Gastric Sonic Hedgehog Acts as a Chemoattractant for Monocytes During Tissue Regeneration

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Background: Re-expression of Sonic Hedgehog (Shh) in *Helicobacter Pylori (H. pylori)* infected patients, following eradication of the infection, results in gastric epithelial regeneration and ulcer repair. Shh regulates multiple events essential to would repair in the cornea and heart. However, in the stomach the role of Shh in gastric wound repair remains unclear. Ulcer repair in the stomach is a complex process that involves inflammatory cell infiltration, cell proliferation, angiogenesis, and re-epithelialization. Macrophages play a principal role in these events and the recruitment of monocytes serves as a potential mechanism through which Shh could drive tissue regeneration.

Hypothesis: Gastric Shh acts as a chemoattractant for monocyte/macrophage recruitment during ulcer repair.

Methods: A mouse model expressing a parietal cell-specific deletion of Shh (PC-ShhKO) was used. Ulcers were induced in control, PC-ShhKO and *H. pylori*-infected mice using acetic acid. Gastric samples were collected from the ulcerated and adjacent uninjured tissue, epithelium enzymatically dissociated and monocyte recruitment analyzed by FACS. Changes in cytokine, chemokine and pro-angiogenic factors were measured using a Luminex®-based multiplex assay. Shh expression was measured by western blot.

Results: In controls, ulcers healed within 7 days post-injury. Tissue regeneration was accompanied by the significant increase in gastric Shh expression within 24 hours and the recruitment of $CD11b^{+}F4/80^{+}Ly6C^{high}$ inflammatory monocytes to the stomach within 48 hours post injury**.** Control mice had elevated expression of cytokines IL-1α, IL-6, IL-1β, chemokines MCP-1 and MIP-2 and pro-angiogenic factor VEGF 1 to 3 days post-injury. PC-ShhKO mice showed complete loss of ulcer repair and the absence of $CD11b^{+}F4/80^{+}Ly6C^{high}$ inflammatory monocyte recruitment to the stomach. PC-ShhKO mice exhibited epithelial hyperproliferation and tumor development 7 days post ulcer-induction. *H. pylori*-infected mice had significantly reduced Shh expression, lack of monocyte recruitment, up-regulation of IFNγ and TNFα accompanied by the suppression of VEGF, MIP-1α and MIP-2 and impaired gastric regeneration.

Conclusions Gastric Shh plays a central role in tissue regeneration by acting as a chemoattractant for monocyte/
macrophage recruitment during ulcer repair. *H. pylori* infection suppresses Shh expression as part of the pathophysiological mechanism that may be detrimental to ulcer healing and a trigger for cancer development.

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