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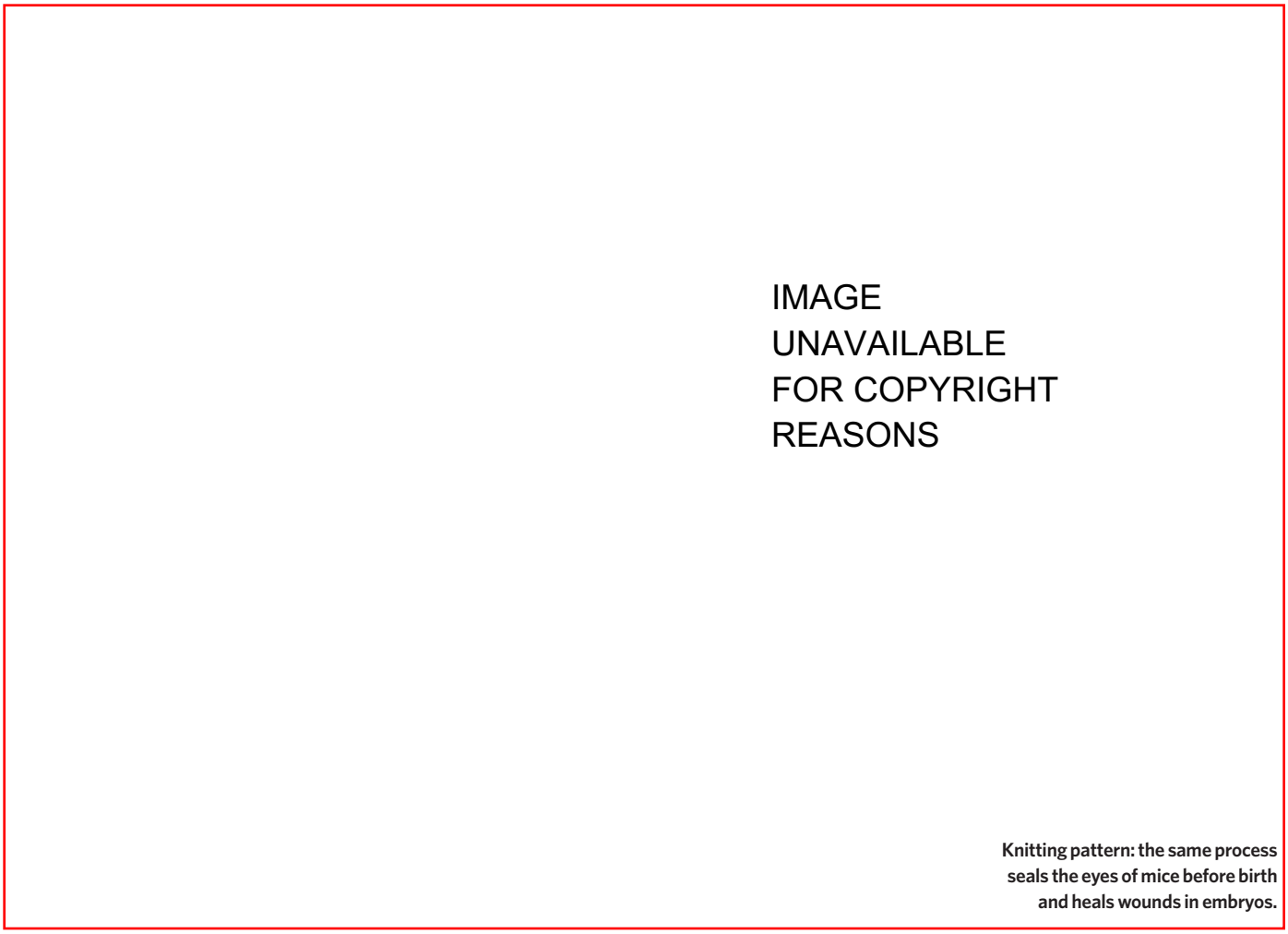


Knitting pattern: the same process seals the eyes of mice before birth and heals wounds in embryos.

The healing touch

Wound an embryo and it heals perfectly, with no scars. Can we teach adult wounds the same trick, asks **Meredith Wadman**.

It was alligators that first drew Mark Ferguson to study human scarring, but not for the reasons you might expect. In the late 1970s, he had become fascinated by cleft palate. Alligators were the obvious research subject: their embryos have palates, and develop in easily accessible eggs. But when Ferguson performed surgery on alligator embryos to mimic cleft palates, the creatures hatched with completely normal, unscarred mouths. "As a surgical model of cleft palate it was perfectly useless," says Ferguson. "As an observation of scar-free healing, it was of great scientific and clinical interest."

Ferguson, who now researches wound healing at the University of Manchester, UK, was not the first to stumble across this phenomenon. As early as 1960 there were anecdotal reports that wounds made early in gestation in embryos of many species, including humans, heal rapidly and perfectly. Over the past 20 years, researchers have been probing the mysteries of this process in the hope of improving adult wound healing and perhaps even making scarless healing a reality. It's no trivial point: delayed wound healing in the US elderly, for instance, is estimated to cost more than $9 billion each year.

Growing realization

Such work has wider implications too. There are striking similarities between the mechanisms embryos use to heal wounds and those they use to knit their body parts together during normal development. Research is throwing light on these basic processes and what might go wrong with them to cause birth defects such as cleft palate.

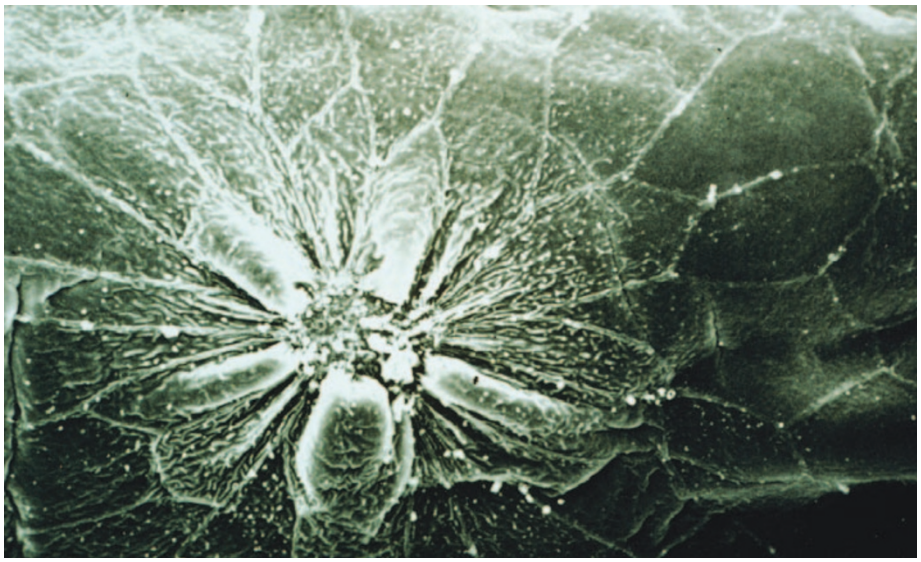
When adult skin is wounded, a blood clot quickly forms to stem the bleeding, then cells underneath move in for the repair job. Some of these cells, called fibroblasts, churn out a matrix of support proteins. Skin cells at the edge of the wound drag themselves over this to close the wound. As immune cells rush to the scene, the underlying connective tissue contracts, leading to scarring.

Embryos do things very differently, says Paul Martin, a cell and developmental biologist at the University of Bristol, UK. A fruitfly embryo closing a wound rapidly assembles a cable of the protein actin in the 'skin' cells at the wound's edges. Contraction of this cable seems to pull the edges together much as a drawstring pulls a bag shut[1]. Once the edges meet, the cells reach out with thin extensions called filopodia, which interlock to seal the wound. Flies unable to make these cell extensions cannot close wounds completely[2].

Intriguingly, this tissue repair uses the same machinery used by embryos to close holes that normally arise as they build their bodies, says Martin. This makes sense, he says: embryos are in the business of building tissue, and wound healing is essentially a process of rebuilding what was damaged. To close a wound, he says, "all they do is reactivate the machinery they're using somewhere else in the body anyway".

For example, a developing fruitfly embryo needs to zip up its outer skin as this grows over its back. This process, dubbed dorsal closure (see graphic), uses the same actin cable and filopodia mechanism. What's more, this mech-

K. WOOLLEY AND P. MARTIN



On the mend: a three-day-old zebrafish embryo draws a wound shut on its outer layer.

anism is remarkably common across the animal kingdom, whether it's in an embryonic mouse knitting its eyelids shut or a worm fusing its outer surface. It is also thought that filopodia are critical for a human embryo to fuse its palate or the tube that forms its brain and spinal cord.

Wrong turning

When these events go wrong in humans, notes Martin, the consequences range from the serious, such as cleft palate, to the devastating, as in anencephaly, when a baby is born missing a large part of its brain and skull. So it is hoped that understanding the process better will mark a beginning in the long journey towards preventing this kind of problem.

The evolutionary conservation of the healing mechanism means that researchers can study its basic biology easily in lab workhorses such as flies and mice. One focus is a signalling pathway between cells known as the JNK cascade, which plays a central role in skin-cell migration both after wounding and during normal development[3]. The genes switched on by this cascade are activated during dorsal closure in flies and within minutes in wounded rat embryos, in response, it is thought, to mechanical stress on cells.

Biologists such as Martin would dearly love to know more about how to activate this signalling, to kick-start healing in chronic wounds such as diabetic leg ulcers. Recent findings that conserved genes also control skin repair once wounds are closed in flies and mammals offers another target for therapy[4,5]. For the moment, such treatments are still a distant dream. But there is one facet of embryonic wound healing that could have clinical relevance sooner: inflammation.

Wounds in adults are red and inflamed, bristling with immune cells summoned by molecular signals from platelets that leak from damaged blood vessels. These signals are also thought to trigger the skin-cell migration and connective-tissue contraction that ultimately lead to scarring. The inflammatory response is a robust one, which makes sense in the face of invading bacteria. Scarring could be the price to pay for protecting against infection.

Embryos cannot mount such a response until late in gestation, and this could be part of the reason they don't scar. Martin and his colleagues recently studied mice genetically incapable of raising an immune response because they lack key immune cells called macrophages and neutrophils[6]. When these mice are wounded as newborns, they not only seem not to scar, they also heal faster.

Gillian Ashcroft, a consultant in tissue repair at the University of Manchester, UK, further bolstered the case when she showed that mice lacking a protein called Smad3, which is involved in inflammation, healed their skin wounds faster than normal mice[7]. Immunologist Luisa DiPietro and her colleagues at Loyola University Medical Center in Maywood, Illinois, have made similar findings in mice lacking neutrophils[8].

DiPietro's team has also looked at the mucous membranes in the mouth. Although this environment is a hotbed of bacteria, wounds in the mouth of mice show less inflammation and heal faster than those in the skin[9]. "The emerging evidence all leads to this idea that in healing wounds, less inflammation might be better," says DiPietro.

To hear Ferguson tell it, the drive to better healing is already under way. In 2000, he co-founded Renovo, a Manchester-based biotechnology company. The firm is well into clinical trials of several drugs aimed at reducing post-operative scarring.

Mimicking embryos

Renovo's drugs stem from Ferguson's work on different forms of a chemical signal, a protein called transforming growth factor β (TGFβ). This plays a key role in scar formation, summoning and activating inflammatory cells, as well as spurring fibroblasts to make matrix. In work on mice and sheep embryos, Ferguson found that embryonic wounds have very high levels of one form of the protein, called TGFβ-3, but very low levels of TGFβ-1 and TGFβ-2. In adult wounds, the profiles of these forms are reversed[10,11]. Ferguson's team discovered that by mimicking the embryonic profiles in adult animal wounds through adding more TGFβ-3 or suppressing TGFβ-1 and TGFβ-2, they could reduce scarring[12].

Today, Renovo's flagship product — a human genetically engineered TGFβ-3 dubbed Juvista, has passed initial safety and efficacy trials in skin wounds. Ferguson is now preparing to publish the trial data, and if all goes well, he predicts that Juvista could be on the market in 2009.

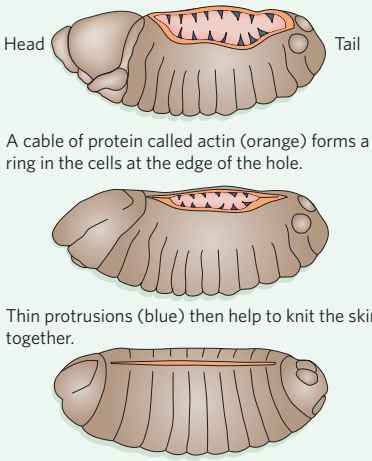
Martin, too, is optimistic. He foresees a day when designer drugs modulating the inflammatory response will improve myriad diseases in which inflammation goes awry, from Crohn's to heart disease.

Getting there, he notes, will take a far better understanding of what he calls "this can of worms we know nothing about". He adds: "Because this is such a complex process, we've got to go back to the embryo, back to a genetically tractable organism like the fly, to start to understand it and to watch it." ■

Meredith Wadman is a freelance writer based in Washington DC.

Drawing on embryos

The way fruitfly embryos 'zip up' their outer skin could give new insights into wound healing.



1. Martin, P. & Lewis, J. *Nature* **360,** 179–183 (1992).
2. Wood, W. *et al. Nature Cell Biol.* **4,** 907–912 (2002).
3. Martin, P. & Parkhurst, S.M., *Development* **131,** 3021–3034 (2004).
4. Mace, K. A. *et al. Science* **308,** 381–385 (2005).
5. Ting, S. B. *et al. Science* **308,** 411–413 (2005).
6. Martin, P. *et al. Curr. Biol.* **13,** 1122–1128 (2003).
7. Ashcroft, G. S. *et. al. Nature Cell Biol.* **1,** 260–266 (1999).
8. Dovi, J. V., He, L.-K. & DiPietro, L. A. *J. Leuk. Biol.* **73,** 448–455 (2003).
9. Szpaderska, A. M., Zuckerman, J. D. & DiPietro, L. A. *J. Dent. Res.* **82,** 621–626 (2003).
10. O'Kane, S. & Ferguson, M. W. J. *Int. J. Biochem. Cell Biol.* **29,** 63–78 (1997).
11. Whitby, D. J. & Ferguson, M. W. J. *Dev. Biol.* **147,** 207–215 (1991).
12. Shah, M., Foreman, D. M. & Ferguson, M. W. J. *Lancet* **339,** 213–214 (1992).