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A. B. SPAN/SPAN (Fold Change)

R-568

CaSR mWNK4 (HA) hSPAK (GFP-HA)
PpSAPK (S373) pERK β-actin

C. D. SPAN-mWNK4-CaSR

R-568

pSAPK (S373) hSPAK (GFP-HA) β-actin
A. 

![Image](image-url)

<table>
<thead>
<tr>
<th>Condition</th>
<th>mWNK4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Empty</td>
<td>-</td>
</tr>
<tr>
<td>CaSR WT</td>
<td>-</td>
</tr>
<tr>
<td>CaSR E228K</td>
<td>+</td>
</tr>
<tr>
<td>CaSR R185Q</td>
<td>+</td>
</tr>
</tbody>
</table>

B. 

![Bar chart](bar-chart-url)

**Klhl3 DNA**
- -
+ +
+ +
+ +

C. 

![Graph](graph-url)

**WNK4/β-actin (Fold Change)**
- Control
- CaSR WT
- CaSR E228K
- CaSR R185Q

D. 

![Graph](graph-url)

**WNK4/β-actin (Fold Change)**
- Control
- CaSR WT
- CaSR E228K
- CaSR R185Q

Klhl3 DNA:
- +
- +
**A.**

Table and graph showing fold change in pRRXS/KLHL3 interaction under different conditions:

<table>
<thead>
<tr>
<th>Condition</th>
<th>Control</th>
<th>CaSR WT</th>
<th>CaSR E228K</th>
<th>CaSR R185Q</th>
</tr>
</thead>
<tbody>
<tr>
<td>pRRXS/KLHL3 (Fold Change)</td>
<td><strong>2.0</strong></td>
<td><strong>1.5</strong></td>
<td><strong>1.0</strong></td>
<td><strong>0.5</strong></td>
</tr>
</tbody>
</table>

**B.**

Graph showing fold change in pRRXS/KLHL3 interaction with BIM treatment:

<table>
<thead>
<tr>
<th>Condition</th>
<th>Control</th>
<th>CaSR E228K</th>
</tr>
</thead>
<tbody>
<tr>
<td>pRRXS/KLHL3 (Fold Change)</td>
<td><strong>2.5</strong></td>
<td><strong>2.0</strong></td>
</tr>
</tbody>
</table>

**C.**

Western blot images showing protein expression for SPAK-WNK4-CaSR complex:

- pWNK4 S1196
- mWNK4 (HA)
- pERK
- SPAP (373)
- hSPAK (HA)
- β-actin

**D.**

Western blot images showing protein expression for CaSR and mWNK4 with and without R-568 treatment:

- CaSR
- mWNK4 (HA)
- pERK
- hSPAK (HA)
- β-actin
Increased calcium and salt delivery to the distal nephron

Hypercalcemia

Salt recovery

Calcium excretion