

Comparative Proteomic Analysis between Neutrophils Primed by PAF and Activated by fMLP

Aquino,E.N¹; Neves,A.C.D¹; Sousa,M.V¹; Castro,M.S¹; Fontes,W¹.

¹Centro Brasileiro de Pesquisas em Proteínas, Laboratório de Bioquímica e Química de Proteínas, UnB, Brazil.

Neutrophils are cells of the innate immune system that exhibit three different phenotypes: quiescent, primed and activated. Several proinflammatory molecules can induce the primed and activated state, including the PAF and fMLP. The PAF, a proinflammatory lipid mediator, induces only priming in neutrophils, however fMLP, a formylated tripeptide derived from bacterial cell walls, can induce both priming and activation, in a dose-dependent way. Both have G-protein coupled receptors, but with different activation pathways. The aim of this study was to compare the proteomic map of neutrophils primed by PAF and neutrophils activated by fMLP and compare both stimulation situations to quiescent neutrophils. Neutrophils from healthy donors were isolated, stimulated by each agent, lysed and the protein extract submitted to 2D-PAGE. Silver stained gel images were processed using ImagemMaster 2DPlatinum. The preliminary comparison between stimulated and quiescent conditions showed that of the 115 spots absent in PAF, 33 were also absent in fMLP, 5 were upregulated in fMLP and 4 downregulated. Of the 43 upregulated in PAF, 8 were absent in fMLP, 5 were also upregulated in fMLP and 2 were downregulated. Of the 10 spots downregulated in PAF, 2 were absent in fMLP, 1 was also downregulated in fMLP and no spots were upregulated. After directly matching PAF and fMLP conditions, 169 spots revealed to be upregulated by PAF stimulation and 78 spots were upregulated by fMLP stimulation, among them 16 proteins identified. This preliminary study shows that there are many differentially expressed proteins between these conditions, that might lead to further elucidation of the differences among the pathways, although both share the same class of receptors.

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