CONCLUSIONS: These findings demonstrate the ability of prehemorrhage to condition the colon integrity and protect it against a subsequent septical insult. Inflammatory cytokine expression was significantly suppressed and resulted in a restored intestinal muscle function that prevented the detrimental consequences of sepsis-induced ileus.

P-selectin glycoprotein ligand-1 (CD162) mediates leukocyte rolling in ischemia-reperfusion and CXC chemokine-induced leukocyte recruitment in the colon

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INTRODUCTION: Leukocyte recruitment is a rate-limiting step in ischemia-reperfusion (I/R)-induced tissue injury. The objective of this study was to determine the potential role of P-selectin-glycoprotein ligand-1 (PSGL-1, CD162) in I/R and chemokine-induced leukocyte-endothelium interactions in the colon.

METHODS: Balb/c mice were exposed to I/R by clamping the superior mesenteric artery for 30 min and leukocyte rolling and adhesion were analysed in the colonic microcirculation by intravital microscopy after 120 min of reperfusion. In separate experiments, mice were challenged with CXC chemokines (MIP-2 and KC). In order to determine the role of PSGL-1, mice were pretreated with antibodies against PSGL-1, P-selectin and an isotype-control antibody. CXC chemokine expression was determined by RT-PCR and ELISA.

RESULTS: I/R caused a profound increase in leukocyte rolling and adhesion in the colon. Pretreatment with the anti-PSGL-1 antibody reduced I/R-induced leukocyte rolling and adhesion by more than 89%. I/R increased the expression of MIP-2 and KC in the colon and immunoneutralization of PSGL-1 reduced chemokine-induced leukocyte rolling and adhesion by more than 85%. Moreover, inhibition of P-selectin abolished both I/R- and chemokine-induced leukocyte rolling and adhesion in the colon.

CONCLUSIONS: Our novel data demonstrate that PSGL-1 is a dominant adhesion molecule supporting I/R- and chemokine-provoked leukocyte rolling in colonic ventules. Moreover, our findings demonstrate that inhibition of PSGL-1, not only reduces rolling, but also abolishes I/R- and chemokine-induced leukocyte adhesion. Thus, PSGL-1 may be a key target to protect against pathologic recruitment of leukocytes and tissue injury in the colon.

Regulation of colonic crypt fluid and electrolyte secretion by the calcium sensing receptor

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INTRODUCTION: Colonic crypts serve in regulating the fluid and electrolyte composition of stool. Secretory diarrhea results when there is excessive fluid secretion because of abnormal chloride (Cl) transport. Recent studies from our laboratory have determined that the Calcium sensing receptor (CaSR) is capable of modulating the activity of these ion exchangers and transporters in the rat colon. The aim of this study was to investigate modulation of fluid and electrolyte secretion by the CaSR in the human colon.

METHODS: Individual human colonic crypts were isolated by microscopic hand dissection. Crypts were loaded with a fluorescent chloride indicator dye, MQAE. Intracellular chloride levels were monitored in various buffer solutions: 0 Cl Hepes, standard Hepes, 0 Cl Hepes and standard Hepes with Gd3+ (potent stimulator of the CaSR), 0 Cl Hepes and standard Hepes with R-568 (calcimimetic). Forskolin was also applied to some crypt cells.

RESULTS: We found that forskolin was able to induce increased chloride and fluid secretion.Activation of the CaSR with 1 mM Ca2+ led to a decrease in basal and forskolin-induced fluid secretion. The use of Gd3+ or the calcimimetic R-568 resulted in an even greater decrease in fluid secretion. The effect with the calcimimetic was atainable at nanomolar concentrations,an effect indicative of CaSR activation.

CONCLUSIONS: The CaSR modulates fluid and electrolyte secretion in human colonic crypts. Activation of the receptor with calcimimetics leads to a reduction in secretagogue-induced fluid secretion. This receptor may be a potent new target for developing therapeutic options for the treatment of secretory diarrhea.

Transforming growth factor-beta (TGF-beta) induces vascular endothelial growth factor (VEGF) and plasminogen activator inhibitor-1 (PAI-1) gene expression through Smad3 transcription factor

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INTRODUCTION: TGF-beta is overexpressed in human colon cancers and associated with advanced stages and decreased survival. PAI-1 promotes tumor cell invasion through degradation of tumor stroma, while VEGF promotes angiogenesis. Both PAI-1 and VEGF are upregulated during colon carcinogenesis. It is not known whether TGF-beta regulates PAI-1 and VEGF expression in the gut. The purpose of this study is to determine whether TGF-beta regulates PAI-1 and VEGF expression in gut epithelial cells through Smad3 transcription factor, one of the mediators of TGF-beta signaling.

METHODS: We generated rat intestinal epithelial cell lines expressing either a dominant-negative Smad3 (RIE-1/Smad3DeltaSSVS) or human Smad3 (RIE-1/Smad3), RIE-1/pBabe cells as vector control. Cells were treated with TGF-beta (40 pM) and real-time RT-PCR was performed to quantify PAI-1 and VEGF mRNA levels. Each experiment was repeated at least twice.

RESULTS: In control cells, TGF-beta induced both PAI-1 and VEGF mRNA in a time-dependent fashion beginning at 1 h. After 5 h of TGF-beta treatment, PAI-1 increased by 23-fold compared to untreated control and VEGF increased 4.3-fold compared to control. Expression of Smad3DeltaSSVS attenuated TGF-beta-induced PAI-1 and VEGF expression compared to control cells (>73%).