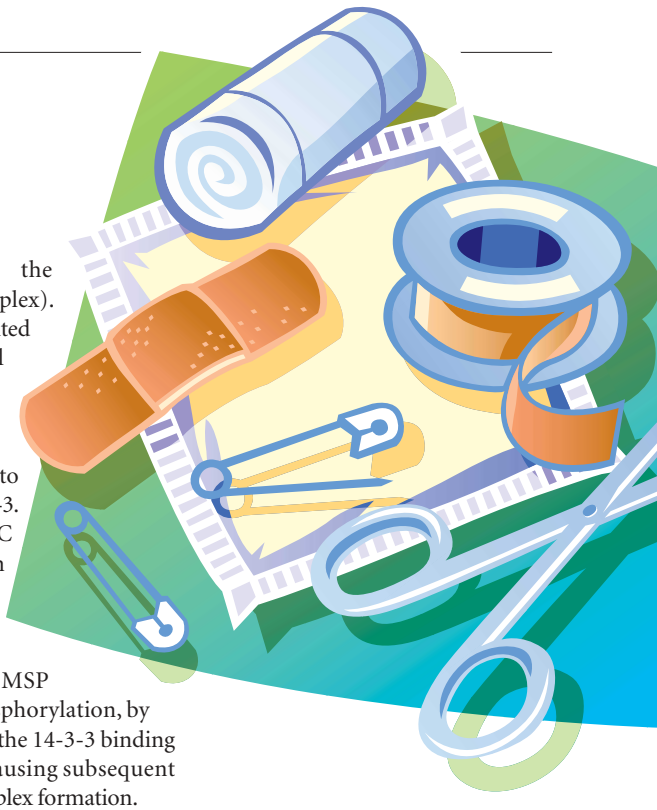
CELL MIGRATION

Healing aid



Growth factors and integrins often join forces to regulate biological processes, but in the process of keratinocyte migration, a third protein — 14-3-3 — puts in an appearance. The growth factor macrophage-stimulating protein (MSP) induces its receptor, Ron, to complex, through 14-3-3, with the $\alpha_6\beta_4$ integrin. This removes $\alpha_6\beta_4$ from cell-adhesion structures and relocalizes it at lamellipodia. In conjunction with increased migration, this implicates Ron and 14-3-3 in wound healing, as reported by Santoro and colleagues.

Serine 1394 in the Ron carboxyl terminus is a potential phosphorylation site for many kinases, but only Akt, through phosphatidylinositol 3-kinase, phosphorylated Ron *in vivo* in response to MSP. Ron's carboxyl terminus also matches consensus motifs for several protein–protein interaction domains — and the phosphorylation-dependent scaffold protein 14-3-3 did, in fact, bind to MSP-mediated, Akt-phosphorylated Ron. 14-3-3 co-immunoprecipitated with Ser1394-phosphorylated Ron, and both proteins colocalized at the leading edge of migrating cells in lamellipodia.

In primary keratinocytes, 14-3-3 associated with Ron in response to MSP. MSP induced keratinocytes to spread and migrate faster on the extracellular-matrix component laminin-5 compared with cells in which Ser1394 was mutated, or in which the carboxyl terminus of 14-3-3 was deleted (both these constructs interfere with the Ron–14-3-3 complex). These effects pointed to a potential involvement of integrins, and Santoro and colleagues found β_4 to interact with 14-3-3. Protein kinase C (PKC) has been shown to phosphorylate β_4, and the authors found that MSP induced the phosphorylation, by PKCα, of $\alpha_6\beta_4$ at the 14-3-3 binding site — thereby causing subsequent $\alpha_6\beta_4$–14-3-3 complex formation.

So, if 14-3-3 binds to Ron and to $\alpha_6\beta_4$, can it mediate a complex between the two proteins? Indeed it can. And as $\alpha_6\beta_4$ is usually found in hemidesmosomes — structures that support cell adhesion — and Ron–14-3-3 in lamellipodia, the authors then assessed the effect of MSP on $\alpha_6\beta_4$ localization. MSP was found to induce $\alpha_6\beta_4$ relocalization from hemidesmosomes to lamellipodia. Concurrent with this, cells adhered to laminin-5 through $\alpha_3\beta_1$ instead of $\alpha_6\beta_4$. As this occurs during wound healing, Santoro and colleagues looked at MSP's role in this process. MSP promoted keratinocyte migration in wounds in mice and in *in vitro* wound-healing assays. This required the formation of a Ron–14-3-3–$\alpha_6\beta_4$ complex and $\alpha_3\beta_1$-dependent cell migration.

When they studied downstream signalling, the authors found that $\alpha_6\beta_4$ was phosphorylated only when a Ron–$\alpha_6\beta_4$ complex was formed, and that this complex activated p38 and nuclear factor κB signalling. Both pathways were required for the transcriptional upregulation, not only of MSP itself, but also of matrix metalloproteinases, which are normally induced during keratinocyte wound healing. So, $\alpha_6\beta_4$ seems to change from being an adhesive molecule to a signalling component during wound re-epithelialization.

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References and links

ORIGINAL RESEARCH PAPER Santoro, M. M., Gaudino, G. & Marchisio, P. C. The MSP receptor regulates α6β4 and α3β1 integrins via 14-3-3 proteins in keratinocyte migration. *Dev. Cell* **5**, 257–271 (2003)