

THE MIDWINTER CONFERENCE OF IMMUNOLOGISTS POSTER ABSTRACT - 2005

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Detection of phospholipase activity in the cytotoxic granules of T and NK lymphocytes. ³

Bryce Alves, William H. Welch, David Tamang, Doug Redelman, Dorothy Hudig. Molecular and Cellular Biosciences. Mail stop 320, University of Nevada, Reno NV. 89557

The cytotoxic granules of T and NK lymphocytes are extremely potent effectors of cellular apoptosis. We examined whether some of the unknown granule proteins could augment to the deadly processes mediated by perforin and granzymes. Phospholipases can increase membrane permeability and/or mediate damage directly. We used the new fluorogenic umbelliferone substrate, O-pivaloyloxymethyl umbelliferone, and observed soluble phospholipase activity from the dense granules of both rat RNK-16 and mouse cytotoxic T cells. The hydrolytic activity from rat granules of about ~12.5 million NK cells was 2 picomoles per minute and equivalent to 4 ug of Pseudomonas sp. phospholipase C (Sigma L9656). The phospholipase activity is extractable with 1 M NaCl, has standard Michaelis-Menten kinetics, and is calcium independent and active with 10mM EGTA. We raise the possibility that this lipase is a functional component of the cytotoxic granules that maybe secreted upon stimulation and may play some role in the clearance of infected of infected cells. Supported in part by NIH R01CA38942.