

EMSL Analytical, Inc.
Microbiology Division
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Certificate of Analysis

Product: Mesosilver®

Project: Antimicrobial effectiveness of colloidal silver

EMSL Reference Number: 090402726

Experimental Design Summary:

Test survival of vancomycin-resistant *Staphylococcus aureus* subsp. *aureus* American Type Culture Collection Strain No. 700699 in two Mesosilver products (20 and 75 ppm) using two (1% and 10%) concentrations of product as supplied. The microorganism was tested for survival at four (0, 2, 5, and 24 hour) time points. A negative control (no product) was included for comparison. The treatments were performed in triplicate in sterile phosphate buffer with MgCl₂ (Hardy Diagnostics) and incubated without continuous mixing at 35°C then plated in duplicate on nutrient agar for 72 hours. Results were reported as mean ± standard deviation. Media sterility controls showed no growth.

Experimental Results Summary:

Vancomycin-resistant *Staphylococcus aureus* subsp. *aureus* inoculated at 2.4×10^8 cells ml⁻¹ was used to determine the effect of Mesosilver colloidal silver on bacterial survival. The results show that both Mesosilver products have a negative impact on the survival of *S. aureus* subsp. *aureus* when used at 1% and 10% concentrations (Tables 1 and 2). After 5 hours, 1% and 10% 75 ppm Mesosilver both reduced the numbers of *S. aureus* subsp. *aureus* cells to below the level of detection. The remaining tested concentrations were successful in reducing the number of cells to below the level of detection within 24 hours.

Analyst _____
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Antimicrobial effectiveness of Mesosilver 20 ppm colloidal silver

20 ppm Mesosilver (%)	CFU ml ⁻¹		
	2h	5h	24h
0	$3.4 \times 10^8 \pm 3.0 \times 10^6$	$3.2 \times 10^8 \pm 1.6 \times 10^6$	$9.8 \times 10^7 \pm 7.6 \times 10^5$
1.0	$3.9 \times 10^7 \pm 3.5 \times 10^6$	$1.8 \times 10^6 \pm 5.8 \times 10^5$	<1
10.0	$5.5 \times 10^6 \pm 1.8 \times 10^6$	$6.7 \times 10^5 \pm 5.8 \times 10^5$	<1

Table 1. Survival of vancomycin-resistant *Staphylococcus aureus* subsp. *aureus* ATCC No. 700699 inoculated at 2.4×10^8 cells ml⁻¹ in the presence of 1% and 10% 20 ppm Mesosilver colloidal silver.

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Antimicrobial effectiveness of Mesosilver 75 ppm colloidal silver

75 ppm Mesosilver (%)	CFU ml ⁻¹		
	2h	5h	24h
0	$3.4 \times 10^8 \pm 3.0 \times 10^6$	$3.2 \times 10^8 \pm 1.6 \times 10^6$	$9.8 \times 10^7 \pm 7.6 \times 10^5$
1.0	$4.2 \times 10^6 \pm 1.9 \times 10^6$	<1	<1
10.0	$5.0 \times 10^5 \pm 5.0 \times 10^5$	<1	<1

Table 2. Survival of vancomycin-resistant *Staphylococcus aureus* subsp. *aureus* ATCC No. 700699 inoculated at 2.4×10^8 cells ml⁻¹ in the presence of 1% and 10% 75 ppm Mesosilver colloidal silver.

Antimicrobial effectiveness of Mesosilver colloidal silver

Treatment	Time Point (hours)					
	2		5		24	
	Colony Count	Dilution Factor	Colony Count	Dilution Factor	Colony Count	Dilution Factor
1.0% 20 ppm Mesosilver-1	34 / 39	1000000	2 / 1	1000000	0 / 0	1000000
1.0% 20 ppm Mesosilver -2	44 / 43	1000000	3 / 2	1000000	0 / 0	1000000
1.0% 20 ppm Mesosilver -3	40 / 38	1000000	2 / 1	1000000	0 / 0	1000000
10% 20 ppm Mesosilver -1	3 / 4	1000000	0 / 0	1000000	0 / 0	1000000
10% 20 ppm Mesosilver -2	6 / 6	1000000	1 / 1	1000000	0 / 0	1000000
10% 20 ppm Mesosilver -3	8 / 6	1000000	1 / 1	1000000	0 / 0	1000000
1.0% 75 ppm Mesosilver -1	2 / 2	1000000	0 / 0	1000000	0 / 0	1000000
1.0% 75 ppm Mesosilver -2	7 / 3	1000000	0 / 0	1000000	0 / 0	1000000
1.0% 75 ppm Mesosilver -3	8 / 3	1000000	0 / 0	1000000	0 / 0	1000000
10.0% 75 ppm Mesosilver -1	1 / 0	1000000	0 / 0	1000000	0 / 0	1000000
10% 75 ppm Mesosilver -2	1 / 1	1000000	0 / 0	1000000	0 / 0	1000000
10% 75 ppm Mesosilver -3	0 / 0	1000000	0 / 0	1000000	0 / 0	1000000

Table 3. Raw data of antimicrobial effectiveness of colloidal silver against *S. aureus* subsp. *aureus*. Average colony count x dilution factor = colony forming units per ml (CFU ml⁻¹).