Hyperhomocysteinemia Exaggerates Adventitial Inflammation and Angiotensin II-induced Abdominal Aortic Aneurysm Formation in Mice

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Despite epidemiological study has revealed an association between hyperhomocysteinemia and abdominal aortic aneurysm (AAA), there is no direct evidence supporting the causal role between each two. Therefore, by use of an Ang II-infused ApoE⁻/⁻ mice, we test the hypothesis that homocysteine contribute to AAA etiology. Homocystine supplement in drinking water resulted in a mild hyperhomocysteinemia in mice, with a plasma concentration around 25 µM. Intriguingly, hyperhomocysteinemia dramatically increased the incidence and severity of Ang II -induced aortic dissection and AAA formation (homocysteine vs. vehicle: 100% vs. 50%, n=16-19 per group, \(P<0.05\)). Histological studies have indicated a markedly exaggerated adventitial inflammation. The increased proinflammatory cytokine IL-6 and MCP-1 were mainly colocalized within adventitial fibroblast (AF) and macrophage, indicating the importance of adventitial fibroblast activation during pathogenesis of homocystine-deteriorated AAA. Indeed, homocysteine in vitro sequentially stimulated AF transformation into myofibroblast, subsequent secretion of MCP-1 and IL-6, and consequently recruitment of monocyte toward AF, which was abolished by NADPH oxidase inhibitor DPI, indicating NADPH-derived ROS mediated homocysteine enhanced AF activation. Further studies revealed that Nox4, but not other homolog of NADPH, was significantly upregulated by homocysteine in AF, whereas Nox4 silencing by siRNA diminished homocystine-induced ROS production, myofibroblast transformation, and recruitment of monocyte. Moreover, mechanistic study has uncovered a quickly p-Smad2/3 activation upon homocysteine changeling. Specific knockdown of Smad2 or Smad3 circumstanced Nox4 upregulation and AF activation. Taken together, our study suggests a promising therapeutic strategy of lowering homocysteine level or alleviating adventitial inflammation for prevention and treatment of clinical AAA patients.