## ABSTRACT

Notch is an intercellular signaling pathway related mainly to sprouting neo-angiogenesis. The objective of our study was to evaluate the angiogenic mechanisms involved in the vascular augmentation (sprouting/intussusception) after Notch inhibition within perfused vascular beds using the chick area vasculosa and MxCreNotch1(lox/lox) mice. In vivo monitoring combined with morphological investigations demonstrated that inhibition of Notch signaling within perfused vascular beds remarkably induced intussusceptive angiogenesis (IA) with resultant dense immature capillary plexuses. The latter were characterized by 40 % increase in vascular density, pericyte detachment, enhanced vessel permeability, as well as recruitment and extravasation of mononuclear cells into the incipient transluminal pillars (quintessence of IA). Combination of Notch inhibition with injection of bone marrow-derived mononuclear cells dramatically enhanced IA with 80 % increase in vascular density and pillar number augmentation by 420 %. Additionally, there was down-regulation of ephrinB2 mRNA levels consequent to Notch inhibition. Inhibition of ephrinB2 or EphB4 signaling induced some pericyte detachment and resulted in up-regulation of VEGFRs but with neither an angiogenic response nor recruitment of mononuclear cells. Notably, Tie-2 receptor was down-regulated, and the chemotactic factors SDF-1/CXCR4 were up-regulated only due to the Notch inhibition. Disruption of Notch signaling at the fronts of developing vessels generally results in massive sprouting. On the contrary, in the already existing vascular beds, down-regulation of Notch signaling triggered rapid augmentation of the vasculature predominantly by IA. Notch inhibition disturbed vessel stability and led to pericyte detachment followed by extravasation of mononuclear cells. The mononuclear cells contributed to formation of transluminal pillars with sustained IA resulting in a dense vascular plexus without concomitant vascular remodeling and maturation.