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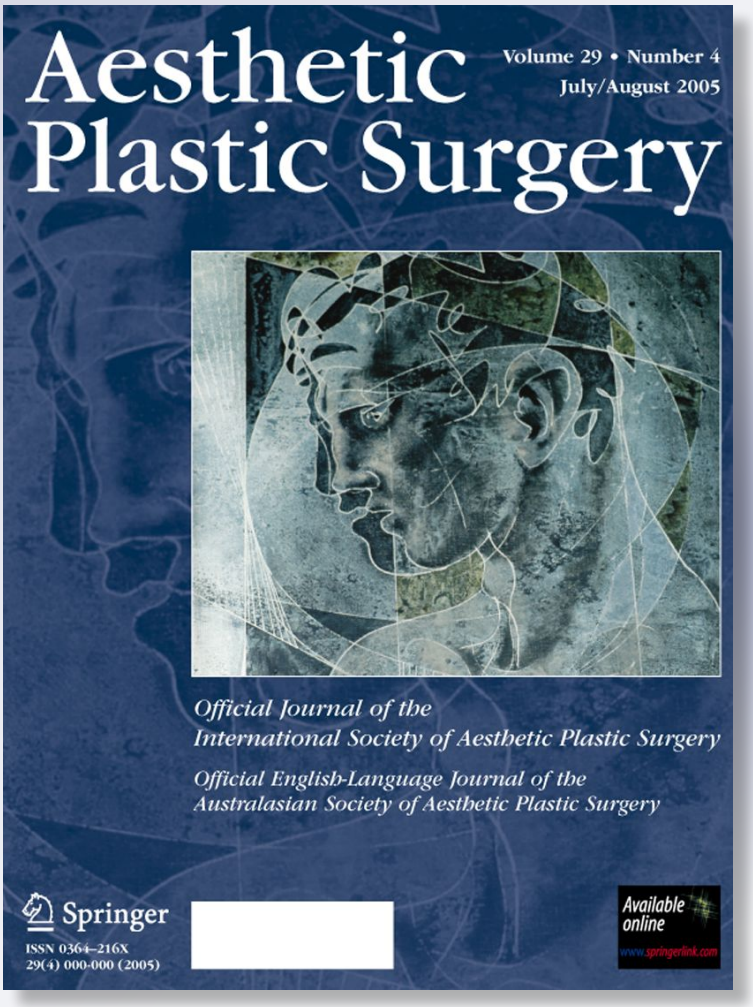

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REVIEW

Current Concepts in Scar Evolution and Control

Alan D. Widgerow

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Abstract The basic principles influencing scar expression and outcome have long been defined. Although these were relatively clear at the time, the exact events at a molecular level were poorly defined. The past decade has delineated the myriad of events that occur in the run-up to scar evolution far more clearly, although the intricate details have yet to be elucidated. What is clear is that a series of conversations and crosstalk takes place in the cell cytosol, in the cellular nucleus, and outside the cell within in the extracellular matrix. This interaction or "dynamic reciprocity" takes place via a series of signals, protein activation, ionic translocations, and receptor transactions. Marrying the previously defined principles with current described cellular/extracellular matrix (ECM) interactions enables us to describe more accurately the crosstalk occurring in scar evolution and possibly to influence the "wording" of that crosstalk to improve scar outcome. Thus, the principles of mechanostimulation and scar support, hydration occlusion, controlled inflammation, and collagen/extracellular remodeling are discussed with possible interventions in each category.

Keywords Cellular crosstalk · Scar · Collagen · Mechanotension · Inflammation · Collagen remodeling · Scar management · Multimodality scar control

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It has been some years since we delineated the principles of scar management. Although the principles have remained relevant, our current understanding of these principles has become much clearer with the advent of in-depth molecular biological sequence identification. This has allowed the choice of agents and the interventions that we use for scar management to be more focused and scientifically validated.

Our initial meta-analysis of the literature identified three principles that governed the successful scar management modalities of the time: scar support, adequate hydration, and hastened collagen remodeling [1]. A few years later we added a fourth principle of controlled inflammation, and the list appeared complete [2]. Based on these principles, a scar management program was developed that was multimodal in nature and directed at the sequential process of scar evolution, with agents selected to specifically impact the principles listed above.

Current progress in molecular biology, cellular signaling mechanisms, and extracellular matrix dynamics has allowed a more detailed molecular explanation for the positive outcomes seen with the interventions described above [2]. This review details the pathophysiology of scar evolution, the principles involved in management, and an explanation of the agents chosen for scar management based on these principles. One of the most exciting developments in our understanding of scar evolution is the interaction that seems to occur at a deeper dermal level even with superficial surface intervention. This appears to take place by intricate signaling mechanisms that interconnect the many layers of the dermis with the outer epidermal layer. If proved correct, this may have far-reaching consequences for certain agents that appear to work on the skin surface but may in fact have impact on the deeper layers of skin. This area of research may have far-reaching implications.

Scar Support

Support of the fresh scar was one of the earliest principles espoused for scar management. Multidirectional vector forces applied to a fresh scar were observed to produce hypertrophy [3, 4]. Observations made by Elliot et al. [3] regarding presternal scars showed that not only was hypertrophy common, but there were significant regional differences, with a tendency to scar hypertrophy overlying the body of the sternum, especially in females. These original observations can be explained by current molecular research. Sensitivity to mechanical tension transmits to the cell via signaling (probably by opening Ca+ channels) to glycoproteins, primarily fibronectin which acts as ligand attaching to integrins transmitting the signal from the extracellular matrix (ECM) into the cytosol [5–7]. From the cellular cytoplasm signal transducers (SMAD) 3/4 signals are stimulated by this tension to form complexes that enter the nucleus, initiating nuclear transcription and resulting in transforming growth factor (TGF)-β1 stimulation, procollagen formation, collagen formation, fibroblast differentiation to myofibroblast, and wound contraction with excess collagen III. Thus, specific focal adhesions at the cellular surface allow mechanical tension generated in the system to be transduced to the cytoskeletal network [8], initiating the synthesis and deposition of collagen. If the cycle is repeated a sufficient number of times, particularly intermittently, the physical representation that results is the hypertrophic scar.

Taping of the scar with microporous tape has proven effective in scar support in numerous publications [9, 10]. Based on the research noted above, one can extrapolate theoretical methods to influence scar mechanotension: these may include SMAD7 inhibition of signaling to TGF-β, connexin (Cx) 43 antagonism, connective tissue growth factor (CTGF) targeting, anti-SMAD2/3/4 complex, decreasing cadherin activation, and, more practical, myofibroblast phenotype induction inhibition by either direct methods or indirect methods by introducing collagen matrices to the wound where appropriate. Suffice it to say, simple support of the wound would appear to be the most logical strategy. This support is applied by taping the wound in a longitudinal direction rather than at right angles; we have also found that keeping the tape in place (including during bathing) for 4–5 days or until spontaneous separation occurs substantially reduces the risk of adhesive sensitivity or tape stripping. The ability to apply active agents to the surface of the tape that have additive effects on scar reduction, such as those described in this paragraph, make the tape–gel combination a very appealing modality [1, 2]. Of course, in many situations, such as small scars, those on the face, and those not exposed to major tension, it may not be necessary to tape the wound (Fig. 1). Adjustment to the formulation of the gel has been made that creates a crusting surface barrier that suffices for support of the scar (without tape) in these lesser-tension areas [2].

Adequate Hydration

Hydration of the scar surface is the basis of action of 90% of scar management systems on the market. Most oils (tissue oils), lotions, and creams have beneficial effects on scars primarily on the basis of their hydrative capacities [11–13]. Normal skin has a mature stratum corneum characterized by minimal transepidermal water loss (TEWL). Dehydration of the stratum corneum initiates signaling, inducing keratinocytes to produce cytokines that activate dermal fibroblasts to synthesize and release collagen. Excessive collagen production leads to abnormal scarring [14, 15]. It would now appear that the keratinocyte on the surface of the skin is capable of orchestrating and initiating the signaling events that culminate in fibroblast TGF-β stimulation of collagen production or cessation. The pro- or antifibrotic status has been shown to directly link up with the hydration or more particularly with the occlusive state of the keratinocyte [16, 17]. This has a major impact on scar management modalities and cosmetic/cosmeceutical formulation as in

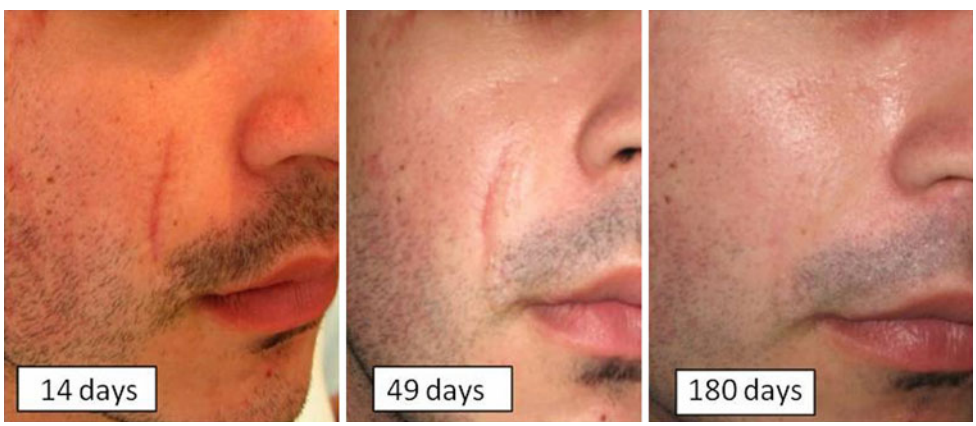

Fig. 1 Scar gel containing *Centella asiatica*, oleuropein, and *Bulbine frutescens* extracts and dimethicone. In this case the preparation was used without tape as support of this wound was relatively unimportant and impractical on the face. Progressive improvement of scar appearance is well demonstrated in this case over time

many cases therapy may be directed superficially with an expectant result in the deep dermis where the impact is desired.

Occlusion hydration results in a decreased activation of keratinocytes resulting in decreased production of IL-1β (and probably other cytokines yet to be identified), increased production of antifibrotic tumor necrosis factor (TNF)-α [14–16], and an increase in TGF-β3 via stimulation of the SMAD7 signaling mechanism [18–23]. This results in extracellular matrix (ECM) remodeling with less inflammation, decreased collagen production, and balanced protease activity collectively encouraging scar maturation.

Hydration of the stratum corneum appears to result in reduced TEWL, reduced inflammatory cytokine release, and reduced TGF-β1 stimulation with direct impact on the scar outcome [13, 15, 17]. This would appear to be the modus operandi of many of the current scar therapies on the market today.

The most effective barrier to TEWL and stratum corneum breach is silicone, in the form of either sheets or gels (dimethicone) [24–27]. In addition, the gel derived from the plant *Bulbine frutescens* has been found to be effective as a hydrating agent; the glycoproteins of the plant extract are large and remain on the surface of the skin long enough to produce effective hydration of the skin [1, 2].

Another novel patented approach to combining scar support and hydration/occlusion is application of a gel that includes dimethicone silicone, hydrating agents, and inflammatory controlling components to the surface of the tape. This results in a saturated tape through which actives of the gel are absorbed creating an ideal occlusive dressing for managing scars [2].

A multitude of antiscarring agents in the marketplace are expected to have some beneficial scar outcome purely on the basis of their hydrative properties. These agents affect merely one aspect of scar control—hydration—as opposed to multimodal formulations aimed at dealing with a number of principles of scar control simultaneously [2].

Controlled Inflammation

Inflammation is a necessary sequence in wound healing. However, exaggerated inflammation appears to be the central problem in most chronic (and many acute) diseases, be it cardiac, vascular, diabetic, or arthritic. Hypertrophic scarring is no exception to this issue: excessive inflammation results in exaggerated scars [28, 29]. Managed inflammation is a sought after principle in scar management. Work on fetal wounds suggests that a very mild inflammatory response may underlie the scarless healing observed [30, 31].

Multiple cytokines are involved in the inflammatory process. The TGF-β family, platelet-derived growth factors (PDGF), and epidermal growth factors (EGF) [28, 32] stimulate fibroblast proliferation and matrix production and induce leukocyte attraction. Leukocytes, in turn, reinforce fibroblast activity by acting through the TGF-β family, fibroblast growth factors (FGF), vascular endothelial growth factors (VEGF), prostaglandins [33], and SMAD activation [34, 35]. Increased levels of TGF-β1 and -β2 and decreased levels of TGF-β3 have been associated with hypertrophic scarring through inflammatory cell stimulation and fibroblast proliferation.

Although a complex array of inflammatory mediators and fibrogenic proteins has been described with multiple release and activation mechanisms, the process is fairly orderly and involves constant crosstalk between keratinocytes and fibroblasts. The keratinocytes primarily monitor the events occurring at a superficial level related to contact with the external environment, while the fibroblasts react to these signals from the keratinocytes and to signals dictated by the status of the extracellular matrix and nature of the granulation tissue. After wounding, stored IL-1 is released by keratinocytes, which activates fibroblasts and adjacent keratinocytes and attracts endothelial cells and lymphocytes to the injured area [36].

An added influence on HT scar formation appears to be the site of the wound, in particular the depth of the wound and components of the wound bed. In full-thickness wounds the fibroblasts that populate the wound area are recruited not only from the surrounding dermis but also from other tissues such as subcutaneous fat. These fibroblasts have been shown to possess increased expression of smooth muscle actin (α-SMA), collagen types I and III, and tissue inhibitors of metalloproteinases (TIMP)s and decreased expression of matrix metalloproteinases (MMPs) in comparison with cultured dermal fibroblasts [37]. This suggests that the myofibroblast from the subcutaneous fat may play a role in hypertrophic scar formation [37], which explains the observation of increased HT scar formation in full-thickness wounds.

Aside from the MMPs, additional mechanisms for clearing the ECM proteins exist. Proteasomes are very large protein complexes located in the nucleus and the cytoplasm [38]. The main function of the proteasome is to degrade unneeded or damaged proteins by proteolysis through their enzyme proteases. Proteasomes are part of a major mechanism by which cells regulate the concentration of particular proteins and degrade misfolded proteins.

In addition to degrading protein fragments, the proteasome plays an important part in deactivating SMAD3/4 signals, stimuli to TGF-β1 production. Several components of TGF-β signal transduction, including both positive and negative transducers, are irreversibly turned over by this

"protein-destroying machine." In a steady state, SMAD3 is constitutively degraded via the ubiquitin-proteasome pathway in the cytoplasm, and in response to TGF-β, it is phosphorylated and translocated into the nucleus, where it is also degraded through the ubiquitin-proteasome pathway. This suggests that not only in response to TGF-β but also in a steady state, the level of SMAD3 is regulated by the proteasome pathway [39].

Collectively, TGF-β signaling is controlled by the proteasome both positively and negatively. Degradation of SMAD7 maintains the signal and production of TGF-β, whereas degradation of the activated receptor complex and R-SMADs turns it off. Overexpression of SMAD7 has been shown to antagonize TGF-β-mediated fibrosis, carcinogenesis, and inflammation, suggesting a therapeutic potential of SMAD7 to treat these diseases [39, 40]. Similarly, a therapeutic possibility to prevent hypertrophic (HT) scar formation is saturation with SMAD7. This may be achieved by simultaneous stimulation of SMAD7 expression and activation of proteasome degradation of SMAD3/4 signals. Certain plant phenols (oleuropein) appear to achieve this goal [41–43].

Finally, concerning the inflammatory response, a short time (i.e., 1 h) after wounding, the endothelial cell's cyclooxygenase-2 (COX-2) enzyme is activated to synthesize prostaglandins. Metabolites and enzymes of the arachidonic acid cascade, including the cyclooxygenase-2 (COX-2) enzyme and its enzymatic product prostaglandin E_2 (PGE_2), are known to be critical mediators of the inflammatory response. Several studies have examined the COX-2 pathway and its part in the regulation of the inflammatory phase of cutaneous wound repair [44–46]. Inhibition of this inflammatory pathway has also been suggested to reduce scar formation [44]. Scarless fetal healing is known to proceed without a significant inflammatory response, which appears to be important in the lack of scarring. Research suggests that the COX-2 pathway is involved in scar production in fetal skin and that targeting COX-2 may be useful for limiting scar formation in adult skin [46].

Maturation of the inflammatory process involves a progressive increase in TGF-β3. This growth factor isoform appears to be involved in cessation of matrix deposition [18]. TGF-β3 reduces fibronectin and collagen deposition and is considered potently antifibrotic [19]. Increased levels are thus desirable in scar control strategies. This can be achieved indirectly via certain plant extracts (*Centella asiatica*) or directly via the human recombinant TGF-β3 avotermin which is still experimental (Juvista, Renovo, Manchester, UK).

We have been particularly interested in plant extracts that have effects on various phases of scar control. Triterpenic fractions of *Centella asiatica* and phenolic extracts of olive oil, e.g., oleuropein, have demonstrated multiple beneficial properties in controlling fibrogenesis. These extracts have been demonstrated to increase SMAD7, increase TGF-β3, decrease TGF-β1, decrease COX-2, increase proteasome activation, and display potent antioxidant effects. The combination is therefore elegant in synergy toward scar control [2, 41–43, 47–65].

Remodeling/Collagen Maturation

The final principle to be discussed that impacts on scar outcome is that of collagen fibrillar arrangement, maturation, and ECM remodeling. During the remodeling phase, myofibroblasts normally replace hyaluronic acid (HA) by proteoglycans such as decorin, which binds TGF-β1 and regulates collagen fibrillogenesis [66]. Decorin presents as a C-shaped structure that imposes itself between collagen fibrils, thus assuring uniform spatial arrangement of these fibrils. In hypertrophic scars, fibroblasts synthesize less decorin than normal dermal fibroblasts [66–68], and expression of decorin in burn scars is suppressed for about 12 months [67]. Decorin inhibits fibroblast proliferation and decreases TGF-β1 production and collagen synthesis in hypertrophic scar fibroblasts [66, 67], emphasizing its possible role in HTS.

In normal skin, collagen fibrils are composed of both type I and type III collagen. Type III comprises almost 20% of the total amount of collagen [69]. It is thought that type III collagen plays a role in fibrillogenesis and determines the collagen fibril diameter [70]. During granulation tissue formation, type III collagen expression increases more than type I expression, resulting in an altered ratio between the two collagen subtypes, changing from 20 to 50% type III collagen [71]. During maturation of the scar, the ratio decreases again to normal levels. Thus, increased amounts of collagen III relative to collagen I depicts an immature scar. Ratios of both collagen subtypes remain high in hypertrophic scars [72].

A change in the ratio of collagen III to collagen I is taking place (back to 20% collagen III from up to 50%) and fibrillogenesis is occurring constantly during the maturation phase. During this process, packaging of new fibers should be uniform and structured in a nonclumped moiety. The process of fibrillogenesis involves the conversion of procollagen to tropocollagen (nonhelical ends cleaved off) to fibrils arranged uniformly by "spacers" preventing collagen clumping. These spacers are normally provided by decorin [73]. Hence, there has been an active interest in decorin-like proteins with tetrapeptides akin to the decorin molecule (*Bulbine frutescens*) for collagen remodeling and structural arrangement [68].

Collagen crosslinking is important in this remodeling phase. Linkages need to be susceptible to MMP breakdown

to ensure balanced degradation and neosynthesis. Pyridinoline crosslinks, not normally seen in skin, have been reported to occur in HTS. They are able to withstand major force and tend to be resistant to MMP-1 degradation [74–76]. This pyridinoline phenomenon has been linked to oxygen radical activity (especially in burns); thus, antioxidants appear to be beneficial in potential scar reduction. The main antioxidant agents such as superoxide dismutase (SOD), catalase, glutathione peroxidase (GSH-Px), glutathione, ascorbic acid, and tocopherol are important for cellular protection because of their ability to eliminate free radicals such as reactive oxygen species (ROS) [76]. There is an increasing interest in the biochemical functions of natural antioxidant extracts from vegetables, fruits, and medicinal plants, which can become candidates to prevent oxidative damage. Extracts of *Centella asiatica* and oleuropein have demonstrated significant antioxidant capacities [57, 65].

Practical Applications

A patented process of applying a cream/gel containing antiscar active agents (*Centella asiatica*, oleuropein, dimethicone, *Bulbine frutescens*) onto the surface of tape has been successfully used for scar management. This creates an ideal occlusive dressing for scars. The gel with its active agents is absorbed through the tape within 2 min but the saturated tape continues to work as a scar dressing (Fig. 2). Thus, all principle requirements are accomplished: support, controlled inflammation, adequate hydration, and scar maturation through collagen remodeling and modulation. The tape remains in place during bathing. It is replaced only once spontaneous separation takes place (typically 3–5 days). Gel is applied to the surface of the tape twice a day and the routine is continued until scar maturation (white color) has occurred. The tape component may be stopped anytime during the 6 weeks after application if the scar is seen to be maturing well; gel is then applied directly to the scar. As some areas are not conducive to taping and support may not critical for the smaller scars, taping can be omitted from the routine. To compensate for this, another modification was made to the formulation whereby a thin film/crust is planned to form when the gel dries. This provides a certain amount of support to the scar but more importantly it serves as a barrier from outside contamination, irritation from the sun, and cosmetics.

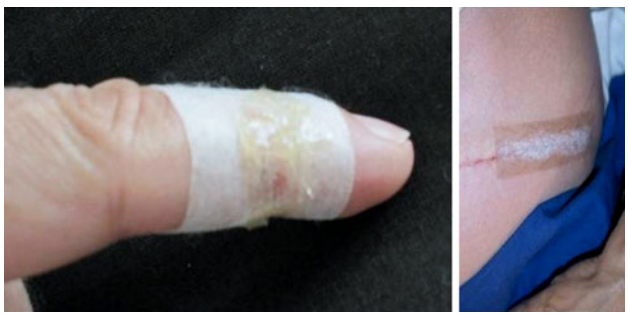

Fig. 2 The tape is applied longitudinally over the length of the scar/wound in areas where excess tension is anticipated or where protection and comfort are provided by the tape (*finger*). The gel is applied to the surface of the tape twice daily until maturation of the scar occurs (*white appearance*). In many cases taping can be discontinued after 6 weeks and gel alone is then applied to the surface of the wound

As with all theoretical postulations, these need to be subjected to scientific validation. A comprehensive trial was undertaken assessing 170 scars in different clinical situations [2]. This trial demonstrated that if scar management is dealt with conscientiously with adherence to the principles elucidated above, hypertrophy could be prevented in more than 80% of cases [2]. This contrasts sharply with a reported series on scar outcome where hypertrophy and exaggeration of the scar was anticipated in 60–80% of cases when there was no management of the scar (Fig. 3) [77].

An added advancement in scar management has been using this application to combat acne scarring. Acne scarring involves a complicated process of hormonal, infective, genetic, and environmental factors that combine to produce devastating outcomes in many patients. The most feared outcome of the process is the ultimate scarring that may result. The formulation described above is non-comedogenic but is not being used to control acne. Rather, it is specifically used as an adjunct to the normal antiacne routine. Thus, when the pustule is free of pus and advances to an inflammatory lesion, the gel is used on each individual lesion. This is a phased approach where each lesion is treated individually depending on its state in the acne evolution. The best results appear to be when treatment is initiated as soon as the infective episode is under control and inflammation predominates. Preliminary results of trials have demonstrated great efficacy at scar prevention, alleviation of redness, and surface filling.

Scar management discussions would not be complete without discussing the concept of keloid scars. These are often confused with hypertrophic scars and arise from completely different circumstances. Keloid scarring is usually a genetic phenomenon where collagen type I is produced in a tumor-like fashion with uncontrolled growth of scar tissue. The history usually involves a wound that is well managed and without infection or any discernable problem that progressively increases in size and reactivity and overflows its boundary. It may be painful, sensitive, and extremely uncomfortable and treatment today (often radiotherapy) is unpredictable and unsatisfactory. The principles described above do not relate to keloid scarring

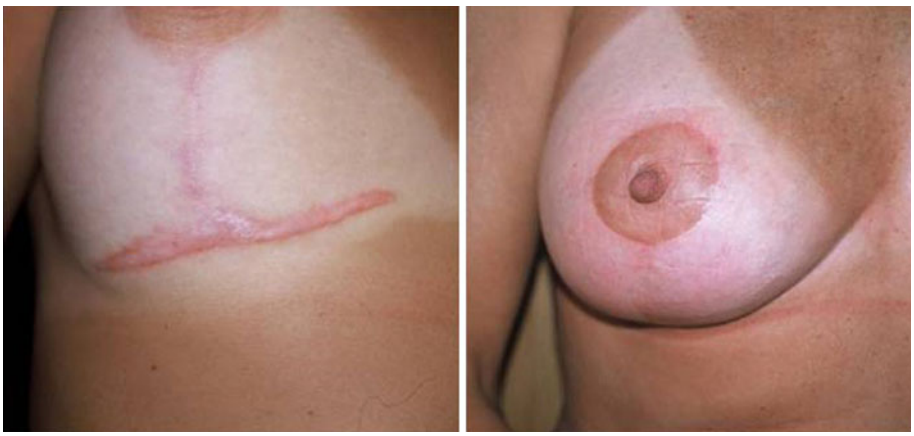

Fig. 3 Contrasting views of unmanaged and managed scars on the breast. Note the thickened, reactive, red scarring seen in the unmanaged breast scar as opposed to thin white flat scar in a patient included in the ScarScience™ (Litha Healthcare) trial

and it is important to recognize that. Any company that claims that keloid and hypertrophic scars can be prevented and treated with the same preparation are displaying ignorance and it should cast great doubt on the efficacy of that product. These are different mechanisms of scarring and need to be dealt with as such.

Summary

Scar control is an area that has received much attention from a patient perspective and fortunately from a research perspective too. Much of the guesswork involved in scar formation and exaggeration has been eliminated and the principles involved have been well defined. It is clear that control needs to come from a number of areas and no single modality will be adequate to ensure good outcome. Those perceptive to acronyms will recognize a perfect example, thus the combination of proven factors relating to Support, Controlled inflammation Adequate hydration, and Remodeling/collagen maturation—SCAR—has proven extremely efficacious in the clinical trials conducted to date. Scar control should be an important aspect of all wound management and can now be scientifically directed to varying clinical situations that may present themselves.

Disclosures Dr. Widgerow serves as an R&D consultant for Litha Healthcare Inc. and receives consulting fees.

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