

mination of minimum inhibitory concentrations (MICs), which may prove useful when studying the effects of antibiotics in the human body but should not be extrapolated to environmental surface disinfection on the macroscopic level. To do so is misleading and erroneous. Fawley et al.¹ state that the mean *C. difficile* sporulation capacity was significantly increased by exposure to neutral detergent, to a combination of detergent and hypochlorite, and to hydrogen peroxide, but the sporulation assay that is described in the article *does not simulate actual facility cleaning and disinfection practices*. The method assumes 72 hours of contact with a highly diluted solution of each germicide tested. However, none of these germicides are intended or directed for use over an extended time period in an extremely diluted form. Thus, the 72 hour incubation period for a combination of the highly diluted germicide and *C. difficile* does not replicate actual practice. Since it is not clearly communicated to clinical readers that the sporulation observed with the tested germicides in this study does not come close to replicating actual clinical conditions, infection control professionals may overlook or stop utilizing products that would fight *C. difficile* safely and effectively.

The message that should be made very clear is that the chlorine-containing germicides, including the sodium hypochlorite-based disinfectant, were shown to inactivate *C. difficile* spores when used at recommended working strength, and these types of germicides should be employed in health-care facilities when *C. difficile* is a problem.

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Reply to Holschlag

TO THE EDITOR—Holschlag¹ has failed to understand the findings of our article² and appears to be suggesting that we were not clear in our experimental methods. This is incorrect. To evaluate the efficacy of germicides and/or cleaning agents against *Clostridium difficile*, we used 3 different measures: the capacity to inhibit vegetative cells, the capacity to prevent spore germination, and the potential to promote sporulation. We clearly stated the concentrations of germicides and cleaning agents that were used in these different experiments; the subinhibitory concentrations that were used in the sporulation experiments are made clear in the Methods section and are reinforced in both the legend to Figure 2 and the Discussion section.

Holschlag¹ questions the relevance of the results that we obtained in our sporulation experiments using subinhibitory concentrations. We addressed this point in the Discussion section, which highlights the potential of environmental stresses (including drying, exposure to air, and exposure to cleaning agents and/or germicides) to influence sporulation. Holschlag¹ failed to point out that our experimental design included the use of fecal emulsions, to test the effects of germicides and/or cleaning agents on *C. difficile* spores. In so doing, we attempted to test the effects of exposure to feces, dilution of the germicides and/or cleaning agents, and prolonged contact with spores, all of which are entirely plausible conditions that may occur in the clinical setting. In many instances in hospitals today, some environmental surfaces are cleaned infrequently or only “terminally.” Even if done frequently for a patient with diarrhea (who will have from several to 10 or more explosive voluminous bowel movements per day), it is likely that environmental cleaning solution will on occasion comprise a mixture of residual waning disinfectant and fecal material from cumulative explosive diarrheal episodes. Thus, our study was intended to at least represent what can occur in practice (eg, rehabilitation, long-term care, and acute care facilities, as well as in the use of physical therapy equipment and stretchers used to transport patients). The results of our study may explain, in part, the rise in rates of *C. difficile* infection over the last decade in North America and in several European countries. We would argue for (at least) daily cleaning and appropriate disinfection for all hor-

izontal surfaces in the rooms of patients with *C. difficile* infection.

It would be obtuse to assume that *in vitro* experiments precisely simulate *in vivo* conditions. Similarly, relying solely on an antimicrobial agent's minimum inhibitory concentration to predict its clinical efficacy against a particular infecting pathogen is ill-advised. Thus, it is standard practice to employ *in vitro* models of infection to simulate, among other factors, waning concentrations of an antimicrobial agent after each dose is administered. The impact of cleaning and disinfecting agents should be viewed no differently. Initial working strength concentrations applied to surfaces do, in fact, wane over time. Subsequently, in germicides and/or cleaning agents, the residual active components are exposed to organic material (eg, feces containing *C. difficile* in both vegetative and spore forms). If working strength concentrations were universally delivered after their initial application, it would be surprising if the environment was ever implicated in the spread of infection. We caution against assuming that use of a germicide or a cleaning agent guarantees effective environmental decontamination; it does not. Thus, it has been shown that as the level of environmental contamination with *C. difficile* increases, so does the magnitude of healthcare worker hand contamination.³

In our article,² we acknowledged that the clinical significance of results showing an increased rate of sporulation associated with use of some cleaning agents and/or germicides is unknown. However, as pointed out by Holtschlag,¹ the US Environmental Protection Agency does not currently recognize a test method for inactivation of *C. difficile* spores. It is logical, therefore, to use different test methodologies and to base any conclusions concerning the potential efficacy of agents against *C. difficile* on all of the results obtained. This is what we did. It would be unwise to pick and choose which results appear more favorable, particularly, as in Holtschlag's case, if there is a potential conflict of interest. Hence, we concluded our report by stating that "the combined body of evidence suggests that dichloroisocyanurate (ie, chlorine-release) germicides currently represent the optimum choice for the removal of *C. difficile* from healthcare environments."^{2(p924)} We went on to say that our results "suggest that compounds that do not kill *C. difficile* spores at working concentrations, such as general-purpose detergents and hydrogen peroxide, may promote the persistence and accumulation of spores in healthcare environments."^{2(p924)} We stand by these comments.

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Should Test Methods for Disinfectants Use Vertebrate Viruses Dried on Carriers to Advance Virucidal Claims?

TO THE EDITOR—The advancements made in microbicidal science in the past decade have raised questions about the appropriateness of the test methods still being used to substantiate microbicidal and virucidal claims globally. The test methods currently being used to evaluate the virucidal activity of disinfectants employ challenge virus that is either dried on prototypical hard surfaces or is in suspension. The latter approach presents a weaker challenge to the formulation that is being tested.^{1,2} Regulatory agencies such as the US Environmental Protection Agency, Canadian General Standard Board, and Australian Therapeutic Goods Administration require that data for virucidal activity be based on carrier test methods that use vertebrate viruses.³⁻⁶ In contrast, European Norms for claims about virucidal activity (both BS EN 14476:2005 and EN 13610) require suspension tests, although EN 13610 specifies bacteriophages, as opposed to vertebrate viruses.^{7,8} We believe that the requirements of both of these European Norms are unrealistic and do not represent field situations where disinfectants are used for decontamination of pathogens dried on hard surfaces in domestic, health care, or extended care settings.

In this letter, we comment on the irrelevance of both these standards (BS EN 14476:2005 and EN 13610:1999) on the basis of our 20 years of experience as manufacturers of microbicidal products and also as developers of methods to test