

Exposure to Platelets Affects the Function of Endothelial Progenitor Cells

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Background: Endothelial progenitor cells (EPC) are bone marrow-derived cells that are mobilized into the circulation in response to tissue or vascular injury. Recent evidence suggests that EPCs have an important role in repair following vascular injury, and that platelets mediate their recruitment to sites of injury. Platelets and EPCs can interact directly via P-selectin – PSGL1 binding, however, the effect of platelets on EPC function remains unclear. Therefore, in this study we aimed to assess the in-vitro effect of platelets on the capacity of EPCs to form colonies, differentiate, migrate and proliferate.

Methods: Human EPCs were isolated from donated Buffy coats and purified on a magnetic separation column specific for CD133 antigen. They were incubated either on traditional fibronectin matrix, or co-incubated with washed platelets (isolated from healthy volunteers), for 7 days. Number of EPC colony forming units (CFU) was quantified, and endothelial cell lineage confirmed by immunostaining with antibodies directed against VEGFR-2, CD31 and Tie-2. Functional properties of the cultured cells were evaluated by MTT - proliferation assay and migration assay using the Boyden chamber.

Results: Co-incubation of EPCs with platelets compared to incubation of EPCs alone (on fibronectin matrix) resulted in a higher number of CFUs after 7 days of culture (6.5 ± 1.3 CFUs/well vs. 3.5 ± 0.5 CFUs/well, respectively, $P < 0.05$). In addition, co-incubation of EPCs with platelets vs. incubation of EPCs alone was associated with higher proportion of living cells, tested by the MTT assay (0.2 ± 0.01 vs. 0.12 ± 0.04 MTT 570nm respectively, $P < 0.05$), and higher number of migrated EPCs, assessed by the migration assay (14 ± 2.12 vs. 5.8 ± 1.8 migrated cells $\times 10^4/20000$ cells, respectively, $P < 0.001$)

Conclusion: In-vitro exposure to platelets promotes the capacity of EPCs to form colonies, proliferate and migrate. Therefore, the interaction with platelets appears to augment EPC functional properties.