The Effectiveness of Chlorine Dioxide in Inactivating Influenza Virus

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ABSTRACT

National outbreaks of avian influenza viruses have been the source of a wide range of adverse effects for the country, including the culling of thousands of birds, economic damage to commercial farms, increased product prices, and blocked international trade. Tennessee is heavily involved with the poultry industry and an outbreak of avian influenza could have serious economic impact. Currently, a standardized system to control and prevent the spread of the pathogenic viruses from entering chicken farms is lacking. This study evaluated the chemical compound chlorine dioxide to inactivate influenza viruses. Seven different strains of influenza were exposed to chlorine dioxide gas and tested at concentrations ranging from 50 parts per million to 500 ppm at time intervals ranging from 30 minutes to 4 hours. As the concentration of chlorine dioxide increased, the length of exposure necessary to bring the virus titer to zero decreased. At 500 ppm, virus was completely inactivated by 30 minutes.

INTRODUCTION

This study was conducted to investigate a potential preventative measure against type A influenza viruses. By June 2015, 22 states had confirmed cases of a high pathogenic avian influenza (HPAI) virus, many of these cases occurring in poultry (1). These viruses have a 90-100% mortality rate in untreated poultry flocks (2). A chicken with avian influenza is seen in Fig 1.

Figure 1. Effect of HPAI on poultry.

Type A viruses are characterized by the proteins on their surface, the Hemagglutinin and Neuraminidase (Fig 2).These proteins are pivotal for infection and spread of the virus by binding to the host cell. Chlorine dioxide inhibits the hemagglutinin protein by changing its conformation, making it unable to bind to the host cell (3). If the hemagglutinin protein is unable to bind, infection cannot occur.

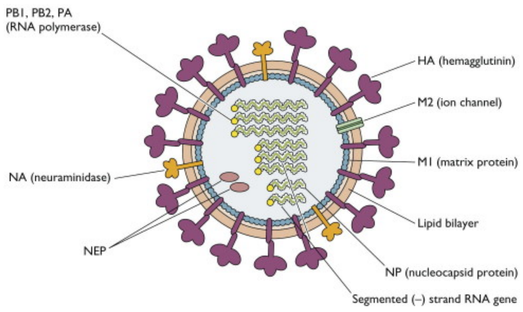


Figure 2: Influenza virus surface membrane proteins.

Seven strains of influenza, 2 avian, 2 human, and 3 swine strains, were used in our trials. A range of chlorine dioxide concentrations and exposure times were evaluated. We determined that chlorine dioxide was very effective at inhibiting all of the tested strains.

MATERIALS AND METHODS

1. Embryonated chicken eggs were infected with viruses to make viral stocks
2. Hemagglutination assays (Fig 3, below) were performed to check viral titers and dilutions were made to get each virus to a standard concentration of 16 (approximately 16 million viruses/mL)

Figure 3. Preparation of hemagglutination assay.

3. Trials with chlorine dioxide were carried out for each virus under the set conditions:
   a. Times of exposure—30 min, 1 hour, 2 hours, 4 hours
   b. Concentration—50 ppm, 100 ppm, 500 ppm
   c. Fig 4 shows experimental apparatus

4. Hemagglutination assays were performed again to determine if viral concentration was reduced and percent reduction calculations were made

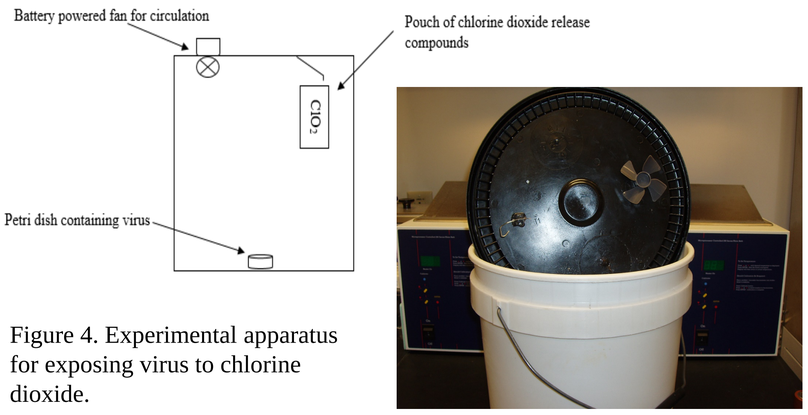


Figure 4. Experimental apparatus for exposing virus to chlorine dioxide.

RESULTS

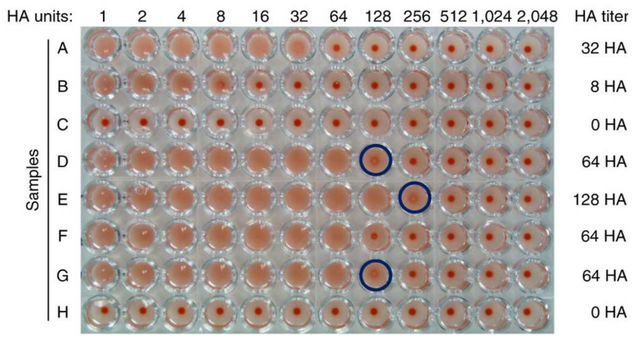
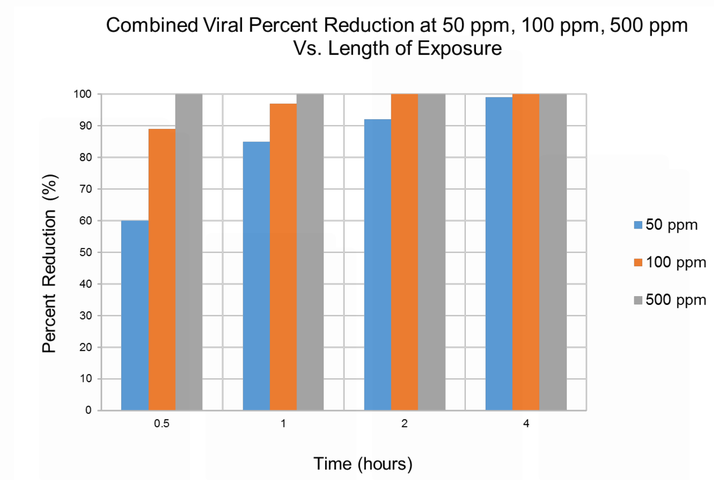
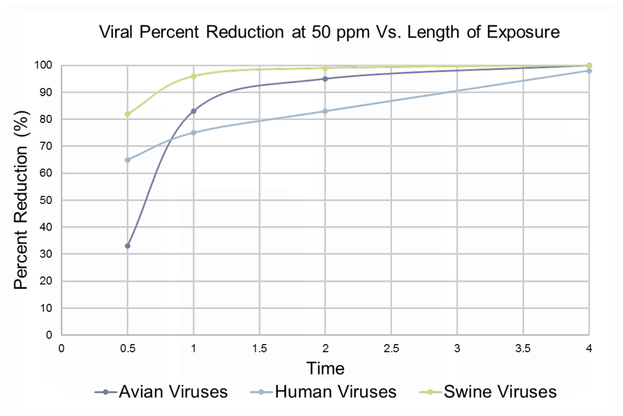


Figure 5. Assay for virus.

Figure 5 above shows results of a hemagglutination assay. The last well with the blood cells in suspension is the virus titer. A red dot indicates the absence of virus.
Chlorine dioxide completely inactivated all influenza strains within 30 minutes at the highest concentration of 500 ppm (Fig 6, below).



Four hours of exposure were required to reduce virus levels below detectable limits when chlorine dioxide was at the lowest concentration of 50 ppm, seen in Fig 7 below. Avian viruses were more resistant than human and swine influenza viruses at 50 ppm.



DISCUSSION

While the inhibitory effects of chlorine dioxide on influenza has been described (3), this is the first study to evaluate different influenza strains against varying concentrations of chlorine dioxide and times of exposure. Based on our results, chlorine dioxide has the potential to be a highly effective way to inactivate influenza viruses and perhaps prevent additional outbreaks among poultry.
Inhalational studies with rats, reported by the EPA, have shown toxic effects with long term exposure (4). Thus, while chlorine dioxide holds promise in relation to threats posed by avian influenza, additional studies with poultry should be undertaken. Chlorine dioxide may best be used to restore a poultry house to a virus-free state following evidence of influenza infection, providing a cost-effective solution to a serious problem.

LITERATURE CITED

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