## Joint Research with Hiroshima University Proves Plasmacluster Technology<sup>\*1</sup> Suppresses Effects of Airborne Fungi Allergens<sup>\*2</sup>

Joint research conducted by Sharp Corporation and Hiroshima University (Graduate School of Advanced Sciences of Matter) has demonstrated that Plasmacluster technology can reduce the amount of a major allergen contained in the crude allergens<sup>\*3</sup> of *Aspergillus fumigatus* (hereinafter, *Aspergillus*), a type of airborne fungus. The research also showed that Plasmacluster technology inhibits the reactivity to IgE antibodies<sup>\*4</sup> in the serum of *Aspergillus* allergy patients.

Fungi are all around us and cause allergic reactions when breathed in. The *Aspergillus* tested in this research is, along with house dust mite, one of the major airborne triggers of allergies.

Sharp has previously proved that Plasmacluster technology can suppress the effects of house dust mite allergens\*<sup>5</sup>. By proving that Plasmacluster is effective in suppressing airborne *Aspergillus* allergens as well, this latest research further raises the promise of Sharp's proprietary air technology for fighting allergies.

Since introducing Plasmacluster technology in 2000, Sharp has conducted academic marketing<sup>\*6</sup> in collaboration with some of the world's leading third-party scientific research organizations. So far,  $23^{*7}$  such organizations have shown the effectiveness of Plasmacluster in not only controlling harmful substances such as viruses, bacteria, and allergens, but also in beautifying people's skin and hair in a proven safe<sup>\*8</sup> manner.

Sharp will continue to create healthy environments by advancing Plasmacluster technology and conducting testing to show its effectiveness.

This research is scheduled to be presented at the EAACI (European Academy of Allergy and Clinical Immunology) Congress 2014 held June 7 to 11 in Copenhagen, Denmark.

# 1. Reduction effect of Plasmacluster ions on the amount of major allergen Asp f 1\*<sup>9</sup> contained in crude *Aspergillus* allergens

Test Space	Bombardment Period	Reduction Rate
8.7-liter cylindrical container measuring 14.5 cm (diameter) x 52.5 cm (length)	Approx. 7 min.	Approx. 76%

## 2. Inhibitory effect of Plasmacluster ions on crude *Aspergillus* allergens (suppression of their binding to IgE antibodies in the sera of *Aspergillus*-allergic patients)

Test Space	Bombardment Period	Inhibition Rate
8.7-liter cylindrical container measuring 14.5 cm (diameter) x 52.5 cm (length)	Approx. 7 min.	Approx. 56%

- \*1 Plasmacluster is a registered trademark of Sharp Corporation.
- \*2 Allergens are substances that cause allergic reactions; they are found in house dust mite, pollen, fungi, and other proallergic substances.
- \*3 Crude allergens of *Aspergillus* contain multiple allergen molecules. By contrast, a purified allergen is a specific allergen molecule that has been purified through biochemical separation procedures.
- \*4 An antibody is a class of protein that specifically binds to certain target antigens. IgE (immunoglobulin E) is an antibody isotype that causes allergic reactions.
- \*5 Announced on September 3, 2003.
- \*6 Collaboration with leading third-party academic research institutions to gather and analyze scientific data in order to apply findings to new products.
- \*7 As of April 10, 2014.
- \*8 According to testing by LSI Medience Corporation (inhalation toxicity test, eye and skin irritation test, and corrosivity test).
- \*9 The major allergen Asp f 1 is one of the main allergens of *Aspergillus* allergens. Its molecular structure has been elucidated, and it has been registered with the WHO (World Health Organization).

## **1.** Reduction effect of Plasmacluster ions on the amount of protein in major allergen Asp f 1 in crude *Aspergillus* allergens

#### Verification methods

- Test institution: Hiroshima University (Graduate School of Advanced Sciences of Matter)
- Test space: 8.7-liter cylindrical container measuring 14.5 cm (diameter) x 52.5 cm (length)
- Allergen tested: Major allergen Asp f 1 contained in crude allergens of Aspergillus fumigatus
- Test device: Plasmacluster ion generating unit (attached to inside of cylindrical container)
- Plasmacluster ion density: Average of 25,000 ions/cm<sup>3</sup> in cylindrical container
- Plasmacluster ion bombardment period: Approx. 7 min.
- Control test: Cylindrical container with no Plasmacluster ion generating unit
- Test and analytical methods:

A Plasmacluster ion generator was attached to the inside of the cylindrical container. The container was filled with Plasmacluster ions. A mist of crude *Aspergillus* allergens was infused into the upper part of the container. The ion-treated and sham-treated allergens were collected at the bottom of the container, and the amount of protein in the major allergen Asp f 1 was quantified using the sandwich ELISA\*<sup>10</sup> method.

#### Results

Compared to the control test (*i.e.* without Plasmacluster ion generating unit), treatment with Plasmacluster ions significantly reduced the amount of protein in the major allergen Asp f 1 (an approximate 76% reduction) in the crude *Aspergillus* allergens.



\*10 Sandwich ELISA method: A method of measuring the amount of biopolymers such as allergens.

# 2. Inhibitory effect of Plasmacluster ions on the binding capacity of crude *Aspergillus* allergens to IgE antibodies in the sera of *Aspergillus*-allergic patients

#### Verification methods

- Test institution: Hiroshima University (Graduate School of Advanced Sciences of Matter)
- Test space: 8.7-liter cylindrical container measuring 14.5 cm (diameter) x 52.5 cm (length)
- Allergen tested: Crude allergens of Aspergillus fumigatus
- Test device: Plasmacluster ion generating unit (attached to inside of cylindrical container)
- Plasmacluster ion density: Average of 25,000 ions/cm<sup>3</sup> in cylindrical container
- Plasmacluster ion bombardment period: Approx. 7 min.
- Control test: Cylindrical container with no Plasmacluster ion generating unit
- Test and analytical methods:

A Plasmacluster ion generator was attached to the inside of the cylindrical container. The container was filled with Plasmacluster ions. A mist of crude *Aspergillus* allergens was infused into the upper part of the container. The ion-treated and sham-treated allergens were collected at the bottom of the container, and their reactivity to IgE antibodies in the sera of *Aspergillus*-allergic patients was analyzed using the ELISA inhibition\*<sup>11</sup> method.

#### Results

Compared to the control test (*i.e.* without Plasmacluster ion generating unit), treatment with Plasmacluster ions significantly impaired the reactivity to IgE antibodies in the sera of *Aspergillus*-allergic patients (an approximate 56% inhibition; assessed at inhibition rate of  $50\%^{*12}$ ).



\*11 ELISA inhibition method: A method in which competitive inhibition of antigen-antibody reactions is used to conduct a quantitative comparative analysis of the antibody binding activity of antigens, including allergens. In these tests, a comparison was done on the reactivity of allergens and IgE antibodies in two cases: one in which exogenously added allergens (herein referred to as 'inhibitor') were collected in the presence of Plasmacluster ions; and the other in which the added inhibitor allergens were collected without Plasmacluster ions. If the allergenicity of the inhibitor is lowered as a result of treatment with Plasmacluster ions, a much larger amount of inhibitor must be exogenously added to achieve the same inhibitory action as that of a sham-treated inhibitor. (*i.e.* The graph shifts to the right.)

\*12 Inhibition rate of 50%: The point at which IgE antibody reactivity (binding activity) to immobilized allergen is inhibited by 50% (reduced by half) upon adding the inhibitor (shown by a dotted line on the graph).

# • Comment from Dr. Seiji Kawamoto, Associate Professor, Graduate School of Advanced Sciences of Matter, Hiroshima University

The increase in allergies is a serious social problem, but no radical treatment is available right now for allergic disorders. Thus, the most effective way to prevent allergies is still avoidance and/or elimination of environmental allergens. Through this joint research, we confirmed that Plasmacluster technology is effective in reducing and inhibiting allergens that originate from fungi. This knowledge provides insight into dealing with indoor-environment allergies.

# • Overview of the Department of Molecular Biology, Graduate School of Advanced Sciences of Matter, Hiroshima University

Research in the Department of Molecular Biotechnology covers the entire spectrum, from fundamental research in the mechanisms of life sciences, to research that applies these fundamental research findings. The department has achieved world-class research successes through its innovative ideas and wealth of research facilities. The department's goal is to foster outstanding researchers and advanced technology specialists by conducting state-of-the-art research aimed at achieving new discoveries and applications in molecular biotechnology that result in improving people's well-being.

### • Associate Professor Seiji Kawamoto

Affiliation: Department of Molecular Biology, Graduate School of Advanced Sciences of Matter, Hiroshima University

Specialties: Immunology, Animal cell technology, Applied microbiology, Applied molecular cell biology

#### History

1996: Research Fellow, Japan Society for the Promotion of Science
1997: Assistant Professor, Faculty of Engineering, Hiroshima University
1999: Assistant Professor, Graduate School of Advanced Sciences of Matter, Hiroshima University
2002: Visiting Research Fellow, Harvard Medical School, USA (Research Fellow, Japan Society for
the Promotion of Science Postdoctoral Fellowships for Research Abroad)
2008: Current position

### • Aspergillus

A common species of fungi occurring in the natural environment; *e.g.* in food, dust, and soil. *Aspergillus fumigatus* is a pathogenic type of *Aspergillus* known to cause allergic bronchitis (bronchopulmonary aspergillosis).

#### **About Plasmacluster Technology**

In Sharp's proprietary air purification technology, positively charged hydrogen ions  $(H^+ (H_2O)_n)$  and negatively charged oxygen ions  $(O_2^{-}(H_2O)_m)$  are discharged simultaneously. These positive and negative ions instantaneously bond on the surface of airborne substances such as bacteria, fungi, viruses, and allergens, becoming highly reactive OH radicals (hydroxyl radicals) that break down the proteins on the surface of these bacteria and other substances. By chemical reaction, the OH radicals work to suppress the activity of those substances.



### How Plasmacluster Ions Are Generated

### Mechanism for Decomposing Allergens\*<sup>13</sup>



As Plasmacluster Ions surround airborne allergens, they are transformed into OH (hydroxyl) radicals, a powerful activated substance. These OH radicals denature the allergen's IgE antibody binding site at the molecular level. Thus, even if these allergens were to enter the body, the body would not react with allergic symptoms.

\*13 July 21, 2006 press release: Mechanism for Suppressing Mite Allergens by Plasmacluster Ions Explained

### **Comparison of Oxidation**

Positive and negative ions bond on the surface of airborne viruses and bacteria and react chemically to form OH radicals, which have high oxidation power (standard oxidation potential 2.81 V). These reduce the contagiousness of airborne viruses and the activity of bacteria.

Active Substances	Chemical Formula	Standard Oxidation Potential (V)
Hydroxyl radicals	ОН	2.81
Oxygen atom	0	2.42
Ozone	O <sub>3</sub>	2.07
Hydrogen peroxide	H <sub>2</sub> O <sub>2</sub>	1.78
Hydroperoxyl radical	ООН	1.7
Oxygen molecule	O <sub>2</sub>	1.23

Source: Fundamentals and Applications of Ozone

## 23 Research Institutes That Provided Data for Sharp's Academic Marketing

Target Substance	Testing and Verification Organization
	Kitasato Research Center of Environmental Sciences, Japan
	Seoul National University, Korea
	Shanghai Municipal Center for Disease Control and Prevention, China
	Kitasato Institute Medical Center Hospital, Japan
Viruses	Retroscreen Virology, Ltd., UK
	Shokukanken Inc., Japan
	Hanoi College of Technology, Vietnam National University, Vietnam
	Pasteur Institute of Ho Chi Minh City, Vietnam
	Public Health Research Foundation, Graduate School of Medicine, Tokyo University
	Graduate School of Advanced Sciences of Matter, Hiroshima University, Japan
Allergens	Department of Biochemistry and Molecular Pathology, Graduate School of Medicine, Osaka City University, Japan
	Soiken Inc., Japan
	Ishikawa Health Service Association, Japan
	University of Lübeck, Germany
Fungi	Professor Gerhard Artmann, Aachen University of Applied Sciences, Germany
	Japan Food Research Laboratories, Japan
	Shokukanken Inc., Japan
	Ishikawa Health Service Association, Japan
	Shanghai Municipal Center for Disease Control and Prevention, China
	Kitasato Research Center of Environmental Sciences, Japan
	Kitasato Institute Medical Center Hospital, Japan
Bacteria	Dr. Melvin W. First, Professor Emeritus, Harvard School of Public Health, US
	Animal Clinical Research Foundation, Japan
	University of Lübeck, Germany
	Professor Gerhard Artmann, Aachen University of Applied Sciences, Germany
	Japan Food Research Laboratories, Japan
	Shokukanken Inc., Japan
Organic chemicals	Sumika Chemical Analysis Service, Ltd., Japan
Odors, pet smells	Boken Quality Evaluation Institute, Japan
	Animal Clinical Research Foundation, Japan
Skin beautifying effects	Soiken Inc., Japan
Hair beautifying effects	Saticine Medical Co., Ltd.
	C.T.C Japan Ltd.

Inhibitory effects on viruses, fungi, and bacteria	Professor Gerhard Artmann, Aachen University of Applied Sciences, Germany
Inhibitory effects on allergens	Graduate School of Advanced Sciences of Matter, Hiroshima University, Japan
Skin moisturizing (water molecule coating) effect	Research Institute of Electrical Communication, Tohoku University, Japan

In collaboration with 23 research organizations, Sharp has proven the efficacy and working mechanism of Plasmacluster ions against 34 types of harmful substances (viruses, allergens, fungi, and bacteria) and in neutralizing five types of odors, beautifying skin and hair, and controlling static electricity. The efficacy of Plasmacluster ions against two types of organic chemicals has also been proven.