Antiviral Effect of Chlorine Dioxide against Influenza Virus and Its Application for Infection Control

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Abstract: Influenza is a respiratory tract infection, causing pandemic outbreaks. Spanish flu (A/H1N1), a pandemic occurred between 1918 and 1919, tolled patients and fatalities of 500 million and 50 million, respectively. Recently, human infection with highly pathogenic avian influenza A/H5N1 and swine influenza [Pandemic (H1N1) 2009] was reported. Because of the population explosion and busy global aircraft traffics, Pandemic (H1N1) 2009 is rapidly spreading worldwide. In addition, it is seriously concerned that H5N1 influenza pandemic would emerge in the very near future. The pandemic will cause the freeze of social activity and the crisis of business continuity, having a serious impact on the global economy consequently. It is fervently desired that efficient methods of infection control against influenza pandemic be developed.

Chlorine dioxide (ClO_2) has a strong antiviral effect, and can disinfect the surface of object and the air in space. In recent study on interaction between ClO_2 and protein, ClO_2 oxidatively modified tyrosine and tryptophan residues, and the protein was structurally denatured. Since hemagglutinin and neuraminidase of influenza virus A/H1N1 were inactivated by the reaction with ClO_2, it is likely that denaturation of the proteins caused inactivation of the virus. A low concentration (0.03 ppm) of ClO_2 gas, where people can stay for a long period of time without any harmful effect, prevented the death of mice caused by infection of influenza virus delivered as aerosol. We review current information based on the efficiency of ClO_2 solution and gas, and also discuss the application of ClO_2 against influenza pandemics outbreak.

Keywords: Chlorine dioxide, influenza, pandemic.

1. INTRODUCTION

We, human being, experienced pandemics caused by influenza virus many times in the history. In the 20th century, three pandemics occurred in 1918, 1957, and 1968. Each pandemic was named as Spanish flu, Asian flu, and Hong Kong flu, among which the subtype of influenza A virus differs as H1N1, H2N2, and H3N2, respectively [1]. The Spanish flu (A/H1N1) pandemic occurred between 1918 and 1919 with three waves, recorded more than the 500 million infected and 50 million deaths, which subsequently reduced the lethality with antigen drift, then it was settled as a seasonal influenza [2]. Thirty eight years after the settlement of the Spanish flu, by antigen shift, the Asian flu (A/H2N2) erupted and became a pandemic once again. The Asian flu also became a seasonal flu and was settled, however, 10 years later, by antigen shift, the Hong Kong flu (A/H3N2) occurred and became a pandemic, which, by antigen drift, became a seasonal influenza and was settled once again. Through the pandemics in the 20th century, it is obvious that, by antigen shift that occurs every few decades, a new subtype of virus was generated and caused a pandemic. Also, it is thought that all these pandemics were originated from the Spanish flu A/H1N1 subtype [3].

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In this new century, a pandemic caused by the infection of influenza virus occurred once again [4]. Since April 2009, an influenza derived from the porcine type A influenza virus (H1N1) originated in Mexico has spread worldwide almost instantly, and resulted in the declaration by the World Health Organization (WHO) of a pandemic on June 11, 2009, 41 years after the Hong Kong flu [5]. According to a surveillance by WHO as of November 22, 2009, the new type influenza [Pandemic (H1N1) 2009] tolled over 622,482 diagnosed infected patients and at least 7,826 deaths, and the infection is still spreading worldwide [6]. The geographical breakdown of the number of diagnosed patients are: 15,503 in African region, 190,765 in American region, 38,359 in the East Mediterranean region, over 154,000 in European region where the spread of infection is still ongoing in Ukraina, 47,059 in Southeast Asian region, and 176,796 in Western Pacific region, indicating a rapid transmission of infection worldwide [6]. Since these numbers are those with a positive result of examination of Pandemic (H1N1) 2009 influenza virus, it is assumed that, among the huge number of influenza-like illnesses (ILIs), a considerable portion of Pandemic (H1N1) 2009 influenza patients are included. It may be said that this is a real example of a long concerned worldwide spread of infection in the contemporary world where the way of transportation by airplane and railroad, etc. is highly developed.

In May 1997, prior to the Pandemic (H1N1) 2009 influenza, a human infection of a highly pathogenic avian influ-

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enza A/H5N1 virus was reported [7, 8], which has been continuously spreading. Influenza A (H5N1) is infected by a direct contact with sick or dead poultry, and a large number of human deaths has been reported. Between November 2003 and December 2007, 349 cases in 14 countries were infected [9], among whom 62% died [10]. Recently, in Egypt, incidence of the influenza A (H5N1) is increasing, and among the 88 accumulated patients, 27 died [11]. So far, the infectivity of the influenza A (H5N1) to human seems relatively low, however, if a mutant virus which is more adaptable to human should emerge, it is easy to expect that the infection spreads worldwide almost instantly, like the case of the Pandemic (H1N1) 2009 influenza. Since the lethality of the influenza A (H5N1) is alarmingly high, it is concerned that there will be a huge number of deaths cases, unprecedented health hazards, and subsequent corruption of social functions as a whole.

In this paper, we focus on chlorine dioxide (ClO_2) which has a potent virucidal activity against influenza virus, and introduce the confirmative results of antiviral effects in the *in vitro* experiments. In addition, we report the details of preventive effect of ClO_2 gas in influenza infection experiments in mice, as well as a recently reported ameliorating effect on the cumulative school absenteeism. Finally, we propose new preventive measures against the flu pandemic utilizing ClO_2 in addition to normal preventive methods.

2. CHLORINE DIOXIDE

ClO_2 (CAS. No. 10049-04-4) is a molecule with the molecular weight of 67.46, and it forms a stable radical [12]. Dauy, a British chemist, first discovered this molecule in 1811, by oxidizing potassium perchlorate [13]. ClO_2 is yellowish gas at room temperature, and relatively easily dissolved in water [solubility in water is 20 g/L (0-5°C and 70-100 mmHg)] [13]. However, since ClO_2 gas dissolved in water dissipates within a short period of time, the dissolved level of ClO_2 gas decreases as time passes [13]. Consequently, ClO_2 solution has been used by generation on site.

ClO_2 is an oxidizer, which is reduced to chlorite ion (ClO_2^-) by capturing an electron ($ClO_2 + e^- \rightarrow ClO_2^-$). The redox potential ($E^o$) is relatively high as 0.95 V. ClO_2^-, in the presence of water, captures four electrons and is reduced to chloride ion (Cl^-)($ClO2^- + 2H_2O + 4e^- \rightarrow Cl^- + 4OH^-$), whose E^o is 0.78 V which is lower than that of ClO_2. Consequently, the oxidative activity of ClO_2 is stronger than ClO_2^-. The E^o of ClO_2 is lower than that of hydroxyl radical ($^{\cdot}OH$, 2.8 V), ozone (O_3, 2.07 V), and hypochlorous acid (HClO, 1.49 V), therefore, it undergoes more selective oxidative reaction than these other molecules [14, 15].

So called "stabilized" chlorine dioxide, which, in fact, is colorless solution of ClO_2^- as the principal ingredient, has been on the market in the Western countries, which is causing a confusion of exact nomenclature [16, 17]. Generally speaking, ClO_2 is synthesized by the reaction formula of (1) - (3) as below [13, 18] ($_{(g)}$ means gas).

$$2NaClO_2 + Cl_{2(g)} = 2ClO_{2(g)} + 2NaCl \quad (1)$$

$$2NaClO_2 + HOCl = 2ClO_{2(g)} + NaCl + NaOH \quad (2)$$

$$5NaClO_2 + 4HCl = 4ClO_{2(g)} + 5NaCl + 2H_2O \quad (3)$$

As a result, the "so called" stabilized chlorine dioxide in which ClO_2^- is the principal ingredient, cannot produce ClO_2 if chlorine gas, hypochlorous acid or acid does not coexist, and the "so called" stabilized chlorine dioxide itself does not possess the selective oxidative activity of ClO_2. Therefore, the efficacy data of the "so called" stabilized chlorine dioxide on microorganisms differ from those of ClO_2 itself, and they need to be carefully interpreted [19-21].

ClO_2 may be used as gas as well as solution, therefore, the field of its application is considerably wide. Since long time ago, principally in the Western countries, ClO_2 has been utilized for the bleaching of paper pulp [22]. ClO_2, which is different from Cl_2, oxidizes lignin selectively among various kinds of carbohydrates existing in the pulp [23, 24]. In addition, ClO_2 has an advantage of reducing the production of dioxins compared with other chlorine type bleaching agents. Therefore, ClO_2, by virtue of its ECF (Elemental Chlorine Free) property without producing dioxins, serves as an alternative to the bleaching by Cl_2 [22]. Also, ClO_2 is allowed by the U.S. Food and Drug Administration (FDA) to be used as a food additive for bleaching of cereal flours [25] and disinfection of vegetables and fruits [26]. Furthermore, the U.S. Environmental Protection Agency (EPA) has allowed ClO_2 to be used as a disinfectant of tap water (less than 0.8 ppm for targeted water quality) [27], and the molecule is exploited as a disinfection method which produce little amount of trihalomethane, a carcinogen, at water purification plants in the U.S.A. [28, 29]. In addition, the EPA has allowed a high concentration ClO_2 gas as a sterilizer for manufacturing and experimental apparatus, clean rooms, environmental surfaces, and tools [30], and recently, the gas was standardized by Annex G of NSF/ANSI 49 as an alternative to formalin fumigation for the sterilization of biosafety cabinets [31]. As will be described later, we confirmed a preventive effect of the low concentration of ClO_2 gas against the influenza virus infection [32]. In the future, we hope that the legal legislation not only for high concentration ClO_2 gas but also for low concentration ClO_2 gas as a preventive measure of infection will take place.

In these current circumstances for ClO_2, we at Taiko Pharmaceutical Co., Ltd. manufacture and market a series of uniquely developed antimicrobial and deodorant consumer products under the brand name of "Cleverin®" in Japan, utilizing ClO_2 [33]. At present, as a products lineup, we have "Cleverin®G" series which disseminate low concentration of ClO_2 gas continuously, "Cleverin®S" and "Cleverin®L" which maintain the ClO_2 concentration (approximately 100 ppm and 500 ppm, respectively) stably in solution for a long period of time. These products look yellowish which is a characteristic of ClO_2 as describe formerly, and exploit ClO_2 gas itself. Also, "Cleverin®S" is a ready-to-use product, without necessitating the ClO_2 gas generation on site. Furthermore, in addition to these products, we manufacture and market LISPASS®, a device which continuously releases low concentration (below 0.1 ppm) of ClO_2 gas. There are two types of LISPASS®: LISPASS®S is a small size portable type for the floor area up to 200 m^2. The other is LISPASS® NEO, which can cover the large floor area up to 16,000 m^2. Recently, we conducted a real size room experiment, and confirmed that, although the level of released ClO_2 gas is affected by humidity and lighting, the apparatus can run

within the safety level below 0.03 ppm [34]. Also, as to the efficacy against viruses, bacteria, and fungi as well as the safety of our ClO_2 products, we have published the research results in the world leading journals [32, 35-39]. In the following section, among these various research results, we focus on the influenza infection, introduce the efficacy data of ClO_2, and, together with the safety, we overview them. Also, we introduce a totally new concept of "three phase protection" to control the influenza infection.

3. POSITIVE EFFICIENCY OF CHLORINE DIOXIDE AGAINST INFLUENZA VIRUS AND INFENCTIOUS DISEASE

3.1. Chlorine Dioxide Solution

There are many reports that ClO_2 solution has a virucidal activity [40-44]. The inactivation concentration against various viruses are: 1-2 ppm in poliovirus [40, 41], 2.19 ppm in coronavirus which causes SARS [42], 7.5 ppm in hepatitis A virus [43], and 0.2 ppm in rotavirus [44]. Recently, we used ten types of viruses and reported the antiviral activity of Cleverin®S and L (hereinafter referred to as Cleverin solution) [36]. The Cleverin solution, at 10 ppm ClO_2 concentration, showed 99.99% or greater antiviral activity against feline calicivirus, human influenza virus A, measles virus, canine distemper virus, human herpesvirus-1, human herpesvirus-2, human adenovirus type 2, canine adenovirus type 2, canine parvovirus and human immunodificiency virus type 1 [36]. Furthermore, under various conditions of the Cleverin solution, we measured the antiviral activity against influenza virus A, and reported the results in comparison with sodium hypochlorite solution [45]. Therefore, in this article, we describe the comparative antiviral activity of the Cleverin solution and sodium hypochlorite solution.

First, in the comparative antiviral activity experiment loaded with fetal bovine serum (FBS), the Cleverin solution, even in the presence of 1% FBS as an interfering organic compound, ClO_2 at 10 ppm showed 99.999% or greater antiviral activity against influenza virus A (Table **1**). On the other hand, under the same condition, 100 ppm of sodium hypochlorite solution was necessary to achieve the same antiviral activity (Table **1**). Therefore, it was shown that, under the serum loaded condition, the Cleverin solution was ten times more effective than sodium hypochlorite solution. Furthermore, since the Cleverin solution containing 100 ppm ClO_2 showed 99.999% or greater antiviral activity under the condition of 10% FBS loading, this product may well show the antiviral activity in the contaminated household environment.

Next, when we compared the antiviral activity in the presence of the high concentration of salt (3 M or greater of NaCl) against influenza virus A, the Cleverin solution at 10 ppm, and sodium hypochlorite solution at 100 ppm showed the antiviral activity (Table **2**). Therefore, same as the FBS loading, in the presence of high concentration of salt, the efficacy of the Cleverin solution was ten times higher than that of sodium hypochlorite solution.

Furthermore, when we compared the antiviral activity at various temperatures (4-50°C) against influenza virus A, the Cleverin solution at 10 ppm and sodium hypochlorite solution at 100 ppm showed the antiviral activity (Table **3**). At various pH (5-6) ranges, the Cleverin solution at 1 ppm and sodium hypochlorite solution at 10 ppm showed the antiviral activity (Table **4**). When we increased the ClO_2 concentration of the Cleverin solution up to 10 ppm, 99.99% of antiviral activity was observed up to pH 10.

Table 1. Relation between Disinfectant Concentration and FBS Added to Inactivate ≥99.999% Influenza A Virus without Added NaCl for 1 min at 25°C

Disinfectant Name	[Disinfectant] (ppm)	[FBS] (%)
Cleverin	1	ND[a]
	10	1
	100	10
NaClO	10	NI[b]
	100	1

[a]Not done, [b]Not inactivated.

Table 2. Relation between Disinfectant Concentration and NaCl Added to Inactivate ≥99.999% Influenza A Virus without Added FBS for 1 min at 25°C

Disinfectant Name	[Disinfectant] (ppm)	[NaCl] (M)
Cleverin	1	1
	10	>3
	100	>3
NaClO	10	NI[a]
	100	>3

[a]Not inactivated.

Table 3. Relation between Disinfectant Concentration and Reaction Temperature to Inactivate ≥99.999% Influenza A Virus without Added FBS and NaCl for 1 min

Disinfectant Name	[Disinfectant] (ppm)	Temperature (°C)
Cleverin	1	10-50
	10	4-50
	100	4-50
NaClO	10	NI[a]
	100	4-50

[a]Not inactivated.

From these results, we confirmed that the Cleverin solution used in the case of contact infection of influenza virus showed ten times higher antiviral activity than sodium hypochlorite solution which is a chlorine type bleaching agent, and its activity as a virucidal agent was not significantly af-

fected by the presence of organic compounds, salt concentration, temperature, and pH.

Table 4. Relation between Disinfectant Concentration and Reaction pH to Inactivate ≥99.99% Influenza A Virus without Added FBS and NaCl for 1 min at 25°C

Disinfectant Name	[Disinfectant] (ppm)	pH
Cleverin	1	5-6
	10	5-10
	100	ND[a]
NaClO	1	NI[b]
	10	5-6
	100	ND[a]

[a]Not done, [b]Not inactivated.

3.2. Chlorine Dioxide Gas

In addition to the Cleverin solution, we focused on the potent virucidal activity of ClO_2 gas, and are extending unique research activities to confirm its efficacy and also to elucidate its mechanism of action. As to the infection of influenza virus, we used an animal (mice) model of infection experiment, and found that ClO_2 gas far below the eight hour average weighted mean exposure level (0.1 ppm) defined by the Occupational Safety and Health Administration (OSHA, U.S.A.) suppressed the infection of influenza virus A [32]. We housed 15 mice (CD-1) in a semi-closed cage, and introduced ClO_2 gas (final average concentration: 0.032 ppm) for 15 minutes simultaneously introducing the aerosol of influenza virus strain A/PR/8/34 (H1N1) (approximately 1 LD_{50}) generated by a nebulizer (treated group). As the control group, fresh air instead of ClO_2 gas was introduced. Three days later, we picked up five mice and measured the pulmonary virus titer ($TCID_{50}$). In the control group, $TCID_{50}$ was 6.7 ± 0.2 log (mean ± SD), while in the treated group, it was significantly reduced to 2.6 ± 1.5 log (P = 0.003, Table **5**). Also, when we compared the mortality of the remaining ten mice until day 16, in the control group, it was 7/10 (number of dead animals/total number of animals), whereas, in the treated group, it was significantly reduced to 0/10 (P = 0.002, Table **5**). From these results, it was suggested that as low as 0.03 ppm of ClO_2 gas prevented the influenza infection in the mouse model.

In addition to this animal model, a retrospective human study revealed that the low concentration ClO_2 gas decreased the cumulative absenteeism of primary school children [35]. When three Cleverin®Gs were placed in a primary school classroom (space volume 230 m^3) for the purpose deodorization (0.01-0.03 ppm), in classes where Cleverin®G was not placed, the cumulative absenteeism rate during 38 consecutive days was 4.0%, while it was reduced to 1.5% in a classroom where Cleverin®Gs were placed (P < 0.00001, Table **6**). The primary reason of absenteeism was cold or influenza. From these experiments, it was suggested that the low concentration of ClO_2 gas prevented the infection of influenza in a semi-closed space.

3.3. Virucidal Mechanism of Chlorine Dioxide

As a mechanism of action of antiviral effect of ClO_2, denaturation of surface proteins, such as hemagglutinin (HA) and neuraminidase (NA), was postulated. Therefore, we investigated the effect of various concentrations of ClO_2 on the activity of HA and NA. As a result, it was revealed that, in accordance with the increase in the ClO_2 level, the activity of HA and NA was decreased [32]. Furthermore, a detailed physicochemical analysis was performed to reveal the interaction of protein and ClO_2, utilizing bovine serum albumin (BSA) and glucose-6-phosphodehydronase (G6PD) as model proteins [46]. As a result, an analysis by nuclear magnetic resonance (NMR) spectroscopy and high performance liquid chromatography-mass spectrometry (HPLC-MS) revealed that ClO_2 performed an oxidative modification of tyrosine residue and tryptophan residue in the protein, and changed the tyrosine residue to 3,4-dihydroxyphenylalanine (DOPA) and 2,4,5-trihydroxyphenylalanine (TOPA), and the trypto-

Table 5. Effect of ClO_2 Gas on Pulmonary Titer, Mortality and Body Mass of Mouse Challenged with Influenza A Virus

[ClO_2 gas] (ppm)	Pulmonary Virus Titer[a] (log_{10})	Mortality[b]	Relative Body Mass[c]
0	6.7±0.2[d]	7/10[e]	0.9±0.04[e]
0.03	2.6±1.5[d]	0/10[e]	1.09±0.08[e]

[a]Virus titer ($TCID_{50}$) was measured 72 hours after a challenge of virus aerosols (n = 5 in each group). [b]Mortality was measured until 16 days after the challenge (n = 10 in each group). [c]Ratio of body mass on day 7 to that on day 0 in each group (n = 5 in each group). [d]P = 0.003, [e]P = 0.002.

Table 6. Effect of ClO_2 Gas on Cumulative Absenteeism Rate

Cleverin® G	Cumulative Attendance Rate (%)	Cumulative Absenteeism Rate (%)
Placed	98.5	1.5[a]
Not placed	96.0	4.0[a]

[a]P < 0.00001.

phan residue to *N*-formylkynurenine. Furthermore, in the peptide derived from HA and NA, we observed that the tyrosine residue and tryptophan residue were oxidized [32]. Therefore, it was suggested that the mechanism of action of ClO_2 to inactivate the influenza virus was specific oxidative modification of these two amino acid residues. Both tyrosine residue and tryptophan residue are contained in HA and NA in Pandemic (H1N1) 2009 virus and highly pathogenic influenza A/H5N1 virus [47-50], suggesting the antiviral effect of a low concentration of ClO_2 gas and ClO_2 solution.

4. SAFETY OF CHLORINE DIOXIDE

4.1. Chlorine Dioxide Solution

ClO_2 has been used to disinfect the tap water in the U.S.A., and a review article was published by EPA [51]. Hereinafter, we extracted the content of this report.

As a result of oral administration of ClO_2 solution for 90 days in rats, the lowest-observed-adverse-effect level (LOAEL) was 25 ppm (2 mg/kg/day). Also, from another experiment in which ClO_2 solution was orally administered for two years in rats, the no-observed-adverse-effect level (NOAEL) was 10 ppm (1.3 mg/kg/day) [51-53]. In an experiment in which African green monkeys were orally administered with ClO_2 solution for six weeks, NOAEL was 30 ppm (3.5 mg/kg/day) and LOAEL was 100 ppm (9.5 mg/kg/day) [51, 54]. Also, a human study was conducted and there was no toxic signs observed after oral intake of tap water containing 5 ppm of ClO_2 for 12 weeks [51, 55]. As to the metabolism of ClO_2, an investigation was made by a single oral administration of radiolabeled $^{36}ClO_2$ in rats, which revealed that the plasma ^{36}Cl level reached the peak two hours after the administration, and 30% of the radioactivity was excreted in urine by 72 hours after the administration [51, 56]. Approximately 80% of ^{36}Cl were in the form of Cl^-, and most of the remaining radioactivity was ClO_2^-, with a small amount of ClO_3^- [57].

We also confirmed the toxicity of Cleverin solution in animals, resulting in the acute oral toxicity more than 5,000 mg/kg, and the inhalation toxicity was more than 12,000 mg/m^3. No dermal irritation was observed and the mucosal irritation was minimal (Table **7**). These experimental results suggest that the safety of both ClO_2 solution and the Cleverin solution containing ClO_2 is relatively high, and may be used in households.

4.2. Chlorine Dioxide Gas

Dalhamn performed an inhalation toxicity study in rats [58]. Some rats died after the exposure of as high as 260 ppm of ClO_2 gas for two hours. On the other hand, after the exposure of ClO_2 gas at 0 or 0.1 ppm (average level during ten weeks, ranging from 0.05 to 0.3 ppm) for five hours a day and seven days a week for ten weeks, neither death nor any toxic signs were observed. Our ClO_2 products have an advantage of controlling the concentration of ClO_2 below 0.1 ppm in the space. This low concentration is lower than the weighted average of eight hours per day allowed by OSHA as the long term exposure, the weighted average of ten hour labor allowed by the National Institute for Occupation Safety and Health (NIOSH, U.S.A.) [59], and the standard level for eight hours a day for 40 hours a week allowed by the American Conference of Governmental Industrial Hygienist (ACGIH, U.S.A.) [60]. From these findings, it may be assumed that there is little toxicity concern about the use of the low concentration of ClO_2 gas below 0.1 ppm.

5. THREE PHASE PROTECTION FOR INFECTION CONTROL OF INFLUENZA

In order to prevent the infection in offices, hospitals, and household rooms, disinfection of the hard surface may not be sufficient to control all the infection routes, and an application which can be applied to every infection route becomes necessary. It is said that the main infection routes of influenza virus to human being are physical contact with the infected individual or *via* the intermediate materials (direct contact infection) and inhalation of droplets containing the virus and exhausted from the infected individual (droplet infection). As to the infection caused by the dried droplets or the aerosol with the size of 5 μm or smaller containing the

Table 7. Toxicology Study of Cleverin Solution[a]

Test	Animal	Result
Acute oral toxicity	Mouse	LD50 > 5000 mg/kg
Acute inhalation toxicity	Mouse	LC50 > 12000 mg/m^3
Single dose skin irritation	Rabbit	No irritation
Multiple dose skin irritation	Rabbit	No irritation
Ophthalmological eye irritation	Rabbit	No irritation
Single dose mucosal irritation	Rabbit	Minimal irritation
Subchronic oral toxicity	Rat	LD50 > 1000 mg/kg
Skin allergic reaction	Guinea pig	No allergic reaction
Micronucleous test	Mouse (bone marrow)	No induction

[a] ClO_2 concentration: 100 ppm.

virus (airborne infection), although a lot of discussion have been made, the conclusion is yet to be reached [61-65]. However, the infection experiments in Guinea Pigs [66] and Ferrets [67] confirmed the airborne infection *via* aerosol. Also, influenza virus was detected in the aerosol sample taken from the ambulatory hospital [68]. Therefore, the existence of influenza virus in the air and the airborne infection by the virus cannot be denied. Hence, we proposed an idea of the three phase protection as a measure to prevent the infection of influenza virus. This concept of infection control system is consisted with the physical surface disinfection against the contact infection (the first phase protection), disinfection of the space against the droplet and airborne infection (the second phase protection), and the control of infection and growth of virus taken into the body (the third phase protection). Examples of the physical protection are the frequent hand washing, the physical shielding of the infection route by wearing a mask or a protective cloth, and the hard surface disinfection of the contact infection routes [69]. For the hard surface disinfection, we can utilize Cleverin®S and Cleverin®L which show highly efficient antiviral effects. As to the space protection, examples are the physical removal of the virus by HEPA filter, and the chemical removal of the virus by gas with a virucidal effect. For the chemical removal of the virus, the choice of Cleverin®G, LISPASS®S, and LISPASS®NEO, all of which generate ClO_2 gas, may be recommended according to the size of the space to be disinfected. For the human body protection, examples are the therapy by administration of anti-influenza drugs [70], the prevention by vaccination [71, 72] and the high dose vitamin D administration which is reported to be effective [73].

Pandemic (H1N1) 2009 influenza is expected to further spread worldwide. Also, highly pathogenic influenza A (H5N1) infection keeps increasing in Egypt [11]. Furthermore, it is not unlikely that a new type of influenza virus may emerge by antigen shift and antigen drift of these two viruses and acquire high morbidity and mortality. We believe firmly that, under the concept of the three phase protection, both the ClO_2 solution and the low concentration of ClO_2 gas which can be used effectively and safely will become an effective measure to prevent not only the current influenza infection but also the new emerging influenza virus.

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