Targeted Sequencing Identifies Missense Variant in the \textit{BEST3} Gene Associated with Antihypertensive Response to Thiazide Diuretics

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Hydrochlorothiazide is one of the most commonly prescribed antihypertensive in the US.

- Poorly understood mechanisms
- Variability in BP response
- Difficult Clinical Prediction

**PHARMACOGENOMICS**

- Elucidation of genetic determinants of HCTZ blood pressure response

**PEAR**
(Pharmacogenomic Evaluations of Antihypertensive Response)

**GERA**
(Genetic Epidemiology of Responses to Antihypertensives)
STUDY DESIGN

PEAR/GERA

Baseline

PEAR -> 9 weeks
GERA -> 4 weeks
Hydrochlorothiazide Treatment

Post Treatment

Genetic variability in this region is determinant of antihypertensive response to HCTZ

Chromosome 12q15 locus associated with BP response to HCTZ

No functional polymorphism was identified in the chromosome 12 locus

No clear understanding of the underlying mechanism could be determined

Successfully replicated in PEAR
### PHASE I - Participant Demographics

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>PHASE I (361)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Responder (%)</td>
</tr>
<tr>
<td>N</td>
<td>181(50.01)</td>
</tr>
<tr>
<td>PEAR participants</td>
<td>97(27.14)</td>
</tr>
<tr>
<td>GERA participants</td>
<td>84(23.26)</td>
</tr>
<tr>
<td>African Americans</td>
<td>122(33.79)</td>
</tr>
<tr>
<td>Baseline SBP (mmHg)</td>
<td>146.66±12.19</td>
</tr>
<tr>
<td>Baseline DBP (mmHg)</td>
<td>95.76±5.81</td>
</tr>
<tr>
<td>Post treatment SBP (mmHg)</td>
<td>125.06±10.54</td>
</tr>
<tr>
<td>Post treatment DBP (mmHg)</td>
<td>80.62±6.52</td>
</tr>
<tr>
<td>Δ SBP (mmHg)</td>
<td>-21.78±9.72</td>
</tr>
<tr>
<td>Δ DBP (mmHg)</td>
<td>-15.04±6.04</td>
</tr>
</tbody>
</table>
Phase I

- Logistic regression was used for analysis and was adjusted for: Baseline BP, age, gender, race and principal components 1 and 2.

- BEST3 was not annotated at the time of target selection
- Only a part of the BEST3 gene was captured in Phase I

**Chr12: 69,357,185 - 70,047,375**

**Missense SNP rs61747221**
• All the participants of phase II were genotyped for rs61747221 and included in phase II data analysis analysis
Validation – Entire Cohorts of PEAR and GERA

- Rs61747221 was tested for association with change in systolic (ΔSBP) and diastolic BP (ΔDBP) response post hydrochlorothiazide treatment.

<table>
<thead>
<tr>
<th>SNP</th>
<th>rs61747221</th>
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</thead>
<tbody>
<tr>
<td>PEAR (N=370)</td>
<td></td>
</tr>
<tr>
<td>DBP P-Value</td>
<td>0.023</td>
</tr>
<tr>
<td>β</td>
<td>-1.08</td>
</tr>
<tr>
<td>SBP P-Value</td>
<td>0.021</td>
</tr>
<tr>
<td>β</td>
<td>-1.60</td>
</tr>
<tr>
<td>GERA (N=571)</td>
<td></td>
</tr>
<tr>
<td>DBP P-Value</td>
<td>0.032</td>
</tr>
<tr>
<td>β</td>
<td>-1.28</td>
</tr>
<tr>
<td>SBP P-Value</td>
<td>0.028</td>
</tr>
<tr>
<td>β</td>
<td>-1.95</td>
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</tbody>
</table>
Minor Allele Frequency = 0.13

- The variant allele carriers were grouped together
- Association analyses using dominant model
<table>
<thead>
<tr>
<th>SNP</th>
<th>Function</th>
<th>Allele Change</th>
<th>Residue Change</th>
</tr>
</thead>
<tbody>
<tr>
<td>rs61747221</td>
<td>missense</td>
<td>CCA → CTA</td>
<td>PRO → LEU</td>
</tr>
</tbody>
</table>

Pathogenicity Prediction using SIFT

SIFT predicted rs61747221 to have an “intolerant/damaging” effect on BEST3 protein

**BEST3 gene - Biological Candidate for HCTZ BP response**

- *BEST3* encodes for bestrophin3 and acts as a **calcium-activated chloride channel**
- **Essential** for the cyclic GMP-dependent **vascular smooth muscle relaxation and maintaining the vaso-motion of blood vessels**
We identified and validated a novel missense SNP in the **BEST3 gene** highly associated with blood pressure response to HCTZ treatment.

**BEST3** is an excellent biological candidate for HCTZ mediated BP regulation.
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