Genetic mechanisms of susceptibility to RSV disease

Steven R. Kleeberger

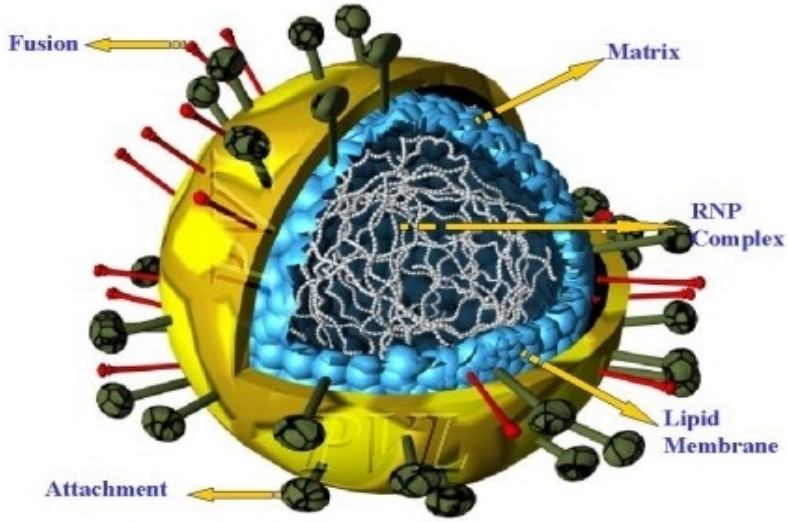
Immunity, Inflammation, and Disease Laboratory

National Institute of Environmental Health Sciences

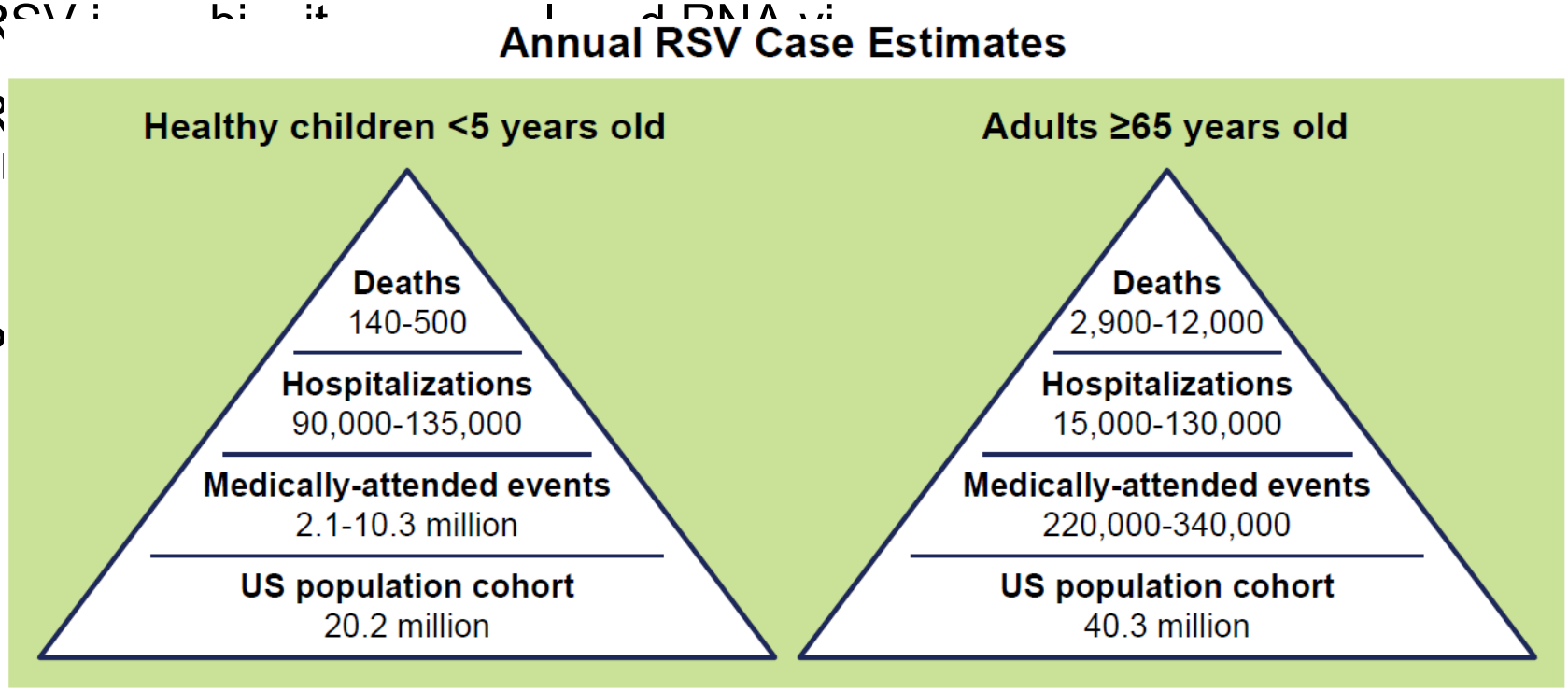
http://www.niehs.nih.gov/research/atniehs/labs/iidl/pi/enviro-gen/

kleeber1@niehs.nih.gov

What is RSV disease?

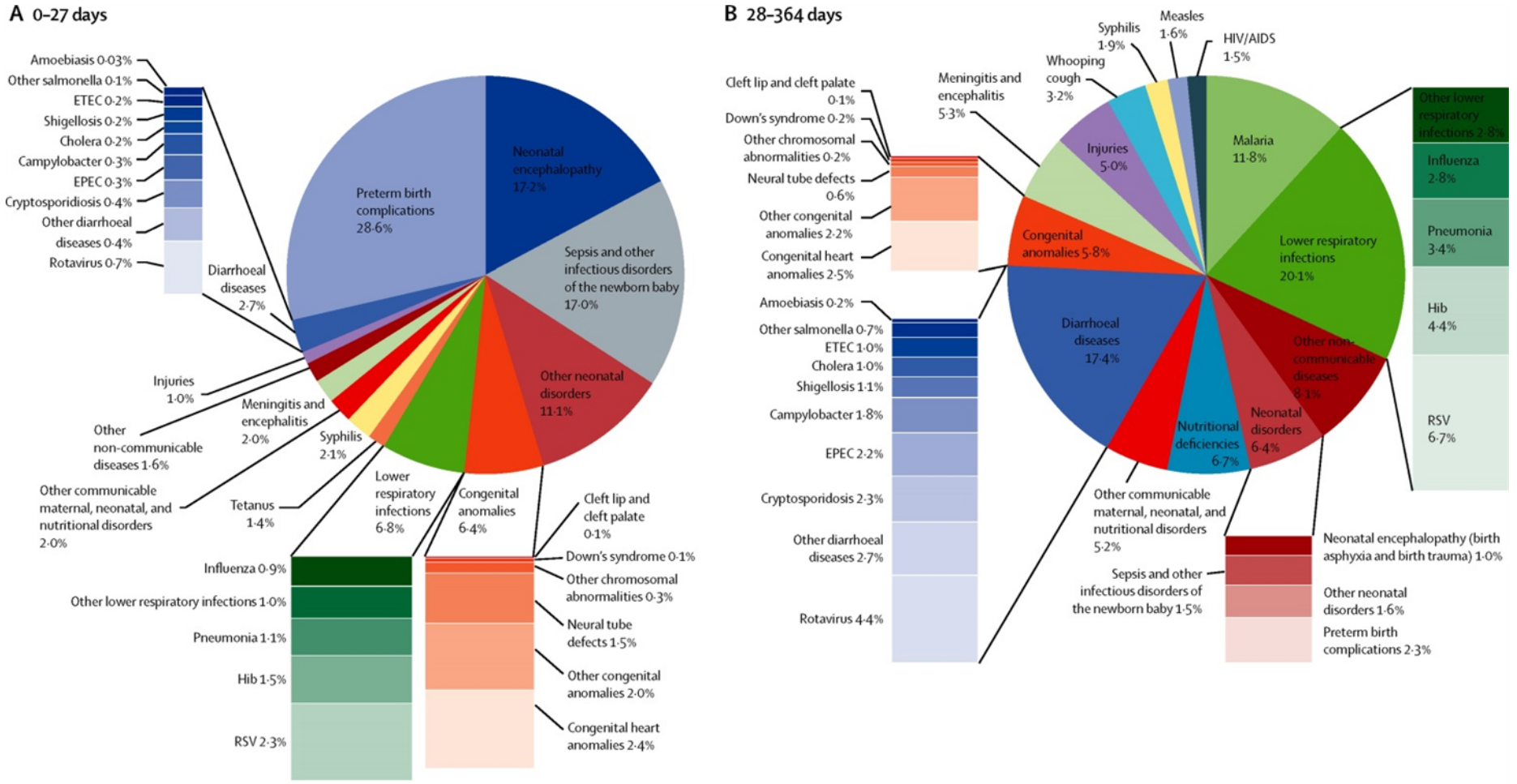


- R
- R
  a
- 7
- P
- No vaccine



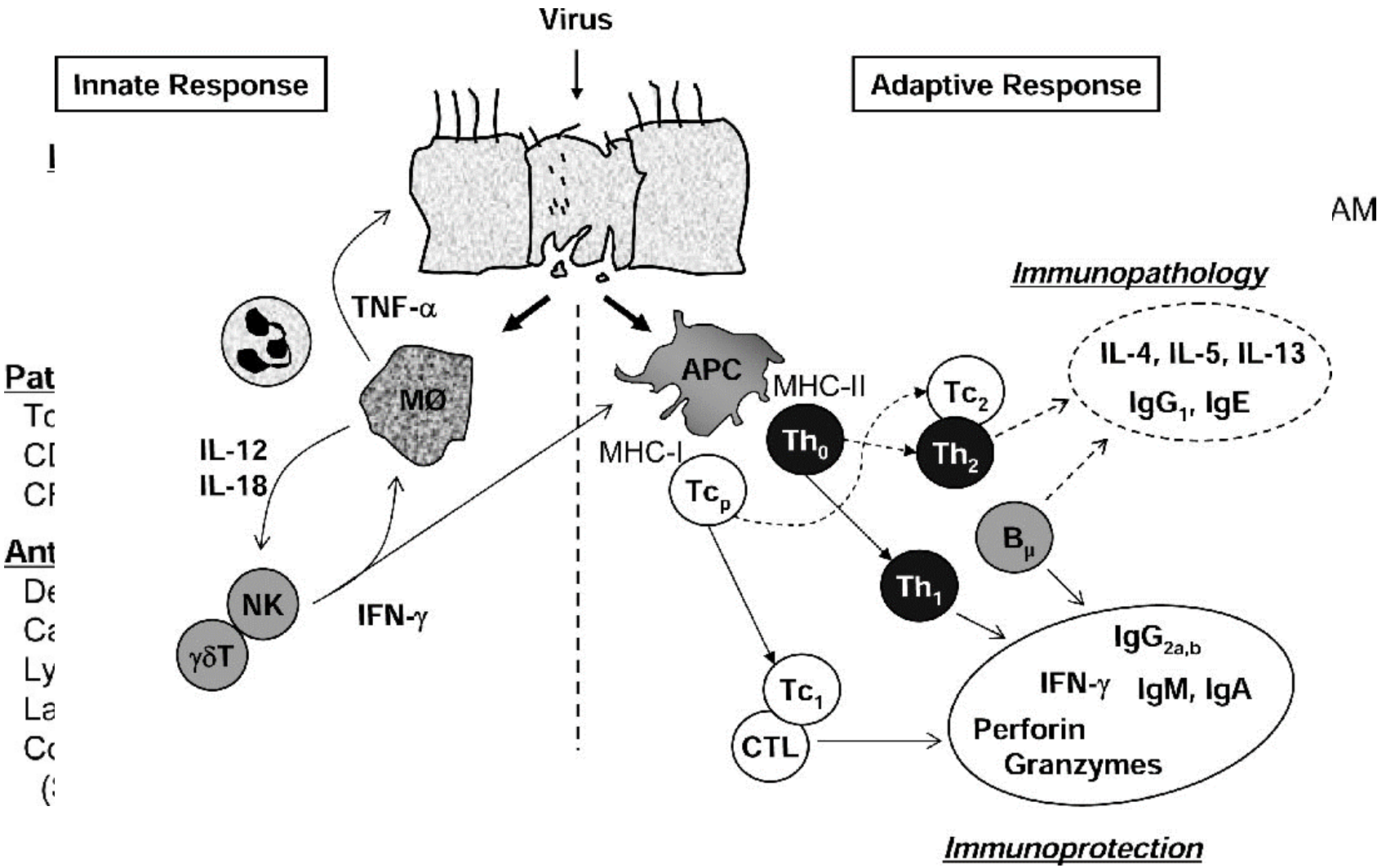
Adapted from Kurzweil et al, 2013

Global Burden of Disease and the Role of RSV



Lozano et al, *Lancet* 2012

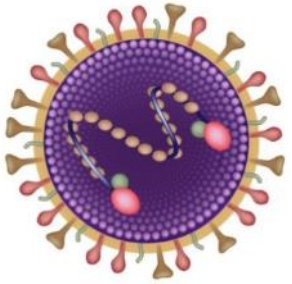
Mechanisms of susceptibility and response to RSV infection



Dakhama et al, 2005

Understanding the genetic basis of susceptibility to RSV disease severity

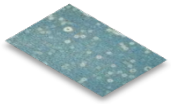
- **Traditional family-based studies, twin studies, and GWAS studies**
- **Association studies are useful, but candidate genes must be selected carefully**
  - **biological plausibility**
- **Animal models have proved to be useful to identify genes that contribute to RSV disease subphenotypes**
  - **expression array studies**
  - **genome scan**



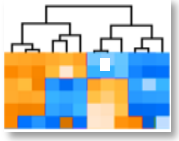
Schematic representation of strategy to identify environmental lung disease susceptibility genes

Study 1. Murine RSV Disease Susceptibility

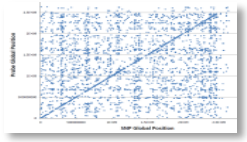
Inbred mouse strains



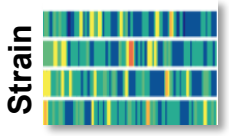
Human cell lines



Global gene expression analyses

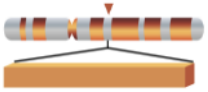


Expression QTL (eQTL) analyses



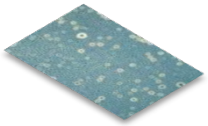
Haplotype association mapping

Study 2. RSV Disease, TLR4, and the environment



Candidate genes and networks

Biologically plausible candidates



Mechanistic functional exposure models

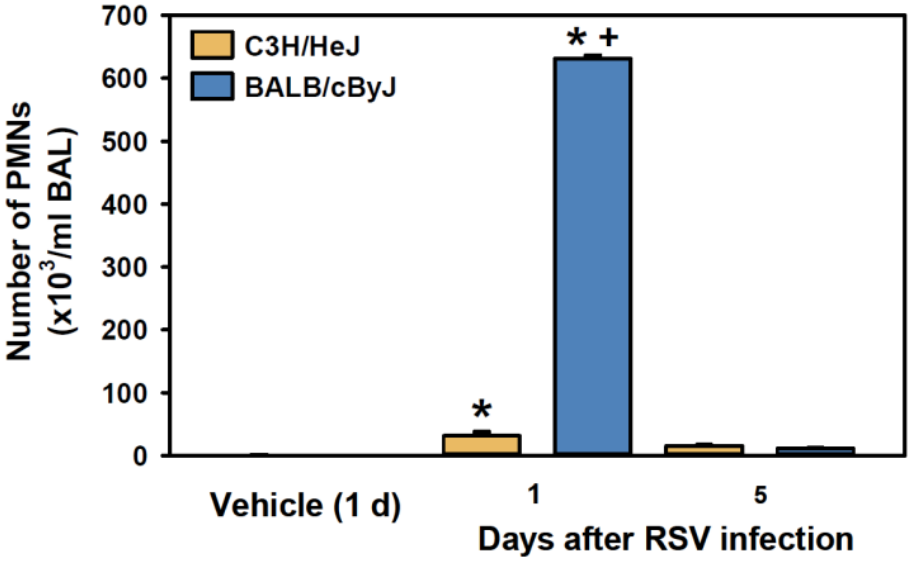
- Targeted deletion mice
- Transgenic mice
- Pharmacological inhibition
- siRNA inhibition

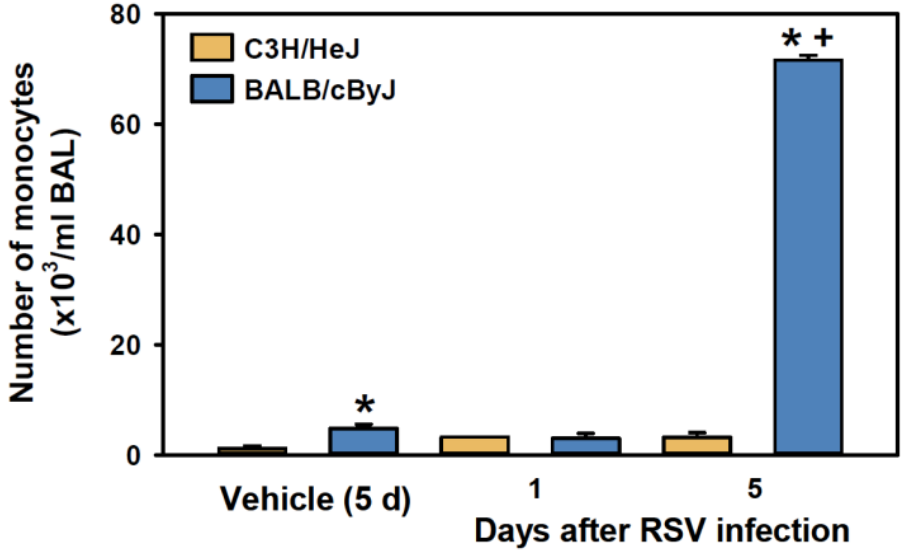
Test candidates in human populations

- Epidemiological studies
- Clinical studies

Study 1. What are the genetic determinants of susceptibility in a mouse model of severe respiratory syncytial virus (RSV)-induced disease?

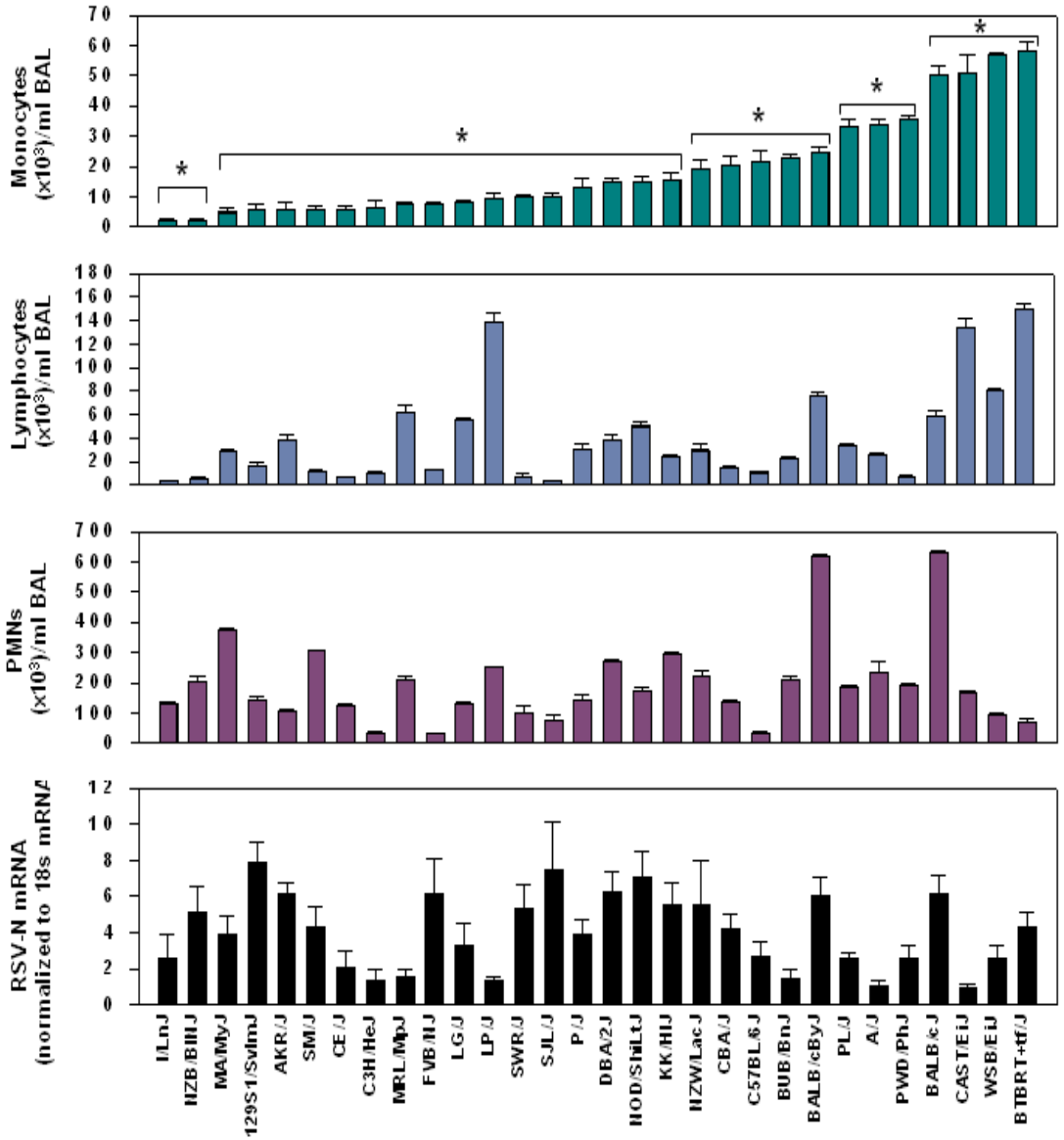
Time course of bronchoalveolar lavage (BAL) polymorphonuclear leukocytes (PMNs) and monocytes after vehicle and RSV infection





* p<0.05 vs. vehicle
\+ p<0.05 vs. C3H/HeJ
n = 7-9/group

Strain distribution patterns for RSV-induced disease phenotypes

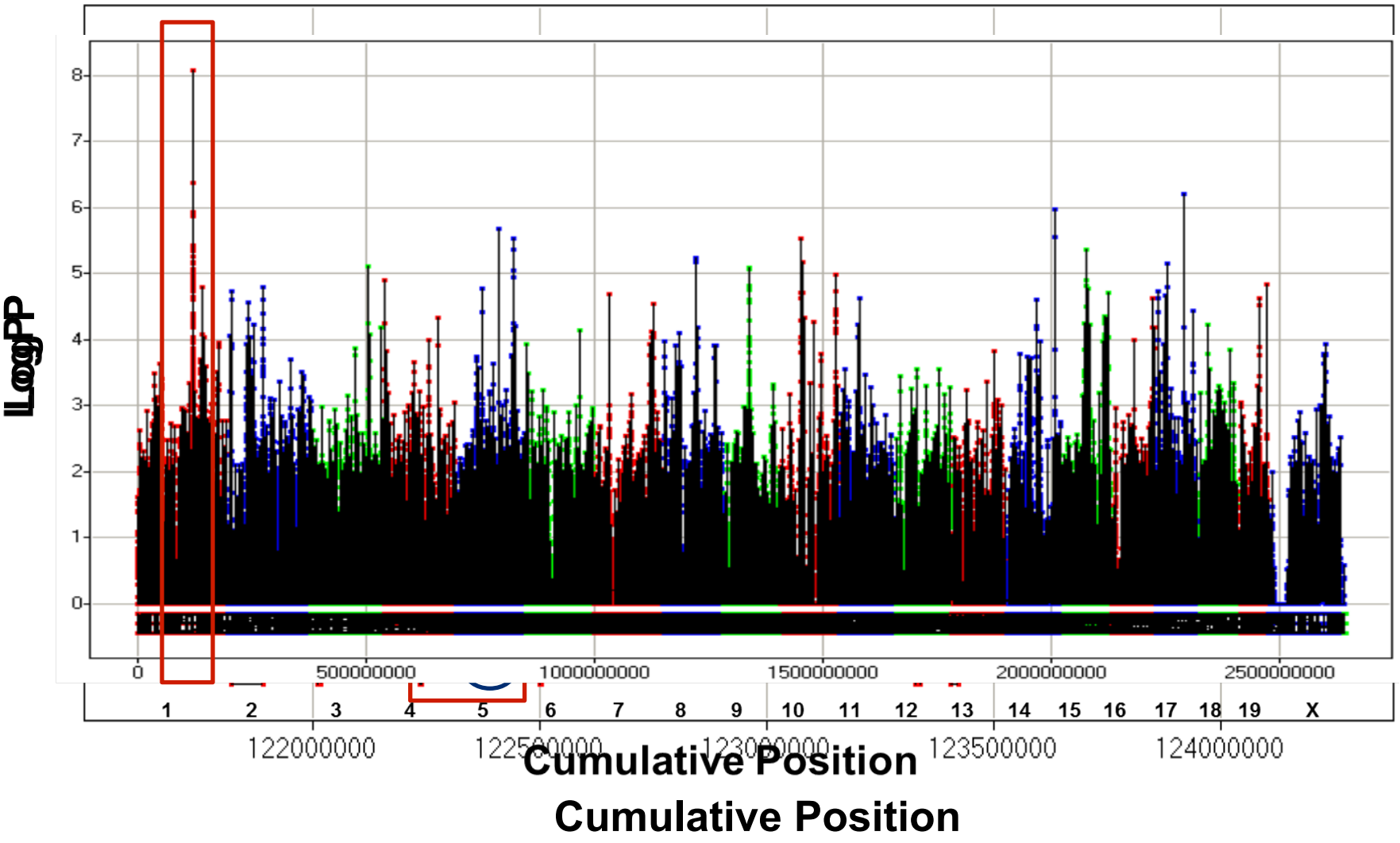


- Inter-strain variation
- Continuous distributions of phenotypes across strains
- Lack of correlation between phenotypes

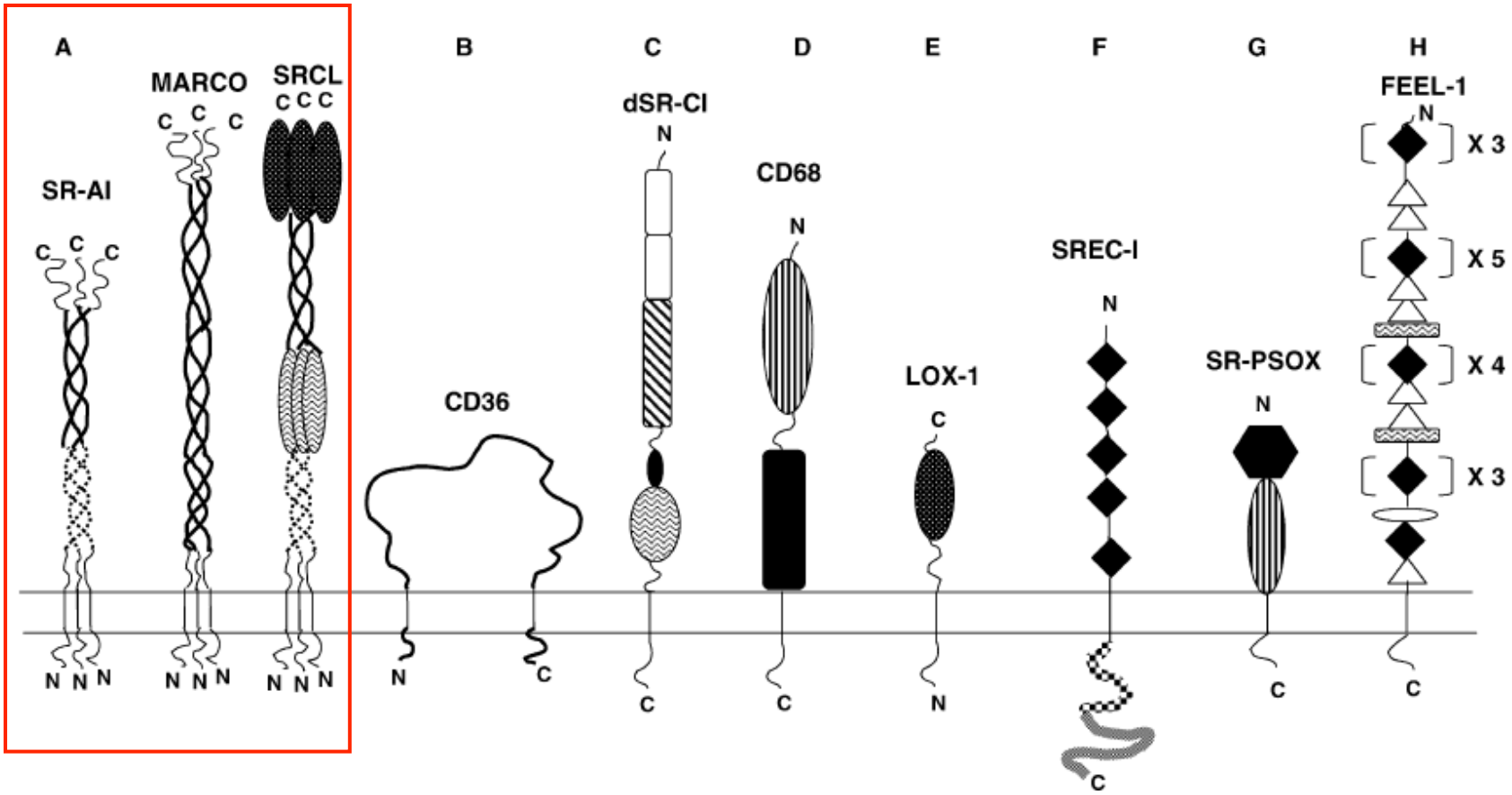
* within group not significantly different from each other

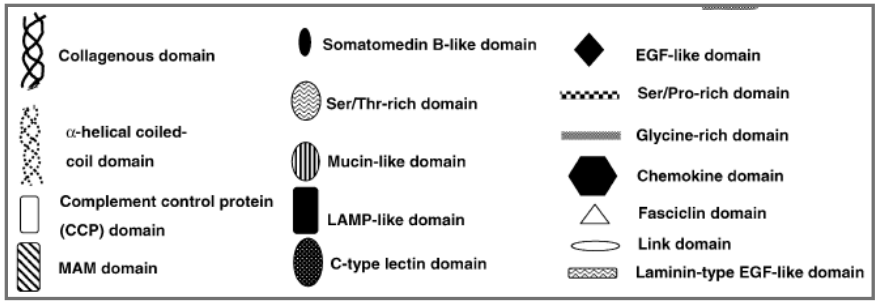
High et al, *EBioMedicine* 2016

Manhattan plot for RSV-induced BAL monocytes in 30 inbred strains of mice (SNPster)



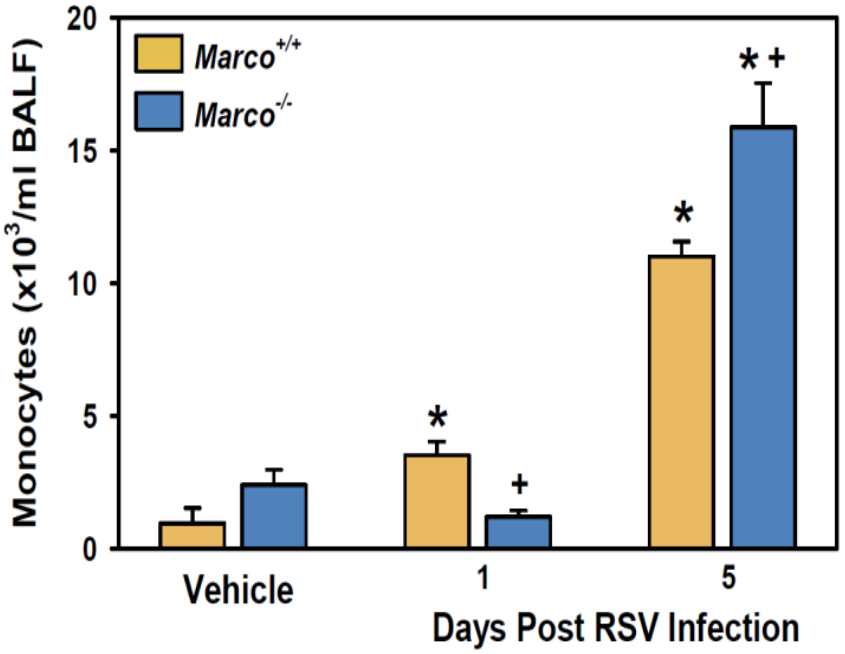
MARCO (macrophage receptor with collagenous structure) is a member of the scavenger receptor family

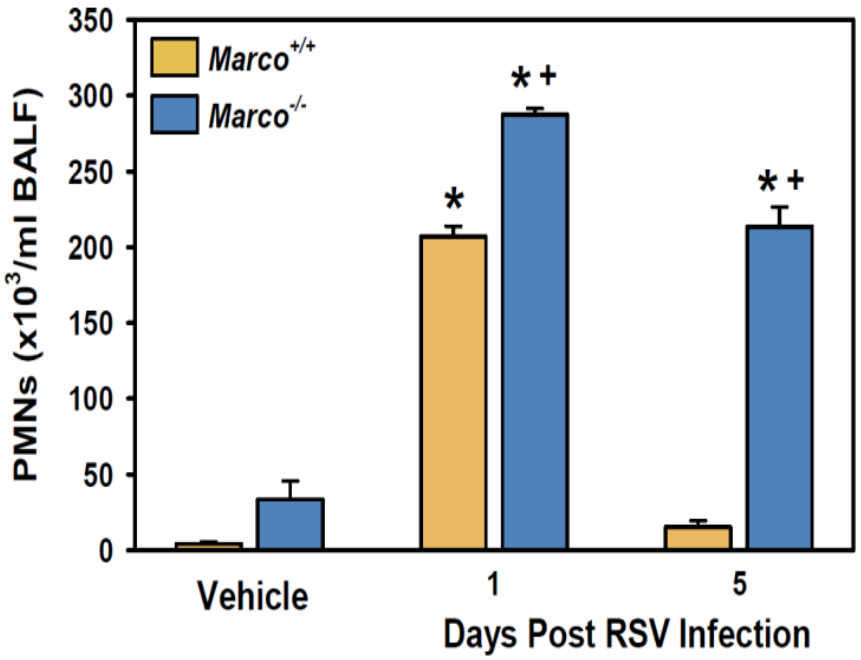




J.E. Murphy et al. / Atherosclerosis 182 (2005) 1–15

Targeted deletion of *Marco* enhances lung inflammation and injury after RSV infection





National Institute of Environmental Health Sciences

High et al, *Ebiomedicine* 2016

What is the role of *MARCO* in human RSV disease severity?

Infant Foundation - Buenos Aires, Argentina

The focus of the Infant Foundation basic and clinical research is centered around children's respiratory diseases.

INFANT Director is Dr. Fernando Polack (Monroe Carell Jr. Children's Hospital, Vanderbilt).

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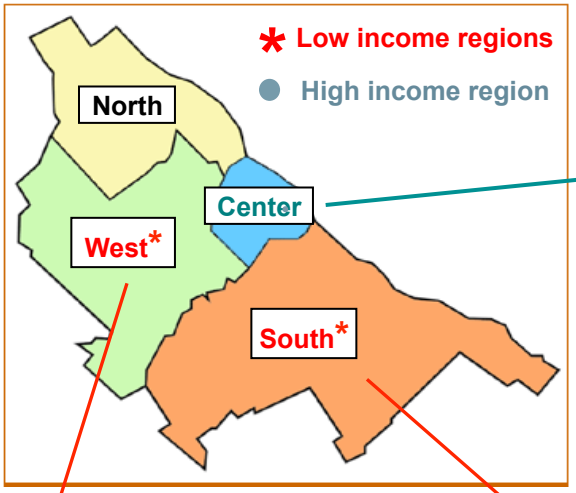
http://www.infant.org.ar

Dr. Fernando Polack

Map of Buenos Aires and its suburbs. The three regions associated with the participating hospitals are displayed along with illustrative pictures



Area of influence of French Hospital

Area of influence of Posadas Hospital

Area of influence of Berazategui Hospital

Clinical outcomes

Primary:

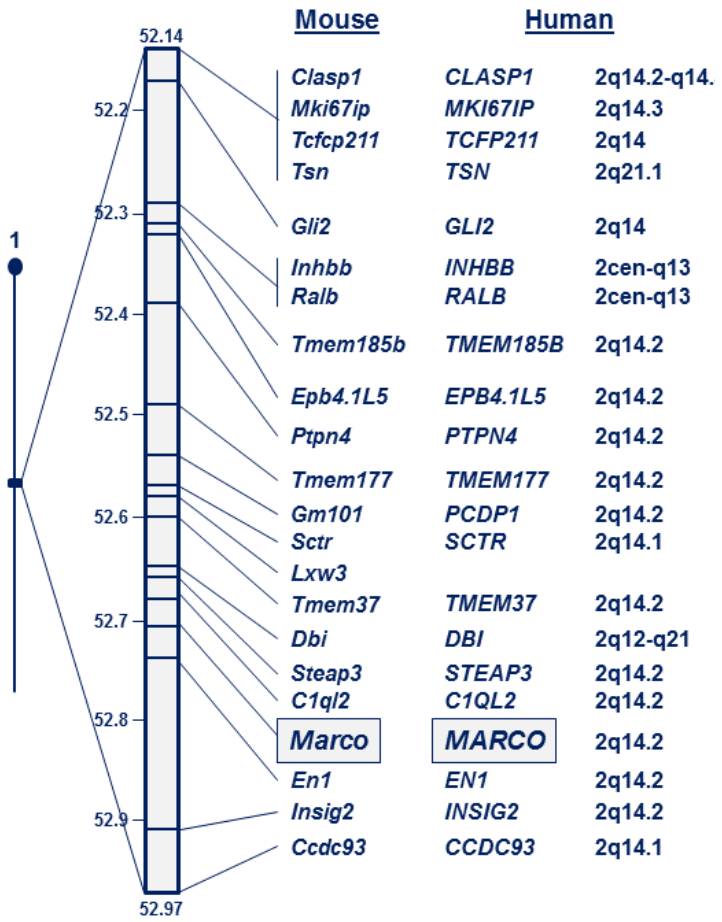
- Severity of disease. Several clinical scores that are hard to validate or impossible to use (i.e.: $pCO_2 > 45$; $O_2 < 87\%$)

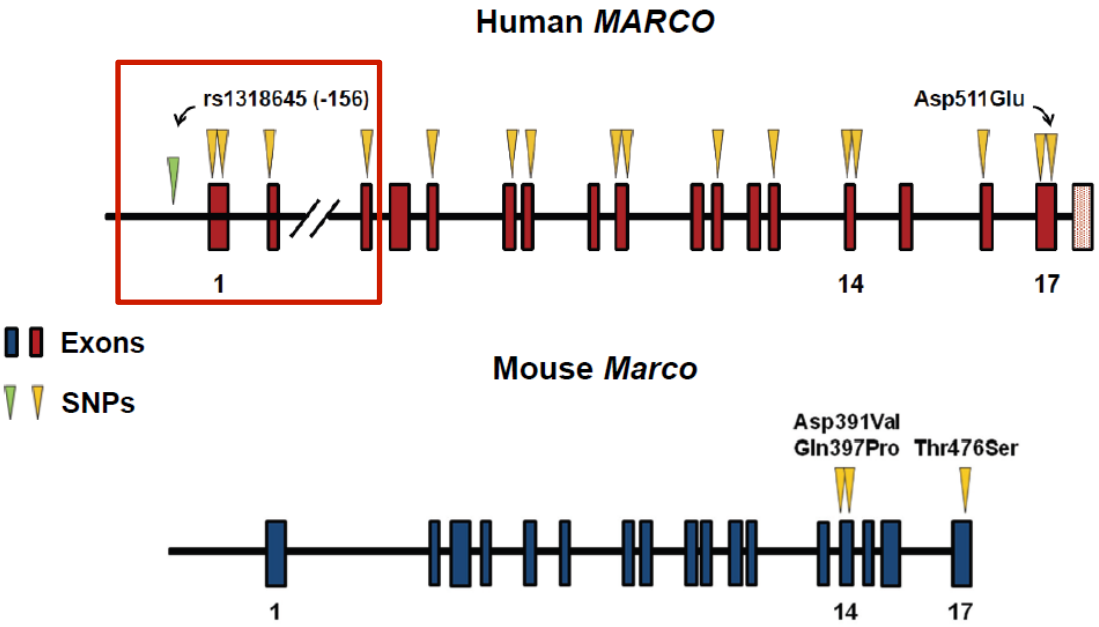
Oxygen saturation <93%

Secondary:

- RSV titer (Guy et al, *J Infect Dis*, 2004)
- Th2 polarization (Choi et al, *J Infect Dis*, 2002)

Human and mouse comparative homology for *Marco*

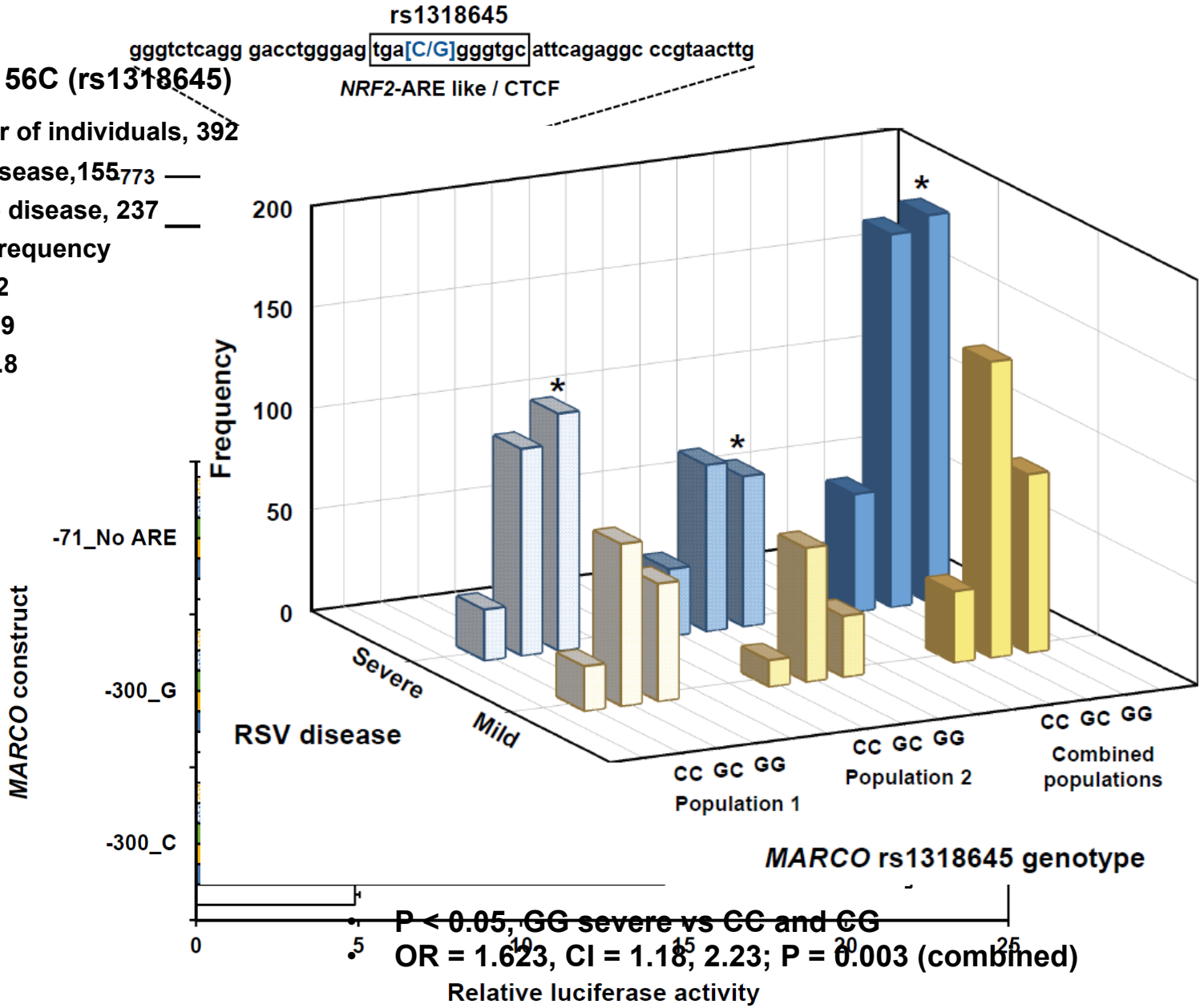




Functional evaluation of the *MARCO* rs1318645 polymorphism

***MARCO* T-156C (rs1318645)**

- Total number of individuals, 392
  - Mild disease,155
  - Severe disease, 237
- Genotype frequency
  - TT, 24.2
  - TC, 45.9
  - CC, 29.8

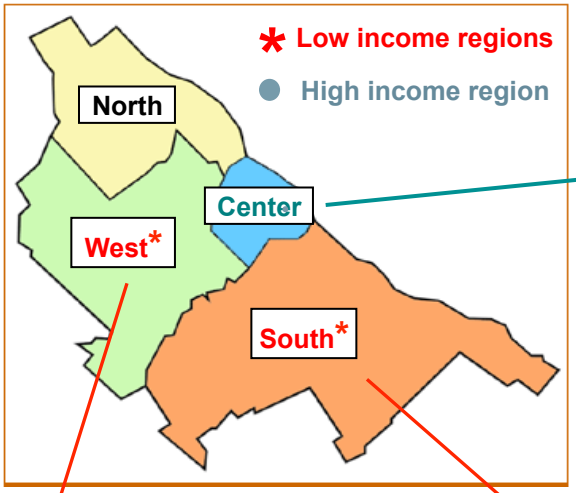


P < 0.05, GG severe vs CC and CG

OR = 1.623, CI = 1.18, 2.23; P = 0.003 (combined)

Study 2: A candidate gene approach - what is the role of interaction between toll-like receptor 4 (*TLR4)* and environmental LPS in human RSV disease severity?

Map of Buenos Aires and its suburbs. The three regions associated with the participating hospitals are displayed along with illustrative pictures



Area of influence of French Hospital

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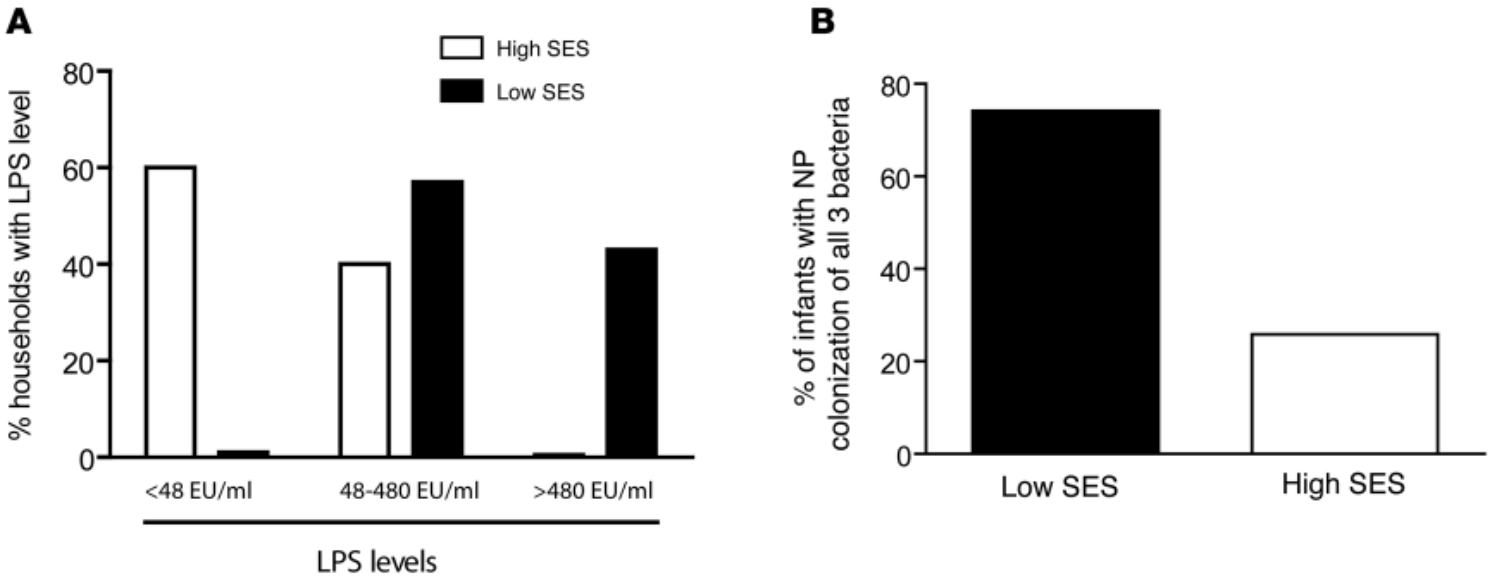
Percent indicators of socioeconomic differences between the hospital-associated regions

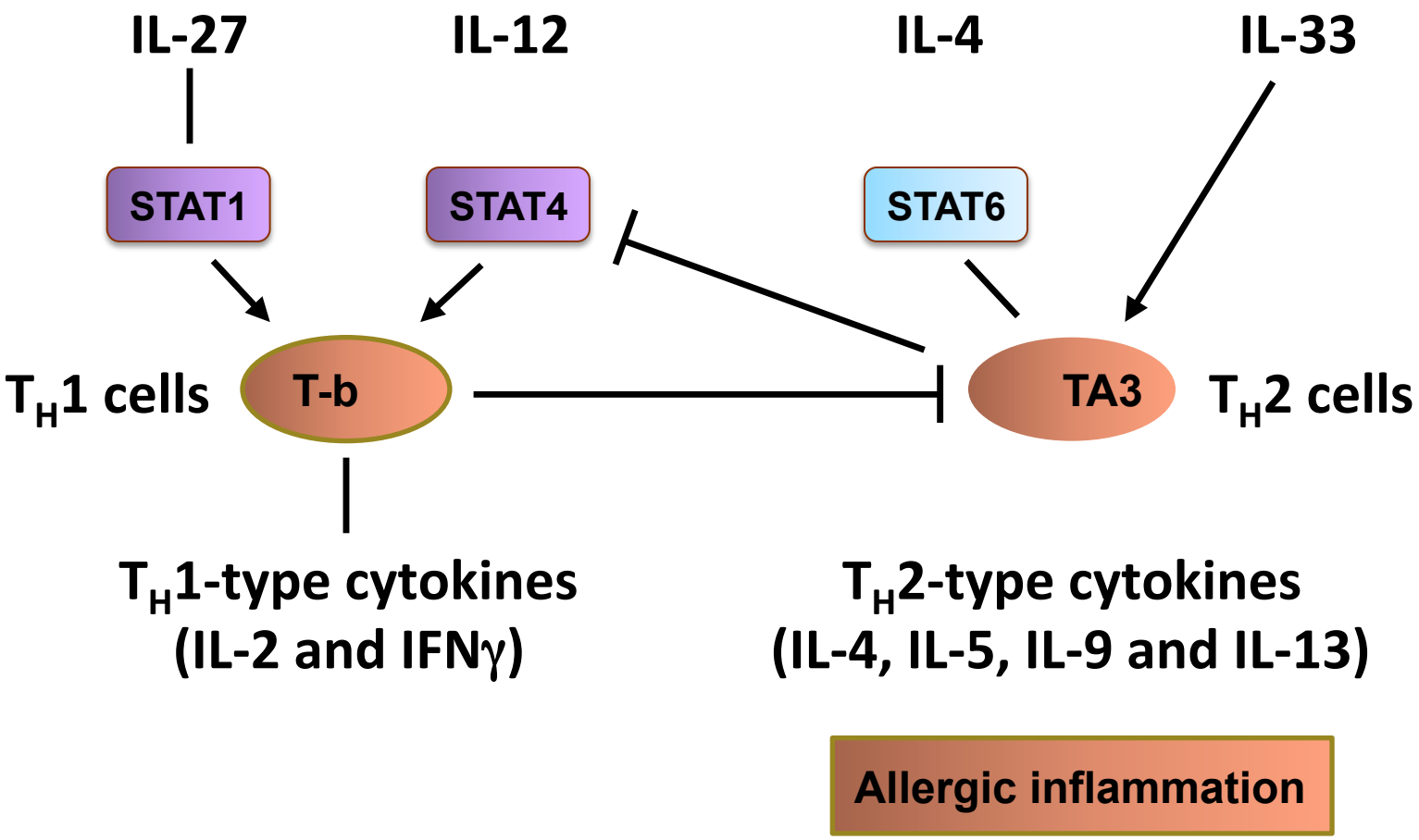
	WEST & SOUTH	CENTER
Household below poverty line	22	8
Illiteracy in older than 20 y	15	5
No private medical insurance	65	36
Well-water	42	2
No sewer	37	3

Evidence for gene-environment interaction and RSV disease severity in infants



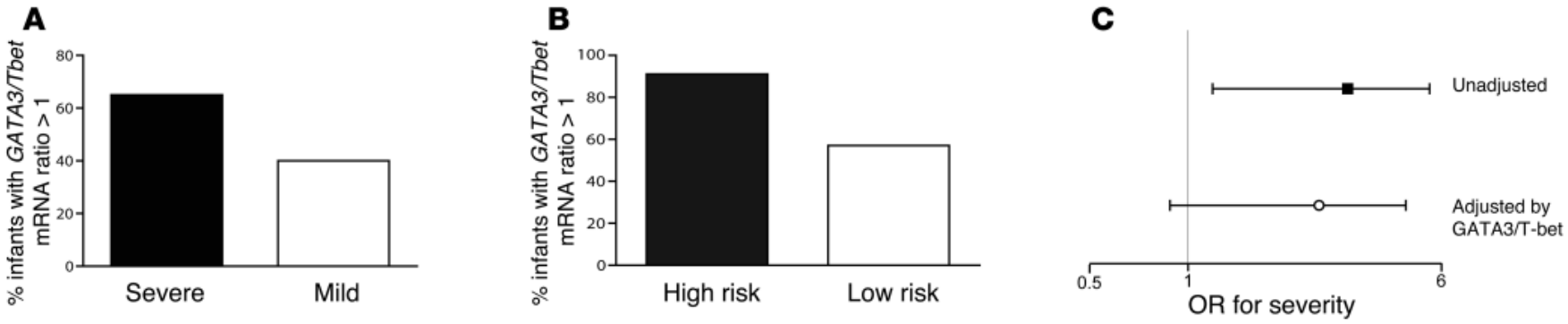
Caballero et al, TLR4 genotype and environmental LPS mediate RSV bronchiolitis through Th2 polarization. *J Clin Invest*, 2015

Interactions between TH1 and TH2 cells in asthma and other allergic diseases



Modified: Barnes, *Nature Reviews Immunology*, 2008

Th2 bias and RSV bronchiolitis in infants with different LPS exposure and socioeconomic status



Caballero et al, *JCI* 2015

What is the public health relevance of these models?

- **Annual global RSV disease burden is over 30 million new acute lower respiratory infection episodes in children under five (WHO).**
- **Severe RSV disease in infancy has also been associated with diseases of childhood and adulthood (e.g. asthma).**
- **A diagnostic panel of genetic SNPs could be designed that would also incorporate environmental exposure status (gene x environment interaction) that may be used to predict disease severity.**
- **Intervention strategies could be put in place for at risk individuals, and thus reduce disease burden.**

ACKNOWLEDGMENTS

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