Nitric oxide stimulates the EGF receptor-signaling pathway promoting angiogenesis of endothelial cells

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Nitric oxide (NO) obtained from exogenous sources stimulated the Ras-MAP kinases ERK1/2 signaling pathway in rabbit endothelial cells (RAEC). Activation of this pathway also involved the transactivation of the EGF receptor (EGF-R) mediated by ERK1/2 (FRBM 35:381; 2003). Now, we evaluate the effects of endogenously generated NO elicited by bradykinin and fluid laminar "shear stress" on tyrosine phosphorylation of the EGF receptor and in the process of angiogenesis. We found that upon stimulation with bradykinin (0.1 ? M) or under shear stress conditions (16 dynes/cm^2) the endothelial isoform of NO synthase was activated in RAEC and in human endothelial cells (HUVEC). Furthermore, increase in NO production correlated with enhanced phosphorylation on tyrosine residues of the EGF-R as seen by immunoprecipitation and westernblot analysis. To determine the angiogenic capacity of the NO-EGF-R signaling pathway, we used the Matrigel®-based *in vitro* assay for angiogenesis. In addition, we analyzed the signaling pathway involved in the process. We showed that bradykinin and shear stress induced the formation of capillary-like structures by HUVEC grown in Matrigel®. HUVEC expressing a mutant of the EGF-R lacking tyrosine kinase activity did not form capillary-like structures upon stimulation with bradykinin or shear stress conditions. Taken together, these findings suggest that the activation of the NO-EGF-R signaling pathway is necessary to promote angiogenesis in HUVEC.

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