

# **Silver Sol Prevents Microorganisms From Becoming Resistant, A Discussion.**

**By**

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## **Abstract**

Silver Sol was compared to several antibiotics against numerous bacterial strains to determine if Silver Sol could destroy the microbes and control mutation better than antibiotics. Silver Sol and the following antibiotics were tested: Ofloxacin, Tetracycline, Penicillin G, and Cefaperazone. The microbial strains tested include: *S. gorondii*, *S. mutans*, *S. pyogenes*, *E. coli*, *S. pneumoniae*, *S. typhimurium*, *Enterobacter*, *Pseudomonas aeruginosa*, *S. faecalis*, *Shigella* and *Staph aureus*. The broth macrodilution susceptibility test was used to determine a minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the silver hydrosol and antibiotic concentrations.

The results include the following observations:

The 10 ppm Silver Sol inhibits MRSA.

The 10 ppm Silver Sol is the minimum inhibitory concentration.

The 15 ppm Silver Sol completely killed the MRSA and prohibited the MRSA from growing for 24 hours.

The 15 ppm Silver Sol controls MRSA and disallows any mutation to grow.

These results indicate that Silver Sol kills MRSA and all other bacteria tested, and disallows any mutation.

## **Procedures**

The broth macrodilution susceptibility test (14) was used to determine the minimum inhibitory concentration (MIC) and the minimum bactericidal concentration (MBC) of the Silver Hydrosol™ and antibiotic preparations. The broth macrodilution susceptibility test was also used to compare the activity of the Silver Hydrosol™ to that of each of the antibiotics.

Streptococcal cultures were prepared by inoculating the bacteria into tryptic soy broth, (TSB), (Difco) and incubating at 37°C in a 5% CO<sub>2</sub> atmosphere until mid-log phase growth was reached. All other bacteria were prepared by inoculating the bacteria in Mueller-Hinton Broth, MHB, (Difco) and incubating at 37°C until mid-log phase was reached. The bacterial suspensions were then adjusted to a 0.5 McFarland Standard using a spectrometer (Spectronic 301, Milton Roy) and diluted one thousand-fold to approximately 10<sup>5</sup> CFU/ml.

One millilitre of culture was then added to each of 10 two-fold serial dilutions in either TSB (streptococcal species) or MHB (all other species) containing the antimicrobial to be tested. This made the concentration of antimicrobial in the first tube 5ppm, 2.5ppm in the second tube, so that each subsequent tube contained half the concentration of antimicrobial that the previous tube contained. Cultures were incubated for 24 hours and the MIC was defined as the lowest concentration of the Silver Hydrosol™ or antibiotic that prevented growth as determined visually by noting the presence or absence of turbidity in the tube after incubation. The MBC's were demonstrated by removing a 0.1-ml aliquot from the non turbid tubes and plating it immediately onto tryptic soy agar (enriched with 5% sheep blood) for streptococcal species, or Mueller-Hinton agar for all other bacteria. The MBC was defined as the lowest concentration of the Silver Hydrosol™ or antibiotic allowing the growth of fewer than 10 colonies.

In order to determine if the bacteria used in this study were able to become resistant to the SilverSol, the following tests were conducted: The bacteria were incubated at the level determined to be their MIC, and then the concentration of the SilverSol. Of particular interest was the Staphylococcus aureus (MRSA strain) since it appears to have a very high rate of mutation. It already had an MIC of 10 ppm, which became the baseline concentration and was titrated up from this starting point. It managed to “grow” (it turns out to just be inhibitory) at that concentration, as determined by starting a new culture at 15 ppm to see if the new “mutant” would grow.

## Materials and Methods

The following antimicrobials and bacterial strains were used: Erythromycin (Westwood Pharmaceuticals, New York), Ofloxacin (Sigma, St. Louis, Missouri), Tetracycline (Sigma), Penicillin G (Sigma), and Cefaperazone (Sigma) were used. Antibiotics were diluted to a concentration of 10 ppm ( $\mu\text{g}/\text{ml}$ ) and used immediately for each test. The Silver Hydrosol™ was obtained from American Biotech Labs (Alpine, UT). Concentrations of silver in the Silver Hydrosol™ were determined by American Biotech Labs using an atomic absorption spectrometer (Perkin Elmer) and the following bacterial strains were used to test the antibacterial activity of the Silver Hydrosol™ and to compare this activity with that of the antibiotics listed above. *Streptococcus gordonii* [*S.gordonii*] (ATCC 10558), *Streptococcus mutans* [*S. mutans*] (ATCC 25175), *Streptococcus pyogenes* [*S. pyogenes*] (ATCC 19615), *Escherichia coli* O157:H7 [*E. coli* O157:H7] (ATCC 43895) were obtained directly from the American Type Culture Collection (ATCC). *Streptococcus pneumoniae* [*S. pneumoniae*] (ATCC 6303), *Klebsiella pneumoniae* [*K. pneumoniae*] (ATCC 13883), *S. typhimurium* (ATCC 14028), *E. coli* (S.E. Luria Strain B ATCC 11303), *Enterobacter aerogenes* [*E. aerogenes*] (ATCC 13048), *P. aeruginosa* (ATCC 27853), *Streptococcus faecalis* (*S. faecalis*), *Shigella boydii* (*S. boydii*) [Utah Valley Regional Medical Centre clinical isolate, Provo, Utah], and *Staphylococcus aureus* (*S. aureus*) [non-haemolytic Utah Valley Regional Medical Centre clinical isolate, Provo, Utah] were obtained from the Brigham Young University, Department of Microbiology, Clinical Laboratory Science bacterial collection. *Klebsiella oxytoca* (*K. oxytoca*) [Provo River, Utah isolate], *Salmonella arizonae* (*S. arizonae*) [Provo River, Utah isolate], *Enterobacter cloacae* (*E. cloacae*) [Provo River, Utah isolate] were gifts from the Central Utah Water Conservancy District and were determined to be either 10 ppm, or 20 ppm ( $\mu\text{g}/\text{ml}$ ) for this study.

## **Results**

The 10 ppm Silver Sol liquid, killed the initial bacteria indicating the Minimum Inhibitory Concentration. In the 15 ppm sample the MRSA and all other bacteria tested completely failed to increase over a period of 24 hours at 37 degrees, indicating that Silver Sol kills MRSA and disallows any mutation.

To emphasize the idea that bacteria cannot become resistant to silver, Roy et; in their paper Materials Research Innovations 2007 Vol 3 no. 1 pointed out that of the 278,000 kg to metallic silver eaten in India each year, they have never found a bacteria that is resistant to it.

Also, in a white paper just published by Prince and Prince in Orthopedic Design and Technology, they determined that it would take at least 6 mutations in the same bacterial cell in order to become resistant to the silver.

## **Conclusion**

Silver Sol kills MRSA and all other bacterial strains tested and prohibits any mutation This is significant because Silver Sol will not be burdened with the same detrimental resistance problems that currently afflict commonly prescribed antibiotics. This means Silver Sol can be safely and effectively used for every day use without causing the bacteria to mutate and become resistant.