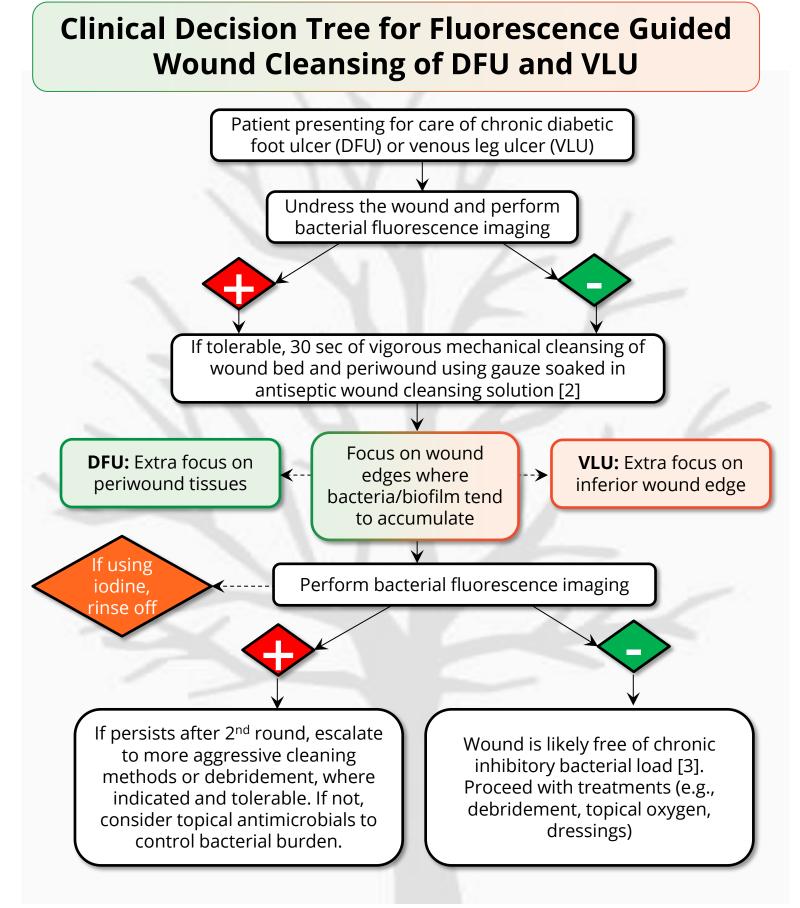
Improving Wound Hygiene Through Fluorescence Imaging: A Focus on Spatial Patterns in Diabetic and Venous Leg Ulcers

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The Modern Era of Wound Hygiene:

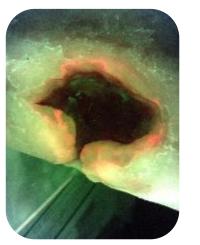
- Wound cleansing is necessary to put stalled wounds back onto a healing trajectory and to prevent infection and its complications.
- The advent of bacterial fluorescence imaging technology (MolecuLight[®]) for wound care has changed our understanding of bacterial distribution in wounds and allows us to evaluate wound hygiene efficacy in real-time.
- Clinical studies show that significant bacterial colonization is frequently left behind following standard cleansing practices [1], highlighting the potential for improvement.
- Objective, reliable methods to improve wound hygiene are much needed. This leads us to present practical guidance on using fluorescence-imaging to accomplish more effective hygiene.



What Does Fluorescence Imaging Tell Us About Bacteria in DFU and VLU?

- For VLUs, cyan fluorescence indicating *Pseudomonas aeruginosa* seems to be more common than red fluorescence. It is often clustered along the wound edge, especially inferiorly, possibly due to gravity.
- Red fluorescence indicating a mixture of bacterial species is more common in **DFUs**, usually forming a "ring" around the wound edge that often extends into the periwound.

Spatial Patterns of Bacterial Fluorescence in <u>DFU</u>



Clinical Case: Fluorescence "ring" in DFU persists after routine hygiene

- from the wound edge (Figure 1A and Figure 2A).
- measures may be recommended if bacterial fluorescence persists.

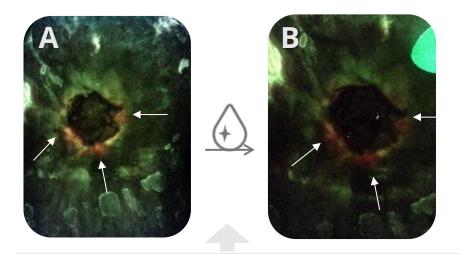


Figure 1 (above): Pre-debridement (A) and post-cleansing and debridement (B) DFU, demonstrating a persistent red fluorescent ring in the periwound.

Figure 2 (right): Fluorescence images of a DFU taken predebridement (A) and post-debridement and cleanse (B), demonstrating significantly less red fluorescence. A few months later, red fluorescence was once again observed (C) and eliminated by fluorescence-targeted cleansing.

Arrows = fluorescence from high bacterial loads (>10⁴ CFU/g)

<u>References: [1]</u> Moelleken et al. Prospective clinical study on the efficacy of bacterial removal with mechanical debridement in and around chronic leg ulcers assessed with fluorescence imaging. Int Wound J., 2020 Aug;17(4):1011-1018. [2] Oropallo et al. An objective comparative study of non-surgical cleansing techniques and cleanser types in bacterial burden management. Int Wound J., 2024 Feb;21(2):e14730. [3] Armstrong et al. Point-of-care fluorescence imaging reveals extent of bacterial load in diabetic foot ulcers. Int Wound J., 2023 Feb;20(2):554-566. [4] Oropallo AR, Andersen C, Abdo R, Hurlow J, Kelso M, Melin M, Serena TE. Guidelines for point-of-care fluorescence imaging for detection of wound bacterial burden based on Delphi consensus. Diagnostics, 2021 Jul 6;11(7):1219.

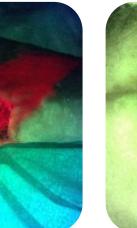
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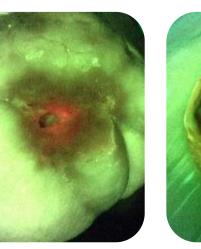
• Chronic VLUs and DFUs frequently display fluorescence signals indicating high levels of bacterial colonization that impede healing and increase infection risk [3].

Conclusions:

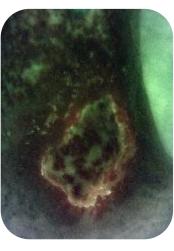
- with published guidelines for fluorescence imaging [4].

Spatial Patterns of Bacterial Fluorescence in VLU



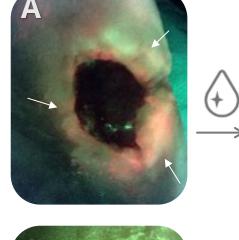






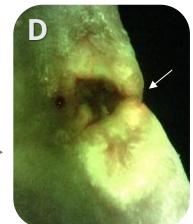
Red fluorescence indicates a mixture of Gram +/- bacterial species at loads >10⁴ CFU/g. In DFUs, red fluorescence is often observed, usually forming a "ring of fire" around the ulcer that extends a few centimeters

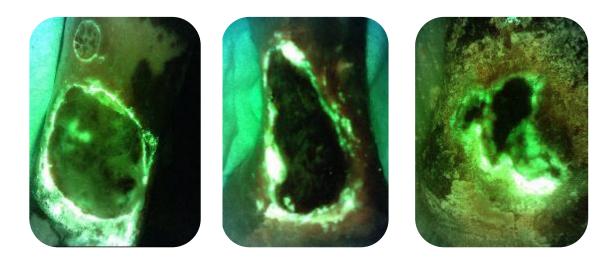
In some cases, cleansing and debridement are insufficient, leaving significant bacterial burden (Figure 1B). It is also not uncommon for debridement to uncover deeper bacterial affection. Fluorescence-guided cleansing towards bacterial-laden regions improves wound hygiene (Figure 2B). However, additional rounds and/or





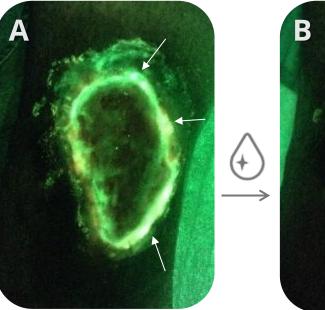






Clinical Case: *Pseudomonas* clusters along inferior edge of VLU

- the inferior wound edge, possibly due to pooling of exudate.
- particularly for *P. aeruginosa*.
- decreased (Figure 3B and Figure 4B).



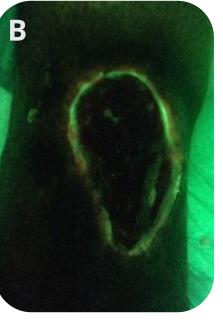


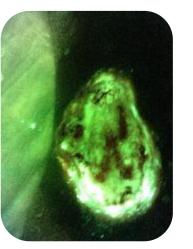
Figure 3: Pre-debridement fluorescent images (A) reveal multiple clusters of cyan fluorescence in a VLU, which was eliminated following debridement and fluorescence-targeted cleansing (B).

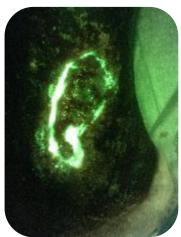


• Fluorescence imaging is a useful technology that can improve the effectiveness of routine wound hygiene at the bedside and supports an objective approach to bacterial-infection management.

Clinically unremarkable VLUs and DFUs display consistent spatial patterns of bacterial fluorescence that can be identified and targeted with fluorescence-guided cleansing.

• When bacterial fluorescence persists after iterative rounds of fluorescence-guided hygiene, additional interventions including debridement and topical antimicrobials may be required. This approach aligns





• Cyan fluorescent signals indicates the presence of *P. aeruginosa*, which is frequently observed VLUs and often clusters along the wound edge (Figure 3 A and Figure 4 A). Cyan fluorescence is also often further concentrated to

• Wound care providers should exercise a high index of suspicion for bacterial colonization in asymptomatic VLUs,

• Following a single round of fluorescence-guided wound cleansing, the presence of cyan fluorescence greatly

• This highlights the efficacy of fluorescence imaging as a tool to manage bacterial loads with proper cleansing.

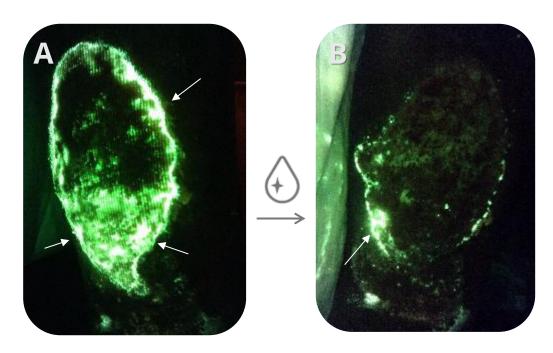


Figure 4: Clusters of cyan fluorescent signals indicative of *P. aeruginosa* observed in a VLU pre-debridement. These fluorescent signals were greatly reduced following fluorescence-targeted cleansing (B).