Vitamin D-Receptor Gene Polymorphisms: Association With Virologic Failure and Mortality

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BACKGROUND
• Host genetic factors play an important role in HIV infection and disease progression.
• Due to its extensive role in the immune system, vitamin D may have a potential role in altering HIV disease progression.
• Vitamin D actions are mediated by the vitamin D receptor (VDR).
• Polymorphic variations exist in the VDR gene, each of which may have different significance in disease susceptibility and progression.
• Different loci of the VDR gene have different functions so that associations of HIV disease progression with specific polymorphisms may indicate unique mechanisms.

OBJECTIVES
• To examine if VDR gene polymorphisms in the 5’ coding region, corresponding to Bsm1 and the 3’ Untranslated region, corresponding to Apa1, Apa1, and Taq1q are associated with virologic failure and mortality among HIV-infected African patients

METHODS
• 142 HIV+ patients initiating ART between November 2008 - March 2009 at the Themba Lethu Clinic, Johannesburg, South Africa.
• Eligibility criteria:
  • age ≥ 18 years
  • CD4 ≤ 350 cells/mm³
  • first-line ART
  • pregnant women were excluded
  • ART naïve patients only

Study outcomes (Disease progression):
1. Failure to achieve viral suppression (<50 copies/ml) within 12 months after initiation of ART.
2. Mortality during the follow-up period

RESULTS

• Genotyping:
  • Genomic DNA was extracted from whole blood using standard methods and stored at -20°C until required.
  • DNA from each person was analysed for the polymorphisms in the VDR gene using polymerase chain reaction – restriction fragment length polymorphism (PCR-RFLP).
  • Restriction digestion of the PCR product was performed according to the manufacturer’s instructions and results visualised on 2% agarose gels incorporating GelRed™ (Anatech, Johannesburg, South Africa).

• Cox proportional regression models constructed for univariate and multivariate analysis
• Multivariate models adjusted for: age, gender, BMI, WHO stage, CD4 count, hemoglobin level, employment status and TB status.
• Tests to estimate Hardy-Weinberg equilibrium conducted by various methods and stored at -2°C until required.

• Statistical analysis:
  • Kaplan-Meier curves and the log-rank test used
  • Logistic regression analysis

• Key findings:
  • Individuals carrying non-bb variants of the Bsm1 polymorphism in the VDR vs. the bb variant:
    • 3.5 fold greater risk of virologic failure
    • 3.5 fold greater risk of mortality
  • We could not demonstrate associations between disease progression and the Apo1, Fok1 and Taq1q variants

CONCLUSIONS
• Patients exhibiting the VDR B allele may be less responsive to the immunoregulatory actions of vitamin D and hence show less favorable disease progression.
• Also, it is possible that the VDR B allele may be a marker allele which is linked to a functional allele located nearby in the same gene influencing HIV disease progression.
• Understanding this role of VDR genotypes among HIV-infected patients may offer new insights for developing therapeutic strategies to alter disease progression.

Table 1: Characteristics of study cohort at baseline and end of follow-up

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Baseline</th>
<th>Follow-up</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CD4 count (cells/mm³)</td>
<td>200</td>
<td>350</td>
</tr>
<tr>
<td>Hemoglobin level (g/dL)</td>
<td>11.1</td>
<td>12.9</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Transfer</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Loss to follow-up</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Death</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total person-years at risk</td>
<td>38.1</td>
<td>40.4</td>
</tr>
</tbody>
</table>

Table 2: Associations between Vitamin D Receptor Gene Polymorphisms and virologic failure within 12 months post-ART initiation

<table>
<thead>
<tr>
<th>VDR Polymorphism</th>
<th>Odds Ratio</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bsm1 bb</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Bsm1 Bb</td>
<td>3.50</td>
<td>1.50-7.90</td>
</tr>
<tr>
<td>Apa1 bb</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Apa1 Bb</td>
<td>3.00</td>
<td>1.50-5.90</td>
</tr>
<tr>
<td>Taq1q bb</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Taq1q Tq</td>
<td>3.00</td>
<td>1.50-6.10</td>
</tr>
</tbody>
</table>

Figure 1: Kaplan-Meier curves for VDR genotype and stratified failure

Acknowledgements: Doctoral Training Grant from the Consortium for Advanced Research Training in Africa (CARTA). CARTA has been funded by the Wellcome Trust (UK) (Grant No: HT92/002), the Department for International Development (UK), the Wellcome Trust, and The University of the Witwatersrand Faculty of Health Sciences Individual Research Grants (Medical Embarkation Award). Contact: Simba Takuva | stakuva@cartafrica.org