Cytometry B Clin Cytom 2014 Jul;86(4):257-62 Epub 2013 Sep 3

PMID: 24002818 DOI: 10.1002/cyto.b.21124

Multivariate statistical data analysis as a tool to analyze ex vivo expansion dynamics of cytokine-induced killer cells

Cristina Zanon, Matteo Stocchero, Elena Albiero, Silvia Castegnaro, Katia Chieregato, Domenico Madeo, Francesco Rodeghiero, Giuseppe Astori

Abstract

Background: Cytokine-induced killer (CIK) cells, obtained after mononucleated cell stimulation with interferon-γ, interleukin-2, and anti-CD3 antibody, are constituted by CD3(+) CD56(+) (CIK) cells and a minority of natural killer (NK; CD3(-) CD56(+)) cells and T-lymphocytes (CD3(+) CD56(-)) with antitumor effect against hematological malignancies, thus representing a promising immunotherapy strategy. To ensure in vivo antitumor activity it is mandatory to maximize the percentage of CD3(+) 56(+) effector cells, which is highly variable depending on the starting sample and the harvesting day. Based on cytofluorimetric data, we have retrospectively applied multivariate statistical data analysis (MVDA) to 30 expansions building mathematical models able to predict the expansion fate and the optimal CIK harvesting day.

Methods: Cell phenotype was monitored during culture; multivariate batch statistical process control was applied to monitor cell expansion and orthogonal projections to latent structures to predict CIK percentage.

Results: Ten expansions had CD3(+) CD56(+) cells ≥ 40% (good batches) and 20 had CD3(+) CD56(+) cells ≤ 40%. In 36.7%, CD3(+) CD56(+) cells reached the highest concentration at day 17 and the others at day 21. We built a highly predictive regression model for estimating CD3(+) CD56(+) cells during culture. Three variables resulted highly informative: NK % at day 0, cytotoxic T-lymphocytes % (CTLs, CD3(+) CD8(+)) at day 4, and CIK % at day 7. "Good batches" are characterized by a high percentage of CTLs and CD3(+) CD56(+) cells at day 4 and day 7, respectively.

Conclusion: By applying MVDA it is possible to optimize CIK expansion, deciding the optimal cell harvesting day. A predictive role for CTL and CIK was evidenced.