

To: Mitsubishi Electric Corporation Nakatsugawa Works

## Test Report

Evaluation of airborne virus (phage) cross contamination  
for the LOSSNAY core

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1-15-1 Kitasato, Sagami-hara-shi, Kanagawa, Japan  
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The contents of this report should not be disclosed to the public without prior consent of the Kitasato Research Center of Environmental Sciences. The study results presented herein only apply to test samples and do not verify the quality of the entire batch (lot) of the test material.

## **1. Object**

The present test was conducted to verify that there is no airborne virus (phage) cross contamination from the outlet air to the inlet air of the LOSSNAY core in the heat exchange process.

## **2. Client**

Name: Mitsubishi Electric Corporation Nakatsugawa Works

Address: 1-3 Komaba-cho, Nakatsugawa-shi, Gifu, Japan

## **3. Institution and Analyst**

Name: Kitasato Research Center of Environmental Sciences

Address: 1-15-1 Kitasato, Sagamihara-shi, Kanagawa, Japan

Analyst: Microbiology Department

Shunji Okuda, Noriko Shimasaki

## **4. Test Period**

December 22, 2004

(Test materials was operated by engineers of your company)

## **5. Test Materials**

New LOSSNAY core "Hyper Element"

## **6. Organism**

### **6-1. Test virus**

*E.coli phage  $\phi$ X174* ATCC 13706-B

### **6-2. Host bacteria**

*Escherichia coli* ATCC 13706

### 6-3. Host bacteria culture

*Escherichia coli* (explained in 6-2.) was inoculated into 0.5% NaCl-added Nutrient Broth(Difco), and was cultivated overnight at 35°C. The resultant medium containing approximately  $10^9$  CFU/ml of host bacteria was used as host bacterium solution.

### 6-4. Test virus solution

*E.coli phage  $\phi$ X174* was mixed with host bacterium solution (explained in 6-3.) and cultivated. The resultant medium was filtrated by membrane filter owing to removal of *Escherichia coli*, and was diluted with sterile ion-exchanged water to obtain test virus solution of approximately  $10^7$  PFU/ml.

## 7. Method

### 7-1. Outline

The test apparatus is schematically shown in Fig. 1. The air flow rate was  $250 \text{ m}^3/\text{hr}$  in the outlet and inlet ducts intersecting each other at the LOSSNAY core. Air-sampling tubes were attached, with their openings against the air flow, at the each center of 4 sites, outlet duct upstream (location A) and downstream (location B) and inlet duct upstream (location C) and downstream (location D) of the LOSSNAY core.

The test was performed as follows: Test virus solution was sprayed from the upstream side of the outlet duct, and a specified quantity of air was then simultaneously sampled with midget impingers at 4 sites, locations A, B, C, and D around the LOSSNAY core to count the number of airborne viruses contained in the air.

### 7-2. Spray of test virus solution

The test virus was sprayed in the outlet duct at a pressure of

1kgf/cm<sup>2</sup> while supplying compressed air from the compressor into the nebulizer containing the test virus solution.

### 7-3. Sampling of airborne viruses

Airborne viruses were collected using the midget impinger as described below. Air in the duct was aspirated at a rate of 5 liters per minute for 4 minutes. Hence, a total of 20 liters of air was collected in 25 ml of sterile ion-exchanged water in the midget impinger.

### 7-4. Method for counting the number of viruses

Ion-exchanged water in the midget impingers, which possibly contained airborne viruses (*E. coli* phages), was used as the sample stock solution, and its 10-fold serial dilutions were then made. 0.2ml of the stock solution and each dilution were mixed with 0.2 ml of host bacterium solution of about 10<sup>9</sup> CFU/ml, and then mixed with 4.0 ml of soft agar for top layer. The mixture was then layered on the surface of 0.5% NaCl-added Nutrient Agar. The resultant medium was incubated for 18 hr at 35°C. The number of plaques formed was counted to determine the number of airborne viruses per 20 L of sampled air.

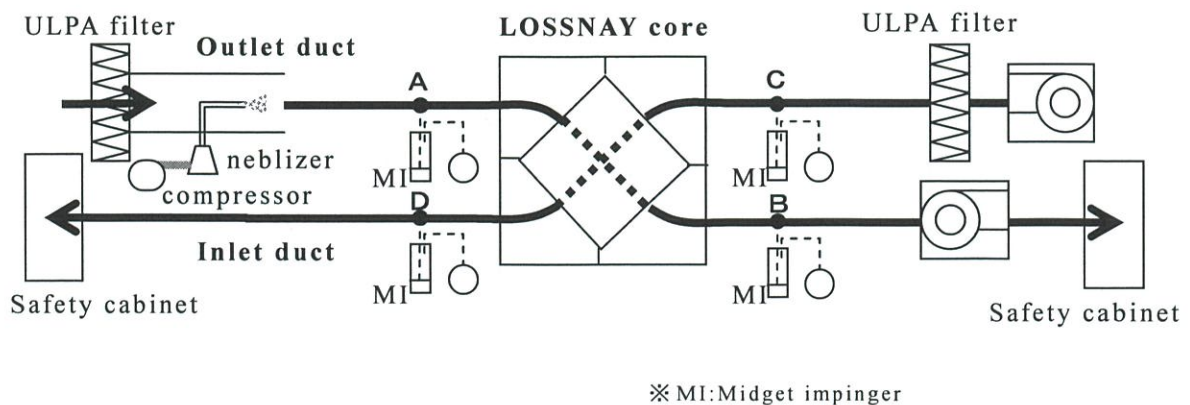


Fig. 1 Schematic diagram of the test apparatus



## 8. Test result

The concentration of test virus solution was  $1.2 \times 10^7$  PFU/ml.

The result of test is shown in table-1.

## 9. Consideration

The test virus was *E. coli phage  $\phi$ X174* with a small viral particle diameter (about 20 nm).

Test viruses were detected at locations A and B on the outlet side, from which the test virus solution was sprayed. In contrast, no test viruses were detected in 20L of sampled air at location C (in the air filtered by the ULPA filter) or location D (in the air crossed in the LOSSNAY core) on the inlet side. Therefore, it can be concluded that airborne viruses in the outlet side will not cross the dividers (specially processed paper) of the LOSSNAY core to the opposite inlet side even when heat is exchanged there.

Table-1 Airborne virus counts on each location

Test virus : *E.coli phage  $\phi$ X174* ATCC 13706-B

Test No.	Location A	Location B	Location C	Location D
1	$3.1 \times 10^2$	$2.8 \times 10^2$	< 1	< 1
2	$4.4 \times 10^2$	$1.2 \times 10^2$	< 1	< 1
3	$1.9 \times 10^2$	$6.2 \times 10$	< 1	< 1
Average	$3.1 \times 10^2$	$1.5 \times 10^2$	< 1	< 1

(Unit of measurement: PFU/20L-air)