1. Introduction/Background

- Selected renal cells (SRC), a renal epithelial cell-enriched platform, are being advanced in a Phase 3 Global Registrational trial for treatment of chronic kidney disease (CKD).
- In CKD models, administration of SRC is associated with improved survival, preservation of renal microarchitecture, and reduced renal dysfunction.
- Preliminary data from a subset of diabetic kidney disease patients suggest randomization to SRC is associated with improvement in glomerular filtration rate.

2. Goal

To test the hypothesis that the renal restorative and reparative effects of SRC are mediated, at least in part, via its nephrogenic potential.

3. Methods

- Bioinformatics with empirical data were coupled to evaluate genes coding for Rack1 (Gnb2l1), Six2, Osr1, Ret, Fgf8, Lhx1, Nphs1 and Nphs2, proteins that are co-expressed by human SRC.
- Genes were seeded into a variety of scaffold systems such as hydrogel, these organoids self-assemble into tubes expressing the marker cytokeratin 18 (A, arrow, 5X). In the presence of a scaffold such as hydrogel, these organoids self-assemble into tubules expressing the marker cytokeratin 18 (B, arrow, 5X). Staining for Keratin-18 antibody in the human healthy kidney is also localized to the tubules (D, The Human Protein Atlas). STRING (A) and GeneMANIA (B) queries suggest that genes coding for SRC proteins are not co-expressed by human tissue. The SRC interactome potentially comprises kireit? (B) whose product (Neph1) participates in glomerular barrier function.

4. Results

- Genes coding for SRC proteins typically do not appear to be co-expressed by human tissue; kireit1, which codes for Neph1, and maintains glomerular barrier integrity, is potentially expressed by SRC.
- Both the SRC gene interactome and SRC gene products are compartmentalized within the tubules and/or glomeruli (podocyte) and participate in kidney development/nephrogenesis.
- SRC cultures form organoids which self-assemble into tubules in the presence of a scaffold.

5. Discussion

- SRC (REACTTM), a renal epithelial cell-enriched platform with a unique protein co-expression profile, is being evaluated in a Phase 3 clinical trial in subjects at increased risk (CKD 3b/4) for kidney failure.
- The SRC gene interactome and SRC proteins appear compartmentalized within tubules and glomeruli (podocytes) and participate in processes associated with kidney development.
- SRC forms organoids which self-assemble into tubules in vitro.

6. Conclusions

The nephrogenic potential of SRC may underlie, at least in part, its renal restorative and reparative activity observed in clinical trials.

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**Gene Ontology Reveals Potentially Unique Mechanism of Action Underlying Selected Renal Cells Bioactivity**

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**Figure 1:** Gene/Protein Co-expression Profile Unique to SRC

**Figure 2:** Tubular and/or Glomerular Compartmentalization of the SRC Gene Interactome

**Figure 3:** Tubular and/or Glomerular Compartmentalization of SRC Proteins

**Figure 5:** SRC Organoids Self-assemble into Tubules: Cytokeratin-18 Expression by SRC Tubules Consistent w/ Observation in Human Healthy Kidney

**Table 1:** SRC Genes Participate in Kidney Development

<table>
<thead>
<tr>
<th>Gene Ontology Biological Process (GO BP)</th>
<th>SRC Proteins</th>
</tr>
</thead>
<tbody>
<tr>
<td>nephron morphogenesis</td>
<td>Rack1</td>
</tr>
<tr>
<td>mesonephric nephron development</td>
<td>Six2</td>
</tr>
<tr>
<td>renal vesicle development</td>
<td>Ret</td>
</tr>
<tr>
<td>mesonephric nephron development</td>
<td>Fgf8</td>
</tr>
<tr>
<td>renal tubule development</td>
<td>Lhx1</td>
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<td>Nphs1</td>
</tr>
<tr>
<td>nephron tubule development</td>
<td>Nphs2</td>
</tr>
</tbody>
</table>

**Figure 4:** SRC Genes Participate in Nephrogenesis

**Figure 6:** SRC Genes Participate in Nephrogenesis