**Introduction**

The HER family consists of four tyrosine kinase receptors designated as HER1, HER2, HER3 and HER4. Although HER3 has little or no kinase activity, it plays an important role in cancer since 1) it heterodimerizes with HER2, 2) it is the primary link to the PI3K/AKT axis and 3) it can mediate resistance to HER1 and HER2-targeted therapeutics.

We and our collaborators have demonstrated previously that a HER3 mRNA locked nucleic acid (LNA)–based antisense antagonist, designated EZN-3920, is able to down regulate HER3, resulting in tumor activity. The aim of this study was to evaluate the effect of EZN-3920 in combination with tyrosine kinase inhibitors such as gefitinib and lapatinib as well as a specific PI3KCA antisense molecule. The studies demonstrate that these combinations are superior to either agent alone, confirming the important role of HER3 and downstream PI3K/Akt signaling in the survival of tumors.

**Methods**

In vitro evaluation

The tumor cells were treated with LNA-oligonudleotides (LNA-ON) without lipofection. Cell proliferation (MTS), mRNA (qRT-PCR), and protein (western blot) examinations were performed 3–5 days after drug incubation. A control oligonucleotide (EZN-SCR) served as a negative control.

**Why Use LNA Technology?**

**HER3 and PI3K/AKT Signaling**

**EZN-3920 is potent in breast and lung cancer cell lines**

<table>
<thead>
<tr>
<th>Tumor Type</th>
<th>Cell line</th>
<th>HER3 expression</th>
<th>HER3 Knockdown %</th>
<th>LNA mRNA</th>
<th>HER3 protein µg</th>
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</thead>
<tbody>
<tr>
<td>Breast</td>
<td>BT474M1</td>
<td><strong>+</strong>/++/+++</td>
<td>+++/+++</td>
<td>EZN</td>
<td>+++/+++</td>
</tr>
<tr>
<td>Breast</td>
<td>HCC827</td>
<td>++/+++</td>
<td>+++/+++</td>
<td>EZN</td>
<td>+++/+++</td>
</tr>
</tbody>
</table>

**EZN-3920 blocks lipatinib-induced HER3 expression in vitro**

**EZN-3920 down-modulates HER3 and PI3K axis signaling**

**EZN-3920 specifically and effectively down modulates HER3 mRNA**

**EZN-3920 specifically potentiates the growth inhibitory effects of lapatinib in vitro**

**Combination of EZN-3920 with gefitinib in HCC827 NSCLC xenograft model. Mice were administered 30 mg/kg EZN-3920 (i.p.) (n = 8 per group) and 10 mg/kg gefitinib (q(3,5,8), p.s.) p<0.05 vs. saline group.**

**Combination of EZN-3920 with lapatinib in HCC827 breast cancer model. Mice were administered 30 mg/kg EZN-3920 (i.p.) (n = 8 per group) and 100 mg/kg lapatinib (q(3,5,8), i.p.) p<0.05 vs. saline group.**

**References**