

Certificate of Analysis

Product: Mesosilver[®]

Project: Phase II: *Staphylococcus aureus*

EMSL Reference: 030319513

Experimental Design Summary:

Test survival of *Staphylococcus aureus* American Type Culture Collection Strain No. 6538 in two Mesosilver products (20 and 75 ppm) using 2 (1 and 10%) concentrations of product as supplied. The organism was tested for survival at 4 (0, 2, 5, and 24 h) time points. A negative control (no product) was included for comparison. All tests were performed in triplicate and plated in duplicate.

Experimental Results Summary:

Staphylococcus aureus at 8.3×10^5 cells ml⁻¹ was used to determine the effect of Mesosilver on bacterial survival. The results show that Mesosilver has a negative impact on the survival of *S. aureus* (Tables 1 and 2), however only the 10% concentration of the 75 ppm product was successful in reducing the numbers of cells to below the level of detection within 24 hours.

Analyst _____
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Date: 12-15-03

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Table 1. Survival of *Staphylococcus aureus* ATCC 6538 inoculated at 8.3×10^5 cells ml⁻¹ in the presence of 1 and 10% 20 ppm Mesosilver[?] colloidal silver.

20 ppm Mesosilver (%)	CFU ml ⁻¹		
	2 h	5 h	24 h
0	$8.9 \times 10^5 \pm 8.1 \times 10^4$	$5.7 \times 10^5 \pm 3.0 \times 10^4$	$9.3 \times 10^2 \pm 5.0 \times 10^2$
1.0	$7.9 \times 10^5 \pm 6.5 \times 10^4$	$4.9 \times 10^5 \pm 6.9 \times 10^4$	$6.8 \times 10^1 \pm 1.4 \times 10^1$
10.0	$2.2 \times 10^5 \pm 2.9 \times 10^3$	$3.3 \times 10^2 \pm 1.1 \times 10^2$	0.5 ± 0.5

All treatments performed in triplicate in 0.35% NaCl incubated without continuous mixing at 35°C. All plate counts performed in duplicate using nutrient agar incubated at 35°C for 72 hours. Results reported as mean \pm standard deviation. Media sterility controls showed no growth.

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Table 2. Survival of *Staphylococcus aureus* ATCC 6538 inoculated at 8.3×10^5 cells ml⁻¹ in the presence of 1 and 10% 75 ppm Mesosilver² colloidal silver.

75 ppm Mesosilver (%)	CFU ml ⁻¹		
	2 h	5 h	24 h
0	$8.9 \times 10^5 \pm 8.1 \times 10^4$	$5.7 \times 10^5 \pm 3.0 \times 10^4$	$9.3 \times 10^2 \pm 5.0 \times 10^2$
1.0	$6.8 \times 10^5 \pm 7.8 \times 10^4$	$2.5 \times 10^5 \pm 1.9 \times 10^4$	$1.9 \times 10^1 \pm 5.4$
10.0	$3.6 \times 10^4 \pm 2.3 \times 10^3$	2.7 ± 1.0	<1

All treatments performed in triplicate in 0.35% NaCl incubated without continuous mixing at 35°C. All plate counts performed in duplicate using nutrient agar incubated at 35°C for 72 hours. Results reported as mean \pm standard deviation. Media sterility controls showed no growth.

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Treatment	Time Point (hours)					
	2		5		24	
	Colony Count	Dilution Factor	Colony Count	Dilution Factor	Colony Count	Dilution Factor
0.35% Saline-1	88/93	10,000	52/57	10,000	5/3	100
0.35% Saline-2	98/95	10,000	63/58	10,000	15/5	100
0.35% Saline-3	81/80	10,000	56/58	10,000	17/11	100
1.0% 20ppm Mesosilver-1	76/81	10,000	36/47	10,000	86/80	1
1.0% 20ppm Mesosilver-2	91/80	10,000	48/62	10,000	74/54	1
1.0% 20ppm Mesosilver-3	75/70	10,000	49/52	10,000	65/48	1
10% 20 ppm Mesosilver-1	19/26	10,000	50/40	10	1/1	1
10% 20 ppm Mesosilver-2	23/21	10,000	46/18	10	0/0	1
10% 20 ppm Mesosilver-3	25/20	10,000	23/23	10	0/1	1
1.0% 75ppm Mesosilver-1	60/60	10,000	20/25	10,000	27/23	1
1.0% 75ppm Mesosilver-2	76/75	10,000	27/24	10,000	18/17	1
1.0% 75ppm Mesosilver-3	73/65	10,000	25/27	10,000	13/16	1
10% 75ppm Mesosilver-1	29/37	1,000	3/4	1	0/0	1
10% 75ppm Mesosilver-2	34/40	1,000	2/1	1	0/0	1
10% 75ppm Mesosilver-3	39/35	1,000	5/2	1	0/0	1

average colony count x dilution factor = colony forming units per ml