

GLUTATHIONE DEPLETION OF STIMULATOR CELLS INHIBITS RESPONDER T CELL IMMUNOGENICITY *IN VITRO* AND PROLONGS ALLOGRAFT SURVIVAL *IN VIVO*

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Objective: To explore the impact of donor cell redox state alteration on allograft immunogenicity.

Background Data: Pretransplant donor organ immunomodulation may attenuate transplant graft rejection allowing for reduction in immunosuppression. As most immunological processes including up-regulation of cell adhesion molecules are redox regulated, altering the redox state of the organ donor cells by glutathione depletion could attenuate donor cell alloreactivity.

Methods: Splenic cells and heart endothelial cells (EC) from Balb/C mice were treated in the presence of 0-1 mM diethylmaleate (DEM, a glutathione depleting agent) and /or 10 μ g /ml of LPS to assess the impact of glutathione depletion on alloreactivity (via mixed lymphocyte reaction using responder T-cells from major histocompatibility complex-disparate C57BL/6 mice), EC adhesion (via T-cell adhesion assay), ICAM-1 expression (via RT-PCR and semiquantitative analysis) and NF- κ B upregulation (via electrophoretic mobility shift assay, EMSA). Glutathione levels were verified through spectrophotometric assays. Heterotopic heart transplants (a model of solid organ transplantation) were performed as an *in vivo* correlate.

Results: Glutathione depletion decreased donor EC and splenic cell alloreactivity ($p < 0.005$), decreased LPS induction of EC ICAM-1 expression through attenuation of NF- κ B activity, and decreased T-cell adhesion to ECs in response to LPS stimulation ($p < 0.001$). These *in vitro* results correlated with a prolongation of DEM treated allograft survival in heterotopic heart transplants compared with untreated controls ($p < 0.001$).

Conclusions: Glutathione depletion may represent a significant immunomodulator of donor antigenicity in the prevention of transplant rejection.