

[gw22-e0312]

THE EFFECTS OF SPLA2-IIA IN HUMAN UMBILICAL VEIN ENDOTHELIAL CELLS

Tan Donghan, He Mingyue, Chen Lianfeng, Fang Quan Peking Union Medical College Hospital, Dongcheng, Beijing, China

10.1136/heartjnl-2011-300867.169

Background Secretory phospholipase A₂ group IIA (sPLA₂-IIA) appears to be an important inflammatory mediator of cardiovascular disease and may play a pivotal role in the pathophysiology of AS. To understand whether the influence of sPLA₂-IIA on proinflammatory effects in vascular endothelial cells may help comprehend the mechanism induced by the sPLA₂-IIA involved in AS.

Objectives To investigate the effect of different concentrations of sPLA₂-IIA on Endothelial Cells, and to investigate the signalling pathway.

Methods (1) HUVEC were cultured in three different concentrations groups (0.01, 0.1, 1 ug/ml) of sPLA₂-IIA. Endothelial cell nitric oxide concentration in the supernatant was detected by kit. The mRNA expression levels of ET-1, eNOS, ICAM-1, VCAM-1 were determined by Real Time-PCR. The protein expression level were measured by Western Blot or by ELISA. (2) HUVEC cells were cultured with 1 ug/ml sPLA₂-IIA and 10 nmol/l 4-BPB (Hydrolysis inhibitor) or 1 umol/l PD98059 (ERK1/2 inhibitor) or SP600125 (JNK inhibitor) or SB203580 (p38 inhibitor) or Bay11-7085 (NF-kB inhibitor) alone or combined. Parameters described in Method step 1 were measured.

Results (1) sPLA₂-IIA increased the mRNA and protein expression of ICAM-1, VCAM-1 and ET-1 and decreased the level of NO in a concentration dependent way. 1 ug/ml of sPLA₂-IIA dramatically decreased the mRNA expression of eNOS. The protein expression of eNOS was not affected by sPLA₂-IIA. (2) 4-BPB abolished the over expression of mRNA and protein of ICAM-1, VCAM-1 and ET-1 induced by the sPLA₂-IIA. 4-BPB also abolished the up-regulated mRNA of eNOS and reduced the down-regulation of NO induced by sPLA₂-IIA. (3) PD98059 abolished the over expression of ICAM-1, VCAM-1 and ET-1 and reduced the down-regulation of NO induced by the sPLA₂-IIA. (4) SP600125 abolished the over expression of ET-1 and reduced the down-regulation of NO induced by sPLA₂-IIA. (5) SB203580 had little effect on the over expression of ICAM-1, VCAM-1 and ET-1 and down-regulation of NO induced by sPLA₂-IIA. 6. Bay11-7085 abolished the over expression of ICAM-1, VCAM-1 and ET-1 induced by the sPLA₂-IIA.

Conclusions (1) sPLA₂-IIA induced the over expression of cytokines including ICAM-1, VCAM-1 and ET-1 in a concentration dependent manner or the repression of eNOS and NO. (2) Hydrolysis of sPLA₂-IIA exactly take a part to regulate the expression of ICAM-1, VCAM-1, ET-1 and eNOS in endothelial cells. (3) sPLA₂-IIA affect endothelial cells adhesion function primarily via ERK1/2 and NF-kB pathway. (4) sPLA₂-IIA affect endothelial cells vasomotor function primarily via ERK1/2, JNK and NF-kB pathway.