Protective role of B7-H3/TLT-2 pathway in acceptance of corneal allografts

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Background:

B7-H3 belongs to the B7 superfamily, a group of molecules that co-stimulate or down-modulate T-cell responses. Recently, triggering receptor expressed on myeloid cells-like transcript-2 (TLT-2) has been identified as a B7-H3 receptor. We have previously reported that B7-H3 is constitutively expressed in ocular tissue and B7-H3/TLT-2 pathway is necessary for corneal allograft survival. The purpose of the present study is to further investigate the mechanisms B7-H3-associated immune suppression.



- TLT-2 is not expressed in normal ocular tissue, but its expression is up-regulated both in corneal tissue and on macrophages and CD4+ T cells, in the allografts. (Taniguchi H, et al, ARVO, 2011)
- TLT-2 expressed in ocular tissue and macrophages within the cornea may contribute to allograft acceptance.

Methods and Results 1: B7-H3-mediated suppression is ACAID-dependent

Allo-antigen-specific ACAID model was used. B6 spleen cells were used as allo antigens and injected into the right anterior chamber (AC) of normal BALB/c eyes. After avertage as an engene and were injected into subcutaneously (SC) to sensitize the mice. After another week, B6 spleen cells were injected pinnae for determination of delayed hypersensitivity (DH) response after 48 h. Treatments with of anti-B7-H3 mAb (MJ18 or MIH35), anti-TLT-2 mAb, or control IgG were applied for 3 weeks after AC injection.



•DH response was induced in sensitized mice without prior AC injection (positive controls) compared with unsensitized naive mice (negative controls) Prior AC injection significantly suppressed the DH response in control IgG-treated mice, indicating induction of ACAID.

ACAID was abolished in the recipients treated with either anti-B7-H3 or anti-TLT-2 mAb.



Methods and Results 2: In vitro assay of corneal endothelial cell destruction by allo-reactive T cells

B6 cornea pre-treated with anti-B7-H3 mAb (MJ18 or MIH35) or control IgG were incubated with CD4+ T cells for 6 h. CD4+ T cells were purified from the spleen of BALB/c that were presensitized by SC immunization with B6 splenocytes or with third-party (C3H/He) splenocytes, or from the spleen of naive BALB/c, B6 or C3H/He mice. Dead CECs stained with PI were counted and compared.



B7-H3 expressed on CECs plays a role in protecting CECs from destruction by activated CD4+T cells.

• The number of dead CECs was significantly larger in anti-B7-H3 mAb-treated corneas than in control IgG-treated corneas after incubation with allo-reactive CD4+T cells.

 The number of dead CECs was also significantly larger in anti-B7-H3 mAb-treated corneas than in control corneas after incubation with CD4+T cells activated against third-party allo-antigens

Conclusions:

- B7-H3/TLT-2 pathway is involved in the induction of ACAID.
- B7-H3 expressed on CECs plays a role in protecting CECs from destruction by activated CD4+ T cells.
- Thus, B7-H3/TLT-2 pathway maintains acceptance of corneal allografts by inducing ACAID as a systemic effect and suppressing allo-reactive CD4+ T cells within the eye as a local effect.