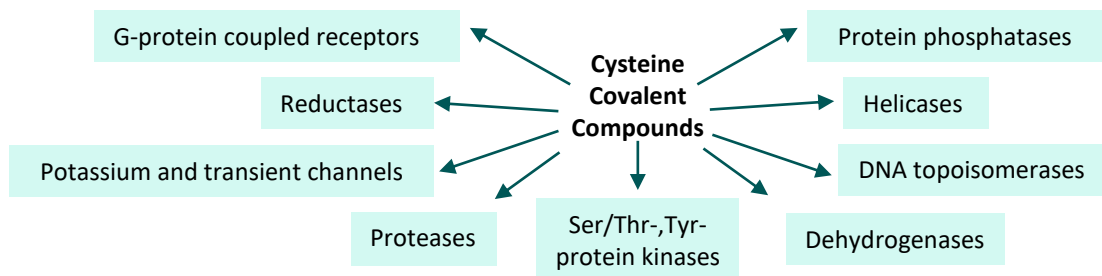


DEVELOPMENT OF COVALENT FRAGMENTS FOR TARGETING CYSTEINE RESIDUES IN DRUG DISCOVERY

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Targeted covalent inhibitors and chemical probes have become integral parts of drug discovery approaches. In recent years, the number of drug candidates with a covalent mechanism of action progressing through clinical trials or being approved by the FDA has grown significantly; around 30% of the marketed drugs are covalent inhibitors. The sustained duration of covalent inhibition provides several potential advantages, including improved biochemical efficiency of target disruption, potential prevention of emergence of drug resistance due to continuous target suppression [PMID: 24314671].

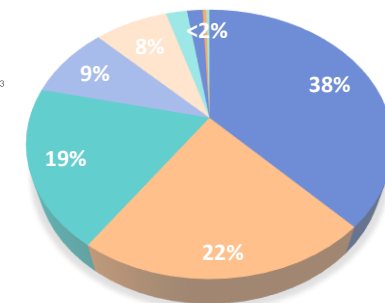
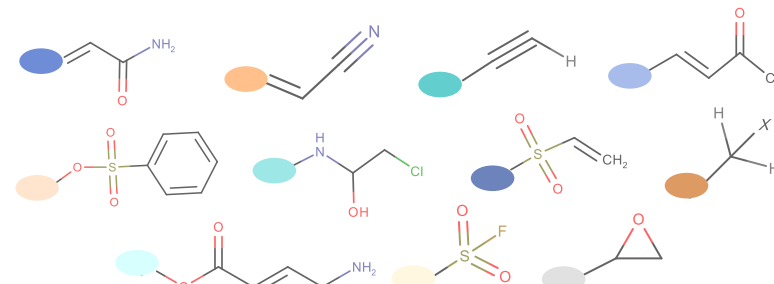


Most popular targets with PubMed BioAssay data less 1mM. Found 706 compounds for 373 proteins

The design of selective covalent, irreversible inhibitors is conceptually very attractive, but hard to achieve in practice because striking the right balance of molecular properties responsible for reactivity and selectivity presents a big challenge [PMID: 32298798, 20640225, 23438744]. In addition to general aspects of the FBDD paradigm, the design of electrophilic fragments requires taking into consideration their reactivity, reversibility, stability, synthetic accessibility and size of the electrophilic functionality [PMID: 32298798].

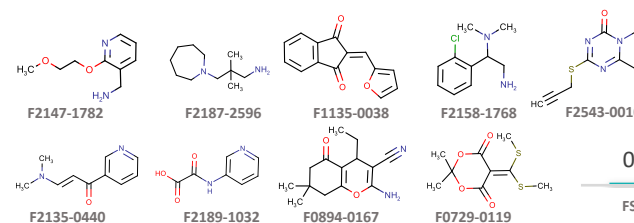


The zoomed view of the structure substrate-binding pocket in complex with random covalent fragments

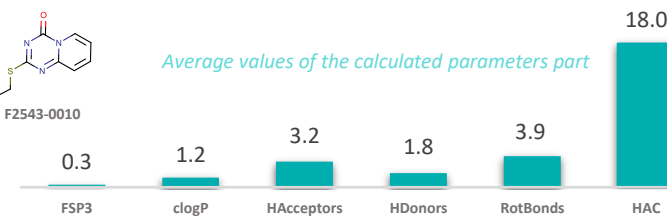


Covalent warheads distribution for compounds in the Cysteine-focused Covalent Inhibitor Library

In response to the growing interest in fragment-based drug discovery and its widespread use we have designed an exclusive collection of covalent inhibitor fragments. A preliminary set of 4,058 covalent fragments was created by selecting compounds with specific structural fragments (functional groups, warheads) that are known to form covalent bonds with amino acid residues in binding sites of targeted proteins, e.g., Lys, Cys, Ser, His and Tyr.



Average values of the calculated parameters part



Representative compounds from the Covalent Fragment Library

Functional group analysis was carried out and their covalent reactivity was confirmed with *in silico* alanine scanning mutagenesis. At the next stage molecules with highly reactive electrophilic and nucleophilic groups, as well as compounds with non-druglike cores, were discarded as non-selective covalent binders. This combined approach allowed us to reduce a previously selected set to analyze only 1,260 Cys-associated covalent compounds. However, the resulting set has not been made Ro5 compliant as in this case many small peptide-mimicking compounds would have been filtered out.

Applying the approach presented above, 157 non-patented (according to the SciFinder) compounds have been selected and prepared for further molecular docking study against a set of protein drug targets.