Check for updates

METHOD ARTICLE

A signaling pathway-driven bioinformatics pipeline for predicting therapeutics against emerging infectious diseases [version 1; peer review: 1 approved, 1 approved with reservations]

Tiana M. Scott , Sam Jensen, Brett E. Pickett

Microbiology and Molecular Biology, Brigham Young University, Provo, Utah, 84602, USA

V1 First published: 29 Apr 2021, 10:330
<https://doi.org/10.12688/f1000research.52412.1>

Latest published: 20 Aug 2021, 10:330
<https://doi.org/10.12688/f1000research.52412.2>

Abstract

Background: Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), the etiological agent of coronavirus disease-2019 (COVID-19), is a novel Betacoronavirus that was first reported in Wuhan, China in December of 2019. The virus has since caused a worldwide pandemic that highlights the need to quickly identify potential prophylactic or therapeutic treatments that can reduce the signs, symptoms, and/or spread of disease when dealing with a novel infectious agent. To combat this problem, we constructed a computational pipeline that uniquely combines existing tools to predict drugs and biologics that could be repurposed to combat an emerging pathogen.

Methods: Our workflow analyzes RNA-sequencing data to determine differentially expressed genes, enriched Gene Ontology (GO) terms, and dysregulated pathways in infected cells, which can then be used to identify US Food and Drug Administration (FDA)-approved drugs that target human proteins within these pathways. We used this pipeline to perform a meta-analysis of RNA-seq data from cells infected with three Betacoronavirus species including severe acute respiratory syndrome coronavirus (SARS-CoV; SARS), Middle East respiratory syndrome coronavirus (MERS-CoV; MERS), and SARS-CoV-2, as well as respiratory syncytial virus and influenza A virus to identify therapeutics that could be used to treat COVID-19.

Results: This analysis identified twelve existing drugs, most of which already have FDA-approval, that are predicted to counter the effects of SARS-CoV-2 infection. These results were cross-referenced with interventional clinical trials and other studies in the literature to identify drugs on our list that had previously been identified or used as treatments for COVID-19 including canakinumab, anakinra, tocilizumab, sarilumab, and baricitinib.

Conclusions: While the results reported here are specific to Betacoronaviruses, such as SARS-CoV-2, our bioinformatics pipeline

Open Peer Review

Approval Status ? ✓ ✓

	1	2	3
version 2			
(revision)			✓
20 Aug 2021			view
version 1	?	✓	
29 Apr 2021	view	view	

1. **José Pedro Cerón-Carrasco** , UCAM
Catholic University of Murcia, Murcia, Spain
2. **Babatunde Joseph Oso** , McPherson
University, Seriki Sotayo, Nigeria
3. **Soumya Lipsa Rath** , McPherson
University, Seriki Sotayo, Nigeria

Any reports and responses or comments on the article can be found at the end of the article.

can be used to quickly identify candidate therapeutics for future emerging infectious diseases.

Keywords

bioinformatics, repurposing, coronavirus, SARS-CoV-2, COVID-19, virus, infection, therapeutic, target

This article is included in the **Pathogens** gateway.

This article is included in the **Bioinformatics** gateway.

This article is included in the **Emerging Diseases and Outbreaks** gateway.

This article is included in the **Cell & Molecular Biology** gateway.

This article is included in the **Coronavirus** collection.

Corresponding author: Brett E. Pickett (brett_pickett@byu.edu)

Author roles: **Scott TM:** Formal Analysis, Investigation, Software, Writing – Original Draft Preparation, Writing – Review & Editing; **Jensen S:** Formal Analysis, Writing – Original Draft Preparation; **Pickett BE:** Conceptualization, Formal Analysis, Investigation, Methodology, Software, Supervision, Writing – Original Draft Preparation, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

Grant information: This research was funded by a startup package from Brigham Young University.
The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Copyright: © 2021 Scott TM *et al.* This is an open access article distributed under the terms of the **Creative Commons Attribution License**, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

How to cite this article: Scott TM, Jensen S and Pickett BE. **A signaling pathway-driven bioinformatics pipeline for predicting therapeutics against emerging infectious diseases [version 1; peer review: 1 approved, 1 approved with reservations]** F1000Research 2021, 10:330 <https://doi.org/10.12688/f1000research.52412.1>

First published: 29 Apr 2021, 10:330 <https://doi.org/10.12688/f1000research.52412.1>

Introduction

Coronaviruses consist of a lipid envelope that contains a single-stranded positive-sense RNA genome that is approximately 30 kilobases in length. Prior to 2019, six human coronavirus species had been discovered including *HCoV-229E*, *HCoV-NL63*, *HCoV-HKU1*, *HCoV-OC43*, *SARS-CoV*, and *MERS-CoV*. Four of these coronavirus species are endemic in humans and typically cause mild respiratory tract infections that present with cold-like symptoms but can cause more severe symptoms in immunocompromised individuals or infants^{1,2}. Two of these four endemic virus species are members of the *Alphacoronavirus* genus (*HCoV-229E* and *HCoV-NL63*), while the other two species are members of the *Betacoronavirus* genus (*HCoV-HKU1* and *HCoV-OC43*). The remaining two novel human coronavirus species discovered during this time are severe acute respiratory syndrome coronavirus (*SARS-CoV*; SARS) and Middle East respiratory syndrome coronavirus (*MERS-CoV*; MERS), which were emergent Betacoronaviruses responsible for epidemics in 2003 and 2012 respectively^{1,3}. Human coronaviruses generally emerge from other animal hosts such as bats or mice, and typically pass through an intermediate host (e.g. civet cats for SARS and dromedary camels for MERS) before infecting a human host^{1,3}.

In December 2019, a novel coronavirus was reported in Wuhan city, Hubei province, China and has since been named severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2). SARS-CoV-2 is highly similar to other SARS-like viruses that have been isolated from bats previously, especially BatCoV RaTG13 with which it shares 96.3% identity^{4,5}. Initial infections with SARS-CoV-2 were traced back to the Huanan Seafood Wholesale Market and likely infected humans via a pangolin intermediate⁶. The global COVID-19 pandemic, as of February 14, 2021, has resulted in over 108 million cases and over 2.3 million deaths worldwide. Approximately 7.6 million of the cases and over 500,000 deaths have occurred in the United States of America⁷.

Common symptoms of COVID-19 include fever, dry cough, dyspnea, sore throat, myalgia, fatigue, and in some cases diarrhea^{4,5,8–11}. SARS-CoV-2 is spread through aerosols, droplets, direct contact between people, and fomites^{4,12}. Other studies suggest a fecal-oral transmission route is possible due to the presence of SARS-CoV-2 in stool samples of infected patients^{12,13}. SARS-CoV-2 infects cells by binding to the membrane-associated Angiotensin-converting enzyme 2 (ACE2) receptor, which generally plays a role in the renin-angiotensin-aldosterone system to regulate blood pressure and fluid balance in the body^{14,15}. ACE2 receptors are known to be expressed in lung, renal, cardiac, vascular, intestinal, and placental tissues^{14,15}.

Both the widespread effects of COVID-19 and the initial highly susceptible population emphasized the need to identify potential drug treatments for emerging diseases—particularly before vaccines become available. The aim of many recent and ongoing clinical trials is to quantify the efficacy of various therapeutics for COVID-19¹⁶. Many vaccines are in various

stages of preclinical (at least 139) or clinical (at least 25) development¹⁷, as well as some that have gained emergency use authorization by the US Food and Drug Administration (FDA).

The COVID-19 pandemic, caused by the SARS-CoV-2 virus, has highlighted the need to quickly and accurately identify therapeutics that can be repurposed to combat the signs, symptoms, and spread of disease. One method for predicting potential therapeutics is to identify host pathways that are dysregulated by infections and then find existing drugs that target those pathways. Programs to perform this analysis include DrugThatGene and GPSnet, which have both been used to identify potential drugs to target cancers^{18,19}. The aim of this study was to construct a bioinformatics workflow that uniquely combines existing tools, databases, and programming libraries with custom scripts to predict potential human therapeutic targets for multiple members of the *Betacoronavirus* genus, which includes the *SARS-CoV*, *SARS-CoV-2*, and *MERS-CoV* species. Our unique combination of tools consists of a consistent and robust RNA-seq preprocessing workflow as well as an intracellular signaling pathway perturbation method that enables us to account for the role of protein-protein interaction networks instead of merely enriching for differentially expressed genes. We then applied this workflow to a use case involving a meta-analysis of coronaviruses and other respiratory viruses. This workflow first performs an analysis of human genes and significant signaling pathways that play a role in infection and pathogenesis. The pathway information is then used to predict relevant human drug targets and the associated small molecules or biologics that bind to the target of interest.

The rationale for identifying drug targets from multiple intersecting signaling pathways is based on the theory that a protein which participates in multiple affected pathways during viral infection has a higher likelihood of playing an important role in viral pathogenesis and replication. Targeting one or more of these proteins that act as “key hubs” with a therapeutic would therefore have a higher chance of reducing viral processes and the ensuing disease. Similarly, host proteins that participate in multiple pathways across various related viruses likely represent an evolutionarily conserved host-pathogen interaction that can be therapeutically modulated. We expect virus resistance to these host-based drugs to be relatively infrequent since they target relevant host processes. Our workflow could therefore be applicable not only to improving therapeutic treatment during infection with existing or emerging coronaviruses, such as SARS-CoV-2, but to rapidly identifying potential treatments for pathogens that may emerge in the future.

Methods

Included datasets

A search of the Gene Expression Omnibus (GEO) database, hosted at the National Center for Biotechnology Information (NCBI; <https://www.ncbi.nlm.nih.gov/geo/>)²⁰, was performed in mid-2020 to find RNA-sequencing datasets for various

viruses including “MERS”, “SARS”, and “coronavirus”. The corresponding sequencing data for four GEO series were retrieved from the Sequence Read Archive (SRA; <https://www.ncbi.nlm.nih.gov/sra>) at NCBI²¹: GSE122876, GSE56192, GSE147507, GSE139516^{22–24}. These datasets were generated from cell cultures or patients infected with one of: respiratory syncytial virus (RSV), influenza A virus (IAV), MERS, SARS, and SARS-CoV-2. The SRA files were downloaded and converted to fastq format using version 2.10.1 of the NCBI sratools software package (<https://github.com/ncbi/sra-tools>).

Differential expression analysis

The Automated Reproducible MOdular Workflow for Preprocessing and Differential Analysis of RNA-seq Data (ARMOR) workflow was used to preprocess and analyze the fastq files against the Ensembl reference transcriptome for *Homo sapiens* build GRCh38, release 98 [GCA_000001405.15]²⁵. Briefly, this automated snakemake-based workflow performs quality control of the reads with fastQC (www.bioinformatics.babraham.ac.uk/projects/fastqc/), trims the adapters and poor-quality regions with TrimGalore! (https://www.bioinformatics.babraham.ac.uk/projects/trim_galore/), maps & quantifies reads to the human transcriptome using Salmon²⁶, performs differential expression using edgeR²⁷, identifies differential transcript usage with DRIMseq²⁸, and enriches on Gene Ontology terms²⁹ and Hallmark gene sets³⁰ using the Camera algorithm to adjust for inter-gene correlation³¹. Differential expression was performed by calculating log2 fold-change and the associated p-values from infected samples versus mock-infected samples. The significance threshold for differential gene expression and Gene Ontology enrichment was defined as an false discovery rate (FDR)-corrected p-value < 0.05. Gene Ontology enrichment results from all datasets were then combined to identify shared terms.

Pathway enrichment analysis

Once the differentially expressed gene lists were constructed, the Ensembl identifiers for these human gene lists were mapped to the corresponding NCBI Entrez Gene identifiers using BioMart and Bioconductor^{32,33} prior to pathway enrichment using Signaling Pathway Impact Analysis (SPIA)³⁴. Briefly, this pathway analysis combines robust statistics and bootstrapping to identify enriched pathways from lists of genes by incorporating the directionality of expression. Public pathway databases in the Graphite package in R Bioconductor were used by SPIA for enrichment with those surpassing a Bonferroni-corrected p-value < 0.05 being retained³⁵. These databases include: KEGG³⁶, Reactome³⁷, Panther³⁸, NCI³⁹, and BioCarta. All pathways from each dataset were then compared to identify those that were shared among the various datasets, as well as those that were unique only to infection with SARS-CoV-2.

Prediction of relevant drug targets

The output from the SPIA pathway enrichment were then used as input for a custom bioinformatics pipeline to identify existing drugs and biologics that could be repurposed to reduce signs, symptoms, and/or replication of coronaviruses. Specifically, this pipeline iteratively 1) retrieves the genes that participate in each statistically significant signaling pathway,

2) maps the gene identifiers to the corresponding UniProt protein identifier⁴⁰, 3) searches the opentargets resource (www.opentargets.org)⁴¹ to identify known drug targets and therapeutics, and 4) generates a table with various attributes of the target and small molecule/biologic treatment. These data were then integrated with the pathway comparisons across the various datasets and analyzed to determine what drugs would affect the pathways dysregulated in the greatest number of viruses and could be used as potential therapeutics for SARS-CoV-2 infection. The results were ranked by how many times a given protein target appeared in the results across the relevant virus taxa. The top ranked small molecules/biologic treatments were analyzed to determine which were predicted to reverse the effects of the viral infection on a given pathway. Manual review of high-ranking hits was then performed to determine the existing indication(s) for each treatment, followed by a literature search to determine which, if any, of the therapeutics identified had previously been used or considered for the treatment of COVID-19. Code for this workflow can be found on GitHub: <https://github.com/bpickett/Pathway2Targets>.

Results

Differential expression analysis

Each of the algorithmic components of our computational workflow was dependent on first calculating differentially expressed (DE) genes for each of the datasets. We consequently began by using the same computational workflow to generate the DE genes from the raw transcriptomic data for human cells infected with one of: RSV, IAV, MERS, SARS, and SARS-CoV-2. Each set of results was specific to the comparison that was performed (e.g. mock-infected vs. infected) and included both log2 fold-change values and FDR-corrected p-values. We then integrated and compared the differential expression results across all virus comparisons to facilitate downstream comparisons.

We began by identifying genes that were either up- or down-regulated across a large number of studies. Specifically, we calculated the number of DE genes (FDR-corrected p-value < 0.05) that significantly changed during infection with influenza A (4205 genes), RSV (3661 genes), MERS (range across studies: 1222-13006 genes), SARS-CoV (range across studies: 2130-5557 genes), or SARS-CoV-2 (range across studies: 427-6933 genes). Out of the 15 studies evaluated, we observed 11 genes that displayed increased expression across 10-13 study comparisons including: TRIM25, C3, NCOA7, PTAFR, TNFAIP3, EIF2AK2, HELZ2, HBEGF, NFKB2, REL and VEGFC. We found an additional 18 genes that were upregulated across 9 of the 15 comparisons including: IFIT2, EGFR, and FOSL1. In contrast, we identified 14 genes that showed decreased expression across 11-12 comparisons including: EIF4H, APH1A, NME2, SEC61B, TUBB4B, CHCHD2, TUBA1C, TP53I3, H2AFZ, PEBP1, HOXB6, TPM2, CBR1, and SIVA1.

We next analyzed the DE lists to identify significant genes that were shared across infections involving only the Betacoronaviruses including SARS-CoV, MERS-CoV, and SARS-CoV-2. This analysis revealed 12 genes that were

upregulated in at least nine of thirteen comparisons including: TRIM25, REL, DNASE1, C3, NCOA7, PTAFR, EIF2AK2, HELZ2, NFKB2, ZNF385C, ZC3H12A, and OR7E122P. In contrast, the 10 genes that were found to be downregulated in ten of the thirteen comparisons involving any of the Betacoronaviruses included: CHCHD2, SMIM3, EIF4H, NME2, CBR1, SPARC, DSTN, CDC123, TIMM17A, and PDAP1.

We then performed a similar analysis to identify the five statistically significant DE genes (FDR-corrected p-value < 0.05) that displayed the highest and lowest average fold-change values during infection across all comparisons of SARS-CoV-2, Betacoronaviruses, or all viruses (Figure 1). This analysis showed that a subset of the genes such as CXCL11, which is induced by interferon and is involved in T-cell signaling, displayed similar fold-change values across multiple comparisons. This finding was somewhat expected since certain genes involved in the host innate immune response are expected to be modulated during virus infection. However, we also observed that genes such as MUC3A, PCSK5, MRC1, and CLEC3B displayed somewhat different average fold-change values across the included comparisons. This observation was also expected given the diverse virus replication requirements and the resulting host intracellular transcriptional response that occurs during infections with human viral pathogens.

Enrichment of annotated terms

Given the number of DE genes involved in the analysis, manual interpretation of the results would be intractable. We

therefore performed an analysis to determine which Gene Ontology (GO) terms and Hallmark gene sets were specifically enriched to better understand which biological processes and molecular functions were represented by the genes in each of the DE gene lists. Overall, we observed a superset of 580 terms that were enriched in any of the included viral infections. The enrichment results for each taxon of virus were then reviewed to identify annotated terms that were shared between multiple types of viral infections, shared amongst coronaviruses, or specific to SARS-CoV-2.

We found several notable terms that were shared among the results obtained from infection of multiple unrelated taxa of viruses. These terms referred to functions such as the host interferon response, regulation of virus response, chemokine activity, and immune cell migration that were positively enriched in both the RSV infection of A549 cells and the SARS-CoV-2 infection of either A549 or NHBE cells. As expected, the statistical significance of these terms supports the important role that the human immune system plays in response to virus infection, which is an expected result and validates the upstream DE gene analysis.

Our method identified no significant GO terms that were shared across all Betacoronavirus comparisons. Interestingly, we found that each coronavirus species had its own set of uniquely enriched terms. In MERS-CoV, “negative regulation of inclusion body assembly” was positively enriched in four of the seven comparisons, while “aggrephagy” and “regulation

Figure 1. Heatmap of human transcriptional biomarkers associated with infection by various virus taxa. The five genes with the highest and lowest log2 fold-change values for each virus comparison were identified and compared against expression in the other datasets. Squares in red or blue represent genes that are upregulated or downregulated (respectively) in the relevant comparison.

of nucleotide binding oligomerization domain containing 2 signaling pathway” were positively enriched in three of the seven comparisons. The SARS-CoV samples had very few significantly enriched annotation terms including the “signal peptidase complex” and “cyclin dependent protein serine threonine kinase activator activity” terms that were each negatively enriched in only one of the four SARS-CoV comparisons.

We also identified 290 terms that were uniquely present among the SARS-CoV-2 comparisons, although no significant terms were identified in comparisons involving either the human post-mortem biopsies or the infected Calu-3 cells. The shared significant terms that we identified in the NHBE- and A549-infected cells included “response to chemokine”, “anti-microbial humoral response”, and “humoral immune response.” Enriched terms in the NHBE-infected cells alluded to the role of the inflammatory response, interleukin-1, interleukin-6, neuroinflammation, arachidonic acid binding, and T-cell activation.

Identification of affected signaling pathways

We then wanted to calculate which intracellular signaling pathways were significantly modulated during viral infection, based on the DE genes identified above. The pathway information used for this analysis was obtained from public databases that manually curate the proteins that participate in conveying a signal from receptors on the cell membrane to transcription factors within the nucleus in order to respond to a stimulus.

We used the results of the differential gene expression analysis as input to the SPIA algorithm. The robust bootstrap-based

approach of this method identified 249 pathways that were significantly perturbed across at least one viral comparison. We subsequently analyzed the lists of significantly perturbed pathways for each comparison to determine those that were shared across virus taxa (e.g. influenza A, respiratory syncytial, MERS-CoV, SARS-CoV, and SARS-CoV-2), shared among coronaviruses, and unique to SARS-CoV-2. This analysis revealed a subset of the total number of pathways to be dysregulated across many of the viral taxa studied. These pathways included translation (affected in 12/17 comparisons and 4/5 viral taxa), cytokine-cytokine receptor interaction (modulated in 11/17 comparisons and 4/5 viral taxa), as well as rRNA processing (modulated in 10/17 comparisons and 3/5 viral taxa). We also observed that some of these pathways were predicted to be activated during infection with certain viral taxa in specific cell types, while inhibited in others.

We did not observe any signaling pathways that were significantly and consistently affected during infection by the individual coronavirus species examined. However, we did detect relevant pathways across multiple virus taxa. Interestingly, we found that the direction (i.e. activated or inhibited) was occasionally dependent on virus taxa or timepoint of infection. For example, the cytokine-cytokine receptor interaction pathway was predicted to be activated during infection with RSV, a subset of comparisons involving MERS-CoV, and many SARS-CoV-2 infections; while the same pathway was predicted to be inhibited during infection with SARS-CoV.

We predicted a total of 38 pathways that were uniquely affected during SARS-CoV-2 infection (Table 1). We noted that 35

Table 1. Intracellular signaling pathways predicted to be significantly affected during infection with SARS-CoV-2 (GSE147507).

Pathway Name	SARScov2NHBE	SARScov2a549	SARScov2calu3	SARScov2Lung
Tuberculosis			U	U
Innate Immune System			U	U
Cytokine Signaling in Immune system		D	D	
Complement and coagulation cascades	U			
Complement cascade		U		
NF-kappa B signaling pathway			U	
Hepatitis B			U	
Inflammatory bowel disease (IBD)			U	
HTLV-I infection			U	
Jak-STAT signaling pathway			U	
MAPK signaling pathway			U	
Osteoclast differentiation			U	
Apoptosis			U	
Measles			U	

Pathway Name	SARScov2NHBE	SARScov2a549	SARScov2calu3	SARScov2Lung
Hepatitis C			U	
Toll-like receptor signaling pathway			U	
Epstein-Barr virus infection			U	
PI3K-Akt signaling pathway			U	
Interleukin-1 signaling			U	
Signaling by Interleukins			U	
Nucleotide-binding domain, leucine rich repeat containing receptor (NLR) signaling pathways			U	
nf-kb signaling pathway			U	
signal transduction through il1r			U	
IL23-mediated signaling events			U	
Angiopoietin receptor Tie2-mediated signaling			U	
IL27-mediated signaling events			U	
IL12-mediated signaling events			U	
IL6-mediated signaling events			U	
Downstream TCR signaling				U
MyD88 dependent cascade initiated on endosome				U
TRAF6 mediated induction of NFkB and MAP kinases upon TLR7/8 or 9 activation				U
Translocation of ZAP-70 to Immunological synapse				U
HDR through Homologous Recombination (HRR)			D	
Class A/1 (Rhodopsin-like receptors)		D		
Peptide ligand-binding receptors		D		
Recognition of DNA damage by PCNA-containing replication complex			D	
Anchoring of the basal body to the plasma membrane			D	
stathmin and breast cancer resistance to antimicrotubule agents			D	

U: upregulated pathway; D: downregulated pathway

of these pathways were only found to be significant in one SARS-CoV-2 comparison and included “NF-kB signaling pathway”, “Interleukin-1 signaling”, “IL6-mediated signaling events”, “PI3K-Akt signaling pathway”, “Jak-STAT signaling pathway”, “Apoptosis”, “Complement and coagulation cascades”, and other processes associated with either the immune system or infectious diseases. The remaining three pathways that were predicted as affected during at least two SARS-CoV-2 comparisons were “Cytokine signaling in Immune system”, “Tuberculosis”, and the “Innate immune system”. In short, these findings indicate a set of signaling pathways that are strongly associated with virus infection and/or immune activation in the host, some of which are uniquely detected during SARS-CoV-2 infection. The “Cytokine Signaling in Immune

System” pathway is stored in the Reactome database and consists of interferon alpha/beta and gamma signaling, interleukin 1, 2, 6, and 10 signaling, and other components³⁷. To better understand the impact of SARS-CoV-2 on interleukin-6 signaling, we overlaid the differential expression data on a representation of the signaling pathway (Figure 2). This analysis revealed six of the eleven total proteins in the pathway were upregulated, while another protein was downregulated during SARS-CoV-2 infection.

Prediction of relevant drug targets

We next wanted to determine whether any of the significantly affected signaling pathways contained known drug targets that could be modulated to reduce infection, virus replication,

Figure 2. Infection with SARS-CoV-2 shows differential gene expression in Interleukin-6 signaling. This signaling pathway is a component of the larger Cytokine Signaling in Immune System pathway in Reactome. Each node represents a protein in the network, while each edge represents a characterized interaction between the proteins. Higher log₂ fold-change values (up-regulation) are represented by increasing saturation of red, while more negative log₂-fold-change values (down-regulation) are colored with increased saturation of blue. White nodes indicate no measured log₂-fold change.

and/or clinical signs and symptoms associated with infection by a panel of Betacoronaviruses or by SARS-CoV-2 alone. To do so we cross-referenced the results from our significant pathways analysis with the drug-target information accessible through an application programming interface (API) to the opentargets.org database, which yielded 179 potential human drug targets (Table 2). We ranked the predicted drugs and their associated targets according to how many coronaviruses shared the same drug target across the various datasets included in our analysis. We performed a separate ranking based on the data obtained solely from the SARS-CoV-2 studies.

After reviewing the results of this analysis, we identified 38 potential human protein targets to counteract MERS-CoV,

SARS-CoV, and/or SARS-CoV-2. Specifically, 38 targets were predicted to be relevant in at least 10 of the 15 coronavirus comparisons (Table 3). Seventeen of these targets were identified across 11 comparisons and included interferon (IFN)-A, IFN A receptors, IL1A and IL12B. Twenty-one additional targets were identified across 10 comparisons and included members of the tumor necrosis factor (TNF) superfamily, CCR4, CCR5, GHR, CXCR4, IL2R, IL4R, IL5, IL5R, IL6R, MPL, and IL20. We also predicted seven targets that were relevant specifically to SARS-CoV-2 including JAK1, FDPS, TLR7, TLR9, PRKCA, TYK2, and VDR.

Our analysis predicted twelve existing drugs that are predicted to be useful as repurposed therapeutics against 73.3%

Table 2. Comparison of predicted human drug targets across multiple datasets.

	GEO Series Identifier																
	GSE122876			GSE139516			GSE56192							GSE147507			
Human Target Symbol	MERS-inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung		
IFNA1	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA4	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA5	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA6	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA7	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA8	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA10	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA13	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA14	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IL1B	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA16	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA17	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNA21	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNAR1	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IFNAR2	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IL1A	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
IL12B	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1		
TNFSF13B	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1		
CCR4	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1		
CCR5	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1		
CSF3R	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1		
IFNLR1	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1		
EPOR	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1		
IL17RA	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1		

GEO Series Identifier																			
GSE122876			GSE139516			GSE56192								GSE147507					
Human Target Symbol	MERS-inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	MERS low24	MERS high24	SARS low48	SARS high48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung			
GHR	1	0	1	0	1	0	1	1	0	1	1	0	1	1	0	1	1		
IL1R1	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
CXCR4	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL2RA	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL2RB	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL3RA	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL4R	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL5	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL5RA	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL6R	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
MPL	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
IL20	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
TNFRSF4	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
TNFRSF8	1	0	1	0	1	0	1	1	1	1	1	0	1	1	0	1	1		
CALM1	0	0	1	0	0	1	1	1	1	0	1	0	0	0	0	1	1		
HCK	0	0	1	0	1	0	0	0	0	1	0	1	0	0	0	1	1		
LYN	0	0	1	0	1	0	0	0	0	1	0	1	0	0	0	1	1		
ROCK1	0	0	1	0	1	0	0	0	0	1	1	0	0	0	0	1	1		
ROCK2	0	0	1	0	1	0	0	0	0	1	1	0	0	0	0	1	1		
PDE1A	0	0	1	0	0	1	1	1	1	0	1	0	0	0	0	0	0		
PDE1C	0	0	1	0	0	1	1	1	1	0	1	0	0	0	0	0	0		
PDE1B	0	0	1	0	0	1	1	1	1	0	1	0	0	0	0	0	0		
GSK3B	0	0	0	0	1	0	0	0	0	1	1	0	0	0	0	1	1		
FGR	0	0	0	0	1	0	0	0	0	1	0	1	0	0	0	1	1		
GSK3A	0	0	0	0	1	0	0	0	0	1	0	1	0	0	0	1	1		
ITK	0	0	0	0	1	0	0	0	0	1	0	1	0	0	0	1	1		

	GEO Series Identifier																
	GSE122876		GSE139516		GSE56192								GSE147507				
Human Target Symbol	MERS-inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung		
PIK3CD	0	0	0	0	1	0	0	0	0	1	0	1	0	0	1	1	
PIK3CG	0	0	0	0	1	0	0	0	0	1	0	1	0	0	1	1	
PRKCZ	0	0	0	0	1	0	0	0	0	1	0	1	0	0	1	1	
SRC	0	0	0	0	1	0	0	0	0	1	0	1	0	0	1	1	
F2R	0	0	1	0	0	0	0	0	0	0	0	1	0	0	1	0	
MAPK11	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	1	
JAK1	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	
CDK6	0	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	
TUBA1B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
FDPS	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	
HDAC1	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1	0	
HDAC2	0	0	0	0	0	0	0	0	0	0	1	0	0	1	1	0	
TUBA3E	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
F2	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	
TUBA4A	0	0	1	0	0	0	0	0	0	0	0	0	0	1	1	0	
TUBA3C	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
TUBA1A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
TLR7	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	
PRKCA	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	
TYK2	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	
TUBA1C	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
CHRM1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
CHRM2	0	0	1	0	0	0	0	0	0	0	0	0	0	1	1	0	
EGFR	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
ERBB2	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	
ERBB3	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	

GEO Series Identifier																		
	GSE122876		GSE139516		GSE56192								GSE147507					
Human Target Symbol	MERS-inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung			
BDKRB2	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0		
PDGFRB	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0		
TLR9	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1		
VDR	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1		
ABL1	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0		
TUBB3	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
TUBB4A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
TUBB4B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
RBX1	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0		
TUBB8	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
TUBB2B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
MYH7B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
TUBB2A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
TUBB1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
TUBB6	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
CYSLTR1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
CHRM3	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
CHRM5	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADORA2A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADORA2B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADRA1D	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADRA1B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADRA1A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADRB1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADRB2	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		
ADRB3	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0		

GEO Series Identifier																
Human Target Symbol	GSE122876		GSE139516		GSE56192							GSE147507				
	MERS-inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	
DRD1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
DRD5	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
AGTR1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
EDNRA	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
F5	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
EDNRB	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
F8	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
PTGS2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0
F9	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
BTK	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0
F10	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
FGF	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
KLKB1	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
HRH1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
SERPINC1	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
HRH2	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
PLAT	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
HTR2A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
PLG	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
HTR2B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
HTR2C	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
HTR4	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
HTR5A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
HTR6	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
HTR7	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
LHCGR	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0

GEO Series Identifier																			
Human Target Symbol	GSE122876			GSE139516			GSE56192								GSE147507				
	MERS-inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung					
OXTR	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
CASP12	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
RRM2	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0
AVPR1A	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
AVPR1B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
PTGER1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
PTGER3	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
PTGFR	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
TACR2	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
TACR1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
TBXA2R	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
PRKCG	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
EPHA2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
FGFR3	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
FGFR4	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
FLT1	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
FLT3	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
FLT4	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
INSR	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
KDR	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
PGF	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
VEGFA	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
VEGFB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
VEGFC	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
ITGB3	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
LCK	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
PPARG	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0

	GEO Series Identifier																
	GSE122876		GSE139516		GSE56192								GSE147507				
Human Target Symbol	MERS-inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung		
CALCR	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
PARP2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
PARP3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
PARP1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
PPARA	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
RXRA	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
CD80	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
CD86	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
ITGAL	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL1A1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL1A2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL4A2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL4A4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL4A5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL4A6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL6A1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL6A2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL6A3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL6A6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
LAMB4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
COL6A5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
ITGA2B	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
ITGA4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	
ITGB7	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	

**KEY: 1 = target was present in comparison, 0 = target was NOT present in comparison

Table 3. Potential drug candidates for repurposing as antiviral prophylactics or therapeutics.

Target (Gene Symbol)	# Comparisons with Target Detected	Name of Therapeutic	Is Small Molecule	Is Antibody	Is Protein
IFNA1	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA10	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA13	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA14	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA16	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA17	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA21	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA4	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA5	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA6	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA7	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNA8	11	SIFALIMUMAB, RONTALIZUMAB		X, X	
IFNAR1	11	PEGINTERFERON BETA-1A, PEGINTERFERON ALFA-2A, INTERFERON BETA-1A, INTERFERON BETA-1B,			X, X, X, X
IFNAR2	11	PEGINTERFERON BETA-1A, PEGINTERFERON ALFA-2A, INTERFERON BETA-1A, INTERFERON BETA-1B,			X, X, X, X
IL12B	11	CANAKINUMAB, USTEKINUMAB, BRIAKINUMAB		X, X, X	
IL1A	11	BERMEKIMAB		X	
IL1B	11	CANAKINUMAB		X	
CCR4	10	MOGAMULIZUMAB		X	
CCR5	10	MARAVIROC	X		
CSF3R	10	FILGRASTIM, PEGFILGRASTIM			X, X
CXCR4	10	PLERIXAFOR	X		
EPOR	10	DARBEPOETIN ALFA, PEGINESATIDE			X, X
GHR	10	SOMATROPIN, PEGVISOMANT			X, X
IFNLR1	10	PEGINTERFERON LAMBDA-1A			X, X
IL17RA	10	BRODALUMAB		X	
IL1R1	10	ANAKINRA			X
IL20	10	FLETIKUMAB		X	
IL2RA	10	DACLIZUMAB, DENILEUKIN DIFTITOX		X,	,X
IL2RB	10	ALDESLEUKIN, DENILEUKIN DIFTITOX			X, X
IL3RA	10	TAGRAXOFUSP			X
IL4R	10	DUPILUMAB		X	
IL5	10	MEPOLIZUMAB, RESLIZUMAB		X, X	
IL5RA	10	BENRALIZUMAB		X	
IL6R	10	TOCILIZUMAB		X	
MPL	10	ELTROMBOPAG	X		
TNFRSF4	10	MEDI-6469		X	
TNFRSF8	10	BRENTUXIMAB VEDOTIN		X	
TNFSF13B	10	BELIMUMAB		X	

X: indicates the type of each therapeutic. Drug types against the same target are indicated with commas.

of all coronavirus taxa evaluated in this work and 100% of the SARS-CoV-2 comparisons included in our analysis (Table 4). Eight of these twelve drugs are used to treat common autoimmune disorders such as systemic lupus erythematosus (SLE), Crohn's disease, and multiple sclerosis (MS). Five of the twelve drugs have been associated or used as antiviral measures largely against hepatitis C virus. Another 27 drugs were predicted to have potential therapeutic activity against 66.7% of all coronavirus infections and 30 were predicted as potential therapeutics against 75% of SARS-CoV-2 infections.

We then analyzed the drugs that were identified as having targets in pathways affected by SARS-CoV-2 to determine which were predicted to reverse the effects of the viral infection on the affected pathway. Of the 42 drugs that targeted SARS-CoV-2 related pathways, 27 were predicted to reverse viral effects on these pathways. We then performed a literature search to determine if any of these drugs had been previously used to treat COVID-19 or had been identified as potential therapeutics by other research groups. We found six of the 12 therapeutics that we predicted to be useful against SARS-CoV-2 had already shown positive results in clinical tests including canakinumab, anakinra, tocilizumab, sarilumab, and baricitinib. These results give further support to the validity of our computational workflow.

Discussion

The computational workflow that we describe in this work predicts human therapeutic targets from signaling pathways and gene expression that are significantly affected during infection. We applied this workflow within the context of a meta-analysis that consisted of multiple public transcriptomic datasets of Betacoronaviruses. We then validated our results by comparing our predictions against recently published studies reporting therapeutics for SARS-CoV-2. Specifically, our downstream analyses enable us to calculate significant signaling pathways from DE genes using the SPIA algorithm as well as to predict potential therapeutics and their respective targets. Our analysis revealed thousands of DE genes, 580 enriched functional terms, as well as 249 significant pathways, including 38 pathways that were specifically affected during infection with SARS-CoV-2. It is important to point out that this workflow focuses on identifying human drug targets for two reasons: 1) to aid in the repurposing of existing drugs against emerging pathogens, and 2) to reduce the likelihood that a pathogen will develop resistance against the therapeutic(s) since they interact with a human protein that is much less likely to mutate than a viral protein.

Our approach differs from prior meta-analyses by focusing on a consistent, robust ARMOR-based RNA-seq preprocessing workflow for all datasets as well as a downstream pathway perturbation analysis. Previous studies have used a variety of approaches to predict possible therapeutics^{42–46}, but none have combined the various aspects that are described in this work. The SPIA algorithm we used in this study has been shown to provide robust statistical results of perturbed pathways while not simply enriching for DE genes³⁴. It

also enabled us to identify protein components in signaling pathways that could be reversed to reduce the adverse signs and symptoms that occur during infection, which differs from simply attempting to target DE genes. This approach drastically differs from some attempts to directly target DE genes without accounting for how a treatment may affect the cellular protein-protein interaction network. In this analysis we have compared significant DE genes and pathways identified in multiple different studies that used different MOIs, timepoints, and cell types. We found many DE genes and pathways that were affected across a variety of samples which increases our confidence in our results as those genes and pathways appear to be affected more by the virus itself than by other variables such as cell type, MOI, or timepoint.

Although our approach was dependent on identifying DE genes using a consistent preprocessing workflow, the focus of our analysis was to identify relevant functions, pathways, and potential drug targets from the DE genes. These processed data could then be used to better understand the underlying biological mechanisms of pathogenesis, but to better identify host-based therapeutic targets. A subset of the enriched annotations identified by the Camera algorithm have been reported to be relevant during infection with SARS-CoV-2 in clinical studies including “response to chemokine”⁴⁷, “humoral immune response”^{48–50}, “chronic inflammatory response”^{51,52}, “toll like receptor binding”^{53,54}, “interleukin-6 production”⁵⁵ and citrate metabolism. A separate study of COVID-19 identified Bradykinin as potentially playing a role in pathogenesis⁵⁶. Interestingly, the annotations for this gene and its receptor include several of the enriched terms we identified such as arachidonic acid, inflammation, and G-protein coupled receptor activity. We interpret these separate studies to validate the findings of our functional enrichment analysis and anticipate that future studies will shed additional insight into the underlying mechanism(s) of viral pathogenesis.

Many of the signaling pathway components that we identified in this study have also been reported previously. One prior proteomics study reported applying translation inhibitors to Caco-2 cells infected with SARS-CoV-2 reduced virus replication⁵⁷, which supports our pathway perturbation results. Other studies have reported that the ORF6, ORF8, and nucleocapsid proteins of SARS-CoV-2 are antagonists of type-I interferon and NF- κ B in HEK-293 T cells⁵⁸ or the induction of apoptosis during viral infection⁵⁹. In contrast, our meta-analysis predicted the type-I interferon pathway to be activated, suggesting either that this response could be dependent on the cell type, or that a potential redundant mechanism in the host cell can still turn on this pathway even if certain components are down-regulated. Our analysis predicted that noncanonical NF- κ B signaling was inhibited during MERS and SARS infection, while being activated during SARS-CoV-2 infection in Calu-3 cells. While it is possible that this difference is due to a cell-specific response from the studies included in our meta-analysis, a NF- κ B inhibitor applied to Vero E6 cells infected with SARS-CoV-2 has been shown to reduce cytopathic effects and virus plaques⁶⁰. This

Table 4. Comparison of therapeutic drugs and small molecules predicted to be repurposed against SARS-CoV-2.

Drug Name	GSE122876			GSE139516			GSE56192								GSE147507							
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549					
SIFALIMUMAB	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
RONALIZUMAB	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
PEGINTERFERON ALFA-2A	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
PEGINTERFERON ALFA-2B	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
PEGINTERFERON BETA-1A	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
INTERFERON BETA-1A	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
INTERFERON BETA-1B	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
INTERFERON ALFA-2B	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
BERMEKIMAB	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
CANAKINUMAB	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
USTEKINUMAB	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
BRIAKINUMAB	1	0	1	0	1	0	1	1	1	0	1	1	1	1	1	0	1					
BELIMUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
MOGAMULIZUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
MARAVIROC	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
FILGRASTIM	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
PEGFILGRASTIM	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
PEGINTERFERON LAMBDA-1A	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
DARBEPOETIN ALFA	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
PEGINESATIDE	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
BRODALUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
SOMATROPIN	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
PEGVISOMANT	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
ANAKINRA	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
DENILEUKIN DIFTITOX	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
DACLIZUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
ALDESLEUKIN	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					
TAGRAXOFUSP	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1					

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507							
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549			
DUPILUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
MEPOLIZUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
RESLIZUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
BENRALIZUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
TOCILIZUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
SARILUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
ELTROMBOPAG	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
FLETIKUMAB	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
MEDI-6469	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
PLERIXAFOR	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
BRENTUXIMAB VEDOTIN	1	0	1	0	1	0	1	1	1	0	1	1	0	1	1	0	1			
MIDOSTAURIN	0	0	0	0	1	0	0	0	1	0	1	0	1	1	1	0	1			
BARICTINIB	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	1			
HYDROXYCHLOROQUINE	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	1			
BENZIODARONE	0	0	1	0	0	1	1	1	0	1	0	0	0	1	1	0	0			
DASATINIB	0	0	1	0	1	0	0	0	1	0	1	0	0	1	1	0	0			
NETARSUDIL	0	0	1	0	1	0	0	0	1	0	1	0	0	1	1	0	0			
LITHIUM CARBONATE	0	0	0	0	1	0	0	0	1	0	1	0	0	1	1	0	0			
PAZOPANIB	0	0	0	0	1	0	0	0	1	0	1	0	0	1	1	0	0			
IDELALISIB	0	0	0	0	1	0	0	0	1	0	1	0	0	1	1	0	0			
DUVELISIB	0	0	0	0	1	0	0	0	1	0	1	0	0	1	1	0	0			
REGORAFENIB	0	0	1	0	0	0	0	0	0	0	1	0	0	1	1	0	0			
VORAPAXAR	0	0	1	0	0	0	0	0	0	0	0	1	0	1	0	0	0			
IMIQUIMOD	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0	1			
TOFACITINIB	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0	1			
PARICALCITOL	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0			
CALCIPOTRIENE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0			
CALCITRIOL	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0			
CHOLECALCIFEROL	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0			
ERGOCALCIFEROL	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	0	0			

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507				
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549
ABEMACICLIB	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0	0
VORINOSTAT	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0	0
PANOBINOSTAT	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0	0
ROMIDEPSIN	0	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0	0
VINFLUNINE	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
TRASTUZUMAB EMTANSINE	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
PACITAXEL POLIGLUMEX	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
BIVALIRUDIN	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0
DABIGATRAN ETEXILATE	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0
ARGATROBAN	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0
DESIRUDIN	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0
GLYCOPYRRONIUM	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ATROPINE	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
BENZTROPINE	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
TERODILINE	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
TROSPiUM	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
SOLIFENACIN	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
OXYBUTYNIN	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
DARIFENACIN	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
TOLTERODINE	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
GEFITINIB	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ERLOTINIB	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
VANDETANIB	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
NINTEDANIB	0	0	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ICATIBANT	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0
RISEDRONIC ACID	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1
ALENDRONIC ACID	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1
ZOLEDRONIC ACID	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1
PAMIDRONIC ACID	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1
DROTRECIGIN ALFA (ACTIVATED)	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507				
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549
EMICIZUMAB	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
EDOXABAN	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
RIVAROXABAN	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
APIXABAN	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
FIBRINOLYSIN, HUMAN	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
APROTININ	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
ECALLANTIDE	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
LANADELUMAB	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
HEPARIN SODIUM	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
ENOXAPARIN SODIUM	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
TINZAPARIN SODIUM	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
AMINOCAPROIC ACID	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
TRANEXAMIC ACID	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
UROKINASE	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
ALTEPLASE	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0
ACETAMINOPHEN	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ACECLOFENAC	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ASPIRIN	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
IBRUTINIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
EMRICASAN	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Small molecule	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Protein	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ERDAFTINIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
SUNITINIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
LENVATINIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
SORAFENIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
AXITINIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
INSULIN GLARGINE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
INSULIN SUSP ISOPHANE BEEF	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
INSULIN GLULISINE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507				
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549
INSULIN LISPRO	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
INSULIN DETEMIR	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
INSULIN PORK	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ANLOTINIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
CABOZANTINIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
AFLIBERCEPT	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
RANIBIZUMAB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
CONBERCEPT	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
TIROFIBAN	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
PIOGLITAZONE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
MESALAMINE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ROSIGLITAZONE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
PRAMLINIDE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
CALCITONIN SALMON RECOMBINANT	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
CALCITONIN SALMON	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
CALCITONIN	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
TALAZOPARIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
NIRAPARIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
OLAPARIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
RUCAPARIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
VELIPARIB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
FENOFRIBRATE	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
FENOFRIBRIC ACID	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ACITRETIN	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
ABATACEPT	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
BELATACEPT	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
EFALZUMAB	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
COLLAGENASE CLOSTRIDIUM HISTOLYTICUM	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
OCRIPLASMIN	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507						
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549		
VEDOLIZUMAB	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	
NATALIZUMAB	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	
PENTOXIFYLLINE	0	0	1	0	0	1	1	1	0	1	0	0	0	0	0	0	0	0	
DIPYRIDAMOLE	0	0	1	0	0	1	1	1	0	1	0	0	0	0	0	0	0	0	
NILOTINIB	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	
IMATINIB	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	
LENALIDOMIDE	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	
THALIDOMIDE	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	
HYDROXYUREA	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	
DOCETAXEL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
CABAZITAXEL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
PACLITAXEL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
COLCHICINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
VINCISTINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
IXABEPILONE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
VINORELBINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
VINBLASTINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
OMECAMTIV MECARBIL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
MONTELUKAST	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
ZAFIRLUKAST	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
IPRATROPIUM	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
TROPICAMIDE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
TIOTROPIUM	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
UMECLIDINIUM	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
FESOTERODINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
CAFFEINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
THEOPHYLLINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
ADENOSINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
CARVEDILOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
NOREPINEPHRINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
EPINEPHRINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507						
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549		
NAPHAZOLINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
TAMSULOSIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
DOXAZOSIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
ERGOTAMINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
ALFUZOSIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
METARAMINOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
PAZOSIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
SERTINDOLE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
ATENOLOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
METOPROLOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
BISOPROLOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
SOTALOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
ALBUTEROL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
TERBUTALINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
FORMOTEROL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
LABETALOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
PROPRANOLOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
LEVOSALBUTAMOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
DROXIDOPA	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
METHYLERGONOVINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
PIMOZIDE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
ERGOLOID	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
AMOXAPINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
VALSARTAN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
LOSARTAN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
TELMISARTAN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
BOSENTAN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
MACITENTAN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
DIPHENHYDRAMINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
CETIRIZINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
FEXOFENADINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0		

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507				
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549
EPINASTINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
OLOPATADINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
DIBENZEPIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
AZELASTINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
TRIPROLIDINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
DESLOPATADINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
RANITIDINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
FAMOTIDINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
NIZATIDINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
CIMETIDINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
TOLAZOLINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
HALOPERIDOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
RISPERIDONE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
CLOZAPINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
OLANZAPINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
QUETIAPINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
AMISULPRIDE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
METHYERGIDE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
DEXFENFLURAMINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
MIRTAZAPINE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
METOCLOPRAMIDE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
PRUCALOPRIDE	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
LUTROPIN ALFA	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
MENOTROPINS	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
GONADOTROPIN, CHORIONIC	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
OXYTOCIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
CARBETOCIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
ATOSIBAN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
DESMOPRESSIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
CONIVAPTAN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Drug Name	GSE122876		GSE139516		GSE56192								GSE147507					
	MERS inf	MERS6	MERS24	MERS high24	MERS low24	MERS low48	MERS high48	SARS low24	SARS high24	SARS low48	SARS high48	SARScov2 NHBE	SARScov2 A549	SARScov2 Calu3	SARScov2 Lung	Flu A549	RSV A549	
VASOPRESSIN	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
ALPROSTADIL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
MISOPROSTOL	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
LATANOPROST	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
TRAVOPROST	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
BIMATOPROST	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
IBODUTANT	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
FOSAPREPTANT	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
APREPTANT	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
NETUPITANT	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
FOSNETUPITANT	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
SERATRODAST	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	

***KEY: 1 = Drug predicted to be relevant in comparison; 0 = drug NOT predicted to be relevant in comparison

result suggests that NF- κ B signaling may be active and contribute to the inflammatory signs and symptoms observed during virus infection, which agrees with our results. The Toll-like receptor and JAK-STAT pathways were previously found to be relevant to SARS-CoV-2 infection in A549 cells, which we also identified in infected Calu-3 cells⁴². Citrate metabolism was also identified as an important pathway by our analysis and has been supported elsewhere⁶¹.

Although the US FDA has only issued emergency use authorization for therapeutic treatment for severe cases of COVID-19, a multitude of studies have reported results from attempting to treat patients with a variety of existing FDA approved therapeutics^{62–69}. We found that 27 of our 42 predicted therapeutics are predicted to “reverse” the effect on the pathways relevant to the viral infections being compared. Twelve of the 27 drugs that were predicted to be potential therapeutics against SARS-CoV-2 and are used to combat autoimmune or inflammatory diseases such as MS, SLE, and rheumatoid arthritis while others have been used in cancer treatments and against viral infections such as hepatitis C and human immunodeficiency virus. Six of these 12 drugs have been used to treat COVID-19 in patients including canakinumab, anakinra, tocilizumab, sarilumab, baricitinib, and hydroxychloroquine⁷⁰. Two other drugs, maraviroc and brodalumab, have been identified as potential treatments via cell cultures and computer models^{71–74}. Others on the list such as benralizumab have been identified through anecdotal data as biologics that potentially exert a prophylactic effect for SARS-CoV-2, when they are taken at the time of infection⁷⁵. Baricitinib is of particular interest as the US FDA has issued emergency use authorization for its use in conjunction with remdesivir in the treatment of COVID-19 patients over the age of two that have been hospitalized and require supplemental oxygen, invasive mechanical ventilation, or extracorporeal membrane oxygenation⁷⁶. Baricitinib has also been shown to be effective against COVID-19 when combined with corticosteroids⁷⁷. A small study involving Tocilizumab has also shown it can be useful in improving the outcome of patients with severe COVID-19⁷⁸.

Conclusions

In conclusion, we developed and applied an important bioinformatics workflow, that combines existing tools with custom scripts, to predict potential human therapeutic targets. This workflow was then validated through a meta-analysis of publicly available transcriptomics data. The multiple Betacoronavirus and SARS-CoV-2 datasets revealed significant genes, annotations, signaling pathways, and human proteins that could be targeted by therapeutics during infection with various Betacoronaviruses. It is important to recognize that many of the predictions made by our workflow have been supported by experimental and clinical work on this virus, which suggests that our approach could enable the rapid identification of relevant therapeutics against future emerging pathogens.

Abbreviations

SARS-CoV-2: Severe acute respiratory syndrome coronavirus-2

COVID-19: Coronavirus disease-2019

GO: Gene ontology

SARS / SARS-CoV: Severe acute respiratory syndrome coronavirus

MERS / MERS-CoV: Middle east respiratory syndrome coronavirus

ACE2: Angiotensin-converting enzyme 2

FDA: Food and Drug Administration

GEO: Gene expression omnibus

NCBI: National center for biotechnology information

ARMOR: Automated Reproducible MODular Workflow for Preprocessing and Differential Analysis of RNA-seq Data

SPIA: Signaling pathway impact analysis

DE: Differentially expressed

FDR: False-discovery rate

Data availability

Underlying data

GEO: Transcriptomic analysis of MERS-CoV infected Calu-3 cell with or without AM580 treatment, Accession number GSE122876: <https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE122876>

GEO: Transcriptomic analysis of the Novel Middle East Respiratory Syndrome Coronavirus (Human, MRC5 cells, Accession number GSE56192: <https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE56192>

GEO: Transcriptional response to SARS-CoV-2 infection, Accession number GSE147507: <https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE147507>

GEO: Transcriptomic Analysis Of circRNAs/miRNAs/mRNAs upon Middle East Respiratory Syndrome Coronavirus (MERS-CoV) infection, Accession number GSE139516: <https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE139516>

Code for this workflow can be found on GitHub: <https://github.com/bpickett/Pathway2Targets>.

Archived code as at time of publication: <http://doi.org/10.5281/zenodo.4706197>⁷⁹.

Code is available under the terms of the [Creative Commons Zero “No rights reserved” data waiver](#) (CC0 1.0 Public domain dedication).

Acknowledgments

We would like to thank the Office of Research Computing at Brigham Young University for their help on this project. We also gratefully acknowledge the original providers of the RNA-seq datasets.

References

- Corman VM, Muth D, Niemeyer D, et al.: **Hosts and Sources of Endemic Human Coronaviruses.** *Adv Virus Res.* 2018; **100**: 163–88.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Wu A, Peng Y, Huang B, et al.: **Genome Composition and Divergence of the Novel Coronavirus (2019-nCoV) Originating in China.** *Cell Host Microbe.* 2020; **27**(3): 325–8.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Cui J, Li F, Shi ZL: **Origin and evolution of pathogenic coronaviruses.** *Nat Rev Microbiol.* 2019; **17**(3): 181–92.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Helmy YA, Fawzy M, Elawad A, et al.: **The COVID-19 Pandemic: A Comprehensive Review of Taxonomy, Genetics, Epidemiology, Diagnosis, Treatment, and Control.** *J Clin Med Res.* 2020; **9**(4).
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Zhai P, Ding Y, Wu X, et al.: **The epidemiology, diagnosis and treatment of COVID-19.** *Int J Antimicrob Agents.* 2020; **55**(5): 105955.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Ye Q, Wang B, Mao J, et al.: **Epidemiological analysis of COVID-19 and practical experience from China.** *J Med Virol.* 2020; **92**(7): 755–69.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Weekly epidemiological update - 16 February 2021.** [cited 2021 Feb 23].
[Reference Source](#)
- Pedersen SF, Ho YC: **SARS-CoV-2: a storm is raging.** *J Clin Invest.* 2020; **130**(5): 2202–5.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Xiong Y, Liu Y, Cao L, et al.: **Transcriptomic characteristics of bronchoalveolar lavage fluid and peripheral blood mononuclear cells in COVID-19 patients.** *Emerg Microbes Infect.* 2020; **9**(1): 761–70.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Wu D, Yang XO: **TH17 responses in cytokine storm of COVID-19: An emerging target of JAK2 inhibitor Fedratinib.** *J Microbiol Immunol Infect.* 2020; **53**(3): 368–70.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Huang C, Wang Y, Li X, et al.: **Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China.** *Lancet.* 2020; **395**(10223): 497–506.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Gu J, Han B, Wang J: **COVID-19: Gastrointestinal Manifestations and Potential Fecal-Oral Transmission.** *Gastroenterology.* 2020; **158**(6): 1518–9.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Ding S, Liang TJ: **Is SARS-CoV-2 Also an Enteric Pathogen With Potential Fecal-Oral Transmission? A COVID-19 Virological and Clinical Review.** *Gastroenterology.* 2020; **159**(1): 53–61.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Hamming I, Cooper ME, Haagmans BL, et al.: **The emerging role of ACE2 in physiology and disease.** *J Pathol.* 2007; **212**(1): 1–11.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Smyth LJ, Cañadas-Garre M, Cappa RC, et al.: **Genetic associations between genes in the renin-angiotensin-aldosterone system and renal disease: a systematic review and meta-analysis.** *BMJ Open.* 2019; **9**(4): e026777.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Search of: **COVID-19 - List Results - ClinicalTrials.gov.**
[Reference Source](#)
- Draft landscape of COVID-19 candidate vaccines.** [cited 2020 Aug 12].
[Reference Source](#)
- Canver MC, Bauer DE, Maeda T, et al.: **DrugThatGene: integrative analysis to streamline the identification of druggable genes, pathways and protein complexes from CRISPR screens.** *Bioinformatics.* 2019; **35**(11): 1981–4.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Cheng F, Lu W, Liu C, et al.: **A genome-wide positioning systems network algorithm for in silico drug repurposing.** *Nat Commun.* 2019; **10**(1): 3476.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Barrett T, Wilhite SE, Ledoux P, et al.: **NCBI GEO: archive for functional genomics data sets—update.** *Nucleic Acids Res.* 2013; **41**(Database issue): D991–5.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Kodama Y, Shumway M, Leinonen R, et al.: **The Sequence Read Archive: explosive growth of sequencing data.** *Nucleic Acids Res.* 2012; **40**(Database issue): D54–6.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Yuan S, Chu H, Chan JFW, et al.: **SREBP-dependent lipidomic reprogramming as a broad-spectrum antiviral target.** *Nat Commun.* 2019; **10**(1): 120.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Blanco-Melo D, Nilsson-Payant BE, Liu WC, et al.: **Imbalanced Host Response to SARS-CoV-2 Drives Development of COVID-19.** *Cell.* 2020; **181**(5): 1036–1045.e9.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Zhang X, Chu H, Wen L, et al.: **Competing endogenous RNA network profiling reveals novel host dependency factors required for MERS-CoV propagation.** *Emerg Microbes Infect.* 2020; **9**(1): 733–746.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Orjuela S, Huang R, Hembach KM, et al.: **ARMOR: An Automated Reproducible MODular Workflow for Preprocessing and Differential Analysis of RNA-seq Data.** *G3 (Bethesda).* 2019; **9**(7): 2089–2096.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Patro R, Duggal G, Love MI, et al.: **Salmon provides fast and bias-aware quantification of transcript expression.** *Nat Methods.* 2017; **14**(4): 417–419.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Robinson MD, McCarthy DJ, Smyth GK: **edgeR: a Bioconductor package for differential expression analysis of digital gene expression data.** *Bioinformatics.* 2010; **26**(1): 139–40.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Nowicka M, Robinson MD: **DRIMSeq: a Dirichlet-multinomial framework for multivariate count outcomes in genomics [version 2; peer review: 2 approved].** *F1000Res.* 2016; **5**: 1356.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- The Gene Ontology Consortium: **The Gene Ontology Consortium. The Gene Ontology Resource: 20 years and still GOing strong.** *Nucleic Acids Res.* 2019; **47**(D1): D330–8.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Liberzon A, Birger C, Thorvaldsdóttir H, et al.: **The Molecular Signatures Database (MSigDB) hallmark gene set collection.** *Cell Syst.* 2015; **1**(6): 417–425.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Wu D, Smyth GK: **Camera: a competitive gene set test accounting for inter-gene correlation.** *Nucleic Acids Res.* 2012; **40**(17): e133.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Durinck S, Moreau Y, Kasprzyk A, et al.: **BioMart and Bioconductor: a powerful link between biological databases and microarray data analysis.** *Bioinformatics.* 2005; **21**(16): 3439–40.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Huber W, Carey VJ, Gentleman R, et al.: **Orchestrating high-throughput genomic analysis with Bioconductor.** *Nat Methods.* 2015; **12**(2): 115–21.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Tarca AL, Draghici S, Khatri P, et al.: **A novel signaling pathway impact analysis.** *Bioinformatics.* 2009; **25**(1): 75–82.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Sales G, Calura E, Cavalieri D, et al.: **graphite - a Bioconductor package to convert pathway topology to gene network.** *BMC Bioinformatics.* 2012; **13**: 20.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Kanehisa M, Goto S: **KEGG: kyoto encyclopedia of genes and genomes.** *Nucleic Acids Res.* 2000; **28**(1): 27–30.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Jassal B, Matthews L, Viteri G, et al.: **The reactome pathway knowledgebase.** *Nucleic Acids Res.* 2020; **48**(D1): D498–503.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Mi H, Huang X, Muruganujan A, et al.: **PANTHER version 11: expanded annotation data from Gene Ontology and Reactome pathways, and data analysis tool enhancements.** *Nucleic Acids Res.* 2017; **45**(D1): D183–D189.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Schaefer CF, Anthony K, Krupa S, et al.: **PID: the Pathway Interaction Database.** *Nucleic Acids Res.* 2009; **37**(Database issue): D674–9.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- The UniProt Consortium: **UniProt: the universal protein knowledgebase.** *Nucleic Acids Res.* 2018; **46**(5): 2699.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Carvalho-Silva D, Pierleoni A, Pignatelli M, et al.: **Open Targets Platform: new developments and updates two years on.** *Nucleic Acids Res.* 2019; **47**(D1): D1056–65.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Fagone P, Ciurleo R, Lombardo SD, et al.: **Transcriptional landscape of SARS-CoV-2 infection dismantles pathogenic pathways activated by the virus, proposes unique sex-specific differences and predicts tailored therapeutic strategies.** *Autoimmun Rev.* 2020; **19**(7): 102571.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Magro G: **SARS-CoV-2 and COVID-19: Is interleukin-6 (IL-6) the “culprit lesion” of ARDS onset? What is there besides Tocilizumab? SGP130Fc.** *Cytokine X.* 2020; **2**(2): 100029.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Sallenave JM, Guillot L: **Innate Immune Signaling and Proteolytic Pathways in the Resolution or Exacerbation of SARS-CoV-2 in Covid-19: Key Therapeutic Targets?** *Front Immunol.* 2020; **11**: 1229.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Drucker DJ: **Coronavirus Infections and Type 2 Diabetes-Shared Pathways with Therapeutic Implications.** *Endocr Rev.* 2020; **41**(3): bnaa011.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Hoagland DA, Clarke DJB, Møller R, et al.: **Modulating the transcriptional**

- landscape of SARS-CoV-2 as an effective method for developing antiviral compounds. 2020 [cited 2020 Sep 17]; 2020.07.12.199687.
[Publisher Full Text](#)
47. Coperchini F, Chiovato L, Croce L, *et al.*: **The cytokine storm in COVID-19: An overview of the involvement of the chemokine/chemokine-receptor system.** *Cytokine Growth Factor Rev.* 2020; **53**: 25–32.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 48. Zhang Y, Xu J, Jia R, *et al.*: **Protective humoral immunity in SARS-CoV-2 infected pediatric patients.** *Cell Mol Immunol.* 2020; **17**(7): 768–70.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 49. Ni L, Ye F, Cheng ML, *et al.*: **Detection of SARS-CoV-2-Specific Humoral and Cellular Immunity in COVID-19 Convalescent Individuals.** *Immunity.* 2020; **52**(6): 971–7.e3.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 50. Kellam P, Barclay W: **The dynamics of humoral immune responses following SARS-CoV-2 infection and the potential for reinfection.** *J Gen Virol.* 2020; **101**(8): 791–7.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 51. Young BE, Fong SW, Chan YH, *et al.*: **Effects of a major deletion in the SARS-CoV-2 genome on the severity of infection and the inflammatory response: an observational cohort study.** *Lancet.* 2020; **396**(10251): 603–11.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 52. Fu Y, Cheng Y, Wu Y: **Understanding SARS-CoV-2-Mediated Inflammatory Responses: From Mechanisms to Potential Therapeutic Tools.** *Viral Sin.* 2020; **35**(3): 266–71.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 53. Bouayad A: **Innate immune evasion by SARS-CoV-2: Comparison with SARS-CoV.** *Rev Med Virol.* 2020; **30**(6): 1–9.
[PubMed Abstract](#) | [Publisher Full Text](#)
 54. Moreno-Eutimio MA, López-Macías C, Pastelin-Palacios R: **Bioinformatic analysis and identification of single-stranded RNA sequences recognized by TLR7/8 in the SARS-CoV-2, SARS-CoV, and MERS-CoV genomes.** *Microbes Infect.* 2020; **22**(4–5): 226–9.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 55. Belhadj Z, Méot M, Bajolle F, *et al.*: **Acute heart failure in multisystem inflammatory syndrome in children (MIS-C) in the context of global SARS-CoV-2 pandemic.** *Circulation.* 2020; **142**(5): 429–436.
[PubMed Abstract](#) | [Publisher Full Text](#)
 56. Garvin MR, Alvarez C, Miller JL, *et al.*: **A mechanistic model and therapeutic interventions for COVID-19 involving a RAS-mediated bradykinin storm.** *eLife.* 2020; **9**: e59177.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 57. Bojkova D, Klann K, Koch B, *et al.*: **Proteomics of SARS-CoV-2-infected host cells reveals therapy targets.** *Nature.* 2020; **583**(7816): 469–72.
[PubMed Abstract](#) | [Publisher Full Text](#)
 58. Li JY, Liao CH, Wang Q, *et al.*: **The ORF6, ORF8 and nucleocapsid proteins of SARS-CoV-2 inhibit type I interferon signaling pathway.** *Virus Res.* 2020; **286**: 198074.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 59. Ren Y, Shu T, Wu D, *et al.*: **The ORF3a protein of SARS-CoV-2 induces apoptosis in cells.** *Cell Mol Immunol.* 2020; **17**(8): 881–3.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 60. Ma Q, Pan W, Li R, *et al.*: **Liu Shen capsule shows antiviral and anti-inflammatory abilities against novel coronavirus SARS-CoV-2 via suppression of NF- κ B signaling pathway.** *Pharmacol Res.* 2020; **158**: 104850.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 61. Overmyer KA, Shishkova E, Miller IJ, *et al.*: **Large-scale Multi-omic Analysis of COVID-19 Severity.** *medRxiv.* 2020; 2020.07.17.20156513.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 62. Zhang B, Liu S, Tan T, *et al.*: **Treatment With Convalescent Plasma for Critically Ill Patients With Severe Acute Respiratory Syndrome Coronavirus 2 Infection.** *Chest.* 2020; **158**(1): e9–13.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 63. Li H, Chen C, Hu F, *et al.*: **Impact of corticosteroid therapy on outcomes of persons with SARS-CoV-2, SARS-CoV, or MERS-CoV infection: a systematic review and meta-analysis.** *Leukemia.* 2020; **34**(6): 1503–11.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 64. Yan Y, Shin WI, Pang YX, *et al.*: **The First 75 Days of Novel Coronavirus (SARS-CoV-2) Outbreak: Recent Advances, Prevention, and Treatment.** *Int J Environ Res Public Health.* 2020; **17**(7): 2323.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 65. Yang X, Yu Y, Xu J, *et al.*: **Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study.** *Lancet Respir Med.* 2020; **8**(5): 475–81.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 66. Messina F, Piaserico S: **SARS-CoV-2 infection in a psoriatic patient treated with IL-23 inhibitor.** *J Eur Acad Dermatol Venereol.* 2020; **34**(6): e254–5.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 67. Caly L, Druce JD, Catton MG, *et al.*: **The FDA-approved drug ivermectin inhibits the replication of SARS-CoV-2 in vitro.** *Antiviral Res.* 2020; **178**: 104787.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 68. Choy KT, Wong AYL, Kaewpreedee P, *et al.*: **Remdesivir, lopinavir, emetine, and homoharringtonine inhibit SARS-CoV-2 replication in vitro.** *Antiviral Res.* 2020; **178**: 104786.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 69. Yao X, Ye F, Zhang M, *et al.*: **In Vitro Antiviral Activity and Projection of Optimized Dosing Design of Hydroxychloroquine for the Treatment of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2).** *Clin Infect Dis.* 2020; **71**(15): 732–9.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 70. Gilzad-Kohan H, Jamali F: **Anti-Inflammatory Properties of Drugs Used to Control COVID-19 and their Effects on the Renin-Angiotensin System and Angiotensin-Converting Enzyme-2.** *J Pharm Pharm Sci.* 2020; **23**: 259–77.
[PubMed Abstract](#) | [Publisher Full Text](#)
 71. Tazikheh-Lemeski E, Moradi S, Raoufi R, *et al.*: **Targeting SARS-COV-2 non-structural protein 16: a virtual drug repurposing study.** *J Biomol Struct Dyn.* 2020; 1–14.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 72. Shamsi A, Mohammad T, Anwar S, *et al.*: **Glecaprevir and Maraviroc are high-affinity inhibitors of SARS-CoV-2 main protease: possible implication in COVID-19 therapy.** *Biosci Rep.* 2020; **40**(6): BSR20201256.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 73. Bulat V, Situm M, Azdajic MD, *et al.*: **Potential role of IL-17 blocking agents in the treatment of severe COVID-19?** *Br J Clin Pharmacol.* 2020; **87**(3): 1578–1581.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 74. Risner KH, Tieu KV, Wang Y, *et al.*: **Maraviroc inhibits SARS-CoV-2 multiplication and s-protein mediated cell fusion in cell culture.** *bioRxiv.* 2020; 2020.08.12.246389.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 75. García-Moguel I, Campos RD, Charterina SA, *et al.*: **COVID-19, severe asthma, and biologics.** *Ann Allergy Asthma Immunol.* 2020; **125**(3): 357–9.e1.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 76. Kalil AC, Patterson TF, Mehta AK, *et al.*: **Baricitinib plus Remdesivir for Hospitalized Adults with Covid-19.** *N Engl J Med.* 2021; **384**(9): 795–807.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 77. Rodriguez-Garcia JL, Sanchez-Nievas G, Arevalo-Serrano J, *et al.*: **Baricitinib improves respiratory function in patients treated with corticosteroids for SARS-CoV-2 pneumonia: an observational cohort study.** *Rheumatology (Oxford).* 2021; **60**(1): 399–407.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 78. Cala-García JD, Sierra-Bretón JD, Cavellier-Baiz JE, *et al.*: **Recovery of COVID-19 acute respiratory distress syndrome with tocilizumab: successful outcome in two critically ill patients.** *Immunotherapy.* 2020; **12**(15): 1127–32.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
 79. Pickett BE: **Pathway2Targets.** *Zenodo.* 2021.
<http://www.doi.org/10.5281/zenodo.4706197>

Open Peer Review

Current Peer Review Status:

Version 1

Reviewer Report 02 August 2021

<https://doi.org/10.5256/f1000research.55686.r89746>

© 2021 Oso B. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Babatunde Joseph Oso

Department of Biochemistry, McPherson University, Seriki Sotayo, Nigeria

The authors presented an original article on the signaling pathway-driven bioinformatics pipeline for predicting therapeutics against emerging infectious diseases. The authors are commended for the clarity and straight-forward style of the article. However, this manuscript needs to be improved regarding the following aspects:

- It would be interesting if a bibliographic review on the importance of bioinformatics workflow and meta-analysis vis-à-vis prediction of potential human therapeutics can be included in the introduction section.
- The argument presented in the introduction section should include a clear description of the statement problem.
- Generally, the sentences should be well structured for a better flow and limit the use of possessive pronouns.

Is the rationale for developing the new method (or application) clearly explained?

Yes

Is the description of the method technically sound?

Yes

Are sufficient details provided to allow replication of the method development and its use by others?

Yes

If any results are presented, are all the source data underlying the results available to ensure full reproducibility?

Yes

Are the conclusions about the method and its performance adequately supported by the findings presented in the article?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Biochemistry

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Reviewer Report 02 June 2021

<https://doi.org/10.5256/f1000research.55686.r85467>

© 2021 Cerón-Carrasco J. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

José Pedro Cerón-Carrasco

Bioinformatics and High Performance Computing Research Group (BIO-HPC), Faculty of Polytechnic Science, UCAM Catholic University of Murcia, Murcia, Spain

I feel this work touches up an alternative approach to the more classical computational protocol of performing virtual screening by docking methods + molecular dynamics to refine binding energies.

Herein, the proposed method uses RNA-sequencing data with a focus on defining differentially expressed genes, enriched Gene Ontology (GO) terms, and dysregulated pathways in infected cells.

On the brightest side, that theoretical framework allows for a simultaneous search of small molecules, antibody and protein. The authors stated that their predictions yield to a final list of 42 therapeutics. 27 of such drugs have been reported to exert an effect against the virus. It remains unrevealed if these other 15 are negative positive or pending possible treatments. A more 'experimental' reader might find a next candidate in that list.

Is the rationale for developing the new method (or application) clearly explained?

Yes

Is the description of the method technically sound?

Yes

Are sufficient details provided to allow replication of the method development and its use by others?

Yes

If any results are presented, are all the source data underlying the results available to ensure full reproducibility?

Yes

Are the conclusions about the method and its performance adequately supported by the findings presented in the article?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Computational Chemistry for Drug Design.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Author Response 17 Aug 2021

Brett Pickett

We thank the reviewer for their helpful comment and agree that researchers would could indeed make good use of the complete list of drugs that we predicted to be useful against SARS-CoV-2. To address this comment we will update Table 4 in the next manuscript version to now include not only all of the predicted drugs, but also summaries of how each drug ranks across all analyzed Coronavirus datasets and also across all SARS-CoV-2 datasets. We believe that this change will improve the readability and relevance of our study.

Competing Interests: No competing interests were disclosed.

The benefits of publishing with F1000Research:

- Your article is published within days, with no editorial bias
- You can publish traditional articles, null/negative results, case reports, data notes and more
- The peer review process is transparent and collaborative
- Your article is indexed in PubMed after passing peer review
- Dedicated customer support at every stage

For pre-submission enquiries, contact research@f1000.com

F1000Research