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HA levels were 201.8 experimental MI induced by stable occlusion of the left anterior descending artery.

A 15, 30 and 60 min of further reperfusion (I/R) was performed at the same flow rate used before ischemia. To investigate the involvement of auto-phagy in degradation of Cx43 triggered by ischemia and I/R, hearts were perfused with KH solution containing pepstatin A and NH4Cl. Altogether, this data demonstrates that degradation of protein Cx43 in I/R results from an increased activation of harmful autophagy.

The effect of ischemia and I/R on the total levels of Cx43 was evaluated by Western blotting. I/R, hearts were perfused with KH solution containing pepstatin A and NH4Cl. In the present study, we sought to determine the role of GSK3β as a molecular target for infarct size limitation by administrating 4 novel indirubins, which were respectively treated with 0.0146 mmoles kg\(^{-1}\) of the novel synthetic GSK3β inhibitor MLS2776 (IC50=0.0134 μM), MLS2777 (IC50=0.0554 μM), MLS2778 (IC50=0.11 μM), MLS2779 (IC50=0.033 μM) and BIO (IC50=0.05 μM), given intravenously at the 20th min of sustained ischemic injury. After the end of rep the I/R, hearts were subjected to mitochondrial fractionation followed by mRNA and protein analysis.

Myocardial infarction (MI) is followed by tightly regulated inflammatory responses and remodeling of the extracellular matrix. With respect to the latter, the initial pro-inflammatory matrix that is formed during and after removal of cellular debris and matrix fragments is likely the matrix that affects the stage for myocardial infarction and further matrix remodeling. Furthermore, the matrix has immunomodulatory functions i.e. through activation of toll-like receptors by matrix fragments. Vice versa, inflammatory mediators likely participate in the regulation of early matrix remodeling after MI. One important matrix component is the polysaccharide hyaluronan (HA) which is synthesized by three hyaluronan synthase (HAS) isoforms (HAS1-3). The aim of the present study was to investigate the possible interaction between induction of MI (I) and early remodeling of the myocardial HA-matrix. After experimental MI induced by stable occlusion of the left anterior descending artery (I), the characteristic rise in plasma IL-6 was accompanied by a dramatic increase in plasma HA levels in male C57BL/6J mice 12 h and 24 h post MI. At 12 h HA levels were 201.8±46.8 ng/ha/ml in sham and 1446.0±301.6 ng/ha/ml in MI animals (n=5), similar levels were measured at 24 h post MI. In parallel, HAS1- and HAS2-mRNA expression were markedly induced in the ischemic left ventricular. Furthermore, in murine primary fibroblasts isolated from male C57BL/6J mice, stimulation with IL-6 plus the soluble IL-6 receptor (sIL-6R) induced HAS1- and HAS2-mRNA expression after 6 h as compared to control (HAS1: 33.47±16.77 fold, HAS2: 54.62±1.284 fold, n=3, p < 0.05). HAS3 was only moderately induced (HAS3 1.739±0.3208 fold of control, n=3). Concomitantly, the amount of HA secreted into the medium was increased by IL-6 + sIL-6R stimulated fibroblasts. Next it was investigated whether a causal relationship exists between IL-6 and upregulation of myocardial HAS-isoenzyme expression and elevated HA plasma levels. For this purpose mice were treated with a neutralizing anti-IL-6 antibody (250 μg/mouse) prior to MI. This prevented the induction of HAS1 and HAS2 gene expression in the ischemic left ventricle and markedly reduced HA plasma concentrations (12h sham: 403.6±72.57 ng H/ml vs. 12 h post MI: 279.3±66.61 ng H/ml, n=5).

In conclusion, the present data strongly suggest (i) that the induction of HA-rich matrix early after MI is driven by IL-6 and (ii) that HA released into the circulation may at least in part be derived from cardiac fibroblasts through HAS1 and HAS2 in response to IL-6 stimulation.

Investigating the role of GSK3β as a molecular target for myocardial infarct size reduction by using novel pharmacological inhibitors

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