DISCOVERY OF A NAPHTHOL MOLECULAR SCAFFOLD WITH INHIBITORY ACTIVITY AGAINST COVID-19 MAIN PROTEASE.

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SARS-CoV-2 is a novel, rapid-spreading coronavirus responsible for a global pandemic of severe acute respiratory syndrome. To date, various viral proteins have been studied as possible drug targets, with SARS-CoV-2 main protease (Mpro) being one of the most attractive enzymes characterized by high druggability. Mpro proved to be an exceptional target for achieving clinically relevant SARS-CoV-2 inhibition with one drug already in therapeutics as well as for confrontation of related novel coronaviruses in the future.

In this study, we present the development of a multiple-step in silico and in vitro methodology that lead to the discovery of a small molecular weight naphthol scaffold which demonstrates inhibitory activity against Mpro. Initially a virtual screening was performed filtering the NCI repository (~260.000 molecules) by means of similarity and docking calculations. A known triarylpyridinone inhibitor was used as a query for identifying compounds of similar structure using both 3-dimensional and 2-dimensional shape- and pharmacophore-similarity methodologies. Consensus ranking was performed by the assistance of R programming language and the ~20.000 top ranked molecules were further explored with docking simulations using GLIDE (Schrödinger software). Sampling of the chemical space was improved by diversity analysis of the hits and 54 molecules were selected. Subsequently, expression and purification of the enzyme was caried out. Experimental evaluation was performed using two orthogonal high-throughput in vitro screening assays, namely Differential Scanning Fluorimetry (DSF) and a FRET based enzymatic assay. A hit compound was discovered of low μM IC50 and confirmed by Isothermal Titration Calorimetry (ITC). Subsequently, a list of 30 available molecular structures, similar to the active compound were also tested and a second hit was identified. Theoretical physicochemical properties and ADMET profile were also defined in order to assist further development in the hit to lead process.