

Fluorescence imaging guided dressing change frequency during negative pressure wound therapy: a case series

Objective: Knowledge of wound bioburden can guide selection of therapies, for example, the use of negative pressure wound therapy (NPWT) devices with instillation in a heavily contaminated wound. Wound and periwound bacteria can be visualised in real-time using a novel, non-contact, handheld fluorescence imaging device that emits a safe violet light. This device was used to monitor bacterial burden in patients undergoing NPWT.

Methods: Diverse wounds undergoing NPWT were imaged for bacterial (red or cyan) fluorescence as part of routine wound assessments.

Results: We assessed 11 wounds undergoing NPWT. Bacterial fluorescence was detected under sealed, optically-transparent (routine) adhesive before dressing changes, on foam dressings, within the wound bed, and on periwound tissues. Bacterial visualisation in real-time helped to guide: (1) bioburden-based, personalised treatment regimens, (2) clinician selection of NPWT, with or without instillation of

wound cleansers, and (3) the extent and location of wound cleaning during dressing changes. The ability to visualise bacteria before removal of dressings led to expedited dressing changes when heavy bioburden was detected and postponement of dressing changes for 24 hours when red fluorescence was not observed, avoiding unnecessary disturbance of the wound bed.

Conclusion: Fluorescence imaging of bacteria prompted and helped guide the timing of dressing changes, the extent of wound cleaning, and selection of the appropriate and most cost-effective NPWT (standard versus instillation). These results highlight the capability of bacterial fluorescence imaging to provide invaluable real-time information on a wound's bioburden, contributing to clinician treatment decisions in cases where bacterial contamination could impede wound healing.

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An optimal wound care treatment plan requires initial patient and wound assessment and comprehensive monitoring to re-evaluate wound status, bioburden and effectiveness of the chosen wound therapies. Negative pressure wound therapy (NPWT) has been established as an advanced therapy which improves wound closure rates by increasing local blood flow and formulation of granulation tissue.¹⁻³

Evidence to date suggests that bioburden control is not a clear benefit of standard NPWT.⁴⁻⁷ Some studies report NPWT suppression of non-fermentative Gram-negative bacilli, for example, *Pseudomonas* spp., but this is associated with enhanced growth of Gram-positive bacterial species.⁸ NPWT with instillation (NPWTi) can be used on challenging wounds which would benefit from a combination of vacuum-assisted closure and constant irrigation with topical wound solutions, such as wound cleansers, antiseptics or saline. Most studies show that this therapy removes bioburden and other contaminants without manual intervention or disruption.^{4,7,8}

The diverse wounds on which experts suggest beneficial use of NPWTi includes chronically infected or contaminated wounds, wounds in patients with diabetes, traumatic wounds, those with exposed bone or underlying osteomyelitis and painful wounds.⁹ Management of bioburden in these wounds is critical and underscores why NPWTi of wound cleansers could be successful, when used appropriately.^{1,10} However, treatment with standard NPWT, a lower cost treatment option,¹¹ is common practice for many of these wound types, often once bioburden has been controlled. Ultimately, treatment selection must be customised on a wound-by-wound basis by the treating clinician after thorough assessment.¹²

Knowledge of when to select NPWTi over standard NPWT, how long to use NPWTi, the ideal length of time between dressing changes and how to optimise these resources has been hampered for several reasons:

- NPWTi is the more novel therapy, therefore evidence is continuously evolving as large clinical studies and retrospective reviews enter the literature^{1,9,13}
- Monitoring of bioburden status via sampling or via clinical signs and symptoms requires removal of the adhesive and NPWT foam, i.e a complete dressing change, which is costly in terms of both materials and clinician time^{14,15}
- Wound sampling is further associated with delays while awaiting microbiological results.

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To address the latter concerns, wounds undergoing NPWT were assessed using a handheld bacterial fluorescence imaging device (MolecuLight Inc, Toronto, Canada) that visualises bacterial fluorescence in real-time.

Bacterial fluorescence imaging is a method to quickly visualise and monitor bacterial presence in and around wounds at the bedside.^{16,17} This is a safe, non-contact and contrast agent-free method to visualise bacteria in real-time using a portable handheld device. The device emits a violet light (405nm), which excites the tissue and bacteria within and around a wound. This causes tissues to fluoresce green while bacteria fluoresce either red (most bacteria species) or cyan (*Pseudomonas aeruginosa*), enabling immediate bacterial localisation.^{16,17} A clinical trial demonstrated that bacterial (red) fluorescence positively predicted the presence of bacteria at loads of clinical concern, $\geq 10^4$ colony forming units (CFU)/g or moderate/heavy growth, in 100% of the 60 studied wounds.¹⁷ This and other clinical trials have demonstrated the bacterial fluorescence imaging device's ability to detect the most common wound pathogens, including *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa*.¹⁶⁻¹⁹ The clinical use of the bacterial fluorescence imaging device has previously been established for chronic wounds,^{16,18,20,21} burn and trauma wounds^{22,23} and surgical wounds.^{24,25} However, its specific usage in conjunction with NPWT has not been described. This case series reports the use of bacterial fluorescence imaging in conjunction with routine wound assessments for clinical signs and symptoms of infection. Real-time information on bioburden was obtained from fluorescence images, including the presence or absence of bacterial fluorescence, its location and observed changes over time. This information significantly impacted treatment decisions and was used to improve resource use.

Methods

Patients

This case series includes both chronic and surgical wounds, diverse wound locations and a variety of negative pressure systems. Informed written consent was obtained from each patient for publication of their wound images and anonymous case information in a scientific publication format. Over a three-month period, all Scarborough Rouge Hospital inpatients and wound care clinic outpatients receiving NPWT from the wound care specialist (author) performing this study were eligible for inclusion.

Wound care patients not undergoing NPWT and any patients unable or unwilling to provide consent were ineligible for this study. All wounds were assessed by the wound care clinician as per standard of care for clinical signs and symptoms of infection.²⁶ The standard of care was based on severity and location of wound and included cleansing with appropriate antiseptic solution, either povidone iodine or

chlorhexidine, and irrigation with antiseptic (solutions such as hypochlorous acid compound from the pharmacy or commercially available) during installation when appropriate and dressing changes between 2–4 days. If dressing changes were less frequent than every two days, imaging was performed daily. Wound assessments by this clinician also routinely included bacterial fluorescence imaging to provide information on bacterial presence and location. Hospital inpatients also received wound care from additional attending wound care clinicians who do not incorporate fluorescence imaging into their wound assessments.

Imaging procedure

Fluorescence images, obtained in real-time, were used to determine if and where significant bacterial loads ($\geq 10^4$ CFU/g)¹⁷ were present in wounds. Images were taken during routine wound assessments, most often at scheduled NPWT dressing changes or immediately before initiation of NPWT. Images were acquired using the MolecuLight i:X Imaging Device (MolecuLight Inc, Toronto, Canada), as previously described in detail.¹⁷ Standard images of the wound were acquired under traditional room lighting conditions, after which the room was made dark, the violet excitation light from the device was turned on and fluorescence images were acquired. When excited by the imaging device's 405nm violet light, tissues fluoresce green while the majority of bacterial pathogens fluoresce red (for example, *Staphylococcus aureus*, *Escherichia coli*, *Enterobacter* spp., *Klebsiella* spp., *Proteus* spp.). *Pseudomonas aeruginosa* uniquely fluoresce cyan in colour. Specialised optical filters reveal these red, green and cyan signals in real-time on the device's display screen.¹⁶⁻¹⁸ A range finder on the device was used to ensure all images were taken at the optimal imaging distance (8–12cm) and a light sensor on the device indicated when the room was dark enough for fluorescence images to be acquired. If sufficient darkness could not be achieved by turning off room lighting, for example in rooms with windows, a disposable dark drape attachment (MolecuLight Inc) was used to provide the required darkness. The dark drape attaches onto the device and blocks all light, establishing an area of complete darkness over the wound for image acquisition in fluorescence mode.

This imaging device was used per its intended use for visualising bacteria within and around wounds. Images acquired before NPWT dressing changes were taken through the sealed, optically transparent adhesives routinely used with NPWT. If a dressing change occurred, additional images were acquired after removal of all adhesives. Wounds exhibiting red fluorescence were considered to have clinically concerning bacterial loads. Any changes to wound management as a result of bacterial fluorescence images were noted.

Results

Patients ranged in age from 18 to 87 years. We assessed

Table 1. Summary of cases in which bacterial fluorescence imaging was beneficially used in combination with NPWT

Patient	Wound type	Time of imaging	Bacterial fluorescence?	Microbiological swab confirmation?	FL-guided treatment change	Effect on short term wound care cost
1	Pressure ulcer (sacral)	At numerous dressing changes	Positive	Heavy growth of <i>Escherichia coli</i> , <i>Enterococcus faecalis</i> , <i>Staphylococcus aureus</i> ; light growth <i>Proteus vulgaris</i>	(1) Prompted early dressing changes (2) Prompted switch to NPWTi (3) Demonstrated effectiveness of instillation (4) Guided targeted cleaning at dressing changes	Increase
2	Necrotising fasciitis (pectoral)	After 2, 4 and 6 weeks of NPWT + instillation	Negative	Negative for bacteria	Negative images led to a 24-hour postponement of dressing change, leaving wound bed undisturbed	Decrease
3	Surgical wound (abdominal)	Before initiating NPWT	Positive	N/A	Images confirming bacterial burden prompted selection of NPWT + instillation, rather than standard NPWT	Increase
4	Necrotising fasciitis (scrotum)	At scheduled dressing changes	Positive	Heavy growth mixed anaerobes; light growth coagulase negative Staphylococci	(1) Confirmed continued presence of bacterial burden, (2) Facilitated education on necessity of dressing change in patient refusing dressing change due to fear of pain	No change
5	Surgical wound (appendectomy abscess)	At scheduled dressing change	Positive	Moderate growth <i>Escherichia coli</i> , light growth <i>Propionibacterium acnes</i> , very light growth <i>Staphylococcus aureus</i>	Guided additional, targeted cleaning during dressing change	Increase
6	Two pressure ulcers (sacral)	At scheduled dressing change	Wound 1: positive Wound 2: negative	N/A	(1) Wound positive for bacterial fluorescence had scheduled dressing change, (2) wound negative for bacterial fluorescence was left undisturbed for another 24 hours	Decrease
7	Breast abscess	At scheduled dressing change	Positive	Heavy growth <i>Diphtheroid bacillis</i> , moderate growth <i>Klebsiella pneumoniae</i>	(1) Led to maintenance on NPWTi (2) Guided additional cleaning during dressing change	Increase
8	Surgical wound (pilonidal sinus)	At scheduled dressing change	Positive	Heavy growth <i>Bacteroides fragilis</i> , <i>Morganella morganii</i> , <i>Streptococcus agalactiae</i>	(1) Guided additional, targeted cleaning during dressing change and (2) Facilitated patient education	Increase
9	Venous leg ulcer (calf)	At scheduled dressing change	Negative	N/A	Clinician confidence to discontinue use of antimicrobials and continue NPWT with regular gauze	Decrease
10	Surgical wound (abdominoplasty)	At scheduled dressing change	Positive	N/A	Guided additional (1) targeted cleaning, (2) prompted earlier dressing changes, (3) prompted use of silver product in combination with NPWT	Increase

FL—fluorescence; NPWT—negative pressure wound therapy; NPWTi—NPWT with instillation

11 wounds undergoing NPWT (10 patients in total) were imaged for bacterial fluorescence as part of this study. A beneficial effect of bacterial fluorescence images on wound management was noted for all 11 wounds; these benefits are described in Table 1 along with information on wound type and any available microbiological findings. Of the 11 wounds, eight were positive for bacterial fluorescence and three were negative for bacterial fluorescence. Wound swabs were taken in six of the 11 wounds and in all six cases culture

results confirmed the bacterial positive or bacterial negative imaging findings. Culture results from fluorescence-positive wounds confirmed the presence of moderate or heavy growth of numerous common wound pathogens (*Escherichia coli*, *Enterococcus faecalis*, *Staphylococcus aureus*, *Diphtheroid bacillis*, *Klebsiella pneumoniae*, *Bacteroides fragilis*, *Morganella morganii*, *Streptococcus agalactiae*, mixed anaerobes).

Images in this study visualised bacteria that had been drawn out from the wound bed and tissues and brought

Fig 1. Detection of bacterial (red) fluorescence in a deep sacral pressure ulcer that had recently begun NPWT. (a–b) Fluorescence images acquired in week two, three days after initiation of NPWT with instillation (NPWTi), detected bacterial (red) fluorescence under sealed, optically-transparent (routine) adhesive before dressing changes (a), within the wound bed and on periwound tissues (b). Regions of red fluorescence in and around the wound bed are highlighted with arrows. The patient was switched to regular NPWT after 10 days, as per NPWT guidelines and standard practice. In week 4 (c–e), widespread bacterial fluorescence was visualised under sealed adhesive (d) just 48 hours after a dressing change. This prompted an early dressing change during which bacterial fluorescence was also present on foam dressing (e). Patient was returned to NPWTi. In week 5, after one week of NPWTi (f–g), bacterial fluorescence was notably reduced to a small region of the packed wound (g, red, circled), demonstrating the effectiveness of instillation treatment. In week 9 (h–k) a small region of red fluorescence (circles) was still detectable under adhesive (h) and was also observed on comparable region of foam dressing (k) and on periwound tissue (j); no red fluorescence was detected on the wound bed. Periwound fluorescence prompted additional cleaning of periwound region (k), better preparing the wound bed for NPWT and maintenance of this patient on NPWTi, rather than the planned return to standard NPWT



into the foam and/or tubing through negative pressure application to the wound. The compelling images of this process demonstrated that NPWT is effective in removing bacterial burden from the wound bed, at least to some extent (Fig 1). In the absence of this drawing out, the bacteria presumably would have remained within the wound tissues, likely delaying healing.

Beneficial effects of bacterial fluorescence imaging on treatment plans were observed for management of both chronic wounds, for example, pressure ulcers (PU), venous leg ulcers (VLU) and of a variety of surgical wounds. Instantaneous information on the absence or presence and location of bacterial (red) fluorescence within the wound and periwound tissue guided numerous treatment decisions. For example, dressing change timing was influenced in five study wounds. Dressing changes were postponed for 24 hours in two inpatient wounds negative for bacterial fluorescence, leaving those wounds to heal without disturbance and saving clinician time and resources, and were expedited in cases where bacterial burden was clearly present. Additional wound management strategies employed as a result of images positive for bacterial fluorescence included:

- Prompted initiation of or maintenance on NPWTi with wound cleansers (3/10 patients)
- Prompted use of antimicrobial products (for example, silver containing products and polyhexamethylene biguanide gauze) in conjunction with NPWT (1/10 patients)
- Led to additional, targeted cleaning during dressing changes (5/10 patients)
- Facilitated patient education (2/10 patients).

The simple colours on images (green–tissue, red–bacterial) also facilitated patient education on the necessity of their dressing changes or on the necessity of improved patient hygiene. Additional details can be found in Table 1. Note that treatment decisions were made after evaluation of fluorescence information as well as standard assessment for clinical signs and symptoms, patient condition and history and before microbiological findings (when applicable). Treatment decisions were based on clinician’s judgement of this combined information.

This study demonstrated that 9/10 patients were likely to have received inappropriately triaged care without the fluorescence information (Table 1). We observed that 60% of patients were to be undertreated; these patients therefore benefited from increased resources in the short term, while 30% of patients were being overtreated and fluorescence information prompted a decrease in resource use. Based on microbiological confirmation (when available) and wound progress in these patients, triaging resources elsewhere did not compromise wound progress.

We will discuss two cases in detail (patients 1 and 2 in Table 1), one PU positive for bacterial fluorescence and one healing necrotising myelitis wound negative for bacterial fluorescence. These wounds were imaged at

numerous timepoints during NPWT treatment and swabs confirmed the findings of fluorescence images.

Case 1

An 87-year-old, female patient fell from a chair in her home while reaching for a ceiling fan chain. She fractured her right femur from the fall, was immobile, and spent three days on the floor before discovery. During this time a deep, unstageable sacral PU developed. The patient was able to keep hydrated with water. At time of admission the patient suffered from rhabdomyolysis (CK=966), renal failure, hypothyroidism (TSH=13.96), and low albumin. She had a history of right nephrectomy and renal cell carcinoma, chronic obstructive pulmonary disease, dyslipidemia, hypertension, and chronic kidney disease. During her 18-week inpatient stay the patient developed pruritis, and anxiety also became a significant issue.

The patient was given a course of systemic antibiotics and her unstageable PU was debrided with hypochlorous acid (1:20) alternated with betadine-soaked gauze. This therapy was deemed ineffective at debridement and, therefore, after 1.5 weeks the patient was switched to NPWTi of hypochlorous acid (1:20) (wound size: 15x9cm, depth not recorded), with good results seen one week later (wound size: 12x8x3.5cm). NPWTi and subsequent dressing changes were extremely painful for this patient; administration of analgesics was required.

Three days after initiation of NPWTi treatment (week 2), fluorescence images acquired at a scheduled dressing change detected bacterial (red) fluorescence. These were detected:

- Before the dressing change under sealed, optically-transparent (routine) adhesive (Fig 1a)
- Within the wound bed (Fig 1b)
- On periwound tissues (Fig 1b).

After 10 days of NPWTi treatment the wound was presumed clean and the patient was switched to standard NPWT. The study clinician was not present at that wound assessment, therefore real-time information on the wound's bioburden via fluorescence imaging was not obtained and evaluated as part of this treatment decision. Rather, this change in treatment plan was made based on standard institutional practice/resource conservation^{1,13} and clinical signs and symptoms during wound assessment.

When next imaged (week 4, Fig 1c), one day before the patient's next scheduled dressing change, red bacterial fluorescence was apparent under the clear adhesive (Fig 1d) and throughout the NPWT foam (Fig 1e). This prompted an immediate dressing change and switch back to NPWTi of hypochlorous acid (1:20). Wound swabs taken at this time confirmed heavy growth of *Escherichia coli*, *Staphylococcus aureus* and *Enterococcus faecalis*, and light growth of *Proteus vulgaris*. Wound size was 11x7.5x2.5cm. After one week (week 5, Fig 1f), the patient was imaged for bacterial fluorescence again 24 hours before a scheduled dressing change. Bacterial fluorescence was notably reduced; however, a small area of red fluorescence was still evident on

images taken through the sealed, optically-transparent adhesive (Fig 1g). The dressing change was therefore expedited and the patient was maintained on NPWTi therapy.

In week 6, after 14 days of NPWTi, the patient's wound size was 8x6x4.5cm (100% granulation) and red fluorescence was present only in periwound tissue (wound bed was negative for fluorescence, images not shown). Periwound tissue received additional cleaning under fluorescence guidance and the patient was switched to standard NPWT. In week 9, a small region of red fluorescence could still be seen under adhesive, on foam and in periwound tissue (Fig 1h-k). Patient was

Fig 2. Fluorescence images confirm the absence of significant bioburden in a pectoral necrotizing fasciitis wound undergoing NPWT. (a–b) Images taken in week two of NPWT with instillation (NPWTi) were negative for bacterial fluorescence (b), as were fluorescence images acquired in week 3 (confirmed via wound culture) (d) and again in week 6 (g). Based on images, clinician delayed several dressing changes by 24 hours, leaving the wound bed undisturbed for better healing and saving clinician time and resources. (h–j) Images taken of the wound bed and foam dressing during a week 6 dressing change also were negative for red fluorescence



maintained on standard NPWT and additional, fluorescence-guided cleaning was performed until periwound red fluorescence was no longer observed (Fig 1k). Wound size was 7x4x1.5cm.

After 14 weeks, the patient was discharged to an alternate level of care and NPWT therapy was ceased due to patient mobility concerns. The wound (5.5x3cm) was treated with methylene blue dressings, and promogram and adjunctive hyperbaric oxygen therapy (HBOT) over the next month. At time of discharge from hospital, after 18 weeks of care, wound size was 6x3x0.3cm. The wound healed fully two months later.

Case 2

A 45-year-old female patient presented at the emergency room with severe right breast pain, worsening over the previous three days. The patient had a past medical history of type II diabetes (uncontrolled and medicated), hypertension, cardiac stenosis and acid reflux. Discomfort rapidly progressed while in emergency care and she became hypotensive and tachycardic. The patient was diagnosed with Group A *Streptococcus* pectoral necrotising fasciitis (blood culture). A CT scan showed extensive oedema and infiltration of the right breast and interior wall. Patient was treated with ampicillin (2g IV q 6 hours) and fluconazole. Exploratory surgery on the chest wall soft tissue and fascia was performed during which time NPWTi was initiated. Patient became palliative after suffering cardiac arrest and anoxic brain injury. The patient was maintained on ampicillin and fluconazole with NPWTi (1:20 hypochlorous acid) dressing changes 2–3 times per week.

Fluorescence images were first acquired two weeks after patient admission and the initiation of NPWT therapy. The wound was scheduled for a dressing change. However, upon seeing that images were negative for bacterial fluorescence (Fig 2b), the clinician decided to delay the dressing change by 24 hours, leaving the wound bed undisturbed. Assessment of clinical signs and symptoms supported this decision. The decision to delay a dressing change was made again in week three when images of the wound acquired through the optically-transparent adhesive were again negative for bacterial fluorescence. A wound swab taken later in the week confirmed the absence of bacteria; only *Candida tropicalis* was present. Wound size was 18x6.5x4.5cm.

In week 6 the wound was healing well (wound size 14x5x1.5cm); images acquired through the adhesive were negative for bacterial fluorescence (Fig 2g), as were images of the wound bed (Fig 2i) and the packing foam (Fig 2j). The wound continued to decrease in size until all treatments were stopped in week 10, for palliative reasons.

Discussion

This case series reports a novel use of a fluorescence imaging device, to facilitate real-time, evidenced-based decision-making around selection of optimal NPWTs and timing of NPWT dressing changes. There is

currently a reliance on manufacturer guidelines, clinician experience and/or institutional practice standards, rather than real-time evidence, for NPWT treatment decisions, for example, time between dressing changes, length of total NPWT treatment. This forces generalisation across patient care practice when, in fact, the World Union of Wound Healing Societies (WUWHS) has stated that a patient-specific treatment plan is required for each wound.¹² Yet, real-time, wound-specific evidence has been unavailable for such decisions, as a clinician cannot assess a wound being treated with NPWT without first removing the vacuum and dressings. This study demonstrates the power of bacterial fluorescence imaging to provide valuable information on bioburden before NPWT dressing removal. The potential benefits of this real-time information on improved decision making, resource management and cost savings are discussed below. This case series also reproduces the findings of other studies on bacterial fluorescence, namely that these images can be used to guide wound cleaning,²⁰ debridement,^{20,22} and patient education.^{20,27}

Case 1 demonstrates the pitfall of guideline-based treatment decisions, rather than real-time evidence. This bioburdened wound was presumed clean after 10 days of NPWTi. Yet, recent studies with NPWTi have demonstrated a bioburden reduction of only 50% in infected wounds after a week of treatment.⁷ Furthermore, because bacterial fluorescence imaging was not standard practice for other members of the wound care team, images were not acquired at the day 10 dressing change to confirm at the bedside that bioburden had been eliminated. The still-contaminated wound was switched to standard NPWT. This decision was likely made to conserve resources: Gupta et al.¹ reported a daily therapy cost for NPWTi as \$194 versus \$106 USD for standard NPWT. However, emerging studies have repeatedly shown that standard NPWT (without instillation) leads to an increase in bioburden over one week of treatment in contaminated wounds.^{4,6,7} Indeed, one week later, images acquired through sealed dressing adhesive clearly demonstrate widespread red (bacterial) fluorescence, suggesting a reversal of some initial benefits of NPWTi. Therefore, if images had been taken earlier, instillation treatment could have been continued, increasing short-term costs of instillation therapy but likely saving costs overall.¹¹ After this patient was returned to NPWTi, bacterial fluorescence images were used to monitor treatment effectiveness. When red fluorescence was observed after a further 10 days of instillation treatment this bioburden-challenged patient was maintained on NPWTi, a patient specific treatment plan based on real-time observation of the wound's bacterial burden.

Cases 2 and 6 (Table 1) demonstrate how incorporation of bedside bacterial fluorescence imaging can lead to cost savings and resource optimisation. NPWT's effectiveness in wound healing is evident from a wealth of studies over many years.^{28–30} However it is still

perceived as a costly therapy in terms of supplies and clinician time. A NPWT dressing change requires skilled personnel³¹ to remove the previous dressing and foam, assess the wound, prepare the wound bed, cut then place foam, apply adhesive, cut and prepare drainage tubing, establish a vacuum seal, program the NPWT device (in some cases) and then test the vacuum seal. In a study of 42 wounds, average time for this dressing change was 31 minutes,¹⁴ at a total labour cost of \$20/dressing change.¹⁴ In contrast, in the current study it took less than one minute to image the wound through the adhesive dressing to determine if a dressing change should be expedited or delayed. The average cost of NPWT supplies per dressing change was reported as \$69 USD in a 2017 retrospective review of >35,000 total days of NPWT occurring over 15 years,¹⁵ for a total cost of \$89 USD per dressing change. NPWT dressing changes were performed every 2–3 days in these studies, as per manufacturer guidelines. However, the ideal interval between dressing changes will vary from wound to wound, with contaminated wounds requiring much more frequent changes. A 72-patient study of non-infected trauma wounds, comparing NPWT dressing changes every three days versus seven days, found no difference in rate of complications, including infections.³² This suggests that wounds free of heavy bioburden, such as the wounds in cases 2 and 6, can receive less frequent changes, so long as they are carefully and objectively monitored by skilled personnel, monitoring that is facilitated by fluorescence imaging information. For example, over the eight weeks of bacteria-free NPWT care in case 2, delaying all dressing changes by 48 hours, in conjunction with daily bacterial fluorescence monitoring, would have decreased the number of dressing changes required from 19 to 11. This treatment plan adjustment would have decreased NPWT care costs by \$1552 USD (eight fewer dressing changes x \$194 USD) in this patient alone. This would free up resources to focus on patients struggling with wounds containing excessive bacterial burden, for example, case 1, and triage treatment resources across all NPWT patients, allocating more appropriate care. Note that while the cost of therapies is an important factor in the total wound care cost, the dominant driver of cost is duration of the wound.³⁵ Wounds harbouring high bacterial burden are associated with longer wound duration.^{12,36} Therefore, facilitating appropriately triaged care for wounds, even with a short-term cost increase, is likely to yield cost savings in the long-term.

Larger studies to validate dressing change postponement based on fluorescence information are required. Due to the novel nature of this study, we were not comfortable delaying dressing changes by more than 24 hours based on negative fluorescence image information alone. Based on the results of this case series, in future we would consider delaying dressing changes by more than 24 hours. This decision would be made on a case by case basis and would take into consideration that leaving the foam too long can

increase growth of the granulation tissue into the foam and thus increase pain at dressing changes.

This case series also demonstrates how incorporation of bedside bacterial fluorescence can facilitate patient education and increase patient comfort. NPWT can be extremely painful and analgesics are often administered before a dressing change in patients with significant pain.¹³ Patients 1 and 4 in this case series experienced pain so severe that they were refusing much needed dressing changes, even with administration of analgesics. Fluorescence images were acquired at bedside and were immediately used to educate these patients about the bacteria that was present. The simple colours were easy for them to understand. After seeing the images of bacterial fluorescence, both patients agreed to have dressing changes, thereby increasing their adherence to the recommended treatment plan. In cases 2 and 6, images consistently free of bacterial fluorescence led to less frequent dressing changes, thereby sparing these patients unnecessary pain.

Although the ability to visualise bacterial fluorescence within NPWT foam through optically transparent sealant has not previously been described, the presence of bacteria within NPWT foam does not come as a surprise. Microbiological analysis of NPWT foams (68 foams from 17 patients, including both polyurethane and polyvinyl alcohol based foams) revealed high bacterial loads, 10⁶CFU/ml or higher in 69% of foams studied.³³ In the current study, polyurethane foams were exclusively used, and can be seen in both Fig 1 and 2. Interestingly, bacterial loads have been reported to be higher in polyvinyl alcohol foam relative to polyurethane,³³ yet polyurethane foam has been speculated to facilitate better removal of bacteria due to higher blood flow increases.³⁴ Regardless, the high prevalence of bacteria, confirmed via microbiological methods in foams from diverse wound types and locations, suggests that real-time bacterial visualisation in NPWT foam could have widespread use.

Limitations

Several limitations of this imaging device warrant discussion. Visualisation of bacteria in and around a wound does not necessarily mean infection is present, therefore this device does not replace the need for clinician judgement and assessment for infection-related signs and symptoms.²⁶ The device also does not indicate which bacterial species are present nor does it provide bacterial antibiotic sensitivities; microbiological culture is still required if the clinician desires that information. However, bacterial fluorescence can identify an ideal location for the clinician to sample. A prospective clinical trial of 60 patients found that regions of red fluorescence predicted the presence of concerning levels of bacteria in 100% of red-fluorescing wounds.¹⁷

Acquisition of fluorescent and standard images in our windowless clinic rooms generally takes less than 20 seconds, unless the wound is in a challenging position to image. However, the required darkness for capturing

fluorescent images is a challenge in inpatient rooms with large windows. To overcome this challenge, a disposable dark drape attachment was used at every inpatient imaging session (six patients, more than 20 imaging sessions in total). The drape attachment was completely effective in achieving the required darkness. Resulting images had no ambient light-induced artifacts and were simple to interpret (all fluorescent images shown in Fig 1 and 2 were taken with drape attachment). Attachment of the drape before imaging did lengthen the total time spent on imaging, by approximately a minute.

Additional studies on the use of this device in conjunction with NPWT systems are warranted. In this small, 10-patient case series we were able to visualise bacteria through the optically transparent seals used with many NPWT systems and within the NPWT packing foam. However, we did not attempt to determine the depth to which bacteria could be imaged through these seals and foams. Furthermore, NPWT systems available on the market with opaque seals and bandages would presumably hinder penetration of the device's violet illumination, therefore the bandage would need to be removed before imaging. These

10 cases cannot be used to generalise findings, but they do serve to demonstrate the clinical usefulness of handheld fluorescence imaging in wound care, supporting previous studies.^{16–18,20–25} These cases also demonstrate, for the first time, how fluorescence imaging of wounds can be used in conjunction with NPWT to guide treatment decisions including timing of dressing changes, treatment selection, and wound cleaning specifically targeted to regions of bioburden.

Conclusions

Bacterial fluorescence images acquired in this series of 10 patients provided real-time information on bacterial contamination in wounds. Having access to this information facilitated immediate, evidence-based and patient-specific changes in treatment plans. Images prompted and otherwise helped guide timing of dressing changes, guided the extent of wound cleaning, and guided selection of NPWT therapies that were most appropriate for patient needs. Larger studies are required to assess whether fluorescence imaging during NPWT-treated wound assessments can improve healing rates and lower total wound care costs. **JWC**

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Reflective questions

- What is the current triage process when it comes to wound clinicians and negative pressure resources in your facility?
- What information do you currently use to assess wounds undergoing NPWT? How could information on wound and dressing bioburden improve your assessment?
- Fluorescence information on bacterial burden helped to adjust treatment plan in 90% of studied wounds. How do you currently determine whether a wound's bioburden is being treated appropriately?

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