**Aims and Objectives**

- Evaluate the effect of GLPG1690 on pro-fibrotic triggering by Transforming Growth Factor beta (TGFβ) in two different cellular models:
  - Normal Human Dermal Fibroblasts (NHDF cells)
  - Lung fibroblasts from an Idiopathic Pulmonary Fibrosis patient (IPF cells)
- Evaluate the activity of GLPG1690 and nintedanib on pro-fibrotic gene expression alone and in combination

**Methods**

- **D0**: NHDF or IPF cells were seeded in 96 well format (see Table 1 for details of seeding medium)
- **D1**: medium was refreshed to 1% charcoal stripped (CS) FBS medium for overnight incubation
- **D2**: cells were pre-treated with compound of interest and treated 1h later with TGFβ (see Table 1 for concentrations)
- **D3/D4**: Supernatant (SN) was harvested and frozen at -20° for later analysis
- **Dx**: SN was analyzed for levels of IL-6 and ET-1 by ELISA and for CTGF by MSD

**Table 1: Experimental details regarding the culturing and the treatment of the cells**

<table>
<thead>
<tr>
<th>Matrix</th>
<th>Seeding medium</th>
<th>Medium change</th>
<th>TGFβ trigger</th>
<th>Incubation time</th>
</tr>
</thead>
<tbody>
<tr>
<td>NHDF cells</td>
<td>FCS2 + 2% FBS</td>
<td>FCS2 + 1% CS FBS</td>
<td>3 ng/ml</td>
<td>24h</td>
</tr>
<tr>
<td>IPF cells</td>
<td>DMEM + 10% FBS</td>
<td>DMEM + 1% CS FBS</td>
<td>10 ng/ml</td>
<td>48h</td>
</tr>
</tbody>
</table>

**References**


**Conclusions**

Both GLPG1690 and nintedanib, the current standard of care, dose-dependently reduce the production of several TGFβ-induced pro-fibrotic mediators like ET-1, IL-6 and CTGF. The inhibitory effects can be seen in both dermal fibroblasts as well as IPF lung fibroblasts (Fig.3). Also pirfenidone dose-dependently inhibited the production of these cytokines (data not shown). GLPG1690 shows the most potent effect on the TGFβ induced ET-1 production with effects in the lower nM range. When suboptimal doses of GLPG1690 and nintedanib are combined, an additive inhibitory effect is seen on each of the three read-outs (Fig. 4). This suggests a promising outlook for combination therapy in the clinic.