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New Developments in Smart Bandage Technologies for Wound Diagnostics

Anna McLister¹, Jolene Phair¹, Jill Cundell², James Davis^{1}*

¹School of Engineering, ²School of Health Sciences, University of Ulster, Jordanstown, Northern Ireland, BT37 0QB, UK

E-mail: james.davis@ulster.ac.uk

Prof J. Davis

School of Engineering, University of Ulster, Jordanstown, Northern Ireland, BT37 0QB, UK

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Abstract: The pH of wound fluid has long been recognised as an important diagnostic for assessing wound condition but as yet there are few technological options available to the clinician. The availability of sensors that could measure wound pH, either in the clinic or in home could significantly improve clinical outcome – particularly in the early identification of complications such as infection. This review identifies new material designs and electrochemical research strategies that are being targeted at wound diagnostics and provides a critical overview of emerging research that could be pivotal in setting the direction for future devices.

1. Introduction

Small cuts and scratches are part and parcel of everyday life and most people, providing the trauma to the skin is relatively minor, will cede responsibility for repair of the wound to the myriad of biochemical processes that govern wound healing. In effect, beyond the simple cleansing of the wound, the body will be expected to act to re-establish the integrity of the skin barrier on its own accord with only the briefest oversight from the patient. This is not however always the case and the healing processes that would normally regulate tissue regeneration can become slowed or stalled^[1-5]. The latter can arise as a consequence of

1 numerous factors such as infection, compromised nutritional status and poor circulatory
2 supply. The development of chronic wounds has long been a major concern for healthcare
3 providers and especially so for those involved in the management of diabetic patients where
4 ulceration is a common and increasingly problematic complication^[1-3]. While advances in
5 sensing technologies have revolutionised the daily management of diabetes through enabling
6 patients to monitor their blood glucose levels, there is an emerging opportunity for these same
7 methods to provide valuable diagnostic insights into the healing process and therein proffer
8 the possibility of markedly aiding the treatment of chronic wounds.
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Chronic wounds are widely regarded as a silent epidemic that affects a significant proportion of the populace and pose a major and, indeed, an ever increasing threat to public health^[1] and while, in recent years, there has been a significant increase in the number and variety of wound dressings^[4], the management of the condition continues to be problematic. In chronic wounds, typified by diabetic foot, venous leg or pressure ulcers, the healing processes are stalled resulting in a wound that can persist for months if not years leaving the patient susceptible to further complications and life threatening events^[1-3]. It is estimated that some 6.5 million people in the US suffer from a chronic wound of one sort or another giving rise to an annual healthcare bill of some \$25 billion ^[6]. Admittedly, the US health system is far from typical, but figures of a similar magnitude can be found in most developed countries. In the UK, some 650,000 patients suffer from some form of chronic wound with published estimates suggesting that the combined cost to the NHS for their treatment reaches approximately £3 billion per year^[1]. Only 5% of the latter is attributed to materials cost with the vast majority – estimated to be around 80% - being staff-related. In a recent audit almost 42% of leg/foot ulcers had not healed in the previous six months and 28% had remained unhealed for a year or longer which, in all cases, requires continuous outpatient management, hospital consultation and treatment^[1]. It must also be noted that the loss of productivity for afflicted individuals and the families that care for them and their diminished quality of life are

1 simply immeasurable and will far outweigh the fiscal concerns of the state. Sadly, the burden
2 of treatment is expected to increase dramatically in the future with ever-rising healthcare costs,
3 an aging population and dramatic increases in diabetes and obesity (critical comorbidities).
4 Similarly, burn injuries are another significant cause for concern for clinicians involved in
5 complex wound care (and NHS budget holders) with over a quarter of a million cases
6 presenting to primary care teams and a further 175,000 passing directly to A&E departments
7 annually^[7].
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10 The important point to note when considering these statistics is that, in the vast
11 majority of cases, the management and treatment of the wounds are largely conducted within
12 the community. There is a dearth of diagnostic options available for the routine, point of care
13 assessment of wound condition with the patient and/or healthcare professional relying on the
14 recognition of subtle local indicators or non-specific general signs (such as loss of appetite,
15 malaise, or deterioration of glycaemic control in diabetic patients). Given the great variation
16 in wound type, ambiguity is an ever-present hazard. The longer a wound takes to heal
17 (irrespective of type/origin), the greater the propensity for complications to arise and, where
18 there is ambiguity or a lack of vigilance in assessing wound condition, delays in seeking
19 medical attention can all too often lead to an irreparable deterioration in the wound condition
20 and further compromise the health of the patient leading to hospitalisation with either limb or
21 life-threatening consequences.
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24 As the majority of chronic wounds are treated within the community, the ultimate aim
25 is for the development of intelligent, decentralised, wound care technologies that can monitor
26 the condition of the wound - reporting directly to the patient and/or healthcare practitioner and,
27 where appropriate, able to act autonomously to facilitate the healing processes or minimise
28 complications such as inflammation or infection. It is easy to envisage therefore that the
29 development of new technologies that can reduce treatment times and minimise complications
30 will have major impact and deliver substantial cost benefits and vastly improve patient
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outcome. One of the key findings in the recent World Union of Wound Healing Societies' report: 'Diagnostics and Wounds: A Consensus Document' was that "diagnostic tools need to be moved into the clinic or the patient's home to ensure optimal care is provided for patients with wounds"^[8]. At present, there are no available technologies to address this recommendation.

In this report, a spotlight is trained on the core issues of chronic and complex wounds which, can be considered to centre on four interdependent themes. These are based on monitoring healing, understanding the dynamics of healing and the material interactions, treatment interventions and the electronics for facilitating the intelligence needed for establishing control over the other three. An indication of the convergence of specialisms that are necessary for the development of a smart dressing are indicated in Figure 1. The difference between a bandage and dressing should be noted as the latter is usually in contact with the wound while the former is responsible for covering and fixing the position of the dressing. The advent of new approaches to monitoring wound status condition can lead to a blurring of these definitions as the integration of functional dressing materials with communication technologies embedded with the bandage requires a much more holistic design strategy.

An extensive range of materials that have been developed for wound dressings in recent years and there has been a gradual evolution from the purely passive, biocompatible absorbant polymers to more functional materials that aim to actively encourage the healing processes and minimise the risk of infection. These tend to incorporate components such as metalloproteinase inhibitors and/or antimicrobial agents such as iodine or silver^[4]. The various strategies available have been critically reviewed by Moura et al, (2013) and it is clear that, despite considerable advances in clinical efficacy, healthcare staff remain effectively blind to the progress of the healing processes until the bandage is removed ^[4,7,8]. A much

1 better solution would be to have a dressing that can proactively monitor the wound and
2 provide an insight into the wound dynamics. The latter is a considerable challenge in terms of
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4 designing smart materials that can function as the interface between the wound and the
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6 electronics and although it is a highly active area, it is one fraught with problems:
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8 biocompatibility, selectivity, sensitivity and sensor lifespan being only a few. The aims of the
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10 present report are to provide a critical insight in to the material approaches being taken at
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12 present in the design of smart dressings and to appraise their translation to the clinic.
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18 2. Wound Healing

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20 Wound healing can be divided into four overlapping phases: coagulation,
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22 inflammation, migration-proliferation and remodelling^[5,9]. Coagulation is needed for wound
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24 protection and hemostasis and occurs in the very early stages of an injury whereby
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26 inflammatory cells are sent to the site of the wound and a protective fibrin plug formed. As
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28 the inflammatory phase subsides, wound contraction is initiated. Matrix proteins provide
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30 substrates for cell movement, which facilitates the change in cell behaviour and structures that
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32 eventually returns the integrity of the tissue^[5,9]. In contrast, chronic wounds seldom progress
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34 beyond the inflammatory stage and can be defined more specifically as “a wound that has not
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36 shown a 20%-40% reduction in area after 2-4 weeks.”^[2,9]. The feedback mechanisms that
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38 would normally end the Inflammatory Stage are short-circuited and are the main reasons for
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40 impaired wound healing^[9]. Although Infection is the primary concern in the management of
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42 chronic wounds, there is a tendency for individuals to seek treatment only once gross
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44 symptoms appear (yellow exudate and red inflammation) by which time bacterial colonisation
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46 will have progressed to the point where more substantial intervention is required. Early
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48 identification of infection is imperative in minimising the need for hospitalisation and
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50 preventing limb threatening events^[10] and it is here that electrochemical sensing systems
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52 could have a major impact on clinical practice.
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3.0 Wound Fluid

The fluid bathing the wound originates from a variety of sources and comprises a highly heterogeneous mixture and its composition is assumed to reflect the clinical condition of the wound at the time of sampling ^[11,12]. The composition of the fluid will vary considerably over the course of the healing process as a consequence of changes in the microenvironment as cell migration and tissue remodeling progresses and knowing what stage the wound is presently at is one of the driving forces behind the development of wound monitoring technologies. Underlying conditions (i.e. diabetes, neuropathy) can also influence the nature of the fluid. Exogenous factors such as bacterial load, treatments applied (either systemic or topical) and the nature of the wound dressing will also contribute to and affect the milieu and again, the ability to assess the changes in the wound environment through simple diagnostics test could revolutionise treatment.

In chronic ulcers, wound fluid is normally defined as an exudate with a high viscosity and protein content that exceeds 30 mg/mL^[11]. Temporal issues are critical when considering the fluid under examination as it has been found by Zillmer et al. and colleagues that immediately following surgery, there can be significant contamination from material being transported directly from the blood stream – accounting for some 30% of the proteinaceous material found with the wound fluid^[13]. It is important to consider the etiologies involved in the samples under consideration (ie comparing acute to chronic wounds) whereby the concentrations of target biomarkers in one particular sample may not correlate to the actual bioactivity in another.

4.0 Potential Biomarkers

Broadbent and co-workers have detailed over 150 proteins and small molecule metabolites whose concentrations are influenced by the cellular/tissue remodeling and

associated inflammatory responses^[14]. The up regulation of proteases and growth factor dysregulation have been identified as offering considerable therapeutic and prognostic value. At present, there is a tremendous effort to elucidate the significance of each of these players and to identify those most likely to be diagnostically useful in the clinical management of chronic wounds. Monitoring markers that can provide key warnings towards the complications, particularly infection, is the critical challenge that faces the development of the next generation of smart dressings. Clinical investigations of wound fluids, exudates and tissues have provided considerable insights and there have been a number of attempts to classify the main protagonists involved in the healing progression of the wound. As mentioned previously, the ability to control the activation of such species has been shown to aid the healing process but such interventions clearly require the provision of analytical tools through which their concentration/activity can be speedily and easily measured at the time of consultation^[8].

Despite an extensive candidate list, the diagnostic community has tended to focus on a more limited group and an overview of the chemical components has been presented by Harding et al^[8]. It must be noted however that although significant strides have been made in terms of characterising the biochemical fluxes that can occur within a wound chronic wound environment there is, as yet, no set of definitive markers.

5.0 Electrochemical Solutions

While there are a large number of systems that could ultimately be applied to wound monitoring^[15], the present report has focused predominantly on electrochemical approaches. The latter include: pH^[16-20], bacterial metabolites^[21-23], endogenous wound biomarkers^[24], volatile organic emissions^[25,26] and temperature^[27]. Many of these have been investigated with the intention of implementation within a clinic setting, but acquiring sufficient selectivity with minimal sample preparation is a severe challenge, especially when considering the

limited time available during a patient consultation. At present, considerable attention is being paid to the measurement of wound pH^[15-20]. The pH of the exudate within a wound site is known to vary according to the stage of healing and an idealized comparison of the pH profiles of acute and chronic wounds is highlighted in Figure 2. It is no surprise therefore that pH, will play a pivotal role in the biochemical reactions taking place and thus being able to monitor it could give the clinician valuable insights into the present state of the wound^[28]. While the natural pH of the skin will vary from person to person, it typically fluctuates within a narrow acidic range between pH 4 and pH 6 that impedes bacterial proliferation. Whenever an injury creates a cutaneous wound, it exposes the underlying tissue which, as it is normally regulated at pH 7.4, can encourage bacterial growth and thereby promote infection. In order to counter this, a temporary acidosis occurs during the initial healing stages^[28]. The pH of the chronic wound, however, oscillates in a weakly alkaline range and increases the susceptibility of the patient to bacterial incursions. Upon infection, bacteria seek to increase the wound pH to create an environment which is more accommodating to their growth^[28,29]. It has been proposed that this increase in pH could be a diagnostic handle through which to identify the onset of infection^[30] and there is an increasing effort to develop disposable pH sensors that could be applied to wound monitoring.

It is clear that the existing pH probe technology is far from suitable for use directly in assessing the pH of wound fluid but there have been considerable developments in recent years with a host of new approaches coming to the fore, largely in response to the limitations of the traditional glass potentiometric systems^[15,28,31]. It must be noted that not all are designed specifically for biomedical contexts, nor indeed are they all appropriate. The implementation of sensor systems within the latter gives rise to numerous concerns where issues over probe size and disposability can be problematic—especially where sample sizes may be limited or in vivo application is desired. A number of research avenues have been explored to counter these issues and both potentiometric^[31-44] and voltammetric^[16-20, 45-51]

methodologies have been pursued. In most cases, a functional material containing pH sensitive components is applied to the surface of an appropriate sensor substrate through a variety of methods that include: Adsorption/monolayer^[39,48], polymer films^[36,53], screen printed inks^[35,38,40,47], covalent attachment^[46,47] or electrodeposition^[32,33,37,42–44,50].

Screen printed electrochemical sensors have been designed specifically for wound pH monitoring applications and shown to be a versatile addition^[16]. Rather than relying on potentiometric detection methodologies, they have exploited voltammetric scanning (typically using squarewave voltammetry) in which an endogenous biomarker such as uric acid (which is ubiquitous within most fluids) is oxidised^[16–20]. The latter process is pH dependent and thus the position of the oxidation peak can be used as indirect measure of pH. An example of the signal output is highlighted in Figure 3. The single shot disposability proffered by standalone screen printed systems does not however adequately meet the objectives of the Harding Consensus document^[8] as it still requires the use of the device by an experienced healthcare provider. It could be argued that although it could be exploited much in the same way as glucose meters, in reality patient compliance will still be a major issue. A much better approach would be to have a system that could periodically measure and autonomously report back – either to the patient or to the clinician. This would require integrating the device within the dressing itself as previously suggested. One innovative approach has been to examine the use of woven carbon fibre as a sensing substrate^[17]. The carbon fibre weave is sufficiently flexible to follow the contours of wounds that can present a highly variable surface morphology and can be manufactured in the volumes necessary to enable the mass production of dressing at relatively low cost. Carbon is already used in wound dressing and thus its biocompatibility could, to a large extent, be assumed to have been proven. The main challenge however relates to the activation of the fibres. The measurement methodology is largely the same as for the screen printed systems^[16] in that urate is the target biomarker but the basal plane nature of the fibres was found to be a considerable impediment to the

acquisition of a measurable voltammetric signal ^[17]. Atmospheric pressure plasma (APP) treatment of the weave however has been investigated as a means of exfoliating the fibres with the aim of improving the electron transfer kinetics – a prerequisite for analytical applicability – and shown to significantly improve performance with a near nernstian response obtained^[17].

The brittle nature of individual carbon fibres however present some additional issues in that although flexible, fracturing and snapping can arise and fragmentation particles could contaminate the wound and induce irritation leading to a prolonging of inflammation within the wound site. The carbon fragments could also be embedded within the wound as tissue remodeling commences leading to a need to debride the wound and further impeding the healing processes. An alternative approach to counter this issue has been investigated through the use of carbon composite polymers based on polyethylene or polycarbonate doped with carbon particles^[18,19]. These films retain the flexibility of the weave without the fear of fracture. More importantly, they can be directly integrated within a conventional wound dressing. It was envisaged that the carbon component would provide the framework for electrochemical transduction enabling quantitative information on key biomarkers associated with wound healing to be extracted through an appropriate electronic monitor^[18] – worn either by the patient or connected at the time of consultation. This would be a more viable solution given the inherent mechanical flexibility of the composite film – a prerequisite given the high degree of morphological variability encountered with diabetic ulcers. Moreover, the film approach would have the critical advantage of component simplicity (requiring only carbon and polyethylene) and hence provide a more inexpensive option when considering manufacture. The possibility of extruding the material in the form of large area films would be particularly amenable to large volume production – especially given the frequency with which dressings would be changed. The electroanalytical performance of these films has

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been shown to accurately measure pH, having been tested in simulated and whole blood samples^[18,19].

Thus far, urate has been used as an indicator of pH yet it has been established in recent years that its quantification is of considerable interest as Fernandez et al.(2012) have shown that its concentration is elevated as a consequence of tissue remodeling within the wound. The cellular trauma and associated healing processes result in the release of adenosine triphosphate (ATP) into the extracellular wound matrix which is subsequently metabolised into uric acid^[24]. Thus, the ability to electrochemically monitor its concentration could be useful in determining the extent of inflammation and the subsequent response to treatment - therein providing insights into the healing process^[24]. This can be achieved through a procedurally simpler approach, involving the amperometric oxidation measurement of uric acid, As a biomarker, it has the advantage of being found in relatively high concentrations and there is an extensive amount of literature on its electrochemical detection^[27]. It is somewhat ironic to note that whilst the ease with which it can be electrochemically oxidised is often considered to be an interference in electroanalytical methods, it is this same property that has made it attractive as a biomarker in the present context. This approach has seen further refinement by Wang and co-workers who have adapted the system for wireless recording of the wound urate concentration through the use of an enzyme based system^[52].

6.0 Summary

Disposable screen printed electrodes have long been suggested as the preferred platform for point of care electrochemical sensors suitable for use within the clinic but while transfer to the home, in a manner analogous to personal glucose monitoring, would be ideal, the implementation is problematic. Chronic wounds invariably require handling under aseptic conditions by trained personnel and it is likely that having such processes conducted by a patient may interfere with the healing process, not only through mechanical disruption of the

wound but also by rendering the exposed tissue susceptible to infection. The “Connected Health” scenario, where electrochemical techniques have the potential to excel, could be more profitably exploited where the sensor substrates are directly integrated within the dressing. These could then perform periodic monitoring of the wound status, typically pH, with minimal intervention and report back to patient or, where appropriate, to a healthcare professional in order effect a more speedy intervention. In many cases, simple amperometric detection strategies could be employed but, even with more complex square wave techniques, it is unlikely that the demands of the instrumentation will be an impediment to the implementation of the technology. The challenge for realistic clinical success however will be to provide economically viable, robust and clinically informative electrode systems that are inherently disposable. This is particularly significant where sensors are embedded within conventional dressings and may need to be changed daily over many months. It could be anticipated that should these hurdles be overcome, the cost of the instrumentation would be more than offset through avoiding the prolonged treatment and limb threatening complications of infection.

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Figure 1. Interdisciplinary nature of smart dressing research

Figure 2. Typical pH profiles observed for acute and chronic wounds.
(Adapted from Scheinder *et al.* 2007[28])

Figure 3. Different material approaches to the design of conductive smart dressings: carbon fibre weave (A), laser etched carbon loaded polyethylene mesh(B), and carbon-polycarbonate film integrated within a conventional dressing (C). The potential implementation of the dressing is indicated in the sketch in (D) and the typical voltammetric pH profile for the oxidation of uric acid is highlighted in (E) [16-20]

The availability of sensors that could measure wound pH, either in the clinic or in home could significantly improve clinical outcome – particularly in the early identification of complications such as infection. This review identifies new materials and electrochemical research strategies that are being targeted at wound diagnostics and the design of smart dressings

Keywords: Electrode;sensors;Wound;Healing;Infection

Anna McLister¹, Jolene Phair¹, Jill Cundell², James Davis^{1*}

New Developments in Smart Bandage Technologies for Wound Diagnostics

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