Abstract

PURPOSE: The authors recently reported that Foxp3(+)CD4(+) CD25(+(Bright)) "natural" regulatory T cells (nT(reg) cells) are abundant in rabbit conjunctiva and suppress herpes simplex virus (HSV)-1-specific CD4(+) and CD8(+) effector T cells (T(eff) cells). However, little is known about the overall regulatory mechanisms of these nT(reg) cells. The authors investigate the regulation of conjunctiva-resident nT(reg) cells through Toll-like receptors (TLRs) and their effect on ocular mucosal T(eff) cell immunity.

METHODS: CD4(+)CD25(+) nT(reg) cells were purified from naive rabbit conjunctivas, and their TLR expression profile was determined. The effects of TLR engagement on nT(reg) cell-mediated suppression of CD4(+) T(eff) cells were determined in vitro and in vivo.

RESULTS: The authors found that conjunctiva-resident nT(reg) cells express high levels of TLR2 and TLR9; exposure to the TLR2 ligand lipoteichoic acid (LTA) led to the increased activation and proliferation of nT(reg) cells, and the addition of autologous APCs further increased nT(reg) cell expansion; in contrast, the TLR9 ligand CpG(2007) inhibited the proliferation of nT(reg) cells, and the addition of autologous APCs had no effect on such inhibition; nT(reg) cells treated with LTA, but not with CpG(2007), expressed IFN-γ and IL-10 mRNA, but not TGF-β; consistent with in vitro data, rabbits immunized by topical ocular drops of HSV-gD peptides + TLR2 ligand (LTA) displayed enhanced CD4(+)CD25(-) T(eff) cell immune responses when compared with HSV-gD peptides + TLR9 ligand (CpG(2007)).

CONCLUSIONS: Although conjunctiva-resident CD4(+)CD25(+) nT(reg) cells express high level of TLR2 and TLR9, their suppressive function is more significantly reversed after the administration of TLR2 ligand (LTA; P < 0.005) than of TLR9 ligand (CpG(200); P > 0.005). These findings will likely help optimize the topical ocular administration of immunotherapies.