Inactivation Test of Influenza A Virus Subtype H1N1 by SPi (Samsung Super Plasma Ion)

September 2009

Veterinary Medicine at Chungnam National University

Research Report

To Samsung Electronics

This document is submitted as the final report of Inactivation Test of Influenza A Virus Subtype H1N1 by SPi (Samsung Super Plasma Ion).

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cases and 161 deaths in Australia: 16,876 confirmed cases and 130 deaths in Thailand: 8,406 confirmed cases and 17 deaths in China: 5,022 confirmed cases and 10 deaths in USA: 7,577 confirmed cases and 7 deaths in Korea as of September 13, 2009.

Oseltamivir (the trade name Tamiflu) and Zanamivir (the trade name Relenza) have been reported to be effective to treat and prevent Influenza A virus subtype H1N1. However, Tamiflu, an antiviral drug and precautionary vaccine are immoderately insufficient to supply and side effects caused by taking Tamiflu are making people easeless.

As it comes a flu season soon in Northern Hemisphere, approximately 2 billion people will be infected and a great number of victims are expected since the medical system will be messed up. There is neither perfect treatment nor vaccine prepared exorbitantly. If any device were developed to inactivate Influenza A virus subtype H1N1 in the air, it would prevent people from being infected with the Influenza A virus subtype H1N1.

Influenza A virus subtype H1N1 has been spreading worldwide and it is at this point that every nation is seeking for precautions against Influenza A virus subtype H1N1. Furthermore, testified that Influenza A (H1N1) virus might be eliminated by 99.7 percent (Japan, Kitasato Environmental Science Center, 2004.07), SPi has been increasingly expected that it could contribute to a measure to prevent and control Influenza A virus subtype H1N1. Therefore, in the first place, necessity and usefulness of SPi should be identified to prevent and control Influenza A virus subtype H1N1 for its application.

Hence, this research is to analyze inactivation of Influenza A virus subtype H1N1 (the WHO Standard Strain, A/California/04/09 (H1N1), Cultivated from CDC, USA) by SPi (Samsung Super Plasma Ion).

2. Materials and Methods

This research was carried out for a strain of Influenza A virus subtype H1N1 under each chamber environment under SPi and non-SPi by using the inactivation test method and the reduction ratio of Influenza A virus subtype H1N1 was evaluated according to SPi application.

2.1. Test Period and Place

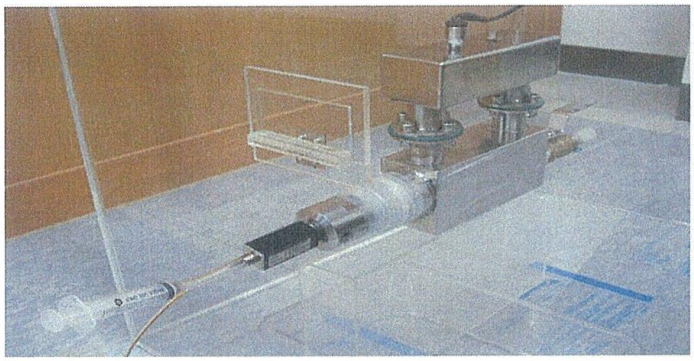
It was carried out in the veterinary medicine at Chungnam National University from July till September 2009. The laboratory for this research is a biosafety level 3 facility specified by the Centers for Disease Control and Prevention (CDC).

2.2. Test Materials

The strain used for this test was A/California/04/09 (H1N1), which is a standard H1N1 virus declared by the WHO, and was cultivated in the Centers for Disease Control and Prevention (CDC, USA).

2.3 Test Plan and Methods

A small chamber into which a SPi device was built was customized for this research. The chamber was made of stainless steel with 500cc capacity, enclosed after finishing an airtight test, and trashed all once it was used.



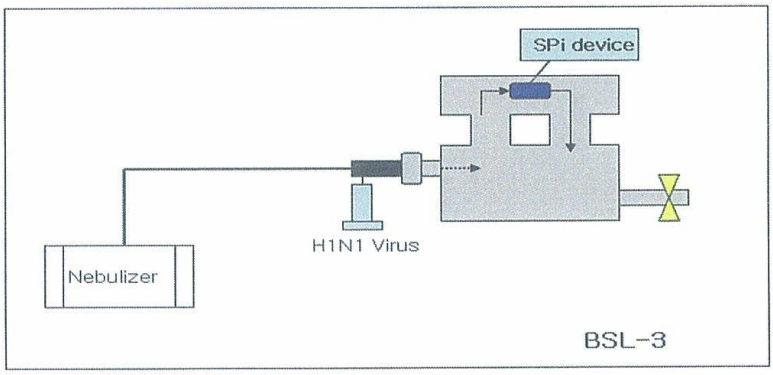
* Test Chamber Developed by Samsung Electroncis, EnHTek, TOPPALLINTECH

<Figure 1> Test Chamber (Stainless steel 500cc)

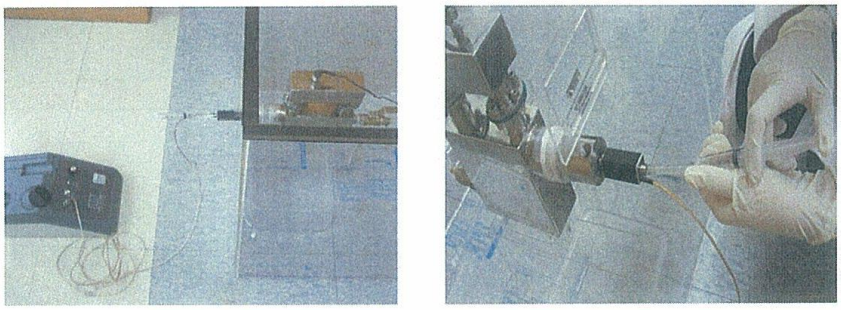
The inactivation test method was used to evaluate virus reduction with the following test procedure.

1. 0.5ml of Influenza A virus subtype H1N1 10^6EID50/ml (10^6 egg infectious dose 50/ml) was sprayed on the equipment with or without built-in SPi through an ultrasonic Nebulizer (Aerosol state less than 0.5 ㎛)

2. Make it react at laboratory temperature for 20 minutes.



<Figure 2> Test Diagram



<Figure 3> Test Procedure (A staged scene for the reason of security in the laboratory)

3. Obtain PBS after spraying 5ml phosphate buffer solution on the test device

4. Inoculate 10-fold diluted PBS into a 10-day chicken embryo.

5. Cultivate the inoculated chicken embryo in an incubator at 35℃ temperature for 48 hours and chill it in a freezer at 4℃ temperature.

6. Measure a virus in the lumbar spinal cord of the chilled chicken embryo by the hemagglutination test and log 10 egg infectious dose 50/ml ($log_{10}EID_{50}$/ml).

3. Results and Conclusions

Table 1 shows the result of four tests.

[Table 1] Result of Inactivation Test of Influenza A virus subtype H1N1

Sampling	Virus Titer [$log_{10}EID_{50}$/ml)				
	1st	2nd	3rd	4th	Average
Chamber under SPi-on	< 1.0	< 1.0	1.0	< 1.0	< 1.0
Chamber under Non-SPi	5.0	5.5	6.0	5.5	5.5

Virus titers ranged from 5.0 to 6.0 $log_{10}EID_{50}$/ml in non-SPi chamber, while all virus titers were less than 1.0 under SPi-on.

On the basis of the values of the virus titers in each SPi and non-SPi chamber, removal rate of Influenza A virus subtype H1N1 was calculated by the following formula under SPi-on.

$$\text{Removal efficiency (\%)} = \frac{EID_{50}\text{/ml of H1N1 (non SPi)} - EID_{50}\text{/ml of H1N1 (SPi)}}{EID_{50}\text{/ml of H1N1 (non SPi)}} \times 100$$

EID50/ml of H1N1 in non SPi Chamber = $10^{5.0}$ ~ $10^{6.0}$
EID50/ml of H1N1 in SPi Chamber = $10^{1.0}$

$$\text{Removal efficiency (\%)} = \frac{10^{5.0} - 10^{1.0}}{10^{5.0}} \times 100 \fallingdotseq 99.99\ \%$$

$$\text{Removal efficiency (\%)} = \frac{10^{6.0} - 10^{1.0}}{10^{6.0}} \times 100 \fallingdotseq 99.99\ \%$$

In conclusion, Influenza A virus subtype H1N1 was inactivated by over 99.99 % in the chamber under SPi-on.