Pooled library construction Single-cell transcriptional

References:


Resolution due to encapsulation of cell doublets

b. the maximum number of cells that can be loaded in the 10X reaction without losing single-cell

cells per 10X reaction had a positive effect on the sgRNA library coverage, without adversely affecting

processes including the regulation of cell proliferation, differentiation, apoptosis, and immune

Tumor necrosis factor-alpha (TNFα) is a proinflammatory cytokine involved in various biological

Perturb-Seq is a single-cell functional genomics platform that combines pooled CRISPR (sgRNA) genetic

Fig. 3 Arrayed, individual CRISPR-sgRNA screen

Fig. 4 Effects of gene knockdown in TNFα-induced transcriptional activation of of IL2 and RELA genes, and a validation of the prior discovery in an independent and orthogonal screen.

Results (continued)

Table 5: Perturb-seq, pooled CRISPR/sgRNA library screens

<table>
<thead>
<tr>
<th>Cell Number</th>
<th>Cells/s/gRNA</th>
<th>Transctional Activation</th>
</tr>
</thead>
<tbody>
<tr>
<td>4,000</td>
<td>10X</td>
<td>Mock 98</td>
</tr>
<tr>
<td>7,500</td>
<td>0.25X</td>
<td>Mock 77</td>
</tr>
<tr>
<td>15,000</td>
<td>0.125X</td>
<td>Mock 77</td>
</tr>
</tbody>
</table>

Fig. 5: TNFα-induced transcriptional activation of of IL2 and RELA genes.

Discussion

• Treating HEK293-Cas9 cells transduced with individual sgRNA with TNFα (10 ng/ml; 24 hours) led to cell survival.

Results

Fig. 6: TNFα-mediated signaling pathway