From 34-mer peptide to small molecule: Using fields to find new actives for a GPCR receptor from the natural peptide ligand

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Abstract

Cresset consultants were asked to identify novel active compounds for a class-II GPCR receptor. There were no known small molecule inhibitors, so the only information available was the natural ligand, a 34-residue peptide. From sparse experimental data, four areas of the native ligand were identified as major binding regions. The field patterns of these regions were determined and used as input to a Blaze virtual screen. A 30% hit rate was achieved, with the hits all being drug like small molecules.

Introduction

The PTH-1 receptor is a member of the class-II (or class B) G-protein coupled receptor (GPCR) subfamily, which includes glucagon, secretin and vasoactive intestinal peptide receptors. No definitive picture of the details of the binding of parathyroid hormone (PTH) with its receptor could be derived at the time of the project – there were no small-molecule inhibitors known and no crystallographic data.

Although PTH is secreted as an 84-amino acid peptide, the first 34 residues have been shown to be fully active. There was reasonable evidence that the hormone remained as a helix when binding. Like other members of the class-II GPCRs, the signal transduction from the PTH-1 receptor can occur via both the cAMP and inositol triphosphate/intracellular calcium second messenger pathways. There is considerable homology with the related VIP (VPAC) receptor agonist family.

A biotech company asked Cresset to find new compounds that would be active at this site.

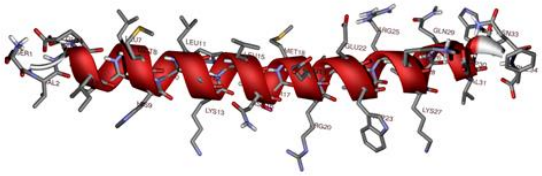

Figure 1. Structure of PTH.

Method

The first step was to identify the binding regions of the hormone.

From sparse NMR, X-ray and mutagenesis evidence, four areas could be tentatively identified as major binding regions (opposite). The first comprised four residues of the N-terminal putative agonist region along with two 4-mers and a 6-mer identified across the first 14 residues from the N-terminal of PTH.

Chosen sequence	Comment
1 SVSE	May be the agonist region
2 RVEW	Suspected binding region
3 WLRKKL	Suspected extended binding region
4 VHNF	Suspected terminal binding region

Figure 2. Binding regions of PTH.

These regions were selected, likely side chain conformations determined and a calculation was performed using Cresset's XED force field to calculate the field patterns. The field patterns were used as input to Blaze to perform a search of Cresset's field database of commercially available compounds. Any compounds whose field patterns would fit the four identified binding regions were returned as hits.

Results

The top 500 ranked results from each of the searches were visually distilled down to a total of 50 by Cresset scientists and submitted to the client. They subsequently purchased 41 compounds for testing – the assay had a low throughput. Although many of the purchases showed probable activity at PTH, the initial assays proved capricious and quantitative results could not be trusted. However, on a related VPAC test, 12 were found to be active below 30µM and six of these were sub-10µM. All were in the 360-500 molecular weight range.

All the active compounds in the final list have been retained as proprietary property by the client.

Conclusion

Cresset's field technology made it possible to find active small molecules for a class II GPCR from analysis of a large natural peptide ligand.

Multiple different series were identified, with several of these falling comfortably inside drug like physicochemical property space. This demonstrates Cresset's capability to locate active small molecules against extremely difficult targets quickly and efficiently.