

TEST REPORT

Test No.29D-BT-388

October 3, 2017

Requested by : PH+Cleanee Inc.

Test sample : JJJee

Test item : Anti-bacterial test

Study director : Noritoshi Ri



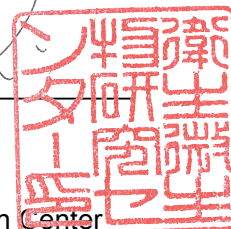
Noritoshi Ri

Director D.V.M., Ph.D.

Hygiene & Microbiology Research Center

21-14, 7-chome, Aoto, Katsushika-ku

Tokyo Japan



Anti-bacterial test

1. Purpose

The test was conducted to estimate the anti-bacterial activity of the sample.

2. Test Sample

JJee

3. Test bacteria

Streptococcus pneumoniae NBRC 102642

4. Test Method

1) Preparation of the cell suspension

The test bacteria was cultured on 5% Sheep blood agar (KOHJIN BIO Co.,Ltd) for 24~48 hours anaerobically at 37°C. After incubation, the culture was suspended in normal saline solution and the solution contain $10^{7\sim 8}$ cells per 1mL.

The dilution was used as the cell suspension for inoculation.

2) Anti-bacterial test procedures

Transfer 0.1 ml of cell suspension to the 10 mL of test solution and incubated at 25°C. After incubations, the 10-fold dilutions of test solution were prepared using sterilized normal saline solution. And each dilution were dispensed to 5% Sheep blood agar. And also, the normal saline solution was used as control. The results were calculated from the numbers of colonies of each test plates by appropriate dilution series.

5. Test Results

The results of the anti-bacterial activity test of the sample were shown in table 1-1, 1-2.

Table 1-1. Results of Anti- bacterial Test of JJJee against *Streptococcus pneumoniae*.

Test sample	Initial	Viable cell count (cfu /mL)		
		10 seconds	30 seconds	1 minutes
JJJee	2.0×10^5	1.4×10^5 (6.7%)	9.1×10^4 (54.5%)	1.3×10^4 (92.8%)
Control	2.0×10^5	1.5×10^5	2.0×10^5	1.8×10^5

Normal saline solution was used as control.

Table 1-2. Results of Anti- bacterial Test of JJJee against *Streptococcus pneumoniae*.

Test sample	Initial	Viable cell count (cfu /mL)		
		15 minutes	30 minutes	1 hours
JJJee	2.0×10^5	$<10^1$ (>99.9%)	$<10^1$ (>99.9%)	$<10^1$ (>99.9%)
Control	2.0×10^5	1.8×10^5	1.3×10^5	1.7×10^5

Normal saline solution was used as control.