Ag OXYSALTS™ Technology – A Novel Silver Antimicrobial

Helen A. Thomason, PhD[1,2]

[1]*KCI, An Acelity Company. King Edward Court, King Edward Road, Knutsford, WA16 0BE, UK.*
[2]*Faculty of Biology, Medicine and Health, University of Manchester, Manchester, M13 9PT, UK.*

Dr Thomason is Director of Research Sciences at KCI, An Acelity Company and holds an honorary position at The University of Manchester where for over 15 years she has conducted her research on skin biology and wound healing. She has published multiple peer-reviewed manuscripts and book chapters focussing on understanding the cellular and molecular mechanisms of healing. Her research centres on developing clinically relevant models of delayed wound healing, including biofilm infection and using these models to understand how wound therapies can stimulate repair.

WOUND INFECTIONS AND BIOFILMS

Wound infection can cause a significant delay or prevent a wound from healing. Therefore, the appropriate management of infection is important to reduce healing times and the associated wound management costs. One of the main complications associated with wound infection is that the infection often manifests as a biofilm.[1] Biofilms differ from free-floating, planktonic bacteria in that the bacteria form communities which adhere to a surface, such as the wound bed. Within these communities, bacteria secrete an extracellular polymeric substance (EPS) composed of polysaccharides, proteins, lipids and extracellular DNA. Bacteria within a biofilm are more tolerant to antimicrobial therapies, in part due to the slow growth rate of the bacteria within the biofilm and the protective nature of the EPS.[2] Therefore, potent antimicrobials are required to effectively kill bacteria within a biofilm.

ANTIMICROBIAL ACTION OF SILVER

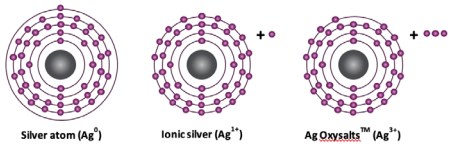
For centuries, silver has been known for its antimicrobial properties[3] and exploited for medicinal use.[4,5] It has a long standing history in the control wound infection, initially in solution or creams for the use in burns and more recently incorporated into wound dressings which deliver controlled and prolonged release of silver.[6] Many different silver delivery systems have been used in wound care products, ranging from colloidal silver, silver sulfadiazine, silver proteins, silver salts, silver compounds, and nanocrystalline silver.

Metal silver (Ag^0) is inert; it must be converted into its metallic state (Ag^+) to exert antimicrobial action. In its metallic form, a silver atom contains a single electron in its outermost shell. When this electron is removed during a process called oxidation the silver becomes positively charged (Ag^{1+}). In its ionic form silver scavenges electrons from bacterial cells. In doing so it disrupts the bacterial cell wall, interferes with DNA replication and denatures proteins involved in bacterial metabolism, ultimately resulting in the bacterial death.

The efficacy of silver depends on a number of factors; the concentration of silver present, its solubility and the composition of the dressing/treatment itself. In addition, once silver is released from the dressing the wound environment can affect its efficacy; protein and chloride ions within wound fluid bind silver and thus reduce the concentration of bioavailability silver. Silver salts became popular in the 1960s when positively charged silver ions were complexed to negatively charged ions to produce electrically neutral compounds (AgCl, $AgNO_3$, $AgSO_4$), providing a more stable delivery system for silver. When exposed to wound fluid, these compounds break down to release ionic silver (Ag^{1+}).

Ag OXYSALTS™ Technology in wound care, silver-based therapies contain metallic silver (Ag^0) or singly ionic silver (Ag^{1+}). More recently a new silver technology, Ag OXYSALTS™ Technology (Ag_7NO_{11}) has been developed which can produce higher ionic states of silver (Ag^{1+}, Ag^{2+} and Ag^{3+}). Higher ionic states of silver are highly reactive and unstable, needing to fill their electron shells to gain stability.[7] However, they can be stabilised with oxygen atoms to form Ag_7NO_{11}. Incorporation of this silver salt into wound dressings allows controlled delivery of higher ionic states of silver to the wound. When exposed to aqueous media, such as wound fluid, Ag OXYSALTS™ Technology break down to produce three ionic states of silver Ag^{1+}, Ag^{2+} and Ag^{3+} **(Figure 1).**[8] The gaining of electrons by silver is known as reduction. The higher the reduction potential the stronger the affinity for electrons. The reduction potential for Ag^{1+} is 0.80 whereas for Ag^{2+} and Ag^{3+} the reduction potential is 1.98 and 1.80, respectively. Thus, these higher ionic states of silver have a greater affinity for electrons.

Figure 1. Atoms are composed of positively charged nuclei surrounded by shells of negatively charged electrons (-). Metallic silver (Ag^0) contains an equal number of positively charged protons within its nuclei and negatively charged electrons in the surrounding shells. When metallic silver loses an electron from its outer most shell it becomes positively charged, ionic silver (Ag^{1+}). Ag OXYSALTS™ Technology lose up to 3 electrons forming higher ionic states of silver (Ag^{1+}, Ag^{2+} and Ag^{3+}).



AG OXYSALTS™ TECHNOLOGY - ANTIMICROBIAL AND ANTI-BIOFILM EFFICACY

Having a greater reduction potential or stronger affinity for electrons would indicate stronger antimicrobial action. In vitro studies have shown that a lower equimolar concentration of Ag OXYSALTS™ Technology is required to eradicate planktonic bacteria, prevent biofilm formation and eradicate established biofilms compare to Ag_2O, AgO, Ag_2SO_4, $AgNO_3$, silver sulfadiazine, and $CuSO_4$.[8] Furthermore, Ag OXYSALTS™ Technology were shown to be superior to other silver compounds at reducing biofilm biomass.[8] Subsequent studies have shown Ag OXYSALTS™ Technology to have superior antimicrobial and anti-biofilm efficacy against dual species biofilms, which are more difficult to eradicate, compared to $AgNO_3$ and $CuSO_4$.[9] In addition, Ag OXYSALTS™ Technology have been shown to be effective against biofilms composed of multi-drug resistant bacteria.[10]

SILVER CYTOTOXICITY

There has been a long-standing concern over the safety of silver therapies in wound care.[11] The concerns are two-fold: firstly, the systemic absorption of silver and secondly local cytotoxic effects which may be detrimental to healing. The effects that silver dressings have on wound cells independent of infection are often overlooked, particularly as antimicrobials

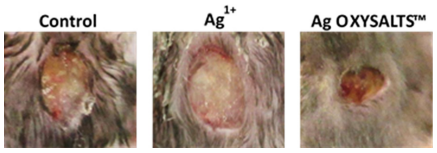
become more potent in attempt to combat biofilm infection. Many chronic wounds are stuck in the inflammatory phase of healing whereby heightened levels of inflammatory cells persist for prolonged periods of time within the wound. These inflammatory cells create tissue damage and a hostile wound environment that delays healing. Antimicrobials therefore need to be potent enough to combat wound infection, but not cause a cytotoxic effect on healing that may exacerbate an already hostile wound environment.

The effects of Ag OXYSALTS™ Technology on healing independent of infection has been examined. Ag OXYSALTS™ Technology had no effect on the closure of fibroblast scratch wounds. In contrast, Ag OXYSALTS™ Technology promoted the healing of keratinocyte scratch wounds.[12] The ability of Ag OXYSALTS™ Technology to promote healing independent of infection was also confirmed in a murine wound model, whereby wounds treated with Ag OXYSALTS™ Technology dressings were significantly smaller, with greater re-epithelialisation and reduced inflammation compared to control treated wounds **(Figure 2).** How Ag OXYSALTS™ Technology promote healing independent of infection has been investigated. Ag OXYSALTS™ Technology were found to be the only silver compound to release oxygen during their breakdown.[12] The release of oxygen may be sufficient to shift wounds out of a hypoxic state or provide additional oxygen to the wound to support cell migration. In addition, dressings containing Ag OXYSALTS™ Technology were able to catalyse the breakdown of hydrogen peroxide to oxygen and water.[12] Hydrogen peroxide plays a vital role in the early stages of wound healing;[13] however, in chronic wounds high numbers of inflammatory cells which release hydrogen peroxide as a mechanism of killing bacteria contribute to damaging levels within the in the wound. Ag OXYSALTS™ Technology may therefore contribute to reducing detrimental levels of hydrogen peroxide within a wound and in doing so release oxygen during this process.

SILVER TOLERANCE AND RESISTANCE

The increased use of silver in wound care has raised concerns over bacteria developing tolerance or resistance to silver therapies. Resistance is defined as the inherited ability of bacteria to grow in high silver concentrations

Figure 2. Ag OXYSALTS™ Technology promote healing independent of infection. Uninfected excisional mouse wounds treated for 3 days with control or silver dressings. In contrast to wounds treated with silver chloride dressings (Ag^{1+}), wounds treated with Ag OXYSALTS™ Technology (Ag^{3+}) show accelerated healing compared to control treated wounds.



irrespective of the duration of treatment.[14] In contrast, tolerance is the temporary ability of bacterial to survive high silver concentrations which would normally be lethal. Tolerance may or may not be inherited; however, unlike resistance tolerance in is often achieved by slowing down bacterial growth and reducing metabolism. Therefore, a longer exposure to silver, rather than a higher concentration, is required to kill tolerant bacteria compare to susceptible bacteria.[14] The molecular basis of silver resistance is well understood. Bacteria display resistance through the expression of silver resistance genes; however, many studies have shown limited presence of these resistance genes in clinical isolates.[15;16] Furthermore, although bacteria may harbour silver resistant genes, in many cases they do not express the genes to a significant level and therefore display little or no resistance to low levels of silver.[17] The likelihood of developing silver resistance is reduced if the silver has rapid and sustained antimicrobial activity.[18] Studies have found that dressings containing Ag OXYSALTS™ Technology were effective against bacteria expressing silver resistant genes when other silver dressings were ineffective.[19]

SUMMARY

In wound care, a wide variety of silver formulations have been used to treat wound infections. There has been a recent increase in the use of wound dressings incorporating silver compounds; however, clinical concerns still remain over their ability to combat biofilm infections, and whether they cause local cytotoxicity or adversely affect healing independent of infection. Dressings containing Ag OXYSALTS™ Technology are the only commercially available silver dressings which produce silver at higher oxidative states (Ag^{2+} and Ag^{3+}). Despite being potently antimicrobial and effectively killing bacteria within a biofilm, Ag OXYSALTS™ Technology do not have an adverse effect on healing; on the contrary, in vivo studies have shown Ag OXYSALTS™ Technology to promote healing independent of infection.

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