augmented. These effects are mediated via PLD-dependent, NADH/NADPH oxidase-sensitive pathways. Thus, increased PLD activity is associated with increased oxidative stress that may underlie enhanced VSMC growth in human hypertension.

Key Words: Oxidative stress; signal transduction; essential hypertension; angiotensin II

B004
UPREGULATION OF SRC PLAYS AN IMPORTANT ROLE IN ALTERED VASCULAR FUNCTION IN GENETIC HYPERTENSION
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We investigated the role of c-Src, a non-receptor tyrosine kinase, in Ang II-mediated Extracellular signal-regulated kinases (ERK1/2) dependent signaling pathways and associated cell growth in vascular smooth muscle cells (VSMCs) from SHR. Mesenteric VSMC, passages 1–4, from WKY and SHR were studied. Activation of c-Src and ERK1/2 were assessed by Western blot using phospho-specific antibodies, and DNA synthesis, index of VSMC proliferation, was determined by measuring 3H-thymidine incorporation. Receptor subtypes through which Ang II mediates effects were determined with the AT1 receptor antagonist, irbesartan, and the AT2 receptor blocker, PD123319. Ang II rapidly increased c-Src phosphorylation with maximal responses obtained within 60 secs of stimulation. These actions were blocked by the selective Src inhibitor, PP2 (10^{-5} mol/L). Ang II-induced activation of c-Src was significantly greater (p < 0.02) in cells from SHR (314 ± 30% of control) than with WKY (191 ± 25% of control). Basal and Ang II-stimulated ERK1/2 phosphorylation was augmented (p < 0.01) in SHR (421 ± 41% vs 172 ± 41%, SHR vs WKY). Effects were abolished by the selective MEK1/2 inhibitor, PD98059 (10^{-5} mol/L). PP2 inhibited Ang II-induced activation of ERK1/2 in WKY (88 ± 18%) and normalized responses in SHR (113 ± 8%). Ang II dose-dependently increased 3H-thymidine incorporation in VSMCs, with greater responsiveness (p < 0.05) in SHR (F_{max} = 311 ± 24% vs WKY (F_{max} = 206 ± 38%). These effects were reduced (p < 0.01) in cells pretreated with PP2 or PD98059. Irbesartan, but not PD123319, abolished Ang II-induced actions. Thus AT_{1}-stimulated c-Src, ERK1/2 and associated DNA synthesis are increased in VSMCs from SHR. Altered regulation of AT_{1}-activated c-Src may be an important upstream modulator of enhanced ERK1/2-dependent VSMC growth in genetic hypertension.

Key Words: Signal transduction; angiotensin II; extracellular signal-activated kinase; PI3 Kinase

B005
PI3 KINASE REGULATES ANG II-STIMULATED ERK ACTIVITY IN VASCULAR SMOOTH MUSCLE CELLS FROM SHR
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This study investigated the role of Ang II in the regulation of extracellular signal-regulated kinase (ERKs) in vascular smooth muscle cells (VSMC) from mesenteric arteries of spontaneously hypertensive rats (SHR) and Wistar Kyoto rats (WKY). ERK activity was determined by Western blot using a phospo-specific ERK antibody. To determine the regulatory role of intracellular Ca2+ in Ang II-stimulated ERK activity, VSMC were exposed to BAPTA-AM (10–5 M). To assess whether PI3 kinase is an upstream regulator of Ang II-activated ERKs, VSMC were treated with a specific PI3 kinase inhibitor, LY294002 (10–5 M). In the basal state, ERK activity, but not protein concentration, was significantly increased (p < 0.05) in SHR (68.9 ± 28.8 units) compared with WKY (21.1 ± 12.2 units). Ang II dose-dependently increased activation of ERK, with the response significantly augmented in SHR (195.4 ± 23.6 units) compared to WKY (81 ± 19 units, p < 0.05). ERK activation was maximal at 5 min in both strains. Responses were sustained at suprabasal levels for up to 20 min in SHR but not in WKY. BAPTA-AM attenuated Ang II-induced effects similarly in VSMC from WKY and SHR. PI3 kinase inhibition completely blocked Ang II-stimulated ERK activation in SHR but not in WKY. Thus intracellular Ca2+ is an important regulator of Ang II-stimulated ERK activity. In VSMC from SHR, augmented Ang II-induced activation of ERKs may be due to PI3 kinase-dependent mechanisms.

Key Words: Signal transduction; angiotensin II; extracellular signal-activated kinase; PI3 Kinase

B006
MOLECULAR AND SURVIVAL EFFECTS OF AMLODIPINE ON ENDOTHELIAL CELLS STRESSED WITH H2O2
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Cellular stress elicits biochemical responses that either enhance cells survival or lead to cells death or apoptosis. The Ca2+ involvement in apoptosis was demonstrated by several experimental studies using calcium channel blockers (CCBs). Thus, Ca2+ increasing could be an important trigger mechanism for apoptosis. However, the CCBs effects on apoptosis, cells proliferation, cells death induced by oxidative stress remains still unclear. The aim of our study was to assess the effects of amlodipine (AML) (a dydropyridine CCBs) on hembulcal vein endothelial cells (HUVEc) stressed with H2O2 (2 mM). We tested the cells death by triphan bleu, and apoptosis by annexin and caspases. The oxidative stress (2 hrs of H2O2 administration) on HUVEc induced a 22% of cells death (Fig. A) and a 20% of apoptosis...