THE EGYPTIAN JOURNAL OF IMMUNOLOGY Vol. 15 (1), 2008
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Epstein-Barr virus-transformed B-cells as efficient antigen presenting cells to propagate Aspergillus-specific cytotoxic T-lymphocytes

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To overcome the cytotoxic T-lymphocytes (CTL) expansion limitations imposed by the lack of sufficient dendritic cells (DC) alternative sources of autologous antigen presenting cells (APC) such as Epstein-Barr virus (EBV)-transformed B-lymphoblastoid cell lines (BLCL), which are easy to establish in vitro, have been considered and studied in the present work. Non-adherent peripheral blood mononuclear cells of three healthy donors were repeatedly primed with autologous Aspergillus fumigatus commercial culture-filtrate antigen-pulsed fast monocyte-derived DC (Aspf-CFA-DC) alone, Aspf-CFA-pulsed BLCL (Aspf-CFA-BLCL) alone or Aspf-CFA-BLCL after one, two, or three primings with Aspf-CFA-DC (1DC/BLCL, 2DC/BLCL or 3DCIBLCL; respectively). After 5th priming, lines generated by Aspf-CFA-BLCL only showed strong/weak lytic activity for EBV/Aspf; respectively. Aspf-specific lytic activity in all donors was increased by increasing the number of primings with Aspf-CFA-DC before switching to Aspf-CFA-BLCL (18.20 +/- 1.65% versus 35.67 +/- 1.02% and 40.03 +/- 1.41% in bulk cultures generated by 1DC/BLCL versus 2DC/BLCL and 3DC/BLCL, respectively). Bulk cultures generated by Aspf-CFA-BLCL after at least two primings with Aspf-CFA-DC showed approximately the same Aspf-specific lytic activity, effector cell phenotype, expansion level and percentage expression of IFN-gamma, CD69 and CD107a without any significant differences ($p > 0.05$) as standard bulk cultures generated by only Aspf-CFA-DC. Thus, this study explored the use of a combined DC/BLCL protocol to establish/propagate Aspf-specific CTL for adoptive immunotherapy to prevent or treat invasive pulmonary aspergillosis.