https://doi.org/10.1038/d41586-019-03602-4

News & views

Medical research

Ready-made cellular plugs heal skin wounds

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The finding that a thin sheet of fibrous tissue under the skin contains a prefabricated, movable cellular sealant that can heal deep wounds might have implications for the treatment of scars and ulcers.

Skin consists of an outer epidermal layer (the epidermis) and an inner dermal layer (the dermis). If you pinch your skin, you can lift it because these two cellular layers move freely above a membranous sheet called the fascia, which contains cells and extracellular-matrix material. This gelatinous tissue creates a frictionless interface between the skin and the more rigid structures beneath it, such as muscle and bone. However, it now seems that the fascia has roles beyond providing a non-stick surface. Writing in *Nature*, Correa-Gallegos *et al.*[1] report that the fascia contains a movable sealant that patches up deep injuries to enable rapid wound repair.

The scar tissue of a healing skin wound contains fibroblast cells, which make and modify extracellular-matrix proteins. These fibroblasts can be identified by their expression of a protein called Engrailed-1, and are termed Engrailed-positive fibroblasts (EPFs). The idea that the fascia might be a repository of cellular components involved in wound healing and scar formation came from a previous study[2], which reported that EPFs reside not only in the skin, as expected, but also in the fascia.

To investigate wound healing in mice, Correa-Gallegos and colleagues grafted fascia containing cells engineered to express green fluorescent protein onto skin cells expressing red fluorescent protein. The authors then wounded this dual-coloured 'fluorescent sandwich' and transplanted it into a healthy mouse. Comparison of the percentages of green and red cells revealed that 80% of cells in the healing wound came from the fascia. Furthermore, the vast majority of many cell types found in the healing injury originated from the fascia, including contractile fibroblasts (or myofibroblasts), blood-vessel cells, macrophages of the immune system and nerve cells.

To confirm that their observations were not due to any peculiarities of this artificial grafted structure, the authors injected a dye into the fascia of mice, and the mice were then given a deep wound that penetrated the animals' skin and fascia. The authors mapped the dye-labelled cells that populated the healing wound and the surrounding scar tissue. More than half of the cells in the healed wound were labelled with the dye, confirming that the fascia is the major source of scar-forming tissue after deep injury.

Deep wounds lead to scars that are larger and harder to heal than those arising from superficial wounds that do not penetrate the fascia[3]. The authors used two-photon microscopy to analyse deep skin wounds in mice engineered[4] to express fluorescent proteins, which can be used to trace scar-forming EPFs. They found that a cellular plug in the fascia, consisting of extracellular matrix, macrophages, blood vessels and nerves, moved upwards into the damaged skin to form a scar. This healing process does not require cell division, indicating that the plug was prefabricated. Importantly, the authors found that key proteins that have been reported to define the types of fibroblast found in scars[5] are expressed at higher levels on fascial than on dermal fibroblasts, consistent with a model in which fascial EPFs are the major source of fibroblasts in healing deep wounds (Fig. 1).

Given that fibroblasts regulate the extracellular matrix, the authors used microscopy to visualize physical features of fibres of the protein collagen, which is a component of the extracellular matrix. Collagen in the fascia was more coiled and immature than were the stretched and interwoven collagen fibres in the dermis. Furthermore, when a fluorescent dye was used to tag collagen in an injured animal, this revealed that the extracellular matrix of

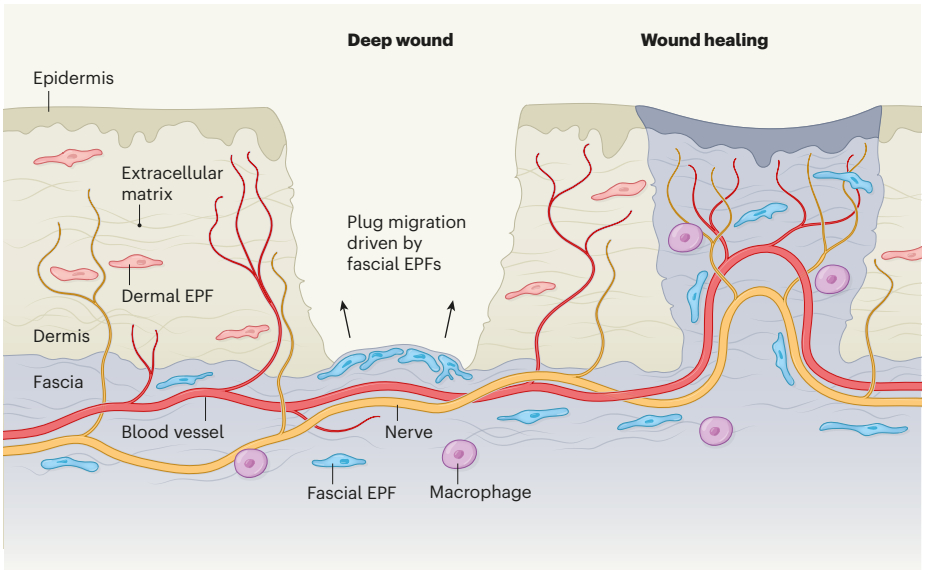


Figure 1 | The healing of deep skin wounds. The skin consists of an outer layer called the epidermis and an inner layer, the dermis. Superficial wounds no deeper than skin level can be repaired by cells called Engrailed-positive fibroblasts (EPFs) in the dermis, which make extracellular-matrix material. Correa-Gallegos *et al.*[1] investigated in mice the healing of deep wounds that penetrated below the skin into a layer known as the fascia. The fascia contains EPFs, extracellular matrix, blood vessels, nerves and immune cells called macrophages. The authors report that prefabricated plug of material from the fascia moves upwards, steered by fascial EPFs, to seal the wound. (Image based on Fig. 6 of ref. 1.)

the fascia moved upwards like a pliable gel into the damaged tissue, to plug and then repair the wound. By contrast, dermal collagen remained immobile.

The authors then tested whether EPFs from the fascia drive the movement of the prefabricated plug. They inserted non-adhesive membranes in mice to separate the fascia from the dermis, which resulted in delayed repair and non-healing wounds that remained open. Animals in which these membranes were not inserted did not show these effects. The removal of fascial EPFs by a genetic approach also resulted in the plug not entering wounds and in poor healing. These findings indicate that fascial EPFs do indeed steer the plug that seals deep wounds.

Although this study has potential relevance for human disease, most of the work was carried out in an artificial mouse model. Moreover, mice have a type of muscle called the panniculus carnosus, which lies between the fascia and the skin and is used to twitch the skin[6]. However, humans lack this twitching ability and have only a small remnant of this muscle. Therefore, the authors needed to determine whether scar formation occurs in a similar manner in humans and mice despite such differences.

The team analysed fascial fibroblasts in human skin and investigated a type of human raised scar called a keloid, which grows bigger than the original injury and can be profoundly itchy, inflamed and painful[7]. Many of the proteins that characterize the mouse fascia were also highly expressed in human fascia and keloid scars. This similarity suggests that the same processes are involved in wound healing and scar formation in both species. However, it is not yet clear whether these findings in mice reveal general principles that are relevant to human skin disease.

The authors' findings provide satisfying potential explanations for some unsolved clinical conundrums. Nerves, blood vessels and macrophages in the prefabricated plug are dragged into the mouse wound; if the same phenomenon occurs in humans, this could explain why keloids itch and are painful. Keloid formation is more common at sites of thicker fasciae (such as the chest, back and thighs) than at sites where the fascia is thinner (for example, the feet), which is consistent with a model in which the fascia drives keloid formation.

Could these discoveries about the skin shed light on other clinically relevant fibrotic diseases (conditions associated with the accumulation of extracellular matrix) that affect organs in which the fascia is not present, such as the lung and liver? Perhaps the mechanisms uncovered in mice might have relevance for the processes underlying skin damage in the leg ulcers that can develop in people who have diabetes. In any case, it is clear that advances made in understanding the biology of the fascia might reveal new targets for treating scarring diseases of the skin.

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1. Correa-Gallegos, D. *et al. Nature* https://doi.org/10.1038/s41586-019-1794-y (2019).
2. Rinkevich, Y. *et al. Science* **17**, aaa2151 (2015).
3. Dunkin, C. S. *et al. Plast. Reconstr. Surg.* **119**, 1722–1732 (2007).
4. Muzumdar, M. D., Tasic, B., Miyamichi, K., Li, L. & Luo, L. *Genesis* **45**, 593–605 (2007).
5. Driskell, R. R. & Watt, F. M. *Trends Cell Biol.* **25**, 92–99 (2014).
6. Stecco, C., Adstrum, S., Hedley, G., Schleip, R. & Yucesoy, C. A. *J. Bodywk Mov. Ther.* **22**, 354 (2018).
7. Peng, G. L. & Kerolus, J. L. *Facial Plast. Surg. Clin. N. Am.* **27**, 513–517 (2019).

Online heading: Ready-made cellular plugs heal skin wounds