Advances in Animal Models of Immune Dysregulation in IBD

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Disclosure

Nothing to report
**‘Dysregulated’**

**Mouse models of IBD**

<table>
<thead>
<tr>
<th>CD45RB transfer model</th>
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<tbody>
<tr>
<td>IL-2, IL-2Rα deficient</td>
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<td>BM -&gt; Tg26 transfer model</td>
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<tr>
<td>IL-10 deficient</td>
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<tr>
<td>CRF 2-4 deficient (IL-10Rβ)</td>
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<tr>
<td>Mφ -PMN Stat 3 deficient</td>
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<td>TGFβ deficient</td>
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<td>TGFβRII deficient</td>
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<td>SMAD3 deficient</td>
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**Multiple cells contribute to intestinal immune regulation**

<table>
<thead>
<tr>
<th>CD4+</th>
<th>Mouse</th>
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<tbody>
<tr>
<td>CD8+</td>
<td>&amp;</td>
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<tr>
<td>NK-T</td>
<td></td>
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<tr>
<td>Tγδ</td>
<td>Human</td>
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<td>CD4-8-</td>
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**Tregs - Common Features**

- Functional activity is antigen-dependent
- Constitutive expression of CTLA4
- Poorly proliferative in vitro
- Suppress proliferative responses of naïve and effector CD4 T cells
- Can act through “bystander” suppression (unlinked antigenic specificities)

**CD4+ Treg lineages**

*Tp* → *Foxp3+CD4+ Thymocyte*

*Foxp3+nTreg* → *Contact* → *IL-10*

*IL-10* → *TGF-β1*

*IL-10* → *Th17*

*“Natural”*

*“Induced”*

*Tp* → *Foxp3+iTreg*

*Foxp3+iTreg* → *Contact* → *IL-10*

*IL-10* → *TGF-β1*

*IL-10* → *TGF-β1*

*“Induced”*
How are Tregs generated in the intestine? TGFβ and Retinoic acid induce Tregs in the gut

RA acts as a cofactor for Foxp3 induction

[Coombes & Powrie Nat Rev Immunol 8:435, 2008]

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Conditioning of LP DCs by epithelial cells

- Retinoic acid
- Polysaccharide A from B. fragilis
- Bacterial DNA via TLR9
- TGFβ
- Apoptotic cells
- Antigen

Induction of Tregs in the Gut

- Stimuli Promoting Gut Lamina Propria Tregs
  - Retinoic acid
  - Polysaccharide A from B. fragilis
  - Bacterial DNA via TLR9
  - TGFβ
  - Apoptotic cells
  - Antigen

[Coombes & Powrie Nat Rev Immunol 8: 435, 2008]

[Barnes, Immunity 31, 2009]
T-regulatory-1 cells

- Human T cells activated in presence of high IL-10 become anergic. Clones of such cells produce high levels of IL-10, some IFNγ, little IL-2 or IL-4 and are suppressive in vitro.
- Mouse T cells activated in the same way have a similar cytokine pattern and suppressive activity in vitro.

Reporter-positive T cells from 10BiT Mice are IL-10-Competent Ex Vivo

Thy1.1 Reporter Identifies Spontaneously Arising IL-10+ T cells In Vivo: Overlapping distributions of Foxp3+ and IL-10+ T cells

Percentages of CD4+Foxp3+ that express Thy1.1

IL-6 plays a crucial role in Foxp3+ vs Th17 balance
Plasticity of CD4 subsets due to epigenetic regulation

IL-27 negatively regulates Th17 cells and induces Tr1-like (IL-10+) CD4 T cells

- IL-27-/- mice have increased LP Th17 and susceptibility to DSS colitis (Troy, 2009)
- IL-27 blocks RORc expression to inhibit lineage commitment of Th17 cells (Diveu, 2009)
- IL-27 induces c-Maf, IL-21, and ICOS that coordinately act together to promote differentiation of IL-10-producing Tr1 cells (Pot, 2009)
- In humans, IL-27 promotes Tr1 and inhibits Th17 T cells (Murugaiyan, 2009)
Mechanisms of Treg Function

![Diagram showing mechanisms of Treg function](image)

Depletion of CD25 Treg cells decreases intestinal IgA anti-CBir1

![Graph showing depletion of CD25 Treg cells](image)

[Cong, PNAS, 2009]
Can Tregs treat colitis?

<table>
<thead>
<tr>
<th>Model</th>
<th>Treg</th>
<th>Reference</th>
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<tbody>
<tr>
<td>CD45RB&lt;sup&gt;hi&lt;/sup&gt; transfer</td>
<td>CD4+ CD25+</td>
<td>Mottet, 2003</td>
</tr>
<tr>
<td>CD45RB&lt;sup&gt;hi&lt;/sup&gt; transfer</td>
<td>Tr1 cells</td>
<td>Foussat, 2003</td>
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IL-10 required for Treg effect
T Cell Regulatory Function: Observations in Humans

1. IPEX syndrome/foxp3 deficiency
2. CTLA-4 blocking antibody and induced colitis
4. SNP flanking the IL10 gene is associated with Ulcerative colitis susceptibility
5. Association of IL-10R mutations (blocked STAT3 signaling) with early-onset IBD (NEJM, 2009 361: 2033)

Gut Mucosal Tregs in Human IBD

To date, no defect in numbers or in vitro suppressive function of tissue-derived Treg cells has been reported in IBD patients.

• Active IBD (CD and UC) not associated with functional defect but with a decrease in PERIPHERAL CD4CD25^{high} and foxp3^{+} cells. Highest Foxp3 expression seen in diverticulitis inflammatory control (Maul et al. Gastroenterology 2005)

• CD4CD25^{high} Treg frequency higher in inflamed UC mucosa compared to non-inflamed or control. No defect in suppressor activity (Holmen et al. Inflamm. Bowel Dis. 2006 12:447)

• Foxp3^{+}CD4 Tregs expanded in LP and MLN (decreased in PB); similar suppressor activity compared to healthy control (Saruta et al. Clin. Immunol. 2007)

• No data on other candidate Treg cells
Protection vs pathology

[Adapted from: Belkaid, Y. Nat Immunol 6:353, 2005]

Conclusions

• Gut Tregs control the innate and adaptive response to the microbiota
• Multiple Treg subsets exist and all are dependent on TGFβ
• IL-6 and IL-27 regulate the balance between Th17 and Tregs
• Tregs are increased in human IBD and are functional ex vivo
• Tregs determine the balance between protection and pathology