A target and two drugs for SARS-CoV-2 found by paralog search

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# Abstract

Using a paralog search pipeline, the author searched the ChEMBL 25 database, screening targets in it against the SARS-CoV-2 genome finding a target that has an identical target sequence. The the target, RNA polymerase, was found to have 100% identity with a gene in the viral genome of SARS-CoV-2.

Three known drugs in the ChEMBL 25 database are associated with the target that was identified. Two of those drugs showed high binding affinity in docking simulations, validating them as promising drug candidates to treat SARS-CoV-2.

# Introduction

(Fig 1.)

An analysis pipeline searches the **chembl\_25** version of ChEMBL’s database for targets and drugs using viral genomic information from Genbank.

SARS-CoV-2, also known as COVID-19, is a virus that causes flu like symptoms including respiratory distress, in many cases requiring respirators to maintain oxygenation in patients. It is highly contagious, and is currently causing pandemic infection, with a fatality rate estimated between 2% and 3% [1]. Persons over 60 have may have much higher fatality rates [2].

A target repurposing strategy might provide drugs more quickly and cheaply than creating new drugs and finding new targets [3]. This strategy could produce treatments to ameliorate the disease until a vaccine becomes available, or in addition to vaccines.

**DISCLAIMER:** The workflow described in this paper was initially conceived as a way to rapidly and cheaply identify targets and drugs for neglected tropical diseases [3, 4]. **This is not a definitive, clinicly approved theraputic option.**  **These methods are intended to identify targets and drugs for further study.** While some dosage and toxicity data are available for the drugs in question from their original usage, the the therapeutic effect of these drugs against SARS-CoV-2 has not been studied.

The workflow uses targets and drugs in databases that are downloadable from the internet, genomic data (in the form of amino acid sequences, (also available from internet sources) and a software stack that can run on a PC, or on a VM hosted on a PC. Specifically, the data and software stack can run on a PC running Windows 10, Centos 7 Linux running in a Virtualbox VM, and a PostgreSQL database running under Centos 7 [5-9]. All software needed are available for free, and run on a PC with at least a 4 core 64 bit Intel compatible CPU, and having 8 GB of ram.

ChEMBL provides a downloadable database that includes drug targets and drug information for those targets, as well as amino acid sequences of the protein targets [10]. Drug targets tend to be proteins that are important enough to the organism to which they belong that they tend to be conserved [11]. If we can find a protein sequence in a disease organism that is sufficiently similar to a known target, the protein may be a promising target in that novel organism, and drugs used against that target in the original organism may be successfully used against the new pathogen.

Genbank provides a nucleotide sequence database containing genomes of many organisms, including the SARS-CoV-2 virus [12].

EMBOSS tools provide the **getorf** utility that finds the genes in the SARS-CoV-2 genome[13]. ORFs (Open Reading Frames) are amino acid sequences, including protein coding genes, that we wish to screen for sequence similarity to targets from ChEMBL.

An ETL (Extract Translate and Load) process downloads the drug target sequences into a file that can be queried with sequence similarity searching software such as **jackhmmer [14].**

**jackhmmer** produced similarity reports and scoring summaries for the ORFs. ETL scripts parsed these results, and uploaded them to supplementary tables in the PostgreSQL database that contained the **chembl\_25** database schema that had been downloaded.

Once the summary information is in the database, statistical methods such as kmeans established a similarity threshold to select the most promising targets and their drugs [15-17].

Swissdock, an opensource docking server, was used to evaluate the identified drugs for docking affinity to the target(s) found to validate whether these drugs might be effective [18].

# Materials and methods

(Fig 2 .) Target and drug analytical workflow.

Before executing the workflow, VirtualBox and the Centos 7 Linux image are installed and executed on the work PC. An empty PostgreSQL database is installed, and a single user named “user” created [5-7, 9].

The PostgreSQL dump archive of the ChEMBL version 25 database is downloaded, decompressed, and restored in the Centos 7 VM.

EMBOSS tools are installed [13].

The genome of SARS-CoV-2 (The COVID-19 virus) was downloaded from Genbank via NCBI’s website as MN908947.3.FASTA. [12].

ORFs were translated using EMBOSS tools [13]. The orfs were translated rather than using curated protein sequences because the translation tool provides the amino acid FASTA sequences in a form convenient for querying. While many ORFs may not be actual proteins, the query that uses these ORFs will eliminate them from consideration.

This command creates a file containing all the open reading frames (ORFs) found and translates the nucleotide sequences into amino acid sequences:

**getorf MN908947.3.FASTA**

This creates file **mn908947.orf**, which contains all the ORFs found for the .FASTA file.

Commands run in R Studio quantify how many ORFs are contained:

|  |
| --- |
| > aa=read.table(file="mn908947.orf",header = FALSE, sep='~', stringsAsFactors = FALSE)  > aa=aa[!is.na(aa[,1]),] # filter out NA  > aa=data.frame(lines=aa, stringsAsFactors = FALSE)  > orf\_headers=aa[substr(aa[,1],1,1)=='>' ,]  > length(orf\_headers)  [1] 1572 |
|  |
| |  | | --- | |  |   1572 ORFs were found. |

The set of target sequences comes from the **ChEMBL\_25** PostgreSQL database and was downloaded by a *psql* script **chembl\_25\_targets.sql** as file **chembl\_targets.txt**.

These targets are converted by a Perl script (**split\_to\_fasta.pl**) creating file **component\_sequences.fa**.

Using **jackhmmer** to provide similarity reports and sequence alignments, a pipeline imported scores showing sequence and structural similarity [14]. This process created files **orf.hmm.txt** and **orf.summary**.

The Perl script **extract\_hmm\_summary.pl** reads the **orf.hmm.txt** file and creates the file **hmm\_stats.tx**t.

From the psql database query prompt, the data were imported into the **chembl\_25** database:

[postgres@osboxes /home/osboxes/genomes] **psql -U postgres -d chembl\_25**

psql (9.2.24)

Type "help" for help.

chembl\_25=# **\i import\_hmmer\_statistics.sql**

TRUNCATE TABLE

INSERT 0 49

chembl\_25=# **update hmmer\_statistics set tax\_id=2697049, organism='SARS-CoV-2’ where tax\_id is null;**

The UPDATE statement is necessary to differentiate the statistics from those of other uploaded genomes.

# Results and discussion

(Fig 3.)

# Conclusions

# References

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# Supporting information

## **S1 Fig. An analysis pipeline searches the chembl\_25 version of ChEMBL’s database for targets and drugs using viral genomic information from Genbank.**

## **S2 Fig. Target and drug analytical workflow.**

## MN908947.3.FASTA.

The nucleotide FASTA formatted genome sequence of SARS-CoV-2.

## chembl\_25\_targets.sql

psql script to download chembl\_25 target sequences.

## chembl\_targets.txt

Downloaded target sequences.

## split\_to\_fasta.pl

Converrts a text file containing sequences to .FASTA format file.

## component\_sequences.fa

Searchable target component file.

## mn908947.orf

ORFs from SARS-CoV-2 genome.

## orf.hmm.txt

A report containing scores and alignments between the ORFs and targets.

ORFs having insufficient similarity have a record that says “[No hits detected that satisfy reporting thresholds].”

## orf.summary

A tab delimited file containing records with statistics of significant matches for ORFs and targets.

## extract\_hmm\_summary.pl

Extracts summary statistics from **orf.hmm.txt** and writes them to file hmm\_stats.txt.

## hmm\_stats.txt

Uploadable statistics file.

## import\_hmmer\_statistics.sql

Uploads statistics file to a work table, and then to **hmm\_statistics** table.

## **S3 Fig. Query joins connecting targets, sequences, and drugs. Tables with blue backgrounds are supplementary tables populated by this workflow.**

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