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Notch signaling is activated in diabetes and contributes to defective wound healing S.-B. Catrina, K. Brismar, V.G. Sunkari, I. Botusan, X. Zheng, J. Grunler Dept of Molecular Medicine and Surgery, Karolinska Institutet, Stockholm, Sweden

Background and aims: Diabetic wounds are characterized by impaired coordination of several mechanisms essential for healing as cell proliferation, cell differentiation and angiogenesis. Notch system together with hypoxia is a major player in cell differentiation and angiogenesis. It consists of several receptors (Notch 1-4) and ligands with a high specific cell-dependent effect. Binding of the ligands to the receptors is followed by proteolytic cleavage of the receptor by γ-secretase complex which results in the release of intracellular domain of the Notch receptor (NICD) that translocates in the nuclei and activates several genes. We have proposed to study the modulation of the Notch pathway in diabetes and its potential relevance in defective diabetic wound healing.**Methods:** The modulation of Notch system by hyperglycemia was studied *in vitro* in human dermal fibroblasts (HDF) and *in vivo* in several animal models (db/db mice and Goto-Kakizaki **(GK) rat)** using the corresponding technique (western blot, transitory transfections with reporter gene assay or evaluation of target genes by quantitative RT-PCR). The functional consequence of the notch system modulation was studied *in vitro* by assessment of the migration of HDF and by angiogenesis assay (in human endothelial cells). Notch pathway inhibition was induced either nonspecific by γ -secretase inhibitors (DAPT, L-685,458) or by specific siRNA silencing of the Notch receptors (1-4). The effect of the notch inhibition on wound healing in diabetes was evaluated in db/db mice by local treatment with γ-secretase inhibitors. **Results:** Hyperglycemia activates Notch pathway at multiple levels. The repressive effect of high glucose on migration (fibroblasts) and angiogenesis (endothelial cells) was cancelled by blocking the notch signaling. Specific modulation of Notch receptors (1-4) using siRNA pointed out on a central role of Notch1 in mediating the repressive effect of hyperglycemia. Moreover local treatment with gamma secretase inhibitors improved the wound healing in db/db mice (percentage of wound closure on day 12 was 60 +/- 2% (DAPT), 70 +/- 4% (L-685,458) and 43 +/- 5 % (Placebo) respectively ($p<0.01$). Blocking Notch signaling in diabetic wounds was followed by increase in granulation and epidermal formation, increase in blood vessel formation and increase in expression of SDF-1 alpha responsible for recruitment of EPCs. **Conclusions:** Hyperglycemia activated Notch system and significantly impaired cell migration and angiogenesis. Blocking notch system improved cell migration, angiogenesis and in consequence wound healing rate in diabetic animals.