ANALYSIS OF MITOCHONDRIAL FUNCTION IN ANTIGEN SPECIFIC NAÏVE, EFFECTOR AND MEMORY CD8⁺ T CELLS

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INTRODUCTION. CD8⁺ T cells play a critical role in the clearance of intracellular pathogens such as viruses and certain bacteria. Before encounter of antigen these cells are naïve. After antigen encounter they become activated, begin to proliferate and acquire effector functions including cytokine production and cytotoxicity. These cells are effector cells. After clearance of the pathogen a death phase ensues. During this time 90-95% of the antigen specific effector cells will undergo apoptosis. The surviving cells are memory cells which can persist at constant numbers for the life of the mouse (1) (2). These cells will rapidly respond to a secondary infection and provide the basis for vaccination.

The mechanisms that control the contraction of the CD8 response remain poorly understood. Because loss of mitochondrial potential is a hallmark of cells preparing to undergo apoptosis we examined mitochondrial potential in antigen specific cells as they passed through all phases of a CD8 response.

METHODS. LCMV infection of mice has been described previously (3). To determine mitochondrial potential splenocytes were incubated in 40 nM 3,3'-dihexyloxacarbocyanine iodide (DiOC6(3)) for 30 minutes. After incubation with (DiOC6(3)) cells were washed once and stained with anti-CD8 α -PE and L^dNP118-126-APC tetramer for 30 minutes on ice. The samples were washed three times and acquired on a FACSCalibur instrument.

RESULTS. Figure 1 shows that naïve CD8⁺ T cells contained a basal mitochondrial potential with a mean fluorescent intensity of 230. After infection, the mitochondrial potential is increased in early effector T cells on day 5 post-infection, but by day 8 the potential in late effectors has returned to a level comparable to naïve cells.

Antigen specific cells in the death phase (day 13) contained a decreased potential relative to naïve cells. The potential in memory cells was indistinguishable from that of antigen specific cells during the death phase.

DISCUSSION. In this short report we present analysis of mitochondrial function in antigen specific cells as they progress from naïve to effector to memory cells. We confirm previous studies that T cells undergoing apoptosis contain a lower mitochondrial potential (4,5) and extend these findings to show that the potential in memory cells does not return to the level found in naïve cells. These findings suggest that memory cells contain some factor that allows them to survive despite a decreased membrane potential.

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