Combined inhibition of PIM and PI3 kinases shows an enhanced efficacy in a number of solid tumour cell lines

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Oncogenicity in a parallel with regulation of cell cycle and proliferation. Over-expression of PIM kinases is associated with oncogenicity in a number of haematological and solid tumours.

Recent evidence suggests that PIM expression is an important parallel pathway to the AKT/ mTOR pathway. Much of the work up to now with PIM has focused on haematological malignancies. This study looked at the effect of combining PIM kinase inhibition with PI3K and PI3K/mTOR inhibition. We have characterized these effects in a range of solid tumour cell lines.

Identification of dual PIM/PI3K and triple PIM/PI3K/mTOR inhibitors

An SAR program was designed to balance the dual (PIM, PI3K) or triple (PIM, PI3K, mTOR) activities and to optimize the drug like properties of the compounds. As a result of this exploration compounds AZD1208 (pan-PIM/PI3K) were identified.

Antiproliferative activity of pan-PIM inhibitors (GDC-0941, PIMi) alone and in combination with BL-202 in colorectal and lung tumour lines.

Table 1: Activities of Inflection Biosciences Ltd and comparator compounds at target kinases

<table>
<thead>
<tr>
<th>Compound</th>
<th>PKI-111801</th>
<th>PD0325901</th>
<th>PD180414</th>
<th>PD167347</th>
<th>PD167347</th>
<th>GDC-0941</th>
<th>AZD1208</th>
<th>BL-202</th>
</tr>
</thead>
<tbody>
<tr>
<td>IC50 (nM)</td>
<td>3.9</td>
<td>0.4</td>
<td>1.2</td>
<td>1.2</td>
<td>3.9</td>
<td>0.4</td>
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Conclusions

Combined inhibition of PIM and PI3 kinases has a synergistic effect on cell proliferation in a range of solid tumour cell lines. This synergistic effect is evident with combinations of molecules that act as selective PIM and PI3 kinase inhibitors and with molecules specifically designed to combine both activities.

Targeting PIM/PI3K activities in the same molecules appears to produce a more potent effect than targeting them with separate agents.

Both AZD1208 (PIMi) and PIMi with PI3K inhibitors alone also induced greater and prolonged antiproliferative activity than PIMi and PI3K selective inhibitors alone. This effect has been shown previously to be correlated with higher induction of apoptosis and strong down-regulation of PI3K, PDK1, mTOR pathway.

Compounds from the PIM/PI3 series show excellent PK and have showed efficacy in vivo. Inflection is currently selecting its candidate for further development.

Data Summary

Table 2: Summary of efficacy of compounds on cell viability in a range of solid tumour lines

<table>
<thead>
<tr>
<th>Cell line</th>
<th>PIMi</th>
<th>PI3Ki</th>
<th>Dual</th>
</tr>
</thead>
<tbody>
<tr>
<td>HCT116</td>
<td>0.12</td>
<td>0.12</td>
<td>0.07</td>
</tr>
<tr>
<td>PC3</td>
<td>0.12</td>
<td>0.12</td>
<td>0.07</td>
</tr>
</tbody>
</table>

Summary statistics and error bars indicate the mean and standard deviation of the triplicate measurements. "n.s." indicates a non-significant effect. *p < 0.05, **p < 0.01, ***p < 0.001. Additional experiments were performed the effect of combinations on solid tumour xenografts in vivo.

C50 PIM2

C50 PIM2

C50 PIM2

C50 PIM2

C50 PIM2

C50 PIM2