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Article *in* Nanomedicine · September 2017

DOI: 10.2217/nnm-2017-0173

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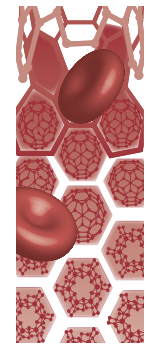

Can regenerative medicine and nanotechnology combine to heal wounds? The search for the ideal wound dressing

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Skin is the outermost covering of the human body and at the same time the largest organ comprising 15% of body weight and 2 m^2 surface area. Skin plays a key role as a barrier against the outer environment depending on its thickness, color and structure, which differ from one site to another. The four major types of problematic wounds include ulcers (diabetic, venous, pressure) and burn wounds. Developing novel dressings helps us to improve the wound healing process in difficult patients. Recent advances in regenerative medicine and nanotechnology are revolutionizing the field of wound healing. Antimicrobial activity, exogenous cell therapy, growth factor delivery, biodegradable and biocompatible matrix construction, all play a role in hi-tech dressing design. In the present review, we discuss how the principles of regenerative medicine and nanotechnology can be combined in innovative wound dressings.

First draft submitted: 12 May 2017; Accepted for publication: 18 July 2017; Published online: 4 September 2017

Keywords: nanotechnology • skin • smart dressings • stem cells • tissue engineering • wound healing

Each year an increasing number of people suffer from problematic skin wounds and other skin diseases that compromise the skin barrier, and wound dressings are the principle therapeutic intervention for these defects (that for one reason or another cannot be treated by skin grafts). The average commercial value of wound dressings is estimated to be around 4–9 billion dollars, and it is projected to grow by 4–7% a year until 2021 [1]. Skin contains three major layers including epidermis, dermis and hypodermis (subcutaneous tissue). The epidermis is the outer layer of skin which forms the main barrier to the outside. This layer has no independent blood supply and is nourished via diffusion of nutrients from the dermis. The epidermis helps the skin to regulate the body temperature and retains moisture inside. The dermis beneath the epidermis provides the main structural elements and imparts strength to the skin. The hypodermis contains fat cells that store energy reserves [2].

Wounds can be divided into two groups, acute and chronic. Acute wounds are formed by traumatic damage to the skin such as injury (incisions, abrasion, etc.), surgery, thermal or chemical burns. Acute wounds generally have a shorter healing time than chronic wounds. Chronic wounds often occur due to metabolic disturbances caused by systemic diseases such as diabetes, vascular insufficiency or obesity. Many deeper acute wounds heal imperfectly, leading to scar formation at the wound site [3]. To overcome this problem, many procedures have been proposed to fully regenerate the skin.

There is now a major epidemic of diabetes in both the developing world, as well as in developed countries. Diabetes is a metabolic disease in which lack of insulin secretion or insulin resistance leads to high blood

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glucose levels. Diabetics have impaired wound healing for a number of reasons including metabolic disturbances, poor circulation, neuropathy leading to loss of sensation, immune system deficiency and propensity to infection. Diabetes is the seventh leading cause of death in the USA, and is the single most important cause of lower limb amputation. A variety of wound dressings have been used for wound healing in diabetic wounds, such as collagen, poly(lactic-co-glycolic acid) (PLGA) [4], chitosan [5], etc. Venous ulcers are caused by impaired blood flow occurring usually in the legs due to failure of the valves in the leg veins allowing blood to pool rather than returning to the heart [6]. The most prevalent dressings that have been utilized for venous ulcers are polyurethane (PU) [7], alginate [8], collagen [9], chitosan [10] and gelatin [11]. Pressure ulcers occur because of the tissue breakdown that occurs under constant pressure, which reduces the blood circulation in that area. Wound dressings for pressure ulcers are hydrocolloids [12], foams [13] and films [14]. In burn wounds, the main goal is to preserve the wound area in optimum condition by preventing water loss and avoiding infection using hydrophilic materials such as hydrogels [15], collagen [16] and peptides [17]. Overall, the pore dimension and thickness are other factors which should be mentioned in the design of wound dressings.

Traditional wound dressing (TWD) was designed to just cover the wound and played no active role in wound healing and skin regeneration. Recent progress in wound dressing technology has led to sophisticated materials with various properties such as exudate absorbance, elastic substrate that conforms to the skin and moisturizing ability. However, even these dressings exhibit the same behavior on every wound. Nanotechnology enables adjustable features to be provided to wound dressings. Nanotechnology-based wound dressings exhibit behavior that can be tailored for each individual wound and can prevent the deterioration of chronic wounds [18]. Nanotechnology-based wound dressings act as a smart substrate in which the release of antibacterial, anti-inflammatory and drug/growth factors (GFs) can be varied in response to the wound environment. Pathogenic bacteria release secreted virulence factors, such as lipase and protease enzymes, that can result in cell death and tissue destruction. With nanotechnology, the antimicrobial materials can be embedded in nanocontainers in which drug release is sensitive to the presence of pathogenic bacteria [19]. TWDs enable antibacterials and drugs to be released in response to an actual infection, but limits the exposure of noninfected wounds to drug doses that may have side effects and tissue toxicity [20]. TWDs can therefore respond to early indicators of infection and can be engineered to change the visible color and act as an 'early warning system' as well as releasing anti-infective compounds on demand [21].

The ideal wound dressing should have sufficient mechanical stability, engineered structure (nanofiber [NF], porosity), absorb wound exudate, moisturize the wound milieu, allow exchange of gases, and release wound healing mediators and antibacterials according to the wound requirements (Figure 1). In this review, we focus on advanced wound dressing technology that incorporates approaches from the field of regenerative medicine and nanotechnology.

Skin defects & the healing process

Defects in the skin occur due to many reasons: various types of wounds such as acute wounds (surgical, traumatic and burns), chronic wounds (diabetic, venous and pressure ulcers, [22]; genetic diseases such as lamellar ichthyosis, Vohwinkel syndrome, etc., can cause skin defects [23]; the aging process causes some defects to skin because of wrinkling, dryness, etc. [24].

Wound healing is a dynamic biologic process which has four phases, which are depicted in Figure 2. Hemostasis; in this stage blood vessels become constricted then a clot forms due to platelet aggregation and degranulation, and activation of the coagulation cascade occurs to form fibrin. Inflammation: in this step neutrophils infiltrate the wound site, followed by monocytes that differentiate to macrophages. Proliferation: in this phase epithelialization occurs, angiogenesis takes place, new collagen is synthesized and the extracellular matrix (ECM) is formed. Remodeling: in this phase, the newly synthesized collagen is remodeled, and the neovasculature matures and regresses. Well-coordinated wound healing requires the sequential occurrence of all these four stages [25,26]. There are many extraneous factors that may affect these phases, thus for better wound healing, the negative factors should be eliminated and the positive factors should be fortified. The application of external sources of physical energy can promote wound healing such as electrical stimulation [27], electromagnetic fields [28], ultrasound [29], cold plasma [30], Light Amplification by Stimulated Emission of Radiation (laser) [31] and light [32]. Biologic and synthetic wound dressings with and without the inclusion of exogenous cells or GFs are useful for wound healing and regeneration. Skin autografting is a procedure in which the epidermal layer or a full-thickness layer including the dermis (depending on the wound) is surgically transferred from a distant donor site to the wound site. Although highly effective in repairing large skin defects, this approach is

limited due to restricted availability of suitable donor sites. Therefore, other methods are preferred such as engineered wound dressings based on nanotechnology materials combined with cells or GFs [26,33].

GFs, exogenous cells & biomolecules

GFs are highly specific proteins that bind to cell-surface receptors and govern the growth, proliferation, migration and differentiation of a wide range of cells. Neutrophils secrete interleukins to activate macrophages and other immune cells, and FGFs to stimulate collagen synthesis and encourage fibroblasts to transform into contractile myofibroblasts [34,35]. EGF, cytokines, etc., are released by activated platelets [36]. Macrophages and keratinocytes release TGFs and VEGF that regulate the proliferation of endothelial progenitor cells and PDGF which also helps in angiogenesis [37,38]. In order to increase the concentration of GFs in the actual wound, these molecules cannot be injected systemically, and must therefore be delivered topically into the wound. This wound-specific delivery has been accomplished using nano-delivery vehicles such as nanoparticles, liposomes, polymersomes and scaffolds [39].

Stem cells (SCs) have impressive self-renewal properties and the ability to differentiate into a wide range of cell types. Miscellaneous types of SCs have been utilized for skin regeneration and wound healing such as embryonic SCs [40], adult SCs [41], induced pluripotent SCs [42], mesenchymal stem cells (MSCs) [43], adipose-derived stem cells [44], human umbilical cord derived SCs [45] and melanocyte SCs [46]. Applying MSCs into the wound area increased re-epithelization and helped angiogenesis [47]. Embedded adipose-derived stem cells within an acellular dermal matrix applied onto the wound led to increased wound healing and better vascularization [44]. Human-induced melanocyte SCs produce hair follicles and epidermal pigment [45]. Skin-derived SCs were utilized for regenerating the neural cells in the skin that could be damaged in burn wounds [48].

Skin aging is an inevitable process of deterioration that is affected by extrinsic factors (exposure to UV radiation) and intrinsic factors such as accumulating oxidative damage. Reactive oxygen species (ROS) and free radicals are important factors in the oxidation process which leads to skin aging [49].

There are some important factors that affect both the endogenous and exogenous modes of skin aging. The molecular mechanisms of the intrinsic (chronological aging) and extrinsic (photoaging) processes have things in common, but there are also differences. The pathological mechanism of intrinsic aging concerns cellular aging, telomere curtailing, DNA/genetic mutations, oxidative stress and hormone levels [50]. Studies have suggested that ROS is the main reason behind photoaging (and to some extent behind chronological aging). ROS production arises from normal oxidative cell metabolism and causes elevated levels of cell damage especially on a cumulative basis. MAPKs are activated by ROS and induce the transcription factor c-Jun to hasten skin aging. This process results in activation of the important transcription factor activator protein 1 (AP-1), which causes expression of matrix metalloproteinases MMP-1 (interstitial collagenase), MMP-3 (stromelysin 1) and MMP-9 (gelatinase b), and thereby prevention of procollagen-1 expression [51]. It has been suggested that the expression of CTGF and TGF-β and the Smad signaling pathway affect the expression of type I procollagen in intrinsically aged skin [52]. UV-light exposure is the main reason behind exogenous skin aging and also photocarcinogenesis. Photocarcinogenesis is related to the process through which living tissue is exposed to illumination (usually sunlight), containing UV radiation which results in generation of free radicals and ROS that react with and damage several essential biomolecules [53]. Moreover, the absorption of short wave UVB by the epidermis assists in formation of photocarcinogenic cyclobutane pyrimidine dimers and 6,4-photoproducts that are considered to be the typical DNA photolesions. It has also been accepted that other cellular chromophores (e.g., melanin precursors and riboflavin) can absorb UVA light and also generate ROS, again with negative consequences to DNA, proteins and lipids [54].

The mechanisms of the skin defense system against oxidative stress are divided into several subgroups including: repair systems (e.g., DNA repair system);

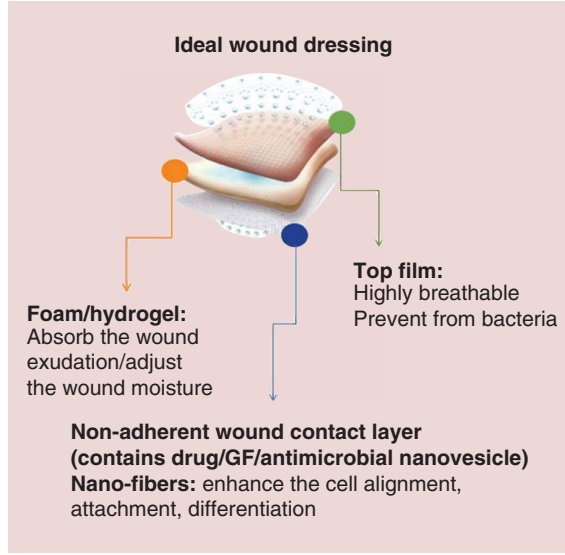

Figure 1. Criteria for an ideal wound dressing.

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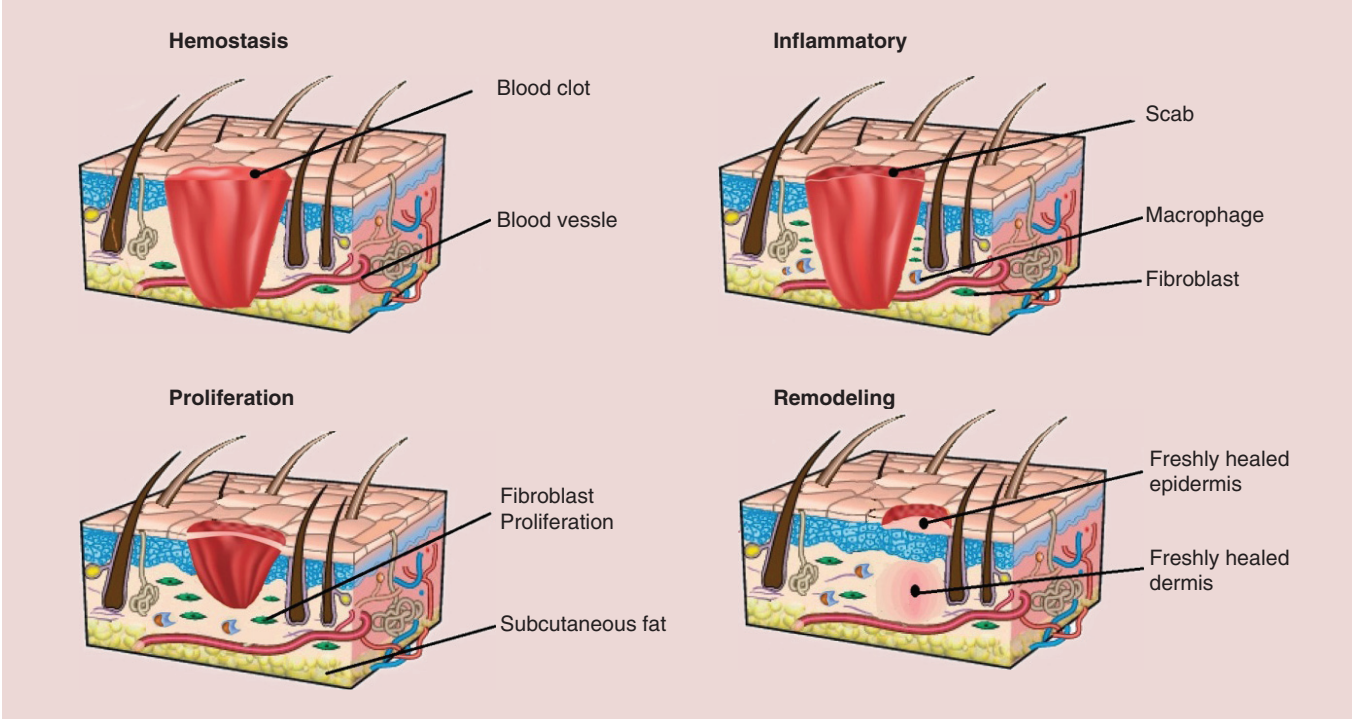

Figure 2. Different stages of wound healing: hemostasis (top left), in which vascular is constricted, then platelets aggregate, degranulate and fibrin is formed; inflammation (top right), in which neutrophil infiltrates, then monocyte infiltrates and differentiates to macrophage; proliferation (bottom left), where epithelialization reoccurs, angiogenesis happens, collagen is synthesized and ECM is formed; remodeling (bottom right), where collagen is remodeled and vascular matures and regresses.

prevention mechanisms (e.g., prevention of the production of reactive ROS); physical defenses (e.g., stabilization of biological sites, steric interface); antioxidant defense (e.g., enzymes, scavengers). Antioxidant (AO) defenses are the most important mechanism which interact with ROS and inactivate free radicals. AOs are classified into two subgroups: low molecular weight antioxidants and enzymes. Low molecular weight antioxidants donate electrons and quench the ROS [55]. Regular wound healing physiology requires a low concentration of ROS for its optimal function, yet not in sufficient quantity or for such a prolonged period to cause oxidative stress. By contrast, the use of an AO can help promote the healing process by decreasing the damage through consumption of oxygen radicals and oxidative stress [56,57].

Preventing oxidative stress results in more healthy skin. Skin has some naturally occurring AO molecules which play a role in reducing oxidative stress like tocopherols [58], ascorbate [59] and superoxide dismutase [60]. Honey [61], vitamins A [62], E [63], C [64], D [65] and selenium derivatives [66] can all act as exogenous AOs. Direct topical application of these substances (either individually or in combination) could be done periodically, but incorporation into engineered dressings or immobilization into scaffolds will result in controlled release and better regulated healing.

Therapy methods

Wound dressings

A typical wound dressing is used as a protection for the wound, preventing dehydration and infection. There are various types of wound dressings available commercially (Figure 3).

Wound dressings are used as a supporting structure for the wound and to immobilize and protect it. The ideal wound dressing should have the correct mechanical features, protection against bacteria, maintenance of the right level of moisture in the wound, gas/fluid permeation characteristics, nontoxic/nonallergenic properties, good handling characteristics, etc. The correct dressing type should be chosen for each wound type (such as diabetic, venous, pressure ulcers, burn, surgical and traumatic, etc.) which can either be exudative or dry. Commercial dressings are usually available in the form of hydrogels, hydrocolloids, hydrofibers, alginate dressings, collagen dressings, foams and films [22]. Other types of dressings contain cells and GFs such as Apligraf® (Organogenesis Inc., MA USA), which is manufactured from human neonatal keratinocytes and fibroblasts. The materials used to make a wound dressing can be classified as either natural or synthetic. Chitosan, alginate and collagen are all naturally occurring polymers, which have a structural elements in common with normal ECM, thus they offer

good cell attachment and preserve cell viability. On the other hand, the properties of synthetic polymers such as PUs can be tailored to provide the optimum physical features for different wound dressings [67,68]. In Table 1, the various synthetic commercially available dressings are summarized with their pros and cons.

Low-power light therapy

Photobiomodulation (PBM) or low-level laser (light) therapy involves the use of nonthermal levels of red or near infrared (NIR) light to stimulate tissue repair and improve healing [69]. Lasers and light-emitting diode (LEDs) are both medically used in PBM. The red/NIR wavelengths can penetrate into tissue [70], where they stimulate mitochondrial respiration and trigger transcription of new gene products [71]. Low-level laser (light) therapy has been used for wound healing since the 1960s [72], but recently the term PBM was internationally adopted [73]. Nowadays, high-frequency pulsed low-power diode lasers have been used due to their better penetration. PBM accelerates epithelialization, wound healing, reduces pain and hastens the resolution of inflammation [70,74]. *In vitro* studies have demonstrated that LED and lasers have a similar effect on cells. *In vivo* wound healing studies have shown fewer inflammatory cells, increased angiogenesis, more collagen production, proliferation of fibroblasts, endothelial cells and keratinocytes and better development of granulation tissue. Silveira *et al.* [75] compared laser and LED for burn wound healing. Results showed that the laser and LED both decreased inflammation and ROS, producing less dermal necrosis, and formation of granulation tissue was accelerated.

Ultrasound & shock waves

Ultrasound (US) is mechanical vibration at a frequency higher than the human hearing range. The vibrations of a mechanical transducer produce an acoustic wave that can travel through tissue. Low-intensity pulsed US (LIPUS) has previously been used in tissue engineering [76], and is often used for healing of bone fractures [77]. Iwanabe *et al.* investigated LIPUS for wound healing in an *in vitro* model [78]. Their results indicated that the LIPUS could accelerate the closure of a scratch in cell cultures, increase the proliferation of gingival epithelial cells and regulate mRNA and protein expression. Several research groups have used low-intensity US for third-degree burn wound healing in rats. In a study by Fantinati *et al.* [79], LIPUS had impressive effects on inflammation, proliferation, and enhancedgranulation tissue by promoting the formation of fibrin-leukocyte crusts. On the other hand, it was not so effective in improving angiogenesis and mononuclear inflammatory infiltration. In another study, LIPUS treatment showed some improvement in body weight and granulation tissue formation, but no difference in the wound healing of burns in rats [80].

A somewhat related intervention, is the application of extracorporeal shock waves (ESW). These are abrupt, high amplitude pulses of mechanical energy, generated by an electromagnetic coil and applied through a pad onto the skin. Originally developed to treat kidney stones (shock wave lithotripsy), ESW is now widely applied in physical therapy for fractures, tendinopathies and wound healing [81]. A clinical study used two ESW therapy sessions in 15 patients with <5% total body surface area deep partial/full thickness burns, on the third and fifth day after injury [82]. A total of 80% of the burns healed by 3 weeks; while 15% required surgical debridement and grafting and 5% developed hypertrophic scarring. After one ESW session, burns had a significant increase in perfusion as shown by laser Doppler imaging. Another clinical study reported on 23 patients with chronic diabetic ulcers who were randomized to receive a series of six ESW treatments over 3 weeks in combination with standard care or standard care alone [83]. ESW therapy was performed using 250 shocks/cm^2 and 500 shocks on arterial beds supplying the ulcer location. Transcutaneous oxygen tension was significantly increased in patients treated with ESW therapy compared with controls. Ulcer area reduction was 34.5% in the intervention group versus 5.6% in the control group at 7 weeks.

Electricity

Several different types of electrical stimulation (ES) have been described for medical applications, including direct current (DC), alternating current, high-voltage pulsed current and low-intensity direct current, pulsed electromagnetic field and transcutaneous electrical nerve stimulation [84]. Electrical stimulation increases chemotaxis, cell-based immunity, vascular perfusion, resistance to infection, epithelial migration/orientation and improves wound healing [84]. Rouabhia *et al.* showed that not only did ES has no harmful effects on fibroblasts (such as cytotoxicity) but ES also stimulated fibroblast growth and migration, GF secretion and boosted the fibroblast to myofibroblast transdifferentiation [85]. It has been reported that, pulsed DC ES causes better wound healing than constant DC ES. Moreover, the application of ES was beneficial for stage III and IV pressure ulcers rather than stage II ulcers [86,87].

Plasma

Plasma is a gaseous mixture of negatively charged electrons and highly charged positive ions, and has been called the 'fourth state of matter'. Incipient plasma

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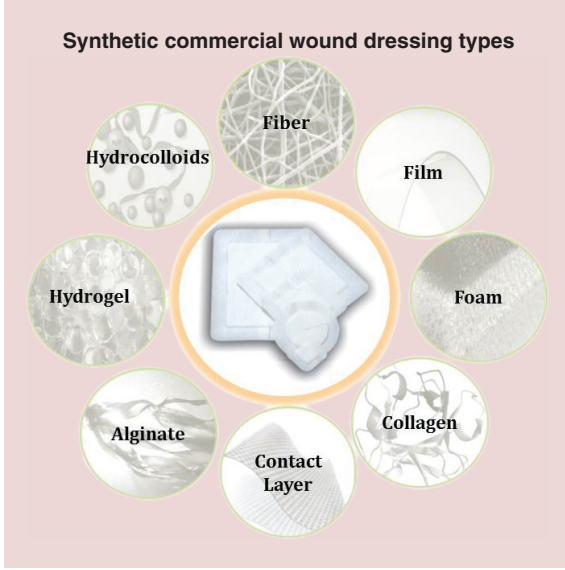

Figure 3. Types of commercially available synthetic wound dressings.

(temperature above 80°C) is formed by laser ablation of aluminum in air, and has been used for sterilization, tissue removal, cutting, etc. On the other hand, cold atmospheric plasma (CAP) has been widely utilized for antimicrobial activity, regenerative medicine and morphogenesis. CAP can be produced by an electric discharge between two electrodes in a flowing stream of gas (argon for instance). CAP has been shown to stimulate keratinocytes and fibroblasts and alter their migration [88,89]. Pathogens are especially vulnerable to CAP and resistance has not been reported [90]. CAP was tested for wound healing in diabetic rats and stimulated epidermal layer formation, cell proliferation, neovascularization and increased levels of TGF- β1 cytokine [91]. Regulating the dosage of CAP is essential, as optimum dosage promotes wound healing, while an excessive dosage causes cell necrosis/apoptosis and hinders wound healing [92].

Advanced wound dressings

A proper scaffold for wound dressings should have good mechanical features, suitable biocompatibility and appropriate structure to provide the best condition for cells (Figure 5). Different morphologies of the scaffold (fiber, foam, etc.) have been used for skin regeneration. Table 2 lists some advanced wound dressingfeatures from a nanotechnology perspective.

Nanofibers

The structural zones of the dermis are divided into three subgroups: a papillary layer with delicate fibers in the 0.3–3 μm width range, which are juxtaposed with the epidermis; a mid-zone composed of a dense layer of concentrated fiber bundles (aligned fine fibers) in the 10–40 μm width range; and a deep-zone with a broad range of fiber bundles. It has been reported that the collagen in the dermis forms 'basket weave' like structures with vertical collagen fibers crossing each other at 90° angles [93]. Various techniqueshave been used to artificially fabricate such basket weave structures from soluble collagen, including electrospinning, braiding [94] and 3D printing [95]. The fibrous structure of the skin ECM results in better compatibility, with NFs, and collagen fibers occupying the dermal matrix. Therefore, ECM-based NFs can recapitulate the skin due to their structural and compositional resemblance to the natural structure [96]. Core-shell polylactic acid (PLA)-aceclofenac and PLA-insulin eluting PLGA fibers exhibited reduced epidermal hyperplasia and increased the migration of skin cells [97]. It is considered desirable for the NFs composing the dressing to mimic as closely as possible the fibers making up the ECM, which is the basic biological material upon which the cells survive and grow. Structural features such as a high surface to volume ratio, high porosity and better alignment result in biomimicking to encourage cell growth, proliferation and differentiation. Moreover, NFs due to their sieve effect with nanopores, hamper the penetration of exogenous bacterial and fungal cells [20]. Pristine NFs promote hemostasis and trigger skin regeneration, moreover they can help to reduce the scar due to their structural alignment with the newly formed collagen fibers [98]. It is noteworthy that NFs exhibit a dynamic interaction with the wound milieu; thus, properties of the NFs such as alignment, diameter, chemistry, loaded drug/GF/antibacterial, all affect the healing process [99]. The NFs are usually fabricated using electrospinning, phase separation or self-assembly. Electrospinning is a simple method in which high voltage is applied to the polymer solution and the fibers are formed on the collector. FGFs are often used for wound healing, however, during the electrospinning process these biomolecules were damaged because of high voltage involved. Overcoming this drawback, the fibers of PLGA were fabricated by electrospinning, then FGF was subsequently coupled to the fibers by polydopamine, thus the FGF preserved its activity [100]. It was hypothesized that use of the blend/emulsion electrospinning method could prevent the degradation of biomolecules by using another molecule as a shield. In this method, an emulsion of water soluble/nonwater soluble molecules disrupts the thermodynamic stability during the process. Bovine serum albumin was used as a carrier and protectant of biologically active molecules including vitamin C, hydrocortisone, insulin, triiodothyronine, EGF and dihydroxyvitamin D. The mixture was electrospun with PLGA/collagen and the release of biomolecules from the fibers stimulated

Table 1. Synthetic commercially available dressings and their pros and cons.

Wound dressing	Material	Application	Advantage	Disadvantage	Company/ brand name
Hydrocolloid	Bilayer structure Inner layer: gelatin, pectin, carboxymethyl cellulose, elastomers Outer layer: waterproof layer	Low or medium exudative wound: pressure, diabetic wounds	Could be removed without pain, maintains the wound moisture, adheres to the dry and humid part	Not suitable for high exudation wounds. Useless for infection wound and necrotic tissues	3M, Tegaderm, comfeel
Hydrogel	Methacrylates, PVP, gelatin, pectin, chitosan	Chronic dry wound (low humid), wound with necrotic tissue, medium or deep wounds: low burning, pressure and abrasion wounds	Provides a humid environment, cools the wound and lowers pain. Useful for necrotic and infectious wounds	Dehydration, low absorbance	Aquaclear, purilon gel, hypergel sterigel
Alginate	Sodium/calcium alginate salts	High and medium exudative wound, deep wound: diabetic, pressure, abrasion, venues, burning type II	Provides a humid environment, low allergic feature, useful for necrotic and infectious wounds	Dehydration, periodic control for deep wound	Algisite, Algoderm, Sorbsan
Foam	Silicone or PU foam	High and medium exudative wound, diabetic, pressure, abrasion, venous, burning type II	Insulates heat and warms the wound, make pressure for venous ulcer	Allergic for delicate skin, useless for the dry and infectious wound	Allevyn, Lyfoam, Tegafoam
Semi-permeable film	Transparent and semi-permeable PU	Used as a second layer for hydrogels and foams, nonexudative wound: surgical wound	Waterproof layer, permeable to oxygen, vapor. Barrier against infections	Useless for bloody/ high exudative, necrotic, infectious wound	Tegaderm, hydrofilm, polyskin

PU: Polyurethane; PVP: Polyvinylpyrrolidone.

proliferation of keratinocytes and human dermal fibroblasts [101]. Different nanoparticles were used in scaffold construction such as ZnO, nanoceria, Ag, TiO_2, Cu, Fe_3O_4, Al_2O_3 and SiO_2. All of these nanoparticles have antibacterial properties. ZnO also has an antioxidative property which enhances the wound healing. Embedded ZnO nanoparticles in polycaprolactone fibers gave increased cell adhesion, fibroblast proliferation and improved wound healing [102]. Metals of the lanthanide group have been utilized in regenerative medicine. Nanoceria is an ROS scavenger material, moreover, it stabilizes HIF-1α expression that causes to stimulate angiogenesis via upregulation of VEGF and modulates the oxygen level in the wound by gene regulation [103,104]. Nanoceria/PU/cellulose acetate fibers were used for wound healing which exhibited antibacterial properties due to the release of free cerium ions [105]. Silver exhibits antimicrobial and antibacterial properties and combats wound infections. Silver/nanosilver has been embedded in various fibers to impart antimicrobial activity [106,107]. Wu *et al.* deposited silver nanoparticles onto bacterial cellulose using a self-assembly process, which led to the toxicity of the nanoparticles being decreased, while they still retained antibacterial properties [108]. Iron oxide nanoparticles added to poly(ethylene oxide)/poly(vinyl pyrrolidone) fibers showed antimicrobial properties and prevented the growth of Gram-positive and Gram-negative bacteria [109]. Gold nanoparticles have been used as a carrier for other molecules such as alpha lipoic acid and epigallocatechin gallate that act to regulate inflammation and angiogenesis. Gold nanoparticles control the production of ROS and show AO properties [104]. Surfactin (an antimicrobial lipopeptide) and 1-dodecanethiol were deposited on gold nanodots and displayed synergistic antimicrobial properties, superior biocompatibility, better epithelialization and collagen deposition [110]. Bio-activated plasma-treated electrospun fibers based on polycaprolactone (PCL)/olfactomedin-like 3 (Olfml3) proteins were fabricated in which the PCL provided a 3D environment and Olfml3 caused cellular migration and endothelial cell attachment (especially in the remodeling stage) [111]. Cell-seeded NFs have been widely utilized in dermal/epidermal

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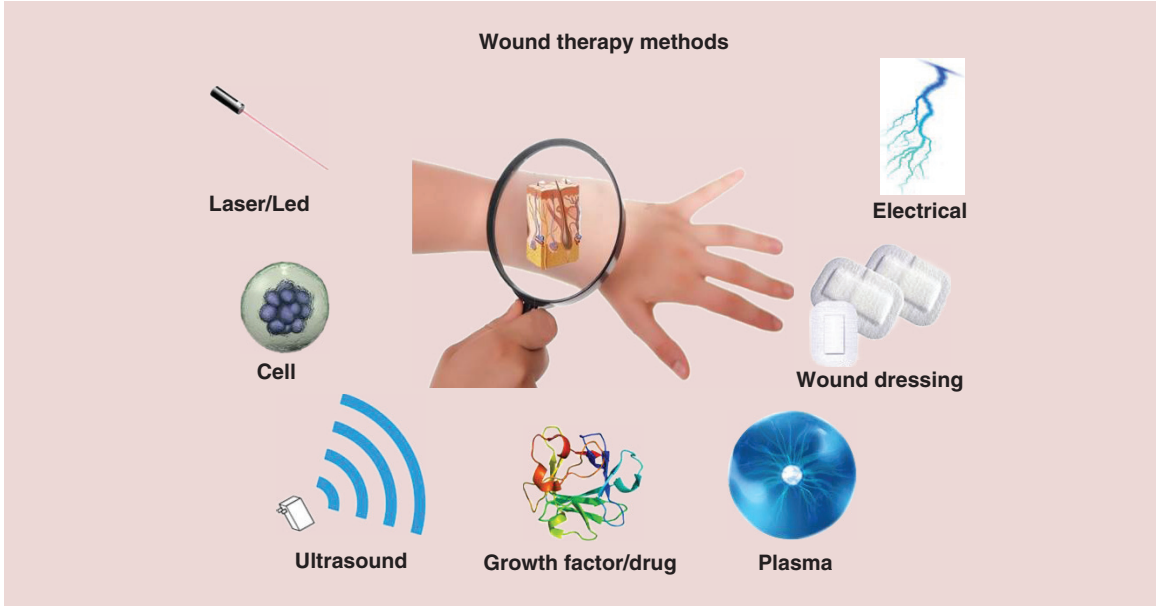

Figure 4. Various therapeutic methods which have been utilized for skin regeneration.

and full-thickness skin regeneration. 3D electrospun NFs can enhance the cellular activity, for example, infiltration and proliferation for dermal regeneration. The ECM secretion by human dermal fibroblast indicated that 3D NFs facilitated the regeneration [112]. Human adipose-derived SCs were seeded on the basketweave structurally aligned NFs, which exhibited good cell proliferation and well-organized distribution, thanks to their nanotophographic surface [95]. Overall, 3D NFs, due to their ECM-like structure, can enhance the cell infiltration leading to better tissue integration and neovascularization after implantation.

MSCs seeded on NF substrates are promising for regeneration; these are good examples of nanotechnology and regenerative medicine combined in wound healing. MSCs participate in all stages of healing; for instance stimulating the vascularization in granulation tissue, enhancing keratinocyte migration and ECM synthesis, moreover MSCs have antimicrobial properties because of antimicrobial protein secretion [113,114]. Direct injection of MSCs into tissue suffers a drawback of excessive cell apoptosis, while NF-seeded MSCs facilitate cell attachment, proliferation and differentiation.

Hydrogels

Hydrogels are a 3D network composed of polymer strands, which can contain up to 1000-times their own weight of water. The 3D structure of the hydrogel is suitable for cell seeding and enhances the cellular activities, such as cell migration and ECM deposition. Hydrogels act as a barrier against bacteria and microorganisms, absorb exudate, provide a moist environment and can cause hemostasis. In general, hydrogels decrease the temperature, reduce pain and promote fibroblast proliferation, keratinocyte migration and wound re-epithelialization and lessen scar contracture. Nanostructured hydrogels with a biologically relevant pore size (around 100 nm) can prevent the bacteria penetration, but still allow the passage of nutrients and biomolecules [115]. Moreover, cellular response depends on pore size and void fraction, both of which can control the rate and depth of cellular ingrowth. Canine dermal fibroblasts (DmFb), vascular smooth muscle cells and micro-vascular epithelial cells are examples of cells with suitable cell sizes so their response was evaluated. It was concluded that 75% void fraction was not suitable for tissue formation, where a thin endothelial lining formed on the scaffold with pore sizes <38 μm [116]. Addition of particles, biomolecules, GFs and cells increase the wound healing properties of hydrogels. A ZnO/chitosan composite hydrogel was utilized as a wound dressing which accelerated wound healing, re-epithelialization and collagen production; moreover, cells seeded on the hydrogel exhibited good attachment and penetration into the scaffold [117]. Loa *et al.* synthesized a peptide self-assembled nanofibrous hydrogel which promoted both epithelial and dermal regeneration (without GFs) in burn wounds [118]. In addition, it had antibacterial and anti-inflammatory properties. Paladini *et al.* synthesized a self-assembling di-phenylalanine hydrogel containing silver which showed good antibacterial properties [119]. In another investigation an ultra-short peptide was self-assembled in the hydrogel and cross-linked by disulfide bonds to produce a transparent dressing thus

the wound could be observed over time. It promoted re-epithelialization and wound healing [120]. Linear and branched polyethylenimine cross-linked onto gelatin fibers exhibited microbicidal properties and could eliminate bacteria and yeasts by rupturing their cytoplasm membrane [121]. Diacrylate poly (ethylene glycol) and heparin were cross-linked to form a biocompatible hydrogel in which EGF was loaded. It had good mechanical properties, water absorbance, permeability for vapor and encouraged wound healing [122]. Soy protein loaded with gentamicin in a dressing was synthesized using glyoxal/l-cysteine as cross-linkers and glycerol/sorbitol as a plasticizer. Soy protein is low cost, stable and has a good storage time, and in addition it is derived from nonanimal origins. Mechanical properties and water uptake were affected by type and amount of cross-linker and plasticizer. The desirable water vapor evaporation was tailored for a suitable evaporation rate of around 2300 g/m^2/day. The release mechanism was diffusion. It was suggested that this dressing was appropriate for burn wounds [123]. Hydrogels based on oligomeric peptides self-assembled in aqueous media such as nanostructured hydrogel are pH-sensitive and thermo-sensitive. These types of nanostructured hydrogel can be used as scaffolds for drug/GF delivery [124]. Drug/GF hydrogels can accelerate wound healing. FGF-2-loaded photo-crosslinked chitosan-stimulated wound healing, wound closure, granulation and epithelialization [125].

PU-based wound dressings

PU is a versatile polymer which is used in various applications such as the packaging industry, electronic industry, medical applications, etc. PU is formed by reaction of di/poly/isocyanates, di/polyol/esters and a chain extender. Diols and diisocyanate form the soft and hard segments, respectively. Interaction of each segment by itself forms domains (soft and hard) which provide the unique properties of PU such as the elastomeric feature formed by interaction between these domains. PU can be fabricated into miscellaneous morphologies such as elastomer, foam, thermoplastic, fiber, etc. [126]. PU is highly practical in wound dressings because of its tailorable properties such as mechanical strength, water absorption, moisture regulation and vapor permeation. Adding nano-platelets of graphene oxide to a PU/siloxane membrane improved the mechanical stability, allowed tunable absorbance of exudate (due to the hydrophilic/hydrophobic behavior) and introduced an antibacterial property to the wound dressing. Dressings with nano-platelet graphene oxide (GO) resulted in better vascularization, re-epithelization (due to controlled moisture) and improved collagen regeneration compared with the non-GO counterpart [127].

Azidophenyl-modified fish gelatin was photocured by UV light on PU foam for protein release to improve wound healing [128]. A rapid hemostatic-enabled wound dressing was synthesized based on PU-urea (PUU) which coagulated blood rapidly and promoted wound healing. PUU exhibited better mechanical properties and hydrophilicity than PU alone, due to the urea hydrogen donors which allowed hydrogen bonding. PUU had better and more rapid water absorbance, and as a result, coagulation occurred as soon as water absorbance [129]. Polyethylene glycol (PEG) is one of the useful polyols used as a soft segment in PU synthesis because of its biocompatibility, low tox-

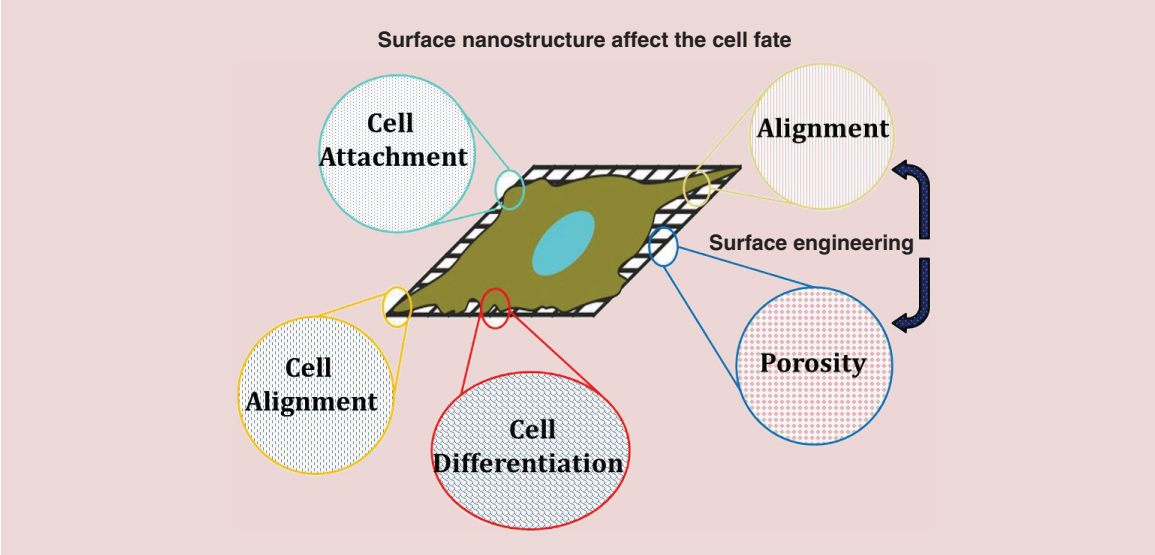

Figure 5. Surface morphology of nanostructures affects cellular behavior.

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icity and elasticity [130]. PLA was used to synthesize PU and improved the PU mechanical features but reduced the water-uptake. The usage of a mixture of PLA/PEG as a polyol exhibited the desired mechanical properties, resilience and water absorption [131]. Poly (ε-caprolactone)/PEG yielded thermo-sensitive PU which was able to provide controlled drug release [132]. A mixture of PEG/glycerol ethoxylate (GE)/pluronic was used as the polyol in a PU foam hydrogel synthesis. The mechanical and thermal properties were tailored by varying the PEG segmental length and pluronic concentration. A long segment of PEG loaded with ciprofloxacin exhibited better antibacterial properties. Moreover, samples showed an initial burst of drug release which was necessary for prevention of infection [133].

Antibacterial/antimicrobial nanoparticle/vesicles embedded in wound dressings

Pathogenic bacterial species which are abundant in acute and chronic wounds include *Staphylococcus aureus, Pseudomonas aeruginosa and Acinetobacter baumannii*. The quaternary amine poly-diallyl-dimethylammonium chloride was attached to PU foam using a 'dip and dry' technique bonded by dehydration. The dressing exhibited good antibacterial properties and inhibited bacterial biofilm formation [134]. Zhou *et al.* synthesized an advanced wound dressing in which an encapsulated antimicrobial (sodium azide) was immobilized in vesicles contained in a nonwoven plasma-treated polypropylene mat [19]. The antimicrobial material was released when the vesicles came into contact with pathogenic bacteria. The pathogenic bacteria ruptured the vesicle; while on the other hand, nonpathogenic bacteria were inert to the vesicles. Bacteria-responsive vesicles were used to detect the infection in an early stage, by employing a dye (fluorescein) encapsulated in the vesicles. After vesicle rupture when it contacted the pathogenic bacteria, the wound dressing color changed, thus, the color change was an alert for bacterial infection [21]. Nanotechnology can assist in material design to endow a tunable antibacterial property to self-assembled polymers, by modification, nanoprecipitation and allow loading of antibacterial materials into the nanopolymer [135]. Figure 6 illustrates the mechanism of the bacterial-responsive wound dressing. In some cases, light-activated drugs (photosensitizers) can be used to eradicate the bacterial infection and overcome bacterial resistance; photodynamic inactivation produces ROS in a photochemical reaction between the excited state of the photosensitizer and ambient oxygen. These ROS are able to kill all known microbial cells without damaging the underlying tissue. The photosensitizers can be incorporated into various nanoparticles such as titania, chitosan and silver which can potentiate the photodynamic inactivation process to kill the microbes [136].

Oxygen-generating scaffolds have been used for tissue regeneration to improve cell proliferation, immune response and new collagen formation. Low levels of oxygen tension in the wound result from impaired perfusion typical of chronic ischemic wounds. A novel wound dressing based on oxygen-generating materials (sodium percarbonate/calcium peroxide) that delivered oxygen through microchannels in the dressing exhibited promising effects on wound healing such as re-epithelization, neovascularization and collagen reconstruction. The *in vivo* tests in a porcine model demonstrated rapid wound healing using these oxygen-generating materials [137–139].

Emerging advanced wound dressings

An adhesive wound dressing that is used postoperatively should have the correct adhesive properties to the wound surface, if it sticks too strongly to the wound it may damage the stratum corneum or healthy skin surrounding the wound, but if it sticks too weakly it is useless. In one study it was shown that self-adhesive PU foam with silicone adhesive produced less damage to the wound compared with hydrocolloid and PU foam with acrylic adhesive [140]. Yoon *et al.* synthesized sprayable wound dressings for diabetic wound healing based on a gelatin hydrogel in which chemokines were incorporated to attract host cells. [141]. The treated hydrogel exhibited better epithelialization, vascularization and collagen deposition. The control of acute hemorrhage is a critical issue, therefore Dowling *et al.* synthesized a sprayable foam to control bleeding without compression based on hydrophobically modified chitosan [142]. Annabi *et al.* synthesized sprayable, elastic and antimicrobial wound dressing based on methacryloylated elastin (GelMA) which could be used for suture-less wound closure to reduce infection and enhance chronic wound healing [143].

Conductive polymers (CPs) promote protein absorbance, and hence cell attachment, proliferation and migration on the substrate; all of which can be enhanced using conducting polymers [144]. Tissue-engineering scaffolds should imitate the normal cell behavior to provide the optimum interaction with tissue. Scaffolds that effectively mimic the biological properties of ECM combined with the ability to conduct electricity can guide tissue to regenerate properly [145–147]. Conductive/electroactive biocompatible scaffolds promote cell growth, proliferation and differentiation with or without ES [148]. CPs such as polyaniline, in addition to their conductivity, exhibit antibacterial and antimicrobial features [149,150]. CPs usually have been

utilized with biocompatible/biodegradable polymers such as chitosan [151]. Electroactive-engineered PUs based on PEG, castor oil, ricinoleic methyl ester and aniline oligomer combine conductivity along with tailored properties such as suitable water absorption and mechanical stability showed good antibacterial/antimicrobial features and promoted collagen deposition, vascularization and improved wound healing [152,153]. Electroactive injectable, self-healing hydrogels with antibacterial, AO and hemostasis features were synthesized based on chitosan/poly(ethylene glycol)-co-poly (glycerol sebacate) with tunable pore size, gel time, adhesiveness and stiffness, and promoted the wound healing process in a full-thickness skin defect model. Chitosan, due to its inherent nature, acts as an antibacterial, while the polymer exhibited a radical scavenging feature endowed with AO properties to the dressing. The major GFs involved in wound healing process are EGF, TGF- β and VEGF. The EGF plays a vital role in early stage of healing with fibroblast activation increment and joints dermal and epidermal intervals. ECM regulation and cell proliferation depend on TGF-β. VEGF controls pathways such as angiogenesis and re-epithelization. The expression of EGF, TGF-β and VEGF were enhanced utilizing electroactive substrates [154–157].

Photosensitive semi-conductive polymer NFs based on poly(*N*,*N*-bis(2-octyldodecyl)-3,6-di(thiophen-2-yl)-2,5-dihydropyrrolo[3,4-*c*]pyrrole-1,4-dione-alt-thieno[3,2-*b*]thiophene) and PCL showed low cytotoxicity and high optical absorbance in the red and NIR spectral region was utilized for evaluating the combination effect of electrical and light stimulation. The results showed better cell proliferation and collagen deposition which produced improved wound healing [158]. A collagen-elastin scaffold was used for skin regeneration. Pore size and cross-linking density had a significant effect on the scaffold function. Pore sizes around 80–100 μm showed the best effect on wound healing. A larger pore size around 120 μm caused more myofibroblast infiltration and presence of giant cells in a foreign body response. Vascularization and myofibroblast numbers showed no change with cross-linking density. However, increasing the cross-linking density resulted in lower proliferation of fibroblasts. Overall, a high cross-linking density decreased the wound-healing process [159].

Sericin is a protein produced by silkworms and together with fibroin makes up silk fibers. Sericin has excellent properties such as antibacterial, UV resistance, oxygen permeability which makes it useful for wound healing. Sericin/PVA/glycerin scaffolds were fabricated using a salt-leaching process which is a solvent-free and time/cost effective method. Compared with a lyophilized sample, the salt-leached sample had larger interconnected pores showing accelerated biodegradation and faster sericin release from the scaffold. Salt-leached scaffold had lower adhesion to the wound meaning its removal was straightforward without pain. The salt-leached sample exhibited better proliferation of fibroblast cells compared with the freeze-dried counterpart [160]. A gelatin/chitosan salt-leached scaffold was investigated in which human fibroblast dermal cells had been embedded in a tissue-engineering approach. The use of gelatin resulted in efficient cell attachment, proliferation and growth; moreover the interconnectivity and uniformity of the pores were beneficial for the wound healing process. *In vivo* evaluation showed that the scaffold increased epithelialization and skin regeneration [161].

Another fabrication method which has been used in wound dressing preparation is phase inversion. Supercritical carbon dioxide assisted phase inversion was used for preparation of poly(vinyl alcohol)-chitosan asymmetrical membranes. Tunable properties such as mechanical strength, hydrophilicity and morphology could be achieved by this technique. This dressing was capable of maintaining moisture levels, absorbing exudate, exchanging gases and preventing microorganism penetration [162]. Asymmetric polyurethane membranes can be fabricated using phase inversion. Compared with conventional commercial PU, asymmetric polyurethane membranes had better moisture maintenance, exudate absorption and gas circulation [163]. The laser excimer technique can create uniform morphology such as pore size/shape but the pore interconnectivity can be low [164].

Silk fibroin combined with PLGA was utilized to fabricate wound dressings via thermally induced phase separation which resulted in a highly porous scaffold with excellent pore interconnectivity, water uptake, moisture vapor transport rate and exudate absorbance [86]. The scaffold properties can be tailored by controlling the factors in the manufacturing process such as temperature, composition, concentration and quenching time [165]. A chitosan scaffold was fabricated by mechanical foaming and thermally induced phase separation with 120-μm pore size and tunable mechanical properties with a compressive modulus in the range of 2.6–25 kPa, thus it can be tailored for soft tissue engineering [166].

Lee *et al.* synthesized a hydrocolloid dressing by dissolution of styrene-isoprene-styrene mixed with carboxymethyl cellulose and silk fibroin nanoparticles [167]. Kim *et al.* synthesized an NF-based hydrocolloid dressing based on PU and carboxymethyl cellulose [168]. Jin *et al.* synthesized a hydrocolloid dressing via hot melting of styrene-isoprene-styrene mixed with poly-

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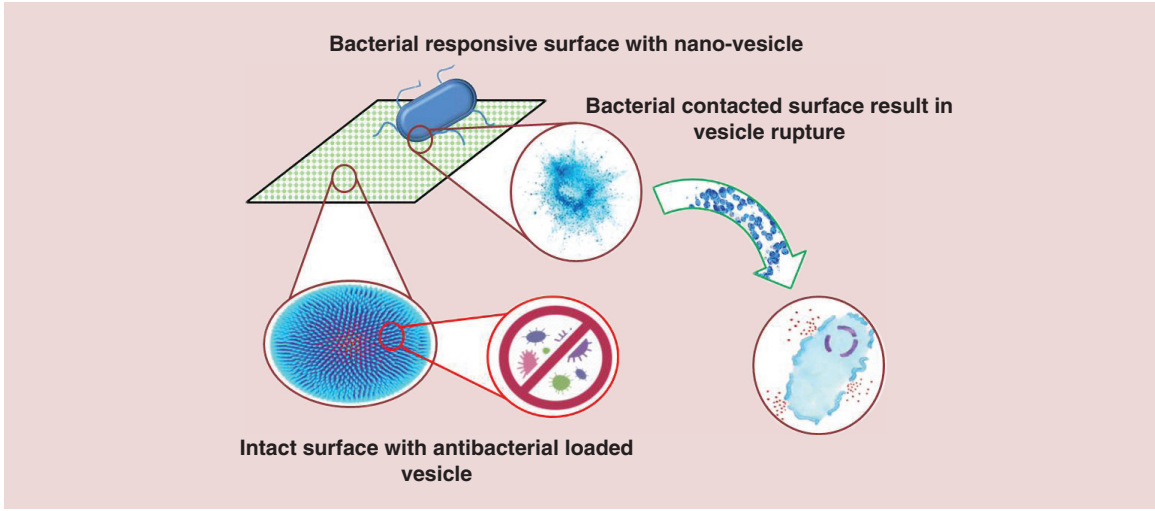

Figure 6. Bacterial-responsive nanostructured wound dressings.

isobutylene (and different types of polymers) providing better swelling, bio-adhesion and mechanical strength than a conventional sodium alginate mixture [169,170].

Bio-printing is a promising new technology that has recently attracted much attention because of its unique attributes. Bio-printed scaffolds can better mimic natural organs and tissues thus accelerating regeneration. Materials which are utilized in bio-printing should have proper biocompatibility and rheological properties to encapsulate cells and form a 3D structure. These materials are often called 'bio-ink'. Regarding these features, material choosing is limited. The majority of materials used as bio-inks are polymers, especially naturally occurring polymers such as collagen, alginate, hyaluronic acid, gelatin, chitosan, etc. [171]. Polypeptides are often used due to their intrinsic features such as the ability to tailor their mechanical properties, stimuli-responsive gelation and good biomimicking of the ECM, which makes them a suitable bio-ink [172]. Liu *et al.* fabricated a gelatin-alginate 3D bio-printed scaffold which promoted the wound healing process by increasing granulation and formation of scar tissue [173]. Ree *et al.* evaluated the features of a carboxymethylated-periodate nanocellulose 3D-printed dressing [174]. It was found that the rheological properties of carboxymethylated-periodate nanocellulose meant that it was a suitable bio-ink with additional antibacterial activity. Some bio-printed and solvent-cast chitosan/gelatin films were compared as a wound dressing. Bio-printed film had better mechanical strength because of the segmental alignment and closely packed polymer chains. On the other hand the water vapor transmission rate and water uptake of the cast film were higher because of its random structure which caused wound dehydration. Bio-printed film exhibited better bio-mimetic properties making it suitable for wound dressings [175].

Exosomes are cell-derived vesicles (diameter between 30 and 100 nm) that are released from cultured cells and can be recovered from the culture medium. Exosomes are released from the cell when multivesicular bodies fuse with the plasma membrane and contain various proteins, mRNA molecules and microRNAs. These matrix-bound nanovesicles were reported to be present in ECM-derived bioscaffolds. Matrix-bound nanovesicles can interact with host cells, causing macrophage activation and neuroblastoma cell differentiation [176]. MSCs implanted into wounds interact with the wound environment by paracrine interactions, promoting secretion of GFs and stimulation of host cells. They can accelerate wound healing by modulating inflammation, collagen regulation, differentiation, angiogenesis, etc. [177–179]. It was reported that exosomes released from SCs accelerated cutaneous wound by accelerating collagen deposition and angiogenesis [180]. MSCs seeded on silk fibroin-chitosan exhibited better wound healing by differentiation to fibrovascular, epithelial and endothelial cells [181]. Exosomes derived from SCs embedded in a chitosan scaffold exhibited a controlled release profile that enhanced epithelialization and angiogenesis [182].

Conclusion & future perspective

Skin has an important role in the body because it acts as a barrier against the external environment and protects the internal organs from water loss, infection and disease. Skin problems such as wounds and structural defects have attracted attention to allow the skin to regenerate as rapidly as possible without causing undesirable scarring. Skin defects occur because of genetic defects, diseases and traumatic accidents. Various energy-based modalities have been used for wounds such as laser and plasma therapy or the use of ES.

Table 2. Advanced wound dressing features from a nanotechnology perspective.

Advanced wound dressing	Advantage	Disadvantage	Features	Ref.
Nanofibers	High porosity, surface volume area	2D structure, low exudate absorbance	Aligned nanostructure promotes cell activity in various fabrication method. Loading different GF/ drugs	[93,99]
Hydrogels	3D structure, moist environment, controllable pore size to prevent the bacteria permeation and controlled release of biomolecules, low adhesion to wound, transparency of dressing through which the wound can be monitored without removing the dressing	Poor mechanical properties	Porosity tuning can be useful for wound repair. Interconnected nanoscale porosity enhances the cellular activity. Different type of nanoparticle can be embedded within hydrogel	[115,117,118]
PU	Adjustable properties, different fabrication types, tunable structure	Synthetic materials cause inflammation	Engineered polymer which can be fabricated in various morphologies to enhance. Self-assembly of soft segments and hard segments produces a nanosized structure	[126,127]
Conductive scaffolds	Promote cellular activity like attachment and proliferation. On-demand release	Cytotoxicity in high concentration, low degradability	Self-assembly like nano particle/ vesicle which is useful for controlled drug release	[147,154,183]
Cell-seeded advanced dressings	Enhancing the healing process, antimicrobial properties	Short storage time	Different type of cells can be seeded on dressing such as MSCs	[144]

GF: Growth factor; MSC: Mesenchymal stem cell; PU: Polyurethane.

Drug therapy *per se* has not been much used for wound healing, but biomolecules such as GFs can be topically applied for wounds provided a suitable delivery vehicle can be devised.

Regardless of which medical therapies are chosen, the exposed area of the wound should be covered with a suitable material generally called a 'wound dressing'. In this review, various wound dressings and their fabrication methods have been summarized. The most important factors that should be addressed in wound dressing design are biocompatibility, noncytotoxicity, mechanical strength (in both the wet and dry state), water uptake, water vapor evaporation rate, gaseous permeation ability, barrier against microorganisms, capacity to absorb exudate, intrinsic antibacterial/antimicrobial properties and appropriate levels of wound adhesion. The optimum wound dressing functionality is likely to combine different materials such as polymers and minerals. For example, polymers such as PU provide a proper environment for the wound, while minerals such as silver and ZnO provide antibacterial features to the dressing.

In recent years the nanotechnology revolution has impacted all areas of human life. The influence that nanotechnology has had on medical therapies has been loosely termed 'nanomedicine'. In the field of wound dressings, nanodimensional materials have special roles to play in several different areas. The high surface area to mass ratio characteristic of nanomaterials enables much better interaction with critical constituents in the wound milieu. Host cells naturally interact with ECM which can be better mimicked by nanoscale materials. Wound dressings must have the correct pore size to allow appropriate diffusion of both water and gases. A large part of effective wound dressings is prevention of infection and destruction of those microbial cells that have managed to penetrate to the wound, and it is well-known that nanoparticles have intrinsic antimicrobial activity. Smart stimulus-responsive nanosystems are a new innovation that will be increasingly applied to wound dressings. Enabling the wound dressing itself to detect and release what needs to be delivered to the wound to combat infection and enhance healing is a significant advance. Delivery of living cells to the wound is a new avenue of regenerative medicine. Undoubtedly, nanomedicine will have a critical role to play in the design and fabrication of wound dressings containing living cells. However, the cost of production and end pricing is an undeniable factor in commercial wound dressings. Advances in large-scale

10.2217/nnm-2017-0173

nanomanufacturing will continue, driven by the wide range of modern industrial applications, and these are expected to provide economic and high-quality wound dressings.

Financial & competing interests disclosure

MR Hamblin was supported by US NIH grants R01AI050875 and R21AI121700. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

No writing assistance was utilized in the production of this manuscript.

Executive summary

- Skin is the body's largest organ whose function as an external barrier is crucial to retain moisture, provide mechanical strength and prevent ingress of pathogenic microorganisms.
- Problematic skin defects with poor healing prognosis include ulcers (diabetic, venous and pressure) and burns (thermal and chemical).
- Various energy-based interventions, application of growth factors and even living cells can be used to stimulate wound healing.
- In cases where skin grafts are not possible, it is necessary to cover up the wound with a well-designed wound dressing to replace the skin's natural barrier.
- Fabricated wound dressings can take advantage of nano-engineered polymers and antimicrobial nanoparticles.
- The appropriate nanofeatures can provide biocompatibility, biodegradability and reduce toxicity of dressing materials.
- Naturally occurring polymers with the correct nanofeatures can better mimic the natural extracellular matrix allowing host cells to repair the wound.
- Well-designed nanofiber-based wound dressings can incorporate living cells to potentiate regenerative medicine.

References

Papers of special note have been highlighted as: • of interest; •• of considerable interest

1 Das, S, Baker AB. Biomaterials and nanotherapeutics for enhancing skin wound healing. *Front. Bioeng. Biotechnol.* 4, 82 (2016).

2 Clark R. *The Molecular and Cellular Biology of Wound Repair*. Springer Science & Business Media (2013).

3 Bryant R, Nix D. *Acute and Chronic Wounds*. Elsevier Health Sciences (2015).

4 Shin YC, Shin DM, Lee EJ *et al.* Hyaluronic acid/PLGA core/shell fiber matrices loaded with EGCG beneficial to diabetic wound healing. *Adv. Healthc. Mater.* 5(23), 3035–3045 (2016).

5 Moura LI, Dias AM, Leal EC, Carvalho L, De Sousa HC, Carvalho E. Chitosan-based dressings loaded with neurotensin – an efficient strategy to improve early diabetic wound healing. *Acta Biomater.* 10(2), 843–857 (2014).

6 Jones JE, Nelson EA, Al-Hity A. Skin grafting for venous leg ulcers. *Cochrane Database Syst. Rev.* (1), CD001737 (2013).

7 Läuchli S, Hafner J, Ostheeren S, Mayer D, Barysch MJ, French LE. Management of split-thickness skin graft donor sites: a randomized controlled trial of calcium alginate versus polyurethane film dressing. *Dermatology* 227(4), 361–366 (2013).

8 O'meara S, Martyn-St James M. Alginate dressings for venous leg ulcers. *Cochrane Library* (4), CD010182 (2015).

9 Marston WA, Sabolinski ML, Parsons NB, Kirsner RS. Comparative effectiveness of a bilayered living cellular construct and a porcine collagen wound dressing in the treatment of venous leg ulcers. *Wound Repair Regen.* 22(3), 334–340 (2014).

10 Stephen-Haynes J, Gibson E, Greenwood M. Chitosan: a natural solution for wound healing. *J. Commun. Nurs.* 28(1), 48–53 (2014).

11 Morimoto N, Yoshimura K, Niimi M *et al.* Novel collagen/gelatin scaffold with sustained release of basic fibroblast growth factor: clinical trial for chronic skin ulcers. *Tissue Eng. Pt A* 19(17–18), 1931–1940 (2013).

12 Srivastava RN, Dwivedi MK, Bhagat AK, Raj S, Agarwal R, Chandra A. A non-randomised, controlled clinical trial of an innovative device for negative pressure wound therapy of pressure ulcers in traumatic paraplegia patients†. *Int. Wound J.* 13(3), 343–348 (2014).

13 Lavery LA, La Fontaine J, Thakral G, Kim PJ, Bhavan K, Davis KE. Randomized clinical trial to compare negative-pressure wound therapy approaches with low and high pressure, silicone-coated dressing, and polyurethane foam dressing. *Plast. Reconstr. Surg.* 133(3), 722–726 (2014).

14 Young SR, Hampton S, Martin R. Non-invasive assessment of negative pressure wound therapy using high frequency diagnostic ultrasound: oedema reduction and new tissue accumulation. *Int. Wound J.* 10(4), 383–388 (2013).

15 Mohamad N, Amin MCIM, Pandey M, Ahmad N, Rajab NF. Bacterial cellulose/acrylic acid hydrogel synthesized via electron beam irradiation: accelerated burn wound healing in an animal model. *Carbohydr. Polym.* 114, 312–320 (2014).

16 Strong AL, Bennett DK, Spreen EB, Adhvaryu DV, Littleton JC, Mencer EJ. Fetal bovine collagen matrix in the treatment

of a full thickness burn wound: a case report with long-term follow-up. *J. Burn Care Res.* 37(3), e292 (2016).

17 Rezzadeh K, Zhou S, Hokugo A, Zhang Z, Segovia LA, Jarrahy R. Peptide amphiphilic (PA) gels enhance burn wound healing *in vitro* and *in vivo*. *J. Am. Coll. Surg.* 221(4), e121 (2015).

18 Andreu V, Mendoza G, Arruebo M, Irusta S. Smart dressings based on nanostructured fibers containing natural origin antimicrobial, anti-inflammatory, and regenerative compounds. *Materials* 8(8), 5154–5193 (2015).

19 Zhou J, Loftus AL, Mulley G, Jenkins ATA. A thin film detection/response system for pathogenic bacteria. *J. Am. Chem. Soc.* 132(18), 6566–6570 (2010).

•• **Dual function system that can both detect and destroy pathogenic bacteria but spare beneficial species.**

20 Said SS, El-Halfawy OM, El-Gowelli HM, Aloufy AK, Boraei NA, El-Khordagui LK. Bioburden-responsive antimicrobial PLGA ultrafine fibers for wound healing. *Eur. J. Pharm. Biopharm.* 80(1), 85–94 (2012).

21 Thet N, Alves D, Bean J *et al.* Prototype development of the intelligent hydrogel wound dressing and its efficacy in the detection of model pathogenic wound biofilms. *ACS Appl. Mater. Interfaces* 8(24), 14909–14919 (2015).

•• **An interesting study about a smart bacterial-responsive wound dressing.**

22 Greer N, Foman NA, Macdonald R *et al.* Advanced wound care therapies for nonhealing diabetic, venous, and arterial ulcers: a systematic review. *Ann. Intern. Med.* 159(8), 532–542 (2013).

23 Lopez-Pajares V, Yan K, Zarnegar BJ, Jameson KL, Khavari PA. Genetic pathways in disorders of epidermal differentiation. *Trends Genet.* 29(1), 31–40 (2013).

24 Gilchrest BA, Campisi J, Chang H, Fisher G, Kulesz-Martin M. Skin aging: molecular mechanisms and tissue consequences. *J. Invest. Dermatol.* 135(4), 950 (2015).

25 Ebaid H, Ahmed OM, Mahmoud AM, Ahmed RR. Limiting prolonged inflammation during proliferation and remodeling phases of wound healing in streptozotocin-induced diabetic rats supplemented with camel undenatured whey protein. *BMC Immunol.* 14(1), 1 (2013).

26 Leonida MD, Kumar I. Wound healing and skin regeneration. In: *Bionanomaterials for Skin Regeneration*. Springer, 17–25 (2016).

27 Watson T. Electrical stimulation for wound healing. *Phys. Ther. Rev.* 4, 10 (2013).

28 Cheing GLY, Li X, Huang L, Kwan RLC, Cheung KK. Pulsed electromagnetic fields (PEMF) promote early wound healing and myofibroblast proliferation in diabetic rats. *Bioelectromagnetics* 35(3), 161–169 (2014).

29 Yadollahpour A, Mostafa J, Samaneh R, Zohreh R. Ultrasound therapy for wound healing: a review of current techniques and mechanisms of action. *J. Pure Appl. Microbiol.* 8(5), 4071–4085 (2014).

30 Miyamoto K, Ikehara S, Sakakita H, Ikehara Y. Low temperature plasma equipment applied on surgical hemostasis and wound healings. *J. Clin. Biochem. Nutr.* 60(1), 25–28 (2017).

31 Aragão-Neto AC, Soares PA, Lima-Ribeiro MH, Carvalho EJ, Correia MT, Carneiro-Da-Cunha MG. Combined therapy using low level laser and chitosan-policaju hydrogel for wound healing. *Int. J. Biol. Macromol.* 95, 268–272 (2017).

32 Houreld NN. Shedding light on a new treatment for diabetic wound healing: a review on phototherapy. *ScientificWorldJournal* 2014, 398412 (2014).

33 Plichta JK, Holmes CJ, Gamelli RL, Radek KA. Local burn injury promotes defects in the epidermal lipid and antimicrobial peptide barriers in human autograft skin and burn margin: implications for burn wound healing and graft survival. *J. Burn Care Res.* 38(1), e212–e226 (2016).

34 Griseri T, Arnold IC, Pearson C *et al.* Granulocyte macrophage colony-stimulating factor-activated eosinophils promote interleukin-23 driven chronic colitis. *Immunity* 43(1), 187–199 (2015).

35 Shimizu K, Nakajima A, Sudo K *et al.* IL-1 receptor type 2 suppresses collagen-induced arthritis by inhibiting IL-1 signal on macrophages. *J. Immunol.* 194(7), 3156–3168 (2015).

36 Nanba D, Toki F, Barrandon Y, Higashiyama S. Recent advances in the epidermal growth factor receptor/ligand system biology on skin homeostasis and keratinocyte stem cell regulation. *J. Dermatol. Sci.* 72(2), 81–86 (2013).

37 Johnson KE, Wilgus TA. Vascular endothelial growth factor and angiogenesis in the regulation of cutaneous wound repair. *Adv. Wound Care* 3(10), 647–661 (2014).

38 Christmann RB, Sampaio-Barros P, Stifano G *et al.* Association of Interferon-and Transforming Growth Factor β–Regulated Genes and Macrophage Activation With Systemic Sclerosis–Related Progressive Lung Fibrosis. *Arthritis Rheumatol.* 66(3), 714–725 (2014).

39 Boateng JS, Matthews KH, Stevens HN, Eccleston GM. Wound healing dressings and drug delivery systems: a review. *J. Pharm. Sci.* 97(8), 2892–2923 (2008).

40 Sennett R, Wang Z, Rezza A *et al.* An integrated transcriptome atlas of embryonic hair follicle progenitors, their niche, and the developing skin. *Dev. Cell* 34(5), 577–591 (2015).

41 Plikus MV, Van Spyk EN, Pham K *et al.* The Circadian clock in skin implications for adult stem cells, tissue regeneration, cancer, aging, and immunity. *J. Biol. Rhythms* 30(3), 163–182 (2015).

42 Streckfuss-Bömeke K, Wolf F, Azizian A *et al.* Comparative study of human-induced pluripotent stem cells derived from bone marrow cells, hair keratinocytes, and skin fibroblasts. *Eur. Heart J.* 34(33), 2618–2629 (2013).

43 Murphy MB, Moncivais K, Caplan AI. Mesenchymal stem cells: environmentally responsive therapeutics for regenerative medicine. *Exp. Mol. Med.* 45(11), e54 (2013).

44 Condé-Green A, Marano AA, Lee ES *et al.* Fat grafting and adipose-derived regenerative cells in burn wound healing and scarring: a systematic review of the literature. *Plast. Reconstr. Surg.* 137(1), 302–312 (2016).

45 Antonyshyn JA, Fitzpatrick LE. Stem cell and stem cell-derived molecular therapies to enhance dermal wound

10.2217/nnm-2017-0173

healing. In: *Microscale Technologies for Cell Engineering*. Springer, 113–141 (2016).

46 Kumar A, Mohanty S, Nandy SB *et al.* Hair & skin derived progenitor cells: in search of a candidate cell for regenerative medicine. *Indian J. Med. Res.* 143(2), 175 (2016).

47 Hocking AM, Gibran NS. Mesenchymal stem cells: paracrine signaling and differentiation during cutaneous wound repair. *Exp. Cell Res.* 316(14), 2213–2219 (2010).

48 Blais M, Parenteau-Bareil R, Cadau S, Berthod F. Concise review: tissue-engineered skin and nerve regeneration in burn treatment. *Stem Cells Transl. Med.* 2(7), 545–551 (2013).

49 Kammeyer A, Luiten R. Oxidation events and skin aging. *Ageing Res. Rev.* 21, 16–29 (2015).

50 Makrantonaki E, Zouboulis CC. Characteristics and pathomechanisms of endogenously aged skin. *Dermatology* 214(4), 352–360 (2007).

51 Chung JH, Kang S, Varani J, Lin J, Fisher GJ, Voorhees JJ. Decreased extracellular-signal-regulated kinase and increased stress-activated MAP kinase activities in aged human skin *in vivo*. *J. Invest. Dermatol.* 115(2), 177–182 (2000).

52 Quan T, Shao Y, He T, Voorhees JJ, Fisher GJ. Reduced expression of connective tissue growth factor (CTGF/CCN2) mediates collagen loss in chronologically aged human skin. *J. Invest. Dermatol.* 130(2), 415–424 (2010).

53 Kohl E, Steinbauer J, Landthaler M, Szeimies RM. Skin ageing. *J. Eur. Acad. Dermatol. Venereol.* 25(8), 873–884 (2011).

54 Klotz L-O, Holbrook N, Sies H. UVA and singlet oxygen as inducers of cutaneous signaling events. In: *Oxidants and Antioxidants in Cutaneous Biology*. Karger Publishers, 95–113 (2001).

55 Kohen R. Skin antioxidants: their role in aging and in oxidative stress—new approaches for their evaluation. *Biomed. Pharmacother.* 53(4), 181–192 (1999).

56 Martin A. The use of antioxidants in healing. *Dermatol. Surg.* 22(2), 156–160 (1996).

57 Fitzmaurice S, Sivamani R, Isseroff R. Antioxidant therapies for wound healing: a clinical guide to currently commercially available products. *Skin Pharmacol. Physiol.* 24(3), 113–126 (2011).

58 Abla M, Banga A. Formulation of tocopherol nanocarriers and *in vitro* delivery into human skin. *Int. J. Cosmetic Sci.* 36(3), 239–246 (2014).

59 Hinek A, Kim HJ, Wang Y, Wang A, Mitts TF. Sodium l-ascorbate enhances elastic fibers deposition by fibroblasts from normal and pathologic human skin. *J. Dermatol. Sci.* 75(3), 173–182 (2014).

60 Holley AK, Xu Y, Noel T, Bakthavatchalu V, Batinic-Haberle I, St. Clair DK. Manganese superoxide dismutase-mediated inside-out signaling in HaCaT human keratinocytes and SKH-1 mouse skin. *Antioxidants Redox Signal.* 20(15), 2347–2360 (2014).

61 Mcloone P, Warnock M, Fyfe L. Honey: a realistic antimicrobial for disorders of the skin. *J. Microbiol. Immunol. Infect.* 49(2), 161–167 (2016).

62 Dos Santos L, Soto CaT, Favero PP, Martin AA. *In vivo* confocal Raman spectroscopy study of the vitamin A derivative perfusion through human skin. Presented at: *SPIE Biomedical Vibrational Spectroscopy 2016: Advances in Research and Industry*. CA, USA, 13–14 February 2016.

63 Sheng X, Fan L, He C, Zhang K, Mo X, Wang H. Vitamin E-loaded silk fibroin nanofibrous mats fabricated by green process for skin care application. *Int. J. Biol. Macromol.* 56, 49–56 (2013).

64 Zhou W, Liu W, Zou L *et al.* Storage stability and skin permeation of vitamin C liposomes improved by pectin coating. *Colloids Surf. B. Biointerfaces* 117, 330–337 (2014).

65 Caini S, Boniol M, Tosti G *et al.* Vitamin D and melanoma and non-melanoma skin cancer risk and prognosis: a comprehensive review and meta-analysis. *Eur. J. Cancer* 50(15), 2649–2658 (2014).

66 Mirastschijski U, Martin A, Jorgensen LN, Sampson B, Ågren MS. Zinc, copper, and selenium tissue levels and their relation to subcutaneous abscess, minor surgery, and wound healing in humans. *Biol. Trace Elem. Res.* 153(1–3), 76–83 (2013).

67 Caló E, Khutoryanskiy VV. Biomedical applications of hydrogels: a review of patents and commercial products. *Eur. Polym. J.* 65, 252–267 (2015).

68 Edmonds M. Apligraf in the treatment of neuropathic diabetic foot ulcers. *The Int. J. Lower Extrem. Wounds* 8(1), 11–18 (2009).

69 Hamblin MR, De Sousa MVP, Agrawal T. *Handbook of Low-Level Laser Therapy*. Pan Stanford Publishing, Singapore (2017).

• **Comprehensive textbook giving state-of-the-art overview of photobiomodulation or low-level laser therapy.**

70 Noda M, Aoki A, Mizutani K *et al.* High-frequency pulsed low-level diode laser therapy accelerates wound healing of tooth extraction socket: an *in vivo* study. *Lasers Surg. Med.* 48(10), 955–964 (2016).

71 De Freitas LF, Hamblin MR. Proposed mechanisms of photobiomodulation or low-level light therapy. *IEEE J. Sel. Top. Quantum Electron.* 22(3), pii:7000417 (2016).

72 Hamblin MR. History of low-level laser (light) therapy. In: *Handbook of Low-Level Laser Therapy*. Hamblin MR, De Sousa MVP, Agrawal T (Eds). Pan Stanford Publishing, Singapore (2017).

73 Anders JJ, Lanzafame RJ, Arany PR. Low-level light/laser therapy versus photobiomodulation therapy. *Photomed. Laser Surg.* 33(4), 183–184 (2015).

74 Dawood MS, Salman SD. Low level diode laser accelerates wound healing. *Lasers Med. Sci.* 28(3), 941–945 (2013).

75 Silveira PC, Ferreira KB, Da Rocha FR *et al.* Effect of Low-Power Laser (LPL) and Light-Emitting Diode (LED) on Inflammatory Response in Burn Wound Healing. *Inflammation* 39(4), 1395–1404 (2016).

76 Ennis WJ, Lee C, Gellada K, Corbiere TF, Koh TJ. Advanced technologies to improve wound healing: electrical stimulation, vibration therapy, and ultrasound—what is the evidence?*Plast. Reconstr. Surg.* 138(3S), S94–S104 (2016).

77 Akiyama H, Hachiya Y, Otsuka H *et al.* Low-intensity pulsed ultrasound therapy stimulates callus formation

between host femur and cortical onlay strut allograft. *Ultrasound Med. Biol.* 40(6), 1197–1203 (2014).

78 Iwanabe Y, Masaki C, Tamura A *et al.* The effect of low-intensity pulsed ultrasound on wound healing using scratch assay in epithelial cells. *J. Prosthodont Res* 60(4), 308–314 (2016).

79 Fantinati MS, Mendonça DEO, Fantinati AMM *et al.* Low intensity ultrasound therapy induces angiogenesis and persistent inflammation in the chronic phase of the healing process of third degree burn wounds experimentally induced in diabetic and non-diabetic rats. *Acta Cir. Bras.* 31(7), 463–471 (2016).

80 Mesquita RL, Silva PICD, Silva SHSDM *et al.* Effect of low-intensity therapeutic ultrasound on wound healing in rats subjected to third-degree burns. *Acta Cir. Bras.* 31(1), 36–43 (2016).

81 Shrivastava SK, Kailash. Shock wave treatment in medicine. *J. Biosci.* 30(2), 269–275 (2005).

82 Arno A, Garcia O, Hernan I, Sancho J, Acosta A, Barret JP. Extracorporeal shock waves, a new non-surgical method to treat severe burns. *Burns* 36(6), 844–849 (2010).

83 Jeppesen SM, Yderstraede KB, Rasmussen BS, Hanna M, Lund L. Extracorporeal shockwave therapy in the treatment of chronic diabetic foot ulcers: a prospective randomised trial. *J. Wound Care* 25(11), 641–649 (2016).

84 Thakral G, Lafontaine J, Najafi B, Talal TK, Kim P, Lavery LA. Electrical stimulation to accelerate wound healing. *Diab. Foot Ankle* 4, 10 (2013).

85 Rouabhia M, Park H, Meng S, Derbali H, Zhang Z. Electrical stimulation promotes wound healing by enhancing dermal fibroblast activity and promoting myofibroblast transdifferentiation. *PLoS ONE* 8(8), e71660 (2013).

86 Liu Y, Sun Q, Wang S *et al.* Studies of silk fibroin/poly (lactic-co-glycolic acid) scaffold, prepared by thermally induced phase separation, as a possible wound dressing. *Sci. Adv. Mater.* 8(5), 1045–1052 (2016).

87 Kloth LC. Electrical stimulation technologies for wound healing. *Adv. Wound Care* 3(2), 81–90 (2014).

88 Schmidt A, Bekeschus S, Wende K, Vollmar B, Woedtke T. A cold plasma jet accelerates wound healing in a murine model of full-thickness skin wounds. *Exp. Dermatol.* 26(2), 156–162 (2016).

89 Heinlin J, Isbary G, Stolz W *et al.* Plasma applications in medicine with a special focus on dermatology. *J. Eur. Acad. Dermatol. Venereol.* 25(1), 1–11 (2011).

90 Tiede R, Hirschberg J, Daeschlein G, Von Woedtke T, Vioel W, Emmert S. Plasma applications: a dermatological view. *Contrib. Plasm. Phys.* 54(2), 118–130 (2014).

91 Fathollah S, Mirpour S, Mansouri P *et al.* Investigation on the effects of the atmospheric pressure plasma on wound healing in diabetic rats. *Sci. Rep.* 6, 19144 (2016).

92 Xu G, Cai J, Chen S *et al.* Effects of low temperature atmospheric pressure plasma on skin wound healing of mice *in vivo*. Presented at: *Plasma Science (ICOPS), 2016 IEEE International Conference on Plasma Science (ICOPS)*. Budapest, Hungary, 9–12 October 2016 (doi:10.1109/PLASMA.2016.7534233).

93 Van Zuijlen PP, Ruurda JJ, Van Veen HA *et al.* Collagen morphology in human skin and scar tissue: no adaptations in response to mechanical loading at joints. *Burns* 29(5), 423–431 (2003).

94 Hu W, Huang ZM. Biocompatibility of braided poly (L-lactic acid) nanofiber wires applied as tissue sutures. *Polym. Int.* 59(1), 92–99 (2010).

95 Xie J, Ma B, Michael PL. Fabrication of novel 3D nanofiber scaffolds with anisotropic property and regular pores and their potential applications. *Adv. Healthc. Mater.* 1(5), 674–678 (2012).

96 Chen S, Liu B, Carlson MA, Gombart AF, Reilly DA, Xie J. Recent advances in electrospun nanofibers for wound healing. *Nanomedicine* 12(11), 1335–1352 (2017).

97 Padmakumar S, Joseph J, Neppalli MH *et al.* Electrospun polymeric core–sheath yarns as drug eluting surgical sutures. *ACS Appl. Mater. Interfaces* 8(11), 6925–6934 (2016).

98 Sell S, Barnes C, Smith M *et al.* Extracellular matrix regenerated: tissue engineering via electrospun biomimetic nanofibers. *Polym. Int.* 56(11), 1349–1360 (2007).

99 Said SS, Aloufy AK, El-Halfawy OM, Boraei NA, El-Khordagui LK. Antimicrobial PLGA ultrafine fibers: interaction with wound bacteria. *Eur. J. Pharm. Biopharm.* 79(1), 108–118 (2011).

100 Sun X, Cheng L, Zhao J *et al.* bFGF-grafted electrospun fibrous scaffolds via poly (dopamine) for skin wound healing. *J. Mater. Chem. B* 2(23), 3636–3645 (2014).

101 Peh P, Lim NSJ, Blocki A *et al.* Simultaneous delivery of highly diverse bioactive compounds from blend electrospun fibers for skin wound healing. *Bioconj. Chem.* 26(7), 1348–1358 (2015).

102 Augustine R, Dominic EA, Reju I, Kaimal B, Kalarikkal N, Thomas S. Electrospun polycaprolactone membranes incorporated with ZnO nanoparticles as skin substitutes with enhanced fibroblast proliferation and wound healing. *RSC Adv.* 4(47), 24777–24785 (2014).

103 Das S, Singh S, Dowding JM *et al.* The induction of angiogenesis by cerium oxide nanoparticles through the modulation of oxygen in intracellular environments. *Biomaterials* 33(31), 7746–7755 (2012).

- **Cerium oxide nanoparticles can increase oxygen in wounds to stimulate healing.**

104 Kalashnikova I, Das S, Seal S. Nanomaterials for wound healing: scope and advancement. *Nanomedicine* 10(16), 2593–2612 (2015).

105 Unnithan AR, Sasikala ARK, Sathishkumar Y, Lee YS, Park CH, Kim CS. Nanoceria doped electrospun antibacterial composite mats for potential biomedical applications. *Ceram. Int.* 40(8), 12003–12012 (2014).

106 Abdelgawad AM, Hudson SM, Rojas OJ. Antimicrobial wound dressing nanofiber mats from multicomponent (chitosan/silver-NPs/polyvinyl alcohol) systems. *Carbohydr. Polym.* 100, 166–178 (2014).

107 Li C, Fu R, Yu C *et al.* Silver nanoparticle/chitosan oligosaccharide/poly (vinyl alcohol) nanofibers as wound dressings: a preclinical study. *Int. J. Nanomed.* 8, 4131 (2013).

108 Wu J, Zheng Y, Song W *et al. In situ* synthesis of silver-nanoparticles/bacterial cellulose composites for slow-released antimicrobial wound dressing. *Carbohydr. Polym.* 102, 762–771 (2014).

109 Burke L, Mortimer CJ, Curtis DJ *et al. In situ* synthesis of magnetic iron-oxide nanoparticle-nanofibre composites using electrospinning. *Mater. Sci. Eng. C* 70, 512–519 (2017).

110 Chen WY, Chang HY, Lu JK *et al.* Self-assembly of antimicrobial peptides on gold nanodots: against multidrug-resistant bacteria and wound-healing application. *Adv. Funct. Mater.* 25(46), 7189–7199 (2015).

111 Dunn LL, De Valence S, Tille JC *et al.* Biodegradable and plasma-treated electrospun scaffolds coated with recombinant Olfactomedin-like 3 for accelerating wound healing and tissue regeneration. *Wound Repair Regen.* 24(6), 1030–1035 (2016).

112 Leong WS, Wu SC, Ng K, Tan LP. Electrospun 3D multi-scale fibrous scaffold for enhanced human dermal fibroblast infiltration. *Int. J. Bioprint.* 2(1), doi:http://dx.doi.org/10.18063/IJB.2016.01.002 (2016).

113 Tartarini D, Mele E. Adult stem cell therapies for wound healing: biomaterials and computational models. *Front. Bioeng. Biotechnol.* 3, 206 (2015).

114 Jackson WM, Nesti LJ, Tuan RS. Concise review: clinical translation of wound healing therapies based on mesenchymal stem cells. *Stem Cells Transl. Med.* 1(1), 44–50 (2012).

115 Madaghiele M, Demitri C, Sannino A, Ambrosio L. Polymeric hydrogels for burn wound care: advanced skin wound dressings and regenerative templates. *Burns Trauma* 2(4), 153 (2014).

116 Zeltinger J, Sherwood JK, Graham DA, Müeller R, Griffith LG. Effect of pore size and void fraction on cellular adhesion, proliferation, and matrix deposition. *Tissue Eng.* 7(5), 557–572 (2001).

117 Sudheesh Kumar P, Lakshmanan V-K, Anilkumar T *et al.* Flexible and microporous chitosan hydrogel/nano ZnO composite bandages for wound dressing: *in vitro* and *in vivo* evaluation. *ACS Appl. Mater. Interfaces* 4(5), 2618–2629 (2012).

118 Loo Y, Wong Y-C, Cai EZ *et al.* Ultrashort peptide nanofibrous hydrogels for the acceleration of healing of burn wounds. *Biomaterials* 35(17), 4805–4814 (2014).

119 Paladini F, Meikle S, Cooper I, Lacey J, Perugini V, Santin M. Silver-doped self-assembling di-phenylalanine hydrogels as wound dressing biomaterials. *J. Mater. Sci. Mater. Med.* 24(10), 2461–2472 (2013).

120 Seow WY, Salgado G, Lane EB, Hauser CA. Transparent crosslinked ultrashort peptide hydrogel dressing with high shape-fidelity accelerates healing of full-thickness excision wounds. *Sci. Rep.* 6, 32670 (2016).

121 Fox SJ, Fazil MHUT, Dhand C *et al.* Insight into membrane selectivity of linear and branched polyethylenimines and their potential as biocides for advanced wound dressings. *Acta Biomater.* 37, 155–164 (2016).

122 Goh M, Hwang Y, Tae G. Epidermal growth factor loaded heparin-based hydrogel sheet for skin wound healing. *Carbohydr. Polym.* 147, 251–260 (2016).

• **The growth factor, EGF, can be loaded into a flexible sheet for a wound-healing dressing.**

123 Peles Z, Zilberman M. Novel soy protein wound dressings with controlled antibiotic release: mechanical and physical properties. *Acta Biomater.* 8(1), 209–217 (2012).

124 Chung HJ, Park TG. Self-assembled and nanostructured hydrogels for drug delivery and tissue engineering. *Nano Today* 4(5), 429–437 (2009).

125 Obara K, Ishihara M, Ishizuka T *et al.* Photocrosslinkable chitosan hydrogel containing fibroblast growth factor-2 stimulates wound healing in healing-impaired db/db mice. *Biomaterials* 24(20), 3437–3444 (2003).

126 Rokicki G, Parzuchowski PG, Mazurek M. Non-isocyanate polyurethanes: synthesis, properties, and applications. *Polym. Adv. Technol.* 26(7), 707–761 (2015).

127 Shams E, Yeganeh H, Naderi-Manesh H, Gharibi R, Hassan ZM. Polyurethane/siloxane membranes containing graphene oxide nanoplatelets as antimicrobial wound dressings: *in vitro* and *in vivo* evaluations. *J. Mater. Sci. Mater. Med.* 28(5), 75 (2017).

128 Woo H-D, Park K-T, Kim E-H *et al.* Preparation of UV-curable gelatin derivatives for drug immobilization on polyurethane foam: development of wound dressing foam. *Macromolecular Res.* 23(11), 994–1003 (2015).

129 Liu X, Niu Y, Chen KC, Chen S. Rapid hemostatic and mild polyurethane-urea foam wound dressing for promoting wound healing. *Mater. Sci. Eng. C Mater. Biol. Appl.* 71, 289–297 (2016).

130 Ulery BD, Nair LS, Laurencin CT. Biomedical applications of biodegradable polymers. *J. Polym. Sci. B Polym. Phys.* 49(12), 832–864 (2011).

131 Lee J-M, Kim S-H, Jeong H-Y *et al.* Preparation and characterization of polyurethane foam using a PLA/PEG polyol mixture. *Fibers Polymers* 15(7), 1349–1356 (2014).

132 París R, Marcos-Fernández Á, Quijada-Garrido I. Synthesis and characterization of poly (ethylene glycol)-based thermo-responsive polyurethane hydrogels for controlled drug release. *Polym. Adv. Technol.* 24(12), 1062–1067 (2013).

133 Lundin JG, Daniels GC, Mcgann CL *et al.* Multi-functional polyurethane hydrogel foams with tunable mechanical properties for wound dressing applications. *Macromol. Mater. Eng.* 302(3), 1600375 (2017).

134 Tran PL, Hamood AN, Souza A *et al.* A study on the ability of quaternary ammonium groups attached to a polyurethane foam wound dressing to inhibit bacterial attachment and biofilm formation. *Wound Repair Regen.* 23(1), 74–81 (2015).

135 Muñoz-Bonilla A, Fernández-García M. The roadmap of antimicrobial polymeric materials in macromolecular nanotechnology. *Eur. Polym. J.* 65, 46–62 (2015).

• **A review of bacteriostatic polymers and nanotechnology used in various applications.**

136 Yin R, Agrawal T, Khan U *et al.* Antimicrobial photodynamic inactivation in nanomedicine: small light strides against bad bugs. *Nanomedicine* 10(15), 2379–2404 (2015).

137 Chandra PK, Ross CL, Smith LC *et al.* Peroxide-based oxygen generating topical wound dressing for enhancing

healing of dermal wounds. *Wound Repair Regen.* 23(6), 830–841 (2015).

138 Li Z, Roussakis E, Koolen PG *et al.* Non-invasive transdermal two-dimensional mapping of cutaneous oxygenation with a rapid-drying liquid bandage. *Biomed. Optics Express* 5(11), 3748–3764 (2014).

139 Lo JF, Brennan M, Merchant Z *et al.* Microfluidic wound bandage: localized oxygen modulation of collagen maturation. *Wound Repair Regen.* 21(2), 226–234 (2013).

140 Matsumura H, Imai R, Ahmatjan N *et al.* Removal of adhesive wound dressing and its effects on the stratum corneum of the skin: comparison of eight different adhesive wound dressings. *Int. Wound J.* 11(1), 50–54 (2014).

141 Yoon DS, Lee Y, Ryu HA *et al.* Cell recruiting chemokine-loaded sprayable gelatin hydrogel dressings for diabetic wound healing. *Acta Biomater.* 38, 59–68 (2016).

•• **A study about a convenient method for sprayable wound dressings formation.**

142 Dowling MB, Macintire IC, White JC *et al.* Sprayable foams based on an amphiphilic biopolymer for control of hemorrhage without compression. *ACS Biomater. Sci. Eng.* 1(6), 440–447 (2015).

143 Annabi N, Rana D, Sani ES *et al.* Engineering a sprayable and elastic hydrogel adhesive with antimicrobial properties for wound healing. *Biomaterials* 139, 229–243 (2017).

144 Wong JY, Langer R, Ingber DE. Electrically conducting polymers can noninvasively control the shape and growth of mammalian cells. *Proc. Natl Acad. Sci. USA* 91(8), 3201–3204 (1994).

145 Murali R, Thanikaivelan P. Bionic, porous, functionalized hybrid scaffolds with vascular endothelial growth factor promote rapid wound healing in Wistar albino rats. *RSC Adv.* 6(23), 19252–19264 (2016).

146 Pramanik N, Dutta K, Basu RK, Kundu PP. Aromatic Π-conjugated curcumin on surface modified polyaniline/polyhydroxyalkanoate based 3D porous scaffolds for tissue engineering applications. *ACS Biomater. Sci. Eng.* 2(12), 2365–2377 (2016).

147 Atoufi Z, Zarrintaj P, Motlagh GH, Amiri A, Bagher Z, Kamrava SK. A novel bio electro active alginate-aniline tetramer/agarose scaffold for tissue engineering: synthesis, characterization, drug release and cell culture study. *J. Biomater. Sci. Polym. Ed.* 28(15), 1617–1638 (2017).

148 Balint R, Cassidy NJ, Cartmell SH. Conductive polymers: towards a smart biomaterial for tissue engineering. *Acta Biomater.* 10(6), 2341–2353 (2014).

149 Shalini A, Nishanthi R, Palani P, Jaisankar V. One pot synthesis, characterization of polyaniline and cellulose/polyaniline nanocomposites: application towards *in vitro* measurements of antibacterial activity. *Mater. Today Proc.* 3(6), 1633–1642 (2016).

150 Soleimani M, Ghorbani M, Salahi S. Antibacterial activity of polypyrrole-chitosan nanocomposite: mechanism of action. *Int. J. Nanosci. Nanotechnol.* 12(3), 191–197 (2016).

151 Singh BK, Dutta PK. Chitin, chitosan, and silk fibroin electrospun nanofibrous scaffolds: a prospective approach for regenerative medicine. In: *Chitin and Chitosan for Regenerative Medicine*. Springer, 151–189 (2016).

152 Gharibi R, Yeganeh H, Rezapour-Lactoee A, Hassan ZM. Stimulation of wound healing by electroactive, antibacterial, and antioxidant polyurethane/siloxane dressing membranes: *in vitro* and *in vivo* evaluations. *ACS Appl. Mater. Interfaces* 7(43), 24296–24311 (2015).

• **A study using a multifunctional conductive polymer with electrical stimulation for wound healing.**

153 Gharibi R, Yeganeh H, Gholami H, Hassan ZM. Aniline tetramer embedded polyurethane/siloxane membranes and their corresponding nanosilver composites as intelligent wound dressing materials. *RSC Adv.* 4(107), 62046–62060 (2014).

154 Zhao X, Wu H, Guo B, Dong R, Qiu Y, Ma PX. Antibacterial anti-oxidant electroactive injectable hydrogel as self-healing wound dressing with hemostasis and adhesiveness for cutaneous wound healing. *Biomaterials* 122, 34–47 (2017).

155 Barrientos S, Stojadinovic O, Golinko MS, Brem H, Tomic-Canic M. Growth factors and cytokines in wound healing. *Wound Repair Regen.* 16(5), 585–601 (2008).

156 Wilson SE, Chen L, Mohan RR, Liang Q, Liu J. Expression of HGF, KGF, EGF and receptor messenger RNAs following corneal epithelial wounding. *Exp. Eye Res.* 68(4), 377–397 (1999).

157 Nissen NN, Polverini P, Koch AE, Volin MV, Gamelli RL, Dipietro LA. Vascular endothelial growth factor mediates angiogenic activity during the proliferative phase of wound healing. *Am. J. Pathol.* 152(6), 1445 (1998).

158 Jin G, Li J, Li K. Photosensitive semiconducting polymer-incorporated nanofibers for promoting the regeneration of skin wound. *Mater. Sci. Eng. C.* 70(2), 1176–1181 (2017).

159 Boekema BK, Vlig M, Damink LO *et al.* Effect of pore size and cross-linking of a novel collagen-elastin dermal substitute on wound healing. *J. Mater. Sci. Mater. Med.* 25(2), 423–433 (2014).

160 Aramwit P, Ratanavaraporn J, Ekgasit S, Tongsakul D, Bang N. A green salt-leaching technique to produce sericin/PVA/glycerin scaffolds with distinguished characteristics for wound-dressing applications. *J. Biomed. Mater. Res. B Appl. Biomater.* 103(4), 915–924 (2015).

161 Pezeshki-Modaress M, Rajabi-Zeleti S, Zandi M *et al.* Cell-loaded gelatin/chitosan scaffolds fabricated by salt-leaching/lyophilization for skin tissue engineering: *in vitro* and *in vivo* study. *J. Biomed. Mater. Res. A* 102(11), 3908–3917 (2014).

162 Morgado PI, Lisboa PF, Ribeiro MP *et al.* Poly (vinyl alcohol)/chitosan asymmetrical membranes: highly controlled morphology toward the ideal wound dressing. *J. Membr. Sci.* 469, 262–271 (2014).

163 Xu H, Chang J, Chen Y, Fan H, Shi B. Asymmetric polyurethane membrane with inflammation-responsive antibacterial activity for potential wound dressing application. *J. Mater. Sci.* 48(19), 6625–6639 (2013).

164 Leonida MD, Kumar I. Nanomaterials, scaffolds, and skin tissue regeneration. In: *Bionanomaterials for Skin Regeneration*. Springer, 103–116 (2016).

10.2217/nnm-2017-0173

165 Akbarzadeh R, Yousefi AM. Effects of processing parameters in thermally induced phase separation technique on porous architecture of scaffolds for bone tissue engineering. *J. Biomed. Mater. Res. B Appl. Biomater.* 102(6), 1304–1315 (2014).

166 Biswas D, Tran P, Tallon C, O'connor A. Combining mechanical foaming and thermally induced phase separation to generate chitosan scaffolds for soft tissue engineering. *J. Biomater. Sci. Polym. Ed.* 28(2), 207–226 (2016).

167 Lee OJ, Kim J-H, Moon BM *et al.* Fabrication and characterization of hydrocolloid dressing with silk fibroin nanoparticles for wound healing. *Tissue Eng. Regen. Med.* 13(3), 218–226 (2016).

168 Kim S, Park SG, Kang SW, Lee KJ. Nanofiber-based hydrocolloid from colloid electrospinning toward next generation wound dressing. *Macromol. Mater. Eng.* 301(7), 818–826 (2016).

169 Jin SG, Kim KS, Yousaf AM *et al.* Mechanical properties and *in vivo* healing evaluation of a novel Centella asiatica-loaded hydrocolloid wound dressing. *Int. J. Pharm.* 490(1), 240–247 (2015).

170 Jin SG, Yousaf AM, Kim KS *et al.* Influence of hydrophilic polymers on functional properties and wound healing efficacy of hydrocolloid based wound dressings. *Int. J. Pharm.* 501(1), 160–166 (2016).

171 Carrow JK, Kerativitayanan P, Jaiswal MK, Lokhande G, Gaharwar AK. Polymers for Bioprinting. In: *Essentials of 3D Biofabrication and Translation*. Atala A, Yoo JJ (Eds). Academic Press, 229–248 (2015).

172 Loo Y, Hauser CA. Bioprinting synthetic self-assembling peptide hydrogels for biomedical applications. *Biomedical Materials* 11(1), 014103 (2015).

173 Liu J, Chi J, Wang K, Liu X, Liu J, Gu F. Full-thickness wound healing using 3D bioprinted gelatin-alginate scaffolds in mice: a histopathological study. *Int. J. Clin. Exp. Pathol.* 9(11), 11197–11205 (2016).

174 Rees A, Powell LC, Chinga-Carrasco G *et al.* 3D bioprinting of carboxymethylated-periodate oxidized nanocellulose constructs for wound dressing applications. *BioMed. Res. Int.* 2015, 925757 (2015).

175 Ng WL, Yeong WY, Naing MW. Potential of bioprinted films for skin tissue engineering. doi:10.3850/978-981-09-0446-3_065 (2014).

176 Huleihel L, Hussey GS, Naranjo JD *et al.* Matrix-bound nanovesicles within ECMbioscaffolds. *Sci. Adv.*2(6), e1600502 (2016).

177 Maxson S, Lopez EA, Yoo D, Danilkovitch-Miagkova A, Leroux MA. Concise review: role of mesenchymal stem cells in wound repair. *Stem Cells Transl. Med.* 1(2), 142–149 (2012).

•• **A comprehensive review about the role of mesenchymal stem cells (MSCs) in wound repair.**

178 Wu Y, Chen L, Scott PG, Tredget EE. Mesenchymal stem cells enhance wound healing through differentiation and angiogenesis. *Stem Cells* 25(10), 2648–2659 (2007).

179 Lee DE, Ayoub N, Agrawal DK. Mesenchymal stem cells and cutaneous wound healing: novel methods to increase cell delivery and therapeutic efficacy. *Stem Cell. Res. Ther.* 7(1), 37 (2016).

180 Zhang J, Guan J, Niu X *et al.* Exosomes released from human induced pluripotent stem cells-derived MSCs facilitate cutaneous wound healing by promoting collagen synthesis and angiogenesis. *J. Transl. Med.* 13(1), 49 (2015).

• **One of the first reports showing that exosomes from MSCs can stimulate wound healing.**

181 Altman AM, Yan Y, Matthias N *et al.* IFATS collection: human adipose-derived stem cells seeded on a silk fibroin-chitosan scaffold enhance wound repair in a murine soft tissue injury model. *Stem Cells* 27(1), 250–258 (2009).

182 Tao SC, Guo SC, Li M, Ke QF, Guo YP, Zhang CQ. Chitosan wound dressings incorporating exosomes derived from microrna-126-overexpressing synovium mesenchymal stem cells provide sustained release of exosomes and heal full-thickness skin defects in a diabetic rat model. *Stem Cells Transl. Med.* 6(3), 736–747 (2017).

•• **Shows that exosomes from MSCs expressing microRNA-126 can be incorporated into a wound dressing that can stimulate diabetic wound healing.**

183 Kim H, Jeong S-M, Park J-W. Electrical switching between vesicles and micelles via redox-responsive self-assembly of amphiphilic rod–coils. *J. Am. Chem. Soc.* 133(14), 5206–5209 (2011).