Optical coherence tomography: a reliable alternative to invasive histological assessment of acute wound healing in human skin?*

N.S. Greaves,1,2,3 B. Benatar,4 S. Whiteside,2 T. Alonso-Rasgado,4 M. Baguneid2 and A. Bayat1

1Plastic and Reconstructive Surgery Research, Manchester Institute of Biotechnology (MIB), University of Manchester, 131 Princess Road, Manchester M1 7ND, U.K.
2University Hospital of South Manchester NHS Foundation Trust, Wythenshawe Hospital, Southmoor Road, Manchester M23 9LT, U.K.
3School of Materials, University of Manchester, Manchester M13 9PL, U.K.
4Department of Histopathology, Royal Oldham Hospital, Pennine Acute Hospitals NHS Trust, Rochdale Road, Oldham OL1 2JH, U.K.

Summary

Background Gold-standard assessment of acute wound healing has traditionally been through histological analysis of biopsied tissue. However, this process is invasive with recognized side-effects. Optical coherence tomography (OCT) is a noninvasive technique generating high-resolution real-time images of cutaneous architecture.

Objectives To compare OCT with histological assessment of in vivo acute wound healing and ascertain the level of agreement between modalities for measurement of defined cutaneous structures.

Methods Punch biopsies (5 mm) were harvested from 50 healthy volunteers. Wounds healed by secondary intention until they were re-excised 7, 14, 21 or 28 days later depending on random group allocation. Wounds were assessed weekly for 6 weeks using OCT and compared with histological findings derived from time-matched biopsies. Dimensions of four cutaneous structures were measured using both modalities and the level of agreement was established by Bland–Altman analysis. The mean greyscale value (MGV) of the upper reticular dermis was derived from OCT images at all time points.

Results Both techniques showed anatomical congruity in normal and wounded skin with correlating architectural changes associated with inflammatory, proliferative and remodelling wound healing phases. MGV was significantly increased 6 weeks after wounding ($P = 0.001$) and may represent a novel measure of wound fibrosis. Despite good association of histomorphometric values with low but consistent bias (range $-4.18$ to $0.43$ $\mu$m), Bland–Altman plots demonstrated poor agreement between OCT and histology.

Conclusions Optical coherence tomography enabled accurate assessment of healing tissue comparable with histological analysis of biopsy specimens. This noninvasive tool is highly suited to wound assessment and may represent a diagnostic alternative to punch biopsies.

What's already known about this topic?

- Optical coherence tomography is an emerging imaging technology that has proved useful in diagnosis and monitoring of inflammatory dermatological conditions.

What does this study add?

- Characteristic architectural changes that correlate with histological phases of cutaneous wound healing can be identified with OCT.
A wound describes a break in cutaneous epithelial continuity characterized by disruption of structure and function of underlying tissues. After injury, skin integrity must be rapidly restored to prevent infection, limit fluid loss and reinstate homeostatic mechanisms. Acute wound healing requires activation, synchronization and tight control of multiple interrelated overlapping biological phases known as haemostasis, inflammation, cellular proliferation and remodelling. Multiple cell types, bioactive molecules and extracellular matrix (ECM) proteins are involved, resulting in characteristic structural changes in the tissue unique to each stage of healing. The failure of any component can result in delayed wound healing, chronic wound formation or abnormal scarring. Histological analysis of biopsied tissue remains the gold standard for assessment and diagnosis of normal and pathological wound environments. It enables clear visualization of cellular behaviour and structural architecture. However, tissue sections provide only snapshots of the healing process. Furthermore, biopsies cannot be repeated at the same site, may alter the original morphology and create a second iatrogenic wound. They are associated with patient-related complications including pain, bleeding, scarring, infection and delay of the healing process. Consequently, a myriad of noninvasive investigative techniques have been developed to aid assessment of wound healing. These include measurement of wound dimensions, analysis of wound exudate, spectrophotometric intracutaneous analysis, laser Doppler imaging, thermography, fluorescence imaging, polarization imaging, high-resolution ultrasound and confocal microscopy. Such modalities allow serial wound assessment over time but may be confounded by external factors, poor resolution and interobserver variability. Optical coherence tomography (OCT) is an emerging noninvasive assessment tool that overcomes many weaknesses inherent in other equivalent modalities and is free of side-effects. It is a real-time tomographic imaging technique using low-intensity infrared light focused within living tissue. Interferometric detection of reflected light enables high-resolution, two- or three-dimensional, cross-sectional visualization of skin and wound morphology analogous to histology. OCT provides depth-resolved images of tissues up to 2 mm deep with lateral resolution of 1 µm in some devices via a handheld instrument placed in contact with the skin. It can accurately delineate wound re-epithelialization, reformation of the dermoepidermal junction, thickening of newly formed epidermis and dermal remodelling. The presence or absence of these variables is useful in determining the likelihood of successful wound healing and in predicting which wounds may become problematic or chronic in nature.

In this unique clinical study, we used OCT to assess cutaneous morphological changes seen during acute wound healing in 50 healthy human volunteers. These findings were compared with time-matched histological analysis of punch biopsies taken from the same patients. Qualitative and quantitative outcome measures were obtained from both modalities in order to compare the feasibility, accuracy and reproducibility of OCT with the current gold-standard diagnostic technique in the assessment of wound healing.

**Materials and methods**

This cohort study was performed in accordance with the Declaration of Helsinki of 1975, National Research Ethics Service (NRES Committee North West – Greater Manchester West, reference no. 12/NW/0078) and local research and development department (University Hospitals of South Manchester NHS Foundation Trust, reference no. 2011BP001) approval. Fifty healthy volunteers were recruited after assessment against inclusion and exclusion criteria (Table 1). Suitable candidates gave written consent, before being randomized into one of five study groups, each containing 10 patients (Fig. 1). Randomization was conducted in nQuery Advisor 7.0 (Statistical Solutions, Cork, Ireland) using a computer-generated permuted block design with mixed block sizes and random seed. The study was conducted at Wythenshawe Hospital in Manchester. Recruitment ran from April to June 2013 and final follow-up for the final patient was in July 2013.

On day 0, a site on the medial aspect of the nondominant upper arm was allocated midway between the axillary hairline and the medial epicondyle (Fig. 2a). This site was visualized using OCT (Vivosight Topical OCT probe; Michelson Diagnostics, Orpington, Kent, U.K.) (Fig. 2b) before a 5-mm full-thickness punch biopsy was taken under local anaesthetic and aseptic conditions (Fig. 2c). After haemostasis was achieved, wounds were covered with an adhesive dressing (Tegaderm®; 3M, Bracknell, U.K.) for 7 days and left to heal by secondary intention.

Patients were followed up weekly for 6 weeks. At each visit an OCT scan was performed and the wound assessed clinically (Fig. 2d). Group 1 patients underwent a 6-mm full-thickness excision biopsy of healing tissue under local anaesthetic on day 7, group 2 on day 14, group 3 on day 21 and group 4...
Table 1  Study inclusion and exclusion criteria

<table>
<thead>
<tr>
<th>Inclusion criteria</th>
<th>Exclusion criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aged ≥ 16 years</td>
<td>Subjects of either sex aged &lt; 16 years</td>
</tr>
<tr>
<td>Able to understand study requirements and attend all follow-up visits</td>
<td>Any subject who, in the opinion of the investigator, is unable to understand fully the requirements of the trial or consent, or is unable to return for follow-up visits and complete the trial</td>
</tr>
<tr>
<td>Able to provide written consent if competent</td>
<td>Subjects who do not give consent or withdraw their consent to take part in the study</td>
</tr>
<tr>
<td>Weight between 40 and 150 kg with a BMI of 20–45 kg m⁻²</td>
<td>Subjects who have a history of keloid or hypertrophic scarring</td>
</tr>
<tr>
<td></td>
<td>Subjects who take medication known to alter/influence the healing of skin</td>
</tr>
<tr>
<td></td>
<td>Subjects who are receiving formal oral anticoagulant therapy (warfarin)</td>
</tr>
<tr>
<td></td>
<td>Subjects who have taken part in clinical studies or received any investigational drugs 2 months prior to day 0</td>
</tr>
<tr>
<td></td>
<td>Subjects who have evidence of drug abuse</td>
</tr>
<tr>
<td></td>
<td>Subjects who have had or are known to have hepatitis B or hepatitis C infection including carriers of hepatitis B surface antigen, hepatitis B core antibodies or hepatitis C antibodies. Previous vaccination against hepatitis B or C is not excluded</td>
</tr>
<tr>
<td></td>
<td>Subjects who have previously had a positive result to the HIV antibody test, or admit to belonging to a high-risk group</td>
</tr>
<tr>
<td></td>
<td>Subjects who become systemically unwell during the research process due to causes external to the study</td>
</tr>
<tr>
<td></td>
<td>Subjects with known allergies to antibiotics</td>
</tr>
<tr>
<td></td>
<td>Pregnant subjects or those likely to become pregnant in the next 3 months</td>
</tr>
<tr>
<td></td>
<td>Subjects with active skin disorders considered by the investigators to affect wound healing adversely</td>
</tr>
</tbody>
</table>

BMI, body mass index.

Fig 1. Cohort study flowchart. A total of 74 volunteers were screened; two subjects did not fulfil recruitment criteria and were excluded and a further 22 declined to take part in the study. Fifty participants were enrolled and randomly allocated to one of five groups each containing 10 patients. All groups had a punch biopsy harvested on day 0. Group number determined the time point for excision biopsies of healing tissue: group 1 at day 7, group 2 at day 14, group 3 at day 21, group 4 at day 28; group 5 did not have repeat biopsies. Complete optical coherence tomography (OCT) and histological assessment for each time point were obtained with no loss to follow-up.
on day 28 (Fig. 2e). Group 5 patients did not have a second excision biopsy. After excision biopsies, wounds healed by secondary intention (Fig. 2f). Biopsies were placed in formalin and later stained with haematoxylin and eosin (HE) enabling histological assessment by a blinded consultant histopathologist.

Qualitative comparison of time-matched OCT and HE samples reporting structural and morphological features were made. Histomorphometric measurements were performed using ImageJ (http://rsbweb.nih.gov/ij/) and Nanozoomer version 2.2.8 software (Hamamatsu Photonics UK Ltd, Welwyn Garden City, U.K.) for OCT and histology, respectively. Repeated measures of epidermal thickness (pre- and post-biopsy), percentage re-epithelialization, papillary dermal thickness and epidermal bulge thickness were taken and averaged. OCT and histological measurements were made by separate clinicians blinded to each other’s results.

The mean greyscale value (MGV) could be obtained only using OCT. Measurements were made using ImageJ from a standardized area of upper reticular dermis at all time points. MGV is a measure of tissue light-scattering properties, which depend on the ratio of refractive indices for scattering centres such as protein aggregates and cellular components compared with interstitial fluid.\(^{17}\) The MGV alters as wound healing progresses secondary to the influx of cellular content as well as deposition and remodelling of ECM proteins.

Vivosight OCT images 60 separate planes within a 6 × 6-mm square of chosen tissue. These are joined to provide a continuous sequence that can be analysed by clinicians using ImageJ software. The lateral and axial resolutions are 7.5 μm and 5.5 μm, respectively.

**Statistics**

Complete OCT and histological data for cutaneous architectural measurements at different time points were provided for 50 patients split into five groups of 10 subjects. This sample size adequately powered a Bland–Altman analysis assessing level of agreement between OCT and histology for measurement of defined structures. Analyses used a conventional two-sided 5% significance level. All summaries, graphs and analyses were produced using SPSS version 20 (IBM, Portsmouth, U.K.).

Pairwise analysis of histological and OCT measurement values assessed the agreement between different modalities using the Bland–Altman method of agreement.\(^{18}\) Outcomes were expressed as mean bias (mean difference of histology – OCT) and 95% limits of agreement. Mean bias represents consistent over- or underestimation of values, and 95% limits of agreement give the extent of discrepancies in terms of predicted extreme limits for discrepancies between modalities. With \(n = 50\), 95% limits of agreement can be estimated with an accuracy of ± 0.5 × SD of simple paired differences.

Data for subjects with values at time points for both histology and OCT were included in analyses, combining all five groups. For percentage re-epithelialization only group 1 data were presented, as other groups had mainly values of 100%. Changes in the MGV from day 0 were analysed using pairwise t-tests, combining groups where appropriate.
Role of OCT in acute wound healing assessment, N.S. Greaves

Results

A total of 74 volunteers were screened for participation in the study; 72 met the suitability criteria but 22 declined to take part. Of the 50 participants, 24 were male and 26 were female. The mean age was 26 years and 92% of participants were white. OCT assessment was performed at all time points for all patients. Day 0 and time-matched tissue biopsies were completed for all patients as determined by study group allocation. No patients were lost to follow-up. There were no adverse events during the study.

Comparative measurement of epidermal thickness on day 0 revealed a significant consistent mean bias of −4.18 μm with 95% limits of agreement ranging from −16.09 μm (25.9% underestimation) to 7.72 μm (12.4% overestimation) (Table 2; Fig. 3a). The mean bias for day 0 papillary dermal thickness was −2.44 μm with 95% limits of agreement from −18.51 μm (23.6% underestimation) to 13.62 μm (17.4% overestimation) (Table 2; Fig. 3b). Bias relating to epidermal thickness on the day of excision biopsies was 0.43 μm; 95% limits of agreement were −18.09 μm (16.8% underestimation) to 18.95 μm (17.6% overestimation) (Table 2; Fig. 3c). The mean bias for epidermal bulge thickness on the day of excision biopsies was 1.10 μm; 95% limits of agreement were −22.61 μm (11.0% underestimation) to 24.81 μm (12.0% overestimation) (Table 2; Fig. 3d). Percentage re-epithelialization (group 1 only) showed a mean bias of 1.07 μm; 95% limits of agreement were −7.23 μm (13.7% underestimation) to 9.38 μm (17.7% overestimation) (Table 2; Fig. 3e).

The MGV on OCT was significantly reduced for 2 weeks after wounding (P < 0.001). Values subsequently increased until they were equivalent to baseline 4 weeks after injury. The MGV at 6 weeks was significantly greater than on day 0 (P = 0.001) (Fig. 4).

OCT clearly demonstrated cutaneous anatomy with delineation of the epidermis, papillary dermis and reticular dermis. The dermoeipidermal junction was noted, as well as dermal appendages including blood vessels, hairs and sebaceous glands (Fig. 5). Details such as rete ridges could be identified in some cases but individual cells could not be resolved. Optical coherence tomography was able to image to a depth of ~1.5 mm.

Table 2 Assessment of congruity between optical coherence tomography (OCT) and histological measurements of cutaneous structural parameters using the Bland–Altman method of agreement expressed as mean bias and 95% limits of agreement for mean bias

<table>
<thead>
<tr>
<th>Structural Parameter</th>
<th>Histology, mean (SD)</th>
<th>OCT, mean (SD)</th>
<th>Mean bias (histology – OCT)</th>
<th>95% limits of agreement</th>
</tr>
</thead>
<tbody>
<tr>
<td>Epidermal thickness (μm), day 0 (n = 50)</td>
<td>60.00 (8.78)</td>
<td>64.18 (6.76)</td>
<td>−4.18</td>
<td>−16.09-7.72</td>
</tr>
<tr>
<td>Epidermal thickness (μm), excision biopsy reading (n = 30)</td>
<td>108.14 (27.47)</td>
<td>107.71 (24.56)</td>
<td>0.43</td>
<td>−18.09, 18.95</td>
</tr>
<tr>
<td>Papillary dermal thickness (μm), day 0 (n = 50)</td>
<td>77.14 (15.29)</td>
<td>79.59 (12.42)</td>
<td>−2.44</td>
<td>−18.51, 13.62</td>
</tr>
<tr>
<td>Papillary dermal thickness (μm), excision biopsy reading (n = 40)</td>
<td>206.54 (47.23)</td>
<td>205.44 (44.77)</td>
<td>1.07</td>
<td>−22.61, 24.81</td>
</tr>
<tr>
<td>Re-epithelialization (%), excision biopsy reading (n = 30)</td>
<td>53.41 (16.59)</td>
<td>52.33 (16.74)</td>
<td>1.07</td>
<td>−7.23, 9.38</td>
</tr>
</tbody>
</table>

Fig 3. Bland–Altman plots for epidermal thickness on day 0 (a), papillary dermal thickness on day 0 (b), epidermal thickness on day of excision (c), epidermal bulge thickness (d) and percentage re-epithelialization (e), illustrating considerable scatter around the mean difference. Despite low bias there was lack of agreement between optical coherence tomography (OCT) and histology, meaning measurements cannot be used interchangeably for these structural parameters.

Both modalities enabled accurate assessment of cutaneous morphology throughout healing. Characteristic architectural features on OCT were noted that correlated with recognized phases of wound healing seen histologically.

**Inflammation**

Subcutaneous fat was seen as a honeycomb structure covered by areas of superficial dense opacification in keeping with inflammatory infiltrate. There was minimal evidence of dermal regeneration after 1 week. Blood vessels adjacent to the wounded area were dilated and there was a thickening of the peripheral epidermis with formation of a migratory epithelial tongue and epidermal bulge (Fig. 6).

**Proliferation**

Images from 2 weeks onwards showed evidence of fibroplasia and angiogenesis with deposition of an increasingly thick ECM forming early dermis (Fig. 7) and granulation tissue (Fig. 8). Differentiation between papillary and reticular dermis was lost. Multiple small vessels were noted. ECM fibres were not orientated in any particular direction. Re-epithelialization was complete 2–3 weeks after injury but was covered by a thick haemostatic crust (Fig. 7).

**Remodelling**

Increasingly thick collagenous ECM was deposited (Fig. 9a). On OCT, this took on a ‘swirled appearance’ initially (Fig. 9b), which was later remodelled to form orientated dense fibrotic tissue after 6 weeks (Fig. 9c). This change was not seen histologically. The protective crust was dislodged to expose a thickened, flattened epidermis while the vessel count was reduced in the scarred tissue.

**Discussion**

OCT has become an established method of clinical and research-based cutaneous assessment in the past decade. Studies have evaluated the effect of age, sex, skin type, anatomical location and physiological conditions on cutaneous morphology using OCT. Furthermore, characteristic changes associated with inflammatory and neoplastic lesions including actinic keratosis, basal cell carcinoma, contact dermatitis and psoriasis have been shown to correlate well with equivalent histological analysis. OCT has also been used to quantify treatment effects of interventions ranging from topical pharmaceutical agents to CO2 laser therapy. Studies utilizing OCT to investigate wound healing are limited. Singer et al. and Cobb et al. used OCT to measure re-epithelialization and monitor structural changes after wounding in porcine and murine models, respectively, while Wang et al. observed healing responses after implantation of collagen dermal scaffolds in full-thickness murine wounds. In one of the few human studies, Kuck et al. serially examined leg ulcers in six patients over 2 weeks. All concluded that OCT was a viable tool for monitoring wound healing noninvasively. However, they were limited by small sample numbers, poor study design and use of animal models limiting translation to human subjects.

In this unique human clinical study, we have demonstrated that OCT can be used to identify normal cutaneous structures and architectural changes associated with each phase of acute wound healing in 50 healthy volunteers. These temporal alterations corresponded to time-matched histological findings. Establishment and progression of re-epithelialization, epidermal bulge development, granulation tissue formation, fibro-
plasia and dermal regeneration were clearly identified via both techniques.

Significant re-epithelialization was seen in the week after injury and the majority of patients had complete epithelial cover after 14 days. After 6 weeks the mature healed epithelium was thickened and homogeneous. Interestingly, both modalities demonstrated minimal evidence of dermal regeneration in the first week after injury. This delay may be secondary to recruitment and migration of fibroblasts from adjacent tissue as well as conversion of circulating fibrocytes to fibroblasts. From 2 weeks after injury, OCT and histology show progressive tissue fibroplasia, remodelling and fibrosis.
Fig 8. Comparison of blood vessel density and diameter in normal skin (a) and granulation tissue (b). Normal skin has multiple small vessels. Granulation tissue is characterized by dilated vessels at the wound periphery and numerous small vessels within the wound bed.

Fig 9. Time-matched images of the proliferative phase of wound healing seen (a) histologically and (b) on optical coherence tomography for the same patient. ‘Swirling’ of the newly deposited extracellular matrix is noted from 3 weeks after injury. This gives way to more ordered dense fibrosis with a reduced vessel count after 6 weeks (c). The epidermis is thickened and lacks defining features such as rete ridges.
However, only OCT demonstrates a ‘swirled’ dermal pattern formed by disordered ECM proteins interspersed with granulation tissue vasculature. This is soon replaced by more ordered collagen fibres at 6 weeks and may represent conversion of type III collagen to type I collagen during wound maturation. MGV was a useful correlating noninvasive measure of fibroplasia and remodelling. Significantly reduced reflectivity at day 7 related to loss of dermal tissue secondary to injury and reflected the lower MGV of subcutaneous fat due to its high water content. Subsequently increased values correlated with fibroblast recruitment to the wound bed seen histologically, resulting in fibroplasia and dermal regeneration eventually ending in tissue fibrosis with formation of mature scar tissue. The statistically significant change in MGV after 6 weeks compared with baseline may represent a novel quantitative measure of tissue fibrosis or scarring density. However, further studies and correlation with collagen-specific immunohistochemistry are required to substantiate this observation.

Comparative histomorphometric studies examining human skin are few in number and limited to measurement of one parameter in small numbers of patients. In contrast, this study assessed four different structures within 50 healing tissue samples. Gambichler et al. compared OCT with histological measurement of epidermal thickness in 16 healthy individuals. They found a mean bias of −3.44 μm, which compares favourably with the day 0 epidermal thickness bias of −4.181 μm described in this study. The mean bias of all other histomorphometric parameters within our study was small (range −2.442 to 0.431 μm). However, Bland–Altman plots consistently demonstrated considerable scatter around the mean difference, and 95% limits of agreement showed unsatisfactory numerical agreement between modalities, suggesting the two cannot be used interchangeably for the parameters measured. Nevertheless, this does not mean OCT is unsuitable for wound assessment. It is unlikely that different investigative methods will give identical results for all individuals, but it is important to recognize that difference limits clinical interpretation. Clearly there is a real but small discrepancy between OCT and histological measurements. However, the modalities correlate strongly despite generating different numerical values. Trends between time points are clear via either modality, while mean bias values are minimal, representing a small percentage of the total measurement. Furthermore, the role of OCT in wound assessment is primarily an observation of changes in cutaneous architecture, not a precise measurement of specific parameters. The consistent bias highlighted does not limit effective wound assessment and therefore, in this clinical context, OCT could be used as an alternative to punch biopsies.

OCT offers advantages to both clinicians and patients (Table 3). Primarily, it is noninvasive and side-effect free. Consequently, it can serially assess a lesion or wound over time without interfering in the disease or healing process. Scans are fast, taking less than 1 min, and images are ready for analysis within seconds enabling rapid diagnosis. OCT has been shown to be precise in terms of repeatability and reproducibility with low variation coefficients and superior resolution compared with other noninvasive modalities, including high-frequency ultrasound. The scanning technique is simple and reproducible. All images can be saved to and accessed from a database. OCT may be utilized in diagnosis and monitoring of dermatological disease, evaluation of response to treatment and interventions, and wound and scar assessment. Furthermore, this safe modality is suitable for use in outpatient settings where scanning can guide further investigations and interventions. Evidence for OCT in the diagnosis of basal cell carcinoma, actinic keratosis, psoriasis and contact dermatitis among other conditions is growing.

Table 3 Advantages and disadvantages of optical coherence tomography (OCT) as an investigative modality in dermatology

<table>
<thead>
<tr>
<th>Advantages</th>
<th>Disadvantages</th>
</tr>
</thead>
<tbody>
<tr>
<td>Noninvasive</td>
<td>Primarily a research tool currently</td>
</tr>
<tr>
<td>Side-effect free</td>
<td>Cost of OCT unit and maintenance</td>
</tr>
<tr>
<td>Enables serial imaging of the same lesion/wound over time</td>
<td>Assessment is operator dependent</td>
</tr>
<tr>
<td>Quick to perform (&lt; 1 min)</td>
<td>Interobserver variability</td>
</tr>
<tr>
<td>Huge numbers of scans can be stored and readily accessed</td>
<td>Operator learning curve</td>
</tr>
<tr>
<td>Images are ready for assessment within seconds of completing the scan, enabling rapid diagnosis</td>
<td>Limited depth of tissue penetration</td>
</tr>
<tr>
<td>Higher resolution than other noninvasive modalities</td>
<td>Lower resolution than histology – cannot identify individual cells</td>
</tr>
<tr>
<td>Suitable for assessment of tissue morphology with evidence for diagnostic use in wound healing, basal cell carcinoma, contact dermatitis and psoriasis</td>
<td>Unsuitable for highly accurate measurement of cutaneous parameters as Bland–Altman analysis has demonstrated poor agreement with histologically derived values</td>
</tr>
<tr>
<td>Cost savings through reduced numbers of biopsies requiring processing, sectioning, staining and pathologist assessment</td>
<td>Dependent on patient cooperation as any movement reduces quality of scan</td>
</tr>
<tr>
<td>Potential roles in:</td>
<td></td>
</tr>
<tr>
<td>Acute and chronic wound assessment</td>
<td></td>
</tr>
<tr>
<td>Diagnosis and monitoring of disease</td>
<td></td>
</tr>
<tr>
<td>Response to treatment or intervention</td>
<td></td>
</tr>
<tr>
<td>Scar assessment</td>
<td></td>
</tr>
</tbody>
</table>
Limitations of OCT in clinical practice are secondary to the depth of tissue penetration (Table 3). The maximum scanning depth is 2 mm but, in practice, resolution over 1.25 mm from the surface is poor. This is compounded by a haemostatic crust or significant epidermal thickening, which may produce shadowing and further reduced resolution. This may limit the diagnostic range of OCT to purely epidermal, papillary dermal and upper reticular dermal pathologies. Furthermore, although lateral resolution is good at 7.5 μm, OCT cannot identify individual cells, unlike histology. Consequently, interpretation is based upon tissue morphology as a surrogate marker of cellular activity. As we have demonstrated, OCT is unsuitable for highly accurate measurement of cutaneous structures where histology remains the gold standard. OCT is an imaging modality with an inherent learning curve for new operators. Experienced clinicians are currently limited in number and a recognized educational programme is not available as OCT is still primarily a research tool. Interpretation of scans remains operator and experience dependent. Initial purchase and maintenance of the OCT unit can be costly. Scans are dependent upon patient cooperation as movement results in low-quality images. As a result, certain patient cohorts may be unsuitable, including those with dementia, movement disorders and psychiatric conditions.

In conclusion, our study is the first to investigate acute wound healing in a large number of human subjects using OCT. We have demonstrated that OCT enables accurate assessment of healing tissue, comparable with histological analysis of biopsy specimens. Morphological changes in cutaneous architecture associated with the inflammatory, proliferative and remodelling phases of wound healing are clearly observed. Measurement of architectural parameters showed good association between OCT and histology with low and consistent bias. However, Bland–Altman analysis found poor agreement, and values obtained were not interchangeable between modalities. OCT represents a novel noninvasive method of wound assessment that could reduce the need for tissue biopsies secondary to its speed, diagnostic capabilities and side-effect-free profile. However, further randomized controlled studies are required to prove the efficacy and accuracy of OCT before it becomes a routinely used diagnostic tool in dermatological practice.

Acknowledgments

The authors gratefully acknowledge the support and help of Paul Cormack and Hamamatsu Photonics U.K. Ltd, who facilitated digitalization of histological sections and Nanozoomer analysis software support. We are also grateful to Adam Mee- kins of Michelson Diagnostics, who provided guidance on the Vivosight scanner and analysis of outcome measures. Finally, we would like to thank Helen Carruthers (UHSM) and Julie Morris (University of Manchester) for graphical and statistical support of this study, respectively.

References