

Introduction Endothelial dysfunction is one of the earliest pathological features in atherosclerosis. Our understanding of endothelial cell biology is derived mainly from studies of human umbilical vein endothelial cells (HUVECs). However HUVECs are not involved in atherosclerosis and provide only limited insight into the pathogenesis of coronary artery disease. We describe a novel method for the isolation of coronary artery endothelial cells from thrombectomy specimens obtained during the treatment of patients for acute myocardial infarction (MI).

Methods Patients presenting with ST-segment elevation MI (n=15) who underwent emergency percutaneous coronary intervention (PCI) and thrombus aspiration were recruited. Thrombus specimens were manually dissected, plated onto collagen-I coated plates and maintained in endothelial cell-specific growth media to encourage coronary endothelial outgrowth (CEO) cells. Growth kinetics were evaluated and phenotype was confirmed by immunocytochemical staining for vonWillebrand factor (vWF). Multiparameter flow cytometric analysis was performed (CD31-FITC, KDR-PE, CD146-PE/Cy7 and CD34-APC/Cy; percent positive expression) and angiogenic potential was assessed using an established assay of tubule formation on a matrigel matrix.

Results Outgrowth of coronary artery endothelial cells was observed in 9/15 samples. CEO cells were maintained for a minimum of 38 days in culture (mean days=45±12). Population doubling times of CEO cells were comparable to HUVECs (mean ±SD: CEO 2.6±0.5, HUVEC 2.3±0.1; p=0.50 student's t-test). CEO cells had typical cobblestone morphology and expressed vWF. Surface expression of CD31 and KDR was comparable in both cell types (CD31 mean±SD: CEO 74.9±21.3 vs. HUVEC 89.3±11.4, p=0.13; KDR mean±SD: CEO 46.7±31.7 vs. HUVEC 22.2±15.9, p=0.11) whereas CD146 and CD34 expression was increased in CEO cells (CD146 mean±SD: CEO 96±5.3 vs. HUVEC 73.5±30.7, p<0.0001).

Conclusion Viable coronary arterial endothelial cells can be isolated from thrombus extracted during emergency PCI. These cells have a mature endothelial phenotype comparable to HUVECs, but have reduced angiogenic potential suggesting they retain the functional characteristics of *in situ* endothelium. This novel approach to isolate dysfunctional endothelial cells may have applications in drug screening and future studies may provide additional insight into the cellular and molecular basis of endothelial dysfunction in patients with coronary artery disease.