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Jagannadha K. Sastry, Ph. D.
Assistant Professor
Department of Veterinary Sciences
UTMDACC, Science Park
Bastrop, TX

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Our proposed experiments included: (a) immunizing mice with synthetic peptides, (b) preparing spleen and lymph node cells, (c) growing them under conventional conditions as well as in the rotatory vessel in appropriate medium reconstituting with synthetic peptides and/or cytokines as needed and (d) comparing at regular time intervals the specific CTL activity as well as helper T-cell activity (in terms of both proliferative responses and cytokine production) using established procedures in my laboratory. We further proposed that once we demonstrated the merit of rotatory vessel technology to achieve desired results, these studies would be expanded to include immune cells from non-human primates (rhesus monkeys and chimpanzees) and also humans.

We conducted a number of experiments to determine CTL induction by the synthetic peptides corresponding to antigenic proteins in HIV and HPV in different mouse strains that express MHC haplotypes H-2b or H-2d. We immunized mice with 100 ug of the synthetic peptide, suspended in sterile water, and emulsified in CFA (1:1). The immune lymph node cells obtained after 7 days were restimulated by culturing in T25 flask, HARV-10, or STLV-50, in the presence of the peptide at 20 ug/ml. The results from the ^{51}Cr-release assay consistently revealed complete abrogation of CTL activity of cells grown in the bioreactors (both HARV and STLV), while significant antigen-specific CTL activity was observed with cells cultured in tissue culture flasks.

This data showing abrogation of CTL activity of cells cultured in the bioreactors under the in vivo mimicking 3D-conditions was rather surprising to us. However, there were few reports in the literature regarding diminished cellular immune responses observed in people after either short- or long-term space travel. But, none of these reports dealt with studies related to either antigen-specificity or CTL activity. Therefore, our results, though disappointing with respect to our intention of using the bioreactor technology for enhancing immune function, are new and informative. We therefore proposed new studies for the TMC-NASA contract round II application, to understand the mechanism of this observed loss of immune function for cells cultured in microgravity. Thus far, it appears that this loss in immune function under microgravity relates to loss of a particular type of immune cell population called antigen-presenting cells. We are conducting detailed studies to further understand this phenomenon.

Thus, overall the data we generated in this study proved the usefulness of the NASA-developed bioreactor technology for understanding the known immune deficiency during space travel. Additionally, this ex vivo microgravity technology since it mimics effectively the in vivo situation, it is also useful in understanding immune disorders in general. Thus, our proposed studies in TMC-NASA contract round II application benefit from data generated in this TMC-NASA contract round I study.

ROUTE 2, BOX 151-B1 • BASTROP, TEXAS 78602 • (512) 321-3991
FAX (512) 332-5208