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SILVER WHITE PAPER

Everything you ever wanted to know about the use of silver in wound therapy

Prepared by Sharon Lindsay of Systagenix

Systagenix

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Contents

Part 1: The use of silver in wound therapy

1. Introduction

Silver has a well established history as a broad-spectrum antimicrobial, with early reports of silver use dating back to ancient times. More recently, silver nitrate and silver sulphadiazine (SSD) have been used for the management of infected wounds and burns. Recent advances in technology have enabled silver to be incorporated into a range of dressing materials. The popularity of using such silver dressings in clinical practice has steadily increased, however, the scientific basis for their use is not always fully understood. The current Silver White Paper summarises the use of silver and silver dressings in wound management. Part 1 provides a general background on silver as an antimicrobial, including details on its mode of action, potential for resistance and safety profile. Part 2 provides an overview of the current available commercial silver dressings.

Silver has a well established history as a broad-spectrum antimicrobial

2. Brief history on the use of silver as an antimicrobial

The use of silver as an antimicrobial agent has an impressive history, going back as far as ancient Greece and Rome when silver coins were used to sterilise drinking water (Figure 1).

The first reports on the use of silver to prevent infection date back to 1834 when the German obstetrician, Crede, used a 1% silver nitrate solution to prevent blindness from post-partum infection in newborns. Following the discovery of antibiotics in the 1930s, the use of silver-based products to combat infection declined. However, over use and mis-use of antibiotics has resulted in the emergence of antibiotic resistant bacteria such as methicillin resistant *Staphylococcus aureus* (MRSA) and vancomycin resistant Enterococcus (VRE). Due to the increase in bacterial resistance towards antibiotics, since the 1960s the use of silver-based products as topical antimicrobials has become increasingly popular.

FIGURE 1. AN ANCIENT ROMAN COIN. THE ROMANS USED SILVER COINS TO STERILISE DRINKING WATER

Silver nitrate and compresses have been used for the management of burn wounds (Moyer et al. 1965; Price et al. 1966; Sawhney et al. 1989). The addition of a sulphonamide antibiotic to silver (SSD) generated a potent antimicrobial and is still used for the topical treatment of burns (Monofo 1996; Sweetman 2004) and infected wounds (Clarke 1999). Silver has also been used to coat catheters to prevent infection (Maki et al. 1988; Tobin and Bambauer 2003).

Despite the historical use of silver nitrate and SSD, these agents are limited by their cytotoxicity including delayed wound healing, electrolyte disturbance, inactivation of patient enzymes and increased occurrence of leucopoenia, argyria and resistance (Lowbury et al. 1976; Heggers and Robson 1978; Maillard and Denyer 2005). The cytotoxicity of SSD has been attributed to the release of the sulphonamide moiety rather than the silver component (Lockhart et al. 1983).

Recently the development of new technologies have enabled various formats of silver to be directly incorporated into a range of dressing materials and benefit from an improved efficacy, safety and resistance profile over silver nitrate and SSD. Use

of such dressings in clinical practice is steadily increasing, with approximately £100 million spent in 2006-2007 on prescribing costs (National Prescribing Centre 2008).

KEY POINTS

- *Silver has a well established history as an antimicrobial.*
- *SSD and silver nitrate are frequently used for the management of burn wounds.*
- *Renewed interest in silver as an antimicrobial has been fuelled by the emergence of antibiotic resistant bacteria.*
- *The development of new materials and technologies has enabled silver to be directly incorporated into a range of dressings and benefit from an improved efficacy, safety and resistance profile over silver nitrate and SSD.*

3. Chemistry and antimicrobial properties of silver

To understand the antimicrobial properties of silver, an appreciation of the chemistry of silver is first required.

Silver is an element represented by the symbol "Ag".

FIGURE 2. THE PERIODIC TABLE SHOWING THE "Ag" SYMBOL FOR SILVER. NOTE THAT THE PERIODIC TABLE INDICATES THAT ELEMENTAL SILVER HAS 47 PROTONS

Similar to other elements, silver consists of three types of subatomic particles: "protons" and "neutrons" (located in the atomic nucleus) and "electrons" (located orbiting the atomic nucleus). Protons and electrons have a positive and negative charge, respectively, whereas neutrons are neutral. The charge of any atom is determined by the number of electrons relative to the number of protons. For example, atoms with more electrons than protons have a negative charge, whereas atoms with more protons than electrons have a positive charge. Conversely, atoms with equivalent number of w2 and protons are neutral. With respect to silver, silver can exist in one of the following two common forms:

- As a neutral atom (with 47 electrons and 47 protons) - referred to as "elemental silver" or "metallic silver" (Figure 2)
- As a positively charged atom (with 46 electrons and 47 protons) - referred to as "ionic silver" or "silver cation" (Ag^{+}).

Silver cations (Ag^{+}/ionic silver) are potent antimicrobials (Ovington 2004; Landsdown and Williams 2004) and may become available when silver is presented in solution (e.g. silver nitrate) or when elemental silver is in the presence of oxygen, as described below.

The oligodynamic effect of silver is well recognised, with as little as 10^{-9} to 10^{-6} mol/L silver cations effective against a broad range of microorganisms including Gram-positive and Gram-negative bacteria, fungi, protozoa and viruses (Russel and Hugo 1994). By comparison, elemental silver is relatively unreactive. However, in presence of oxygen from the air or dissolved in aqueous environments such as body fluids and wound exudates, elemental silver oxidises to form silver oxide ($Ag^{+}_{2}O^{2-}$). On dissolution in fluid, silver oxide dissociates into its separate components releasing the antimicrobial silver cations. Thus, irrespective of the presentation of silver in wound care products, silver achieves its antimicrobial effect by releasing silver cations. Common microorganisms that silver products can kill are detailed in Figure 3.

Once silver is in the ionic form, how does silver confer antimicrobial activity? Despite the wide spread use of silver as an antimicrobial, the exact mechanism(s) of action is yet to be fully determined (Drug and Therapeutics Bulletin, 2010). Silver cations are thought to interact with multiple sites within the target cell

(Figure 4). A likely mechanism of action is that the positively charged silver cations bind to negatively charged components of the bacterial cell. Binding of silver cations to the negatively charged cell wall and membrane will induce structural changes and cell lysis. Likewise, binding of silver cations to negatively charged proteins, enzymes, DNA and RNA will interfere with bacterial electron transport, cell division and cell replication (Lansdown 2002). Similarly, silver is likely to demonstrate antimicrobial activity against fungi and viruses by binding to negatively charged moieties.

KEY POINTS

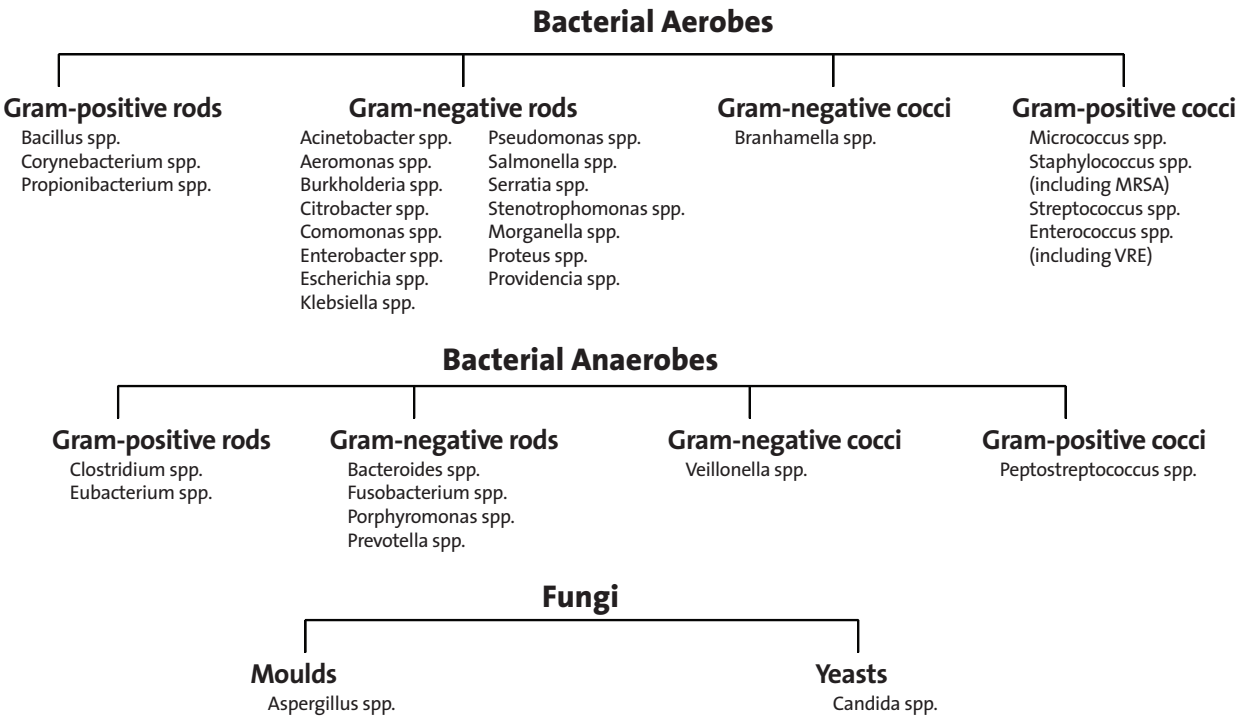
- *Silver can exist in "elemental" or "ionic" form*
  - *Silver ions are also referred to as "Ag^+" or "silver cations".*
- *All silver-containing products, whether elemental or ionic, achieve their antimicrobial effect via the action of silver cations.*
- *Elemental silver exists as a neutral atom and is relatively unreactive.*
  - *In the presence of oxygen, elemental silver oxidises to form silver oxide and upon dissolution in fluid (such as wound exudate), silver oxide dissociates into separate components releasing antimicrobial silver cations.*
- *Ionic silver is positively charged and has antimicrobial activity.*
- *The broad-spectrum antimicrobial activity of silver is thought to be attributed to the silver cations interfering with electron transfer and binding to the following negatively charged moieties in the target cell:*
  - *Cell wall*
  - *Cell membrane*
  - *DNA*

4. Silver in wound care

Wounds may arise through a range of circumstances such as surgical incisions or trauma, or arterial, venous or diabetic foot ulcers, and may be either acute or chronic. A brief description of acute and chronic wounds and suitable use of silver for each condition are provided in Section 4.1 and 4.2, respectively.

4.1 Overview of acute wounds

Most wounds, regardless of the aetiology, heal without difficulty (Grey et al. 2006) and are frequently called "acute" wounds. In such wounds, healing normally progresses via a series of phases (haemostasis, inflammation, granulation and remodelling) to restore the integrity of the skin. However, transient microbial flora/infection may be problematic in acute wounds, particularly if trauma has occurred from a contaminated object. Under such circumstances, application of a silver dressing may be recommended



MRSA: Methicillin resistant *Staphyloccus aureus*.
VRE: Vancomycin resistant Enterococcus.

FIGURE 3. BROAD-SPECTRUM ANTIMICROBIAL ACTIVITY OF SILVER. THE FIGURE DETAILS THE ANTIMICROBIAL ACTIVITY OF THE SILVERCEL® ANTIMICROBIAL ALGINATE DRESSING

either prophylactically or to control wound infection.

KEY POINTS

- *The healing process occurs via the following phases: haemostasis, inflammation, granulation and remodelling.*
- *Transient microbial flora/infection may be problematic in acute wounds: application of silver dressings can control wound infection in acute wounds.*

4.2 Overview of chronic wounds

In some patients, wounds fail to progress to healing in a predictable amount of time for the given wound type and are termed "chronic". Factors that may impede the wound healing process include: inadequate blood supply, obesity, smoking, malnutrition, advancing age, immobility and microbial infection (Grey et al. 2006).

The majority of non-healing or "chronic" wounds are colonised by bacteria, although high numbers of bacteria are required for the wound to be considered clinically "infected". Most wound infections are "polymicrobial" in nature, meaning that the infection is attributed to a number of different species of microbes. Aerobic pathogens such as *Pseudomonas aeruginosa* and *Staphylococcus aureus* are among the most frequently cited as the cause of delayed healing (Bowler et al. 2001). Historically, it is considered that bacterial levels over 10^5 colony-forming units per gram of tissue indicate an infected wound and may impair the wound healing process (Bendy et al. 1964; Teplitz et al. 1964). More recently, the polymicrobial interactions and presence of bacteria-specific virulence factors within wounds have also been implicated in hindering wound healing (Trengove et al. 1996; Bowler, 2003).

Chronic wounds are "stuck" in the inflammatory phase and typically show high levels of pro-inflammatory cytokines (IL-1 and TNF-alpha) and proteases (matrix metallo proteases [MMPs] and elastase), and low levels of protease inhibitors and active growth factors. Bacterial endotoxins or lipopolysacharides (LPS) from bacteria present at the wound site also induce the production of pro-inflammatory cytokines, further increasing the inflammatory status of chronic wounds (Falanga 2004; Moseley et al. 2004; Schultz et al. 2004; Quatresooz et al. 2002).

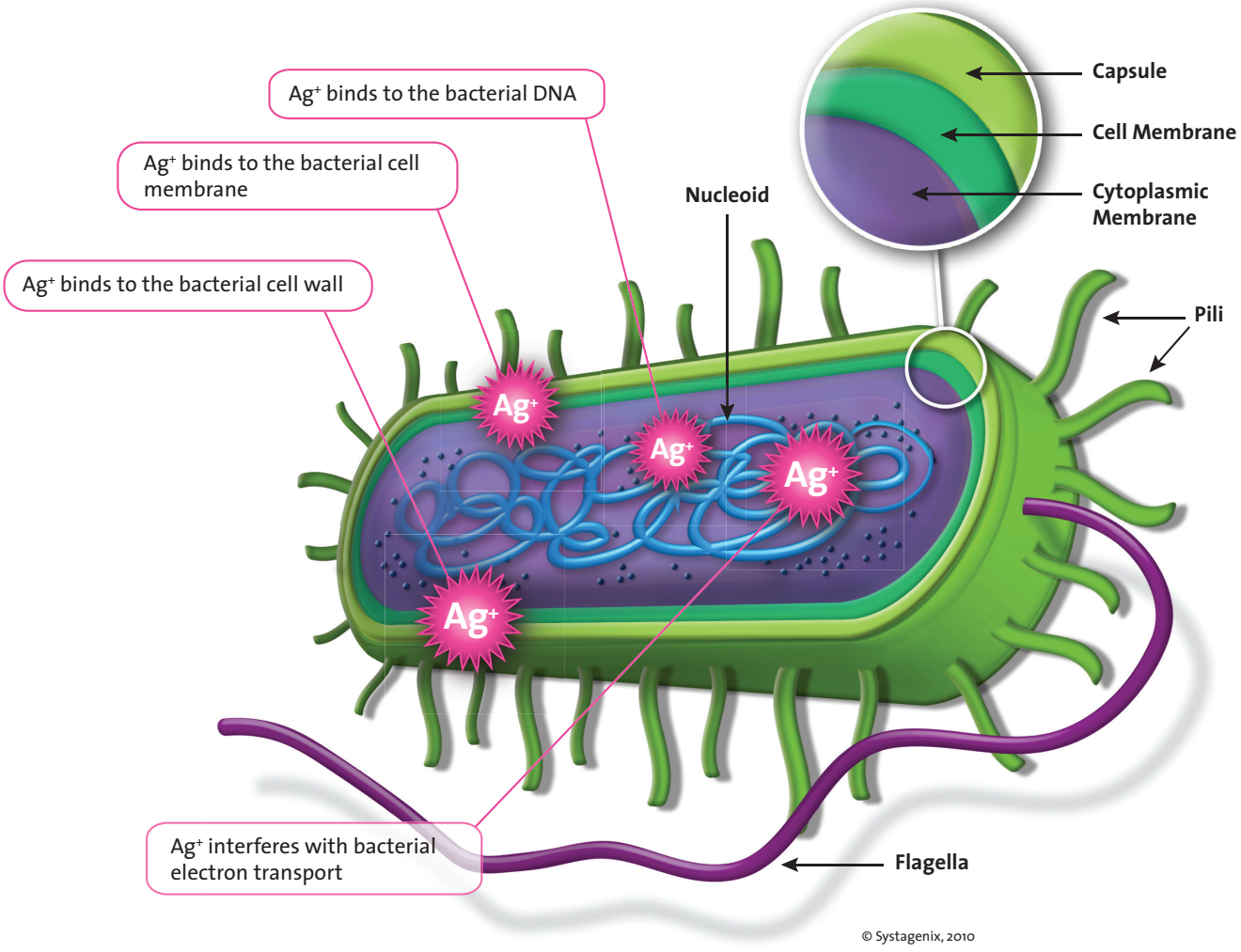


FIGURE 4. BACTERIAL TARGET SITES OF SILVER CATIONS (Ag^+)

Application of silver dressings to chronic wounds may aid wound healing by controlling wound bioburden, thereby contributing toward progressing the wound beyond the inflammatory phase.

KEY POINTS

- *Historically bacterial levels over 10^5 colony-forming units per gram of tissue are considered to impair the wound healing process.*
- *Most chronic wound infections are polymicrobial.*
- *Chronic wounds are "stuck" in the inflammatory phase: application of silver dressings can control wound bioburden and contribute toward progressing the wound beyond the inflammatory phase*

4.3 Silver dressings

The development of innovative and sophisticated materials together with the use of new technologies has increased the number of silver dressings available on the market (Cutting et al. 2009). Silver materials include: alginate, activated charcoal, carboxymethylcellulose (CMC), films, hydrocolloids, nanocrystalline/nanoparticles and polyurethane foams (Pal et al. 2009; Thomas et al. 2009), and also collagen, hydrofibre and hydrogel.

Silver is typically presented in dressings in either elemental or compound form (Lansdown and Williams 2004). (A description of elemental silver is provided in Section 3; the term "compound" silver refers to the presentation of silver in the active, ionic form. This may be achieved, for example as silver nitrate or SSD). Examples of dressings containing elemental or compound silver are listed below. Further examples of silver dressings are provided in Part 2.

- Elemental silver dressings
  - Coated fibres: SILVERCEL® Antimicrobial Alginate Dressing (SILVERCEL®)
  - Nanocrystalline coating: ACTICOAT
  - Silver and charcoal combination: ACTISORB® Silver 220 Activated Charcoal Dressing (ACTISORB® Silver 220)
- Silver compound dressings
  - SSD: Urgotul SSD/S.Ag and Allevyn Ag
  - Silver-oxidised regenerated cellulose (ORC) salt: PROMOGRAN PRISMA® Wound Balancing Matrix (PROMOGRAN PRISMA® Matrix)
  - Silver-CMC salt: AQUACEL Ag

Irrespective of the presentation of silver in dressings, silver confers its antimicrobial effect by releasing silver cations (see Section 3 for details on silver chemistry). Of note, however, is that different brands of silver dressings will vary with respect to their silver release profile due to the presentation of silver and the initial silver content. In addition, the amount of silver cations released into the wound environment will be affected by the production and viscosity of wound exudates, extracellular matrix components and the frequency of dressing changes.

Elemental silver dressings typically contain high levels of silver. This results in a sustained silver release, with the dressing acting as a reservoir for the formation and release of silver cations. By comparison, dressings with silver compounds usually contain lower levels of silver and are likely to release silver over a shorter time frame in the wound or be depleted, depending on wound exudates levels. In the presence of fluid, the separate components of compound silver dissociate and release the antimicrobial silver cations.

KEY POINTS

- *Silver materials include: alginate, activated charcoal, CMC, chitosan films, collagen, films, hydrocolloids, hydrofibre, hydrogel, nanocrystalline/nanoparticles and polyurethane foams.*
- *Silver is typically presented in dressings in either elemental or compound form.*
- *Irrespective of the presentation of silver in dressings, silver confers its antimicrobial effect by releasing silver cations.*
- *Dressings with silver compounds usually contain lower levels of silver than elemental silver dressings and are likely to release silver over a shorter time frame in the wound.*

5. Assessing the *in vitro* silver release profile of dressings

Healthcare companies devote a considerable amount of research into monitoring the *in vitro* release of silver from dressings. Dressings that release silver in a controlled and sustained manner rather than a short burst of silver benefit in that they provide continuous antimicrobial activity and minimise potential adverse events. In clinical practice, it is also important to remember that wound exudate is often produced and the sustained efficacy of a formulation depends on the bioavailability of silver ions under these conditions.

Silver released from dressings *in vitro* is typically expressed in "ppm", i.e. one part per million, equivalent to 1 mg/L. However, currently there is no standard method to evaluate the silver release from dressings. Method variation in different wound care companies include: the size of dressing, the type of solution and volume of solution used to simulate wound exudate and incubation time. In addition, silver concentration may be analysed by either atomic absorption (total silver) or use of an electrode (ionic silver only).

Variations in experimental design will invariably make cross study comparisons difficult, therefore caution must be exercised when comparing the silver release profile of dressings derived from different studies. Lindsay et al. (2010) noted that the amount of silver released from a range of dressings was consistently lower for dressings immersed in de-ionised water or saline solutions than solutions containing albumin. This is likely to be due to albumin enhancing the solubility of silver. The authors argue that a simulated wound fluid containing approximately 2% albumin should be used in *in vitro* evaluations to reflect the protein concentration observed in the exudate of a chronic wound. This concentration of albumin is supported by clinical assessments of wound fluid exudates (Falanga 1992; Harris et al; 1995; James et al. 2000; Trengrove et al. 2000).

KEY POINTS

- *The ideal silver dressing releases silver in a controlled and sustained manner.*
- *Silver released from dressings is typically expressed in ppm.*
- *Method variation among different wound care companies can result in conflicting results, even when the same dressings are tested. Variations in experimental design make cross study comparisons difficult.*
- *A simulated wound fluid containing approximately 2% albumin should be used for in vitro evaluations to reflect the protein concentration observed in the exudate of a chronic wound.*

6. Microbiological methods to assess efficacy of silver dressings

Microbiological *in vitro* assays to assess the efficacy of silver dressings include the zone of inhibition assay and the $\log_{10}$ reduction assay and are described below. These methods have been adapted from recognised international standard methods frequently used to assess the efficacy of suspensions of antimicrobials and disinfectants.

6.1 Zone of inhibition assay

The zone of inhibition assay provides a qualitative assessment of the susceptibility of a surface-cultured microorganism to an antimicrobial agent. In this assay, a dressing sample is placed in the middle of the pre-inoculated plate and incubated. If the strain is susceptible to the dressing no growth, or a "zone of inhibition", will be observed surrounding the sample. An indication of the longevity of antimicrobial action of the dressing can be achieved by re-challenging the dressing over a number of days, whereby the dressing sample is transferred to a fresh inoculated plate and similarly incubated. The bactericidal activity of the dressing can also be observed by taking a swab from underneath the dressing and streaking onto fresh agar: no growth following incubation indicates that the dressing is bactericidal.

Variations in the zone of inhibition method between different companies include pre-wetting the dressing with simulated wound fluid to simulate an exudative wound, inoculum concentration and incubating the inoculated plates prior to exposure to the dressing. These variations in experimental design may ultimately generate different results when evaluating the same dressings.

6.2 $\log_{10}$ reduction assay

The $\log_{10}$ reduction assay provides a quantitative assessment of the performance of an antimicrobial agent against a planktonic ("free-floating") suspension of microorganisms. This assay investigates how much of the microbial population is killed when incubated in the presence of the test agent within a set time frame (e.g. 60, 120 and 180 minutes). Results are expressed in $\log_{10}$ reduction values ($\log_{10}$ initial count – $\log_{10}$ final count).

Variations in the $\log_{10}$ reduction method among different companies include inoculum concentration, exposure time to the antimicrobial and use of a "neutraliser" to inhibit the action of the antimicrobial after the given exposure time. Similar to variations within the zone of inhibition test, variations in experimental design may ultimately generate different results when evaluating the same dressings.

KEY POINTS

- *Antimicrobial activity of wound care products can be assessed by zone of inhibition and $\log_{10}$ reduction assays.*

- *Variations in experimental design of the zone of inhibition method and the log_{10} reduction method may ultimately generate different results when evaluating the same dressings.*

7. Concentration of silver cations required to exert an antimicrobial effect

Some authors argue that silver concentrations as low as 1 ppm are capable of exerting an antimicrobial effect, whereas others suggests that much higher concentrations of silver are required (Brett 2006). Details of Systagenix silver dressings and their antimicrobial activity are provided in Part 2.

But is there a correlation between the amount of silver release and the antimicrobial effect? An *in vitro* comparison of a range of silver dressings demonstrated that there was no relationship between silver release and antimicrobial activity (White and Cutting 2006; Jones et al. 2005; Parsons et al. 2009).

7.1 Clinical practice

While the abundance of silver dressings has increased the therapeutic options in wound care, this is often confounded by confusion over selection of the most suitable dressing. According to Maillard and Denyer (2005) the ideal antimicrobial dressing should have sustained antimicrobial action over the entire surface of the wound, provide a moist wound healing environment, enable monitoring of the wound with minimal interference, manage wound exudate, be comfortable and conformable, provide an effective microbial barrier, adsorb and retain microorganisms and avoid trauma upon removal. A number of elemental and compound silver dressings have been developed which address these considerations. But with so much choice, how does the clinician know which dressing to select?

Managing wound infection involves careful assessment of patients and their wounds, appropriate care planning and selection of dressings. In short, the needs of the patient together with the individual characteristics of the dressing must be considered. Clinicians need to consider manufacturers´ recommendations for product use, for example some products have to be pre-wetted prior to use, whereas some products are more suited for highly exuding wounds while others are more suitable for low exuding wounds (Moore and Romanelli 2006). If a patient has a highly exudative wound, an alginate may be appropriate. If the presence of bacterial toxin or malaodour is a problem, a dressing with activated charcoal may be considered. For irregular shaped wounds, enhanced conformability of the dressing, reducing the occurrence of "dead space" where bacteria may flourish (Cutting et al. 2009) could be addressed by using a dressing that transforms into a gel or foam within the wound bed. A further aim may be to reduce the occurrence of pain upon dressing removal, particularly for friable tissue or painful venous leg ulcers and arterial ulcers. Under such circumstances a non-adherent dressing that minimises trauma and pain during application and removal should be chosen. It may also be appropriate to use a dressing that is capable of treating infection since wound infection can also be a source of pain (Mudge and Orsted, 2010).

KEY POINTS

- *Managing wound infection involves careful assessment of patients and their wounds, appropriate care planning and selection of dressings.*
- *The ideal antimicrobial dressing should:*
  - *Provide sustained antimicrobial action*
  - *Provide a moist wound healing environment*
  - *Enable monitoring of the wound with minimal interference*
  - *Manage wound exudate*
  - *Be comfortable and conformable*
  - *Provide a microbial barrier*
  - *Adsorb and retain bacteria*
  - *Avoid trauma upon removal.*

8. Safety considerations and appropriate use

8.1 Cytotoxicity

The cytotoxic effect of silver dressings can be determined *in vitro* with fibroblasts and keratinocytes cultivated as a monolayer. Of note, however, that cross study comparisons are difficult due to the varying cell culture conditions that may be used in different laboratories, e.g., the keratinocyte cell type and the methodology used. Despite the cytotoxicity, it is important to highlight that there is a role for silver in wound management. In a heavily infected wound, the priority must be to clear the infection rather than to try to heal the wound. Once the bioburden level is reduced, the wound and dressing

selection can then be re-evaluated to generate an appropriate treatment regimen.

Like other biocides, silver is non-specific in action and is cytotoxic to both microbial and mammalian cells, including cells present at the wound site such as fibroblasts and keratinocytes. In other words, silver dressings cannot discriminate between pathogenic bacteria and healthy cells involved in wound healing. While this may not be problematic for wounds that are overtly infected or heavily colonised, silver containing products should be used cautiously on wounds with a low bioburden and on epithelising wounds. Innes et al. (2001) showed that re-epithelisation was significantly slower in wounds treated with silver dressings compared with non-antimicrobial dressings (14.5 +/- 6.7 days versus 9.1 +/- 1.6 days; p=0.004). Moreover, in a comparative *in vitro* study investigating the cytotoxicity of the silver dressing AQUACEL Ag, ACTICOAT, PolyMem Silver and Urgotul SSD, all dressings investigated were shown to be cytotoxic for fibroblasts and keratinocytes (Burd et al. 2007). In addition, all dressings tested induced a significant delay in epidermal cell proliferation (Burd et al. 2007). The authors also showed that ACTICOAT and Contreet foam inhibited wound epithelisation when tested *in vivo* using mice models (Burd et al. 2007). Similarly, Van Den Plas and colleagues (2008) showed that silver dressings induced apoptosis in cells involved in wound healing and concluded that such dressings should only be used on critically contaminated wounds.

PROMOGRAN PRISMA® Matrix dressing provides an alternative for controlling bioburden in wounds with low levels of infection or prophylactically. This dressing has been shown to provide a simultaneous antimicrobial effect without causing injury to host cells. Under *in vitro* conditions, PROMOGRAN PRISMA® Matrix exerted a positive effect on cell proliferation of host cells such as keratinocytes and endothelial cells (ETRS Newsletter, 2005). PROMOGRAN PRISMA® Matrix dressing, together with other Systagenix silver dressings, is discussed further in Sections 13 and 14.

With respect to the use of silver dressings in clinical practice, Thomas and McCubin (2005) analysed the wound exudates and tissue from seven patients and showed that silver accumulation is proportional to the viscosity and protein content of such material. These results indicate that "excess" silver cations are bound to protein and other ions present at the wound site. The authors concluded that this may protect the cells involved in wound healing from the cytotoxicity of silver.

KEY POINTS

- *In a heavily infected wound, the priority must be to clear the infection rather than to try to heal the wound.*
- *Like other biocides, silver is non-specific and can interact negatively with both mammalian and microbial cells.*
- *Silver containing products should be used cautiously on wounds with a low bioburden and on epithelising wounds.*

8.2 Absorption

Lansdown (2005) showed that silver is naturally found at low concentrations in individuals without prior exposure to silver, although the metal has no recognised physiological function. Reported concentrations of silver were as follows: blood <2.3 µg/L; urine 2 µg/day; liver 0.05 µg/g wet tissue and kidney 0.05 µg/g wet tissue (Lansdown and Williams 2005).

With regard to systemic absorption of silver in patients treated with silver dressings, absorption of silver appears to be proportional to the wound area and dressing application. In a study involving patients with burns (n=30), the maximum serum silver concentration was related to the wound area exposed to the silver dressing and frequency of dressing application (Vlachou et al. 2007). It was observed in the follow-up visit three months later that the silver serum concentrations had returned to near-baseline levels in the majority of patients. Moreover, although use of silver in burns and chronic wounds may lead to circulatory silver absorption and deposition in organs including the liver and kidney, the risks of prolonged tissue damage is considered low (Lansdown and Williams 2004).

KEY POINTS

- *Silver is found in the human body at low concentrations but has no recognised physiological function.*
- *Absorption of silver appears to be proportional to the wound area and dressing application.*
- *Risk of prolonged tissue damage from silver absorption and deposition is considered low.*

8.3 Argyria and silver deposition

Argyria, a general term used to describe a grey-blue discolouration of the skin and mucus membranes, is caused by deposition of silver. It is not considered harmful by the Toxic Substances and Disease Registry, but considered a cosmetic issue that many people may find undesirable and socially debilitating.

Argyria is an adverse event that may be associated with environmental exposure, ingestion of silver or extensive SSD use (Walker et al. 2006). The amount of discolouration depends upon the route of silver delivery, together with the individual's ability to excrete silver (Walker et al. 2006).

There have been limited reports in the literature of argyria-like symptoms observed following use of modern wound care dressings such as ACTICOAT. Instances include a patient with 30% burns treated with ACTICOAT who was noted to have argyria to the face and lips, and blood and urine levels of 107 µg/g and 28 µg/g, respectively (Lansdown 2002). Following discontinuation of treatment with ACTICOAT, blood and urine silver levels returned to normal and the wound healed (Lansdown 2002). Wang et al. (2009) observed skin discolouration following ACTICOAT application to porcine deep dermal partial thickness burns, with the severity of discolouration correlating with the length of time of application.

Silver deposition has also been reported in heavily exudating wounds in patients treated with ACTICOAT and Contreet foam whereby grey-black deposits were observed in the wound bed (Lansdown 2002; Clennett and Hoskin 2003; Lium 2003; Lansdown and Williams 2004). However, these deposits were removed following washing the wounds (Lansdown 2002; Clennett and Hoskin 2003; Lium 2003; Lansdown and Williams 2004). In addition, in *ex vivo* wound models using human skin collected from surgical waste, the wound models treated with ACTICOAT showed more deposits than other treatments tested (AQUACEL Ag, FLAMAZINE, PolyMem Silver and SilvaSorb, and also silver nitrate). Black discolouration was observed in the epidermal layer when exposed to ACTICOAT for 14 days (Fredriksson et al. 2009). Black or grey deposits were also noted in cells at the wound margins and around the blood vessels in the dermal tissue.

There have been no reports of argyria or silver deposition for SILVERCEL®, ACTISORB® Silver 220 or PROMOGRAN PRISMA® Matrix in clinical practice.

KEY POINTS

- *Argyria is an adverse event associated with environmental exposure or ingestion of silver or extensive SSD use.*
- *Argyria-like symptoms have been observed on occasion with modern silver dressings such as ACTICOAT.*
- *Silver deposition has been observed with some modern silver dressings but the condition is temporary.*
- *There have been no reports of argyria or silver deposition for SILVERCEL®, ACTISORB® Silver 220 or PROMOGRAN PRISMA® Matrix in clinical practice.*

8.4 Potential limitations of use

Silver dressings have few contraindications for use. However, SSD dressings are contraindicated in pregnancy and in neonates, and in patients with severe renal or hepatic impairment. SSD is also contraindicated in patients sensitive to sulphonamides or those with large wounds (Joint Formulary Committee 2009; Drug and Therapeutics Bulletin, 2010).

A further consideration is that silver should be used cautiously on wounds with a low bioburden or epithelialising wounds. In addition, as a precaution, silver products should not be used during magnetic resonance imaging (MRI) scans or on patients with a hypersensitivity toward silver other components of the product.

9. Resistance profile

Extensive and uncontrolled use of silver in medical products and consumer products such as silver-coated mints in Japan and supermarket-available colloidal and "silver-gelatine" for washing vegetables in Mexico, have led to concerns over the development of silver-resistant bacteria (White and Cutting, 2006; Silver 2003).

Bacterial resistance may be genetic. In genetic resistance, resistant genes may be passed from parent to offspring (vertical genetic transfer). In addition, some bacteria have the ability to "acquire" resistance by a process of horizontal genetic transfer from one bacterium to another, as has been reported for antibiotic resistance (Percival et al. 2005). The transfer of the resistant genes may take place via plasmids or transposons. Molecular biology techniques such as

polymerase chain reaction (PCR) have been developed to identify the presence of resistance genes within the bacterial genome. However, such techniques are limited in that the choice of primers dictates what genes are targeted.

In addition to genetic resistance, resistance may also be phenotypic. Phenotypic resistance is the result of changes in the expression of a gene. Specifically, phenotypic resistance may be due to a range of factors including: reduction of update, reduction of the agent to a less toxic state, expression of efflux pumps or the production of neutralising compounds. With respect to silver, these phenotypic responses could result in silver resistance by:

- Decreasing intracellular accumulation of silver, either by reduction of uptake of silver cations or by actively increasing the efflux of silver cations (efflux pumps)
- Increasing production of neutralising compounds such as chelation of the silver cations sulfyhydril groups of metal binding proteins
- Reduction of the silver cations to the metallic form, establishing a less toxic oxidation state (Clennett et al. 2003).

Silver resistance has been documented in a range of bacteria from patients treated with SSD and silver nitrate, most of which have been isolated from burn wounds (Gupta and Silver 1998; White and Cutting 2006). Silver resistance has also developed *in vitro*. By exposing *Escherichia coli* to sequential increases in silver nitrate and SSD, Li and colleagues (1997) demonstrated that *E. coli* was able to tolerate high concentrations of silver (>1024 ppm). The authors suggest that efflux of silver cations may have attributed to the silver resistance.

Despite the selection of silver resistant bacteria *in vitro*, there have been limited instances of silver resistance isolated from clinical samples. Indeed, Chopra et al. (2007) emphasise that there have been fewer than 20 publications of bacterial silver resistant clinical isolates since 1975. In 1998, for example, Gupta and Silver isolated a silver resistant Salmonella isolate from a burns ward and determined that silver resistance was attributed to a plasmid containing seven genes and two open reading frames encoding a silver binding protein (SilE), a two component mRNA regulatory system (SilS and SilR) and efflux pumps (SilCBA and SilP). The authors also noted that closely related genes are found in other bacteria from clinical and environmental samples and conclude that uncontrolled use of silver may result in the development of silver resistant bacteria. In addition, Lansdown and Williams (2007) isolated bacterial cultures from 30 patients with chronic leg ulcers. All bacterial isolates were cultured on agar containing sequential increases in the concentration of silver nitrate. All isolates, with exception to an *Enterobacter cloacae* strain, were inhibited by 1 mM silver nitrate. The *E.cloacae* strain was isolated from a 79 year old lady with venous leg ulcers who had previously been treated with silver dressings (the *E.cloacae* strain is currently being analysed further at the genetic level). Despite the isolation of the silver resistant *E.cloacae* strain, the authors note that for the other bacterial isolates, prolonged exposure did not lead to silver resistant bacteria. However, the authors also emphasise that the full extent of bacterial resistance is still not fully understood and the lack of evidence and technical expertise in wound clinics to analyse silver resistant bacteria may contribute to bacterial resistance.

Given the limited publication of silver resistance from clinical isolates, the overall consensus would appear that silver resistance is generally considered rare due to its multiple target sites (see Section 3 for details on mode of action of silver). The dissemination of silver resistant bacteria among the community would appear unlikely since repeated sub-culture of silver resistant bacteria *in vitro* usually renders the isolates sensitive to silver (Silver 2003). Moreover, since silver has multiple points of attack, the development of a series of mutations that result in resistance to all mechanisms of action in a single generation seems unlikely (Li 1997; Ovington 2004; Percival 2005; Chopra 2007) and to date, no transfer of silver resistance has been reported. Importantly, silver demonstrates antimicrobial action in resistant bacteria including MRSA, VRE and bacteria with silver resistant genes (Percival et al. 2008; Loh et al. 2009; McInroy et al. 2009).

KEY POINTS

- *The extensive use of silver in medical and consumer products has lead to concerns over the development of silver resistant bacteria.*
- *Resistance can be genetic or phenotypic.*
  - *Resistance genes may be passed vertically from parent to offspring or distributed between bacteria*

by a process of horizontal gene transfer (genetic resistance)

  - *Resistance may be attributed to changes in gene expression, e.g. reduction of update, reduction of the agent to a less toxic state, or expression of efflux pumps or production of neutralising compounds (phenotypic resistance).*

- *Despite the selection of silver resistant bacteria in vitro, there have been limited instances of silver resistant bacteria isolated from clinical samples.*
- *Silver resistance is considered rare due to its multiple target sites.*
- *Silver demonstrates antimicrobial action in resistant bacteria including MRSA, VRE and bacteria with silver resistant genes.*

10. Biofilms

In addition to antimicrobial activity being reduced by the occurrence of genetic or phenotypic resistance (Section 9), the presence of a biofilm within a wound may impede the action of antimicrobials.

It has become apparent over the past thirty years or so that in nature, bacteria exist predominately as biofilms, whereby bacteria are found in association with surfaces enclosed within an exopolymer matrix (Costerton et al. 1978; 1987; Gilbert et al. 2002). Indeed, it is estimated that over 80% of chronic infections are caused by biofilms (Lewis 2001). Moreover, biofilms have been recently associated with chronic wound infections (James et al. 2008; Percival et al. 2008; McInroy et al. 2009; Hill et al. 2010; Lipp et al. 2010; Werthén et al. 2010).

What makes biofilms so significant in a clinical setting is their extreme tolerance towards antimicrobial treatment agents and ability to resist the host immune defences, such as in a chronic wound environment. Data derived from *in vitro* models suggest that silver is readily antimicrobial against biofilms that are a few days old. For example, a 90% kill of *in vitro* biofilm-associated *P.aeruginosa, S.aureus* or *E.cloacae* was achieved following a 24 hour exposure to a silver hydrofibre dressing (AQUACEL Ag); total kill was achieved within 48 hours (Percival et al. 2008). Similar results were obtained with SILVERCEL® NON-ADHERENT Dressings (SILVERCEL® NON-ADHERENT) tested against 24 hour *in vitro* "peg-based" (MBEC model) biofilms of *P.aeruginosa*, MRSA and VRE (McInroy et al. 2009, 2010). In addition, using a drip flow *in vitro* biofilm model, Lipp et al. (2010) demonstrated that SILVERCEL® has a favourable antibiofilm formation activity against *P.aeruginosa* and MRSA *in vitro*.

Hill et al. (2010) investigated the effect of silver dressings on more mature biofilms. Using the constant depth film fermenter (CDFF), the authors observed limited antimicrobial activity on silver dressings when tested on *in vitro* seven day-old biofilms, highlighting the recalcitrant properties of well-established biofilms.

KEY POINTS

- *In nature bacteria exist predominately as biofilms.*
- *It is estimated that over 80% of chronic infections are caused by biofilms.*
- *Biofilms demonstrate an extreme tolerance towards antimicrobial treatment agents and ability to resist the host immune defences.*
- *Numerous companies are currently testing the antimicrobial activity of their silver dressings against biofilms.*
- *Silver dressings have demonstrated various degrees of antimicrobial activity against in vitro biofilms.*
- *Limited data are available on the antimicrobial action of silver in relation to chronic wounds; available in vitro data suggest that silver is less effective of eradicating mature biofilms compared with younger biofilms.*

11. Overview: The use of silver in wound therapy

Silver has a well established history as an antimicrobial and is currently receiving increasing interest within the medical community for the topical treatment of wound infections due to its favourable efficacy, safety and resistance profile. Moreover, developments in technology have enabled several silver types to be incorporated into a range of different dressing materials and benefit in an improved safety profile over formulations of silver nitrate and SSD. However, as a non-specific antimicrobial agent, silver dressings should be replaced with non-antimicrobial dressings or dressings with low silver concentrations (such as PROMOGRAN PRISMA® Matrix) once the wound bioburden is reduced.

There are currently a number of silver dressings available on the market; these are described in Part 2.

Part 2: Commercial silver dressings

12. Commercial silver dressings

Clinical use of silver dressings for the treatment of chronic wounds has dramatically increased in recent years, with an estimated £100 million spent in 2006-2007 on prescribing costs (National Prescribing Centre 2008).

A number of silver dressings are available on the market. An overview of some of these dressings commercially available at the time of print, together with other silver-containing wound care products such as gels and powders, is provided in Appendix 1. Systagenix silver dressings are detailed in Section 13.

13. Systagenix silver dressings

Launched over twenty years ago, ACTISORB® Silver 220 was the first elemental silver dressing on the market. Since then, Systagenix (formerly Johnson and Johnson) have launched three further silver products: PROMOGRAN PRISMA® Matrix, SILVERCEL® and SILVERCEL® NON-ADHERENT.

Launched over twenty years ago, ACTISORB® Silver 220 was the first elemental silver dressing on the market

Table 1 summarises the main differences between Systagenix silver dressings. Further information for each dressing, including information from the package inserts, is detailed in Sections 13.1 to 13.4.

13.1 ACTISORB® Silver 220 Activated Charcoal Dressing

ACTISORB® Silver 220 activated charcoal dressing (ACTISORB® Silver 220) is a dressing composed of a layer of pure activated charcoal impregnated with elemental silver (0.22% [w/w], equating to 33 µg/cm²). The activated charcoal layer, upon absorption of wound exudates, traps bacteria and removes them away from the wound bed (Figure 5).

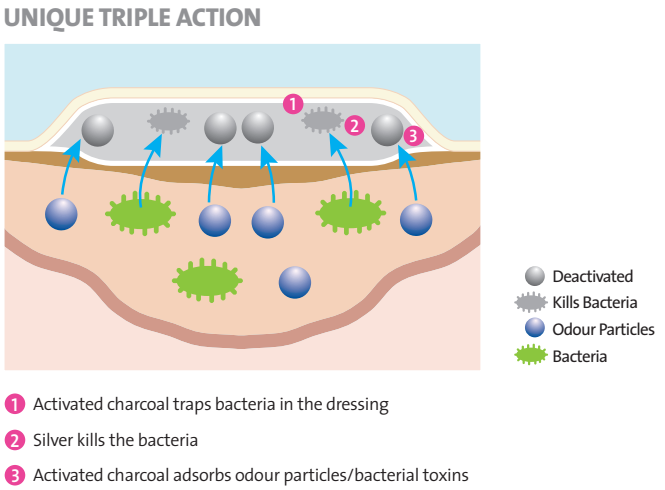


FIGURE 5. MECHANISM OF ACTION OF ACTISORB® SILVER 220

ACTISORB® Silver 220 represents an exception to most elemental silver dressings with the silver irreversibly bound to the activated charcoal. The silver cations that act locally within the dressing exert a broad-spectrum antimicrobial effect. Moreover, the charcoal helps to reduce wound malodour. The dressing has been shown to be effective against over 150 clinically relevant pathogens, including Gram-positive and Gram-negative aerobic bacteria, Gram-positive and Gram-negative anaerobes, yeasts, and the resistant bacteria species MRSA and VRE (Frost. 1984; De Voy 1985; Rudolph et al. 2000; Boothman 2002; Rennison et al. 2003).

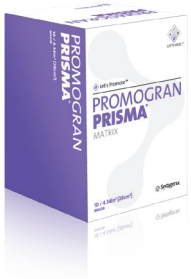
Indications

ACTISORB® Silver 220 is indicated for fungating carcinomas, ulcerative traumatic and surgical wounds where bacterial contamination, infection or odour occurs.

Silver concentration and release

ACTISORB® Silver 220 represents an exception to most other elemental silver dressings as the silver is in much lower concentrations. The dressing absorbs wound fluid and exudates containing infectious organisms into the dressing fabric, where the silver exerts its antimicrobial action (Lansdown et al. 2005). Specifically, any microorganisms present at the

TABLE 1. DIFFERENTIATING ATTRIBUTES FOR SYSTAGENIX SILVER DRESSINGS

ATTRIBUTE	SYSTAGENIX SILVER DRESSING			
	ACTISORB® SILVER 220	PROMOGRAN PRISMA® MATRIX	SILVERCEL®	SILVERCEL® NON-ADHERENT
				
Dressing composition	Activated charcoal impregnated with elemental silver	ORC (44%) and bovine collagen type I and III (55%). The dressing also contains 1% silver-ORC	Non-woven pad composed of a high G (guluronic acid) alginate, CMC and silver-coated nylon fibres	Non-woven pad composed of a high G (guluronic acid) alginate, CMC and silver-coated nylon fibres, laminated to a perforated, non-adherent EMA wound contact layer
Presentation of silver	Elemental	Compound (silver salt)	Elemental	Elemental
Silver concentration (mg silver/ 100 cm^2)	3.3	1.6	111	111
Indication	Fungating carcinomas, ulcerative traumatic and surgical wounds where bacterial contamination, infection or odour occurs	Wounds that are clear of necrotic tissue including: diabetic ulcers, venous ulcers, pressure ulcers, ulcers caused by mixed vascular aetiologies and traumatic and surgical wounds. Has shown haemostatic properties and can be used under compression therapy	Moderate to heavily exuding partial and full thickness wounds including: decubitus (pressure) ulcers, venous leg ulcers, diabetic ulcers, donor sites, traumatic and surgical wounds. Management of infected wounds, or wounds in which there is an increased risk of infection	Moderate to heavily exuding partial and full thickness wounds including: decubitus (pressure) ulcers, venous leg ulcers, diabetic ulcers, donor sites, traumatic and surgical wounds. Management of infected wounds, or wounds in which there is an increased risk of infection

CMC: Carboxymethylcellulose; EMA: Ethylene methyl acrylate; ORC: Oxidised regenerated cellulose.

wound site are absorbed and bound to the activated charcoal where they are exposed to the silver cations. The silver cations act locally within the dressing, eliminating the adsorbed microorganisms.

13.2 PROMOGRAN PRISMA® Wound Balancing Matrix

PROMOGRAN PRISMA® Wound Balancing Matrix (PROMOGRAN PRISMA® Matrix) is a platform derivative of PROMOGRAN Protease Modulating Matrix in a double density format. PROMOGRAN PRISMA® contains 44% ORC and 55% bovine collagen type I and III. PROMOGRAN PRISMA® Matrix also contains 1% silver-ORC, which equates to 0.25% (w/w) silver in the final product. Thus, PROMOGRAN PRISMA® Matrix retains all the properties of PROMOGRAN Matrix but has the added benefit of silver.

PROMOGRAN PRISMA® Matrix is a topically-applied wound therapy. The product is a sterile, freeze-dried, composite of ORC, collagen and silver-ORC (a compound of silver and ORC). In the presence of wound exudates, the PROMOGRAN PRISMA® Matrix transforms into a soft and conformable biodegradable gel which enables contact with all areas of the wound. Saline or Ringers´ solution should be used to hydrate PROMOGRAN PRISMA® Matrix on dry wounds.

The PROMOGRAN PRISMA® Matrix modulates and re-balances the wound environment by the unique combination of binding and inactivation of proteases (i.e. MMPs, elastase and plasmin) which have been shown to be detrimental in excess in chronic wounds. The dressing also binds and protects naturally occurring growth factors that may be degraded by these proteases. These growth factors are released back into the wound.

Indications

PROMOGRAN PRISMA® Matrix is indicated for the management of all wounds healing by secondary intent which are clear of necrotic tissue including diabetic ulcers, venous ulcers, pressure ulcers, ulcers caused by mixed vascular aetiologies and traumatic and surgical wounds.

PROMOGRAN PRISMA® Matrix has shown haemostatic properties and can be used under compression therapy.

Contraindications

PROMOGRAN PRISMA® Matrix is contraindicated in patients with known hypersensitivity to the components of this product, i.e. ORC, collagen or silver.

Silver concentration and release

As PROMOGRAN PRISMA® Matrix biodegrades, the silver cations derived from the silver-ORC in the product are released through dissolution in fluid to exert the antimicrobial activity.

Use of dressings containing high levels of silver on wounds with low bioburden may have a negative effect on wound repair (Innes et al. 2001). The formulation of PROMOGRAN PRISMA® Matrix provides a simultaneous antimicrobial effect and bioburden control without causing injury to the host cells. This unique product property was confirmed in cell culture experiments with fibroblasts, keratinocytes and endothelial cells incubated in the presence of chronic wound fluid and dressing samples. In these experiments, PROMOGRAN PRISMA® Matrix was compared with several silver dressings such as AQUACEL Ag, ACTICOAT 7, Contreet and Urgotul S.Ag. PROMOGRAN PRISMA® Matrix demonstrated an ability to balance the chronic wound fluid, thereby providing an environment favourable for cell proliferation (ETRS Newsletter 2005).

13.3 SILVERCEL®

SILVERCEL® Antimicrobial Alginate Dressing (SILVERCEL®) is an antimicrobial dressing with elemental silver-coated nylon fibres. Specifically, it is a non-woven pad composed of a high G (guluronic acid) alginate, carboxymethylcellulose (CMC) and silver-coated nylon fibres. It has a high capacity of absorption, derived from the calcium alginate and carboxymethylcellulose fibres. The unique composition of the dressing manages exudates in moderate to heavily exuding wounds, which creates a favourable environment for effective wound management. The silver fibres kill a broad-spectrum of microorganisms associated with the bacterial colonisation and infection of wounds. In moderate to heavily exuding wounds, the dressing maintains a moist wound healing environment and allows for intact removal.

Indications

SILVERCEL® is intended for use in the management of all moderate to heavily exuding partial and full thickness chronic wounds including decubitus (pressure) ulcers, venous ulcers, diabetic ulcers, donor sites, traumatic and surgical wounds. As the product contains alginate it may assist in supporting the

control of minor bleeding in superficial wounds. It is also suitable for use, under medical supervision, in the management of infected wounds or wounds where there is an increased risk of infection.

Contraindications

SILVERCEL® is not indicated for surgical implantation or for patients with a known sensitivity to silver.

Precautions

SILVERCEL® is not intended:

- To control heavy bleeding
- For direct application on dry/low moisture wounds. As wound conditions improve and exudates levels decrease, it may be preferable to use a NON-ADHERENT wound contact layer (e.g. N-A® Ultra or ADAPTIC® Non-Adhering Dressing) or switch to a more appropriate dressing.

Silver concentration and release

- *In vitro* experiments using simulated wound fluid (a saline solution with albumin) demonstrated that SILVERCEL® provides a sustained release of silver cations (approximately 20 ppm) up to seven days into the simulated wound fluid (Addison et al. 2006; Clark et al. 2009b).
- The total content of elemental silver in SILVERCEL® is 8% (w/w), equating to 111 mg/100 cm^2 (Addison et al. 2006).
- Silver cations are released from the elemental silver-coated nylon fibres.

13.4 SILVERCEL® NON-ADHERENT

SILVERCEL® NON-ADHERENT Dressing (SILVERCEL® NON-ADHERENT) is a non-woven pad composed of a high G (guluronic acid) alginate, carboxymethyl cellulose (CMC) and silver-coated nylon fibres, laminated to a perforated, non-adherent ethylene methyl acrylate (EMA) wound contact layer. Thus, with exception to the EMA wound contact layer, SILVERCEL® NON-ADHERENT is the same as the original SILVERCEL® dressing.

The unique composition of the dressing manages exudates in moderate to heavily exudating wounds, which creates a favourable moist wound healing environment for effective wound management and allows intact dressing removal. The silver fibres kill a broad-spectrum of microorganisms associated with the colonisation and infection of wounds. Odour reduction results from the antibacterial effect in the dressing.

Indications

As detailed for SILVERCEL® (Section 13.3).

Contraindications

SILVERCEL® NON-ADHERENT is not intended for use for patients with a known sensitivity to alginates, EMA or silver, for pregnant or lactating women due to the absence of specific information, or for surgical implantation.

Precautions

- SILVERCEL® NON-ADHERENT is not intended to control heavy bleeding.
- As wound conditions improve and exudates levels decrease, it may be preferable to switch to a more appropriate dressing or moisten the dressing with saline solution prior to application.
- The dressing must be removed prior to patients undergoing MRI examinations.
- Avoid contact with electrodes or conductive gels during electronic measurements, e.g. ECG and EEG.

Silver concentration and release

- Similar to SILVERCEL®, SILVERCEL® NON-ADHERENT combines a sustained and controlled release of silver cations (derived from the elemental silver-coated nylon fibres) up to seven days (Clark et al. 2009d).
- *In vitro* experiments using simulated wound fluid demonstrate that SILVERCEL® provides a sustained release of silver cations (approximately 20 ppm) into the simulated wound fluid (Stephens et al. 2009).
- Silver cations are released from the elemental silver-coated nylon fibres.

14. Evidence-based medicine: Systagenix dressings

There is considerable "evidence-based medicine" for use of Systagenix silver dressings, ranging from *in vitro* data to clinical trials and post marketing surveillance studies. Figure 6 shows the hierarchy of *in vitro*, *in vivo* and *ex vivo* data and clinical evidence-based medicine.

Supporting evidence for each Systagenix silver dressing-type is discussed below in Sections 14.1

to 14.4.

14.1 Evidence-based medicine for ACTISORB® Silver 220 Activated Charcoal Dressing

ACTISORB® Silver 220 is an exception to most other elemental silver dressings in that it has a low silver content and does not release silver cations. However, *in vitro* and *in vivo* tests show that the dressing is effective at eradicating wound pathogens despite its comparatively low silver concentration (Wunderlich and Orfanos 1991; Tebbe and Orfanso 1996; Johnson and Johnson, Data on file 2001). Given that ACTISORB® Silver 220 was launched over twenty years ago, there is a wealth of publications available supporting the use of this product in the treatment of infected chronic wounds. These publications relate to data collected from a range of studies, including *in vitro* laboratory studies, clinical observations and post-marketing surveillance studies (Milward 1991; Wunderlich and Orfanos 1991; Tebbe and Orfanos 1996; Hametner 2000; Rudolph 2000; Johnson and Johnson, Data on file 2001; Müller et al. 2003).

The silver fibres kill a broad-spectrum of microorganisms associated with the colonisation and infection of wounds.

Noteworthy evidence-based medicine for use of ACTISORB® Silver 220 includes results from a comparative, randomised controlled clinical study involving patients with leg ulcers or pressure sores (n=40) treated with either ACTISORB® Silver 220 or a control therapy (zinc paste) (Wunderlich and Orfanos 1991). Results indicated that patients receiving ACTISORB® Silver 220 demonstrated a statistically significant reduction in wound area compared with patients in the control group, with 6/19 patients in the treatment group experiencing full wound closure versus 2/19 patients in the control group ($p<0.05$). These results were supported by two separate randomised controlled trials (RCTs) involving patients with chronic venous leg ulcers or pressure ulcers receiving either a hydrocolloid dressing in the control group, or ACTISORB® (without silver) or ACTISORB® Silver 220 in the treatment groups (Kerihuel, 2010). In addition, results from a prospective multi-centre observational study (n=224) showed that treatment with ACTISORB® Silver 220 for four weeks resulted in a reduction of wound size by up to 50% (Tebbe and Orfanos 1996). Moreover, results from a post-marketing surveillance study involving patients with pressure sores, venous leg ulcers, diabetic foot ulcers or traumatic wounds (n=12,444) further demonstrated the efficacy of ACTISORB® Silver 220. In this study, the overall healing rate of wounds was 35.5% after six-weeks of treatment (Johnson and Johnson, Data on file 2001). In addition, in a retrospective study conducted with patients with colonised or infected wounds, ACTISORB® Silver 220 combated infection, reduced pain and promoted healing (Krammerlander et al. 2008). A review of ACTISORB® Silver 220 based on evidence from comparative and non-comparative trials involving over 12000 patients similarly concluded that this silver dressing was effective in reducing wound malodour, effective in promoting wound healing and safe (White et al. 2001).

14.2 Evidence-based medicine for PROMOGRAN PRISMA® Wound Balancing Matrix

There is an abundance of evidence to support the use of PROMOGRAN PRISMA® Matrix for the treatment of chronic wounds.

PROMOGRAN PRISMA® Matrix is capable of exerting a potent antimicrobial effect. In an *in vitro* study investigating the antimicrobial properties of PROMOGRAN PRISMA® Matrix, application of the $\log_{10}$ reduction test demonstrated that PROMOGRAN PRISMA® Matrix has antimicrobial activity, even against the resistant strains MRSA and VRE (ETRS Newsletter 2005).

The low concentration of silver together with the unique dressing composition of ORC and bovine collagen in PROMOGRAN PRISMA® Matrix is particularly beneficial to patients where there are low levels of bacterial infection or a potential for wound infection. Under *in vitro* conditions, PROMOGRAN PRISMA® Matrix demonstrated antimicrobial activity

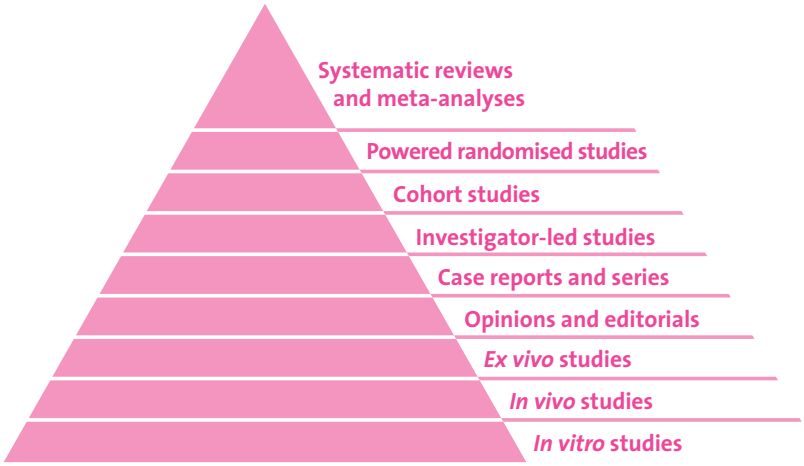


FIGURE 6: ILLUSTRATION DEMONSTRATING THE HIERARCHY OF *IN VITRO*, *IN VIVO* AND *EX VIVO* DATA AND CLINICAL EVIDENCE

and also had a positive effect on the cell proliferation of fibroblasts, keratinocytes and endothelial host cells (ETRS Newsletter 2005). In addition, Cullen et al. (2010) report that collagen/ORC (a component of PROMOGRAN PRISMA® Matrix) reduced elastase, MMP-2, MMP-8 and MMP-9 activity *in vitro*.

Results from a randomised, prospective, open-labelled, multicentre, comparative trial including patients with venous leg ulcers (n=49) showed that for the patients receiving PROMOGRAN PRISMA® Matrix there was a trend towards more rapid wound closure compared with the control treatment. This was particularly apparent during the first four weeks of therapy (Hanf et al. 2007). In addition, in a randomised prospective controlled pilot study patients with venous leg ulcers (n=30) were treated with either PROMOGRAN PRISMA® Matrix and compression therapy or the standard of care (moist wound healing and compression therapy) for 12 weeks. Following treatment, patients who received compression therapy with PROMOGRAN PRISMA® Matrix were four times more likely to heal compared with those patients who received the standard of care ($p<0.04$) (Lanzara et al. 2008).

A further randomised, prospective, controlled clinical study involving patients with diabetic foot ulcers (n=40) treated with either PROMOGRAN PRISMA® Matrix or the standard of care for 14 weeks showed that PROMOGRAN PRISMA® Matrix stimulated healing while protecting the wound from infection (Gottrup et al. 2010). Of those patients receiving PROMOGRAN PRISMA® Matrix, significantly more experienced at least a 50% reduction in their wound area (Margolis Index) at Week 4 compared with the control group (70% vs 43%; $p=0.035$).

Observations from a collection of a number of case studies involving patients with venous leg ulcers, pressure sores or traumatic wounds treated with PROMOGRAN PRISMA® Matrix suggested that the dressing helped to initiate the healing process of chronic wounds trapped in a non-healing inflammatory status (ETRS Newsletter, 2005; Cullen et al. 2010).

14.3 Evidence-based medicine for SILVERCEL®

A number of *in vitro* studies (Addison et al. 2005; Addison et al. 2006; ETRS Newsletter 2005; Meaume and Vallet 2005) and clinical evidence (Meaume et al. 2005; Teot et al. 2005) support the use of SILVERCEL® for the management of infected chronic wounds.

In vitro experiments demonstrate that SILVERCEL® is antimicrobial against over 150 microorganisms including *Candida albicans*, *E.coli*, *Klebsiella pneumonia*, MRSA, *P.aeruginosa*, *S.aureus*, *Staphylococcus epidermidis*, *Streptococcus pyrogenes* and VRE (Addison et al. 2006). In addition, *in vitro* data indicate that SILVERCEL® has excellent fluid handling properties in managing high levels of exudates when tested in combination with an appropriate secondary dressing such as TIELLE® or TIELLE® Plus Hydropolymer Adhesive Dressings (Addison et al. 2005). Clinical observations showed similar results in that SILVERCEL® was shown to absorb wound exudates and combat infection (Meaume and Vallet 2005; Teot et al. 2005). SILVERCEL® was also shown to maintain close contact with the wound bed, support the formation of new granulation tissue and improve wound healing rates (Meaume and Vallet 2005). Indeed, wound healing rates were found to be double that of a non-silver control dressing (Meaume and Vallet 2005).

Clinical evidence for SILVERCEL® includes results from a collection of case studies confirming a favourable

profile for exudate handling, tensile strength and antimicrobial activity in wounds with high levels of exudates (Teot et al. 2005). These findings were confirmed in a RCT where SILVERCEL® was found to be well tolerated, able to manage high levels of wound exudate, provide a moist wound environment and easily removed after saturation. In addition, SILVERCEL® was considered to promote wound cleansing, control wound bioburden and improve the healing rate (Meaume and Vallet 2005). Similar results were also observed by Meaume et al. (2005), where 4/38 (10.5%) patients in the control group were treated with systemic antibiotics at the final visit compared with 0/40 patients receiving SILVERCEL® (p=0.053). In addition, fewer wounds developed a clinical infection over the four-week follow-up in the treatment group (33% versus 46%; p=0.223), and the four-week closure rate was statistically greater in the treatment group (0.32 +/- 0.57 cm^2/day versus 0.16 +/- 0.40 cm^2/day; p=0.024).

14.4 Evidence-based medicine for SILVERCEL® NON-ADHERENT

Launched in 2009, there is already clinical data to support the use of SILVERCEL® NON-ADHERENT for the treatment of chronic wounds: these data are supported by numerous *in vitro* studies. There is also a considerable amount of clinical evidence to support the use of SILVERCEL®, the derivative dressing of SILVERCEL® NON-ADHERENT Dressing (Section 14.3).

Using a series of *in vitro* assays, Clark et al. (2009a) demonstrated that the addition of a perforated film to SILVERCEL® to generate the non-adherent dressing "SILVERCEL® NON-ADHERENT" does not compromise the absorbency, wet tensile strength or antimicrobial properties of the dressing. Additional *in vitro* assays investigated the adherence levels of SILVERCEL® NON-ADHERENT and demonstrated that SILVERCEL® NON-ADHERENT was less adherent than competitor alginate/fibrous based dressings tested. In these experiments the force required to separate the dressing from a fibrin clot was <160 gf for SILVERCEL® NON-ADHERENT, whereas a force of 384-940 gf was required for the competitor dressings (Clark et al. 2009a). Further publications also support these favourable physical properties and also demonstrate antimicrobial activity (including antimicrobial activity against biofilms) (Clark et al. 2009b, 2009c, 2009d; Stephens et al. 2009; McInroy et al. 2009; McInroy et al. 2010). Moreover, SILVERCEL® NON-ADHERENT was found to shed less fibres after *in vitro* dressing application assays than other commercially available wound dressings tested (AQUACEL Ag, ACTICOAT Absorbent, Sorbsan Silver and Urgosorb Silver) (Clark et al. 2009d).

In vivo experiments using a porcine partial-thickness exudating wound model showed that SILVERCEL® NON-ADHERENT performed more favourably than AQUACEL Ag (Hart et al. 2009). Specifically SILVERCEL® NON-ADHERENT showed lower wound surface adherence, reduced dressing debris deposition and reduced wound tissue disruption compared with AQUACEL Ag.

Dressing removal and pain reduction

Inappropriate dressing selection may lead to dressing-wound adherence, causing trauma and pain upon dressing removal. SILVERCEL® NON-ADHERENT is particularly beneficial for patients with infected wounds with moderate to heavy exudate and where there may be a risk of damage to the surrounding skin (Russell, 2009). In a clinical study involving 20 patients with chronic wounds, SILVERCEL® NON-ADHERENT and a commercially available alginate dressing were assessed (Stephens et al. 2010). SILVERCEL® NON-ADHERENT was found to be less adherent and less painful at dressing changes compared with the competitor dressing. In addition, in a study by Hart and Bell (2009), SILVERCEL® NON-ADHERENT exhibited lower wound adherence and reduced debris deposition compared with a control silver hydrofiber dressing.

15. Cost effectiveness of silver dressings

Conflicting opinions exist within the literature regarding the cost effectiveness of silver dressings, with some publications supporting the use of these products while others do not. An overview of some of the current opinions in the scientific literature is provided below.

In a non-blinded RCT involving patients with mid-dermal or mixed partial-thickness burns (n=84), treatment with AQUACEL Ag was found to be less costly than 1% SSD cream (Caruso et al. 2006). Although the initial cost of AQUACEL Ag was more than that for 1% SSD cream, the cost of pain medications, secondary dressings was lower and fewer dressing changes were required.

Conversely, results from the VULCAN study did not support the use of silver dressings (Michaels et al. 2009). In this non-blinded RCT the cost-effectiveness analysis of silver dressings was investigated. Inclusion

criteria included presence of a leg ulcer on the lower leg for over six weeks. Exclusion criteria included insulin-controlled diabetes mellitus, ankle brachial pressure index less than 0.8 in the affected leg, atypical ulcers including those with a suspicion of malignancy, and patients receiving antibiotic treatment. The primary endpoint was complete ulcer healing at 12 weeks. Secondary endpoints included time to healing, quality of life and cost-effectiveness.

In the VULCAN study, a total of 213 patients with venous leg ulcers were recruited and treated with compression bandaging and either a silver dressing (n=107) or a non-antimicrobial low adherence dressing (n=106); At 12 weeks, no difference between the two types of dressings was noted with respect to the primary endpoint of complete ulcer healing (59.6% and 56.7% for silver and non-antimicrobial dressings, respectively). Similarly, no significant differences were noted between the dressing types for health related quality of life at the follow-up times of one, three, six or twelve months. However, treatment with the silver dressings was more expensive than the non-antimicrobial dressings. Compared with the control group, the silver dressing group had an additional cost of £98 and an additional quality-adjusted life year gain of 0.0002. However, it is important to consider that in clinical practice the wound area would be continuously assessed and treatment with silver dressings would not necessarily continue for 12 weeks: treatment with silver dressings should be discontinued if a wound showed signs of healing or reduced bioburden. In addition, recruitment of patients with heavily infected wounds was not a prerequisite for enrolment to the VULCAN study, suggesting that dressings were applied to patients who did not have overtly infected chronic wounds. White and Kingsley (2010) also note that in the VULCAN study, silver dressings were placed on wounds without a clinical justification for use and used for a prolonged period of time, which is contrary to current clinical practice.

A recent review assessed the quantity and quality of RCTs conducted using silver dressings and silver-based topical agents (Chambers et al. 2007). The rate of healing, proportion of ulcers completely healed and change in ulcer size were evaluated. Out of all the available RCTs, only nine were considered eligible for evaluation. The authors concluded that there is poor evidence to support routine use of silver dressings for leg ulcer treatment.

Despite these disparaging publications on the use of silver dressings in the treatment of chronic wounds, numerous publications highlight the benefits of such treatments. In addition, there is a wealth of evidence and literature to support the use of Systagenix silver dressings for the treatment of chronic wounds as described in Section 14.

KEY POINTS

- *The VULCAN study suggested that there was no evidence to support the use of silver dressings underneath compression dressings for the treatment of venous leg ulcers as cost-effective. However, the VULCAN study was flawed in the trial design in that patients with non-infected wounds received silver dressings.*
- *There is a wealth of evidence and publications to support the use of Systagenix silver dressings for the treatment of chronic wounds.*

16. Overview: Commercial silver dressings

Use of silver dressings has increased substantially over recent years, with numerous silver dressings now available on the market. The scientific literature is punctuated with a plethora of publications supporting the use of silver dressings to promote wound healing and enhance patient quality of life. Despite a limited number of controversial studies suggesting that use of silver dressings is not cost-effective, such antimicrobial dressings continue to be used widely in clinical practice, supporting the view that they are generally accepted as efficacious. In addition, it is also important to consider that many of these controversial studies state total healing as a primary endpoint and place limited emphasis on returning the wound environment to a normal healing trajectory, as evidenced by the degree of wound reduction or assessing other factors that impact upon patients lives such as high bioburden, wound exudate, odour or pain. With respect to Systagenix silver dressings, there is a wealth of *in vitro*, *in vivo* and clinical evidence to support their safe and effective use in wound management

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17. Appendix 1: Commercially available elemental and compound silver wound care products

PRODUCT FORMAT	PRODUCT [1]	MANUFACTURER	PRESENTATION OF SILVER
Adhesive strips	Silverlon Adhesive Strips	Argentum Medical, LLC	Elemental
Calcium alginate/ alginate based	ACTICOAT Absorbent	Smith and Nephew	Elemental
	ALGICELL Ag	Derma Sciences	Specified as: "silver"
	Algidex Ag	DeRoyal	Specified as: "ionic silver"
	ALGISITE Ag	Smith and Nephew	Specified as: "silver"
	Arglaes Island	Medline	Specified as: "silver"
	Askina Calgitrol Ag	Braun	Compound (calcium alginate and silver alginate with 10% of bounded water)
	Invacare Silver Alginate	ISG	Compound (silver sodium hydrogen zirconium phosphate)
	Maxorb Extra Ag	Medline	Compound (silver sodium hydrogen zirconium phosphate)
	Melgisorb Ag	Molnycke	Specified as: "silver"
	Restore Calcium Alginate	Hollister Wound Care LLC	Specified as: "ionic silver"
	SeaSorb Ag	Coloplast	Compound (specified as: "ionic silver complex" only)
	SILVERCEL®	Systagenix	Elemental
	SILVERCEL® NON-ADHERENT	Systagenix	Elemental
	Silverlon Calcium Alginate	Argentum Medical, LLC	Elemental
	SILVASORB	Medline	Specified as: "ionic silver"
	Sorbsan Silver	Unomedical	Compound (silver Sorbsan Silver is made from the calcium salt of alginic acid)
	Suprasorb A + Ag	Activa Healthcare	Specified as: "silver"
	Tegaderm Alginate Ag Silver	3M	Compound (carboxymethylcellulose and alginate fibre formula)
	Urgosorb Silver/Ag	Urgo Medical	Specified as: "ionic silver complex"
Cream	Flamazine Cream	Smith and Nephew	Compound (SSD)
Collagen based	BIOSTEP Ag	Smith and Nephew	Compound (silver chloride)
	CollaGUARD Ag	Innocoll	Specified as: "silver"
	COLACTIVE Collagen with Silver	Smith and Nephew	Compound (silver lactate)
	PROMOGRAN PRISMA® Matrix	Systagenix	Compound (silver salt)
	PURACOL PLUS Ag^{+}	Medline	Specified as: "silver"
	CovaClear Ag	Covalon	Specified as: "silver"
Fibrous/ cloths, miscellaneous	ACTICOAT	Smith and Nephew	Elemental
	ACTISORB® Silver 220	Systagenix	Elemental
	Atrauman Ag	Hartmann Group	Elemental
	Mepilex Ag	Molnycke	Specified as: "silver"
	Physiotulle Ag / Altreet – Ag	Coloplast	Compound (SSD)
	Silver Cloth Island	Ferris Mfg. Corp.	Specified as: "silver"
	Silverlon	Argentum Medical, LLC	Compound (silver oxide)

PRODUCT FORMAT	PRODUCT [1]	MANUFACTURER	PRESENTATION OF SILVER
Fibrous/ cloths, miscellaneous	SILVERSEAL	Derma Sciences	Compound (silver oxide)
	Tegaderm Ag Mesh Dressing with Silver	3M	Compound (silver sulfate)
	Urgotul SSD	Laboratoies Urgo	Compound (SSD)
	Vliwaktiv Ag, Absorbent Activated Charcoal	Lohmann and Rauscher	Specified as: "silver"
	Vliwaktiv Ag, Activated Charcoal Rope with Silver	Lohmann and Rauscher	Specified as: "silver"
Film/mesh	ACTICOAT 7	Smith and Nephew	Elemental
	Arglaes film	Medline	Specified as: "silver"
	Restore Contact Layer with Silver	Hollister Wound Care LLC	Compound (AgCl)
Foam	ACTICOAT Moisture Control	Smith and Nephew	Elemental
	Allevyn Ag	Smith and Nephew	Compound (SSD)
	Biatain Ag	Coloplast	Specified as: "silver"
	Mepilex Ag	Molnlycke	Specified as: "silver"
	OPTIFOAM Ag Adhesive	Medline	Specified as: "ionic silver"
	OPTIFOAM Ag Non-adhesive	Medline	Specified as: "ionic silver"
	PolyMem Silver Island	Ferris Mfg. Corp.	Elemental
	PolyWic Silver	Ferris Mfg. Corp.	Elemental
	Restore non-adherent foam with silver	Hollister Wound Care LLC	Specified as: "silver"
	Silverlon Negative Pressure	Argentum Medical, LLC	Specified as: "ionic silver"
	SilverSite	Centurion	Compound (silver alginate)
	UrgoCell Silver/Cellosorb Ag	Urgo Medical	Compound (silver salts)
	V.A.C GranuFoam Silver	KCI	Specified as: "silver"
Gauze	Urgotul SSD/S.Ag	Urgo Medical	Compound (SSD)
Hydrocolloid	Contreet Hydrocolloid	Coloplast	Specified as: "silver"
	SILVERSEAL Hydrocolloid	DermaSciences	Specified as: "silver"
	SureSkin	EuroMed	Compound (silver zeolite)
Hydrofibre	AQUACEL Ag	ConvaTec	Compound (specified as "ionic silver")
Hydrogel	Elta Silvergel	Elta	Specified as: "silver"
	ExcelGinate Ag	MPM	Specified as: "silver"
	Gentell Ag Hydrogel Wound Dressing	Gentell	Compound (SSD)
	SILVASORB Gel	Medline	Specified as: "ionic silver"
	SilverMed Antimicrobial Silver	MPM	Specified as: "silver"
	SILVERSEAL	DermaSciences	Compound (silver oxide)
	Silver-Sept Antimicrobial Gel	Anacapa Tech Inc	Compound (silver salt)
Powder	Arglaes Powder	Medline	Specified as: "silver"
Wash	SilverMed Antimicrobial Wound Cleanser	MPM	Specified as: silver microparticles

[1] Brands marked with ® or ™ are trademarks of Systagenix. All other products referenced herein are acknowledged to be trademarks of their respective owners.

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