Evaluating antimicrobial efficacy of new commercially available silver dressings

Marion H Cavanagh, Robert E Burrell, Patricia L Nadworny

ABSTRACT
Prevention and treatment of bacterial colonised/infected wounds are critical. Many commercially available silver dressings claim broad-spectrum bactericidal activity over days and are indicated for serious conditions including burns and ulcers. However, there is no peer-reviewed literature available for many newer dressings. This study compared the activity of some of these dressings. Six silver-containing dressings were compared using log reduction, silver release and corrected zone of inhibition assays. Only the nanocrystalline silver dressing was bactericidal against Staphylococcus aureus, and the only other dressing that produced any log reduction was a silver collagen matrix dressing. These two dressings and a silver alginate dressing produced zones of inhibition, although the collagen matrix and alginate dressings had decreasing zone sizes over time, and the latter liquefied after five transfers. The remaining dressings (two ionic silver foam dressings and a silver sulphate dressing) did not produce zones of inhibition. For the foam, alginate and collagen matrix dressings, antimicrobial activity was related to silver release. The silver sulphate dressing released large quantities of silver, but only through the dressing edges, as the wound-contacting surface appeared to be hydrophobic. The results of this study emphasise the importance of confirming product claims regarding silver dressing efficacy.

Key words: Advanced wound dressing • Bactericidal • Bacteriostatic • Silver

INTRODUCTION
Chronic and acute wounds are often heavily colonised or infected by bacteria (1) which interfere with wound healing processes by inducing an inflammatory host response (2,3). This makes prevention and treatment of bacterial colonisation a critical part of wound care. Traditional methods of controlling bacterial colonisation involve the use of topical antimicrobial agents such as silver nitrate and silver sulphadiazine, as well as a variety of other antiseptics, such as chlorhexidine, quaternary ammonium compounds and povidone-iodine. However, over the past decade, a large number of advanced silver-containing dressings have become commercially available. The majority of these dressings are marketed as effective against a broad range of bacteria over multiple days and are indicated for a variety of serious conditions including partial thickness burns, ulcers of various aetiologies, donor and graft sites, traumatic and surgical wounds, dermatologic disorders and skin tears. However, there are little data available in the peer-reviewed literature regarding these dressings, particularly those which have been released recently.

The purpose of this study was to compare the activity of some recently available silver-containing dressings. Six dressings were...
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tested – a silver sulphate dressing, a silver alginate dressing, a silver collagen matrix dressing, a nanocrystalline silver dressing and two ionic silver foam dressings.

MATERIALS AND METHODS

Materials

All dressings used were obtained free from the companies which produce them. All dressings were stored as indicated on the packaging and tested within their expiry dates. Unless mentioned, all other materials were purchased from Fisher Scientific Canada, Inc.

Dressings tested in this study were as follows:

- A silver sulphate (Ag₂SO₄) dressing (Mepilex® Ag Antimicrobial Soft Silicone Foam Dressing, Molnlycke Health Care LLC, Sweden). Product information indicates that this dressing inactivates a wide range of bacteria, including *Staphylococcus aureus*, within 30 minutes; provides a rapid sustained silver release; can be worn for 7 days and does not stain. The dressing is indicated for low to moderately exuding wounds such as partial thickness burns, leg ulcers, foot ulcers and pressure ulcers (4).

- A silver alginate dressing (Algicell™ Ag, Derma Sciences, Inc., Ontario, Canada). Product information indicates that this dressing contains 1.4% ionic silver; has a kill rate of 99.99%, 99.91% and 97.46% at days 1, 3 and 5 of challenge, respectively, for *S. aureus* and behaves as a bacterial barrier with controlled sustained silver release. The dressing is indicated for diabetic foot ulcers, leg ulcers, foot ulcers and pressure ulcers (4).

- A silver collagen matrix dressing with calcium alginate and ethylenediaminetetraacetic acid (EDTA) (Biostep™ Ag, Smith and Nephew, Inc., Largo, FL). The silver is in the form of silver chloride. Product information for this dressing indicates that it has antibacterial activity, targets and deactivates excess matrix metalloproteinases and has a 6-day wear. The silver in the dressing is intended to prevent colonisation of the dressing (7). The dressing is indicated for management of full-thickness and partial-thickness acute and chronic wounds including pressure ulcers, diabetic ulcers, mixed vascular etiology ulcers, venous ulcers, first- and second-degree burns, donor or graft sites, abrasions, dehisced surgical wounds and traumatic wounds (7).

- Two ionic silver foam dressings (PolyMem® Silver Non-Adhesive Pads, and PolyMem® Silver Shapes, Ferris Mfg. Corp., Burr Ridge, IL). Product information indicates that both of these dressings kill 99.9% of bacteria tested (including *S. aureus*), do not stain skin and can be worn for 3 days. These dressings are indicated for first- and second-degree burns, leg ulcers, diabetic ulcers, venous ulcers, donor and graft sites, traumatic and acute wounds, surgical wounds, dermatologic disorders and skin tears (6).

- A silver collagen matrix dressing with calcium alginate and ethylenediaminetetraacetic acid (EDTA) (Biostep™ Ag, Smith and Nephew, Inc., Largo, FL). The silver is in the form of silver chloride. Product information for this dressing indicates that it has antibacterial activity, targets and deactivates excess matrix metalloproteinases and has a 6-day wear. The silver in the dressing is intended to prevent colonisation of the dressing (7). The dressing is indicated for management of full-thickness and partial-thickness acute and chronic wounds including pressure ulcers, diabetic ulcers, mixed vascular etiology ulcers, venous ulcers, first- and second-degree burns, donor or graft sites, abrasions, dehisced surgical wounds and traumatic wounds (7).

Bactericidal efficacy – log reduction assay

Log reductions were used to determine the ability of commercial silver dressings to kill bacteria in 30 minutes, using the methods of Gallant-Behm et al. (9) All tests were performed in triplicate. Briefly, *S. aureus* ATCC 25923 [three colonies of a Mueller-Hinton agar (MHA) plate or 1 ml of a 4- to 5-hour growth culture grown as described below] was used to inoculate 100 ml of tryptic soy broth (TSB) and was grown overnight at 37°C and 0.409 g. One milliliter of this culture was used to inoculate another flask of TSB (100 ml) and was grown under the same conditions for 4-5 hours to ensure the bacteria were in log-phase growth [~10⁸ colony-forming units (CFU)/ml]. Using
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Aseptic technique, dressings were cut into various sizes, with dressings that expand considerably upon saturation being cut to smaller sizes than those that do not. The nanocrystalline silver and silver alginate dressings were cut into 2.54 × 2.54 cm pieces. Silver collagen matrix dressings were cut into 2.54 × 1.27 cm pieces. Silver sulphate dressings and both ionic silver foam dressings were cut into 1.27 × 1.27 cm squares. The dressing pieces were placed on thin sheets of plastic (aseptically cut into 3.8 × 3.8 square cm pieces) in the inverted lid of a sterile Petri dish. Experimental dressing pieces were then moistened with the required moistening volume of sterile distilled water. To determine the moistening volume, the saturation volume of each dressing was determined by a simple water holding test in which the dressings were weighed, submerged in water for 2 minutes, carefully removed (without squeezing), allowed to drip for 10 seconds and then re-weighed (Table 1). Then the inoculum volume was subtracted from 90% of the saturation volume to obtain the moistening volume. Control dressing pieces were moistened with the moistening volume of STS, a detergent which inactivates silver (6.0% [w/v] NaCl, 1% [v/v] polysorbate 20 and 0.1% [w/v] sodium thioglycolate for S. aureus (10)). Both control and experimental dressing pieces were then inoculated with 75 μl of inoculum per 1.27 cm square. A second sheet of plastic was then laid on top of the dressings to contain the inoculum, followed by the Petri dish base placed upright to ensure good contact between the bacteria and the dressings. The inoculated dressings were then incubated at 37°C for 30 minutes. After removal of the dressings from the incubator, they were placed in STS in order to achieve a 1:10 dilution of the original inoculum. The dressings in STS were then vigorously vortexed, and the resulting solutions containing the recovered bacteria were serially diluted in phosphate-buffered saline (pH 7.0), containing 8.5 g/l NaCl, 0.61g/l KH2PO4 and 0.96 g/l K2HPO4. Three 20 μl drops from each dilution were plated on MHA. The plates were then incubated at 37°C and after 24 and 48 hours the numbers of bacterial colonies were counted. The counts generated from the experimental pieces of dressings were used to calculate the surviving number of CFU, whereas the counts generated from the control pieces of dressings were used to calculate the numbers of bacterial CFU in the original inoculants that were not trapped in the dressings. The log10 of the starting numbers and surviving numbers of bacteria were then determined. Log reductions were then calculated as the difference between the log of the initial number of bacteria and the log of the final surviving number of bacteria.

### Silver dissolution assay

The release of silver from each dressing was determined with static dissolution tests using methods similar to those of Wright et al. (11) except that corrections were made for dressing saturation volume and dressing breakage. Briefly, one 2.54 × 2.54 cm piece of dressing per 5 ml of sterile distilled water plus the dressing saturation volume was placed in a sealed vial wrapped in aluminium foil to prevent silver precipitation. The vial was then incubated at 37°C for 24 hours. Afterwards, the dressings were aseptically removed from the vials, allowed to drip into the vial for 10 seconds and then disposed off. The remaining solutions were immediately filtered with a 70-μm filter if pieces of the dressings had broken off in solution (both of the ionic silver foam dressings, the silver collagen matrix dressings and the silver alginate dressings) in order to prevent further leaching of silver from the pieces after the 24-hour time point and to facilitate atomic absorption spectroscopy (AAS). The solutions were then acidified in 9% nitric acid/0.9% tartaric acid to ensure that all silver released remained in solution. The solutions were then submitted for total silver analysis by AAS. For AAS, a Varian 220 FS double-beam Atomic Absorption Spectrophotometer (Varian Inc., Palo Alto, CA) was used, with the following

### Table 1 Dressing saturation volumes

<table>
<thead>
<tr>
<th>Dressing description</th>
<th>Average saturation volume (μl/cm², ±SD, n = 3)</th>
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<tbody>
<tr>
<td>Silver sulphate dressing</td>
<td>808 ± 43</td>
</tr>
<tr>
<td>Non adhesive ionic silver foam dressing</td>
<td>594 ± 43</td>
</tr>
<tr>
<td>Shaped ionic silver foam dressing</td>
<td>172 ± 66</td>
</tr>
<tr>
<td>Silver collagen matrix dressing</td>
<td>176 ± 5</td>
</tr>
<tr>
<td>Silver alginate dressing</td>
<td>151 ± 11</td>
</tr>
<tr>
<td>Nanocrystalline silver dressing</td>
<td>53 ± 1</td>
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</table>
instrument parameters: an Ag hollow cathode lamp with a wavelength of 328.1 nm and a lean air-acetylene flame. A calibration plot was generated using silver standards of 0.5, 1.0, 3.0 and 5.0 ppm, prepared from a silver standard stock solution of 1000 ppm. If dressings had released more than 5 ppm into solution, the solutions were diluted as necessary with distilled water until they were in the linear range for silver analysis (0.1–5 ppm).

**Bacteriostatic longevity: day-to-day corrected zone of inhibition assay**

The bacteriostatic longevity of the dressings was assessed using day-to-day transfer corrected zone of inhibition (CZOI) assays. The method used for this procedure, a modified form of the Kirby–Bauer assay, is similar to that of Wright et al. (11), with modifications as described below. Briefly, 100 μl of *S. aureus* taken from an overnight culture was spread onto MHA plates, and silver dressing pieces pre-moistened with their saturation volume of distilled water were then placed onto the centre of the plates. The original dressing placement was traced onto the bottom of the Petri plate to correct for dressings which shrank over time (as with the silver collagen matrix dressing). The plates were incubated overnight at 37°C and then the zones of bacterial inhibition and dressing widths (or tracings as appropriate) were measured in two perpendicular directions. The CZOI was calculated by subtracting the dressing width from the zone width, and the results for the two directions were averaged. After zone measurement, the dressings were then transferred to new bacteria-seeded MHA plates, and this procedure was repeated for a total of 9 days. During this period, if all three dressing pieces of an experimental group ceased to produce any zone of inhibition, they were eliminated from the procedure. The shaped ionic silver foam dressing was not tested in this protocol (see Discussion).

**Statistics**

One-way analysis of variance tests with Tukey–Kramer multiple comparisons post-tests were performed for all assays in which more than two dressings were compared (log reductions, absorptive capacity, silver release and CZOI assays up to day 6). For days 7–9 in the CZOI assays, where only the nanocrystalline silver dressing and the silver collagen matrix dressing were still active, they were compared using unpaired *t*-tests with Welch corrections. All statistical analyses were performed using Graphpad InStat Version 3.06 (GraphPad Software, San Diego, CA; © 2003, www.graphpad.com).

**RESULTS**

Table 1 shows the dressing saturation volumes for each commercially available silver dressing tested. There were significant differences in absorptive capacity between dressings (*P* < 0.0001). The silver sulphate dressing had a significantly higher absorptive capacity than all other dressings tested (*P* < 0.001), while the non adhesive ionic silver foam dressing had a significantly higher absorptive capacity than all the remaining dressings (*P* < 0.001). The only other significant differences were that the silver collagen matrix dressing, and the shaped ionic silver foam dressing had significantly higher absorptive capacities than the nanocrystalline silver dressing (*P* < 0.05).

Table 2 shows the log reductions measured for each commercially available silver dressing tested. There were significant differences in the activity of the various silver-containing dressings (*P* < 0.0001). The nanocrystalline silver dressing produced significantly higher log reductions than all other dressings tested (*P* < 0.001). It was also the only dressing to produce a total kill, and the only dressing that was bactericidal, where the definition of bactericidal is a dressing capable of producing a log reduction greater than three (9). The silver collagen matrix dressing produced significantly higher log reductions than the remaining four dressings (*P* < 0.001). The remaining four dressings showed no significant differences.

<table>
<thead>
<tr>
<th>Dressing description</th>
<th>Average log reduction (±SD, n = 3)</th>
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<tr>
<td>Silver sulphate dressing</td>
<td>−0.61 ± 0.08</td>
</tr>
<tr>
<td>Non adhesive ionic silver foam dressing</td>
<td>−0.03 ± 0.01</td>
</tr>
<tr>
<td>Shaped ionic silver foam dressing</td>
<td>−0.33 ± 0.08</td>
</tr>
<tr>
<td>Silver alginate dressing</td>
<td>−0.08 ± 0.30</td>
</tr>
<tr>
<td>Silver collagen matrix dressing</td>
<td>1.18 ± 0.29</td>
</tr>
<tr>
<td>Nanocrystalline silver dressing</td>
<td>&gt;3.46 ± 0.00</td>
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</tbody>
</table>
from one another, and all produced no positive log reductions.

Table 3 shows the silver release from the commercially available silver-containing dressings into distilled water after a 24-hour period. There were significant differences between groups ($P < 0.0001$). The silver sulphate dressing released significantly more silver than all other dressings ($P < 0.001$), and the nanocrystalline silver dressing released significantly more silver than the remaining dressings ($P < 0.001$). The other dressings were not significantly different from one another.

The CZOIs over a 9-day period are shown in Figure 1 for the nanocrystalline silver dressing ($n = 3$), the silver collagen matrix dressing ($n = 2$) and the silver alginate dressing ($n = 3$). The silver sulphate dressing ($n = 3$) and non adhesive ionic silver foam dressing ($n = 2$) did not generate any zones of inhibition on the first day of testing and had S. aureus growing under the dressings. Therefore, no transfers were performed for these dressings. The nanocrystalline silver dressing generated consistent zone sizes until the experiment was terminated after 9 days. The silver collagen matrix dressing zone sizes began to decrease after day 7 ($P < 0.05$) but were still present out to day 9. The silver alginate dressing zone size decreased after day 1 ($P < 0.001$). Smaller zones were generated by the silver alginate dressing out to day 6, by which point the dressing completely liquefied, making further transfers impossible. There were significant differences ($P < 0.05$) in zone sizes between dressings on all days up to day 6, with the silver alginate dressing having the largest zone on the first day, and the smallest zone on all subsequent days. On all days but days 4 and 5, the silver collagen dressing produced smaller zones than the nanocrystalline silver dressing, but there were no significant differences in zone sizes between these two dressings except on day 1.

**DISCUSSION**

In 30 minutes, only the nanocrystalline silver dressing was bactericidal against S. aureus, contrary to the product information for some of the other dressings. This corresponded to release of greater concentrations of active silver compared with most of the other dressings tested. Interestingly, the silver released by the nanocrystalline silver was about 6% of traditional silver treatments which release Ag$^+$ only. Silver nitrate is provided at 2.95–5.91 mg/cm$^2$/day, while silver sulphadiazine is provided at 1.21 mg/cm$^2$/day in burn treatment (12,13). The nanocrystalline silver dressing also showed an inhibitory effect for over 9 days of in vitro challenges with S. aureus, indicating sustained release of active silver. These results are in good agreement with those of previous studies (9,14,15). Gallant-Behm *et al.* (9) found that of eight silver-containing dressings tested, only nanocrystalline silver dressings produced bactericidal activity, which was related to its rapid sustained release of active silver. Thomas and McCubbin (14,15) gave nanocrystalline silver dressings the highest score when comparing 10 silver-containing dressings, and attributed its antimicrobial activity to the rapid release of active silver. Wright *et al.* (16) found that relative to silver nitrate, silver sulphadiazine and mafenide acetate, nanocrystalline silver provided the fastest, broadest spectrum fungicidal activity. Yin *et al.* (17) compared these same dressings against five wound pathogens and came to the same conclusions. Another study by Wright *et al.* (11), comparing the nanocrystalline silver dressing to a controlled release silver film dressing, found that the nanocrystalline silver showed more rapid bactericidal activity against a broader spectrum of organisms. Wright *et al.* (18) compared nanocrystalline silver dressing with polyhexamethylene biguanide (PHMB), and they both showed

<table>
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<tr>
<th>Dressing description</th>
<th>Average silver release (ppm ± SD)</th>
<th>Average silver release (mg/cm$^2$ ± SD)</th>
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<tbody>
<tr>
<td>Silver sulphate</td>
<td>$318.63 ± 15.72$</td>
<td>$0.69808 ± 0.03124$ (n = 3)</td>
</tr>
<tr>
<td>Non adhesive ionic silver foam</td>
<td>$0.10 ± 0.02$</td>
<td>$0.00014 ± 0.00004$ (n = 3)</td>
</tr>
<tr>
<td>Shaped ionic silver foam</td>
<td>$0.13 ± 0.11$</td>
<td>$0.00009 ± 0.00007$ (n = 2)</td>
</tr>
<tr>
<td>Silver alginate</td>
<td>$2.58 ± 1.58$</td>
<td>$0.00401 ± 0.00245$ (n = 2)</td>
</tr>
<tr>
<td>Silver collagen matrix</td>
<td>$0.26 ± 0.11$</td>
<td>$0.00040 ± 0.00018$ (n = 2)</td>
</tr>
<tr>
<td>Nanocrystalline silver</td>
<td>$92.87 ± 5.88$</td>
<td>$0.14398 ± 0.00911$ (n = 3)</td>
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potent in vitro bactericidal activity. However, in a porcine full-thickness infected wound model, PHMB reduced the bacterial population initially, but could not keep it in check to the same degree as the nanocrystalline silver dressing (18). The PHMB-containing dressing was also highly inflammatory in the wound bed, delaying healing (18), whereas nanocrystalline silver shows anti-inflammatory/prohealing activity (19). The antimicrobial efficacy of nanocrystalline silver dressings showed in these studies has been corroborated by other in vivo and clinical studies (20–23). This suggests that the species of silver released, as well as the
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Total silver released, appear to be important factors for activity, because the dressing showed good antimicrobial activity despite releasing 6% of the amount of silver provided by traditional silver treatments. Based on the product information, nanocrystalline silver dressings were the only dressings in this study to release species other than ionic silver ($\ce{Ag^+}$), also releasing metallic silver ($\ce{Ag^{0}}$, likely in cluster form) and a higher oxidation state species (24). This dressing, which is designed to be a porous moisture wicking wound interface, has a low absorptive capacity – significantly lower than the other dressings except the silver alginate dressing – because the only absorptive material is the middle gauze layer, and therefore it requires a secondary dressing for clinical use. It did not shed dressing components during any of the assays.

Despite the fact that the silver sulphate dressing released high quantities of silver compared with the other dressings (about 30% of the amount provided by traditional silver-containing burn treatments), it was not able to generate any log reduction in 30 minutes, or any CZOI, with bacteria growing under the wound-contacting surface of the dressing, suggesting it had no bacteriostatic activity. Both the top surface and the wound-contacting surface of the dressing were hydrophobic – they both repelled water droplets – preventing fluid from entering, or silver from being released from, the dressing through these surfaces. Silver could, however, be released by cut dressing edges (Figure 2). Thus, during the log reduction assay, when bacteria were placed on the wound-contacting surface, they did not contact silver within the dressing. Similarly, when the wound-contacting surface was placed down onto the agar seeded with $\ce{S. aureus}$, no liquid transfer occurred between the plate and the dressing, no silver was released onto the plate and therefore no CZOI was produced. The silver released into solution during the silver dissolution assay was released through the edges of the dressing – the only portion of the dressing which allowed for fluid exchange. This was confirmed by cutting a silver sulphate dressing into strips, moistening the strips to their saturation volume and placing them onto plates seeded with $\ce{S. aureus}$ such that the dressings contacted the plate cut edge down. The plates were then incubated overnight at 37°C, and in this orientation, the dressing was able to produce CZOIs. Unfortunately, this orientation is not clinically relevant, as the edges/interiors of the dressing are unlikely to contact the wound. In its current configuration, minimal silver would be released into the wound and likely no bacteria would be taken into the dressing because of no fluid exchange. The saturation volume measured is not clinically relevant either, because the dressing was submerged and fluid could enter through the edges. The absorption capacity in a wound environment for this dressing configuration appears to be essentially zero. Overall, although the dressing contains a large quantity of soluble silver, in its current configuration, it has neither wound fluid absorption capacity nor antimicrobial activity. Product information for the dressing confirms that its wound-contacting surface is hydrophobic, despite its claims of absorbency, as it indicates that ‘The [wound-contacting] layer seals the wound edges, preventing the exudate to [sic] leak onto the surrounding skin, thus minimizing the risk of maceration.’ (4)

There are no studies concerning the silver sulphate dressing in the peer-reviewed literature, to the knowledge of the authors. A handful of case studies (25–29) and one open non comparative multicentre investigation (30) have been presented as posters, which did not show direct measures of antimicrobial activity. One poster on an open non comparative single-centre study did include microbiological culture swabs and concluded that the dressing showed antimicrobial efficacy. However, their tabulated data showed that 6 of 11 patients initially colonised with $\ce{S. aureus}$...
aureus, and 9 of 11 patients initially colonised with Pseudomonas aeruginosa, had little or no reduction of these species after 30 days treatment (31). Another poster on an in vitro study indicated that the dressing was bactericidal against various wound pathogens within 30 minutes to 3 hours and could be challenged daily for 7 days and retain its activity (32). However, for all tests they put 1 g of dressing in 50 ml of media plus bacteria. One gram of this dressing has an area of 5.9 cm², and thus, in their assay, the dressing was instantly exposed to approximately 8.5 g/cm² of fluid. Because wound exudate is classified as mild (0.25 g/cm²/24 hours), moderate (0.5 g/cm²/24 hours) or heavy (1.0 g/cm²/24 hours) (33), the method used above would instantly expose the dressings to 8.5 times the 24-hour fluid volume of highly exudative wounds, suggesting that the results of their study may not be indicative of the clinical efficacy of the dressings (34). Furthermore, because this study was performed with the dressings submerged, this would allow for contact between bacteria and silver through the absorbent edges of the dressing, explaining the discrepancy between that study (32) and the current study.

The silver collagen matrix dressing was not bactericidal in half an hour. It was bacteriostatic for 9 days. The silver released was significantly lower than that of the nanocrystalline silver dressing, releasing 0.02% of the Ag⁺ provided by traditional silver-containing burn treatments. This suggests at least a partial explanation for its lack of bactericidal efficacy. To the knowledge of the authors, there are no studies concerning the silver collagen matrix dressing in the peer-reviewed literature. There are a handful of case studies which have been presented as posters (35-39), which do not focus on the antimicrobial activity of the dressing. In the CZOI assay, the dressing edges left behind material from the fifth transfer on, and also left material in solution during the total silver assay, suggesting that material could be left behind in a wound bed, which could result in generation of a foreign body response, depending on dressing bioabsorbability. The absorptive capacity of the silver collagen matrix dressing was mid-range. Overall, although the silver collagen matrix dressing did show bacteriostatic longevity, it did not appear to be the best product examined in this study in terms of its antimicrobial activity, its absorptive capacity or its use. However, it is only indicated for prevention of bioburden in the dressings, unlike the other materials tested in this study, which should be taken into consideration.

The only other dressing showing any antimicrobial activity in vitro was the silver alginate dressing. Although the dressing was unable to generate any log reduction in 30 minutes against S. aureus, it was able to generate CZOIs for 6 days. The zone size was much higher on the first day than on subsequent days, suggesting an initial dump of silver, followed by a lower sustained release. Thus, the dressing was not bactericidal in this study, but it was bacteriostatic. There are no studies concerning the silver alginate dressing to the knowledge of the authors – their product claims, which do not match the results of this study, are based on data the company has on file. The results of this study may have serious consequences, as studies have indicated that when an antimicrobial agent is provided in concentrations such that bacterial inhibition occurs but the bacteria are not actually killed, selection for resistant organisms occurs (40,41). The antimicrobial activity of this dressing appears to be related to its silver release, which was significantly lower than that of the only bactericidal dressing, but was higher than some dressings which were unable to generate bacteriostatic activity. The silver alginate dressing released approximately 0.2% of the Ag⁺ released by traditional silver-containing burn treatments. Its absorptive capacity was low and highly variable. As well, portions of the dressing flaked off during the total silver assay, and fibres were left behind after each transfer in the CZOI assay. Furthermore, the dressings began to fall apart at the third transfer, and after the fifth transfer, they liquefied completely. This suggests that – contrary to claims in the product information indicating that the dressing does not leave any silver coated nylon thread residue in the wound and remains intact, facilitating ease of removal (5) – fibres could fall off in the wound, potentially causing a foreign body response and/or delayed wound healing. Furthermore, the dressings may become more difficult to remove with time, as they may start to break apart or liquefy. It is anticipated that the dressing breakdown observed could occur more quickly in the relatively
harsh and dynamic conditions of the wound environment. It is uncertain what the wound environment would then be exposed to in terms of liquefaction products.

The product information for the shaped ionic silver foam dressing and the non adhesive ionic silver foam dressing indicates that the dressings have the same silver technology (6), and therefore they should have equal performance. This was confirmed in this study, where both dressings performed poorly, with log reductions of zero. As well, the dressings released about the same level of silver into the solution during a 24-hour period, at approximately 0.005% of the Ag$^+$ released by traditional silver-containing burn treatments. The shaped foam dressing had a significantly lower absorptive capacity than the non adhesive silver foam, possibly because of the adhesive added to the shaped dressings. The non adhesive ionic silver foam dressing was not able to generate CZOIs (bacteria grew under the wound-contacting surface), and therefore, the shaped ionic silver foam dressing is not expected to generate CZOIs either. Thus, neither of the ionic silver foam dressings showed bactericidal or bacteriostatic activity against S. aureus, corroborating the results of previous studies: There are a variety of case studies concerning the ionic silver foam dressings which have been presented as posters (42–50), which do not show direct antimicrobial measures, but there are also two in vitro studies in the peer reviewed literature (51,52) that tested an ionic silver foam dressings, along with a variety of other silver containing dressings, for antimicrobial activity using methods similar to those of Taherinejad et al. (32), described above. Both studies concluded that the ionic silver foam dressing was ineffective against P. aeruginosa and only marginally effective against S. aureus (51,52).

One of the above studies also tested the nanocrystalline silver dressing and concluded that it had a broad spectrum of bactericidal activity (52). The clinical relevance of the antimicrobial testing performed in these studies remains uncertain because of the methods used (34), however, the results of both those studies and this study indicate that the lack of activity corresponded to low silver release, as the silver released in this study was barely detectable using AAS. As well, although these dressings have a fairly high absorptive capacity, the dressings appeared to shed pieces, both into solution during the silver dissolution assay and onto plates during the CZOI assay, suggesting that in a clinical situation, dressings pieces could be left in the wound. Thus, the ionic silver foam dressings showed the worst performance overall, with the shaped dressing being inferior to the non adhesive dressing.

This study did not examine the benefits or disadvantages related to other physical or chemical components of the dressings (e.g. the glycerol and cleansing agents present in the ionic foam dressings, or the EDTA and carboxymethylcellulose present in the silver collagen matrix dressing), but these components should also be taken into consideration when deciding on the appropriateness of a dressing for a clinical situation.

Although caution must be exercised when extrapolating the results of in vitro studies to the clinical environment (15), important differences were detected in the antimicrobial efficacy of the dressings tested, with the nanocrystalline silver dressing having stronger antimicrobial activity than any of the other dressings. The silver alginate and silver collagen matrix dressings have some bacteriostatic activity, but this may not be sufficient support for immunocompromised patients indicated for treatment with these dressings. Furthermore, exposure of bacteria to silver concentrations which are bacteriostatic but not bactericidal creates a high-risk situation in terms of development of silver-resistant bacteria (40,41). The silver sulphate dressing does not appear to have clinically relevant antimicrobial activity in its current configuration because of the hydrophobicity of the wound-contacting surface, while the ionic silver foam dressings do not appear to contain sufficient silver for antimicrobial activity. The latter three dressings, which were the only dressings in the study described as non staining, were the dressings that showed neither bactericidal nor bacteriostatic activity, suggesting that non staining Ag$^+$-containing products may not release enough silver to be effective.

The results of this study emphasise the importance of confirming claims made in product information through simple in vitro efficacy tests. If a dressing does not show efficacy in the relatively benign environment of a Petri plate, it is unlikely to do so in the wound environment. Finally, this study corroborates the conclusions of others that silver release, log reduction and day-to-day transfer CZOI assays
should be used in conjunction to analyse silver-containing dressings (9,17). Sufficient information about the antimicrobial activity of the dressings examined in this study, and more specifically why certain dressings were active while others were not, could not have been gained from running only one of the above assays.

ACKNOWLEDGEMENTS
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