Effects of neutrophil ERK/MAPK phosphorilation inhibition on cell adhesion to HUVECs and PMN surface molecules expression

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MAPK family constitutes the major pro-inflammatory intracellular signaling pathway. One of its main groups is ERK, involved in growing, proliferation, and cell survival. This path has been related to cell adhesion and transmigration processes, and implicated in neutrophil (PMN) CD11b/CD18 expression regulation. The study of ERK/MAPK influence on cell adhesion and PMN surface molecules expression was performed using a selective inhibitor of ERK phosphorilation, PD-98059. Adhesion assays were performed with isolated PMNs co-incubated with cultured HUVECs for 60 minutes, and quantified by colorimetric method. Prior to adhesion, PMNs were incubated for 30 minutes with 50µM PD-98059 (n=8). Surface molecules expression was evaluated using flow cytometry techniques. Whole blood was co-incubated with pre-marked antibodies against CD11a, CD11b, CD18 e CD62-L, with or without PD-98059, 20 or 50 µM. Adhesion results showed decreased adherence of PD-98059-incubated PMNs in comparison to the incubation control ($p<0,01$), indicating that MEK/ERK pathway may be important to leukocyte adhesion to endothelial cells. Surprisingly, CD11b demonstrated a dose-dependent increased expression ($p=0,033$) in the presence of PD-98059, while none of the other molecules showed any significant differences. These apparently controversial results may be due to intracellular mechanisms which might have a compensatory role for ERK inactivation, once its activation is responsible by several normal cell functions. Possible mechanisms include NADPH oxidase lower activation by the decreased phosphorilation levels in the cell in presence of PD-98059, thus reducing superoxide production and increasing relative NO availability, providing an anti-adhesive environment independent of cell surface adhesion molecules expression.

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