

**Manage TB**  
**Dr. Subash Babu**  
**National Institute for Research in Tuberculosis, Chennai**


**Lecture – 05**  
**Pathogenesis of Tuberculosis**  
**Session 02**

Welcome to the second session on the Pathogenesis of Tuberculosis.

(Refer Slide Time: 00:17)

**Reactivation of TB**

- Reactivation of latent TB reflects progression to active, symptomatic disease
- Reactivation TB must be distinguished from re-infection with a second strain of bacteria
- Most cases of TB in adults are attributable to reactivation
- Reactivation TB is widely attributed to 'weakened' immunity, minority of cases are attributable to well-characterized defects in immunity




Manage TB  
ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

In this session we are going to mainly look at factors that impinge on the reactivation of TB. So, reactivation of latent TB pretty much reflects progression to active symptomatic disease from latent TB. And it must be distinguished from reinfection, which is usually with the second strain of bacteria.

Most cases of TB in adults are attributable to reactivation while reactivation is widely attributed to the so called weakened immunity, only a minority of cases are directly attributable to well characterized defects in immune responses.

(Refer Slide Time: 00:50)



Manage TB

### Mechanisms underlying Reactivation of TB

- Quantitative and qualitative CD4<sup>+</sup> T cell defects - occur in people with HIV infection
- Therapeutic neutralization of TNF, especially by monoclonal antibodies
- The effects of TNF blockade that account for reactivation TB
  - ✓ decreased macrophage-mediated anti-mycobacterial activity and the subsequent death of macrophages
  - ✓ induction of a higher frequency of regulatory T cells
  - ✓ depletion of a subset of CD45RA<sup>+</sup> effector memory CD8<sup>+</sup> T cells that contain granulysin

ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

So, when we look at the mechanisms that underlie reactivation of tuberculosis, the commonest a mechanism that underlies reactivation of TB is the quantitative and qualitative T cell defects that occur in people with HIV infection, and we also know that therapeutic neutralization of TNF alpha especially by monoclonal antibodies, can also cause reactivation.


And the effects of TNF alpha blockade that account for reactivation TB include decreased macrophage mediated antimicrobial micro bacterial activity and subsequent death of macrophages, induction of a higher frequency of regulatory T cells, depletion of a subset of effector memory T cells that contain granulysin etcetera.

(Refer Slide Time: 01:39)

Manage TB  
ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

## HIV and TB coinfection

- HIV-1 infects CD4<sup>+</sup> T cells and macrophages
- *M. tb* primarily infects macrophages, which require CD4<sup>+</sup> T cells to augment intracellular clearance of microbial pathogens
- Depletion of CD4<sup>+</sup> T cells that is associated with HIV-1 infection is thought to have a major role in the increased risk of TB in individuals infected with HIV-1



Coming to HIV and TB coinfection, HIV one in fact, CD 4 T cells as well as macrophages while *M. tb* primarily infects macrophages which require CD 4 positive T cells, to augment the clearance of microbial pathogens.


Therefore, the definition of CD 4 positive T cells that is associated with HIV infection is thought to have a major role in the increased risk of TB in individuals with HIV.

(Refer Slide Time: 02:07)

Manage TB  
ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

## HIV and TB coinfection

- HIV targets and depletes *M. tuberculosis* antigen-specific CD4<sup>+</sup> T cells at a greater frequency than CD4<sup>+</sup> T cells specific for other antigens
- Progressive HIV-1 disease and TB are both characterized by chronic inflammation driven by the failure to clear either pathogen
- The chronic nature of these responses may undermine host protection by promoting an immunoregulatory phenotype that is characterized by attenuated T cell responses



Moreover HIV targets and depletes *M. tb* antigen specific T-cells at a much greater frequency, than CD 4 T cells specific for other antigens. And finally, both progressive

HIV disease and TB are characterized by chronic inflammation driven by the failure to clear either pathogen and therefore, the chronic nature of these responses may undermine host protection by promoting an immunoregulatory phenotype that is characterized by attenuated T cell responses.


(Refer Slide Time: 02:42)

Manage TB

ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

## Reactivation of TB - Common causes

- Diabetes
- Malnutrition
- Smoking
- Alcohol consumption
- Treatment with glucocorticoids
- Silicosis
- Hematological malignancies
- Uremia
- Indoor air pollution



So, in terms of the other common causes of reactivation TB; diabetes, malnutrition, smoking, alcohol consumption, treatment with glucocorticoids silicosis hematological malignancies uremia and indoor air pollution are some.


(Refer Slide Time: 03:01)

Manage TB

ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

## Diabetes and TB

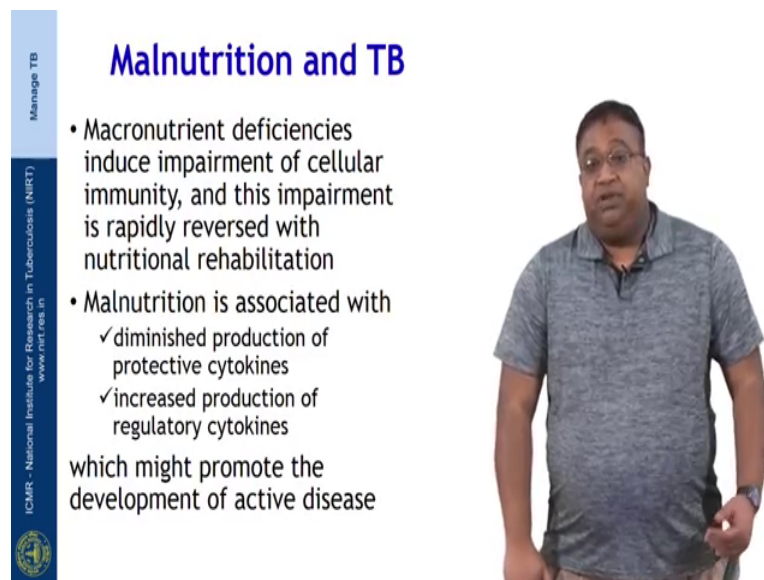
- In addition to increasing the risk to develop active TB, DM increases TB disease severity
- Diabetes is associated with
  - ✓ diminished Th1 and Th17 responses in latent TB, which might confer increased susceptibility to development of disease
  - ✓ exaggerated pro-inflammatory response in active disease, which might worsen TB pathology



So, let us look at a few of those in a little more detail in terms of the immune response and pathogenesis. So, we know that diabetes is commonly associated with an increased risk for TB. In addition to increasing the risk to develop active TB, diabetes mellitus also increases TB disease severity.

And diabetes is typically thought to be associated or shown to be associated with diminished Th 1 and Th 17 responses in latent TB infection, which might confer increased susceptibility to development of disease. And diabetes is also associated with exaggerated pro inflammatory responses in active disease, which might actually worsen TB pathology in those with pulmonary tuberculosis.

(Refer Slide Time: 03:47)



The slide features a vertical blue bar on the left with the text 'Manage TB' at the top, 'ICMR - National Institute for Research in Tuberculosis (NIRT)' in the middle, and 'www.nirt.res.in' at the bottom. The main title 'Malnutrition and TB' is in blue. The content includes a bulleted list and a concluding sentence. To the right of the text is a photograph of a man in a grey polo shirt speaking.

**Malnutrition and TB**

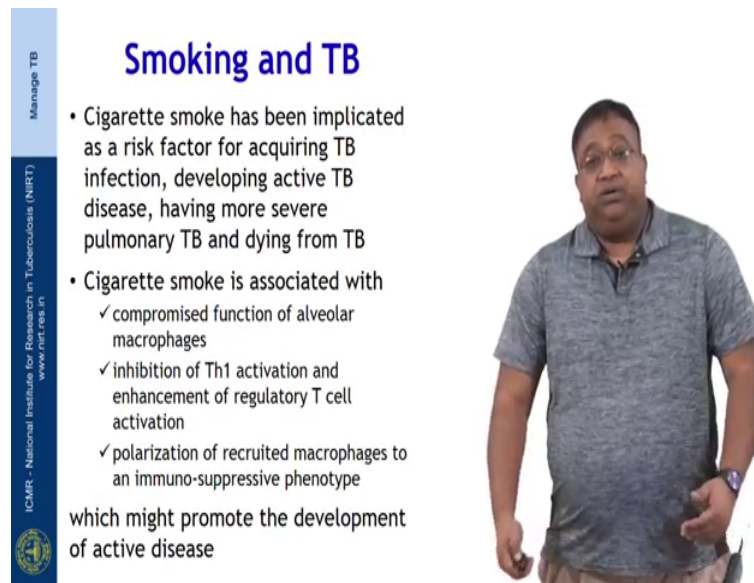
- Macronutrient deficiencies induce impairment of cellular immunity, and this impairment is rapidly reversed with nutritional rehabilitation
- Malnutrition is associated with
  - ✓ diminished production of protective cytokines
  - ✓ increased production of regulatory cytokines

which might promote the development of active disease

And we look at malnutrition, micronutrient deficiencies are known to it cause impairment of cellular immunity and this impairment can be rapidly reversed with nutritional rehabilitation.

Moreover malnutrition is also associated with diminished production of protective cytokines, an increased production of regulatory cytokines, both of which might contribute to the promotion of the development of active disease.

(Refer Slide Time: 04:16)



The slide features a vertical blue bar on the left with the text 'Manage TB' at the top, 'ICMR - National Institute for Research in Tuberculosis (NIRT)' in the middle, and the website 'www.nirt.res.in' at the bottom. The main title 'Smoking and TB' is in blue. The content includes two main bullet points with sub-points, and a concluding sentence. A man in a grey polo shirt is standing on the right side of the slide.

## Smoking and TB

- Cigarette smoke has been implicated as a risk factor for acquiring TB infection, developing active TB disease, having more severe pulmonary TB and dying from TB
- Cigarette smoke is associated with
  - ✓ compromised function of alveolar macrophages
  - ✓ inhibition of Th1 activation and enhancement of regulatory T cell activation
  - ✓ polarization of recruited macrophages to an immuno-suppressive phenotype

which might promote the development of active disease

The other factor which is of importance in TB reactivation is smoking; cigarette smoke has been implicated as a risk factor for acquiring TB infection, for developing active TB disease, for having more severe pulmonary TB and also for dying from TB

We know that cigarette smoke is associated with a compromised function of alveolar macrophages, it is associated with inhibition of Th 1 activation and enhancement of regulatory T cell activation. And also is associated with the polarization of recruited macrophages to an immunosuppressive phenotype. So, all of the above basically contribute to the promotion of development of active disease in the case of smoking and tuberculosis.



(Refer Slide Time: 05:05)

Manage TB


ICMR - National Institute for Research in Tuberculosis (NIRT)

www.nirt.res.in

## Alcohol and TB

- Alcohol consumption, especially heavy consumption is estimated to increase the risk of incident TB and death due to TB
- Alcohol consumption is associated with
  - ✓ suppressed mobilization and adherence of macrophages
  - ✓ suppressed phagocytosis of macrophages
  - ✓ antigen - specific T cell activation

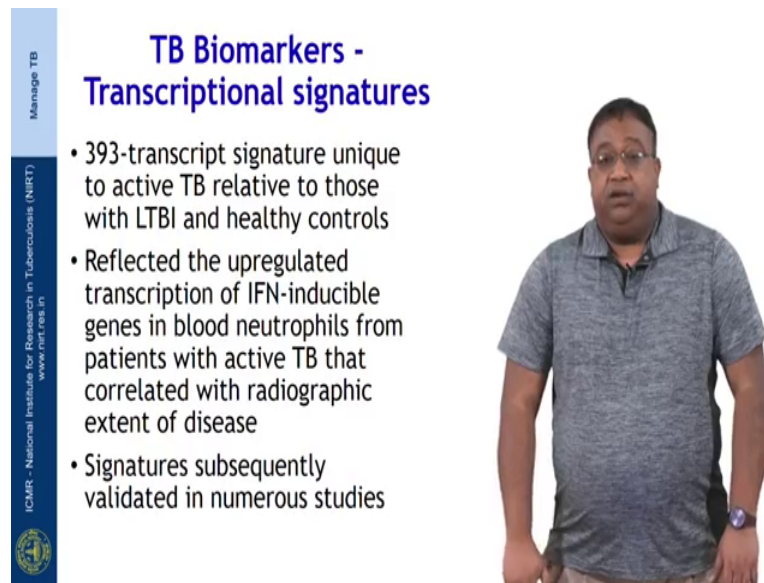
which might promote the development of active disease



Finally, if we look at alcohol, we also know that alcohol consumption especially heavy consumption is estimated to increase the risk of incident TB as well as the risk of death due to tuberculosis.

And this is typically thought to be associated again with the impairment of certain immune responses and these include suppressed mobilization and adherence of macrophages, suppress phagocytosis of macrophages and diminished antigen specific T cell activation, all of which might contribute to the development of active disease as well.

(Refer Slide Time: 05:40)




**TB Biomarkers -  
Transcriptional signatures**

- 393-transcript signature unique to active TB relative to those with LTBI and healthy controls
- Reflected the upregulated transcription of IFN-inducible genes in blood neutrophils from patients with active TB that correlated with radiographic extent of disease
- Signatures subsequently validated in numerous studies

ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

Manage TB



So, switching gears we are now going to look at TB biomarkers, which are basically going to be discussed under three modules, transcriptional signatures, correlates of risk and immune biomarkers. And we are interested in biomarkers in TB mainly because of the fact that the diagnosis and prognosis of TB is a very difficult phenomenon and TB biomarkers could aid in both diagnostic purposes as well as for therapeutic monitoring.

So, in terms of therapeutic the transcriptional signatures, it was initially discovered that there was a 393 transcript signature, which was unique to active TB and help distinguish those with active TB from those with LTBI and healthy controls.

The signature basically reflected an up regulated transcription of interferon inducible genes mainly, in the blood neutrophils from patients with active TB, and it turned out that the signature actually correlated with the extent of radiographic disease as well.




(Refer Slide Time: 06:44)

Manage TB  
ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

## TB Biomarkers - Transcriptional signatures

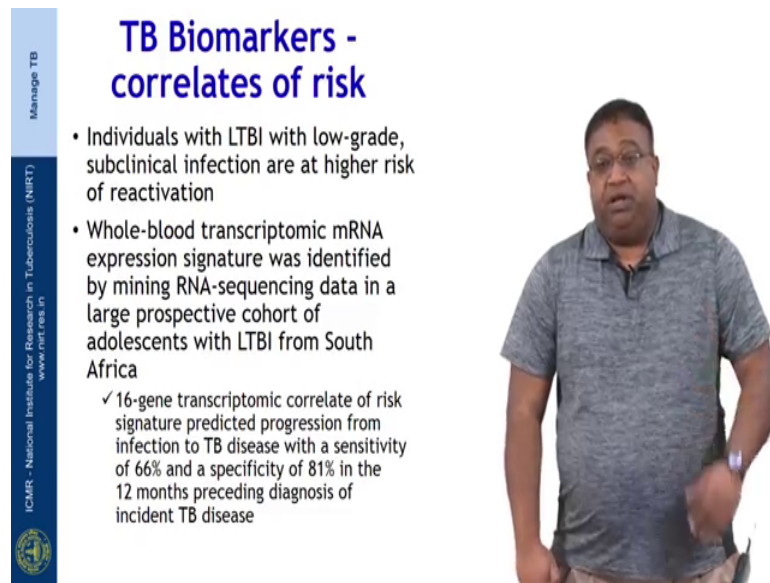
- Blood gene-expression profiles in patients with PTB at diagnosis and throughout TB treatment and within 1 week of TB treatment - expression of more than 1,200 genes was markedly downregulated
- Marked changes in gene transcription profile at the end of treatment
- Transcriptional changes also reflect alterations in blood cell numbers including neutrophils (increased), monocytes (increased) and lymphocytes (decreased) in TB



The signature is now subsequently been validated in numerous studies and we also know that the blood gene expression profile that is present at M tb at PTB in diagnosis is actually changed over the course of treatment such that, within a week of TB treatment there is a massive down regulation of more than 1200 genes when we look at transcriptional signatures. And in fact, these changes in gene transcription profile continue till the end of treatment.

So, transcriptional changes are also thought to basically reflect, changes in blood cell numbers that occur during TB infection and disease, including neutrophils which are increased, monocytes which are increased and lymphocytes which are decreased in TB disease.

(Refer Slide Time: 07:27)



Manage TB

ICMR - National Institute for Research in Tuberculosis (NIRT)  
www.nirt.res.in

## TB Biomarkers - correlates of risk

- Individuals with LTBI with low-grade, subclinical infection are at higher risk of reactivation
- Whole-blood transcriptomic mRNA expression signature was identified by mining RNA-sequencing data in a large prospective cohort of adolescents with LTBI from South Africa
  - ✓ 16-gene transcriptomic correlate of risk signature predicted progression from infection to TB disease with a sensitivity of 66% and a specificity of 81% in the 12 months preceding diagnosis of incident TB disease

So, when we look at correlates of risk. The idea behind correlates of risk is trying to understand or identify which individuals with LTBI who have low grade subclinical infection are in the highest risk of reactivation. So, if we can identify people with the highest risk of reactivation, they can be selectively targeted for therapy or for prophylactic treatment.

So, one of the few studies that have actually been able to establish a correlate of risk was a study looking at whole blood transcriptomic mRNA profile, wherein a signature was identified by mining RNA sequencing data, from a large prospective cohort of adolescents with latent tuberculosis from South Africa.


It was found that these individuals actually exhibited a 16 gene transcriptomic signature, which turned out to be a good quadrant of risk and predicted progression of infection from TB disease to from infection to TB disease with a sensitivity of about 66 percent and a specificity of about 81 percent. At least 12 months preceding the diagnosis of incident TB.

(Refer Slide Time: 08:40)

Manage TB

## TB Biomarkers - Immune biomarkers

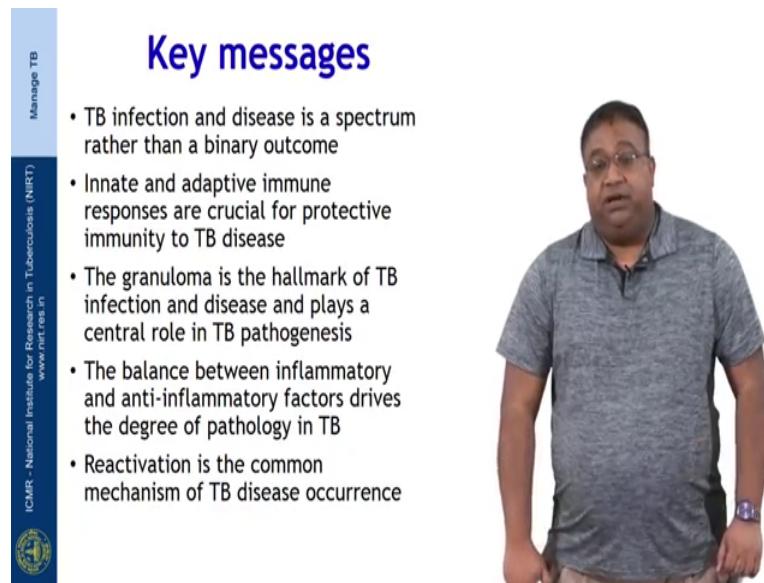
Immune markers	Description
Cytokines	IFN- $\gamma$ , CXCL10, CXCL8 and CCL8, IFN- $\gamma$ /IL-4 ratio
Receptors and soluble receptors	sUPAR, sICAM1, sIL-2R, sTNFR1, sTNFR2, CD11c, LAG3
Inflammation markers	CRP, neopterin, procalcitonin, ADA and Granzyme B
Immune cells and their markers	polyfunctional T cells, Single positive TNF-expressing CD4+ T cells, monocyte/lymphocyte ratio, Foxp3, CXCL8 and IL-12 $\beta$



Finally we look at TB biomarkers the other area of major interest is of course, looking at immune biomarkers and these include, certain cytokines which have been associated with TB disease and this typically involves interferon gamma, CXCL 10, CXCL 8, CCL 8 and interferon gamma and IL 4 alpha ratio, the presence of receptors and soluble receptors including soluble upar, soluble icam one soluble IL 2 IL 2receptor soluble TNF alpha receptor 1 and 2, CO 11 c and lag 3.

Inflammation markers which are typically nonspecific, but also associated are associated typically with TB disease and these include C reactive protein, neopterin, procalcitonin ada and granzyme B. And finally, immune cells themselves and their markers can also be associated with active TB and help distinguish active TB from latent infection and other diseases; these include poly function of T cells, single positive TNF expressing T cells, monocyte lymphocyte ratio, Foxp 3 CXCL 8 and IL 12 beta.

(Refer Slide Time: 09:54)



The slide features a vertical blue bar on the left with the text 'Manage TB' at the top, 'ICMR - National Institute for Research in Tuberculosis (NIRT)' in the middle, and 'www.nirt.res.in' at the bottom. To the right of the bar is the title 'Key messages' in blue. Below the title is a list of five bullet points. To the right of the text is a photograph of a man with glasses wearing a grey polo shirt.

**Key messages**

- TB infection and disease is a spectrum rather than a binary outcome
- Innate and adaptive immune responses are crucial for protective immunity to TB disease
- The granuloma is the hallmark of TB infection and disease and plays a central role in TB pathogenesis
- The balance between inflammatory and anti-inflammatory factors drives the degree of pathology in TB
- Reactivation is the common mechanism of TB disease occurrence

So, to conclude the key messages that I want everybody to take home from the session on the pathogenesis of TB are as follows, TB infection and disease is actually a spectrum rather than a binary outcome, innate and adaptive immune responses are crucial for a protective immunity to TB disease.

The granuloma is the hallmark of TB infection and disease and plays a central role in TB pathogenesis, the balance between inflammatory and anti-inflammatory factors drives the degree of pathology in TB and finally, reactivation is the common mechanism of TB disease occurrence in addition to this. This we come to the end of the session.

Thank you very much.