Effects of Fluid Shear on Endothelial Cell Response to Inflammation

by

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List of Abbreviations

ACD	Acetate-citrate-dextrose
BAECs	Bovine aortic endothelial cells
CAM	Cell adhesion molecule
CB1R, CB2R	Cannabinoid Receptor 1 and Cannabinoid Receptor 2
CVD	Cardiovascular Disease
CHX	Cycloheximide
DPBS	Dulbecco's phosphate buffered saline
EC	Endothelial cells
FITC	Fluorescein isothiocyanate
HAECs	Human aortic endothelial cells
HSA	Human serum albumin
HUVECs	Human umbilical vein endothelial cells
I/R	Ischemia/reperfusion
ICAM-1	Intercellular cell adhesion molecule-1
IL-1R	Interleukin-1 receptor
IL-1β	Interleukin-1beta
LPS	Lipopolysaccharide
MMP	Matrix metalloproteinase
NF-κB	Nuclear factor kappa B
NK	Natural killer
PECAM-1	Platelet-endothelium cell adhesion molecule-1
PPFC	Parallel plate flow chamber
PS	Preshear, or precondition
RBCs	Red blood cells
RFI	Relative fluorescent intensities
SF	Shape factor
TNFR	Tumor necrosis factor receptor
TNFα	Tumor necrosis factor alpha
VCAM-1	Vascular cell adhesion molecule-1
Δ^9-THC	Delta-9-tetrahydrocannabinol

ABSTRACT

Effects of Fluid Shear on Endothelial Cell Response to Inflammation

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The endothelium is a thin monolayer of cells (*e.g.* endothelial cells, ECs) that regulates several important processes including maintaining blood pressure, clotting, angiogenesis, barrier function, and inflammation. Inflammation, a natural process by which foreign particles are eliminated from vascular tissue, is of considerable importance due its prevalent role in a number of disease pathologies. Endothelial response to inflammation results in the differential expression of cell adhesion molecules, such as E-selectin, on the apical surface facing the bloodstream. Patterns and duration of adhesion molecule expression is critical to the balance between healthy and pathogenic inflammatory response. Chronic inflammation, due to endothelial dysfunction leads to a number of diseases, which include neurological disorders, cancers and metastatic tumor growths, and cardiovascular diseases. Despite its prominent roles in the disease pathogenesis, endothelial response has yet to be fully understood due to current *in vitro*

models failing to fully replicate relevant endothelial inflammatory response under human physiological conditions.

The influence of fluid shear stress on E-selectin expression due to inflammatory activation is investigated through simultaneous co-stimulation with fluid shear and cytokine interleukin-1β of naïve and preconditioned ECs using a novel laminar flow apparatus designed to study the broader time frame over which chronic inflammation is relevant. Naïve cells exposed to shear-cytokine activation display high E-selectin expression for up to 24 hr with peak expression occurring after 8-12 hr of activation contrary to the commonly observed 4-6 hr peak in statically activated cells. High shear preconditioned cells exhibited either elevated or muted E-selectin expression during acute and chronic time frames, respectively, depending on the preconditioning and subsequent shear-cytokine activation durations. The 8-12 hr peak E-selectin expression time in shear-cytokine activated cells coincides with the time frame observed for shear-exposed ECs to shift from a cobblestone to elongated morphology, highlighting a role for cell morphology determining EC response. Overall, the presented data suggest that a high laminar shear enhances acute EC response to interleukin-1ß in naïve ECs as may be found in the pathological setting of ischemia/reperfusion injury while in preconditioned ECs, high laminar shear confers rapid E-selectin downregulation to protect against chronic inflammation. However, high laminar shear is protective against TNFα-induced acute and chronic inflammatory response.