TLR1 GENE IS ASSOCIATED WITH LEPROSY RISK AMONG BRAZILIANS

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Leprosy: a complex disease

- The course of leprosy infection is under environmental and host influence

Which are the genes/genetic markers associated with leprosy susceptibility?
Genetic variations regulate immunity and leprosy outcome
Toll-like receptor 1 (TLR1) and leprosy

**TLR1:**
- Plays a fundamental role in pathogen recognition and activation of inflammatory response;
- *M. leprae* activates TLR1/TLR2 dimer, inducing the antimicrobial response in human macrophages (Krutzik et al., 2003; Liu et al., 2006)

**TLR1 : Genetically associated with Leprosy**

- N248S: risk to leprosy - Bangladesh (Schuring et al., 2009)
- I602S: protection to leprosy - India (Wong et al., 2010)
GOALS

1. Evaluate the association of SNPs at Toll-like receptor 1 gene and leprosy in a Brazilian sample

2. Investigate the Toll-like receptor 1 genotype-phenotype correlation
Genetic Study in Brazilian Samples

Case-control study

**Rio de Janeiro - RJ**
- Cases: 584
- Controls: 564
- N= 1,148

**Rondonopolis - MT**
- Cases: 402
- Controls: 413
- N= 815

**Bauru - SP**
- Cases: 450
- Controls: 376
- N= 826

Family-based study

**Almenara - MG**
- Trios: 125
- Offspring: 147
- N= 447
RESULTS

Bauru-SP Case-Control: TLR1 248S was associated with leprosy risk

**TLR1 N248S**
- NS: 1.58, p = 0.01
- SS: 1.81, p = 0.004

**TLR1 I602S**
- IS: 0.79, p = 0.15
- SS: 0.83, p = 0.40
Almenara-MG Family-Based Study: TLR1 248S was also associated with leprosy risk

Transmission Disequilibrium Test of N248S at *TLR1* in familial sample from Almenara, Brazil.

<table>
<thead>
<tr>
<th>SNP</th>
<th>Alleles</th>
<th>Frequency</th>
<th>Transmitted</th>
<th>Untransmitted</th>
<th>z (p-value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>N248S</td>
<td>N</td>
<td>0.55</td>
<td>34</td>
<td>53</td>
<td>2.15 (0.05)</td>
</tr>
<tr>
<td></td>
<td>S</td>
<td>0.45</td>
<td>53</td>
<td>34</td>
<td></td>
</tr>
<tr>
<td>I602S</td>
<td>I</td>
<td>0.67</td>
<td>52</td>
<td>30</td>
<td>2.16 (0.06)</td>
</tr>
<tr>
<td></td>
<td>S</td>
<td>0.33</td>
<td>30</td>
<td>52</td>
<td></td>
</tr>
</tbody>
</table>

RESULTS
RESULTS

Replication case-control studies: confirmed the association of 248S with leprosy susceptibility

Rio de Janeiro

TLR1 N248S

<table>
<thead>
<tr>
<th>OR</th>
<th>NS: 0.83, p = 0.23</th>
<th>SS: 1.59, p = 0.006</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>protection</td>
<td>risk</td>
</tr>
<tr>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
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</table>

Rondonopolis

TLR1 N248S

<table>
<thead>
<tr>
<th>OR</th>
<th>NS: 1.47, p = 0.03</th>
<th>SS: 1.56, p = 0.03</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>protection</td>
<td>risk</td>
</tr>
<tr>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
RESULTS

Combined Brazilian Sample (Bauru, Rio de Janeiro and Rondonopolis)

Combined results for TLR1 N248S in Brazilian case-control studies

<table>
<thead>
<tr>
<th>N248S Genotype/allele</th>
<th>Cases</th>
<th>Controls</th>
<th>OR (p-value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>280 (0.22)</td>
<td>354 (0.27)</td>
<td>reference</td>
</tr>
<tr>
<td>AG</td>
<td>622 (0.49)</td>
<td>659 (0.48)</td>
<td>1.22 (p= 0.03)</td>
</tr>
<tr>
<td>GG</td>
<td>374 (0.29)</td>
<td>340 (0.25)</td>
<td>1.51 (p= 0.0002)</td>
</tr>
<tr>
<td></td>
<td>1276</td>
<td>1353</td>
<td></td>
</tr>
<tr>
<td>A</td>
<td>1182 (0.46)</td>
<td>1367 (0.51)</td>
<td>reference</td>
</tr>
<tr>
<td>G</td>
<td>1370 (0.54)</td>
<td>1339 (0.49)</td>
<td>1.23 (p= 0.008)</td>
</tr>
<tr>
<td>G carriers</td>
<td>996 (0.78)</td>
<td>999 (0.73)</td>
<td>1.31 (p= 0.003)</td>
</tr>
</tbody>
</table>

a N (frequency). b adjusted for gender, ethnicity and Brazilian region by logistic regression analysis

The TLR1 248 S variant was associated with leprosy susceptibility in Brazilians
RESULTS

Genotype-phenotype correlation: 248S effect over TLR1 structure

Effect of N248S in electrostatic profile at TLR1 by molecular dynamics analysis. The white circles encompass the N248 or S248 region. Molecular dynamics analysis by Pymol and Gromacs softwares.

The TLR1 248 S variant modified the electrostatic profile: more electronegative
RESULTS

Genotype-phenotype correlation: 248S effect over TNF/IL10 production

TNF/IL10 ratio in non-carriers (NN) and carriers (NS + SS) of S allele at N248S. PBMCs from healthy subjects stimulated with several stimulus in two independent experiments (A: 18h, B: 24h). Cytokines production was evaluated in supernatants by ELISA.

The TLR1 248 S variant was correlated with reduced inflammatory profile
CONCLUSIONS

TLR1 - 248S

- Associated with leprosy risk in Brazilians
- Modifies electrostatic profile
- Reduces inflammatory profile
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Thank you!