

**Historical Perspective of HIV-exposed
Seronegative Individuals (ESN):
Has nature done the experiment for us?
(Notes from the 20th century)**

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First report of T cell responses in PBMC of HIV-exposed, Ab-negative individuals:

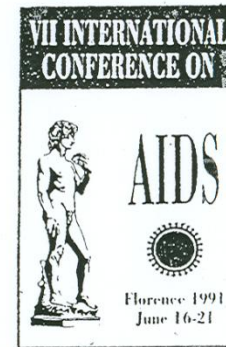
**T cell responses towards HIV in infected individuals with and without zidovudine therapy, and in HIV exposed sexual partners.
A Ranki *et al.* *AIDS* 1989; 3: 63-9**

Abstracts from Winnipeg group (1990-93)

- K Fowke *et al.* Detection of HIV sequences in lymphocyte DNA from African specimens by the polymerase chain reaction. 5th Int Conf AIDS. Kinshasa. Oct 1990. Abst #FPC 29**
- K Fowke *et al.* Heterogeneity in susceptibility to HIV-1 in continuously exposed prostitutes. VIII Int conf AIDS/III STD World Cong Amsterdam. July 1992. Abst #PoC 4026**
- F Plummer *et al.* Heterogeneity in susceptibility HIV-1. Laboratory Tumor Cell Biol Ann Meeting, Bethesda. Aug 1992**
- N Simonsen *et al.* Evidence for resistance to HIV-1 among continuously exposed prostitutes. Res Conf Immunologic & Host Genetic Resistance to HIV infection and Disease. Bethesda, Feb 1993**
- K Fowke *et al.* Evidence for resistance to HIV-1 among continuously exposed prostitutes. Keystone Symp Frontiers HIV Pathogenesis. Albuquerque, Mar-Apr 1993. Abst #Q215**
- F Plummer *et al.* Evidence of resistance to HIV among continuously exposed prostitutes in Nairobi, Kenya. 9th Int Conf AIDS/ IV STD World Cong. Berlin, June 1993. Abst #WA-A07-3**

VII International Conference on AIDS (Florence, 1991)

M Clerici *et al.* T helper cell assays detect exposure to HIV-1 antigen earlier than demonstration of infection by serum antibodies or polymerase chain reaction.
Abst # WA 76



M Clerici *et al.* Exposure to HIV-1 indicated by HIV-specific T helper cell responses before detection by polymerase chain reaction and serum antibodies. *J Infect Dis.* 1991; 164: 178-82

Question

If HIV-specific cell-mediated immunity (CMI) can be detected in HIV-exposed individuals before seroconversion,.....

Can HIV exposure result in CMI without detection of serum Ab?

.... also from the Florence AIDS Conference

JV Giorgi *et al.* HIV-specific cellular immunity in high-risk HIV-1 seronegative homosexual men. Abst # WA 120

**M Clerici *et al.* Specific T cell immunity to HIV-1 envelope peptides in seronegative individuals with recent exposure to HIV-1.
J Infect Dis. 1992; 165: 1012-9**

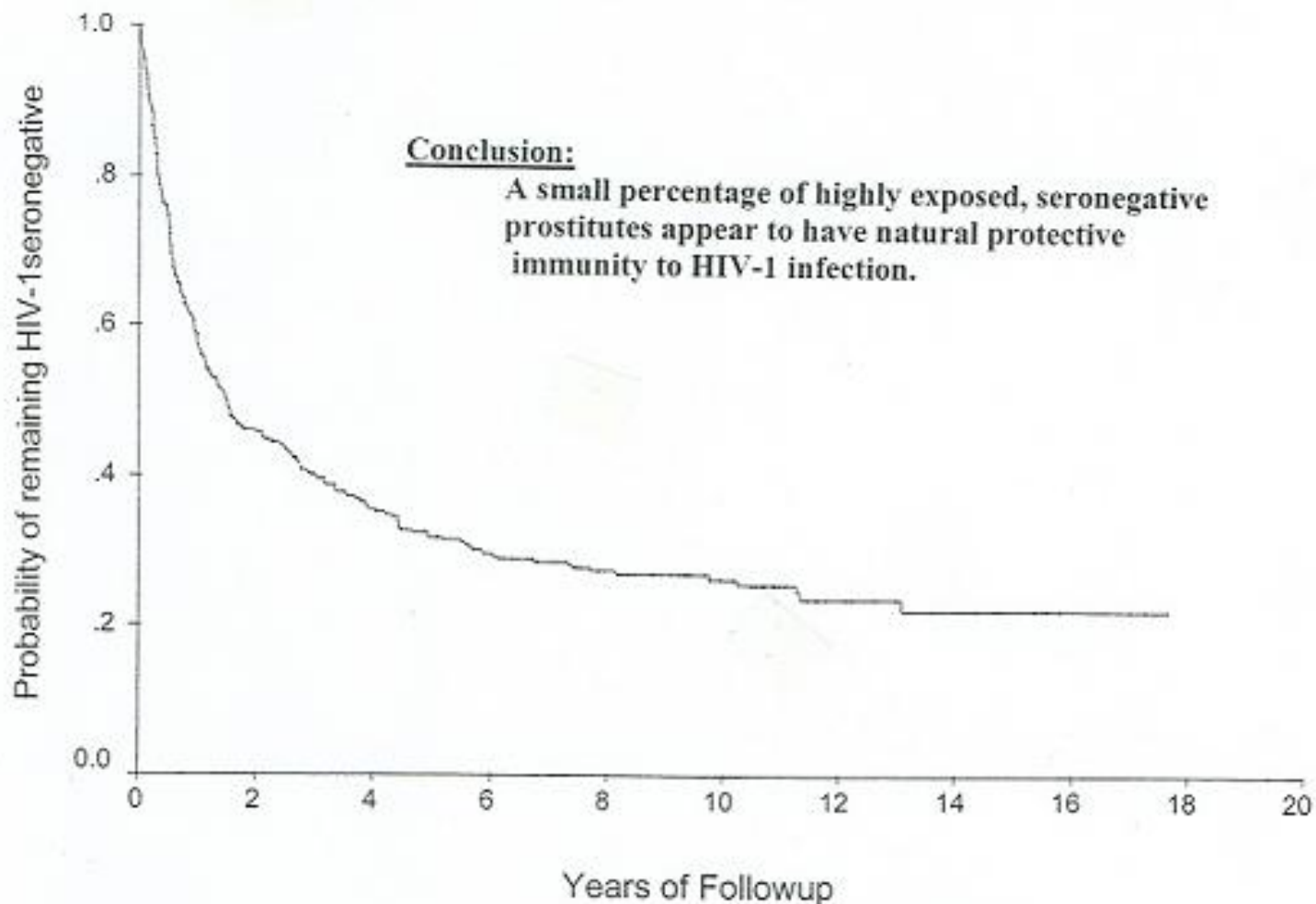
ESN generate T helper cell responses to HIV antigens

H Kelker *et al.* Lymphocytes from some long-term seronegative heterosexual partners of HIV-infected individuals proliferate in response to HIV antigens. *AIDS Res Hum Retrovir.* 1992; 8: 1355-9

Resistance to HIV-1 infection among persistently seronegative prostitutes in Nairobi, Kenya

Keith R Fowke, Nico J D Nagelkerke, Joshua Kimani, J Neil Simonsen, Aggrey O Anzala, Job J Bwayo, Kelly S MacDonald, Elizabeth N Ngugi, Francis A Plummer

The Lancet. 1996; 348: 1347-51



ESN generate HIV-specific CTL responses

- P Langlade-Demoyen *et al.* Human immunodeficiency virus (HIV) nef-specific cytotoxic T lymphocytes in noninfected heterosexual contact of HIV-infected patients. *J Clin Invest.* 1994; 93: 1293-7**
- S Rowland-Jones *et al.* HIV-specific cytotoxic T-cells in HIV-exposed but uninfected Gambian women. *Nature Medicine.* 1995; 1: 59-64**
- S Rowland-Jones *et al.* Cytotoxic T cell responses to multiple conserved HIV epitopes in HIV-resistant prostitutes in Nairobi. *J Clin Invest.* 1998; 102:1758-65**

ESN are found among intravenous drug abusers

W Barcellini *et al.* *In vitro* production of type 1 and type 2 cytokines by peripheral blood mononuclear cells from high-risk HIV-negative intravenous drug users. *AIDS*. 1995; 9: 691-4

Neonates and ESN

Prior to the use of antiretroviral therapy in pregnant women, vertical transmission of HIV (maternal → fetal/perinatal) transmission was 25-30%.

Neonates (70-75%) who did not become HIV infected comprise an ESN cohort that was continuously exposed

Are neonates who escaped vertical transmission examples of ESN?

▪

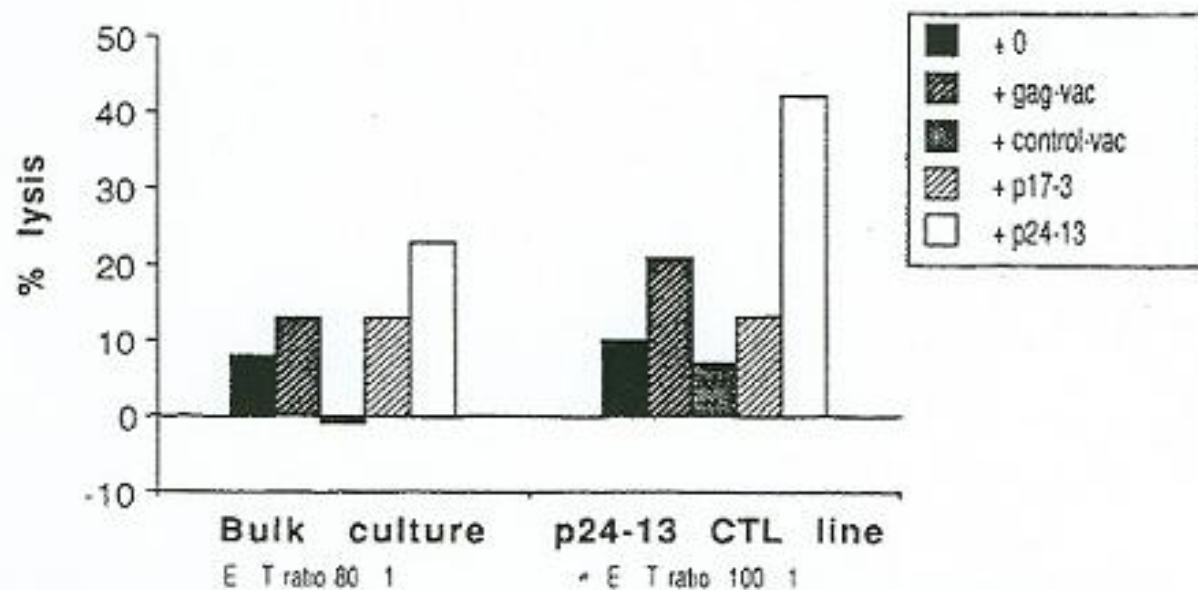
Newborns of HIV-infected mothers can generate T helper and CTL responses to HIV antigens

- M Clerici *et al.* Cellular immune factors associated with mother-to-infant transmission of HIV. *AIDS*. 1993; 7: 427-33**
- R Cheyneier *et al.* Cytotoxic T lymphocyte responses in the peripheral blood of children from human immunodeficiency virus-1 infected mothers. *Eur J Immunol*. 1992; 22: 2211-7**
- S Rowland-Jones *et al.* HIV-specific cytotoxic T-cell activity in an HIV-exposed but uninfected infant. *The Lancet*. 1993; 341: 860-1**
- A DeMaria *et al.* Occurrence of HIV-1 specific cytolytic T cell activity in apparently uninfected children born to HIV-1 infected mothers. *J Infect Dis*. 1994;170: 1296-1303**

HIV-specific cytotoxic T-cell activity in an HIV-exposed but uninfected infant

SARAH L. ROWLAND-JONES
DOUGLAS F. NIXON MARIAN C. ALDHOUS
FRANCES GOTCH KOYA ARIYOSHI
NICHOLAS HALLAM J. SIMON KROLL
KARIN PROEBEL ANDREW MCMICHAEL

Lancet 1993; 341: 860-1



CTLs from the baby recognise HLA-B8-restricted antigen in HIV gag.

Mucosal IgA antibodies in ESN

S Mazzoli *et al.* HIV-specific mucosal and cellular immunity in HIV-seronegative partners of HIV-seropositive individuals. *Nature Medicine*. 1997; 3: 1250-7

Anti-HIV soluble factors in ESN

- J Levy et al. CD8 cell noncytotoxic antiviral activity in human immunodeficiency virus-infected and –uninfected children. *J Infect Dis.* 1998; 177:470-2**
- S Stanford et al. Lack of infection in HIV-exposed individuals is associated with a strong CD8+ cell noncytotoxic anti-HIV response. *Proc Natl Acad Sci USA.* 1999; 96:1030-5**
- L Purci et al. Antigen-driven C-C chemokine-mediated HIV-1 suppression by CD4+ T cells from exposed uninfected individuals expressing the wild-type CCR5- allele. *J Exp Med.* 1997; 186:455-60**

What is the minimum number of HIV exposures required to induce CMI in an HIV-naïve individual?

Tested PBMC from single needle-stick health care workers for CMI to HIV *env* peptides

M Clerici *et al.* HIV-specific T-helper activity in seronegative health care workers exposed to contaminated blood. *JAMA*. 1994; 271: 42-6

LA Pinto *et al.* ENV-specific cytotoxic T lymphocyte responses in HIV seronegative health care workers occupationally exposed to HIV-contaminated body fluids. *J Clin Invest*. 1995; 96; 867-76

Conclusion:

One needle stick is sufficient to induce HIV-specific T helper and CTL responses, some responses lasting for 12 months.

Question:

Would a single parenteral exposure protect against HIV infection?

**Are there observations in the
SIV/macaque model that resemble
the human ESN findings?**

Macaques inoculated with subinfectious doses of SIV appeared resistant to challenge with an infectious dose of SIV.

M Clerici *et al.* T-cell proliferation to subinfectious SIV correlates with lack of infection after challenge of macaques. *AIDS*. 1994; 8: 1391-5

Titration of SIV_{mne}CLE11S from 0.001-to-100 AID on 3-28-1990

Mag#	IV exposed	IR challenge	<u>Viral isolation/PCR (wks)</u>						Pathology
	3-28-1990	7-29-1991	2	4	8	14	27	80	
89078	0.1 AID	100 AID	-/-	+/+	-/-	-/-	-/-	-/-	None
F90184	None	100 AID	-/+	+/+	+/+	+/+	+/+	+/+	CD4 depld.
89143	0.001 AID	10 AID	-/-	-/-	-/-	-/+	-/-	-/-	None
J90219	None	10 AID	+/+	+/+	+/+	+/+	+/+	NT	AIDS

?? = virus isolation/ PCR signal

At 51wk after IR, test T cell responses to 9 SIV peptides, H3N2 FLU, PHA

Mag#	J90044	89078	F90184	89143
Peptide response:	0/9	5/9	0/9	6/9

Categories of ESN cohorts

Repeated unprotected sexual HIV exposure

Promiscuous sex

Stable discordant couples

Repeated intravenous HIV exposure

I.V. drug abusers

Limited number of HIV exposures

Hemophiliacs

Continuous HIV exposure

Fetal/perinatal exposure

Single accidental HIV exposure

Health care workers

Laboratory workers

Using history to speculate...

“Moving forward” by glancing back....

Questions

Is the ESN phenomenon due to:

protective genetics?

activated immunity?

anti-viral mechanisms?

combinations of above?

Does the homozygous CCR5 32-bp ($\Delta 32/\Delta 32$) deletion account for the ESN phenomenon?

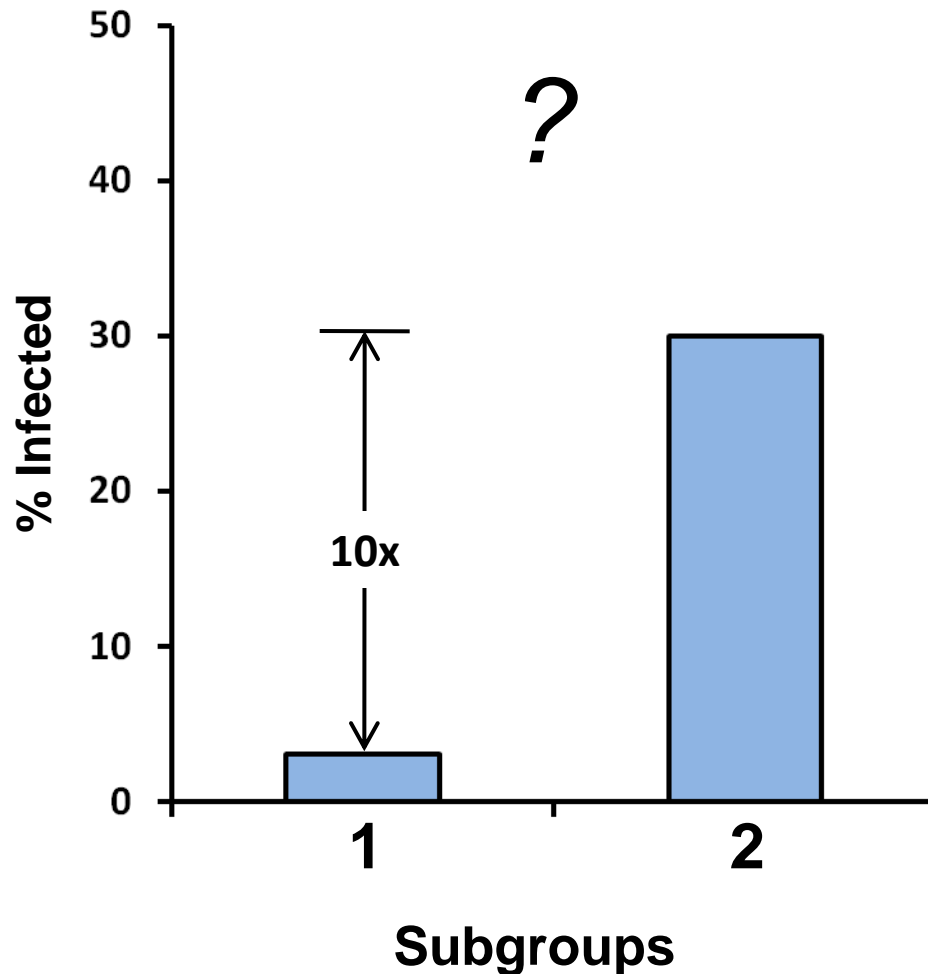
R Liu *et al.* Homozygous defect in HIV-1 coreceptor accounts for resistance of some multiply-exposed individuals to HIV-1 infection. *Cell*. 1996; 86: 367-77

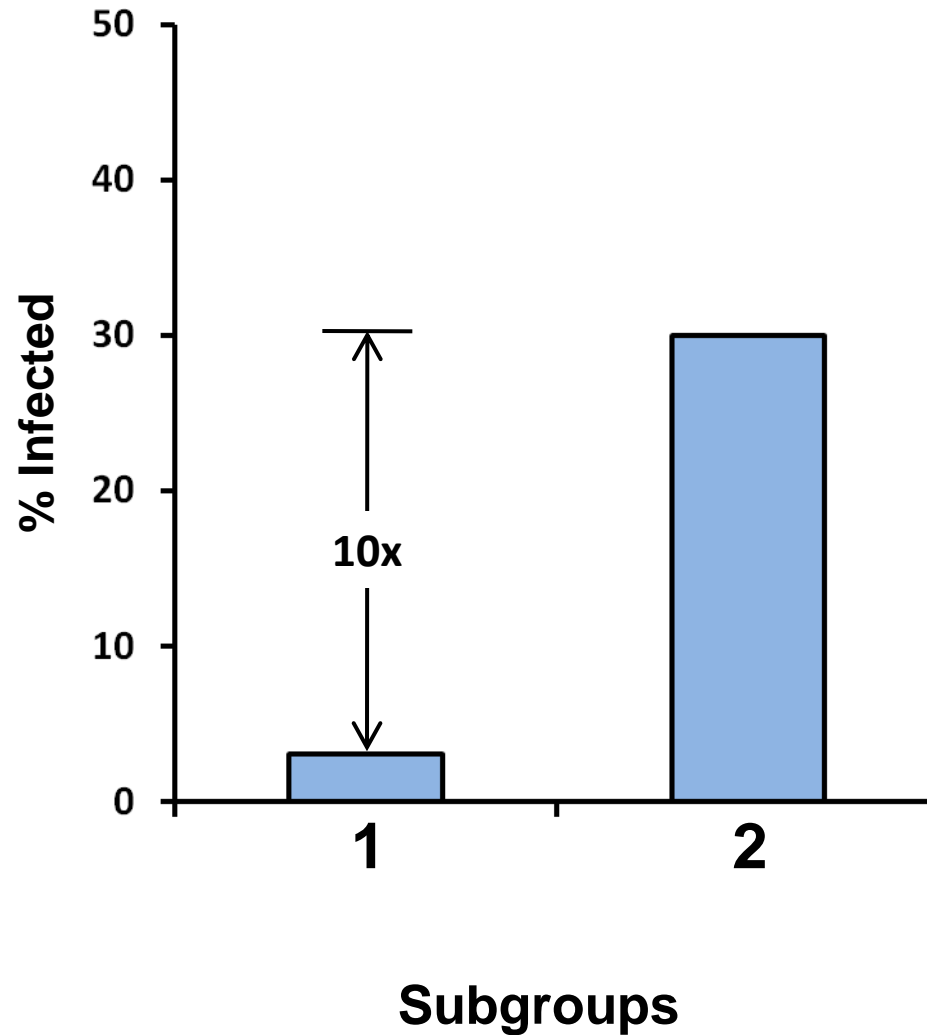
W Goh *et al.* Protection against human immunodeficiency virus type 1 infection in persons with repeated exposures: Evidence for T cell immunity in the absence of inherited CCR5 coreceptor defects. *J Infect Dis*. 1999; 179: 548-57

**Back to the maternal→fetal/perinate
model of ESN,...**

**to consider an unexpected genetic
effect, and possibly other
complexities.**

Could an ESN group be divided into subgroups with different infection risks?

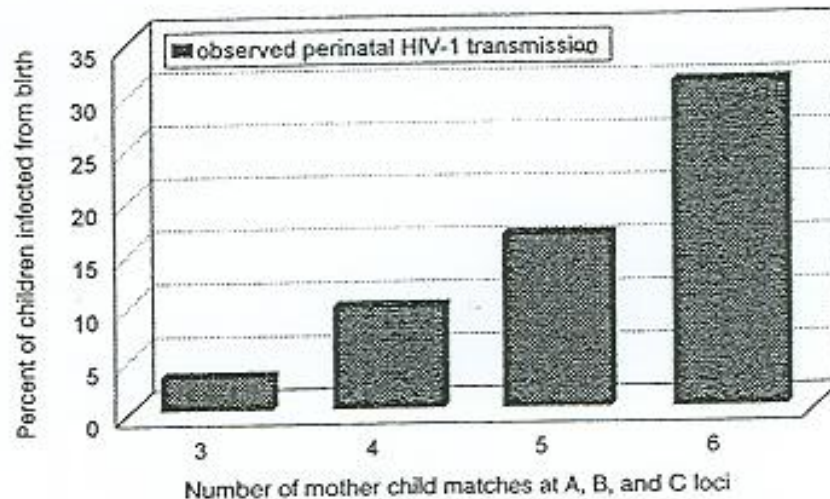




Would the 3% infected frequency be due to homozygosity for a resistance gene?

Mother-Child Class I HLA Concordance Increases Perinatal Human Immunodeficiency Virus Type 1 Transmission

Kelly S. MacDonald, Joanne Embree, Simon Njenga, Nico J. D. Nagelkerke, Irene Ngatia, Zeena Mohammed, Brian H. Barber, Jeckoniah Ndinya-Achola, Job Bwayo, and Francis A. Plummer



Mother-child class I HLA concordance and perinatal HIV-1 transmission.

MacDonald *et al. J. Infect. Dis.* 1998; 177: 551-556

Polycarpou *et al. AIDS Res Hum Retrovir.* 2002 ; 18 : 741-6

In the MacDonald (Kenya) and Polycarpou (USA) cohorts, multiple mechanisms appear to contribute to resistance against vertical transmission:

One (affecting ~70% uninfected) appears to be independent of maternal/fetal HLA I matching.

The second (affecting remaining susceptibility) is regulated by maternal/fetal HLA-I concordance.

**These findings suggest that MHC
allorecognition is, in itself, an
evolutionary driving force.**

Do anti-viral and/or immunologic mechanisms contribute to the ESN phenomenon?

Back to the SIV/macaque experiment

This single SIV/macaque experiment would need to be repeated (with controls) and expanded to verify the 1990-92 findings.

If confirmed, this result would suggest an inducible protective effect, because macaques are susceptible to AIDS.

This approach could open mechanistic analyses studies not possible in humans.

In contrast to long-term non-progressors (LTNP) and elite controllers (EC), ESN have passed the first test by resisting infection.

Have these potentially-relevant cohorts been overlooked in the search for AIDS vaccines?

ESN was a major reason for suggesting that CMI contributes to protection against HIV infection.

A Th1/Th2 switch is a critical step in the etiology of HIV infection. M Clerici, GM Shearer. *Immunology today*. 1993; 143; 107-11.

The Th1/Th2 hypothesis of HIV infection: new insights. M Clerici, GM Shearer. *Immunology today*. 1994;45; 575-81

At least 3 factors contribute to failure of CMI to protect against HIV infection

- **HIV rapidly invades mucosal immunologic tissue.**
- **HIV rapidly activates multiple mechanisms of immune dysregulation and CD4 T cell depletion.**
- **HIV can rapidly mutate away from CTL epitopes used in AIDS vaccines.**

Would solving the ESN mystery be useful in developing an effective AIDS vaccine?

Depends on whether the ESN phenomenon includes inducible(?) immunologic and or anti-viral mechanisms, and whether these can be identified and elucidated.

Suggestion

Specific experiments should be developed:

1] to establish whether the ESN effect has inducible components,.....

and if it does,

2] to determine their mechanisms of activation and protection.

**As would be demonstrate in the 21st
century....**

**Noninfectious interactions between HIV and
CD4+ cells can result in immunopathogenesis.**

Herbeuval & Shearer. *Clin Immunol.* 2007

Boasso & Shearer. *Clin Immunol.* 2008

**So,.. could noninfectious interactions between
HIV and CD4+ cells also result in immune
protection against HIV infection?**

Protective immunity against HIV infection:

has nature done the experiment for us?

Shearer & Clerici. *Immunol today*. 1996

APOBEC3G?

- **Pido-Lopez *et al.* *Eur J Immunol.* 2009**
- **Biasin *et al.* *J Infect Dis.* 2007**
- **Soros *et al.* *PLoS Pathology* 2007**
- **Rugeles *et al.* *AIDS* 2003**

- **MacDonald *et al.* *J Infect Dis.* 1998**
- **Bedoya *et al.* *Current HIV Res.* 2008**