D Mevorach
Hadassah-Hebrew University, Medicine, Jerusalem, Israel

Introduction:
Allocetra™, donor leukocytes containing early apoptotic cells and no necrotic cells, was shown as safe and potentially efficacious for the prevention of aGVHD (Mevorach et al. BBMT 2014). We tested the effects of early apoptotic cells on cytokines/chemokines of patients with aGVHD, and in mice treated with LPS and IFN-g.

Methods:
LPS and IFN-g were used to trigger cytokine/chemokine release in vitro and in vivo in mice, and in patients treated for aGVHD. Cytokines/chemokines were evaluated in 13 patients. Mouse and human IL-1β, IL-2 to 10, IL-12p70, IL-13, IL-15, IL-17A, IL-22, IL-27, IL-31, IL-32, IP-10, RANTES, GRO, IFN-g, GM-CSF, TNF-a, MIP-1a, MIP-1β, MIP-2, MCP-1, MCP-3, MIG, ENA-78, were evaluated (Luminex technology, Merck Millipore). The IFN-g effect was evaluated by STAT1 phosphorylation.

Results:
Significant downregulation (p<0.01) of about 30 pro- and anti-inflammatory cytokines, including IL-6, IP-10, TNF-a, MIP-1a, MIP-1β, IL-10, was documented. IFN-g effect on macrophages and dendritic cells was inhibited at the level of phosphorylated STAT1. IFN-g-induced expression of CXCL10 and CXCL9 in macrophages was reduced. Patients treated in vivo with higher dosages of apoptotic cells had lower cytokine/chemokine levels compared to those treated with lower levels, and in inverse correlation to aGVHD staging. In vitro binding of apoptotic cells to LPS was documented.

Conclusion:
The cytokine storm is significantly modified towards homeostasis following apoptotic cell treatment. The mechanism is multifactorial and was shown to include TAM receptor triggering, NFκb inhibition, and LPS binding. These results together with previous studies showing significantly higher murine survival in sepsis models of LPS and cecal ligation puncture suggest that apoptotic cells may be used to treat patients with sepsis. A multicenter clinical trial in septic patients is planned in 2018.