Abstract
Microbiologically based diseases continue to pose serious global health problems. Effective alternative treatments that are not susceptible to resistance are sorely needed, and the killing of photosensitized bacteria through photodynamic therapy (PDT) may ultimately emerge as such an option. In preclinical research and early in vivo studies, PDT has demonstrated the ability to kill an assortment of microorganisms. Antimicrobial PDT has the potential to accelerate wound healing and prevent clinical infection, particularly in patients with chronic leg ulcers. Larger trials are needed to confirm its early promise and suggest its ultimate role in caring for chronic wounds. (JNCCN 2012;10[Suppl 2]:S80–S83)

“A huge number of papers now in the literature show that it is very easy to kill microorganisms in vitro with photodynamic therapy [PDT],” announced Stan Brown, PhD, Director of the Centre for Photobiology and PDT and Professor of Biochemistry at the University of Leeds in the United Kingdom. However, little work has been done in vivo with antimicrobial PDT, but encouraging clinical phase II data are emerging. Although PDT has had perhaps its biggest success in the management of macular degeneration and has obtained a few approved indications for cancer treatment, Dr. Brown offered a glimpse at the rationale behind using PDT in the treatment of wound infections caused by various microbes and its early encouraging results in small numbers of patients with chronic leg ulcers.

Antimicrobial PDT in the Era of Antibiotic Resistance
The problems associated with antibiotic resistance are well-known. Because antimicrobial PDT does not target a specific pathway, it is not associated with the development of resistance. In addition, it readily kills antibiotic-resistant organisms, because it has a totally different mechanism of action, declared Dr. Brown. However, antimicrobial PDT has some disadvantages, he admitted. First, it is a local not systemic technique, so “it only works where you shine the light.” Second, it is a 2-component technique (drug administration and light therapy).

The killing of photosensitized microorganisms has been the subject of investigation for years.1–3 Jori et al determined that PDT may be an effective alternative modality for the treatment of localized microbial infections through the in situ application of a photosensitizer followed by irradiation of the photosensitizer-loaded infected area. Although any photosensitizer would work to kill gram-positive bacteria, Dr. Brown considers phenothiazinium photosensitizers to be an excellent choice for broad-spectrum killing of microorganisms. For gram-negative bacteria, which are more difficult to eradicate, PDT consisting of disrupting agents (eg, polymyxin) plus almost any cationic photosensitizer is worth considering, according to Dr. Brown.

In turning to the use of antimicrobial PDT in vivo, relatively little work has been done, acknowledged Dr. Brown. Thus far, the 3 main areas of research include for oral infections,4 Helicobacter pylori infections,5 and chronic
wounds, although other preclinical applications are being explored (Figure 1). For instance, PDT for dental plaque–related diseases and infections has several advantages: 1) bacteria can be eradicated quickly (within seconds or minutes); 2) the development of resistance in the target bacteria is unlikely; and 3) disruption of the normal microflora can be avoided. Wilson mentioned another possible benefit of using PDT to treat oral infections: through replacing the antimicrobial agents currently being used for these purposes, it might help conserve the dwindling supply of these agents for effective treatment of serious systemic infections. (The use of pyropheophorbide-a [PPA904] in reducing levels of bacteria in chronic leg ulcers is discussed later in more detail.)

Photoactivation of several organisms, including *Escherichia coli*, *Staphylococcus aureus*, methicillin-resistant *S aureus* (MRSA), *Pseudomonas aeruginosa*, and *Candida albicans*, has been demonstrated, stated Dr. Brown. With the use of PPA904-mediated PDT for *S aureus* and MRSA, “you are getting cell kill in a few minutes with very low doses compared with what we are using in cancer,” he explained. “In all the families of bacteria that we looked at, including anaerobic and aerobic, every organism was killed by PDT with PPA904.” In terms of adverse events associated with antimicrobial PDT, “there were no significant safety issues from the toxicology studies,” reported Dr. Brown. No genotoxicity (in the presence or absence of light), no repeat-dose sensitivity, and no systemic exposure occurred after topical application to a wound, he added.

**Rationale for Studying PDT for Chronic Ulcers**

“Chronic ulcers are not a sexy area but nonetheless are a huge problem for many people,” pronounced Dr. Brown. Chronic leg lesions and diabetic foot ulcers may be contaminated, colonized, or clinically infected by bacteria. Moreover, despite the absence of an overt clinical infection, increased bacterial load (> 10^5 colony-forming units [CFUs]/g) correlates with poor healing, he added.

“Infection in diabetic foot ulcers can be very serious, as approximately 15% of infected diabetic foot ulcers lead to amputation and in turn about 15% of these amputations lead to death,” revealed Dr. Brown. Therefore, a clear need exists for a nonantibiotic treatment, perhaps PDT, to reduce the bacterial load and aid in wound healing. Furthermore, the prophylactic use of PDT to prevent infection in surgical wounds before incisions are closed is also under investigation.

The potential mechanisms of PDT-induced wound healing were briefly discussed by Dr. Brown. First, PDT has antimicrobial effects. It is commonly accepted that microbiologic loads greater than 10^5 CFUs/g may inhibit healing. Generally, wound bed preparation for healing may be improved through reduction in bacterial load. Second, PDT may have an effect on the host tissue. Growing evidence seems to suggest that PDT itself may enhance wound healing independent of its antimicrobial abilities. It has been proposed that PDT may stimulate growth factors and an immune response, although these effects have yet to be definitively confirmed.

A clinical trial algorithm for PDT treatment is illustrated in Figure 2. “Basically, we apply the drug in a gel, occlude the wound from light, wait 15 minutes for the bacteria to take up the drug, illuminate with the
super light source, and then do wound swabs,” detailed Dr. Brown.

**Preliminary Phase II Clinical Trial Results of PDT for Leg Ulcers**

In the first human study of PDT in patients with leg ulcers or diabetic foot lesions, 10 subjects took part. According to Dr. Brown, antimicrobial PDT was well tolerated by all of these patients, with no pain experienced nor safety issues noted. The single PDT treatment caused a significant reduction in the bacterial load (up to 2 log) of both gram-positive and gram-negative bacteria immediately after treatment, he added.

A phase IIa, randomized, placebo-controlled trial conducted in 2 centers (one in Manchester, England, and the other in Dundee, Scotland) investigated antimicrobial PPA904-mediated PDT for treating chronic leg ulcers and diabetic foot lesions in 32 patients with chronic wounds. The first part of this study consisted of a single treatment with the gel and light. Half received the experimental PDT treatment and the other half served as the control group. The active group underwent standard bandaging and PPA904 in vehicle plus light, whereas the placebo group had standard bandaging plus vehicle only and light.

In the PPA904 group, bacterial counts were significantly lower immediately after PDT compared with before PDT ($P < .001$), and no significant bacterial reduction was seen in the corresponding placebo group. In the 16 patients with diabetic foot ulcers, 80% had MRSA; after treatment with PPA904, a reduction was seen in the MRSA load. Three months after treatment, 4 of the 8 chronic leg ulcers treated with PPA904 healed, compared with 1 of the 8 ulcers treated with placebo. Although Dr. Brown acknowledged that this is not statistically significant, he said it was “slightly encouraging.”

In turning to the effect of PDT on wound healing, Dr. Brown showed the promising results obtained in one patient 2 months after PPA904-mediated PDT (Figure 3). Again, neither serious treatment-related adverse events nor pain were experienced during treatment.

In the second part of this phase II placebo-controlled clinical trial, repeated PDT treatment in 48 patients with chronic leg ulcers was tested; antimicrobial PDT was used once a week for 12 weeks. Up to 10 sites in the United Kingdom were involved in this study. Dr. Brown showed how the treatment was given (with the PPA Lux 680; Figure 4); “the leg is a curved surface, and so the system is contoured to bend approximately to the shape of the leg,” he explained.

The primary study end point was to determine whether repeat-dose antimicrobial PDT can cause reduction in the bacterial content of chronic leg ulcers, with measurements of total bacterial load of the ulcer taken immediately before and after each treatment. The secondary study end points included noting the levels of specific bacteria before and after each dose, measuring the ulcer area weekly for 12 weeks, and assessing ulcer pain before treatment for 12 weeks. Among the inclusion criteria were wound duration of at least 3 months and not more than 3 years, and total bacterial load of $10^4$ CFUs/g or more determined within 2 weeks of the start of the first treatment. Use of systemic antibiotics was prohibited during the study.

Interim data on 24 patients showed that 4 of 12 ulcers in the PDT arm closed completely, compared with 0 of 12 ulcers in the placebo arm. Dr. Brown found these early results “quite encouraging,” although no significant difference was seen at this
stage. The final results indicate that the primary end point of the trial was met: a statistically significant greater reduction in the wound bacterial load was seen in persons receiving weekly PDT treatment with PPA904 and light compared with weekly placebo and light, he reported. “We looked at different species—anaerobic, aerobic, MRSA, Pseudomonas, Staphylococcus, and Streptococcus—and they were all killed, at slightly different levels,” revealed Dr. Brown. Furthermore, when considered individually, the PDT effect was similar at each weekly treatment.

In terms of wound healing, the PDT arm showed a slight increase in healing against a greater barrier. Although Dr. Brown said that these findings were not as encouraging at the end as at the interim analysis, “with such tiny numbers, we certainly need a study of 200 or 300 patients in order to get meaningful statistics.” The literature indicates that wound healing is most difficult in large wounds that have been present for a long time, and our data support this conclusion, he continued.

PDT treatment with PPA904 has the potential to keep bacterial loads below key clinical thresholds, Dr. Brown surmised. Fifty-five percent of patients treated with PDT had one or more posttreatment time-varying covariant (TVC) measurements greater than $10^5$ CFUs/g, compared with 83% of patients treated with placebo ($P = .0354$), which is statistically significant. In addition, 18% of patients treated with PDT had one or more posttreatment TVC measurements greater than $10^6$ CFUs/g, compared with 63% of patients treated with placebo ($P = .0026$). “In terms of our rationale, [PDT] appears to be a good approach to wound healing. Patients who have a viral load above $10^6$ are heading into the region at risk of clinical infection. This treatment may assist with prevention of infection,” he explained.

**Conclusions**

Phase II clinical trial findings have shown that topical PPA904-mediated PDT for the treatment of chronic ulcers is a safe, well-tolerated option that significantly reduces the bacterial load of the ulcer (immediately after treatment). Furthermore, PDT has the potential to accelerate wound healing and prevent clinical infection, which is a key factor, particularly for patients with diabetic foot ulcers. It also seems to be able to treat clinical infection and kill antibiotic-resistant organisms, without the development of resistance. Based on these encouraging preliminary results with PDT for wound care, further studies with larger numbers of patients are warranted and should help to clarify the true emerging role of antimicrobial PDT.

**References**