des racines pour la vie • roots for life

HBV and T cell Exhaustion

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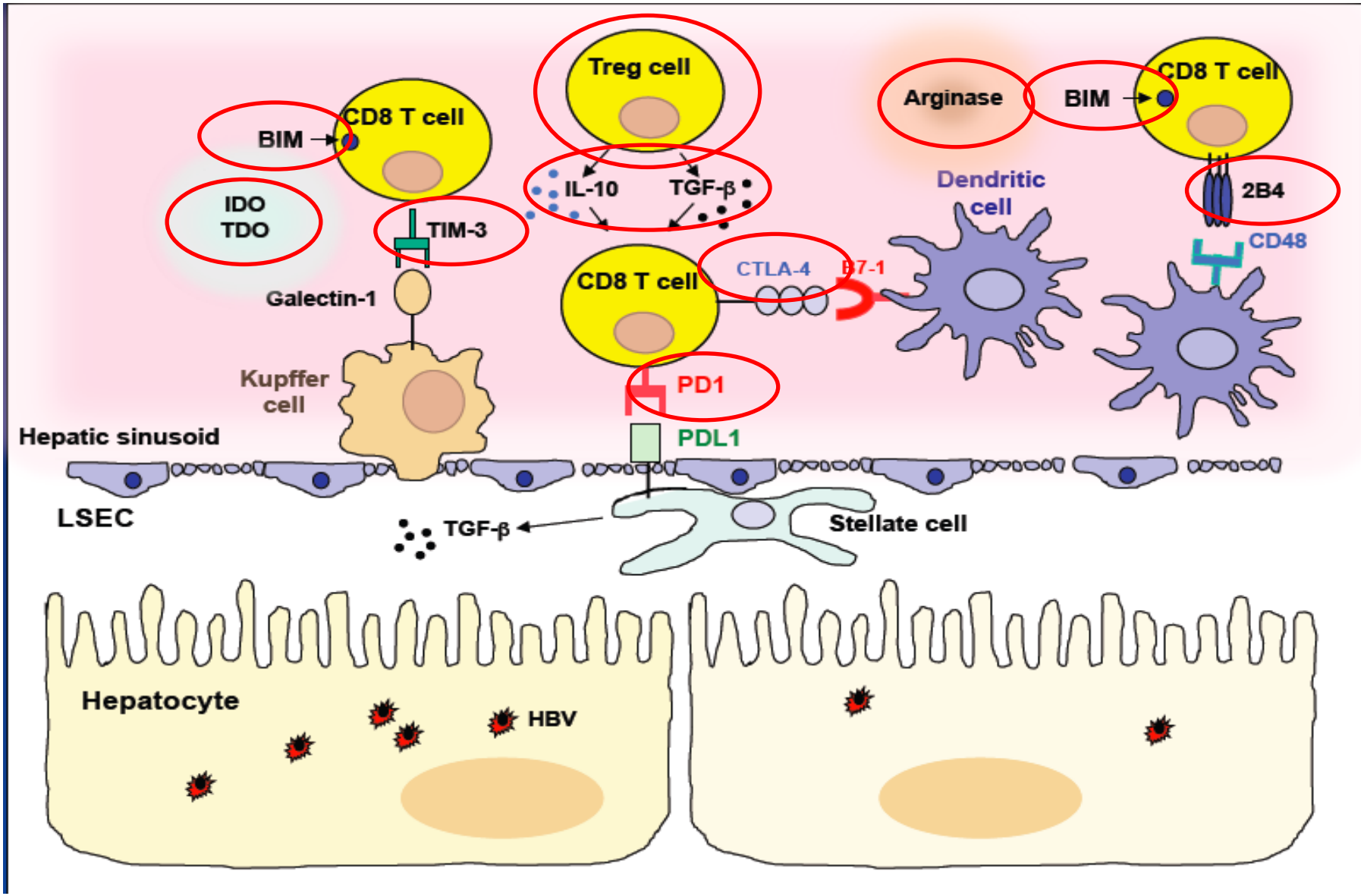
INTRAHEPATIC INHIBITORY MECHANISMS

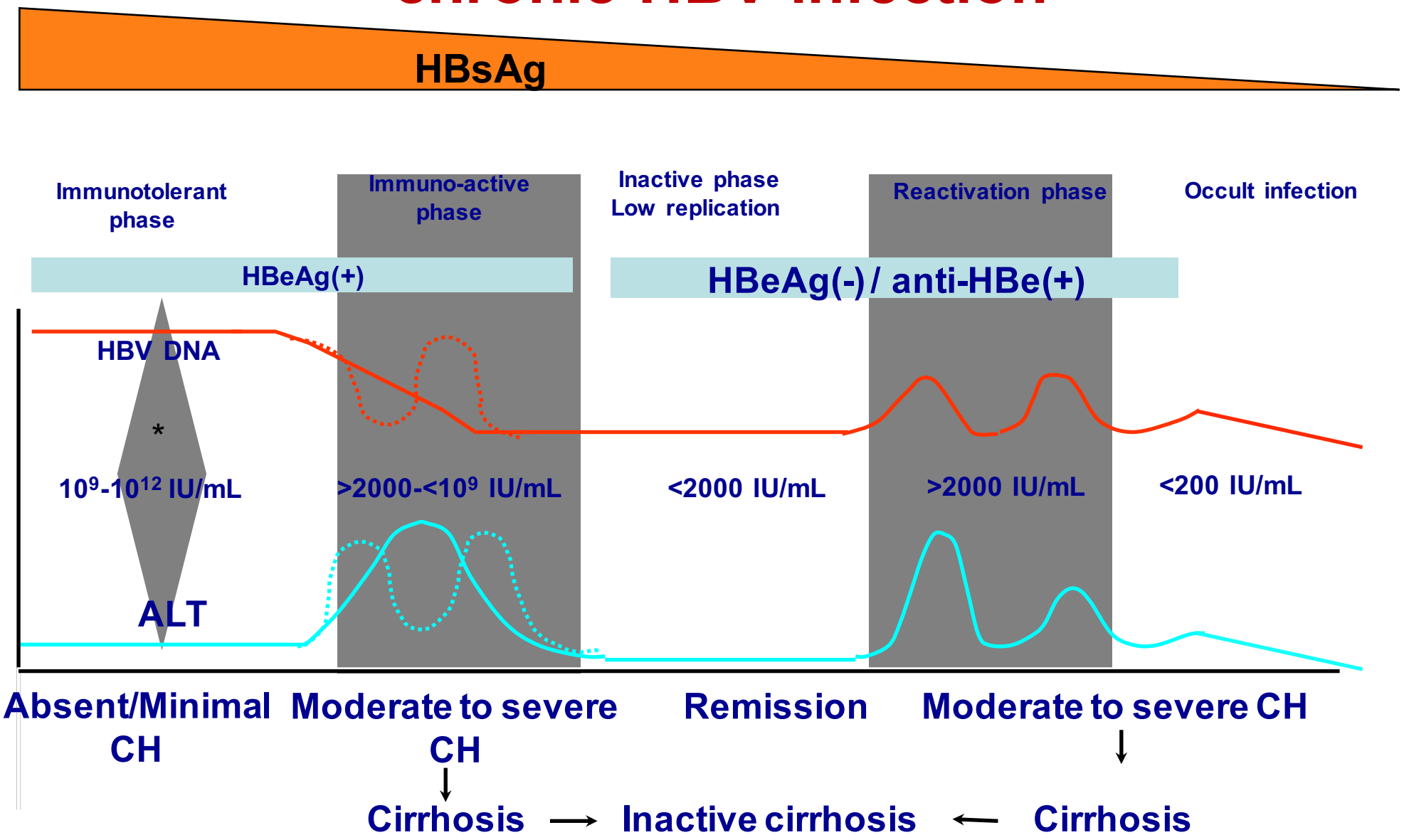


Modified from U. Protzer et al. Nature Reviews in Immunology 2012

List of topics

- HBV-specific T cells in chronic infection
- NK cell response and its regulatory role on HBV-specific T-cell response
- Potential strategies to reconstitute the anti-viral T cell function
- Molecular basis of CD8 cell dysfunction in chronic HBV infection

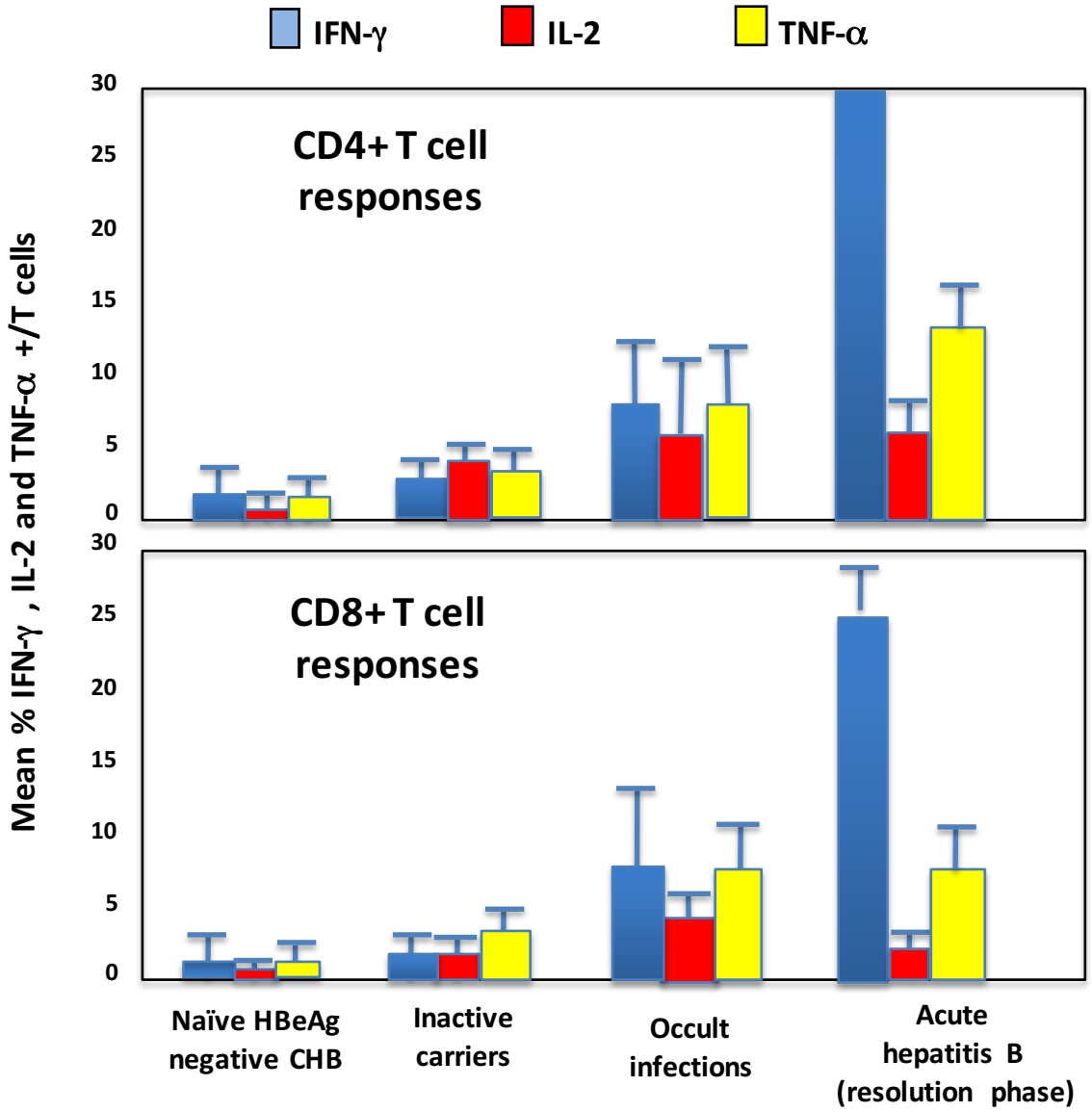
The five phases of the natural history of chronic HBV infection



Adapted from Fattovich G. Sem Liver Dis. 2003

HBV-specific T cells in chronic infection

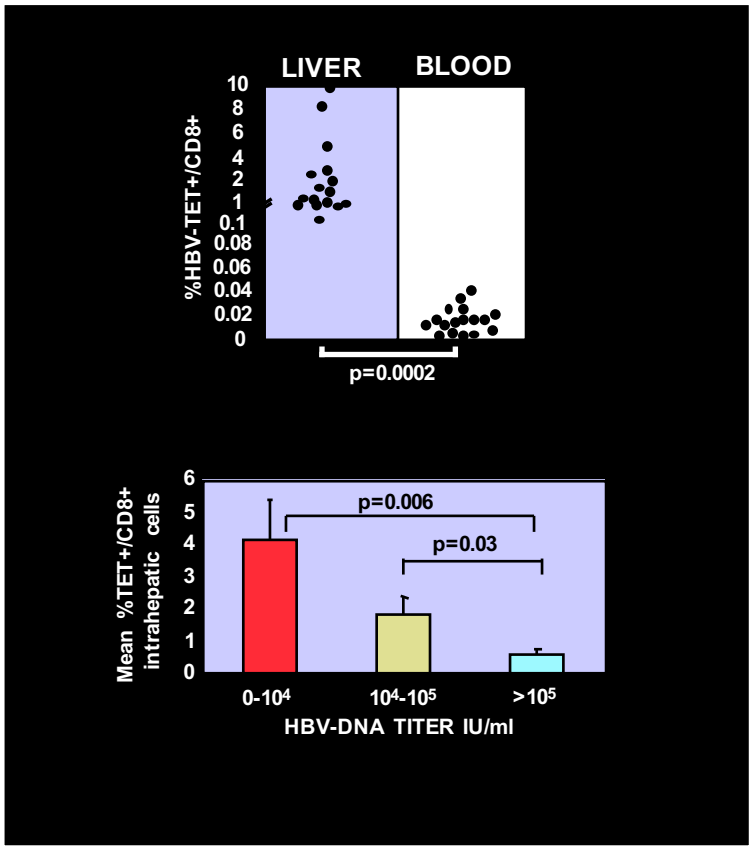
Different levels of functional T cell efficiency in different conditions of HBV control



Boni C, Laccabue D et al. Gastroenterology 2012;143:963–973

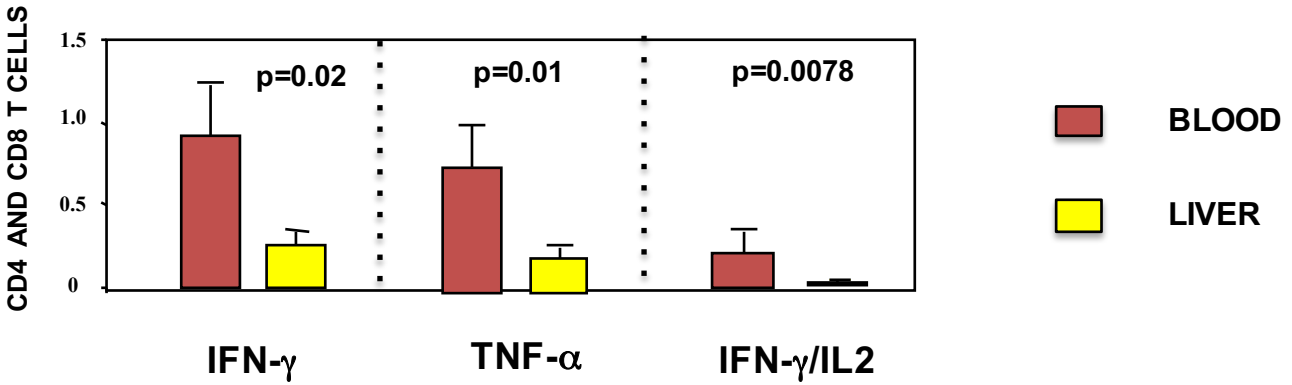
HBV-SPECIFIC CD8 CELLS ARE PREFERENTIALLY CONCENTRATED WITHIN THE LIVER IN PATIENTS WITH CHRONIC HBV INFECTION

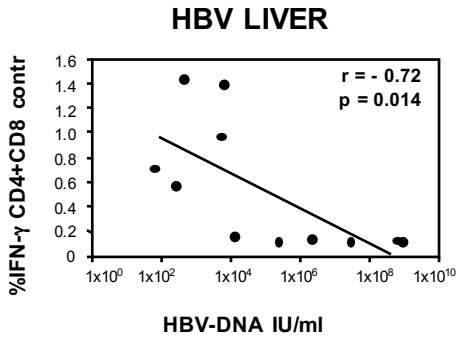
(Fisicaro P. et al. Gastroenterology 2010)



INTRAHEPATIC HBV-SPECIFIC T CELLS ARE MORE DEEPLY EXHAUSTED THAN THEIR PERIPHERAL BLOOD COUNTERPARTS IN CHRONIC HBV INFECTION

(Fisicaro P. et al. Gastroenterology 2012)

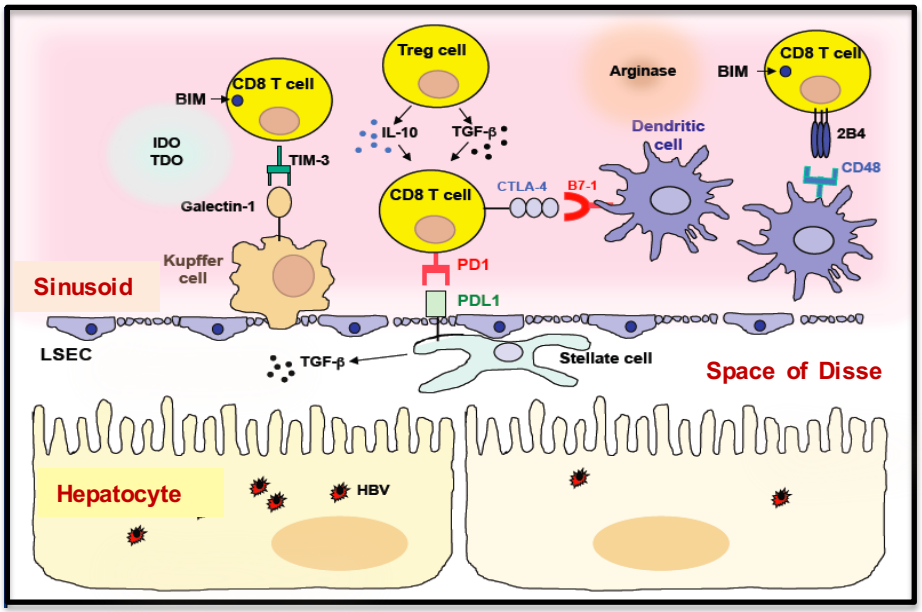




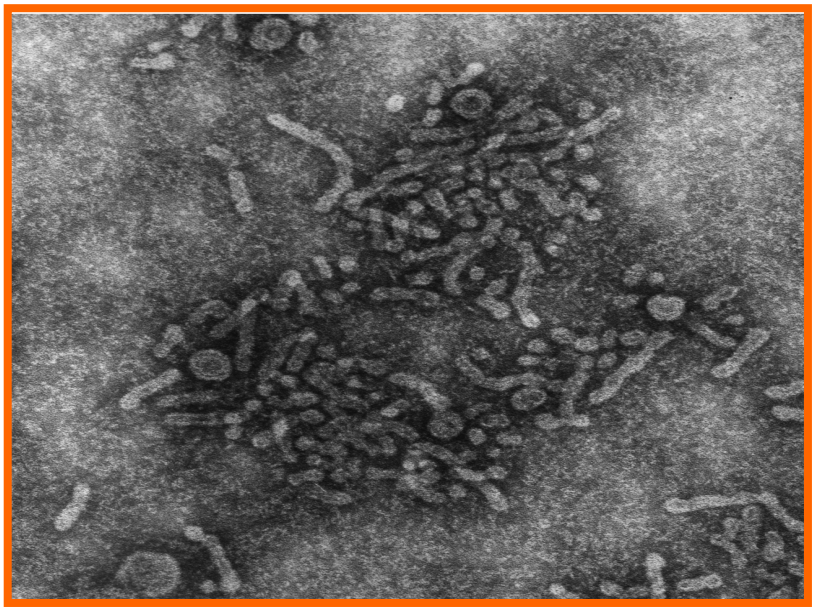
Is it possible to restore innate and adaptive immunity for HBV?

PUTATIVE MECHANISMS OF T CELL EXHAUSTION IN HBV INFECTION

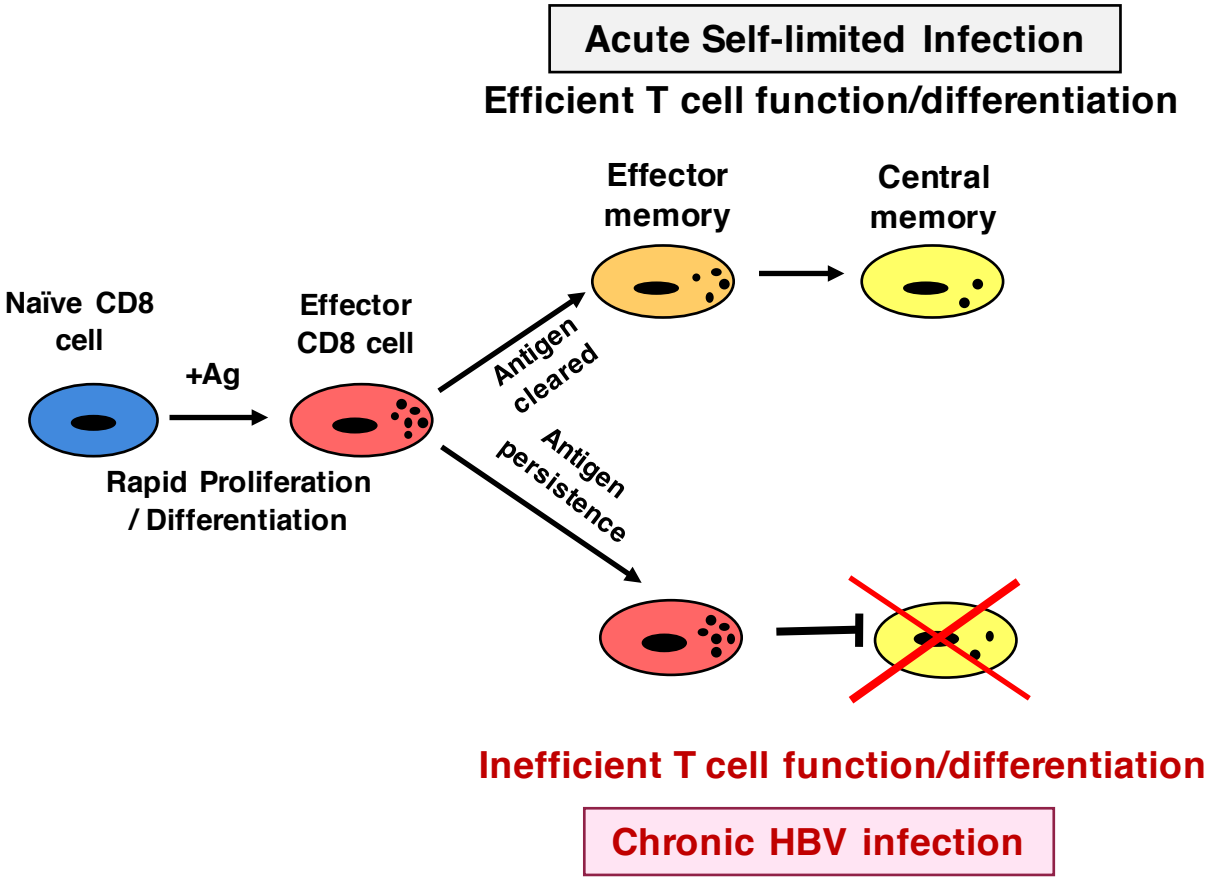
Tolerizing liver environment



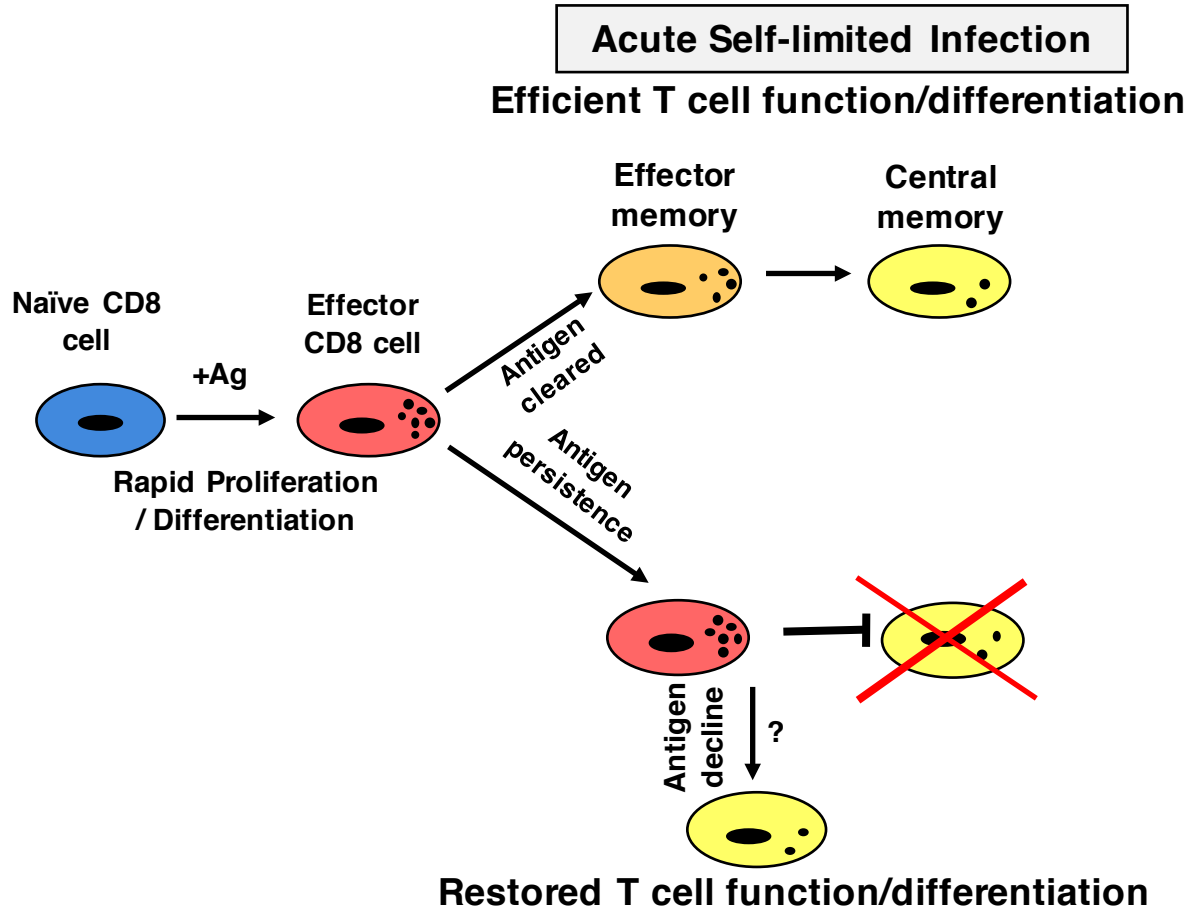
High viral load with massive production of secretory proteins (HBsAg, HBeAg)



T CELL FUNCTIONAL IMPAIRMENT IN CHRONIC HBV INFECTION

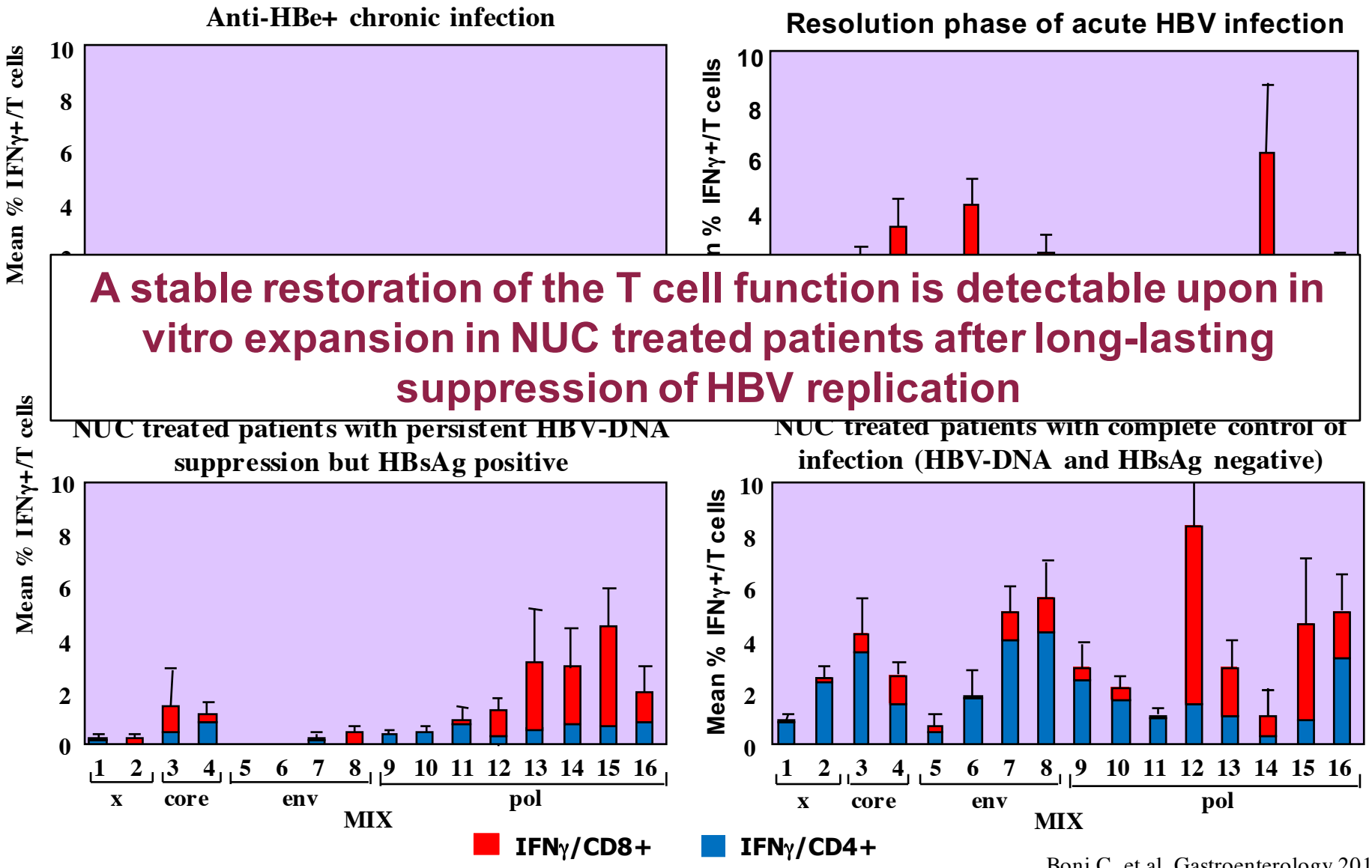


CAN THE HBV-SPECIFIC T CELL FUNCTION BE RESTORED BY ANTIGEN DECLINE IN CHRONIC HBV PATIENTS?



Effect of long-term NUC therapy on T cell responses

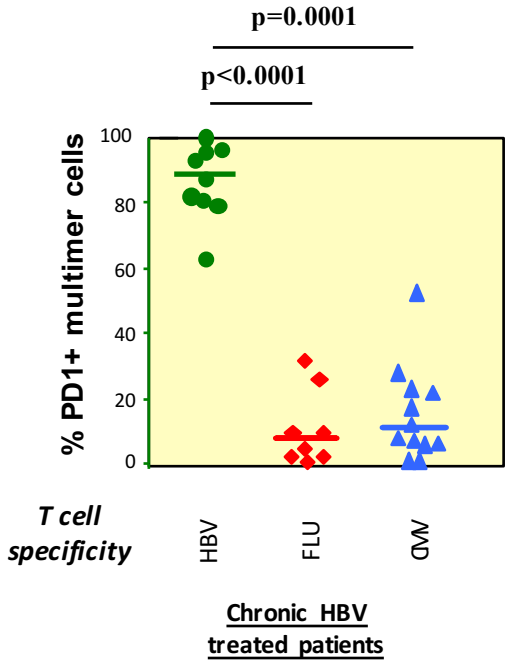
Stable restoration of the T cell function after long-lasting suppression of HBV replication induced by NUC therapy and detected following expansion in vitro

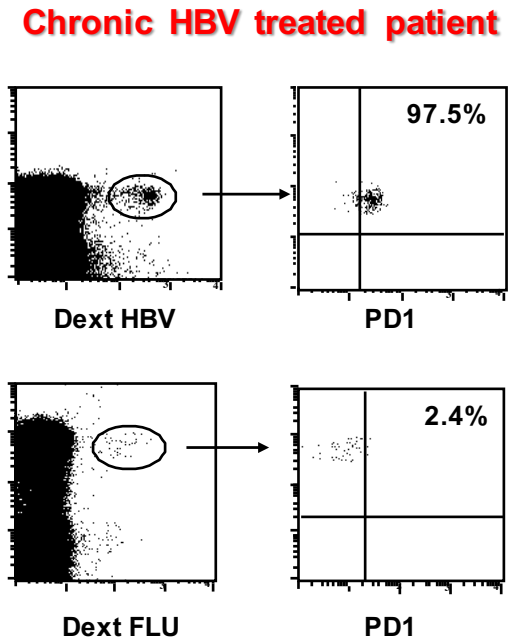


Boni C. et al. Gastroenterology 2012

PD-1 expression by HBV-specific CD8 cells in NUC treated patients

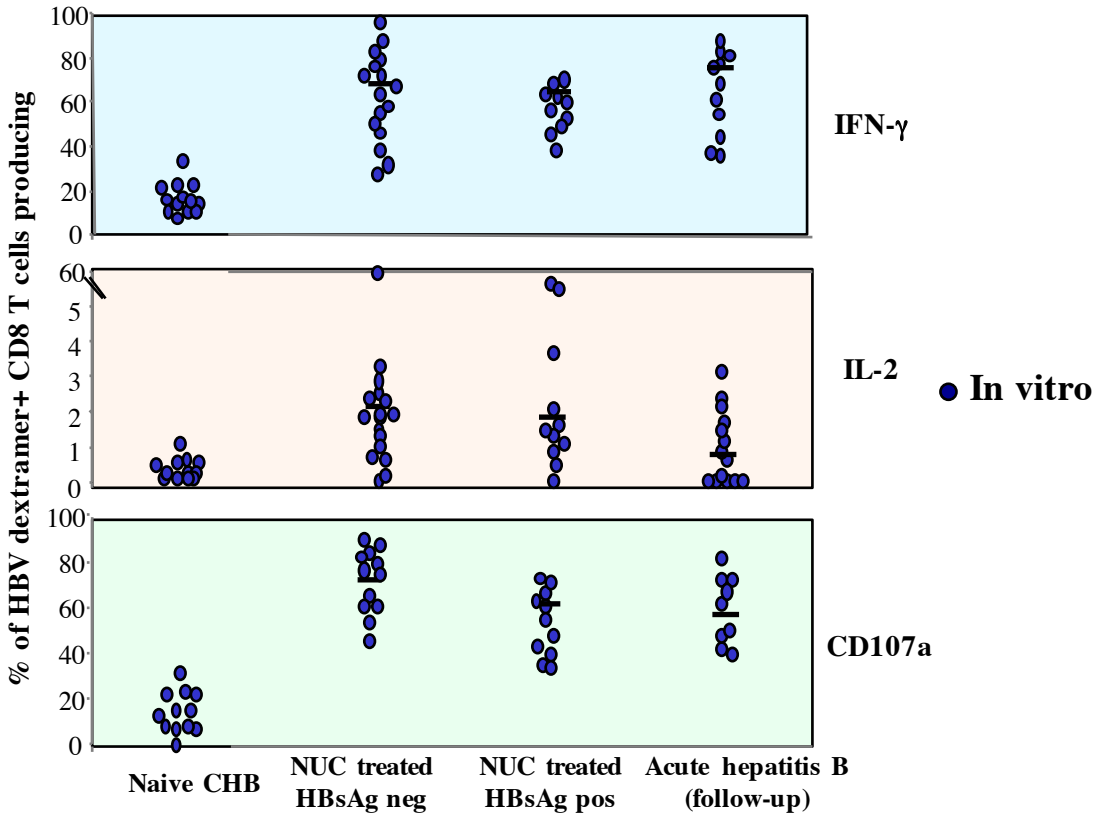
Boni C. et al. Gastroenterology 2012

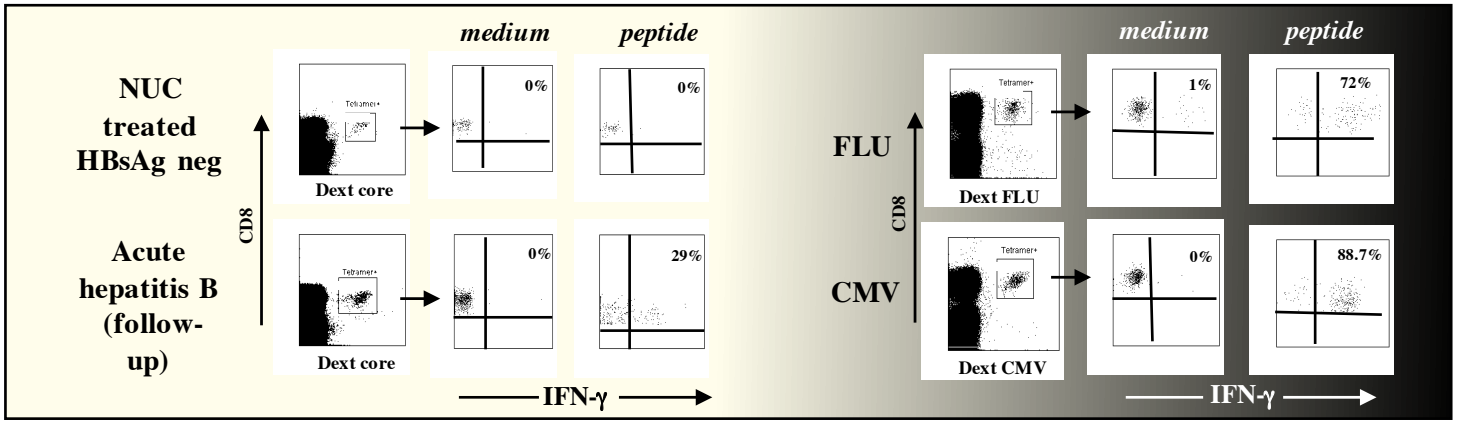




T cell restoration following NUC treatment is efficient in vitro

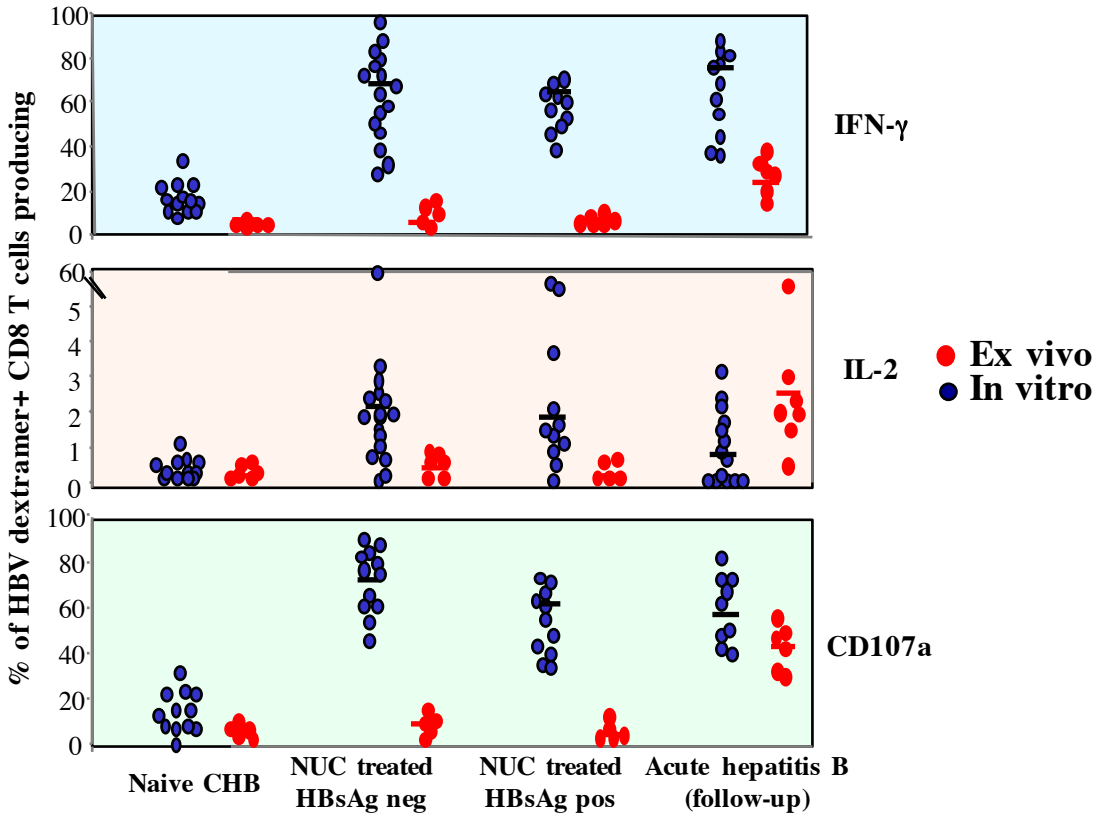
Boni C. et al. Gastroenterology 2012

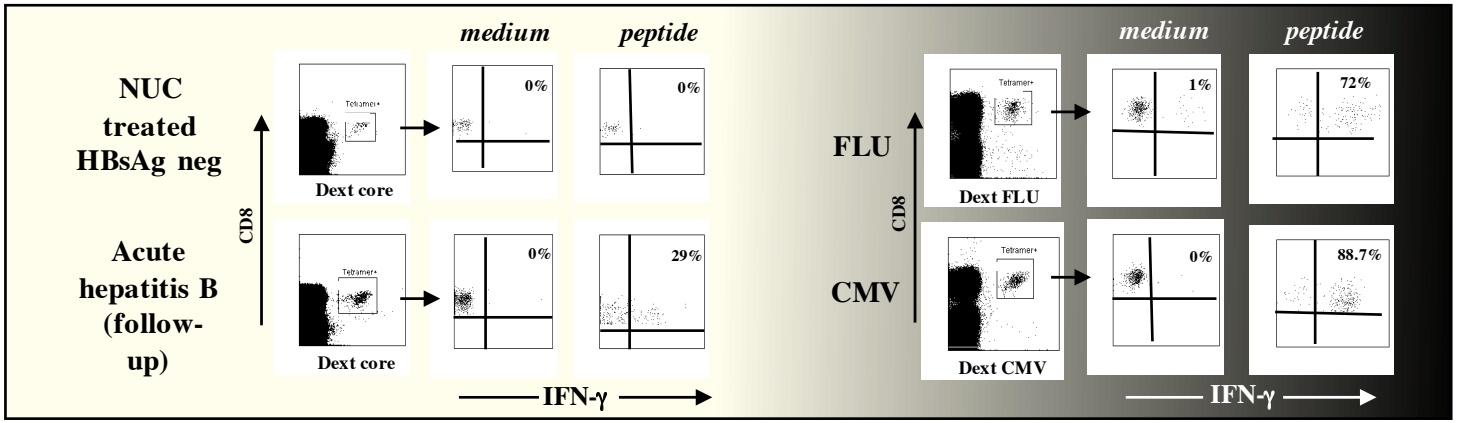




T cell restoration following NUC treatment is partial ex vivo

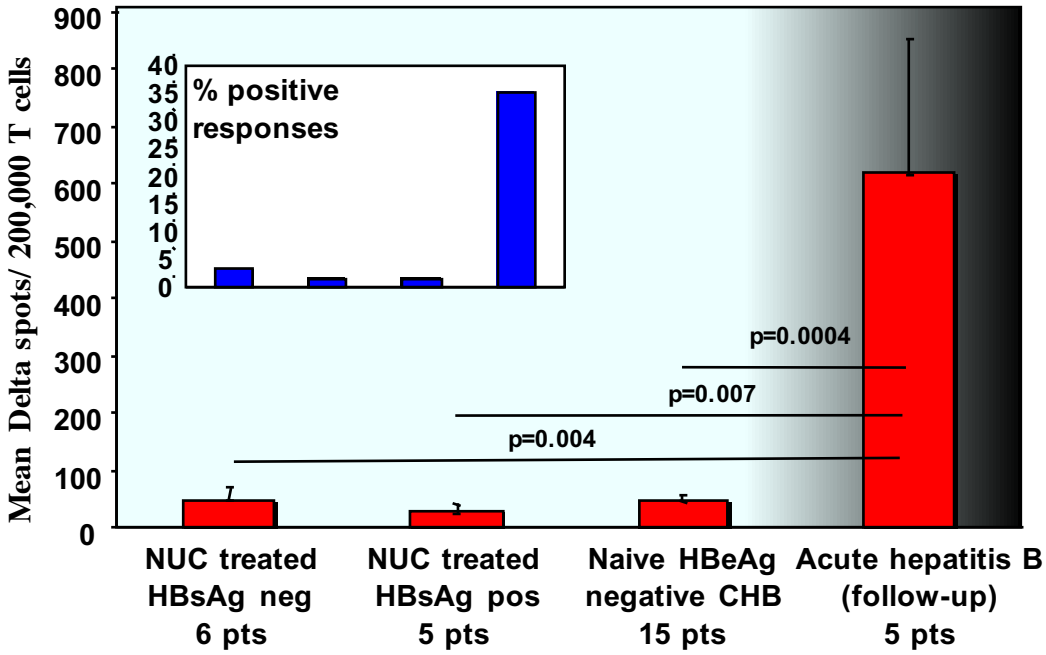
Boni C. et al. Gastroenterology 2012





Ex vivo frequencies of IFN-γ producing HBV-specific T cells after peptide stimulation

(ELISPOT analysis)



Boni C. et al. Gastroenterology 2012

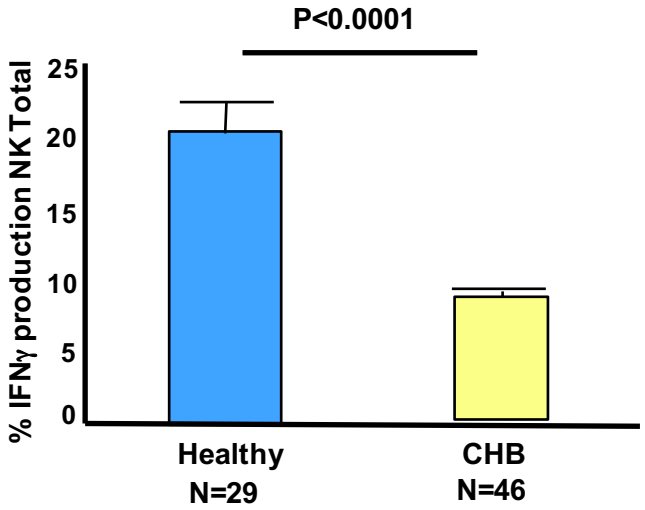
NK cell function in NUC treated patients

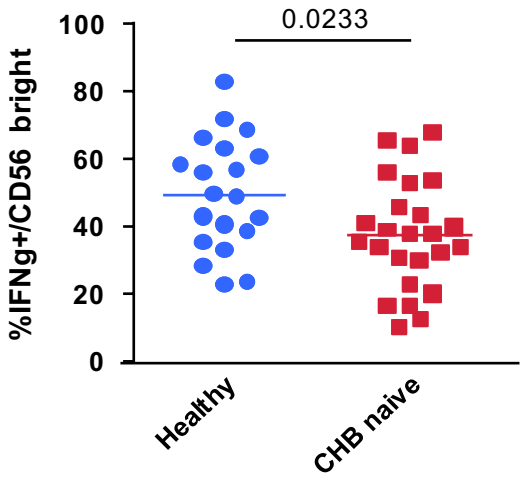
Is functional HBV-specific T cell restoration associated with a parallel improvement of NK cell function in NUC treated patients?

Functional NK cell dichotomy in chronic HBV infection

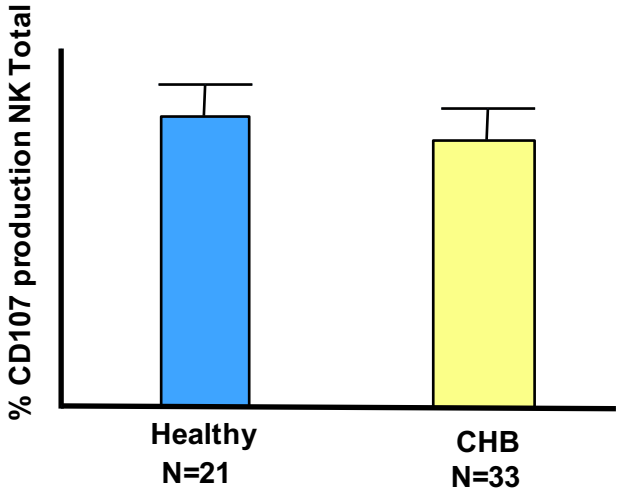
Impaired IFN-γ production with normal cytotoxicity

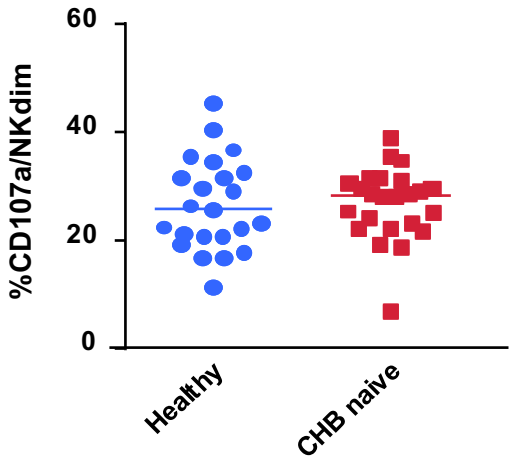
IFN-γ





CD107a

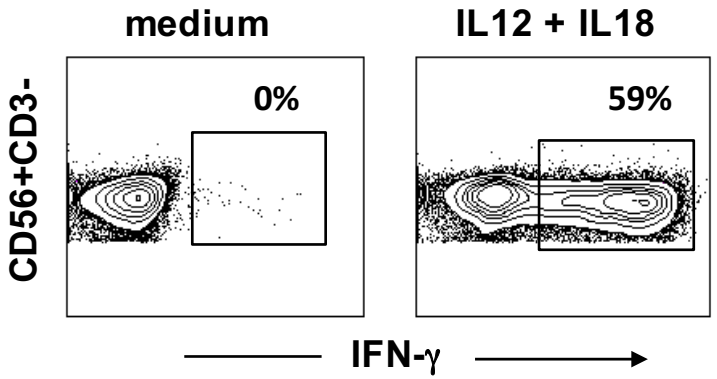


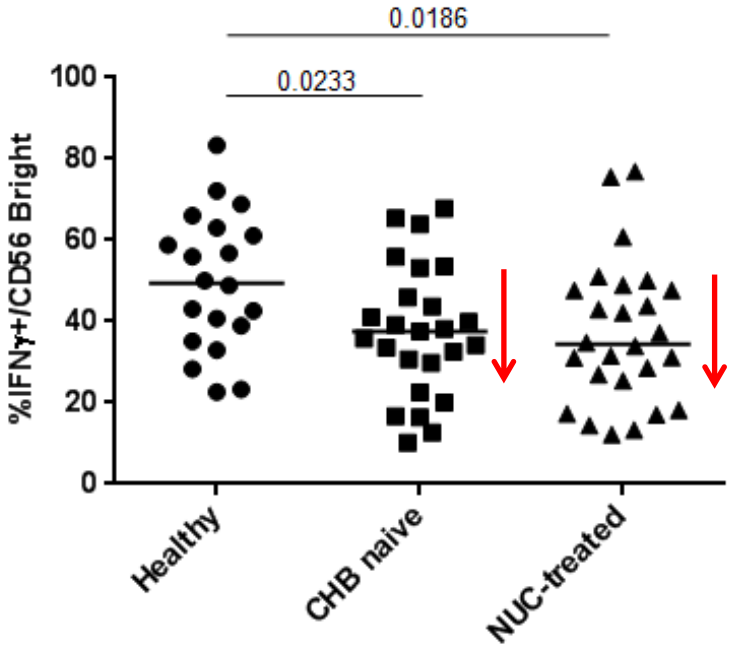


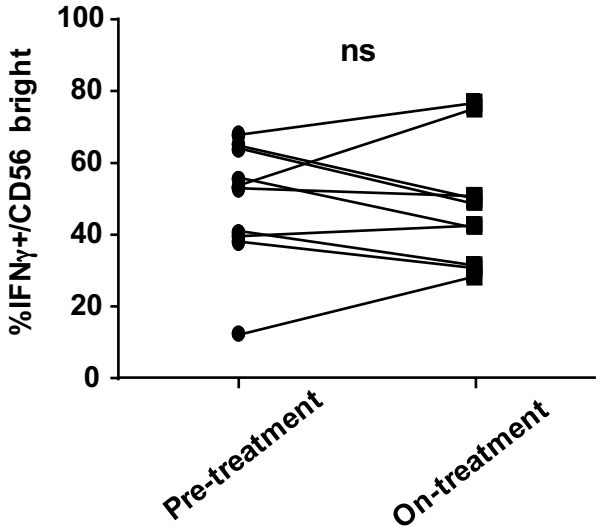
Peppa D. et al Plos Pathogens 2010

Boni C. et al. Hepatology 2015; 62:1697-709

IFN-γ production by NK cells is not significantly improved by NUC therapy

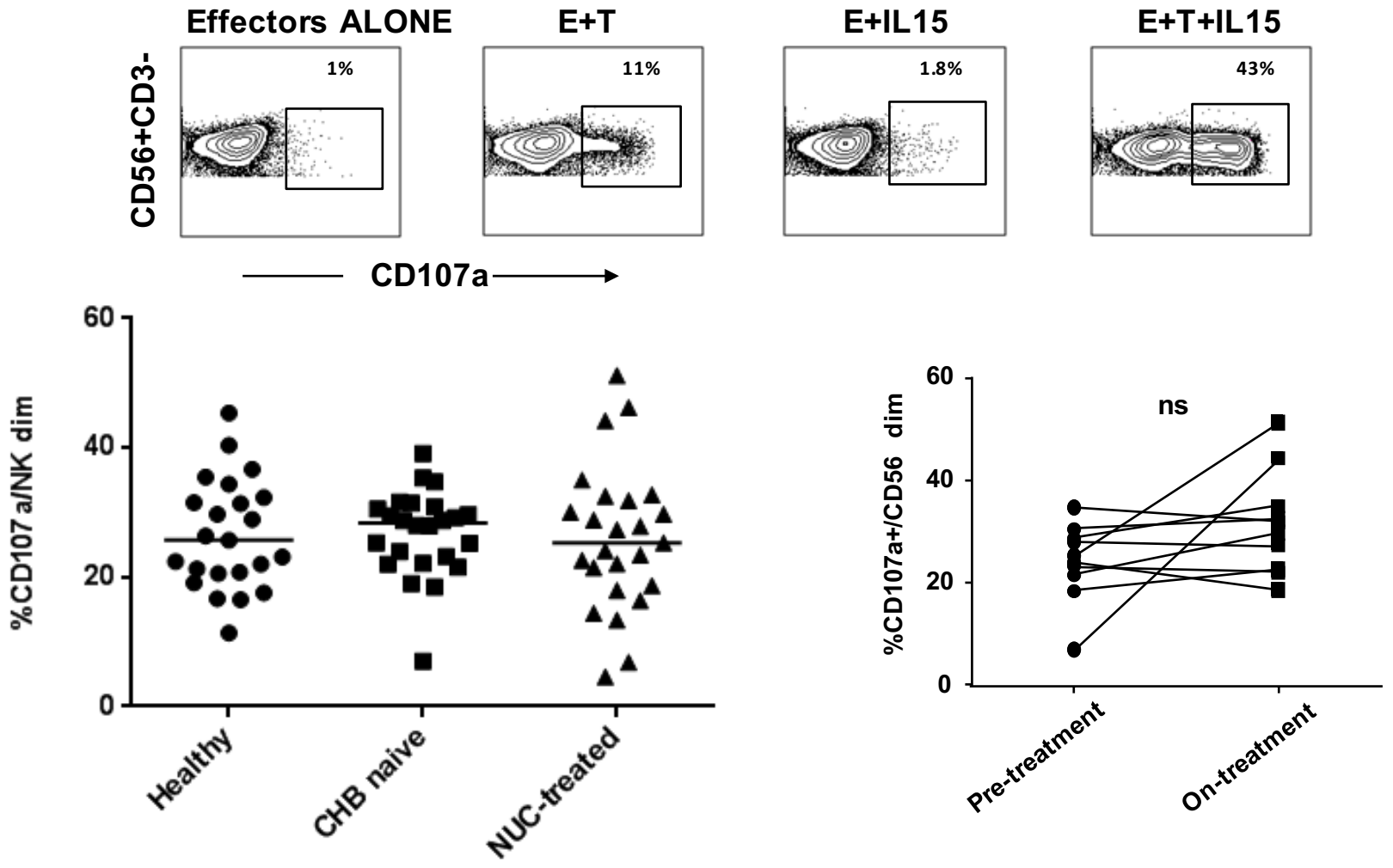






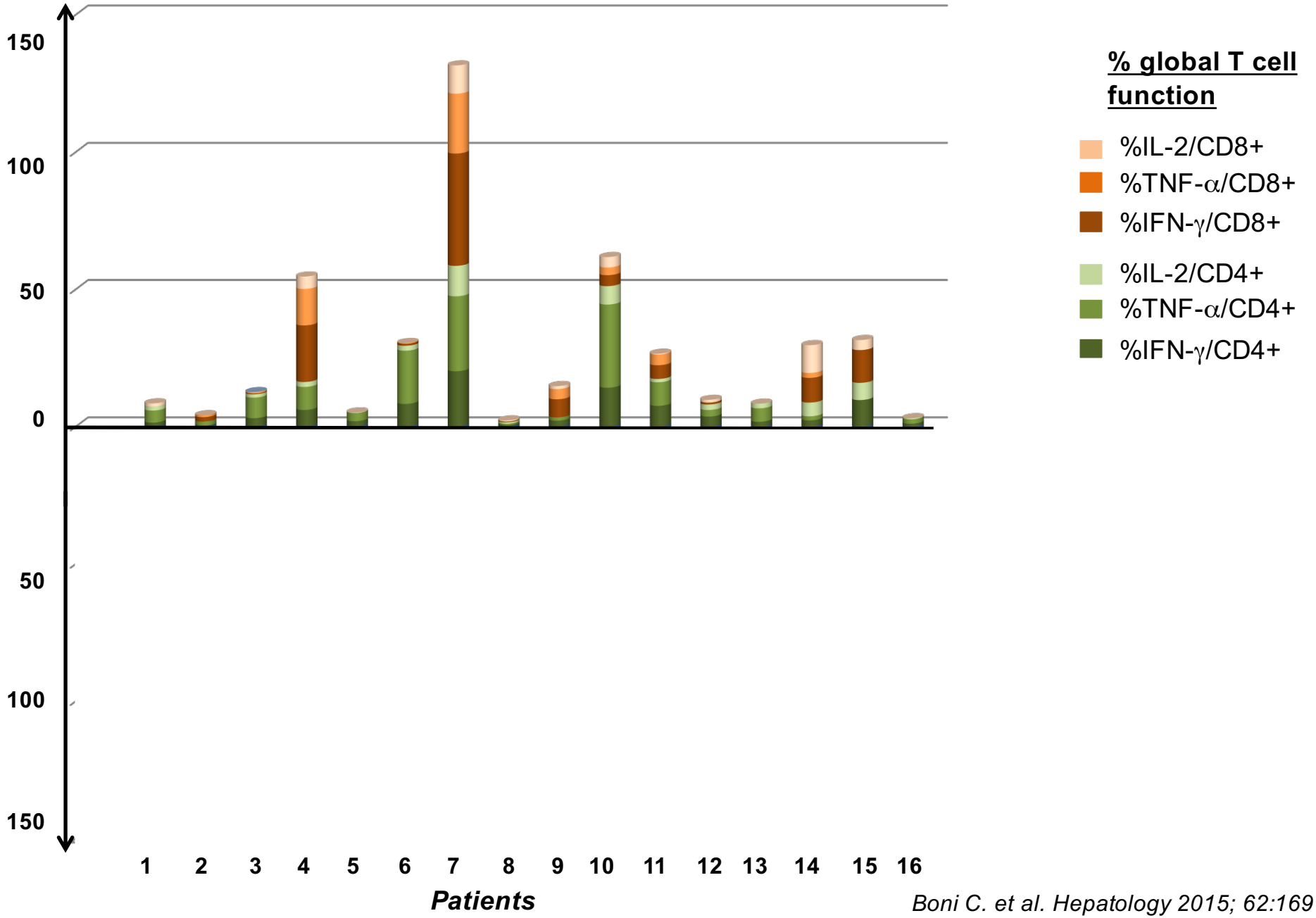
Boni C. et al. Hepatology 2015; 62:1697-709

Capacity to degranulate of NK cells is not significantly affected by NUC treatment



Boni C. et al. Hepatology 2015; 62:1697-709

Reciprocal behaviour of NK cell and HBV-specific T cell responses In NUC-treated patients



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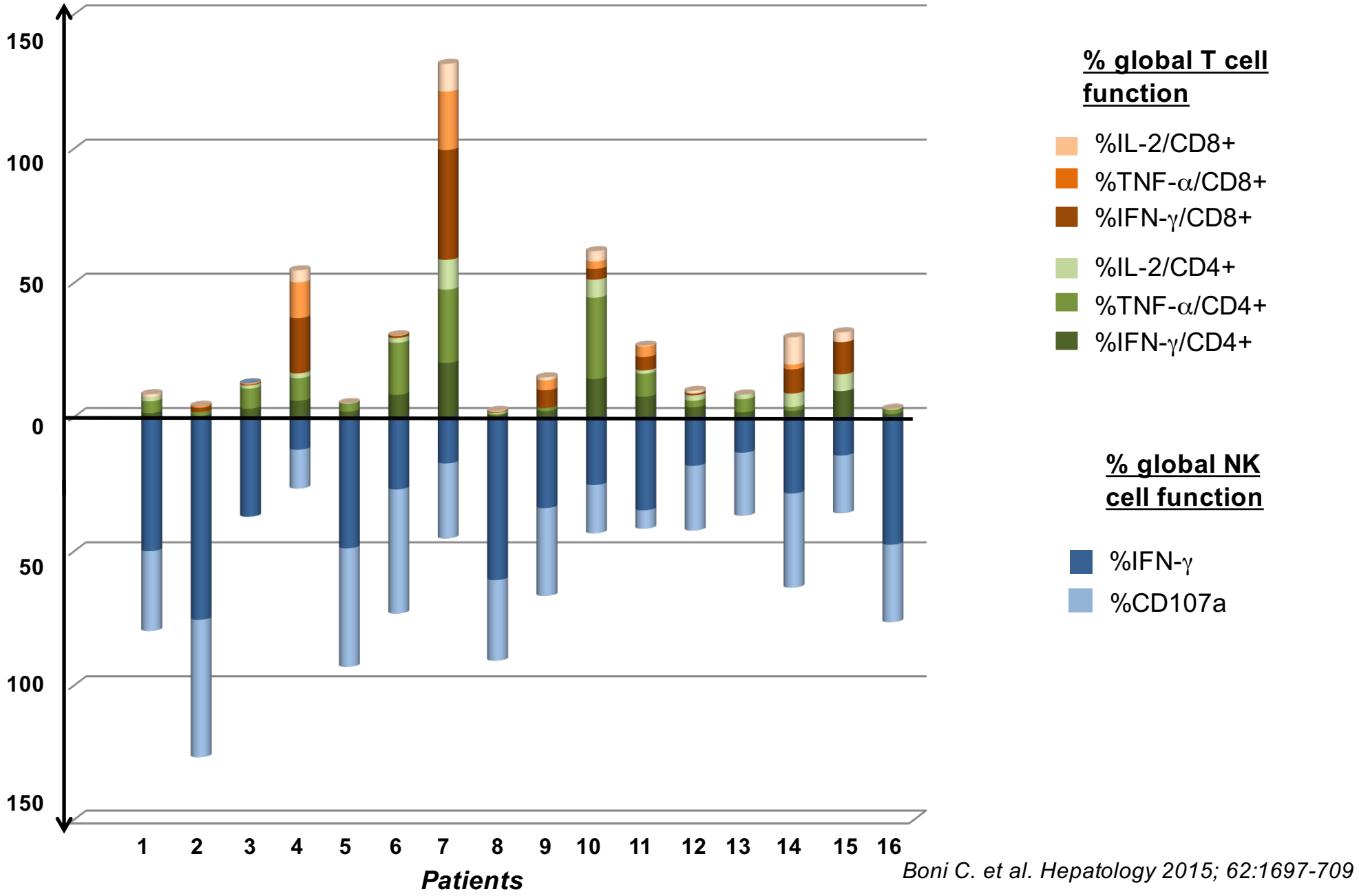
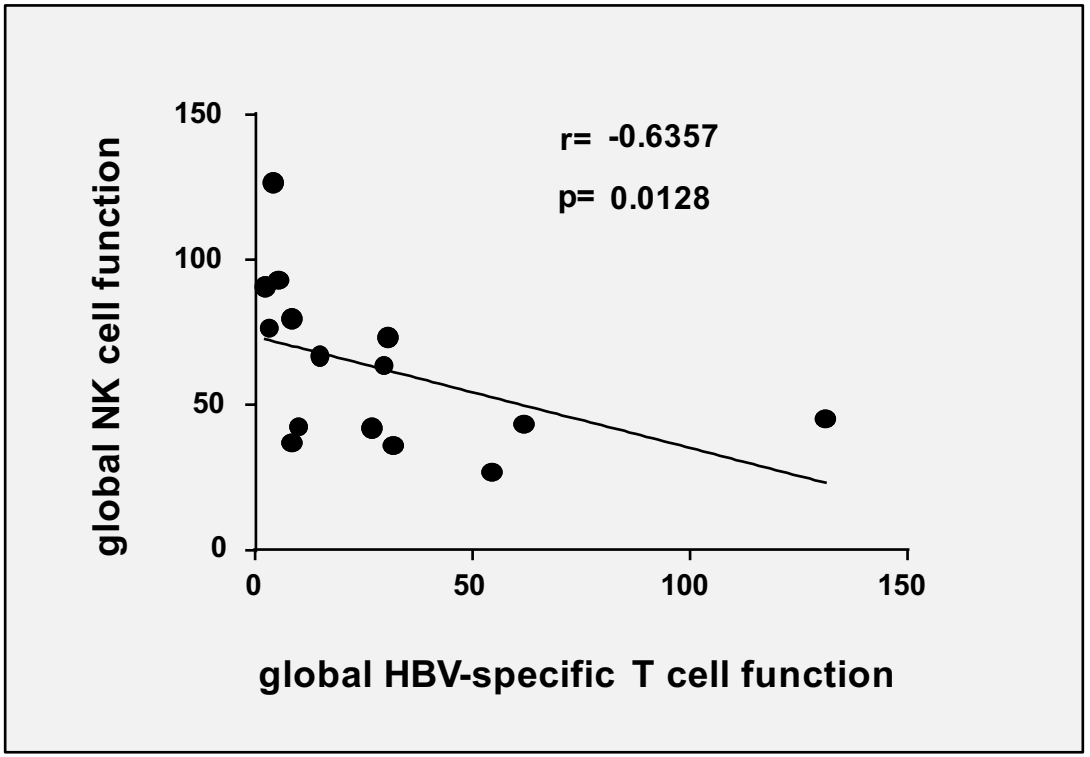


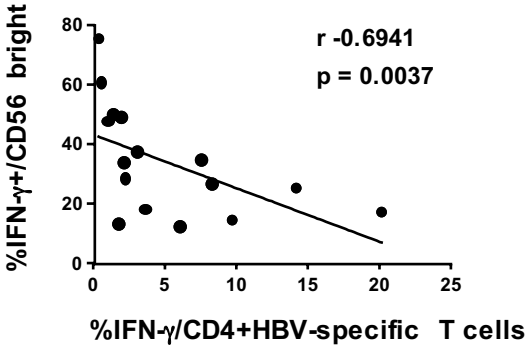
Boni C. et al. Hepatology 2015; 62:1697-709

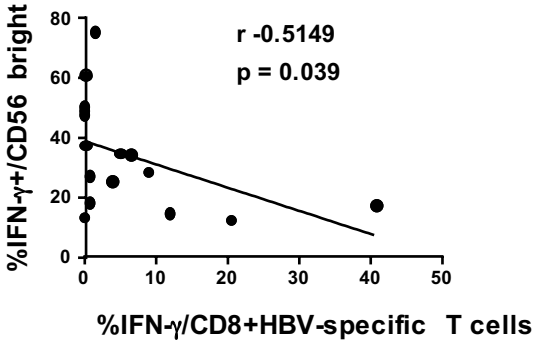
Inverse correlation between global NK cell and global HBV-specific T cell functions in 16 NUC-treated patients

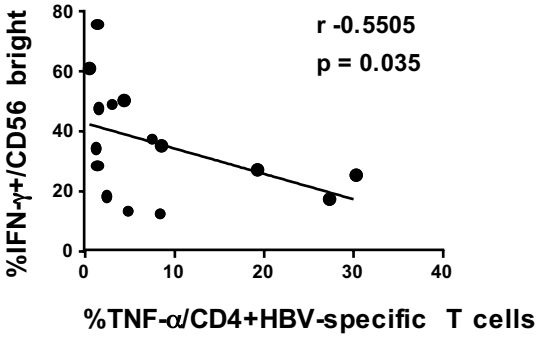


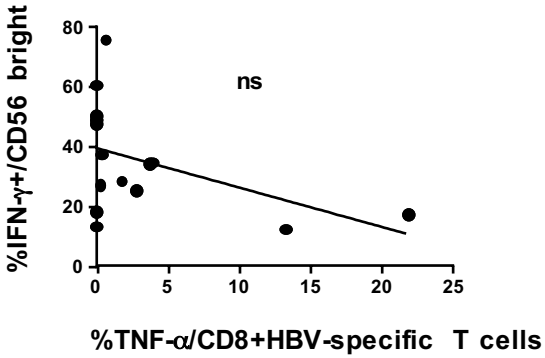
Boni C. et al. Hepatology 2015; 62:1697-709

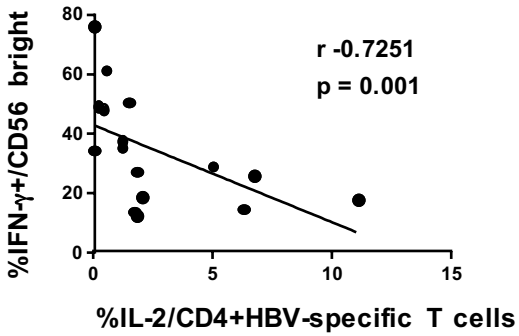
Inverse correlation between NK cell IFN-γ production and HBV-specific T cell cytokine production in NUC-treated patients

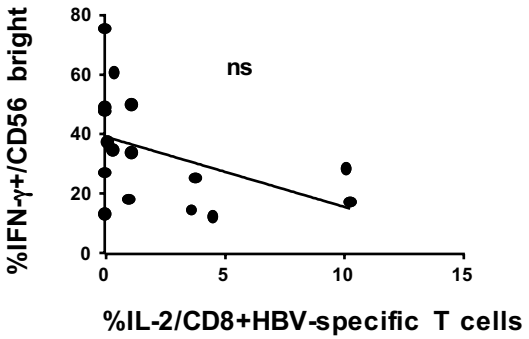




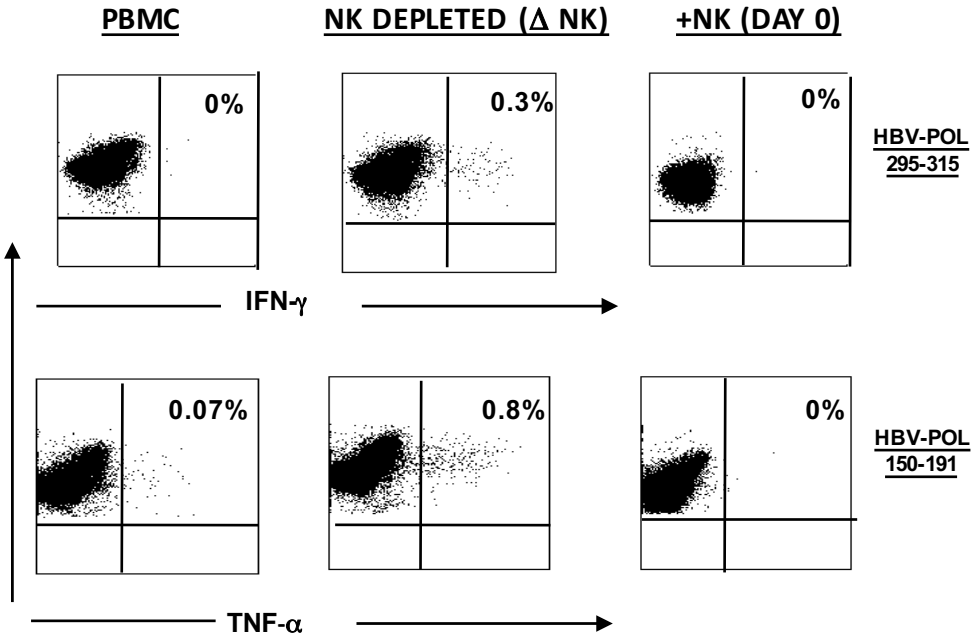


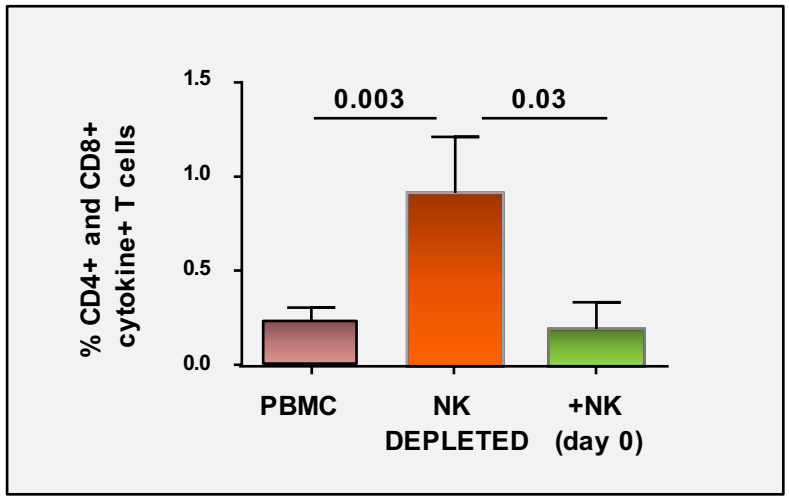




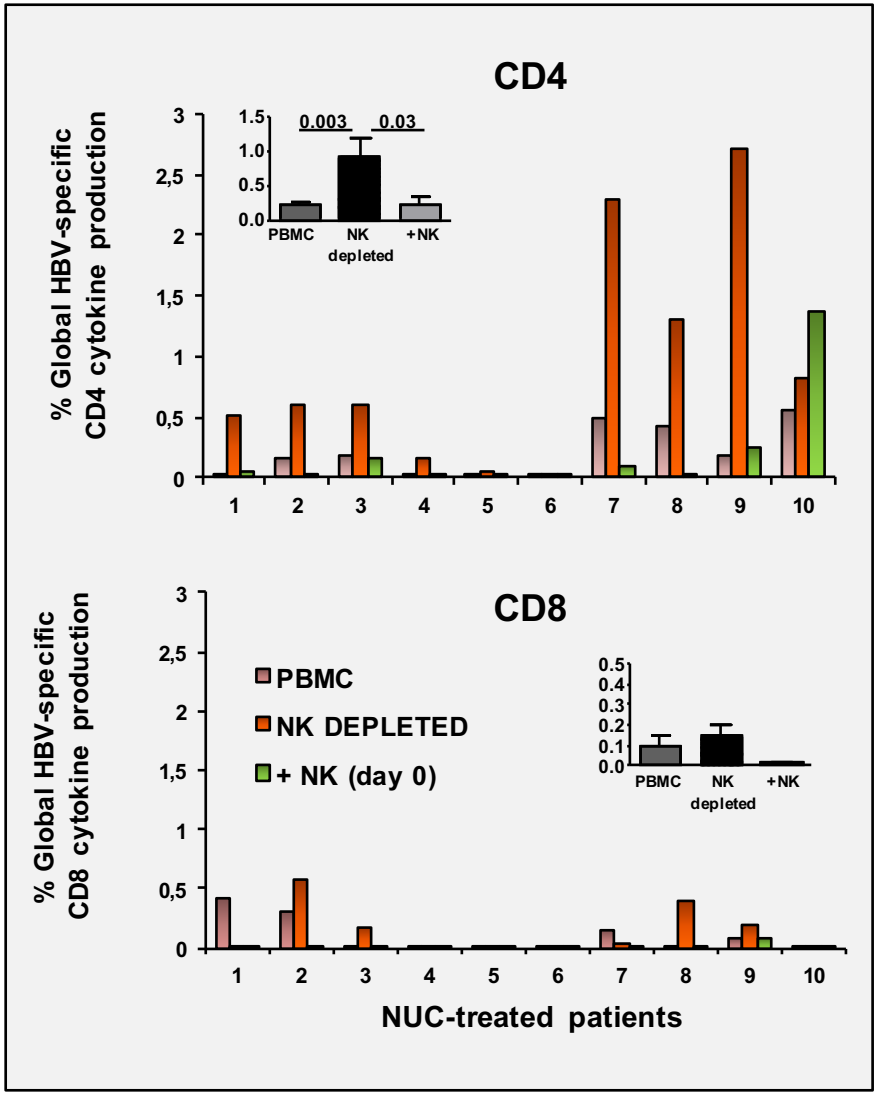


Improvement of HBV-specific T cell function after NK cell depletion by cell sorting in NUC-treated patients (1)



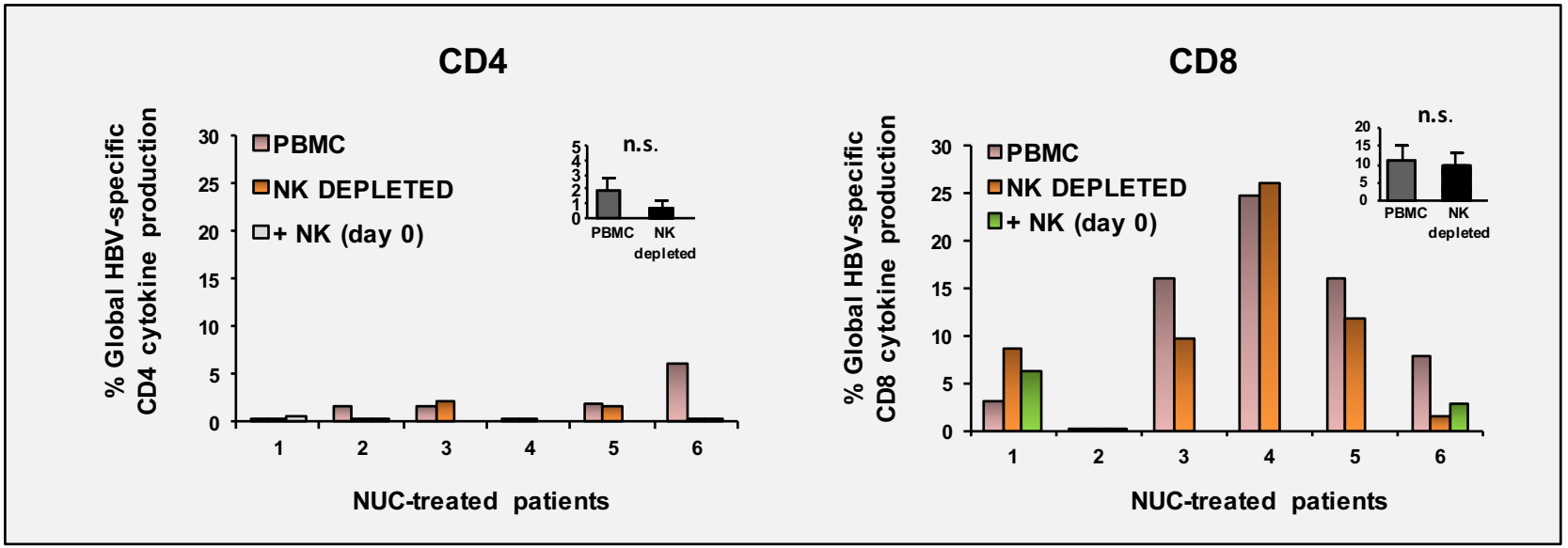


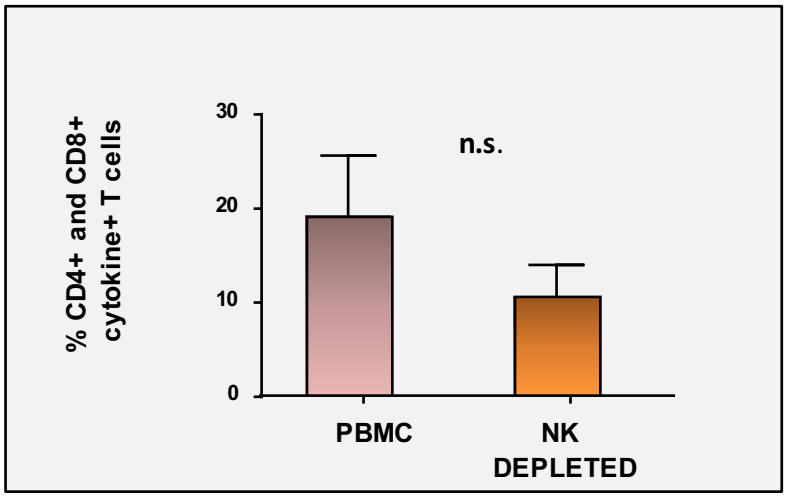
HBV-Core and Polymerase



NK cell depletion does not affect the function of T cells of different specificity in NUC-treated patients

CMV, EBV, FLU



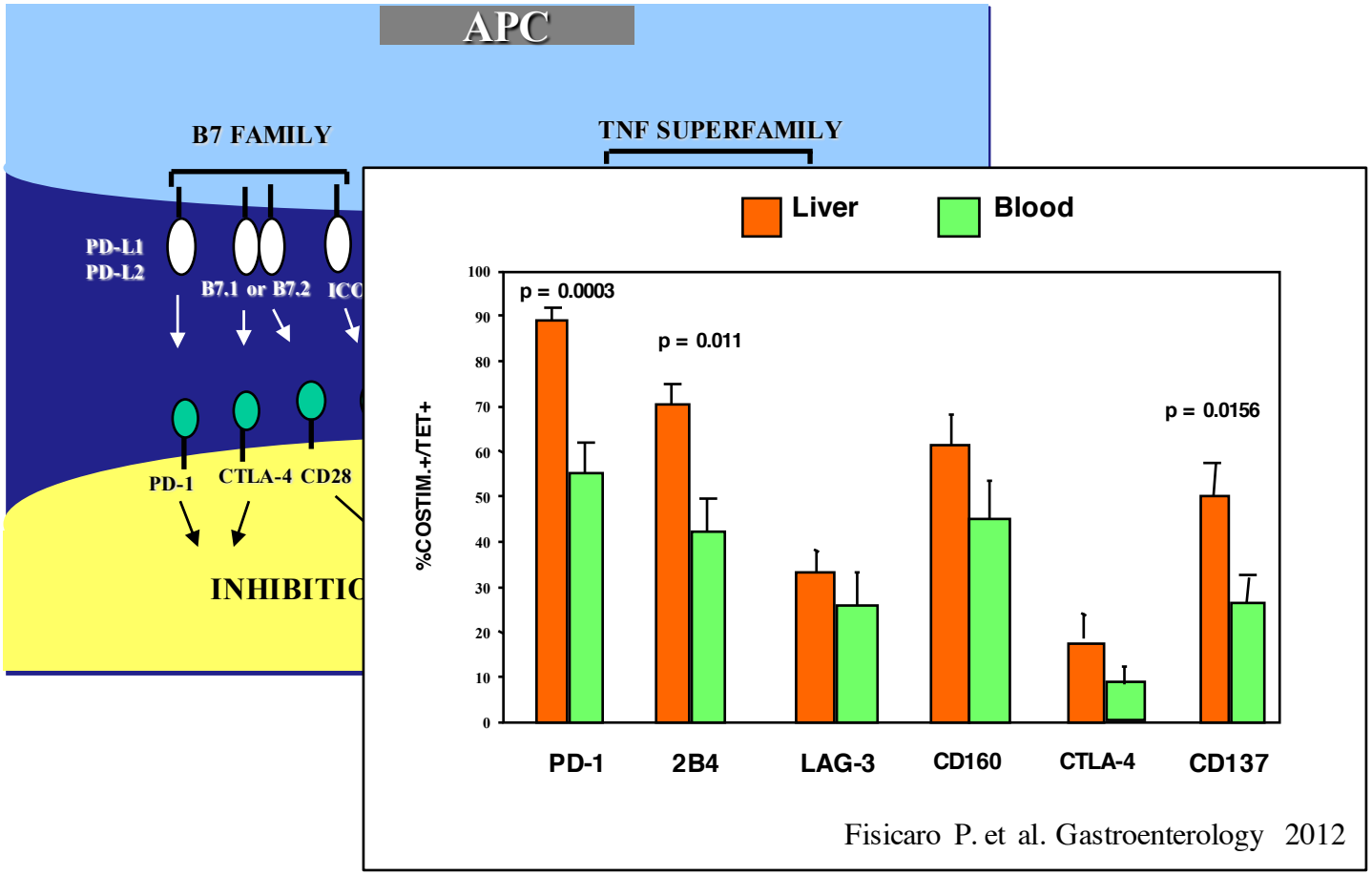


Boni C. et al. submitted

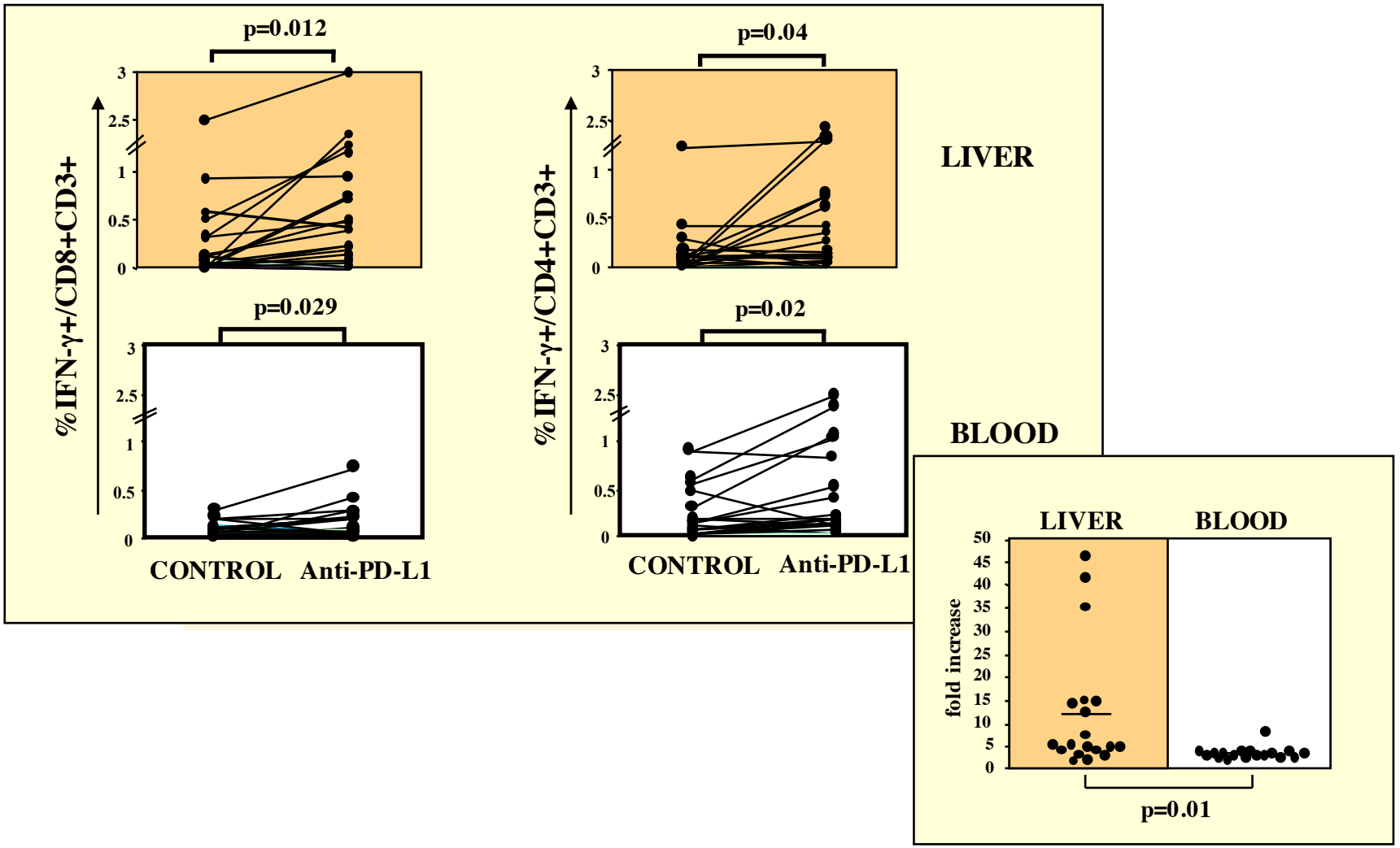
List of topics

- HBV-specific T cells in chronic infection
- NK cell response and its regulatory role on HBV-specific T-cell response
- **Potential strategies to reconstitute the anti-viral T cell function**
- Molecular basis of CD8 cell dysfunction in chronic HBV infection

Expression of various inhibitory receptors on circulating and intrahepatic virus-specific CD8 cells of patients with chronic HBV infection



Expression of PD-1 and CD127 on circulating and intrahepatic virus-specific CD8 cells of patients with chronic HBV infection and effect of PD-1/PD-L1 blockade on the T cell function



Fisicaro P. et al. Gastroenterology 2010, 2012

MESSAGE

Strategies based on the correction of individual dysregulated pathways can only induce a partial restoration of the T cell function

HBV-specific T cells

↓

Genome-wide expression profiling

↓

Mysregulated genes and pathways associated with T cell exhaustion

↓

Correction strategies to restore anti-viral T cell functions

TRANSCRIPTOME STUDY IN ACUTE, RESOLVED AND CHRONIC HBV INFECTION

Patient	infection	ALT
A1	ACUTE	1785
A2	ACUTE	998
A3	ACUTE	659
A4	ACUTE	1118
A5	ACUTE	211
R1	RESOLVED HEP B	16
R2	RESOLVED HEP B	17
R3	RESOLVED HEP B	20
R4	RESOLVED HEP B	12
CH1	CHRONIC	40
CH2	CHRONIC	96
CH3	CHRONIC	68
CH4	CHRONIC	63

CONTROLS		CELL SPECIFICITY
H1	HEALTHY	FLU
H2	HEALTHY	FLU
H3	HEALTHY	FLU
H4	HEALTHY	FLU
H5	HEALTHY	FLU

→ Isolation of HBV/FLU-specific CD8+ T cells by cell sorting

↓

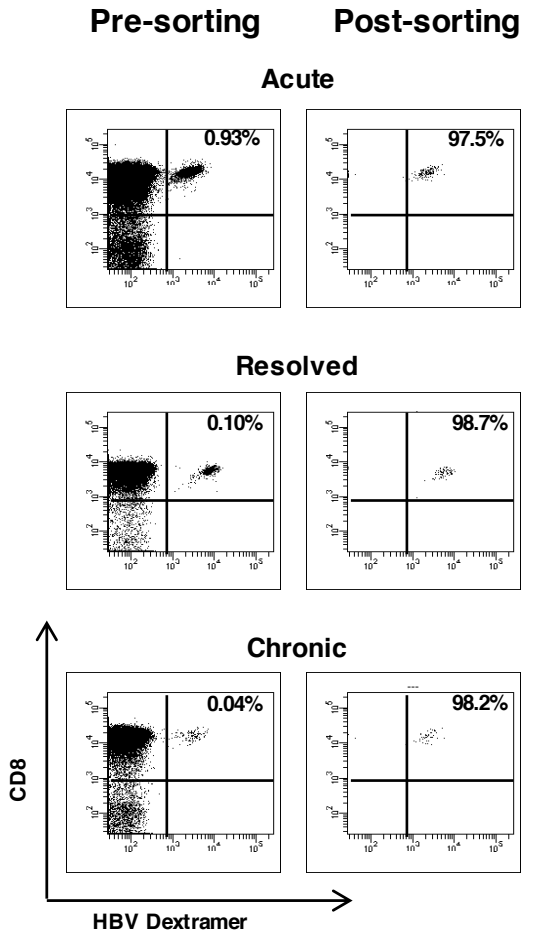
RNA extraction and amplification

↓

Gene expression by microarray analysis (4x44K Agilent)

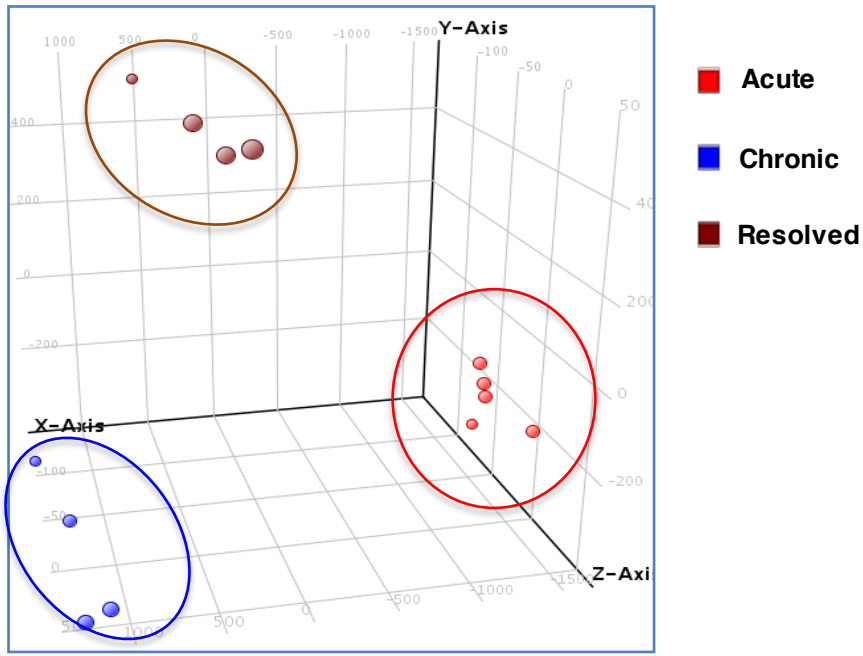
↓

VALIDATION AND DISCOVERY OF NEW TARGETS



TRANSCRIPTOME STUDY IN ACUTE, RESOLVED AND CHRONIC HBV INFECTION

Visualization of overall sample distribution and distances between groups of patients by principal component analysis (PCA)



Fisicaro P. et al. submitted

Results and conclusion of molecular analysis of HBV-specific CD8 T-cells

1. Downregulation of genes involved in mithocondrion function
2. Dna repair
3. Proteasome

Exhausted HBV-specific CD8 cells are deeply impaired at a metabolic and energetic level with a prevalent down-regulation of various core cellular processes centered on extensive mitochondrial alterations.

A significant improvement of mitochondrial and antiviral CD8 functions was elicited by mitochondrion-targeted antioxidants

Main message

A deep metabolic and energetic impairment is typical of exhausted T cells

Question

Is restoration of an efficient anti-viral T cell function an achievable objective?

Evidence

Multiple levels of correction will likely be needed to restore an efficient anti-viral T cell function

(unless correction affects specific core central processes able to indirectly affect other distal regulatory pathways)

Acknowledgments

C. Boni
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