Comparative pathology of tuberculosis following infection from mice to men.

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Milestones...

- 125th anniversary of the discovery of TB bacillus
- Decades ago, TB started disappearing, thanks to antibiotic era.
- Promised to eliminate it from the globe by the turn of the 20th century.
Today...

• But today, TB still remains a major disease afflicting all of mankind

• New forms of TB have appeared in many developed countries.

• And in developing nations it is spreading even faster, with two million people dying every year from the disease.
Two reasons

• Emergence of drug resistance
  – MDR and XDR TB

• HIV/AIDS epidemic
Tuberculosis: Medical Need

- "Worldwide Pandemic" - WHO
- Kills 5000 persons every day
- Incidence increasing by 1% / yr
- Leading cause of Death in HIV
- Drug Resistance in all countries
- 5% refractory to present therapy
- No new drug in the last 40 years
Distribution of TB Cases

GLOBAL EPIDEMIOLOGY OF TUBERCULOSIS

per 100,000 pop

- < 25
- 25 - 49
- 50 - 99
- 100 - 299
- 300 or more
- No estimate

© WHO 2004
Prevalence and Deaths

By 2015 (worldwide):
10 million cases
1 million deaths


AFR- Africa
AMR- America
EMR- Eastern Mediterranean
EUR- Europe
SEAR- South East Asia
WPR- Western Pacific
MDG- Millennium goal

STPI-Oct 23-25, 2008
Complexities of tuberculosis

- Slow growing pathogen
- Granulomatous inflammation.
- Latency. Ability of the host to contain the organism.
- Spectrum of histological changes—varies with species.
- Immunological status of individual.
Complexity in TB Treatment

Why 6 month Rx with 4 drugs-HRZE

Initial Phase [2 months]
Isoniazid, Rifampicin, Ethambutol, Pyrazinamide

Relapse ~ 70%
Relapse ~ 40%
Relapse ~ 6%

Continuation Phase [4 months]
Isoniazid, Rifampicin

>90% cured

Persisters

Time (months)

STPI-Oct 23-25, 2008
First-Line Treatment of Tuberculosis (TB) for Drug-Sensitive TB

- **Isoniazid (1952)**
  - Inhibits cell wall synthesis

- **Ethambutol (1961)**
  - Inhibits cell wall synthesis

- **Pyrazinamide (1952)**
  - Exact Target Unclear
  - Disrupts Plasma Membrane
  - Disrupts Energy Metabolism

- **Rifampin (1966)**
  - Inhibits RNA synthesis

**Mycobacterium tuberculosis**

- **Cell Wall Synthesis**
  - Acyl Lipids
  - Mycolic Acid
  - Arabinogalactan
  - Peptidoglycan
  - Plasma Membrane

- **DNA Coiling, Transcription, and Translation**
  - DNA Polymerase
  - DNA Gyrase
  - mRNA
  - Ribosome
  - Protein

- **ATP Synthesis**
  - Cell Wall
New Tuberculosis (TB) Drugs Under Development

**Cell Wall Synthesis**
- Acyl Lipids
- Mycolic Acid
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- Plasma Membrane

**DNA Coiling, Transcription, and Translation**
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**Mycobacterium tuberculosis**

**SQ-109***
- Inhibits cell wall synthesis

**Nitroimidazoles** (E.g. PA-824*, OPC 67683)
- Novel, complex mechanisms of action
- Inhibit cell wall synthesis AND inhibit cell respiration

**Macrolides**
- Inhibit protein synthesis

**Oxazolidinones**
- Inhibit protein synthesis

**Pyrroles**
- Mechanism of action uncertain

**ATP Synthesis**
- Diarylquinoline (TMC 207)
- Inhibits ATP synthase

*STPI-C*
Variations in histopathology

- Nonnecrotizing granuloma which is dominated by the epitheloid histiocytes.
- Suppurative granuloma with a necrotic centre.
- Fibrotic/sclerotic granuloma with concentric fibrosis with minimal inflammatory cells.
- Calcified granuloma with a few cells at the periphery.
- Tuberculous pneumonia characterised by filling the airspaces by inflammatory cells.
Mouse lung granuloma

BALB/c mouse infected with *M. tuberculosis* aerosol infection. 8 wks. Post infection
Granuloma from BALB/c mouse. 8 weeks post infection. Nonnecrotising granuloma, predominantly lymphocytic.
Granuloma from BALB/c mouse with calcification and cholesterol clefts. 21 weeks post infection.
Granuloma from BALB/c mouse 21 weeks post infection. Aggregation of epithelioid histiocytes. Foamy macrophages.
BALB/c mouse infected with *M. tuberculosis* H37Rv

Predominantly lymphocytes, epitheloid and foamy macrophages

8 wks PI. H&E

1000X

8 wks PI. ZN

STPI-Oct 23-25, 2008
BALB/c mouse infected with *M. tuberculosis* H37Rv

34 wks PI- Trichrome staining

Scanty connective tissue interspersed within the granuloma. Foamy macrophages and epitheloid cells are present.
8 wks post infection. Consisting of Infiltration of predominantly lymphocytes and macrophages and no necrosis.

Randall J. Basaraba, Tuberculosis, 2008
IFN-G KO mouse showing extensive lesion with infiltration of neutrophils

Randall J. Basaraba, Experimental tuberculosis, Tuberculosis, 2008, 88, suppl. 1, S35-S47
Lungs from *M. tuberculosis* infected G.pigs. Primary granuloma showing Dystrophic calcification.

Lenearts et al, AAC, 2007
G. pig lung infected with *M. tuberculosis*

dystrophic calcification at the centre and delineated by acellular rim that blends with fibrous capsule which contains predominantly Lymphocytes and fewer macrophages.

Lenearts et al, AAC, 2007
Cotton rats infected with *M. tuberculosis*

Immunologically naïve cotton rats show - necrotic lesion in 30 days.

Randall J. Basaraba, Tuberculosis, 2008
Non-human primate (Rhesus macaque, *Macaca mulatta*) -

Form a large cavity as a result of lung tissue necrosis. Liquefactive necrosis

Rabbit infected with *M. tuberculosis* H37Rv

5 wks post infection

10 wks post infection

15 wks: caseous center has been replaced by epitheloid macrophages

15 wks: Intraalveolar plug of Epitheloid macrophages- after Immunosupression.

Manabe et al. Tuberculosis: 2008
Rabbit lungs - cavitary lesion

18 wks post infection - high dose aerosol infection

Paul J. Converse et al., Infection and Immunity, 1996
Cavitary lesion

- Seen in human tuberculosis.
- Rabbit and non-human primates develop this lesion, however there is a difference in pathogenesis.
  - Rabbits develop cavitary lesion as apart of primary lesion.
  - In humans, the cavitary lesions typify post primary or reactivation TB that can develop decades after initial infection.
Human Lesions

Human TB case- Nonnecrotic granuloma with Giant Cell (GC)
Surrounded by lymphocytic infiltration.

Human granuloma with necrotic center
Detection of Mtb by ZN technique.
Some acid fast bacilli are not detected by ZN stain
(are they in a different state- cell wall deficient state?)

Summary

• TB is not a single disease entity
• Multiplicity of physiological states of the bacilli
  - Extracellular, Intracellular, Hypoxic etc
• Multiplicity of pathophysiological conditions in the lesions
  - Necrotic, Nonnecrotic, fibrous, calcification and cavitary lesions etc.
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