





TOP > Available Issues > Table of Contents > Abstract

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A possible role of phospholipase C and phospholipase D in chemotaxis

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Abstract Cell chemotaxis plays a role in many cellular or individual actions suited to accomplishing a variety of purposes. These actions include activities of bacteria and Protista, cellular slime molds' migration to food, starvation-induced aggregation of cellular slime molds and mobilization of immunocytes to infectious or inflammatory sites in higher order animals. Macrophages, neutrophils, C3H10T1/2 clone 8 (10T1/2 (SIGMA, USA)) cells, etc. are chemotactic cells. Macrophages and neutrophils are terminally differentiated cells, while 10T1/2 cells are undifferentiated mesenchymal cells. However, currently there is almost no research in which chemotaxis of terminally differentiated cells and undifferentiated mesenchymal cells are comparatively evaluated. Therefore, in this article, the effects of phospholipase C (SIGMA, USA) or phospholipase D (SIGMA, USA) on chemotaxis were evaluated using these 3 types of cells in different differentiation states. The chemotaxis of each cell was evaluated using D609 (SIGMA, USA) and ET-18-OCH₃ (SIGMA, USA) (which are phospholipase C inhibitors), and suramin (SIGMA, USA) and D-erythro-Sphingosine (SIGMA, USA)(which are phospholipase D inhibitors). As chemotactic factors, ZAS was used for macrophages and neutrophils, and platelet-derived growth factor (PDGF) was used for 10T1/2 cells. The chemotaxis of each cell was assessed using 96hole chemotaxis chambers. Firstly, chemotactic factors were added only to the lower chambers, and chemotaxis was then measured. In macrophages, D609, ET-18-OCH₂, suramin and D-erythro-Sphingosine significantly inhibited chemotactic activity. In neutrophils and 10T1/2 cells, D609 and ET-18-OCH₃ significantly inhibited chemotactic activity. To study these inhibiting mechanisms, further evaluation was performed. ET-18-OCH₃ significantly and dose-dependently inhibited random migration when chemotactic factors were added to both upper and lower chambers. However, ET-18-OCH₃ did not

significantly affect chemokinesis where no chemotactic factor had been added. Secondly, in 10T1/2 cells, D609 significantly reduced random migration and chemokinetics, but ET-18-OCH $_3$ did not significantly change random migration and chemokinesis, even though it slightly reduced them.

Key words 10T1/2, Chemotaxis, Phospholipase, Neutrophil

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