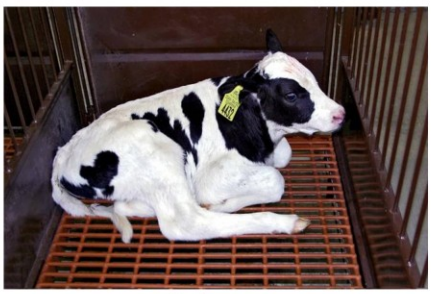
Comparative Immunology of Tuberculosis

Ray Waters, DVM, PhD
Tuberculosis Research Project
National Animal Disease Center
Ames, Iowa



USDA United States Department of Agriculture ARS Agricultural Research Service NADC National Animal Disease Center

Immune Response - Relevance

1. Diagnostics: Is the animal infected?

2. Vaccination: Did the vaccine elicit a response and is this response protective?

3. Correlations to:

- **Pathology**: response after infection compared to disease
- **Protection**: response after vaccination compared to efficacy
- **Infection**: Active vs latent? Progressive, resolving, or cured?

Robert Koch, 1890 - develops tuberculin

Immune Response – *One Size Does Not Fit All!*

Host Factors:

• Poorly organized vs organized granulomas, badgers vs cattle

• Multibacillary vs paucibacillary disease, mice vs humans/cattle

Pathogen Factors:

• *M. bovis*, *M. tuberculosis*, *M. africanum*, *M. caprae*, *M. pinnipedii*, *M. mungi*, *M. microti*, dassie bacillus, etc

• Hopkins Model - Cavitation in rabbits upon *M. tb* challenge requires prior sensitization to heat killed *M. bovis*

Host / Pathogen Interactions:

• Host adaptation led to speciation

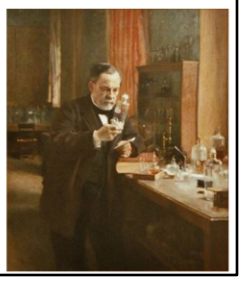
• Speciation partially defined by transmission capacity

Environmental Factors:

• *M. mungi* – prevalence dramatically increases with dry season / Badgers and underground environment

• Nutrition, exposure, over-population, stress, etc.

Louis Pasteur - 1860's, germ theory of tuberculosis & pasteurization



Humans

IFN-γ: humans with defective receptors develop severe disease

TNF-α: essential for proper granuloma formation – TNF inhibitors used for Crohns Disease, rheumatoid arthritis, etc. promote transition from latent to active TB

CD4+ cells: HIV patients more susceptible

IL-12: humans with defective IL12p40 subunit or receptors develop severe disease

Antibody: associated with active disease; sensitivity associated with smear positive cases which correlate with pathology – likely due to antigen load

Latent disease common – latency not documented in other species! Generally only CMI (undetectable antibody) with latent disease.

Chemokines, Cathelicidins, CD8+ cells, IL-17, etc.

Emil von Behring
Nobel Prize, 1901
Also, *M. tb* to cattle

"I need hardly add that the fight against cattle tuberculosis only marks a stage on the road which leads finally to the effective protection of human beings against the disease."

Laboratory Models of TB

Mice: multibacillary, **high mycobacterial burden** with progressive, less-organized pathology and more neutrophils than many other species

IFN-γ > NO > $\alpha\beta$ TCR > MHC II > MHC I > WT and $\gamma\delta$ TCR **- Th1 immunity crucial**

Kinetics of Infection: **2 wk delay** in activation of T cells, optimal antigen presentation in lymph nodes draining the lungs, then migration of specific T cells back to the lungs

Apoptosis of infected macrophages critical for efficient priming of T cells via uptake of apoptotic vesicles and antigen presentation by dendritic cells **-** *M. tb* complex mycobacteria actively inhibit apoptosis

Guinea Pigs, Rabbits: pathology similar to humans, models for evaluating DTH and cellular immunity to TB

Skin test in Mice – sensitivity to tuberculin 1000X lower than humans (***differences exist in application of diagnostic tests***)

Hosts of Veterinary Significance

Cattle: DTH, IFN-γ (Bovigam), poor antibody response,

Cervids: DTH (? Accuracy), moderate antibody response

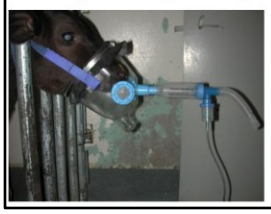
Eurasian Badgers: Poor DTH response, antibody to MPB83 correlates w/bacterial load & ability to transmit, pathology – poor granuloma, cell-mediated immunity less vigorous yet good enough (low EC response)

Elephants: Robust antibody response, poor DTH (difficult to apply)

South American Camelids: antibody response, poor DTH

Wildboar, Spain: Oral heat-killed *M. bovis* vaccine is effective?

Antigen recognition profiles vary for different species



Correlates of Protection (the Holy Grail)

DTH (skin test) is indicative of prior exposure to *Mycobacteria* spp. but is NOT indicative of disease severity or protection elicited by vaccination

IFN-γ responses, especially to specific antigens such as ESAT-6/CFP10, are indicative of infection but do not necessarily correlate to protection elicited by vaccination

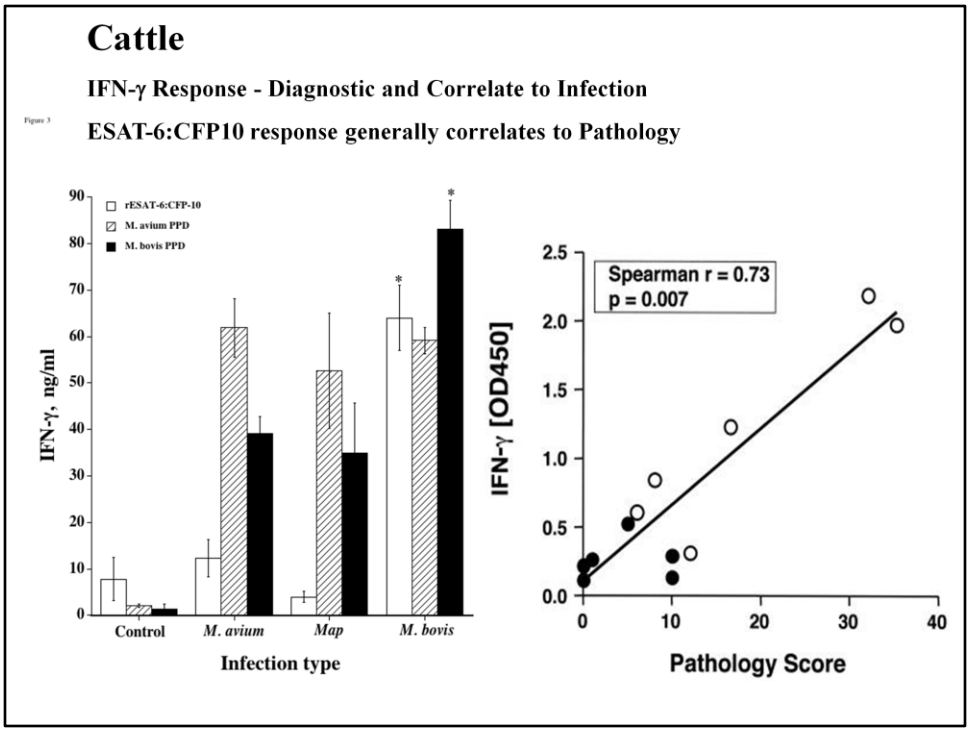
Central Memory Responses (TcM) – correlate to reduced bacterial burden and reduced pathology

IL-17 – correlate to pathology, pre-challenge responses may also correlate to protection.

Multi-functional T cells - IFN-γ TNF-α, IL-2

Patterns of response (multi cytokine / chemokine / etc. profile) – outcome of RNA sequencing studies

Leonard Pearson 1892 - 1st injection of tuberculin to US cattle for TB test



M avium/Map – PPDa > PPDb, no EC
M. Bovis PPDb > PPDa, +EC
EC response correlates to pathology

Cattle

The IFN-γ Response to PPDb does not always Correlate to Pathology; however, it is a good Correlate to Infection

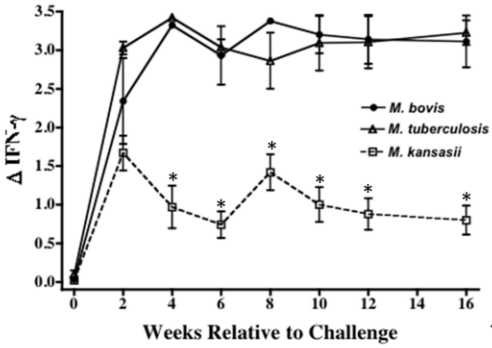
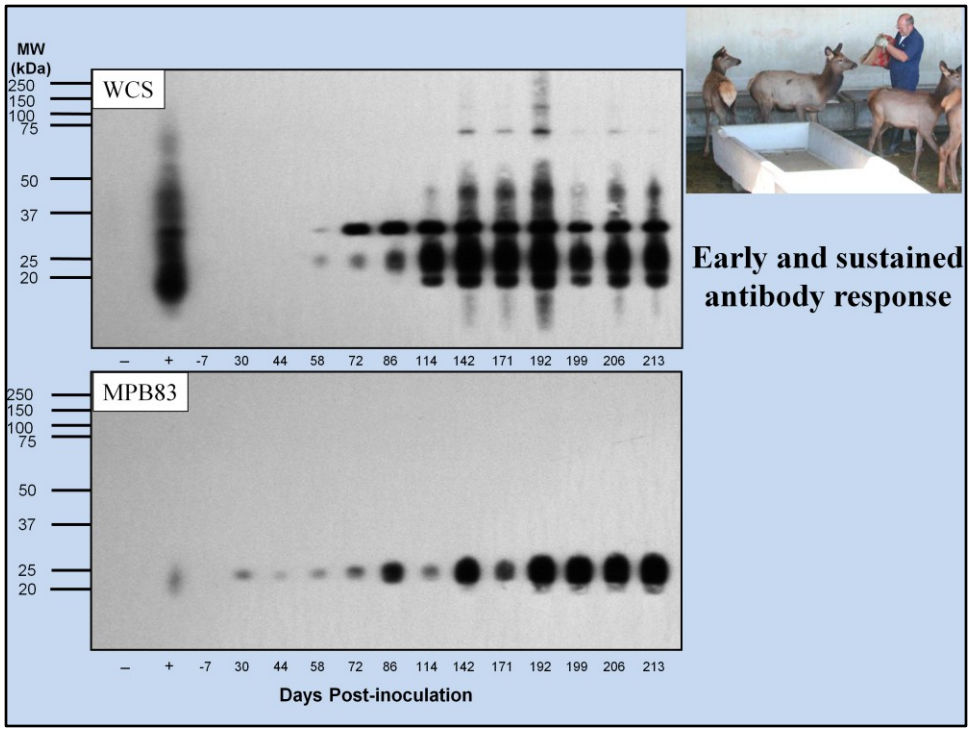
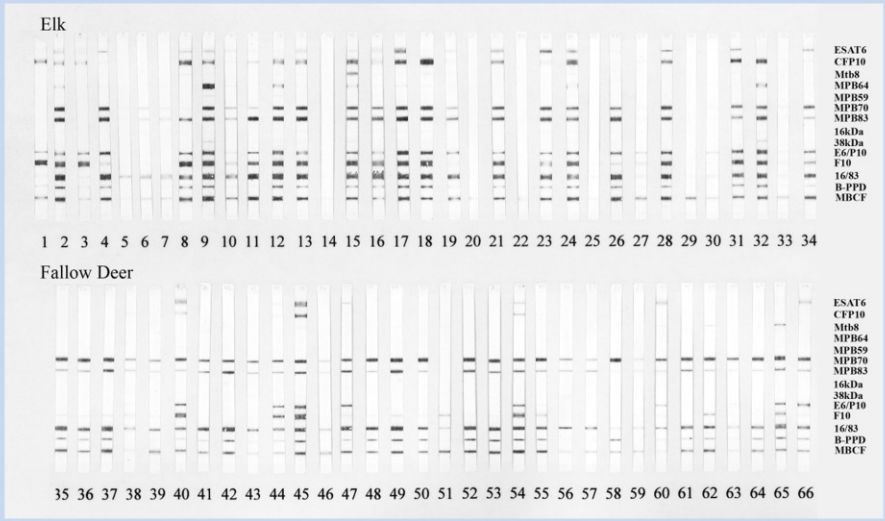


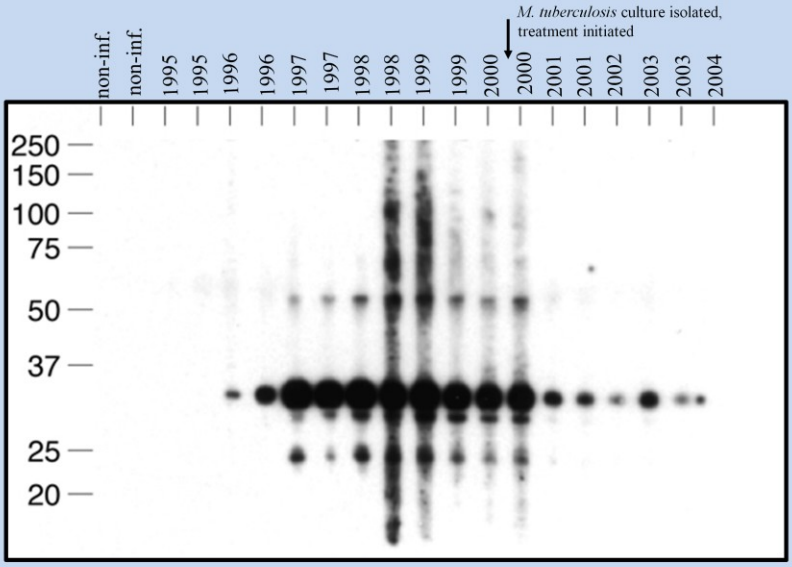
Table 1. Disease expression upon mycobacterial inoculation.

Group	Gross Pathology[a]	Culture[b] *
M. bovis	All positive	27.2 ± 7.3
(n = 5)		
M. tuberculosis	All negative	13.9 ± 5.5
(n = 5)		
M. kansasii	All negative	0 ± 0
(n = 4)		


MW
(kDa)
250
150
100
75
50
37
25
20
WCS
– + -7 30 44 58 72 86 114 142 171 192 199 206 213
MPB83
250
150
100
75
50
37
25
20
– + -7 30 44 58 72 86 114 142 171 192 199 206 213
Days Post-inoculation
Early and sustained antibody response


Elk
ESAT6
CFP10
Mtb8
MPB64
MPB59
MPB70
MPB83
16kDa
38kDa
E6/P10
F10
16/83
B-PPD
MBCF
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34
Fallow Deer
ESAT6
CFP10
Mtb8
MPB64
MPB59
MPB70
MPB83
16kDa
38kDa
E6/P10
F10
16/83
B-PPD
MBCF
35 36 37 38 39 40 41 42 43 44 45 46 47 48 49 50 51 52 53 54 55 56 57 58 59 60 61 62 63 64 65 66

Immunoblot


M. tuberculosis culture isolated, treatment initiated
non-inf.
non-inf.
1995
1995
1996
1996
1997
1997
1998
1998
1999
1999
2000
2000
2001
2001
2002
2003
2003
2004
250
150
100
75
50
37
25
20

Conclusions: Comparative Immunology to TB

- **Similar, yet different, immune responses between hosts / pathogens**

- **Opportunities:**
  1. TcM
  2. Specific antibody
  3. Cytokine profile comparisons
  4. Host / pathogen comparisons
  5. Interactions
  6. Field Application

- **In regards to the immune response to TB, generalizations and extrapolations between hosts is risky!!!**