Two-component systems (TCSs) are the main signal transduction pathways used by bacteria to regulate a variety of processes including bacterial development, metabolism, virulence mechanisms, and resistance to antibiotics. TCSs consist of a homodimeric membrane-bound sensor enzyme, a histidine kinase (HK), and a cognate effector, a response regulator (RR). A high degree of sequence conservation in the catalytic domain (CA) of HKs especially in the ATP-binding pocket and their essential role in bacterial signal transduction make an attractive target in broad-spectrum anti-virulence. It has been theorized that eliminating bacterial virulence would be a promising alternative to current antibiotic strategy. Detailed mechanistic and structural insights into ligand-domain binding in HKs is urgently needed as the development of potent inhibitors that modulate bacterial signal transduction could lead to a new mechanism for treatment of infectious diseases. Structure-activity relationship (SAR) studies have been performed with compounds that target the CA domain that our group identified through a small molecule high-throughput screening (HTS) campaign against HK853 (Thermotoga maritima). The most potent compounds discovered in these studies possess IC$_{50}$ values in the low μM range, while also exhibiting activity against two additional HKs: VicK (Streptococcus pneumoniae) and CheA (Escherichia coli). These compounds have shown to be effective in whole cells, with anti-virulence activity against Pseudomonas aeruginosa, MRSA, Salmonella and Vibrio cholerae. Docking studies suggest the preference in potent of an exocyclic nitrogen for hydrogen bonding in the HK active site through a conserved aspartate residue confirmed by SAR studies. A recent screen of eukaryotic kinase inhibitors against HK853 showed minimal activity except for one inhibitor that presents hydrogen bonding to three conserved residues in the active site in a similar way than previously reported inhibitors.