TB Screening and Interferon Gamma Release Assays

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Overview

- Public health goals of TB screening
- Tools of Screening
- IGRAs: current evidence
- Clinical scenarios
Is latent TB really latent?

Granulomas are dynamic lesions

- Constant rotation of new and dying immune cells
- Equilibrium of activity of dividing M.tb and non-replicating persistence
- Tumor necrosis factor key in maintenance

Consensus Statement, Eur Respir J 2009; 33: 956-973
Screening Goals and Methods

1. Find cases

2. Prevent cases
I would insert a slide here on your former position and being on the receiving end of immigrants and refugees screened overseas. Then discuss important identification of LTBI cases can be for health departments. This will lead into a nice transition to the next slide.
Importance of LTBI detection and treatment

Expected success if active TB is diagnosed and treated compared with also finding and treating LTBI.

For major impact on TB disease rates we must address latent TB infection

Adapted from Abu-Raddad et al. Epidemiological benefits of more-effective tuberculosis vaccines, drugs, and diagnostics. PNAS. 2009; 106; 13980-5.
Tuberculosis is the Tip of the Iceberg

Globe: ~9 million Active cases

2 Billion With LTBI*

Silent carriers

Congregate settings
Weak Infection control
Urban crowding

HIV
Diabetes
Immunosupression

Total Population 7 billion (2011)
Worldwide: 2 billion infected with TB
…. Have germ, will travel

Migrating Populations in the 1990s
*Compared to 1960-75, four-fold increase in migration*

Source: Population Action International 1994

Slide originally created by Jenny Flood, M.D, California Dept. of Public Health
Panel Physicians plays a critical role

Low burden countries benefit from preventing active cases from entry
- Identification of active TB and treatment
  - stops the chain of transmission
  - early case finding – decreases morbidity/mortality
  - demonstrates state of the art care in high burden setting

…and preventing cases through LTBI treatment after entry
- Identification of LTBI (B2)
  - addresses the future cases of reactivation TB that will occur among young immigrants with LTBI and older immigrants with inactive fibrosis*

*Note: A chest radiograph can only be said to have reactivation fibrosis if smears and cultures are negative
Diagnostic tools for TB infection

TB Skin Test (TST)  Interferon Gamma Release Assays (IGRAs)
TST and IGRAs are indirect methods of detection
NO GOLD STANDARD for LTBI

Presentation of mycobacterial antigens

Antigen presenting cell

Memory T-cell

TB testing: How good are our tests?

Facts:

- TST and IGRAs are indirect methods and are dependent on a healthy immune system.
- No gold standard to compare.
- Accuracy of tests depends on the prevalence of infection.
**Current IGRAs**

**QuantiFERON® (QFT):** Whole blood incubated with MTB specific antigens → free IFN-γ release is measured (ELISA)

**T-Spot®.TB:** T-cells incubated with MTB specific antigens; IFN-γ releasing cells are counted (ELISpot-based technology)

Adapted slide: originally created by Annie Leutkemeyer, M.D, UCSF
# IGRAs: Interpreting Results (US standards)

<table>
<thead>
<tr>
<th></th>
<th>Positive</th>
<th>Negative</th>
<th>Gray Zone</th>
<th>Indeterminate</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>QuantiFERON®-TB In-Tube</strong></td>
<td>≥0.35*</td>
<td>&lt;0.35 *</td>
<td>None</td>
<td>Controls fail: High Nil</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Poor Mitogen response</td>
</tr>
<tr>
<td><strong>T Spot TB™</strong></td>
<td>≥8 spots*</td>
<td>&lt; 8 spots*</td>
<td>5-7 spots*</td>
<td>same as above</td>
</tr>
</tbody>
</table>

*(TB Ag - Nil) and assumes appropriate control responses*
Indeterminate Results

- Is meaningful and does not necessarily indicate a failed test

- Indicates one of the following
  - Technical error
  - May indicate immune suppression
  - High background IFN-γ levels
# Indeterminates

<table>
<thead>
<tr>
<th>No. of subjects</th>
<th>No. Indeterminates</th>
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</thead>
<tbody>
<tr>
<td>22,752</td>
<td>445</td>
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</table>

**Indeterminate Rate (%)** 1.96

*Data source:*
More than 70 publications using QFT-Gold In-tube
Indeterminate results: Test vs. host failure

High background gamma interferon

- Intercurrent illness (random)
- Faulty collection tube lots (sudden high rate of indeterminates from a single site)
- Mitogen put in the wrong well (nil value high and mitogen low)

Low mitogen control

- Transient or chronic immune suppression
- QFT-GIT: overfilling, or inadequate shaking, switched tubes when running ELISA
- Tspot: no mitogen in control well
Indeterminate Results: What to do

- REPEAT the QFT: Our SF data (healthy ambulatory population) tells us that you will get a valid result (usually negative) >80% of the time

- Low mitogen indeterminate in very young kids (<5), very ill or debilitated elderly: maximize sensitivity
  - use other available tests (e.g., TST, other available IGRAs)

- Work with your lab: report runs that have >5% indeterminate results
Interferon Gamma Release Assays vs. Tuberculin Skin Test

**IGRA**

- *In vitro*
- Single antigens
- Automated results
- Not affected by BCG
- Result with one patient visit
- Minimal inter-reader variability
- Results: one standard for all QFT, gray zone for Tspot

**TST**

- *In vivo*
- Multiple antigens
- Manual reading and entry
- BCG may affect results
- Two patient visits required for result
- Significant inter-reader variability
- Results: different cut points based on risk
# Specificity of IGRA

## Species Specificity of IGRA TB Antigens

<table>
<thead>
<tr>
<th>Tuberculosis Complex</th>
<th>ESAT-6</th>
<th>CFP-10</th>
<th>TB7.7</th>
<th>Environmental Strains</th>
<th>ESAT-6</th>
<th>CFP-10</th>
<th>TB7.7</th>
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<tbody>
<tr>
<td>M tuberculosis</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>M abcessus</td>
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<tr>
<td>M africanum</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>M avium</td>
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<td>M bovis</td>
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<td>+</td>
<td>+</td>
<td>M branderi</td>
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<tr>
<td>BCG substrain</td>
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<td></td>
<td>M szulgai</td>
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<td>M xenopi</td>
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</table>

IGRAs do NOT react to BCG or NTM!
Which test do I use?
2010 IGRA CDC Guidelines

- IGRAs can be used in all situations where the skin test is currently being used

- IGRAs preferred:
  - BCG vaccinated persons
  - Persons unlikely to return for a TST reading
  - Low risk individuals

- Like the TST, clinical judgment required when interpreting IGRA results in children <5yrs, immunocompromised persons, and TB suspects

- TST preferred in children <5yrs

- When maximum sensitivity needed → acceptable to use both TST and IGRA (Culture and DOT TB TI currently allow use of either, but not both tests)

- Lab should report quantitative results
Peer reviewed IGRA publications over time

Source: PubMed 17 Aug 11 ("20xx"[Entrez Date] : "20xx"[Entrez Date]) AND Quantiferon
<table>
<thead>
<tr>
<th>Performance Characteristics</th>
<th>TST</th>
<th>QFT-Gold IT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Est. sensitivity</td>
<td>67-72%*</td>
<td>78-83%*</td>
</tr>
<tr>
<td>Est. specificity</td>
<td>59% (+BCG)**</td>
<td>98-100%*</td>
</tr>
<tr>
<td>Correlates with exposure</td>
<td>Often no</td>
<td>Yes</td>
</tr>
</tbody>
</table>

*Diel, Chest published online 12/09  **Pai et al 2008
Predictive power of QFT for development of active TB
Diel, Loddenkemper et al., AJRCCM, 27 August 2010

954 close contacts

198 QFT-positive

142 QFT-positive/TST-positive
Not treated
17 developed active TB

5 QFT-positive TST-negative
Not treated
2 developed active TB

51 QFT-positive (49 TST-positive)
Chemoprophylaxis RIF and/or INH
No active TB

756 QFT-negative

413 TST positive
Not treated
No active TB

343 TST negative
Not treated
No active TB

Mean follow-up >3.5 yr
Predictive value of interferon-gamma release assays and tuberculin skin testing for predicting progression from latent TB infection to disease state: a meta-analysis

R. Diel, R. Loddenkemper and A. Nienhaus

_Chest; Prepublished online April 5, 2012; DOI 10.1378/chest.11-3157_
Results: Diel et al, 2012 CHEST

Pooled positive predictive value (PPV) for progression:
Commercial IGRAs was 2.7%.
TST was 1.5%.

PPV for progression in high risk groups:
IGRAs was 6.8%.
TST 2.4%

Pooled values of negative predictive values (NPV) for progression
IGRAs: 99.7% [p<0.01
TST: 99.4%
What happens in pediatric populations when IGRAs are used?

- **517 healthy children** without immunosuppression: 1 month to 18 years (34% under <5)
- 355 (68.7%) BCG vaccinated: mostly adopted Asian and Latino children
- 434 (84%) TST+
- 25/434 (5.8%) QFT+
- 25 (5.4%) indeterminate
- Children <5 years of age: Adequate mitogen and Ag responses found
- Conclusion: **Majority of TST+ children represents false positive results**

Clinical Scenarios

Warning: Diagnosis and Prevention requires thinking TB!

No TB diagnostic can “rule out” TB by itself!

*Our tools are only as smart as the person that uses them*
Can our tools rule out active TB?
NEVER use a TST or IGRA to:

“Rule out” disease in TB suspects (symptoms, abnormal CXR, or physical finding suspicious of TB)

“Rule out” LTBI in immunocompromised individuals in the setting of high exposure and high risk of disease progression (e.g., HIV, children under 5, transplant patients, those on immunosuppressive drugs)

NO TEST CAN REPLACE CLINICAL JUDGMENT!!!!
Can our tools falsely “rule in” LTBI?
False-positive Results

TST

- Cross-reactions and boosted reactions (NTMs and BCG)
- Misinterpretation of immediate hypersensitivity to tuberculin
- Switching tuberculin products standardized to RT-23

IGRA

- Cross-reactions from *M. kansasii*, *M. szulgai* and *M. marinum*
- Technical problem
Can our tools result in false negative results?
TST and IGRA test interpretation: False-negative or indeterminate results

Host factors affecting TST and likely IGRA

- HIV - low CD4, no ARVs
- Recent TB infection (<10 weeks)
- Infections (viral, fungal, bacterial)
- Other illness affecting lymphoid organs
- Live virus vaccination
- Immunosuppressive drugs
- Overwhelming TB
- Age (newborn, elderly)

TST technical factors

- Improper storage, contamination
- Improper method of administration, reading and/or recording of results

IGRA technical factors

- QFT: Insufficient mixing
- Overfill of mitogen
- Lab errors
Common Questions

- When should an IGRA be repeated in contact investigation?
- Is it okay to do an IGRA after a childhood vaccination?
- Should I treat a positive IGRA result more seriously in low risk patients? Do they need to be treated?
- How often do you screen with an IGRA?
- Should we test those who have been treated with INH or for active TB?
- Will a TST boost an IGRA response?
- Should we test those who have been treated with INH or for active TB?
When in doubt, follow guidelines and expert advice for TST

- Impact from vaccinations: delay test for 10-12 weeks or do test on the day of vaccination
- Follow-up testing in contacts: 8-10 weeks
- A TST cannot boost a reaction in an uninfected person but may if there is prior infection
- Always test prior to start of immunosuppressive therapy
- Frequency of testing should be determined by the risk of exposure

Above all, do not test low risk populations!!!
Using Quantitative IGRA data: What is currently known

IFN-γ levels ↓ with treatment of LTBI or TB disease but reversion is not consistent

_Don’t use levels to determine treatment efficacy_

Variability of levels seen in individuals on different days

_This may impact individual having serial testing_

In serial testing, higher conversion/reversion rates are seen with IGRAs compared to TST

_A quantitative definition of conversion is needed_

Lower levels may be seen in immunocompromised persons
Other Common Questions

Can we use the IGRA to monitor LTBI treatment and to see if treatment is working?

I heard IGRAs measure recent infection and not remote infection. Is that true?

Once positive, always positive? Is that true?
Other Common Questions

Can we use the IGRA to monitor LTBI treatment and to see if treatment is working?

**NO, current research does not validate this practice.**

I heard IGRAs measure recent infection and not remote infection. Is that true?

**NO, this is a theory and the jury is still out.**

Once positive, always positive? Is that true?

**NO, IGRA research and review of old TST studies show otherwise.**
You are now an IGRA expert.
Would an IGRA be useful in the following scenario?

• MD wants QFT approval for a 61 y/o Nepali patient who is in the hospital for 1/2 cup hemoptysis and lingula infiltrate. Prior TST+ and vague history of taking multiple TB meds.

• 39 y/o pregnant diabetic AA US-born TST converter (last TST done at a community site). Prior to pregnancy, had INH stopped because of recurrent nausea and vomiting.

• 2.5 y/o healthy asymptomatic Chinese-born male with +TST (12 mm). History of BCG at birth. He is hyperactive and mom wants to avoid giving unnecessary medication.
Summary

• IGRAs are a significant advance because of its high specificity and operational advantages to the TST

• Like the TST, it is not a “rule out test” for active TB or immunocompromised persons with LTBI

• IGRAs are powerful epidemiologic and clinical tools

• Cost effective: Eliminates unnecessary CXRs, medical evaluations, treatment and toxicity

• Research and experience is growing rapidly on serial testing, young children and risk of progression
Thank you