

# Higher Concentrations of Plasmacluster Ions<sup>®</sup>\*<sub>1</sub> Boost Virus Inactivation and Elimination, Inhibit 99.9% of Airborne H5N1 Avian Influenza (“Bird Flu”) Virus

Verified in Collaboration with Retroscreen Virology Ltd. \*<sub>2</sub> of the UK, August 27, 2008

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Sharp Corporation, working in collaboration with Retroscreen Virology Ltd. founded by Professor John S. Oxford of the University of London, UK, has demonstrated that Plasmacluster Ions (PCIs) inactivate and eliminate the airborne, highly pathogenic H5N1 avian influenza virus by 99.9% in ten minutes at an ion concentration of approximately 50,000 ions/cm<sup>3</sup> in a box having a volume of 1 m<sup>3</sup>. In 2005, Sharp and Retroscreen Virology previously verified that Plasmacluster Ions inactivate and eliminate the same virus by 99% in ten minutes at a concentration of approximately 7,000 ions/cm<sup>3</sup>. Now, the use of Plasmacluster Ions in higher concentrations has been proven to inhibit infection in cells at an even higher rate.

Plasmacluster Ions are a revolutionary air purification technology in which **positive ions** [H+(H<sub>2</sub>O)<sup>n</sup>] and **negative ions** [O<sub>2</sub><sup>-</sup>(H<sub>2</sub>O)<sup>m</sup>] are released into the air simultaneously. These **positive and negative ions** instantly recombine on the surface of bacteria, mold fungus, viruses and allergens floating in the air to form hydroxyl (OH) radicals, which have extremely high oxidation ability, and this chemical reaction decomposes proteins on the surface of bacteria and other pathogens, thereby inhibiting their activity. Sharp is using the fact that higher ion concentration results in greater inactivation and elimination of the H5N1 influenza strain as a springboard to push ahead with further R&D on high-concentration Plasmacluster Ion generators.

Details of these findings will be presented at BirdFlu2008: Avian Influenza and Human Health, the first international avian influenza conference, which will be held in the UK on September 10, 2008.

In the eight years since developing Plasmacluster Ion technology, Sharp, in a "collaborative research approach to product marketing"<sup>\*<sub>3</sub></sup> based on working together with academic research organizations around the world, has demonstrated that this technology can inactivate and eliminate 27 different harmful microbes, including MRSA<sup>\*<sub>4</sub></sup>. The efficacy of Plasmacluster Ions for inactivating and eliminating airborne viruses has been proven against the H1N1 strain of human influenza virus, as well as Corona, Polio, and Coxsackie viruses. In addition, in 2005, Sharp, working together with a number of academic institutions<sup>\*<sub>5</sub></sup>, elucidated the mechanism behind the ability of Plasmacluster Ions to destroy the spike-like proteins on the virus surface, which are the triggers for infections. In addition, the safety of high concentrations of Plasmacluster Ions has also been confirmed<sup>\*<sub>6</sub></sup>.

Sharp will continue its initiatives to contribute to society through the further evolution of Plasmacluster Ions technology and additional demonstrations of its efficacy with the aim of lessening the anxiety about new viruses that may emerge in the future.

## Comments by Professor John S. Oxford

I founded Retroscreen Virology Ltd. in 1989, and since then, have been pursuing R&D and conducting verification testing related to viruses, drugs, vaccines, and advanced face masks for medical use. At present, mutations in the avian influenza virus pose a risk that the infection will spread from birds to humans, leading to a worldwide pandemic. Currently, sophisticated face masks are worn to prevent infection in individuals. But along with such masks, we can anticipate that Plasmacluster Ions technology will contribute to combating a potential future influenza pandemic.

- \*1 Plasmacluster and Plasmacluster Ions are trademarks of Sharp Corporation.
- \*2 The OECD (Organisation for Economic Co-operation and Development) Principles of GLP (Good Laboratory Practice) is a set of standards intended to ensure the generation of high-quality and reliable test data through periodic reviews of operational organization and management, test apparatus and materials, study designs, internal audit controls, quality assurance systems, test data, etc., at all test facilities. Re-certification is required every three years.
- \*3 The "collaborative research approach to product marketing" verifies the effectiveness of a technology based on scientific data developed in collaboration with leading-edge academic research institutions. New products are then brought to market based on the results.
- \*4 MRSA is an acronym for methicillin-resistant *Staphylococcus aureus*, a bacterium responsible for difficult-to-treat infections in humans. MRSA typically infects humans with weakened immune systems, for example, patients in hospitals, and its resistance to a large group of antibiotics is a serious problem.
- \*5 Joint research conducted with Professor Gerhard Artmann, of Aachen University of Applied Sciences (2005).
- \*6 Testing conducted by Mitsubishi Chemical Safety Institute Ltd. (inhalation toxicity, as well as eye and skin irritation/corrosion tests).

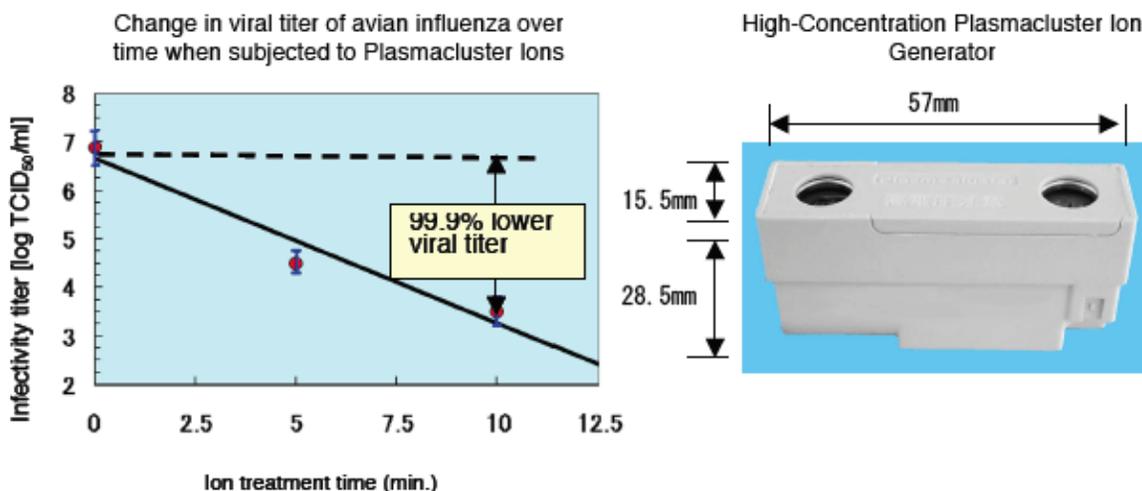
### Method of Testing Efficacy Against Airborne H5N1 Avian Influenza Virus

A Plasmacluster Ion generators were placed in a box having a volume of 1m<sup>3</sup>.

Plasmacluster Ions were generated (concentration: approximately 50,000 ions/cm<sup>3</sup>) and the H5N1 avian influenza virus was sprayed into the box.

Airborne viruses inside the box were sampled five minutes after the spraying was complete and then sampled again 10 minutes after the spraying was complete. Infectivity (viral infectivity titer <sup>\*7</sup>) was studied using the TCID50 method <sup>\*8</sup> commonly used in the virology research field.

After activating the Plasmacluster Ion generator, the infectivity titer of the virus had been reduced by 99.9% after 10 minutes with respect to the virus concentration initially introduced.



- \*7 A value indicating the capacity of a virus to infect cells.
- \*8 50% Tissue Culture Infective Dose method; a test protocol that examines the amount of a virus that will produce pathological change in 50% of cell cultures inoculated with a virus suspension diluted in stepwise increments.

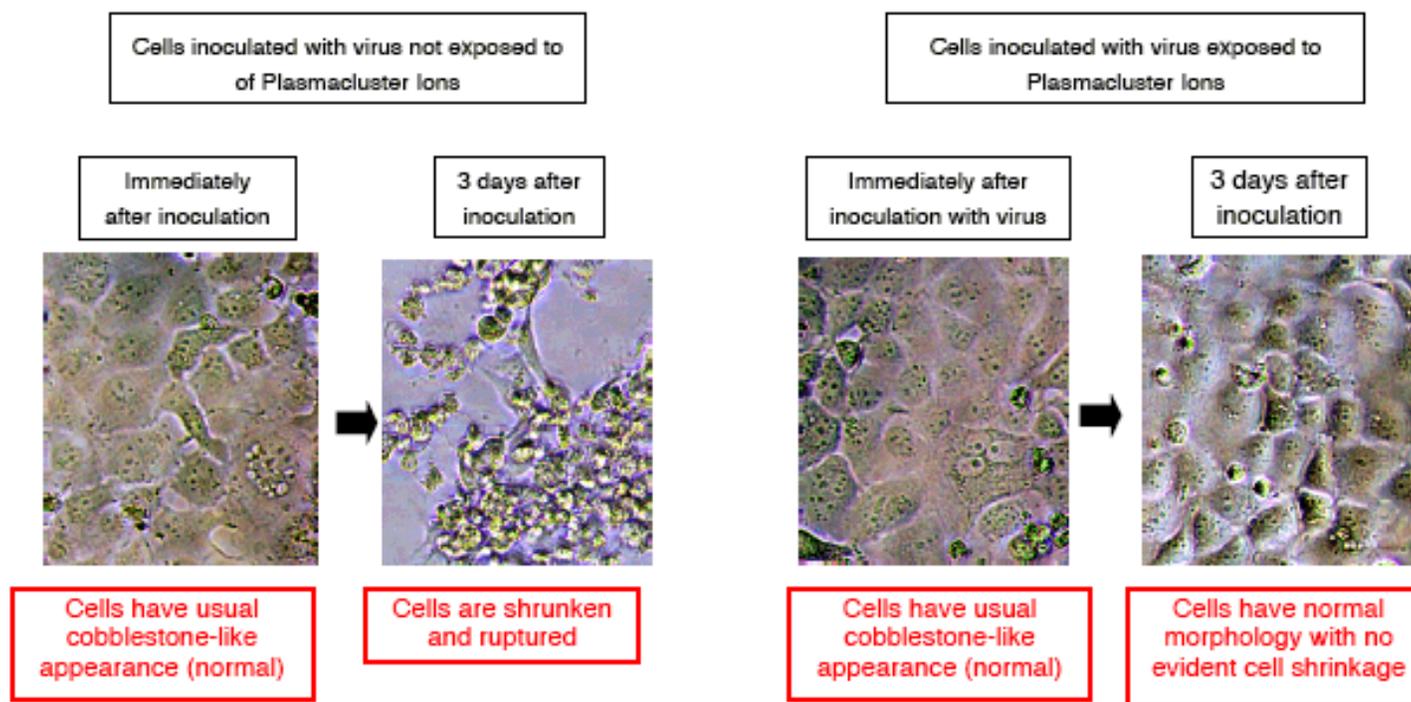
### Infectivity Tests of Virus Using Cells

A Plasmacluster Ion generator was placed in a box having a volume of 1m<sup>3</sup>. Plasmacluster Ions were generated (concentration: approximately 50,000 ions/cm<sup>3</sup>) and the H5N1 avian influenza virus was sprayed into the box. The air in the box containing the airborne virus was sampled 10 minutes after spraying was complete. Airborne virus cells were inoculated with these virus samples over a three-day period and changes in the cells studied.

Cells inoculated with the virus but not exposed to Plasmacluster Ions had shrunk and ruptured after three days. In contrast, cells inoculated with the virus that had been exposed to Plasmacluster Ions were largely not shrunken and retained their normal morphology.

These findings confirm that Plasmacluster Ions can inhibit the ability of a virus to infect cells.

**Photomicrographs of cells (MDCK\*<sup>9</sup> cells) inoculated with the H5N1 virus (40X magnification)**



\*<sup>9</sup> Madin-Darby canine kidney (MDCK) cell line; an animal-derived cell system used in research.

**Oxidizing Substances Produced by Plasmacluster Ions**

Plasmacluster Ions adhere to airborne viruses, and the positive and negative ions react to form OH (hydroxyl) radicals, which have the most powerful oxidation ability (standard oxidation potential of 2.81 V), thereby inhibiting the infectivity of the airborne virus.

Active Substance	Chemical Formula	Standard Oxidation Potential [V]
Hydroxyl radical	•OH	2.81
Oxygen atom	•O	2.42
Ozone	O <sub>3</sub>	2.07
Hydrogen peroxide	H <sub>2</sub> O <sub>2</sub>	1.78
Hydroperoxide radical	•OOH	1.7
Oxygen molecule	OO <sub>2</sub>	1.23

## About the H5N1 Strain of Avian Influenza

(Source: Japanese Prime Minister's Office website)

Avian influenza is an infection that spreads from birds to other birds, however, humans can also be infected through close contact with living birds. There is also the possibility of the virus mutating into a new type of highly infectious virus capable of being transmitted from human to human.

## Efficacy of Plasmacluster Ions Against Various Pathogens Confirmed Through Collaborative Research

Target Substance	Species	Testing & Verification Organization	Date of Announcement
Bacteria	<i>Serratia</i> bacteria	Harvard School of Public Health (Dr. Melvin W. First, Professor Emeritus), United States	March 2007
	Coliform bacteria ( <i>E. coli</i> )	Ishikawa Health Service Association, Japan	September 2000
	<i>E. coli</i> , <i>Staphylococcus aureus</i> , <i>Candida</i>	Shanghai Municipal Center for Disease Control and Prevention, China	October 2001
	<i>Bacillus subtilis</i>	Kitasato Research Center of Environmental Sciences, Japan	September 2002
		CT&T (Professor Gerhard Artmann, Aachen University of Applied Sciences), Germany	November, 2004
	MRSA (methicillin-resistant <i>Staphylococcus aureus</i> )	Kitasato Research Center of Environmental Sciences, Japan	September 2002
		Kitasato Institute Medical Center Hospital, Japan	February 2004
	Pseudomonas, Enterococcus, Staphylococcus	University of Lübeck, Germany	February 2002
	Enterococcus, Staphylococcus, Sarcina, Micrococcus	CT&T (Professor Gerhard Artmann, Aachen University of Applied Sciences), Germany	November 2004
	Allergens	Mite allergens, pollen	Graduate School of Advanced Sciences of Matter, Hiroshima University, Japan
Airborne allergens		Asthma Society of Canada	April 2004

Fungi	Cladosporium	Ishikawa Health Service Association, Japan	September 2000
		University of Lübeck, Germany (growth-suppressing effect)	February 2002
		CT&T (Professor Gerhard Artmann, Aachen University of Applied Sciences), Germany	November 2004
	Penicillium, Aspergillus	University of Lübeck, Germany (growth-suppressing effect)	February 2002
	Aspergillus, Penicillium (two species), Stachybotrys, Alternaria, Mucorales	CT&T (Professor Gerhard Artmann, Aachen University of Applied Sciences), Germany	November 2004
Viruses	H1N1 human influenza virus	Kitasato Research Center of Environmental Sciences, Japan	September 2002
		Seoul University, Korea	September 2003
		Shanghai Municipal Center for Disease Control and Prevention, China	December 2003
		Kitasato Institute Medical Center Hospital, Japan	February 2004
	H5N1 avian influenza virus	Retroscreen Virology, Ltd., London, UK	May 2005
	Coxsackie virus	Kitasato Research Center of Environmental Sciences, Japan	September 2002
	Polio virus	Kitasato Research Center of Environmental Sciences, Japan	September 2002
	Corona virus	Kitasato Institute Medical Center Hospital, Japan	July 2004

Note: *Efficacy in inhibiting activity of the airborne target substances noted above was verified by exposing the substances to an ion concentration of at least 3,000 ions/cm<sup>3</sup>.*

## Profile of Professor John S. Oxford

- Professor of Virology in the Institute of Cell and Molecular Science at St. Bartholomew's and the Royal London Hospital, Queen Mary's School of Medicine and Dentistry, University of London, UK
- Founder and Scientific Director of Retroscreen Virology Ltd.
- Specialization: Virology

## Publications

- Scientific papers: Approximately 250
- Texts: Three texts on virology

1. *Influenza, the Viruses and the Disease*
2. *Human Virology: A Text for Students of Medicine, Dentistry and Microbiology*
3. *Conquest of Viral Diseases*

## Other

- Has chaired numerous international academic conferences and meetings

## About the University of London

Established by Royal Charter in 1836, the University of London is a federation of 19 Colleges and Institutes and has more than 115,000 students, making it one of the largest universities in the world.

Queen Mary, University of London, is composed of the UK's oldest medical school, London Hospital Medical College (est. 1785), and St. Bartholomew's Hospital Medical College (est. 1843). It has approximately 8,800 undergraduate students and offers a wide range of fields of study including not only medicine and dentistry, but also biology, chemistry, physics, electrical engineering, computer science, law, literature, political science, and others. It has also produced seven Nobel Prize winners.

## Retroscreen Virology Ltd.

Retroscreen Virology Ltd. was founded by Professor John Oxford in 1989 to conduct R&D and verification testing related to viruses, drugs, and vaccines, and is well known as one of the leaders of its field. It is certified under GLP (Good Laboratory Practices), an international set of standards for maintaining high levels of reliability and safety in trials involving chemical substances. It is also ISO 9001-certified.

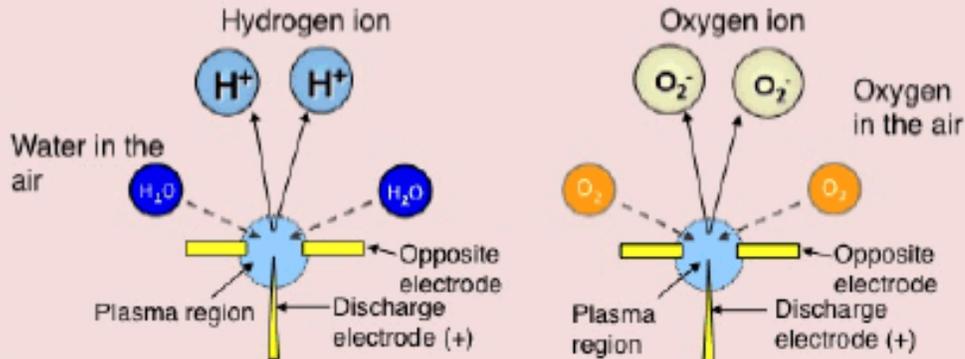


## Website Set Up by Professor John Oxford

A new website entitled "A New Approach to Fighting H5N1" has been set up for R&D based on new approaches, and oriented toward mitigating the risk of infection by the avian influenza virus. On the site, Professor Oxford will also be describing the testing and evaluation of Plasmacluster Ions he has performed.

## Plasmacluster Ion Generation

Applying positive and negatively charged voltages to discharge electrodes electrically decomposes water molecules in the air into hydrogen molecules and oxygen molecules. Positive hydrogen ions ( $H^+$ ) and negative oxygen ions ( $O_2^-$ ) are generated in this way.



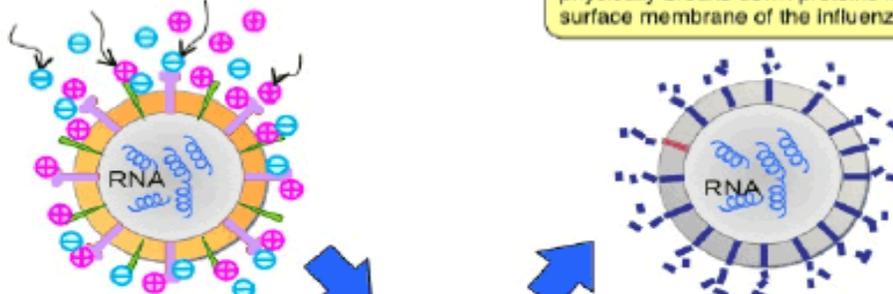
Water molecules in the air cluster around the ions like a bunch of grapes. Each ion forms part of a stable "bunch of grapes" or ion cluster.



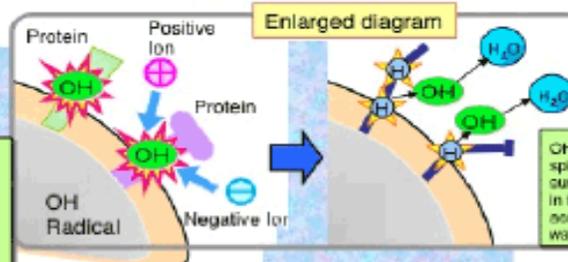
## Working Mechanism to Inhibit Infection by Airborne Viruses

Positive and negative ions surround the surface membrane of the airborne virus.

A chemical reaction takes place that physically breaks down proteins in the surface membrane of the influenza virus.



Through a chemical reaction occurring on the virus membrane surface, the ions are transformed into OH radicals, which are powerfully active but unstable.



(Information taken from collaborative research done in association with Professor Gerhard Artmann of Aachen University of Applied Sciences in Germany.)

OH radicals steal hydrogen atoms from the spike-like proteins that protrude from the surface of the virus membrane, opening holes in the membrane. When the OH radicals acquire a second hydrogen atom, they form water ( $H_2O$ ).

## Efficacy of PCIs on Various Viruses

Species of Virus	Method of Testing/Efficacy	Testing and Verification Organization	Explanation of Virus
H5N1 influenza virus	Testing volume: 1m <sup>3</sup> box Dispersal time: 10 minutes Inactivation ratio: 99.0%	Retroscreen Virology, Ltd., London, UK Study No. PNT-PCS-001(2005)	Virus extracted from humans that causes avian influenza
H1N1 influenza virus	Testing volume: 1m <sup>3</sup> box Dispersal time: 25 minutes Inactivation ratio: 99.7%	Kitasato Research Center of Environmental Sciences, Japan Kitasato Institute Medical Center Hospital, Japan Study No. 00313 (2004)	Virus causing influenza infection in humans
Corona virus	Testing volume: 1m <sup>3</sup> box Dispersal time: 35 minutes Inactivation ratio: 99.7%	Kitasato Institute Medical Center Hospital, Japan Study No. 00313 (2004)	Virus causing inflammation of the stomach lining in cats; same family as the SARS virus
Coxsackie virus	Testing volume: one-pass test Dispersal time: 3.3 seconds Inactivation ratio: 98.9%	Kitasato Research Center of Environmental Sciences, Japan Kitasato Study No. 13-0214-2 (2002)	Virus causing summer colds
Polio virus	Testing volume: one-pass test Dispersal time: 3.3 seconds Inactivation ratio: 98.9%	Kitasato Research Center of Environmental Sciences, Japan Kitasato Study No. 13-0214-2 (2002)	Virus causing paralysis in children
SARS Virus	Testing volume: one-pass test Dispersal time: 3.3 seconds Inactivation ratio: 73.4%	Retroscreen Virology, Ltd., London, UK Study No. PNT-PCS-003 (2005)	Virus causing SARS

The following information is true and accurate at the time of publication. Manufacture, sale, price and specifications of products may be subjected to change.