

Efficacy of Virkon™ against mineralised Nanobacteria

The term Nanobacteria is short for its scientific genus & species name *Nanobacterium Sanguineum* ('blood nanobacteria'). Nanobacteria were discovered in 1988 by Nobel Prize Nominees Dr. Neva Ciftcioglu, PhD and Olavi Kajander, MD, PhD as a contaminant in mammalian cell cultures that killed the mammalian cells in their cell culture research. Nanobacteria are from 20-200 nanometers in size (a nanometer is 1 billionth of a meter and or the width of ten hydrogen atoms side-to-side) and are the smallest known self-replicating bacteria.

Nanobacterium sanguineum is newly recognized as an emerging infectious disease. Nanobacteria have been shown to cause coronary artery disease and vascular disease. Its DNA, RNA and Lipopolysaccharide (LPS) profiles have been accurately mapped.

Nanobacteria can be cultured from the blood of humans and mammals. When compared to regular bacteria, Nanobacteria are 1/100 to 1/1,000 the size, allowing them to easily move around into other cells and invade/infect them. Nanobacteria have been shown to kill human cells, including immune cells and/or other bacteria. Nanobacteria cause alteration of cellular RNA and DNA gene-expression patterns in cells they infect. This process can lead to genetic alteration, abnormal cell growth and proliferation. When compared to other bacteria, Nanobacteria grow very slowly, only reproducing every 3 to 6 days, whereas regular bacteria reproduce in minutes or hours. Because of this and their small size, Nanobacteria can only be seen using electron and atomic force microscopes. Nanobacteria cannot be grown in standard culture media and can only be grown in mammalian blood or serum.

Nanobacteria are pleomorphic which means that they have different physical forms during their life cycle and can change appearance and physical form during their growth, development or in response to changing environmental factors. Nanobacteria have a distinctively unique and unusual cell membrane structure. They secrete a calcific biofilm around themselves that provides protection as well as allowing for multiple nanobacteria to connect, collaborate and function together as a unit or colony. The calcific biofilm allows the Nanobacteria to act like slime molds that can expand, contract and move. The calcific layer is carbonate apatite, a calcium phosphate mineral ($\text{Ca}_5(\text{PO}_4, \text{CO}_3)_3\text{F}$) similar to bone (which is hydroxyapatite)

In order to eradicate nanobacteria, one must first dissolve their protective calcium shells from around them and then kill them.

An acidic oxidising biocide such as Virkon would be expected to be effective on two fronts. Firstly, the acidity will cause the calcific layer to dissolve. Secondly, oxidising biocides are effective at destroying biofilms. Recent published work confirms the efficacy of Virkon against nanobacteria.

Efficacy of Chemical Reagents against Nanobacteria (mineralised)

Chemical	Exposure Time	
	10 min	30 min
70 % Ethanol	NE	NE
2 % Glutaraldehyde	NE	NE
4 % Formaldehyde	NE	NE
0.5 % Hypochlorite	NE	NE
3 % Peroxide	NE	NE
1M HCl	nd	RS *
1 M NaOH	NE	NE
1 % SDS	NE	NE
1 % Tween 80	NE	NE
1 % Triton X-100	NE	NE
3M Guanidium - HCl	nd	MRS*
3 M Urea	NE	NE
1 % Virkon ®	nd	NS*
1.5 % Erifenol ®	NE	NE
1 % Klorilli ®	NE	NE
3 % Buraton ®	nd	RS*

Key :

NE No Effect

RS Reduces Survival

MRS Markedly Reduces Survival

NS No Survival

nd Not Determined

* Partial or total deattachment on exposure

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