A Practical Approach to Calculating Dose Values for Water Disinfection

Introduction

Clean, disinfected water is necessary to minimize the potential transmission of **pathogens** from water to produce, between produce within a lot, and between lots over time. Water-borne microorganisms, whether post-harvest plant pathogens or agents of human illness, can be rapidly acquired and taken up on plant surfaces. Natural plant surface contours, natural openings, harvest and trimming wounds, and scuffing can be points of entry for microbes. Within these protected sites, microbes are largely unaffected by common postharvest water treatments such as chlorine, chlorine dioxide, ozone, peroxide, and peroxyacetic acid. It is essential, therefore, that the water used for washing, cooling, water-mediated transporting (flumes), postharvest treatment drenches, or other water procedures be maintained with sufficient **sanitizer** to kill microbes before they can attach or become internalized in produce that is being washed. The standards for microbial quality of the water should increase as product moves from the field to final processing. However, it is important to recognize that excessive treatment, particularly hyperchlorination (use of high levels of chlorine), has several known or potential negative **product sensory**, environmental and human health impacts. We, therefore, find it desirable to optimize water treatment to minimize the effective dose for microbial **disinfection**. These minimum effective doses are typically represented as a combination of *C*oncentration and *T*ime of exposure, or *Ct*. In this note, we introduce a different term, using the same principles, to help guide our management of water quality: D_h, or **Disinfection Hurdle**.

Chlorine and Hypochlorite (Bleach) Treatment

Ease of use and relative low cost makes hypochlorite (usually liquid sodium hypochlorite) a very common water treatment in the produce industry. The **antimicrobial activity** of chlorine compounds depends largely on the amount of hypochlorous acid (HOCl) present in the water. This, in turn, depends on the pH of the water, the amount of organic material in the water and, to a more limited extent, the temperature of the water. Above pH 7.5, very little (<50%) chlorine exists as active HOCl, but rather as far less active hypochlorite (OCl^-). With very long contact time, OCl- does have some antimicrobial activity, but this would not be expected to result in beneficial control in typical postharvest handling systems. Below pH 6.0, noxious chlorine gas (Cl_2) is formed. Of the many possible forms of chlorine, HOCl is the form most readily transferred across a microbial cell wall to begin the killing process. Thus, in our management of chlorine, we want to maximize HOCl concentrations and minimize all the other forms of chlorine. Therefore, it is highly desirable to keep the pH of the water between 6.0 and 7.5 to ensure adequate activity without the formation of chlorine gas, which is irritating to workers and more **corrosive** to equipment.

The amount of HOCl needed to maintain the most active antimicrobial action in water, which comes into contact with produce, depends on several dynamic factors. Chlorine is very **reactive**, combining with almost any oxidizable substrate to form secondary compounds. The amount of chlorine needed for disinfection of water depends on the pH as well as amounts and kinds of inorganic (particularly ammonia, nitrites, iron, and manganese) and organic materials (particularly amino acids and simple proteins) present in the water. Because chlorine is rapidly used up by organic and inorganic molecules in the wash water, a minimum level of chlorine, the “chlorine demand”, must first be satisfied in the water before sufficient amounts of “free available chlorine” will be present to kill microorganisms. The required treatment times of fruits and vegetables in water at **peak chlorine demand** (generally caused by soil, plant “trash”, and exudates from cut surfaces) are usually very short. To minimize this “chlorine demand” in recirculating systems, periodically replacing or filtering water and blending with **potable water** is important.

Calculating Required HOCl Additions

In clean water, very low levels of HOCl will inactivate most bacteria and some viruses, with 1 to 2 ppm for less than 2 minutes being sufficient contact time. As water quality decreases and complexity increases, contact time or concentration must increase to maintain adequate microbial kill. In general, contact times during postharvest handling are relatively fixed and are determined by product flow requirements. Therefore, we generally work with the concentration of added disinfectant. Effective concentrations of HOCl (or other forms of chlorine) must be determined and validated within each system. We recognize that water quality management is perceived as a time consuming and costly venture; however, we strongly recommend a high level of attention to this area.

As a starting point, the following simple calculations can be made based on an adaptation of tables developed for treatment of water to achieve potable water quality standards. The goal is to determine the minimum concentration of "free available chlorine" as HOCl in wash or cooling water that is needed to kill free-floating pathogenic bacteria and viruses. Higher levels or other treatments would be needed for the more resistant *Bacillus* or *Clostridium* spores and parasites like *Giardia* and *Cryptosporidium*.

From the table below, we calculate the verifiable concentration of HOCl for the contact time of our system that, taken together, establishes the effective delivered dose.

Example: Based on the contact time of our system, we calculate the HOCl concentration necessary for an effective dose. For washing and cooling, our system has a 5-minute residence time (t) and we have a water pH that remains around 7.5 to 7.8 without adjustment when product is running through the system. Our water temperature is maintained at 34-38°F (1.1 to 3.3°C). From the table we know that the Disinfection Hurdle, D_h, is 20. Using these known values in the following equation we can solve for C, our minimal HOCl concentration.

$$\boxed{C = D_h / t \text{ or } 20 \div 5 = 4 \text{ mg /L (4 ppm)}}$$

Using this result, we can measure free chlorine using a titration kit or colorimeter specific for "free available chlorine". At pH 7.8 and 34-38 °F, only 50% of the measured "free available chlorine", by the commonly used methods, is in the desired HOCl form. Therefore, a minimum reading of 8 ppm is needed to hit the target.

Remember, the calculated amount may be very different from the actual dose of total hypochlorite solution added to the system at peak demand. The target D_h will be determined by the sensitivity of the most resistant microbe you are trying to manage (*Erwinia* soft rot bacteria and *E. coli* are relatively sensitive, *Geotrichum* Sour Rot and *Rhizopus* are much more resistant). This has to be determined by direct testing until sensitivity charts for important decay fungi that are calibrated to this system of calculating effective doses become available.

It is easy to see the impact of water pH on C. In the same example, adding citric acid to maintain pH 7.0, C = 2.75 **ppm** (HOCl is 87% of free chlorine at pH 7.0). If the pH increases to 8.5 during hypochlorite treatment, a common outcome, then C = 41 ppm (HOCl is 17.5% of free chlorine at pH 8.5).

It is important to remember that test kits for measuring 'free available chlorine' are only suitable for concentrations up to 4 ppm. It will be necessary to dilute any treated water with distilled water to bring it into a measurable range. Typical dilutions are 1:10, although a dilution of 1:100 may be necessary where control of fungal spores is important. Always follow the instructions provided by the test kit supplier.

It is easiest to adjust and standardize water pH to 6.5 to 7.0, making the majority of free available chlorine in the HOCl form. Consulting a second table of pH and temperature becomes unnecessary. Measurements of HOCl in water may also be adequately determined by using a calibrated ORP (oxidation reduction potential) sensor. In our experience, 4-6 ppm HOCl, as in the first example, typically gives a sensor reading of 725-750 millivolts.

Table 1: Guidelines for Meeting the Disinfection Hurdle* in Postharvest Water Treatment

Water pH range	Value of $\boldsymbol{D_h}$ in $C \times t = D_h$	
	32 – 41°F	50°F
7.0 - 7.5	12	8
7.5 – 8.0	20	15
8.0 – 8.5	30	20
8.5 – 9.0	35	22

- Values given are the product of concentration of HOCl and time of exposure of a diversity of microbes in water to achieve greater than 99% kill. The value *t* is determined by the specific process or operation and assumes adequate mixing to accomplish uniform exposure.

Table 2 : Current Projected Value of D_h in Postharvest Water at pH 7.0

Target microorganism	32 – 41°F	Typical Contact Time (min)
Non-spore-forming Bacteria	3 - 6	1-5
Many Viruses	3 - 10	1-5
Many Yeast	75 - 100	10 - 30
Spore-forming Bacteria	150 - 250	15 - 60
Fungal Spores	150 - 500	15 – 60
Parasite Spores		
Giardia	30 – 100	5 – 10
Cryptosporidium	Highly Tolerant	Use UV or Ozone

Tables are modified from White, 1992 and reflect results from our laboratory and field research experience.

GLOSSARY OF TECHNICAL TERMS:

Antimicrobial activity: The effectiveness with which a sanitizer or disinfectant can kill microorganisms.

Corrosive: Elements, in this case chemical, that has the power to weaken or eat away at equipment, especially metal.

Disinfection: The act of adding or applying a sanitizer in order to kill microorganisms that may cause decay in produce or illness in humans.

Disinfection hurdle: A descriptive concept term that symbolizes the minimum effective exposure to achieve microbial kill. Disinfection is one of several hurdles in a prevention, reduction, and contamination control program. The “disinfection hurdle” will be different for different types or classes of microorganisms.

Pathogens: Microorganisms such as bacteria, fungi, parasites, and viruses that can cause disease in humans or plants.

Peak chlorine demand: The time in a batch of water when material, inorganic and organic, will “use up” the maximum amount chlorine that has been added. After the peak chlorine demand is known, it can be better established how much *more* chlorine or *more* clean water should be added to maintain target disinfectant concentrations. Additional steps, such as minimizing adhering soil, a pre-wash, or filtration may be necessary to reduce the chlorine demand .

Potable water: Water that is clean enough to be considered drinkable.

ppm: Parts per million.

Product sensory: Characteristics of a product, in this case fresh produce, related to smell, taste, appearance and texture.

Reactive: A chemical that is especially reactive is one that doesn’t stay in one form for very long. In the case of chlorine, it is important that chlorine stay in a particular form in order to be effective, so the reactivity of chlorine is of particular interest.

Sanitizer: A chemical that is added or applied, in this case to water, in order to kill pathogens. A surface or water may be sanitized, free of pathogens, but not sterile, free of microorganisms.

Sensitivity: A system or test’s sensitivity refers to the lowest concentration that the system can detect or respond to. For example, if a chlorine test system can only detect concentrations of chlorine at or higher than 1 ppm, the system’s senstivity is said to be at 1 ppm.

Additional Background Information: There are numerous articles and scientific research journal papers and reviews that relate to this broad topic. A few lay-technical introductions and one comprehensive book that are easy to access are listed below.

Suslow, T. 1997. Postharvest Chlorination: Basic Properties and Key Points for Effective Sanitation. DANR PUB # 8003 at http://vric.ucdavis.edu

Suslow, T. 1998. Introduction to ORP as the standard of postharvest water disinfection monitoring. at http://vric.ucdavis.edu

Suslow, T. 1998. Prevention of postharvest water infiltration into fresh market tomatoes: Food safety and spoilage control practices. at http://vric.ucdavis.edu

White, G.C. 1992. Handbook of Chlorination and Alternative Disinfectants. 3rd Edition. New York: Van Nostrand Reinhold, 1308 pp.

Prepared by Trevor V. Suslow, Ph.D.
Postharvest Specialist
Postharvest Quality and Safety from Seed to Shelf
One Shields Ave.
University of California
Dept. of Vegetable Crops, Mann Lab
530-754-8313
530-752-4554 fax
tvsuslow@ucdavis.edu
http://vric.ucdavis.edu
http://postharvest.ucdavis.edu