi Algae Viruses	Widely used, well characterized and easy to implement Less hazardous than chlorine gas (Cl_2) Can be generated on site; reducing need for transport and handling of hazardous chemicals	

DBPs: disinfection byproducts; THMs: trihalomethanes; HAAs: haloacetic acids; LED: light emitting diode.

use in organic treatment regimens and largely used in conventional closed greenhouse systems (Dorais et al., 2016).

All currently available technologies have several advantages and disadvantages (Table 2), such that it is difficult to provide generalized recommendations. The main advantages of the physical/mechanical treatments is that they do not form DBPs; disadvantages include the lack of residual disinfectant, and the requirement for pretreatment to reduce the potential for clogging with filtration and increase the efficacy of UV treatment. The advantages of chemical sanitation treatments are that, in some cases, residual disinfection can be maintained throughout the distribution system, thus reducing the risk of pathogen regrowth. The main disadvantages of chemical sanitation treatments are 1) the formation of DBPs, which are generally formed during the reaction of oxidants with organic matter; 2) the maintenance of residual disinfection during storage; 3) the handling and transport of dangerous chemicals, and 4) the expertise required to run and maintain complex water treatment technologies. The application of chemical disinfectants requires careful monitoring and process control to ensure suitable residual disinfectant concentration and avoid phytotoxicity (Allende & Monaghan, 2015). In addition, depending on the source, irrigation water might have high organic matter content, meaning that the formation of DBPs and their potential for plant accumulation should be carefully considered when selecting treatment technologies.

Given the above, the identification and investigation of treatment technologies that 1) generate no or minimal DBPs; 2) provide some residual disinfection without resulting in phytotoxicity or increasing the risk of antimicrobial resistance; and 3) are simple to implement with no additional chemicals required, is a high priority in a world with increasing regulation of irrigation water and food production. The potential for point-of-use water treatment is also appealing, so that storage/transfer time is minimized and the water is of the highest quality directly prior to crop application in the field.

2.2. Potential irrigation water treatment technologies

We have identified several treatment technologies (Table 3) that have the potential to address some of the concerns outlined above, whilst also acknowledging that in many cases multiple treatment technologies in combination will likely be the best scenario for effective irrigation water treatment. The common feature through all of the methods outlined below is that they have an element of advanced oxidation processes, because of the generation of reactive oxygen species (ROS), particularly hydroxyl radicals (HO^\bullet), for the degradation/oxidative attack on organic material including

Table 3. Potential irrigation water treatment technologies.

Treatment type	Active constituent(s)	Target organism(s)	Advantages	Disadvantages
Hydrodynamic cavitation	Mechanical generation and subsequent collapse of vapor bubbles, causing aggressive physico-chemical environments, under high temperature and pressure	Bacteria and viruses Cyanobacterial cell disruption Removal of pharmaceuticals and pesticides Degradation of organic pollutants (e.g. textile dyes)	Can be combined with UV treatment and other AOPs Reactor design is simple, scalable and easy to operate High energy efficiency No chemicals required	Most effective under acidic conditions which favors generation of hydroxyl radicals Requires optimization of configuration for each application No residual No available research for irrigation water use
Electrolyzed oxidizing (EO) water	Hypochlorous acid (HOCl) Hypochlorite (ClO^-) Hydrogen peroxide (H_2O_2) Ozone (O_3)	Bacteria Biofilms Fungi Algae Viruses	Wide applicability in health, food, agriculture Scalability for small, medium and large applications Some EO water technologies such as NEW are pH neutral and require no hazardous chemical usage, therefore non-corrosive and non-hazardous NEW is nontoxic, the electrodes do not contain ruthenium	High initial set-up cost Inactivation in the presence of high organic matter, which might require more brine and increase cost Potential different kill rates/concentration against a variety of micro-organisms Formation of DBPs (THMs, HAAs) in reaction with organic matter
Electrochemical treatment	Hypochlorous acid (HOCl) Hypochlorite (ClO^-) Hydrogen peroxide (H_2O_2) $\bullet\text{OH}$ Ozone (O_3)	Bacteria Biofilms Fungi Algae Viruses	Easy set up Requires no hazardous chemical usage Transport, storage or dosage with chemicals is not required Disinfection strength can be adjusted according to on-site demand	High initial reactor cost Efficacy dependent on water pH, temperature, suspended solids, microbiological load, organic matter The amount of chloride ions needed will vary depending on the water quality Potential for DBPs untested Level of on-going maintenance uncertain

DBPs: disinfection byproducts; THMs: trihalomethanes; HAAs: haloacetic acids; NEW: neutral electrolyzed water; AOP: advanced oxidation process; UV: ultraviolet.

pathogenic microorganisms. The HO^\bullet molecule has the highest oxidizing potential of all oxidizing agents used in water treatment (Deng & Zhao, 2015).

2.2.1. Hydrodynamic cavitation

Hydrodynamic cavitation (HC) is a technique with a range of potential applications in water treatment and environmental remediation (Zupanc et al., 2019). First characterized in the 19th century, research has shown that HC treatment can generate localized high temperature and pressure hot spots under nearly ambient ‘bulk’ conditions. Previously, ultrasound was the main method used for producing cavitation but the adoption by industry has been poor because of the cost and extensive expertise required to operate the equipment successfully. HC is a cheaper and simpler alternative than the ultrasound-based process; the cavitation is produced by the rapid constriction and subsequent expansion of a liquid through a Venturi or orifice plates under controlled conditions (Ciriminna, Albanese, Meneguzzo, & Pagliaro, 2016; Dular et al., 2016). As the fluid flows through the constriction, HC occurs in regions where the (hydro)static pressure drops below the vapor pressure of water, causing evaporation and the formation of vapor bubbles (Figure 2). On return to regions of normal static pressure, vapor re-condenses and cavitation bubbles collapse, leading to the formation of very short lived (μs) but also very aggressive physico-chemical microenvironments characterized by very high temperature ($>1,500^\circ\text{C}$), pressure ($>69\text{ MPa}$), and turbulence (100 m s^{-1} micro jets; Tao, Cai, Huai, Liu, & Guo, 2016), all while the bulk water environment remains at ambient conditions. Reactive oxygen species (ROS; including HO^\bullet and HO_2^\bullet radicals), while generated during cavitation, can also be added (H_2O_2 , O_3) to further enhance organics removal during water treatment applications (Jusoh, Aris, & Talib, 2016; Raut-Jadhav et al., 2016; Tao et al., 2016).

Research has shown the potential beneficial uses of HC for remediation of contaminated waters, with applications including: elimination of refractory organic pollutants (Petkovšek et al., 2013; Tao et al., 2016); disinfection and pathogen destruction (Dular et al., 2016; Li, Song, & Yu, 2014; Tao et al., 2016; Torabi Angaji & Ghiaee, 2015); removal of oxyanions (As, Se) and pharmaceuticals (Zupanc et al., 2013, 2014); and recovery of base/precious metals from mine waters (Kirpalani, Singla, Lotfi, & Mohapatra, 2016).

HC can be used as a stand-alone process or in conjunction with UV (Zupanc et al., 2013), and H_2O_2 treatments (Rajoriya, Carpenter, Saharan Virendra, & Pandit Aniruddha, 2016). The main drawback of this treatment technology is the lack of residual disinfection, which might mean that it is best used in combination with another form of disinfection, or

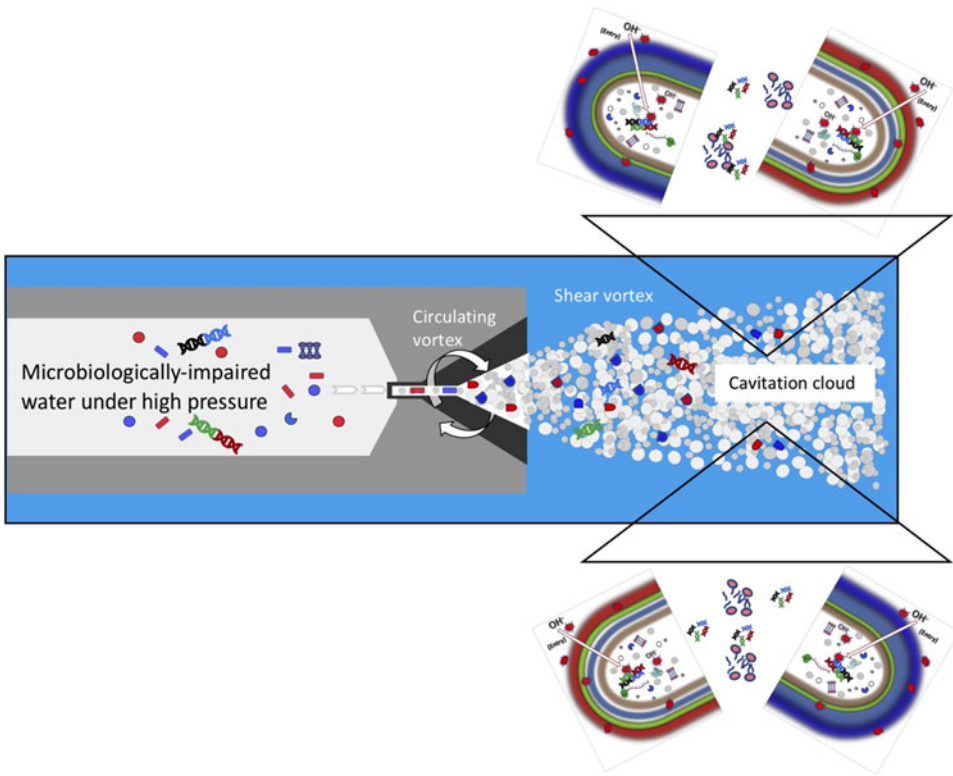


Figure 2. Principles of hydrodynamic cavitation. Formation and collapse of vapor bubbles from liquids in orifices or Venturi occur rapidly under very high temperature and high pressure changes, resulting in very high energy densities and generating hydroxyl radicals, leading to pathogen destruction.

implemented as a point-of-use water treatment. Also, given the paucity of reports in the literature, various issues such as the potential for clogging at the constriction point and the durability of the cavitation chamber need to be considered. On the other hand, the simple reactor design, easy operation, high energy efficiency and scalability have made this technology attractive for deployment (Tao et al., 2016). The review by Zupanc et al. (2019) summarized recent research on the effects of cavitation on a range of organisms, including bacteria (both Gram negative and Gram positive), cyanobacteria, algae, fungi, yeast and viruses, whilst also highlighting the many limitations of research in this area. Despite the potential of this technology, much research is required to optimize HC treatment for application to irrigation water and ensure optimal pathogen inactivation.

2.2.2. Electrolyzed oxidizing (EO) water

EO water is obtained through the electrolytic treatment of brine (water containing NaCl or KCl salts; Bakhir, 1985). In the presence of chloride,

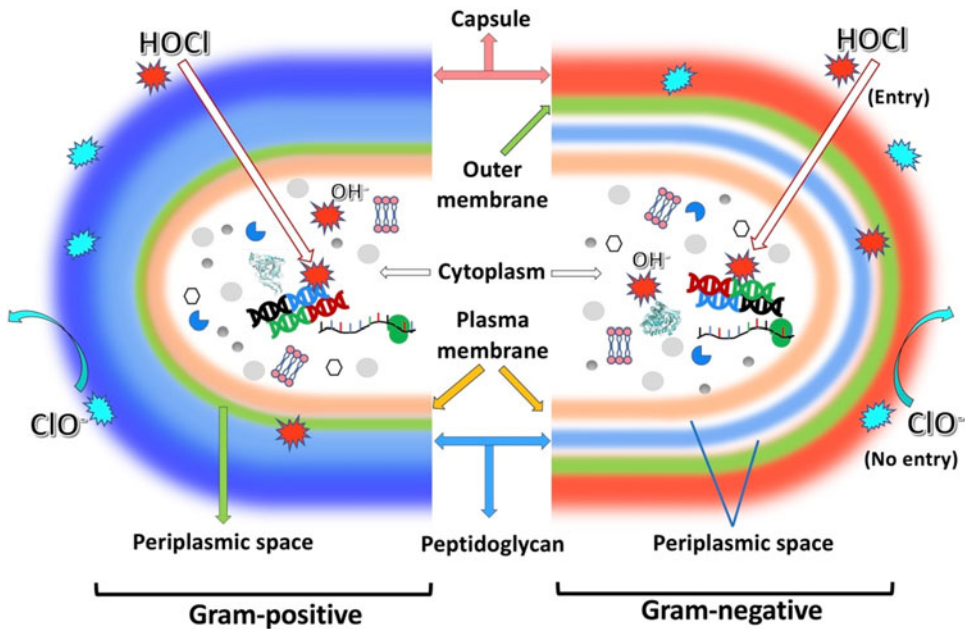


Figure 3. Schematic representation of the abilities of hypochlorous acid (HOCl) and hypochlorite (ClO^-) to kill Gram-positive and Gram-negative bacteria. The potent activity of HOCl is due to its dual cidal action on bacterial cells: HOCl is electrically neutral and can passively diffuse through the cell wall and plasma membrane into the cytoplasm where it attacks constituents including nucleic acids, proteins and lipids. HOCl is also able to directly destroy the cell wall and plasma membrane through its oxidizing action. However, ClO^- is unable to penetrate the cell and only exerts its cidal action on the bacterial cell surface.

active chlorine (sodium hypochlorite or hypochlorous acid) and ROS (O_3 , H_2O_2) are formed, which are toxic to microorganisms (Figure 3). The resulting concentrated solution ($300\text{--}500\text{ mg l}^{-1}$ active chlorine) can then be diluted into water for disinfection treatment. Electrolysis in a 2-chamber system generally results in both an acidic anolyte and an alkaline catholyte, while a 4-chamber system produces a pH-neutral anolyte, NEW (Bohnstedt, Surbeck, & Bartsch, 2009; Ferro, 2015; Migliarina & Ferro, 2014; Quadrelli & Ferro, 2010).

The main active component in the disinfection activity of EO water is free chlorine. ROS are also produced but their action is limited by their short half-life. The EO water activity will largely depend on the pH, oxidation reduction potential (ORP) and available chlorine concentration (Rahman, Khan, & Oh, 2016). Similar to traditional chlorination treatments, the optimal activity of free chlorine generated in the electrolytic process occurs when the pH of the EO water is around 6. Of the various types of EO water available, NEW (pH 6.5–7.5) is arguably the most promising as it contains predominantly HOCl. This compound is uncharged and poorly solvated by water molecules and as such it is able to penetrate

bacterial cell walls and oxidize polysaccharides (Bonfatti et al., 2000). EO waters with extremes of pH are likely to damage infrastructure and cause phytotoxicity and are therefore less suitable for agricultural applications.

Several studies have described the activity of EO waters against suspensions of target human pathogens (*E. coli*, *Salmonella* spp. and *Listeria* spp.; [Supplementary information Table S1](#)) where substantial log reductions in viable microorganisms were obtained with treatment under a range of conditions of exposure time, pH, temperature, available chlorine and ORP (Rahman et al., 2016). However, there are limited published applications of the use of EO technology in treating irrigation water. Grech and Rijkenberg (1992) found that micro-emitter-based irrigation to treat citrus root pathogens with acidic EO water at 40–50 $\mu\text{g ml}^{-1}$ active chlorine did not result in chlorine-induced phytotoxicity in field-grown plants. Similarly, the use of acidic EO water as a foliar spray (free chlorine of 54–71 mg l^{-1}) on a variety of bedding plants grown under greenhouse conditions demonstrated very little to no phytotoxicity to the plants while exhibiting rapid killing of pathogenic fungi such as powdery mildews and gray molds (Buck, van Iersel, Oetting, & Hung, 2003). Zarattini, De Bastiani, Bernacchia, Ferro, and De Battisti (2015) reported that the use of NEW at up to 500 mg l^{-1} on tobacco plants and apple trees produced no phytotoxic effects but unexpectedly triggered the molecular defenses of plants. NEW was effective at inactivating norovirus, showing >5-log reduction in suspension with NEW at 250 mg/l free chlorine, but increasing organic load or reduced NEW concentrations were less effective at reducing the viral load (Moorman, Montazeri, & Jaykus, 2017).

Similar to other chlorination treatments, organic matter has a detrimental effect on the efficacy of EO water (Jo, Tango, & Oh, 2018; Stevenson, Cook, Bach, & McAllister, 2004) and can result in the formation of DBPs, although few studies have investigated this in detail (López-Gálvez, Andujar, et al., 2018). Chlorates can also be produced during the electrolysis process itself; this can be controlled by the choice of electrode material, electrolyte composition, applied current, pH and temperature (López-Gálvez, Andujar, et al., 2018). As an alternative to traditional chlorination treatments, the technology is easy to implement and safe to use, with no dangerous chemicals required; however, the production of DBPs is still a concern and further research is required to determine the type and levels of DBPs produced and their potential accumulation in plants.

2.2.3. Electrochemical disinfection

Electrochemical disinfection is achieved by passing an electric current through the water under treatment, using suitable electrodes, without the addition of exogenous salts (Kraft, 2008). At the phase boundary between

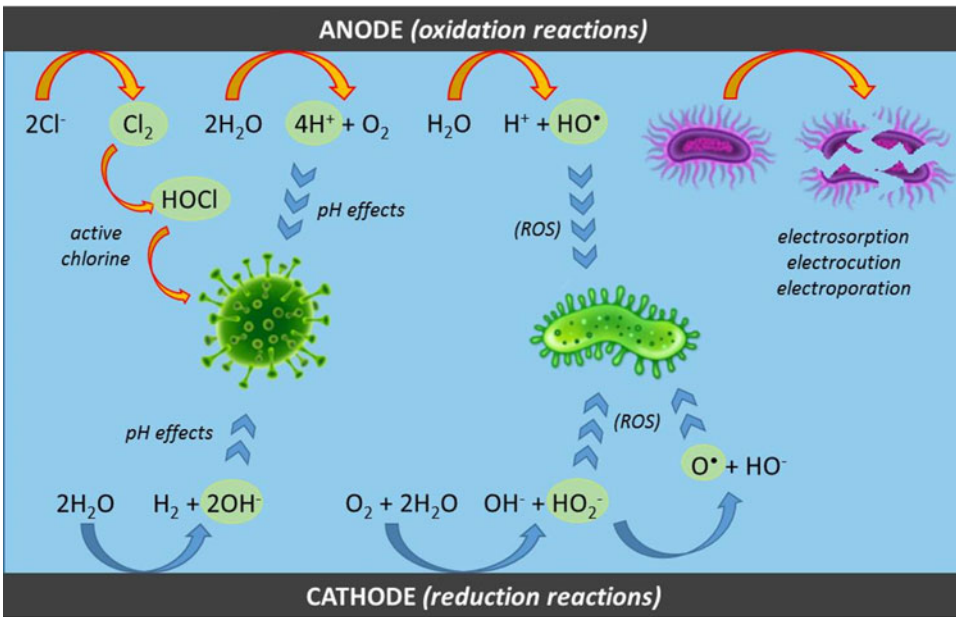


Figure 4. Reactions that occur at the anode and cathode during electrochemical disinfection of water. ROS: reactive oxygen species.

the electrodes and the water, the electric current leads to the electrochemical production of disinfecting species from the water itself (for example, ROS) or from species dissolved in the water (most notably chloride is oxidized to free chlorine; Figure 4; Kerwick, Reddy, Chamberlain, & Holt, 2005). Sufficient free chlorine can be produced to efficiently disinfect water even at low chloride concentrations (less than 100 mg l^{-1} ; Kraft, 2008). The disinfection efficacy of the electrochemical approach is thought to be higher than that of chlorination due to the formation of ROS such as hydroxyl radicals ($^\bullet\text{OH}$), atomic oxygen ($^\bullet\text{O}$), H_2O_2 , and O_3 (Delaedt et al., 2008; Diao, Li, Gu, Shi, & Xie, 2004). Yet, the short lifetime of most of the ROS in solution means that they are only active inside the electrochemical reactor. While most disinfecting agents are produced at the anode, H_2O_2 may also be produced at the cathode, as a product of oxygen reduction (Stoner, Cahen, Sachyani, & Gileadi, 1982).

The inactivation efficacy of electrochemical disinfection systems depends on several factors, including the electrochemical cell configuration, electrode material, water composition, the nature of the target microorganism, flow rate and current density (Jeong, Kim, & Yoon, 2009; Martínez-Huitle & Brillas, 2008). The main process leading to electrochemical water disinfection relies on the electrosynthesis of disinfecting agents, however other phenomena such as the electrosorption of bacteria on the electrode surface (with consequent direct interaction), electrocutation, and electroporation might play a role in the process (Matsunaga, Nakasono, Kitajima, &

Horiguchi, 1994; Matsunaga, Okochi, & Nakasono, 1995; Nakasono, Nakamura, Sode, & Matsunaga, 1992). After electrosorption, inactivation of microorganisms can result from the direct electrochemical oxidation of intracellular coenzyme-A, leading to decreased respiration and consequent cell death (Matsunaga et al., 1992). Electrochemical treatment was shown to result in oxidation of viral capsid proteins, leading to loss in structural integrity and viral inactivation (Shionoiri, Nogariya, Tanaka, Matsunaga, & Tanaka, 2015).

An interesting feature of the electrochemical disinfection approach is that the local concentration of the active agents (i.e., within the diffusion layer that forms at each electrode surface) can exceed the average concentration found in the water leaving the reactor by one or two orders of magnitude (Stoner et al., 1982). Consequently, the local concentration can be high enough to destroy highly resistant microorganisms, even if the concentration of active species in the treated water is kept at a low level. When compared with chemical disinfection methods, electrochemical water disinfection has the advantage that no transport, storage or dosage with disinfectants is required. In addition, the disinfection strength can be adjusted according to the on-site demand by adjusting the current. The technology is easy to install and could be integrated into irrigation systems where required. While electrochemical water disinfection has great potential for point-of-use irrigation water treatment, the amount of chloride ions needed, the effect of the water pH, temperature, presence of suspended solids, microbiological load, high organic matter, nature of the electrode material and the potential to produce DBPs need to be carefully evaluated. De Battisti, Formaglio, Ferro, Al Aukidy, and Verlicchi (2018) observed the formation of chlorate and perchlorate during electrochemical disinfection of groundwater, but that the concentrations of these DBPs was lower than the appropriate guideline values.

3. Other considerations in the choice of irrigation water treatment methods

There are many other issues that should be considered when choosing an appropriate irrigation water treatment method. These include potential health risks such as antimicrobial resistance and DBP accumulation, and application concerns such as cost, water quality and application methods.

The treatments described in this review have generally focused on bacterial pathogens, however, the control and treatment of other pathogen types is important. Viral pathogens such as hepatitis A and norovirus have been associated with several recent outbreaks on fresh or frozen berries and other fresh produce such as leafy greens and salads (Chatziprodromidou,

Bellou, Vantarakis, & Vantarakis, 2018). Viral pathogens can be introduced to fresh produce during preharvest operations (from contaminated irrigation water) or during postharvest manipulation (from infectious food handlers or contaminated process/washing water). Thus the role of irrigation water in virus transmission and the efficacy of the disinfection treatments investigated in this review against viral pathogens should be an important focus of future research (Hedberg, 2016).

Disinfection is a key component in successfully controlling pathogen populations in water but has also been linked in some studies to the selection of antimicrobial resistance (Rizzo et al., 2013) and reduced efficacy/increased resistance over time. Recent studies have implicated both DBPs and residual disinfectants in the induction of antimicrobial resistance and horizontal transfer of antimicrobial resistance genes (Li & Gu, 2019). Multidrug resistant opportunistic and human pathogens are an emerging worldwide threat to human health that can be transmitted through a variety of sources, including as foodborne pathogens (Baker, Thomson, Weill, & Holt, 2018). These risks should be carefully considered in the risk–benefit analysis of any proposed disinfection strategy, particularly where this is linked directly to human food.

The accumulation of DBPs in plants and potential health effects also need to be carefully considered (Dannehl, Schuch, Gao, Cordiner, & Schmidt, 2016; López-Gálvez, Andujar, et al., 2018) and this is an area that would benefit from more research. Dannehl et al. (2016) found that using potassium hypochlorite as the disinfectant in a recirculating hydroponic system, resulted in higher chlorate content in the tomatoes being grown than the current European maximum residue limit. Similarly, overhead irrigation with EO treated water resulted in accumulation of chlorates in lettuce to above the maximum residue limit (López-Gálvez, Andujar et al., 2018).

Cost is obviously an important factor in the decision-making process (Raudales, Fisher, & Hall, 2017; Van Haute et al., 2015), but because of its variability at local, national and international scale, it is difficult to draw broad conclusions. Significant research gaps also exist in terms of the practical application of water treatment to irrigation water and potential impacts in the field and beyond. The variability of irrigation water quality and quantity, crops and scale of production also makes it difficult to identify an optimal treatment arrangement that will be suitable for all potential users. For each water source and treatment configuration, the efficacy (in terms of pathogen reduction) and safety (in terms of DBPs production and/or accumulation in plants) should be independently verified to ensure compliance with the relevant guidelines. For instance, water with high turbidity might not be suitable for UV treatment; and water with high

dissolved organic matter content could be problematic for chlorine treatment because of the potential for DBPs and high chlorine demand, resulting in reduced disinfection efficacy. The irrigation method (i.e., drip *vs.* overhead) might also considerably affect the risk of pathogen or DBP uptake from treated water.

Below we provide some perspective on two important considerations for both human and plant health, which are induction of the viable but non-culturable (VBNC) state in microbial populations, and the potential effect of treated waters on soil and plant microbial communities.

3.1. Induction of VBNC microorganisms

Microbial populations can exist in a VBNC state, a survival strategy used by many Gram-positive and Gram-negative bacteria in response to adverse environmental conditions (Ferro, Amorico, & Deo, 2018; Ramamurthy, Ghosh, Pazhani, & Shinoda, 2014). There have recently been several works published investigating the potential for induction of VBNC cells during water disinfection processes (Lin, Li, Gu, Zeng, & He, 2016; López-Gálvez, Gil, Meireles, Truchado, & Allende, 2018; Zhang, Ye, Lin, Lv, & Yu, 2015). This might be of particular concern in low-quality irrigation waters, where disinfection efficacy is compromised by organic matter content or other factors. Hence, investigation of irrigation water disinfection using only conventional microbial culturing techniques might overestimate the efficacy of the disinfection treatment if VBNC organisms are not specifically considered. This is because VBNC organisms do not grow when plated on culture media that would normally support their growth *in vitro*, rendering them difficult to detect by conventional means.

VBNC microbes have lipid-rich membranes, tend to be smaller than their non-VBNC counterparts, exhibit reduced metabolic activity, and display altered cellular changes including cell leakage, depletion of energy pools, and altered gene expression and DNA replication (Arzanlou, Chai, & Venter, 2017; Trevors, Bej, Mojib, van Elsas, & Van Overbeek, 2012). Importantly, under favorable conditions (such as through expression of a resuscitation-promoting factor), these organisms can be revived. For example, it has been shown that *L. monocytogenes* treated with distilled water entered into the VBNC state and became virulent after resuscitation using embryonated eggs (Cappelier, Besnard, Roche, Velge, & Federighi, 2007). It therefore cannot be excluded that VBNC pathogens may be present in treated irrigation water and that they may become virulent again at a later stage. Furthermore, several studies have shown that pathogens may still exert detrimental effects even when in a VBNC state. For instance, laboratory-induced VBNC *E. coli* O157:H7 cells produced Shiga-like toxins

in a vero-cell microplate cytotoxicity assay, demonstrating a potential health hazard (Liu, Wang, Tyrrell, & Li, 2010). It has also been demonstrated that the VBNC state in *S. epidermidis* contributes to the formation and persistence of biofilms, resulting in tolerance to multiple antimicrobials and immune evasion (Cerca et al., 2011).

VBNC bacterial cells can be induced by many factors, including water sanitation treatments with H₂O₂ (Arana, Muela, Iriberry, Egea, & Barcina, 1992), chlorination (Oliver, Dagher, & Linden, 2005), high/low temperature (Patrone et al., 2013; Pawlowski et al., 2011), UV irradiation (Zhang et al., 2015), peroxide-based disinfectants such as PAA (Park, Lee, Bisesi, & Lee, 2014) and high-pressure CO₂ (Zhao, Bi, Hao, & Liao, 2013). A recent study showed that *E. coli* O157:H7 treated with acidic (pH 2.7–2.9 or pH 5.6–6.3) EO water could become VBNC and be resuscitated at available chlorine concentrations that resulted in no viable counts (30 mg l⁻¹; Zhang, Chen, Xia, Li, & Hung, 2018). Much higher concentrations of available chlorine (50 mg l⁻¹) were required to remove all VBNC cells. Green fluorescent protein-tagged *L. monocytogenes* and *S. enterica* Thompson became VBNC upon exposure to 12 mg l⁻¹ and 3 mg l⁻¹ chlorine, respectively (Highmore, Warner, Rothwell, Wilks, & Keevil, 2018). Thus, it is critical to investigate whether and under which conditions the various water treatment regimens induce VBNC cells in a microbial community and whether these organisms can become active again on crops or fresh produce postharvest. To fully characterize the induction of VBNC status by the various water treatment technologies, a combination of macromolecular and cellular techniques such as real-time PCR (DNA), transcriptomic (RNA) metabolic activity (protein, lipid, luminescence) measurements, fluorescence-based imaging flow cytometry, as well as morphometric analyses by transmission and scanning electron microscopy will be essential.

3.2. Effects of treated irrigation waters on soil and plant microbial communities

Soil-borne microbes constitute a major proportion of the resident organisms (the “microbiome”) identified on fruit and vegetables. The vast majority of these are not responsible for spoilage but rather act as a “natural biological barrier” against plant opportunistic pathogens, which are often a smaller subset of the entire soil microbial community (Andrews & Harris, 2000; Barth, Hankinson, Zhuang, & Breidt, 2009; Janisiewicz & Korsten, 2002). Indeed, it has been shown that an inverse relationship exists between soil microbial diversity and the survival of an invading pathogen (van Elsas et al., 2012). Hence, it is important that the irrigation with treated water does not negatively alter the microbial ecology of soils as this could directly

influence the plant microbiome (by altering the plant endophytic and phyllosphere microbial community) or indirectly by compromising organisms important for soil health (fertility and biocontrol) and thereby decreasing the health status of plants.

Many factors contribute to changes in the microbial ecology of soil, vegetables and fruits, including soil characteristics, climatic conditions and agronomic practices (Allende & Monaghan, 2015; Barth et al., 2009; Becerra-Castro et al., 2015; Berg & Smalla, 2009; Cluff, Hartsock, MacRae, Carter, & Mouser, 2014; Frenk, Hadar, & Minz, 2014; Zheng et al., 2017). Irrigation water quality also contributes to changes in microbial communities in soil and plants, especially in copiotrophic environments/ecosystems. For instance, Mañas, Castro, and de Las Heras (2009) reported significant increases in fecal streptococci, *Salmonella* spp., sulfite-reducing *Clostridium* spp. as well as total and fecal coliform counts in lettuce irrigated with minimally treated wastewater (using trickling filters), relative to control plants receiving potable water (groundwater). These findings indicate the potential deleterious effects of microbiologically impacted irrigation water on fresh produce. However, the use of tertiary water treatment regimes, such as final disinfection using UV light, chlorination and/or ultrasound, have been shown to effectively remove indicator microorganisms and pathogens to below limits of detection at the point of discharge (Pachepsky, Shelton, McLain, Patel, & Mandrell, 2011; Villanueva, Luna, Gil, & Allende, 2015). Therefore, it is critically important that good agricultural practices are implemented before, during and after harvest to maintain soil health and promote a balanced and functioning microbial community. These practices are defined in the Codex General Principles on Food Hygiene (Codex Alimentarius Commission, 2003) and aim at maximizing the quality of the crop harvested. However, a search through the literature reveals very few original manuscripts and/or reviews pertaining to changes in the microbial ecology of soil and foliar tissues after irrigation with treated irrigation water. Chevremont, Boudenne, Coulomb, and Farnet (2013) documented the changes in microbiological properties of soils irrigated with UV-LED treated wastewaters over a one-year period. When compared with watering with untreated wastewater, watering with the UV-LED treated wastewater resulted in decreased occurrence of fecal coliforms, and showed no deleterious effects on overall microbial diversity and function. Truchado, Gil, Suslow, and Allende (2018) recently investigated the effect of a low residual ClO_2 concentration (approx. 0.25 mg l^{-1}) in irrigation water on the soil microbiome and baby spinach phyllosphere bacterial community. Next generation sequencing demonstrated that while the composition of these microbiomes was not significantly altered, the relative abundance of specific bacterial genera was influenced. In particular, the relative abundance of

Pseudomonaceae and *Enterobacteriaceae* significantly decreased when the water was treated with ClO_2 .

Our overall knowledge of how the microbial ecosystems in the soil and on the surface of each produce type are influenced by the treatment of irrigation water, especially when disinfectant residues are present, is still very limited. Considering the importance of the soil and plant microbiomes to directly and indirectly control the occurrence of both human and plant pathogens, more research effort is needed in this regard.

4. Conclusions

The need to utilize water bodies and sources with sub-optimal microbiological characteristics is anticipated to increase in line with increased demand for water by the agricultural sector and society in general. In the case of fresh produce, it is of paramount importance that the microbiological quality of the water is optimized to minimize the potential for pathogen outbreaks. A significant number of treatment technologies are available for the treatment of irrigation water and they include both physical and chemical treatments. At present, the use of sodium hypochlorite and UV disinfection are widely applied because of both cost and convenience. However, other treatments such as EO water and electrochemical water disinfection (which do not require addition of chemicals) could provide interesting alternatives. Hydrodynamic cavitation should also be considered and further investigated as, in addition to not requiring chemicals due to it being a “mechanical treatment process,” it may also mitigate disinfection-induced selection of resistant bacteria (which are often pathogenic), particularly if it is proven to also destroy resistance genes and not induce the VBNC state. As noted above, however, it is generally advisable that multiple treatments are used in conjunction in high-risk settings (e.g., salad crop production), in order to ensure continuity of high water quality even in the event of total or partial failure of individual treatment barriers. We propose the concept of multistep irrigation water treatment that could be implemented for on-farm sanitation, which could vary depending on the physico-chemical parameters of the water to be treated, level of contamination and the size and cost implications of the approach to be adopted.










While there is a significant body of work on the relative efficacy of various water treatments for production of clean water, there is little direct information on the microbial profiling of irrigation water. More critically, there is little data on the effects of microbiologically impacted irrigation water on the quality of fresh produce or its effects on soil microbial communities. Direct evidence, via specific in-field experiments and advanced molecular and cellular techniques, showing the effects of the various

irrigation treatment regimens on the VBNC state as well as their effects on the dynamics of soil and microbial communities, particularly on high-risk vegetables, is warranted and paramount. Equally important is a thorough evaluation of the long-term effects and benefits of the irrigation treatment methods on soil sustainability, produce quality and overall farm productivity. Moreover, judicious implementation of environmentally-friendly treatment technologies that can effectively remove antibiotics and other antimicrobial agents, their metabolites, antimicrobial resistant microorganisms and antimicrobial resistance genes in irrigation water will improve the overall safety and value of minimally-processed foods.

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Supporting Information

Table S1: Efficacy of electrolyzed oxidizing water treatments on specific pathogens in suspension

Pathogen	Matrix/Active agent	Dose/contact time	Log reduction	Reference
<i>Escherichia coli</i>	PBS/ SAEW (20:1)	5–10 min; 60 ppm ACC; ORP +910 mV; pH 6.4; volume ratio 20:1	~8 log CFU/mL	Ye et al 2017
<i>E. coli</i>	TSB/ NEW (0.1/9.9 mL)	10 min; 20–100 ppm total residual chlorine; ORP +800-900 mV; pH 6.3– 6.5; 25°C	6.1–6.7 log CFU/mL	Guentzel et al 2008
<i>E. coli</i>	0.85% NaCl/ LcEW (1/9 mL)	1 min; 5–10 mg/L ACC; ORP +660–700 mV; pH 6.8–7.4	4.9–5.3 log CFU/mL	Rahman et al 2012
<i>E. coli</i> O157:H7	^a Culture/sterile water/NEW (1/1/8 mL)	5 min; 89 mg/L ACC; pH 7.99–8.19; ORP +745– 771 mV; 23°C	>6 log CFU/mL	Deza et al 2003
<i>E. coli</i> (range of strains)	NECAW	30 s; 100 ppm FAC; ORP +864 mV; pH 7.0	>5 log CFU/mL	Yang et al 2013
<i>Salmonella</i> (range of strains)	NECAW	30 s; 100 ppm FAC; ORP +864; pH 7.0	>5 log CFU/mL	Yang et al 2013
<i>Salmonella enteritidis</i>	Culture/sterile water/NEW (1/1/8 mL)	5 min; 89 mg/L ACC; pH 7.99–8.19; ORP +745– 771 mV; 23°C	>6 log CFU/mL	Deza et al 2003
<i>Listeria monocytogenes</i>	Culture/sterile water/NEW (1/1/8 mL)	5 min; 89 mg/L ACC; pH 7.99–8.19; ORP +745– 771 mV; 23°C	>6 log CFU/mL	Deza et al 2003

<i>Listeria monocytogenes</i> (range of strains)	PW/NECAW (1/99 mL)	30 s; 50–100 ppm FAC; ORP +824–864; pH 7.0 10 min; 150 ppm ACC;	>5 log CFU/mL	Yang et al 2013
<i>Listeria innocua</i>	Cells resuspended in NEW	ORP +840 mV; pH 6.9; 23°C	2.7 log CFU/mL	Feliciano et al 2012
<i>Listeria innocua</i>	Cells resuspended in AEW	10 min; 150 ppm ACC; ORP +1100 mV; pH 2.7; 23°C	4.7 log CFU/mL	Feliciano et al 2012
<i>Listeria monocytogenes</i>	TSB/ NEW (0.1/9.9 mL)	10 min; 20–100 ppm total residual chlorine; ORP +800–900 mV; pH 6.3– 6.5; 25°C	6.1–6.7 log CFU/mL	Guentzel et al 2008
<i>Listeria monocytogenes</i>	0.85% NaCl/ LcEW (1/9 mL)	1 min; 5–10 mg/L ACC; ORP +660–700 mV; pH 6.8–7.4	5.2–5.6 log CFU/mL	Rahman et al 2012
<i>Listeria monocytogenes</i>	0.85% NaCl/NEW (1/9 mL)	30 s; 20 ppm total chlorine concentration; ORP +1100 mV; pH 7.0; 30°C	≥5 log CFU/mL	Arevalos-Sanchez et al 2012

NEW: neutral electrolyzed water; AEW: acidic electrolyzed water; SAEW: slightly acidic electrolyzed water; CFU: colony forming unit; ACC: available chlorine concentrations; NECAW: neutral electrochemically activated water; LcEW: low concentration electrolyzed water; ORP: oxidation-reduction potential; PBS: phosphate buffered saline; TSB: trypticase soy broth; PW: peptone water

^a details of culture medium not provided.

References to Table S1:

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