Guideline for Disinfection and Sterilization in Healthcare Facilities, 2008

William A. Rutala, Ph.D., M.P.H.[1,2], David J. Weber, M.D., M.P.H.[1,2], and the Healthcare Infection Control Practices Advisory Committee (HICPAC)[3]

[1]Hospital Epidemiology
University of North Carolina Health Care System
Chapel Hill, NC 27514

[2]Division of Infectious Diseases
University of North Carolina School of Medicine
Chapel Hill, NC 27599-7030

Chlorine and Chlorine Compounds

***Overview*.** Hypochlorites, the most widely used of the chlorine disinfectants, are available as liquid (e.g., sodium hypochlorite) or solid (e.g., calcium hypochlorite). The most prevalent chlorine

products in the United States are aqueous solutions of 5.25%–6.15% sodium hypochlorite (see glossary), usually called household bleach. They have a broad spectrum of antimicrobial activity, do not leave toxic residues, are unaffected by water hardness, are inexpensive and fast acting [328], remove dried or fixed organisms and biofilms from surfaces[465], and have a low incidence of serious toxicity [515-517]. Sodium hypochlorite at the concentration used in household bleach (5.25-6.15%) can produce ocular irritation or oropharyngeal, esophageal, and gastric burns [318, 518-522]. Other disadvantages of hypochlorites include corrosiveness to metals in high concentrations (>500 ppm), inactivation by organic matter, discoloring or "bleaching" of fabrics, release of toxic chlorine gas when mixed with ammonia or acid (e.g., household cleaning agents) [523-525], and relative stability [327]. The microbicidal activity of chlorine is attributed largely to undissociated hypochlorous acid (HOCl). The dissociation of HOCl to the less microbicidal form (hypochlorite ion OCl^-) depends on pH. The disinfecting efficacy of chlorine decreases with an increase in pH that parallels the conversion of undissociated HOCl to OCl^- [329, 526]. A potential hazard is production of the carcinogen bis(chloromethyl) ether when hypochlorite solutions contact formaldehyde [527] and the production of the animal carcinogen trihalomethane when hot water is hyperchlorinated [528]. After reviewing environmental fate and ecologic data, EPA has determined the currently registered uses of hypochlorites will not result in unreasonable adverse effects to the environment [529].

Alternative compounds that release chlorine and are used in the health-care setting include demand-release chlorine dioxide, sodium dichloroisocyanurate, and chloramine-T. The advantage of these compounds over the hypochlorites is that they retain chlorine longer and so exert a more prolonged bactericidal effect. Sodium dichloroisocyanurate tablets are stable, and for two reasons, the microbicidal activity of solutions prepared from sodium dichloroisocyanurate tablets might be greater than that of sodium hypochlorite solutions containing the same total available chlorine. First, with sodium dichloroisocyanurate, only 50% of the total available chlorine is free (HOCl and OCl^-), whereas the remainder is combined (monochloroisocyanurate or dichloroisocyanurate), and as free available chlorine is used up, the latter is released to restore the equilibrium. Second, solutions of sodium dichloroisocyanurate are acidic, whereas sodium hypochlorite solutions are alkaline, and the more microbicidal type of chlorine (HOCl) is believed to predominate [530-533]. Chlorine dioxide-based disinfectants are prepared fresh as required by mixing the two components (base solution [citric acid with preservatives and corrosion inhibitors] and the activator solution [sodium chlorite]). In vitro suspension tests showed that solutions containing about 140 ppm chlorine dioxide achieved a reduction factor exceeding 10^6 of *S. aureus* in 1 minute and of *Bacillus atrophaeus* spores in 2.5 minutes in the presence of 3 g/L bovine albumin. The potential for damaging equipment requires consideration because long-term use can damage the outer plastic coat of the insertion tube [534]. In another study, chlorine dioxide solutions at either 600 ppm or 30 ppm killed *Mycobacterium avium-intracellulare* within 60 seconds after contact but contamination by organic material significantly affected the microbicidal properties[535].

The microbicidal activity of a new disinfectant, "superoxidized water," has been examined The concept of electrolyzing saline to create a disinfectant or antiseptics is appealing because the basic materials of saline and electricity are inexpensive and the end product (i.e., water) does not damage the environment. The main products of this water are hypochlorous acid (e.g., at a concentration of about 144 mg/L) and chlorine. As with any germicide, the antimicrobial activity of superoxidized water is strongly affected by the concentration of the active ingredient (available free chlorine) [536]. One manufacturer generates the disinfectant at the point of use by passing a saline solution over coated titanium electrodes at 9 amps. The product generated has a pH of 5.0–6.5 and an oxidation-reduction potential (redox) of >950 mV. Although superoxidized water is intended to be generated fresh at the point of use, when tested under clean conditions the disinfectant was effective within 5 minutes when 48 hours old [537]. Unfortunately, the equipment required to produce the product can be expensive because parameters such as pH, current, and redox potential must be closely monitored. The solution is nontoxic to biologic tissues. Although the United Kingdom manufacturer claims the solution is noncorrosive and nondamaging to endoscopes and processing equipment, one flexible endoscope manufacturer (Olympus Key-Med, United Kingdom) has voided the warranty on the endoscopes if superoxidized water is used to disinfect them [538]. As with any germicide formulation, the user should check with the device manufacturer for

compatibility with the germicide. Additional studies are needed to determine whether this solution could be used as an alternative to other disinfectants or antiseptics for hand washing, skin antisepsis, room cleaning, or equipment disinfection (e.g., endoscopes, dialyzers) [400, 539, 540]. In October 2002, the FDA cleared superoxidized water as a high-level disinfectant (FDA, personal communication, September 18, 2002).

Mode of Action. The exact mechanism by which free chlorine destroys microorganisms has not been elucidated. Inactivation by chlorine can result from a number of factors: oxidation of sulfhydryl enzymes and amino acids; ring chlorination of amino acids; loss of intracellular contents; decreased uptake of nutrients; inhibition of protein synthesis; decreased oxygen uptake; oxidation of respiratory components; decreased adenosine triphosphate production; breaks in DNA; and depressed DNA synthesis [329, 347]. The actual microbicidal mechanism of chlorine might involve a combination of these factors or the effect of chlorine on critical sites [347].

Microbicidal Activity. Low concentrations of free available chlorine (e.g., HOCl, OCl^-, and elemental chlorine-Cl_2) have a biocidal effect on mycoplasma (25 ppm) and vegetative bacteria (<5 ppm) in seconds in the absence of an organic load [329, 418]. Higher concentrations (1,000 ppm) of chlorine are required to kill *M. tuberculosis* using the Association of Official Analytical Chemists (AOAC) tuberculocidal test [73]. A concentration of 100 ppm will kill ≥99.9% of *B. atrophaeus* spores within 5 minutes [541, 542] and destroy mycotic agents in <1 hour [329]. Acidified bleach and regular bleach (5,000 ppm chlorine) can inactivate 10^6 *Clostridium difficile* spores in ≤10 minutes [262]. One study reported that 25 different viruses were inactivated in 10 minutes with 200 ppm available chlorine [72]. Several studies have demonstrated the effectiveness of diluted sodium hypochlorite and other disinfectants to inactivate HIV [61]. Chlorine (500 ppm) showed inhibition of *Candida* after 30 seconds of exposure [54]. In experiments using the AOAC Use-Dilution Method, 100 ppm of free chlorine killed 10^6–10^7 *S. aureus*, *Salmonella choleraesuis*, and *P. aeruginosa* in <10 minutes [327]. Because household bleach contains 5.25%–6.15% sodium hypochlorite, or 52,500–61,500 ppm available chlorine, a 1:1,000 dilution provides about 53–62 ppm available chlorine, and a 1:10 dilution of household bleach provides about 5250–6150 ppm.

Data are available for chlorine dioxide that support manufacturers' bactericidal, fungicidal, sporicidal, tuberculocidal, and virucidal label claims [543-546]. A chlorine dioxide generator has been shown effective for decontaminating flexible endoscopes [534] but it is not currently FDA-cleared for use as a high-level disinfectant [85]. Chlorine dioxide can be produced by mixing solutions, such as a solution of chlorine with a solution of sodium chlorite [329]. In 1986, a chlorine dioxide product was voluntarily removed from the market when its use caused leakage of cellulose-based dialyzer membranes, which allowed bacteria to migrate from the dialysis fluid side of the dialyzer to the blood side [547].

Sodium dichloroisocyanurate at 2,500 ppm available chlorine is effective against bacteria in the presence of up to 20% plasma, compared with 10% plasma for sodium hypochlorite at 2,500 ppm [548].

"Superoxidized water" has been tested against bacteria, mycobacteria, viruses, fungi, and spores [537, 539, 549]. Freshly generated superoxidized water is rapidly effective (<2 minutes) in achieving a 5-log_{10} reduction of pathogenic microorganisms (i.e., *M. tuberculosis*, *M. chelonae*, poliovirus, HIV, multidrug-resistant *S. aureus*, *E. coli, Candida albicans*, *Enterococcus faecalis, P. aeruginosa*) in the absence of organic loading. However, the biocidal activity of this disinfectant decreased substantially in the presence of organic material (e.g., 5% horse serum) [537, 549, 550]. No bacteria or viruses were detected on artificially contaminated endoscopes after a 5-minute exposure to superoxidized water [551] and HBV-DNA was not detected from any endoscope experimentally contaminated with HBV-positive mixed sera after a disinfectant exposure time of 7 minutes[552].

Uses. Hypochlorites are widely used in healthcare facilities in a variety of settings. [328] Inorganic chlorine solution is used for disinfecting tonometer heads [188] and for spot-disinfection of countertops and floors. A 1:10–1:100 dilution of 5.25%–6.15% sodium hypochlorite (i.e., household bleach) [22, 228, 553, 554] or

an EPA-registered tuberculocidal disinfectant [17]has been recommended for decontaminating blood spills. For small spills of blood (i.e., drops of blood) on noncritical surfaces, the area can be disinfected with a 1:100 dilution of 5.25%-6.15% sodium hypochlorite or an EPA-registered tuberculocidal disinfectant. Because hypochlorites and other germicides are substantially inactivated in the presence of blood [63, 548, 555, 556], large spills of blood require that the surface be cleaned before an EPA-registered disinfectant or a 1:10 (final concentration) solution of household bleach is applied [557]. If a sharps injury is possible, the surface initially should be decontaminated [69, 318], then cleaned and disinfected (1:10 final concentration) [63]. Extreme care always should be taken to prevent percutaneous injury. At least 500 ppm available chlorine for 10 minutes is recommended for decontaminating CPR training manikins [558]. Full-strength bleach has been recommended for self-disinfection of needles and syringes used for illicit-drug injection when needle-exchange programs are not available. The difference in the recommended concentrations of bleach reflects the difficulty of cleaning the interior of needles and syringes and the use of needles and syringes for parenteral injection [559]. Clinicians should not alter their use of chlorine on environmental surfaces on the basis of testing methodologies that do not simulate actual disinfection practices [560, 561]. Other uses in healthcare include as an irrigating agent in endodontic treatment [562] and as a disinfectant for manikins, laundry, dental appliances, hydrotherapy tanks [23, 41], regulated medical waste before disposal [328], and the water distribution system in hemodialysis centers and hemodialysis machines [563].

Chlorine long has been used as the disinfectant in water treatment. Hyperchlorination of a *Legionella*-contaminated hospital water system [23] resulted in a dramatic decrease (from 30% to 1.5%) in the isolation of *L. pneumophila* from water outlets and a cessation of healthcare-associated Legionnaires' disease in an affected unit [528, 564]. Water disinfection with monochloramine by municipal water-treatment plants substantially reduced the risk for healthcare–associated Legionnaires disease [565, 566]. Chlorine dioxide also has been used to control *Legionella* in a hospital water supply. [567] Chloramine T [568] and hypochlorites [41] have been used to disinfect hydrotherapy equipment.

Hypochlorite solutions in tap water at a pH >8 stored at room temperature (23°C) in closed, opaque plastic containers can lose up to 40%–50% of their free available chlorine level over 1 month. Thus, if a user wished to have a solution containing 500 ppm of available chlorine at day 30, he or she should prepare a solution containing 1,000 ppm of chlorine at time 0. Sodium hypochlorite solution does not decompose after 30 days when stored in a closed brown bottle [327].

The use of powders, composed of a mixture of a chlorine-releasing agent with highly absorbent resin, for disinfecting spills of body fluids has been evaluated by laboratory tests and hospital ward trials. The inclusion of acrylic resin particles in formulations markedly increases the volume of fluid that can be soaked up because the resin can absorb 200–300 times its own weight of fluid, depending on the fluid consistency. When experimental formulations containing 1%, 5%, and 10% available chlorine were evaluated by a standardized surface test, those containing 10% demonstrated bactericidal activity. One problem with chlorine-releasing granules is that they can generate chlorine fumes when applied to urine [569].