Regulation of CNS Autoimmunity

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Myelin is the target of the autoimmune response in Multiple Sclerosis (MS) and Experimental Autoimmune Encephalomyelitis (EAE)

Frohman et al., N Engl J Med 354:94202
Peripheral Lymphoid Tissue

CD8+ Th1 Th17

APC

TGF-β (IL-6/IL-21/IL-1β) + IL-23

IL-12

INFLAMMATORY CELLS

Blood Brain Barrier

Central Nervous System

Axonal and myelin damage

Initiation of inflammatory cascade

TGF-β

Microglia / Dendritic cells

Treg

FoxP3+

Th3

IL-6

TNF-α

Tr1

Axonal and myelin damage

Initiation of inflammatory cascade

TGF-β

Microglia / Dendritic cells

TGF-β + IL-27

TGF-β

IL-10

Peripheral Lymphoid Tissue

Blood Brain Barrier

Central Nervous System

B cell

T cell

Th1

CD8+

Teff

Microglia / Dendritic cells

IL-10

IL-35

INFLAMMATORY CELLS

REGULATORY CELLS
Alterations in the balance between Tregs and Teffs play a central role in multiple sclerosis

IFN-β induces IL-27 expression and promotes the differentiation of Tr1 cells

Mitsdoerffer and Kuchroo 2009
IL-27 signaling and biological effects

Type 1 regulatory T (Tr1) cells

FoxP3- suppressive CD4+ T cells that produce IL-10.

IL-27 promotes the differentiation of Tr1 cells.

IL-21 is an autocrine growth factor for Tr1 cells.

c-Maf controls IL-10 production in Tr1 cells.
**IL-27 promotes Tr1 cell differentiation via AHR signaling**

- **Environment**
  - Pollutants
  - Diet
  - Commensal flora
- **AHR ligands**
- **AHR**
  - Association
  - Interaction
- **AHR/c-Maf complex**
- **pSTAT3**
- **IL-10 promoter**
- **IL-21 promoter**
- **IL-10**
- **IL-21**
- **Tr1 cell differentiation**

**Dendritic cell**

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**IL-27 produced by DCs promotes Tr1 cell differentiation**

- **Dendritic cell**
- **IL-27**
- **Tr1 cell differentiation**

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The receptor for IL-27 is expressed by DCs

IL-27RA mRNA Relative expression

pDC cDC

What are the autocrine effects of IL-27 signaling in DCs?

Effects on DCs?

Tr1 cell differentiation

Dendritic cell

T cell

IL-27

IL-27 modulates the APC function of DCs

DCs + MOG + 2D2 T cells

[Graphs showing CPM (x10^3), IFN-γ, IL-17, IL-10, and TGF-β levels in response to IL-27 and ecLPS treatment for Teffs and Tregs]


Generation of mice lacking IL-27RA expression in DCs

[Diagram showing the generation process from CD11c-DTR BM to WT or IL-27RA-KO DCs and the effect on T cell responses and EAE]

IL-27RA in DCs limits EAE development

Increased Teff activation by DCs from DC_{IL-27RA-KO} mice

IL-27 acts on cDCs in vivo to limit the development of encephalitogenic T cells and EAE.
**Summary I**

IL-27 signaling in cDCs decreases the differentiation of Th1 and Th17 cells while it boosts the differentiation of Tr1 and FoxP3+ CD4+ T cells.

IL-27 signaling in cDCs *in vivo* limits the development of encephalitogenic T cells and EAE.

**Computational modeling of the transcriptional response of DCs to IL-27 identifies ENTPD1 (CD39)**

(RNA expression + ChIPseq)

CD39: Degradation of extracellular ATP

IL-27 signaling in DCs regulates *Entpd1* (CD39) expression

*In vitro*

![Graph showing the relative expression of Entpd1 mRNA over time.](image)

*In vivo*

![Graph showing the relative expression of Entpd1 mRNA and CD39 in DCs.](image)

**CD39 (ENTPD1) mediates the tolerogenic effects of IL-27 conditioned DCs**

![Bar graph showing the CPM values for different conditions.](image)

DCs + MOG + Naïve 2D2 T cells

CD39 (ENTPD1) degrades extracellular ATP

Extracellular ATP produced by DCs activates the NLRP3 inflammasome in an autocrine manner

Does IL-27-induced CD39 regulate inflammasome activation in DCs?
IL-27-induced CD39 on DCs controls extracellular ATP


IL-27-induced CD39 controls NLRP3 activation

CD39 expression in DCs limits the encephalitogenic Th1 and Th17 T-cell response and the development of EAE.
Extracellular ATP produced by activated DCs acts on an autocrine manner to activate the NLRP3 inflammasome

IL-27 up-regulates ENTPD1 expression in DCs
DC-expressed ENTPD1 degrades extracellular ATP and reduces NLRP3 inflammasome activation

Can we exploit this pathway therapeutically?
Vaccination with IL-27-conditioned DCs suppresses EAE

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Vaccination with IL-27-conditioned DCs limits the encephalitogenic T-cell response

PLP (139-151)

PLP (178-191)

Vaccination with IL-27-conditioned DCs limits epitope spreading

Antigen microarrays

Summary II

- IL-27 signaling in DCs limits the effector T-cell response and CNS autoimmunity
- IL-27 up-regulates CD39 expression in DCs via STAT3
- The induction of CD39 expression in DCs by IL-27 decreases extracellular levels of ATP, reducing NLRP3 inflammasome activation and Teff differentiation
- Vaccination with IL-27-conditioned DCs halts established chronic EAE.
Transcriptional effects of IL-27 on DCs

AHR activation in DCs promotes the differentiation of regulatory T cells

Swiss Med Wkly. (2012); 142:w13592


Proc Natl Acad Sci USA 107, 19961-19966 (2010).
AHR activation induces tolerogenic DCs

DCs + MOG + Naive 2D2 T cells

Control
ITE

Targeting of AHR in DCs with Nanoparticles to co-deliver autoantigens and AHR agonists

PNAS 107, 20768-20773 (2010).
PNAS 107, 19961-19966 (2010).
Targeting of AHR in DCs with Nanoparticles to Co-deliver a myelin antigen and activate AHR signaling

Tolerogenic dendritic cells

Control of autoimmunity

Targeting of AHR in DCs with Nanoparticles to Co-deliver a myelin antigen and activate AHR signaling

Tolerogenic dendritic cells

Control of autoimmunity

PNAS 109, 11270-5 (2012)
Construction of NP containing MOG_{35-55} and ITE

PNAS 109, 11270-5 (2012)

NP_{ITE+MOG} suppress CNS inflammation in an experimental model of Multiple Sclerosis (EAE)

PNAS 109, 11270-5 (2012)
EAE inhibition by NP<sub>ITE+MOG</sub> is mediated by AHR in DCs

Summary III

1- AHR controls the APC function of DCs.

2- NP<sub>ITE+ANTIGEN</sub> induces tolerogenic DCs that favor Treg generation.

3- NP<sub>ITE+ANTIGEN</sub> suppress the development of EAE and T1D.
## Acknowledgments

<table>
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<tr>
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<tr>
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http://brighamandwomens.org/research/labs/quintana/default.aspx