International Journal on Infectious Disease and Epidemiology

A New Iodine-Based Functional Material for Infectious Diseases Prevention: Germicidal Properties and Potential Applications

Research Article

Volume 3 Issue 3- 2022

Author Details

Katsuyoshi Tatenuma[1], Yoshiaki Kinase[1,2], Hitomi Kakuta[1,2] and Fabio Spaziani[1,3]*

[1]*Kaken Inc, 1044 Hori, Mito 310-0903, Japan*
[2]*Iode-Labo Inc, 1044 Hori, Mito 310-0903, Japan*
[3]*ENEA (CR Casaccia), Via Anguillarese 301, 00123 Rome, Italy*

*Corresponding author

Katsuyoshi Tatenuma, Kaken Inc, 1044 Hori, Mito 310-0903, Japan

Article History

Received: June 04, 2022 Accepted: June 06, 2022 Published: June 20, 2022

Abstract

An Iodic Acid-Based solution (hereinafter referred to as "Iodox solution") was investigated for its disinfectant and deodorant properties. The experimental results showed that the bactericidal ability is high both in a wet and dry state and stronger than commercial chlorine-based disinfectants. Excellent antimold and deodorant activities were also proved. Additionally volcanic pumice granules impregnated with Iodox solution called "Iodox soil" were tested for their virucidal activity on avian influenza in order to evaluate its application in breeding farms for livestock infectious diseases prevention. The results showed that Iodox soil completely inactivated the virus (residual virus titer 1/10 million or less) in a short time (about 10 minutes). Exhaustive tests, according to OECD guidelines established the safety of the product. Being the Iodox solution colorless and odorless and having a bactericidal effect equivalent to the widely used povidone-iodine it has the potential to be employed as a topical disinfectant (both for medical applications and daily antiseptic necessities) and for numerous applications in various fields.

Keywords: Iodic acid; Iodine; Disinfection; Infectious diseases; Antibacterial; Antiviral; Antimold; Deodorant

Introduction

Iodine is one of the halogens a subset of very chemically reactive elements (all act as strong oxidizing agents) and is well recognized as an efficient germicidal agent used against numerous pathogens that constitute a major health risk to humans. Similarly to chlorine-based ones there are free-iodine disinfectants (such as aqueous iodine solution, iodinated phenol, iodine glycol) and combined-iodine disinfectants (such as iodoform, bismuth formic iodide, thymol iodide, iodocresol, iodophors) according to the solvent and substances interfering (by complexing) with iodine species [1,2]. Iodine germicidal activity depends on its chemical form which is regulated by the chemical conditions of the system. As an example, iodine in an aqueous solution shows a germicidal efficiency depending on the pH and Eh (Figure 1). Generally in an acidic to neutral aqueous solution molecular iodine (I_2) has the strongest efficiency while Hypoiodous acid (HIO, IO^-) is slightly weaker. In alkaline aqueous solution, Iodine and Iodate ions (such as I^-, IO^-, IO_3^-) have very low or no germicidal activity. In previous studies we incorporated the germicidal activity of molecular iodine on activated carbons and successfully tested the potential use of such iodine-based materials for numerous applications [4,5]. In the present study instead we focused our attention on a different type of iodine chemical form the iodic acid and specifically an iodic acid-based aqueous solution (hereinafter referred to as "Iodox solution").

Germicidal Activity of Iodox (Iodic Acid-Based Products)

Antibacterial and Antiviral Activity

The antibacterial activity of Iodox solution was tested and compared to a commercially available chlorine-based disinfectant. An Iodox solution (containing HIO_3 600ppm, adjusted to pH 3 to 4 with phosphate buffer) and a hypochlorite solution (containing NaClO 200ppm) were used as germicidal samples and tested against Escherichia Coli. The experimentation was conducted under wet and dry conditions. In the wet experiments 0.2ml of each germicidal sample was pipetted

in the center of a Petri dish (9cm diameter) and 0.2mL of Escherichia Coli solution (10^5-10^6CFU/mL) was then injected into the same location (center of the Petri dish) in order to obtain a total volume of 0.4ml. In the dry experiments 0.4ml of each germicidal sample was pipetted in the center of a Petri dish and was left air-drying and then 0.4ml of Escherichia Coli solution was injected in order to obtain again a total volume of 0.4ml. After this preliminary step a 4cm × 4cm polyethylene film was placed to cover the solution and all the Petri dishes were cultured at about 35°C for 20 hours. Afterward each Petri dish was rinsed with 10mL of sterile saline solution which was retrieved and diluted 10 times with new aliquots of sterile saline solution. 1ml of the diluted solution was added to standard agar medium and cultured at about 35°C for 22 hours. Both the wet and dry experiments were conducted using untreated Escherichia Coli culture of about 10^7CFU as control. The results showed that the Iodox solution has a strong bactericidal activity both in wet and dry conditions resulting in the complete absence of bacteria (Figure 2). The chlorine-based solution instead exhibited a weaker bactericidal effect in the wet experiment and was completely inefficacy when tested under dry conditions.

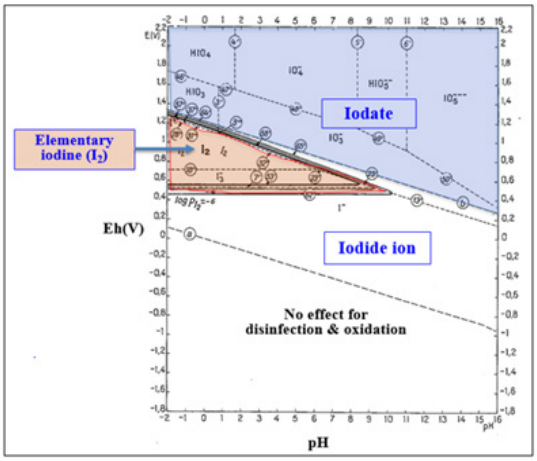

Figure 1: pH-Eh equilibrium diagram for aqueous iodine species, at 25°C (adapted from [3]). According to the diagram, iodate species (such as IO_3-, IO_4-) has strong germicidal activity and oxidizing power at high Eh and within a wide pH range. By keeping iodate compounds in an insoluble state, the germicidal activity can be preserved for a long period. Elementary iodine (I_2) has high germicidal activity at high Eh and low pH. Iodine activity is especially reduced at low Eh and high pH (alkaline condition).

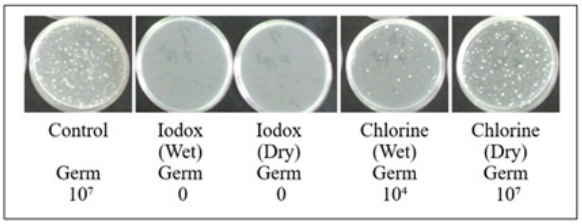

Figure 2: Antibacterial test results of Iodox solution vs. chlorine-based disinfectants.

Antiviral tests were designed in order to evaluate the application of Iodox solution in the development of a granular scatter disinfectant for long-lasting livestock infectious diseases prevention. For this purpose volcanic pumice granules (such as Hyuga soil and Kanuma soil which are abundant in Japan having a specific gravity of about 0.5 and a weakly acidic pH) were impregnated with Iodox solution (to attain a 1-5%w/w amount of the Iodox's iodine component retained in the granules). The resulting granular product (Figure 3) here in after called "Iodox soil" was tested against avian influenza specifically the "A/whistling swan/Shimane/499/83/(H5N3)" strain (a lowly pathogenic strain isolated in 1983 from the feces of Tundra swans wintering in Shimane Prefecture Japan that was confirmed to become highly pathogenic through successive passages in chicks [6,7]. The virus was inoculated in the allantoic cavity of 10-day-old embryonated chicken eggs cultured at 35°C for 2 days and the allantoic fluid was finally collected and used to prepare the virus solution employed in the experiments. The 50% Egg Infectious Dose (EID50) was calculated and adjusted to about $10^{7.5}$EID50/0.2mL with sterile Phosphate-Buffered Saline (PBS). Iodox soil and the virus solution were inserted in a test tube using a 1:10 ratio (1cc of Iodox soil in 10cc of virus solution) and mixed for 10minutes. Once the mixing was completed the solution was serially diluted with PBS (1:10, 1:100, 1:1000, etc.) and 0.2 mL of each dilution stage was inoculated into the allantoic cavity of 3 eggs (10-day-old embryonated eggs) and cultured at 35°C for 2 days. The residual virus titer was calculated as EID50 using the Reed and Muench method [8]. The results evidenced the germicidal activity of Iodox on avian influenza since a residual virus titer of 1/10 million or less was registered.

Figure 3: Iodox soil (pumice granules impregnated with Iodox solution).

Antimold Activity

The antimold activity of Iodox solution was assessed by comparison with commercially available chlorine-based products. Three Iodox solutions (containing respectively HIO_3 3ppm, 30ppm and 300ppm) adjusted to pH 3 to 4 with phosphate buffer a hypochlorite-based disinfectant (NaClO 200ppm) and a chlorine dioxide-based disinfectant (ClO_2 100ppm) were used. Sterile gauze squares (5x5cm) were lightly impregnated with the abovementioned solutions and placed in sterile Petri dishes. A solution containing black mold was injected into each gauze and the Petri dishes were stored at about 26°C for 14 days. The results (Figure 4) showed that the mold did not grow on the gauzes impregnated with the Iodox solutions and with the hypochlorite-based product while abundantly bloomed on the gauzes treated with the chlorine dioxide-based disinfectant. A further comparison was conducted on strawberries (used because it is a fruit that is easily damaged by mold). Iodox solutions and the abovementioned commercial products were sprayed on the fruits. The strawberries treated with Iodox solutions were the ones that showed the most durable freshness since were the last to get moldy.

Deodorant Activity

Iodic acid has the ability to decompose smelly substances such as ammonia and hydrogen sulfide through its oxidizing power. The deodorant ability of Iodox solution was primarily tested against ammonia. Ammonia water (concentration 200ppm) and Iodox solution (concen-

tration 1000ppm) were mixed in a 1:1(v/v) ratio and after stirring the odor was evaluated using human panels. Ten panelists were involved in the test and all of them reported that the samples not treated with Iodox solution emanated a strong pungent odor while the ammonia odor was not perceived in the samples blended with Iodox solution. The results therefore confirmed the ability of Iodox solution to decompose odorous substances such as ammonia. Iodox solution efficiency was also compared with commercial deodorant products evidencing excellent and better deodorizing activity even against trimethylamine and hydrogen sulfide which are malodorous components difficult to eliminate.

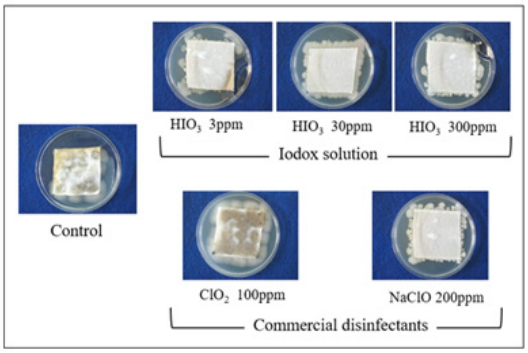

Figure 4: Antimold test results of Iodox solution vs. chlorine-based commercial disinfectants.

Safety

In order to assess the safety of Iodox solution the following exams were executed according to the "OECD Test Guidelines for Chemicals" [9] acute oral toxicity test, skin irritation test, eye irritation test, skin sensitization test and bacterial reverse mutation test. The tests were performed using an Iodox solution having a 3000ppm concentration of iodic acid which is 5 times the concentration practically used in germicidal applications. The results did not show any abnormalities in any of the tests thus confirming the safety of Iodox solution (Table 1).

Table 1: Safety test results of Iodox solution (3000ppm).

Test	Results
Acute oral toxicity on mouse	• No deaths observed during the observation period • No abnormalities found by observing the general conditions • Good weight gain • The median lethal dose (LD50) after a single oral dose was > 2000 mg/Kg
Skin irritation on rabbit	• No skin reaction observed • No skin irritating or corrosive effects on the skin • GHS classification: Not classified
Eye irritation on rabbit	• No stain (damage) on the cornea • It was concluded that the product has no seriously damaging or irritating effects • GHS classification: Not classified
Skin sensitization on mouse	• No skin reactions observed • It was judged to be a no skin sensitizing substance • GHS classification: Not classified
Bacterial reverse mutation	• No increase in the number of mutant colonies above a threshold of 2-fold in all the strains was observed as compared to the concurrent negative controls • No growth inhibitory effect • At the time of colony counting, no precipitation of the test substance was observed in any of the treatment methods • Judged as negative from the above results

Potential Applications of Iodox Products and Future Directions

The results obtained overall confirmed the germicidal activity of Iodox solution, thus indicating its direct applicability as a liquid and spray disinfectant. Being non-irritating, colorless and non-staining it can be utilized in several fields and can replace disinfectants characterized by drawbacks (such as the staining effect of povidone-iodide). Its confirmed safety also makes it suitable for the disinfection needs of generic users (such as for first aid or household disinfection). Moreover, it was also ascertained the possibility of using Iodox solution in conjunction with other ingredients in order to develop materials for very specific applications. As an example the Iodox soil obtained by treating natural pumice granules with Iodox solution and efficaciously tested against the avian influenza virus is suitable for the disinfection of places such as land and livestock farms.

Additional applications currently under testing are food preservation and the use of Iodox-based material in air-conditioners (for mold and odors prevention). A summary of potential applications of Iodox solution and Iodox-based materials is reported in (Table 2).

Table 2: Potential application of Iodox products.

Field	Application examples	Effect
Environment	Place sterilization (e.g. after sewer flooding), algal bloom	Disinfection and infectious diseases prevention
Drinking water	Water purifying, arsenic oxidation (AsIII to AsV)	Safe drinking water production, increase arsenic removal (since AsV is more efficiently removed by sorbents)

Tables continues

Zootechnics	Breeding farm area disinfection, drugs, eggs washing	Infectious diseases (Avian influenza etc.)/ Zoonosis prevention
Medicine and Healthcare	Intestine sterilization treatment, blood sterilization treatment, pre-surgical tools sterilization, antibacterial bandages/gauzes/ cotton	HIV inactivation, sapraemia treatment, ulcerative colitis treatment, skin disinfection
Defense	Military germicidal mask	Protection against biological weapons
Indoor air treatment	Air conditioning filters, Aseptic room filters (laboratory, hospital, etc.)	Air disinfection and deodorization
Food industry	Food packaging and storage	Keeping of freshness, food poisoning prevention
Electrical appliances	Water-related devices (pool water treatment, bath water treatment, etc.)	Disinfection
Housekeeping	Clothes, furniture, toys, etc.	Disinfection and deodorization
Construction	Surface, floor, wall treatment	Antibacterial and antimold prevention
Aquaculture	Aquaculture fish tank	Fish diseases prevention
Import & Export	Post-harvest treatment for imported citrus, etc.	Mold prevention during transportation
Agriculture	Agricultural product	Replant disease prevention

Conclusion

Iodox solution a product based on iodic acid was tested concerning its germicidal activity and deodorant activity. Rigorous experiments involving Escherichia Coli, Avian influenza and black mold demonstrated that Iodox can eradicate a wide range of pathogens. Our experiments were also focused on the exploitation of Iodox solution as an ingredient for functional materials. From this point of view we successfully produced and tested a granular material prepared from natural pumice granules and incorporating the Iodox solution properties which could be used for the prevention of infectious diseases in livestock farming areas. These preliminary results open the way for numerous applications in many fields also including healthcare (such as wound disinfection and home sanitization), medicine (such as pre-surgical skin antiseptic), agriculture and zootechnics (such as disinfection of breeding farm and agriculture products preservation).

Acknowledgment

The authors gratefully thank the following: Mr. Tsutomu Kojima and Mr. Kazuyuki Someya (Kaken Inc.) who were in charge of the experiments concerning the basic research the analysis and the development of Iodox; Dr. Sayoko Yano and Ms. Chiemi Ueba (Kyoto Prefectural Agriculture, Forestry and Fisheries Technology Center) and Mr. Koichi Endo (A&A Material Corporation) who discovered the practical functions of pumice granules impregnated with Iodox solution; Ms. Toshiyo Yabuta, Dr. Hiroki Takakuwa, and Dr. Koichi Otsuki (Avian Influenza Research Centre, Kyoto Sangyo University) for their implementation and guidance of antiviral tests using the avian influenza virus "A/whistling swan/Shimane/499/83/(H5N3)" strain.

References

1. Kaiho T (2014) Physical Properties of Iodine. Iodine Chemistry and Applications 7-14.
2. Gottardi W (2014) Iodine as Disinfectant. Iodine Chemistry and Applications 375-410.
3. Pourbaix M. Atlas of electrochemical equilibria in aqueous solution. 2nd ed. National Association of Corrosion Engineers; 1974: 614-626.
4. Spaziani F, Natori Y, Kinase Y, Kawakami T, Tatenuma K (2019) Elementary Iodine-Doped Activated Carbon as an Oxidizing Agent for the Treatment of Arsenic-Enriched Drinking Water. Water 11(9): 1778.
5. Natori Y, Kinase Y, Ikemoto N, Spaziani F, Kojima T, et al. (2021) Activated Carbon Impregnated with Elementary Iodine: Applications against Virus- and Bacteria-Related Issues. C (Journal of Carbon Research) 7(4): 86.
6. Otsuki K, Takemoto O, Fujimoto R, Yamazaki K, Kubota T, et al. (1984) Isolation of H5 influenza viruses from whistling swans in western Japan in November 1983. Acta Virol 28(6): 524.
7. Otsuki K, Takemoto O, Fujimoto R, Yamazaki K, Kubota N, et al. (1987) Isolation of influenza A viruses from migratory waterfowl in San-in District, western Japan in the winter of 1983-1984. Res Vet Sci 43(2): 177-179.
8. Reed LJ, Muench H (1938) A simple method of estimating fifty percent endpoints. Am J Hyg 27(3): 493-497.
9. OECD Guidelines for the Testing of Chemicals, Section 4.