Heterodimeric IL-15 Induces Effector Cell Activation and Trafficking to the Germinal Centers of SIV infected Macaques

Barbara K. Felber & George N. Pavlakis
National Cancer Institute - USA
Disclosures

Barbara Felber & George Pavlakis are co-inventors in hetIL-15 patents held by NIH–USA and licensed to NOVARTIS
IL-15 Is A Heterodimeric Cytokine (hetIL-15)

- Co-expression of IL-15 and IL-15Rα produces a stable heterodimer
  - Natural way of cytokine production in vivo
  - Essential for large scale production in bioreactors
- IL-15 single chain is unstable, not found in vivo

Bergamaschi, Blood 2012; Chertova, J.Biol Chem 2013;
Thaysen-Andersen Glycoconjugate J. 2015
Functions of hetIL-15

**Lymphocyte Expansion and Differentiation**
NK, CD8, effector CD4, γδ T cells

**Enhanced Activation, Cytotoxicity**
Increases expression of granzymes, perforin
Enhances ADCC

**Migration**
Rapid extravasation of lymphocytes to tissues
- Increases CD8 Effectors in tumors

hetIL-15 is A Unique Immune System Agonist For Cancer and AIDS Immunotherapy
Proposed Clinical Uses of hetIL-15

• In cancer immunotherapy
  - phase I clinical trial (NCT02452268) in adults with metastatic cancer at the NIH Clinical Center (with Admune/Novartis)

• As vaccine enhancer

• For HIV eradication
  • To induce cytotoxic cells and promote delivery into areas of virus reservoirs and sanctuaries
2-week Administration of SC hetIL-15
Similar to the clinical schedule

Blood, LN, tissue collection BEFORE

Day 1  Day 3  Day 5  Day 8  Day 10  Day 12

Blood, LN, tissue collection AFTER

Day 15

- High doses of hetIL-15 able to increase total body lymphocytes can be delivered without toxicity
- Important for combination of hetIL-15 with other drugs
hetIL-15 Increases CD8 Effector Cells (CD95^+CD28^-) In Lymph Nodes of Macaques
hetIL-15 Triggers a Cytotoxic Commitment (increased Granzyme B) in CD8\(^+\), CD4\(^+\), and NK Cells in Tissues, Including Lymph Nodes

Example CD8 T cells
hetIL-15 Treatment Increases SIV-specific T Cells Within the Lymph Nodes

CD3^+ T lymphocytes in LN

- Untreated
- hetIL-15

CD8

CM9 Tetramer+

Granzyme B

Cytotoxic Potential

Ki67

EXPANSION

Untreated

hetIL-15
Multiplex Confocal Imaging and Histocytometry to Provide Spatial Information about the Localization of Cells Within the Lymph Nodes After hetIL-15 Treatment
hetIL-15 induces an Accumulation of CD3$^+$ and GrzB$^+$ cells in Lymph Nodes
hetIL-15 Increases CD3+ GrzB+ Lymphocytes in Germinal Centers
hetIL-15 Induces Accumulation of CD3+ and GrzB+ Cells in Lymph Nodes

**CD3+ Cells**

- **Whole LN**
  - Before: 1000 cells/mm²
  - +hetIL-15: 2000 cells/mm²
  - Statistically significant: $p=0.0317$

- **Follicles**
  - Before: 500 cells/mm²
  - +hetIL-15: 500 cells/mm²
  - Statistically significant: $p=0.0159$

- **Germinal Center**
  - Before: 250 cells/mm²
  - +hetIL-15: 250 cells/mm²
  - Statistically significant: $p=0.0995$

**CD3+GrzB+ Cells**

- **Whole LN**
  - Before: 100 cells/mm²
  - +hetIL-15: 100 cells/mm²
  - Statistically significant: $p=0.0095$

- **Follicles**
  - Before: 10 cells/mm²
  - +hetIL-15: 10 cells/mm²
  - Statistically significant: $p=0.009$

- **Germinal Center**
  - Before: 5 cells/mm²
  - +hetIL-15: 5 cells/mm²
  - Statistically significant: $p=0.009$

F CD3+, $p=0.0317$; F GrB+, $p=0.0159$
GC CD3+, $p=0.0095$; GC GrB+, $p=0.009$
On-Going Studies: cART+hetIL-15

- Measure virus reservoirs in LN/tissues
- Measure virus suppression after STI
- Combine hetIL-15 with other strategies
  - Therapeutic vaccination + hetIL-15
Efficient Long-term Control of Viremia in SIV_{mac25}-infected Macaques under cART treatment for > 6 months

Log RNA copies/ml plasma

7 months of cART
3-drug combination:
Tenofovir (TFV), emtricitabine (FTC), Integrase inhibitor Dolutegravir in a single combination (sc daily)

Treatment with hetIL-15 2-week cycles

Threshold 50 copies/ml

Weeks of infection

0 2 4 6 8 10 12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44 46
hetIL-15 Treatment of SIV-infected cART-Treated Macaques Does Not Increase Plasma Virus Load

![Graph showing RNA copies/ml plasma over time for saline and hetIL-15 treatments.](image)

Threshold 2 copies/ml
Hypothesis:
Broad Effective and Non-escaped Immunity during ART contributes to cure

- Induction of potent cytotoxic T cells by **DNA vaccine** targeting the ‘Achilles’ heel of the virus, i.e., **the highly conserved regions**

- Enhancing immunotherapy by **hetIL-15**
Therapeutic Vaccination of SIVmac Infected cART Treated Macaques with SIV p27CE pDNA Induces Robust T Cell Responses

- Induction of broad effective and non-escaped immunity by Conserved Element (CE) pDNA vaccine during cART treatment may contribute to cure (Oral presentation Thursday THAA02 14.30)

Macaques were
- infected with SIV<sub>mac251</sub> for ~3 months
- treated with cART (Gilead) for 9-11 months
- Vaccinated 3x with p27CE pDNA vaccine (N=4) or sham DNA (N=2) via IM route followed by electroporation
Heterodimeric IL-15 is A Promising Agent For Virus Control And For Eradication Approaches

• hetIL-15 increases cytotoxic CD8 effector cells in Lymph Nodes and Germinal Centers, a known HIV/SIV reservoir/sanctuary

• hetIL-15 increases also the virus-specific CD8 effectors

• hetIL-15 affects the organization of Germinal Centers

• Combination of Flow and Histo-cytometry revealed:
  – Increase of CD3+ and CD8+ cells in the follicles and GCs
  – Increase of the number of CD3+ GrzB+ cells in follicles and GCs

• High and effective doses of hetIL-15 can be safely administered with long-term ART without toxicity
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