Meta-Data Analysis to Explore the Hub of the Hub-Genes That Influence SARS-CoV-2 Infections Highlighting Their Pathogenetic Processes and Drugs Repurposing

Md. Parvez Mosharaf 1,2, Md. Kaderi Kibria 1, Md. Bayazid Hossen 1, Md. Ariful Islam 1, Md. Selim Reza 1, Rashidul Alam Mahumud 3, Khorsheed Alam 2, Jeff Gow 2,4 and Md. Nurul Haque Mollah 1,* 1

Bioinformatics Lab, Department of Statistics, University of Rajshahi, Rajshahi 6205, Bangladesh; parvez.mosharaf@usq.edu.au (M.P.M.); s1510124113@ru.ac.bd (M.K.K.); s1710624176@ru.ac.bd (M.B.H.); s1511124151@ru.ac.bd (M.A.I.); selim@siat.ac.cn (M.S.R.)
2 School of Business, Faculty of Business, Education, Law and Arts, University of Southern Queensland, Toowoomba, QLD 4350, Australia; khorsheed.alam@usq.edu.au (K.A.); jeffrey.gow@usq.edu.au (J.G.)
3 NHMRC Clinical Trials Centre, Faculty of Medicine and Health, The University of Sydney, Camperdown, NSW 2006, Australia; rashed.mahumud@sydney.edu.au
4 School of Accounting, Economics and Finance, University of KwaZulu Natal, Durban 4001, South Africa

* Correspondence: mollah.stat.bio@ru.ac.bd

Abstract: The pandemic of SARS-CoV-2 infections is a severe threat to human life and the world economic condition. Although vaccination has reduced the outbreak, but still the situation is not under control because of the instability of RNA sequence patterns of SARS-CoV-2, which requires effective drugs. Several studies have suggested that the SARS-CoV-2 infection causing hub differentially expressed genes (Hub-DEGs). However, we observed that there was not any common hub gene (Hub-DEGs) in our analyses. Therefore, it may be difficult to take a common treatment plan against SARS-CoV-2 infections globally. The goal of this study was to examine if more representative Hub-DEGs from published studies by means of hub of Hub-DEGs (hHub-DEGs) and associated potential candidate drugs. In this study, we reviewed 41 articles on transcriptomic data analysis of SARS-CoV-2 and found 370 unique hub genes or studied genes in total. Then, we selected 14 more representative Hub-DEGs (AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNF, TP53, UBA52, UBC, VEGFA) as hHub-DEGs by their protein-protein interaction analysis. Their associated biological functional processes, transcriptional, and post-transcriptional regulatory factors. Then we detected hHub-DEGs guided top-ranked nine candidate drug agents (Digoxin, Avermectin, Simeprevir, Nelfinavir Mesylate, Proscillaridin, Linifanib, Withaferin, Amuvatinib, Atazanavir) by molecular docking and cross-validation for treatment of SARS-CoV-2 infections. Therefore, the findings of this study could be useful in formulating a common treatment plan against SARS-CoV-2 infections globally.

Keywords: SARS-CoV-2 infections; selection of drug targets and agents; drug repurposing; molecular docking and dynamic simulation

1 Introduction

The epidemic of coronavirus diseases (COVID-19) has now passed the initial phase due to inconsistent RNA patterns that lead its spreading status to be out of control. Although several vaccines including BBIBP-CorV, Pfizer, BBV152, Moderna, AstraZeneca, EpiVacCorona, Sputnik, Ad5-nCoV, WIBP, and CoronaVac are now available and being dosed against SAR-CoV-2 [1,2], scientists around the world are anxious regarding their effectiveness because of the unstable pattern of RNA sequence of SARS-CoV-2 virus. For instance, recently it has been observed that fully vaccinated people are also being affected by novel SARS-CoV-2 variants around the world. In this regard, the distinct types of well-established drugs could be a supportive tonic in the treatment of supplementing vaccines against coronavirus to reduce infection rates and save lives.
However, using traditional methods, novel drug discovery is a tremendous thought-provoking, time consuming, and expensive task. The foremost challenge is to explore drug target receptors and drug candidate small molecules. Now, transcriptomics data analysis is a widely used popular approach to explore genomic biomarkers [3–7]. Modern high throughput technology and the gene expression data analysis techniques have revealed the potential biomarker-induced proteins which are being considered as the key drug target receptors. The repurposing of existing drugs against a disease could outweigh the de novo drug development by reducing the time and cost. Ongoing research proposes several sets of transcriptome-guided targeted genes/receptor proteins as well as different drug candidate molecules [8–28]. Nonetheless, the comparative discussion regarding the consistency and inconsistency of the drug target molecules (i.e., Hub-DEGs) among the distinct articles of COVID-19 has not been presented yet. In our literature review, we also noticed that there was not any set of genes/receptor proteins and/or drug molecules regarding COVID-19 infection that are commonly reported across studies. It may happen due to study design, data collection methods, and the environment.

Therefore, it may be difficult to develop a common therapeutic treatment plan against SARS-CoV-2 infection. To make a comparatively more common therapeutic development strategy for all, it is required to find more representative Hub-DEGs set which will be the global key drug target molecules for the human body. There are several research groups working with COVID-19 research [8–28], but so far this issue has not been reported yet. The goal of this study was to provide more representative Hub-DEGs from all published studies by means of hub of Hub-DEGs (hHub-DEGs) and the associated potential drug candidates. The pipeline of this study is given in Figure 1.

Figure 1. The road map of this research.

2. Materials and Methods

In this project, the necessary meta-genes and re-purusable meta-drug agents were collected from different online sources and published articles. Then, the integrated bioinformatics analysis approach was utilized to investigate the most effective drug target biomolecules and potential drug agents for SARS-CoV-2.
2.1. Metadata Sources and Descriptions

In this study, we collected metadata for both drug targets and drug agents that were associated with SARS-CoV-2 as described below:

2.1.1. Collection of Hub-DEGs to Explore Drug Targets

Several research groups have already published different sets of Hub-DEGs that are associated with SARS-CoV-2 infections. Here, Hub-DEGs indicates the key differentially expressed genes between SARS-CoV-2 infections and control samples. We collected 41 published articles that suggested SARS-CoV-2-causing Hub-DEGs through the Google Scholar search engine. We found in total 370 unique Hub-DEGs/studied genes (Table 1) by reviewing those 41 articles.

Table 1. Hub genes/proteins that were associated with SARS-CoV-2 infections in different studies.

| SL | Articles & Datasets | Hub-Genes/Proteins | Number of Proteins |
|----|---------------------|--------------------|--------------------|
| 1  | Caradonna, A et al., 2022 [29] | ACE2, APP            | 2                  |
| 2  | Hanming Gu et al., 2020 [30]  | NFKBIA, C3, CCL20, BCL2A1, BID | 5                  |
| 3  | Kang Soon Nan et al., 2021 [18] | ALB, CXCL8, FGF2, IL6, INS, MMP2, MMP9, PTGS2, STAT3, VEGFA | 10                 |
| 4  | Hanming Gu et al., 2020 [31]  | CDC20, NCBP1, POLR2D, DYNLL1, FBXW5, LRRC41, FBXO21, FBXW9, FBXO44, FBXO6 | 10                 |
| 5  | Rahila Sardar et al., 2020 [19] | HMOX1, DNM1, PLAT, GDF1, ITGB1 | 5                  |
| 6  | Hanming Gu et al., 2020 [21]  | FLOC, DYNLL1, FBXL3, FBXW11, FBXO27, FBXO44, FBXO32, FBXO31, FBXO9, CUL2 | 10                 |
| 7  | Tian-Ao Xie et al., 2020 [8]   | CXCL1, CXCL2, TNF, NFKBIA, CSF2, TNFAIP3, IL6, CXCL3, CCL20, ICAM1 | 10                 |
| 8  | Jung Hun Oh et al., 2020 [9]   | GATA4, ID2, MAFA, NOX4, PTBP1, SMAD3, TUBB1, WWOX, TP53, HRAS, CTNNB1, FYN, ABL1, STAT3, STAT1, JAK2, CIQB, XBPI, BST2, CD99, IFT35, MAPK11, RELA, LCK, KIT, EGR1, IL20, ILF3, CASP3, IL19, ATG7, GIP, SIPRI | 25                 |
| 9  | Basavaraj Vastrad et al., 2020 [20] | STAT1, IRF7, IFIHI, MX1, ISG15, IFIT3, OAS2, DDX58, IRF9, IFIT1, OAS1, OAS3, DDX60, OASL, IFIT2, MYC, HDAC9, NCOA3, CEBPB, VEGFA, BCL3, SMAD3, SMURF1, KLHL12, CBL, ERBB4, CRKL, MARCO, VCAN, AFB8, LGLAL5, HMOX1, TIMP1, OAS2, GAPDH, MSH3, FN1, NPC2, JUND, CHI3L1, GPNB4, SYTL2, CASP1, S100A8, MYO10, IGBP3, APCDD1, COL6A3, FABP5, PRDX3, CLEC1B, DDI4, CXCL10, CXCL8 | 27                 |
| 10 | Kartikay Prasad et al., 2020 [22] | STAT1, IRF7, IFIHI, MX1, ISG15, IFIT3, OAS2, DDX58, IRF9, IFIT1, OAS1, OAS3, DDX60, OASL, IFIT2, MYC, HDAC9, NCOA3, CEBPB, VEGFA, BCL3, SMAD3, SMURF1, KLHL12, CBL, ERBB4, CRKL, MARCO, VCAN, AFB8, LGLAL5, HMOX1, TIMP1, OAS2, GAPDH, MSH3, FN1, NPC2, JUND, CHI3L1, GPNB4, SYTL2, CASP1, S100A8, MYO10, IGBP3, APCDD1, COL6A3, FABP5, PRDX3, CLEC1B, DDI4, CXCL10, CXCL8 | 27                 |
| 11 | Gurudeeban Selvaraj et al., 2021 [32] | VEGFA, AKT1, MMP9, ICAM1, CD44 | 5                  |
| 12 | Md. Shahriare Satu et al., 2021 [24] | MX1, IRF7, BST2 | 3                  |
| 13 | Tasnimul Alam Taz et al., 2020 [25] | BIRC3, ICAM1, IRAK2, MAP3K8, S100A8, SOCS3, STAT5A, TNF, TNFAIP3, TNIP1 | 10                 |
| 14 | Mohammad Ali Moni et al., 2020 [26] | JUN, XPO1, NPM1, HNRNPA1 | 4                  |
| 15 | Tania Islam et al., 2020 [27]   | MMP13, NLRP3, GBP1, ADORA2A, PTAFR, TNF, MLNR, IL1B, NFKBIA, ADRB2, IL6, IGFB2, HINT1, MAPK10, SGCE, HDAC5, SGCA, SGCB, CFD, ITSNI1, EHMT2, CLU, ILSR, PGM5, ANK2, HDAC9, SYT11, MDH1, SCDPDH, SIRT6, DNTA, FN1, ARRB1, MAGED2, TEX264, VEGFC, HK2, TXNL4A, SLCL16A3, NUDT21, TRA2B, HNRNPA1, CDC40, THOC1, PFKFB3 | 34                 |
| 16 | Ge C et al., 2020 [10]     | STAP1, CASP5, FDCSP, CARD17, ST20, AKR1B10, CLC, KCNJ2-AS1, RNASE2, FLG | 10                 |
| 17 | Aishwarya et al., 2020 [11]  | STAP1, CASP5, FDCSP, CARD17, ST20, AKR1B10, CLC, KCNJ2-AS1, RNASE2, FLG | 10                 |
| 18 | Saxena, A. et al., 2020 [12]  | STAP1, CASP5, FDCSP, CARD17, ST20, AKR1B10, CLC, KCNJ2-AS1, RNASE2, FLG | 10                 |
| 19 | Tao Q et al., 2020 [13]      | MAPK3, MAPK1, MAPK8, IL10, TNF, CXCL8, IL6, PTGS2, TP53, CCL2, CASP3, IL1B | 12                 |
| SL | Articles & Datasets | Hub-Genes/Proteins | Number of Proteins |
|---|---|---|---|
| 21 | Zhang N et al., 2020 [14] | CXCL10, ISG15, DDX58, MX2, OASL, STAT1, RSAD2, MX1, IRF7, OAS1 | 10 |
| 22 | Han L et al., 2020 [15] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CAPS3, PTGS2, TP53, MAPK1 | 10 |
| 23 | Tian J et al., 2020 [33] | CXCL8, CXCL2, CXCL10, ADRA2A, ADRA2C, CHRM2, PTGER3, OPRM1, OPRD1, UNJ, SMAD3, STAT1, SH3KB1, HDGC, TUBB, NFkB2, ETS1, UBC, TUFM, TRAF3, CCT5, RPL9, TUBB4B, CSNK1E, S100A9 | 15 |
| 24 | Jha PK et al., 2021 [34] | ELANE, MPO, ARG1, DEFA4,CAMP, MMP9, LTF, LCN2,PGLYRP1,HP | 10 |
| 25 | Ramesh P et al., 2020 [35] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 11 |
| 26 | Tian J et al., 2020 [33] | CXCL8, CXCL2, CXCL10, ADRA2A, ADRA2C, CHRM2, PTGER3, OPRM1, OPRD1, UNJ, SMAD3, STAT1, SH3KB1, HDGC, TUBB, NFkB2, ETS1, UBC, TUFM, TRAF3, CCT5, RPL9, TUBB4B, CSNK1E, S100A9 | 15 |
| 27 | Li Zhonglin et al., 2020 [36] | DDOST, UPF1, HIST2H2A, ITGAL, EGFR, CXCL1, DYNLL1, POLR2F, RPL13A, FBXO11, CSNK1E | 11 |
| 28 | Prasad K et al., 2021 [38] | MOV10, NFX1, APP, ELAVL1, CUL3, XPO, TP53, EGFR, MCM2, MYC, COPS5, ESR1, UBC, FN1, CUL7, VCAM1, RNF2, CUL1, SIRT7, CAND1, OBSL1, HSP90AA1, CDK2, NPM1, GRB2, FBXO6, CDC5L, GABARAPL2, VCP, CCD8, GABARAPL1, CUL2, SNA1, ACTB, CFB, LCA1, CETP, TLN1, SAA2, FGL1, CF1, YWHAZ, YWHAE, AZGP1, S100A8, CFHR1, CFHR3, PON3, PRDX6, ARHGID1B, TAGLN2, TRIM33, TUBB1, SH3BGR1L3 | 35 |
| 29 | Fangzhou Liu et al., 2021 [39] | AKTI, TP53, TNF, IL6, BCL2L1, ATM | 6 |
| 30 | Prasad K et al., 2020 [40] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 11 |
| 31 | Ke-Ying Fang et al., 2021 [41] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 11 |
| 32 | Zhen-Zhen Wang et al., 2021 [47] | TNF, EGFR, CASP9, EGFA, NFKB1, TP53, IL6, CASP3, MAPK8, PTGS2, GAPDH, CCL2, NFkBIA, MMP9, MMP2, CCND1, MCL1, MAPK1, MYC, CXCL8, JUN, CASP8, PPARG, IL1B | 24 |
| 33 | Shenglong Li et al., 2020 [43] | RELA, TNF, IL6, IL1B, MAPK14, TP53, CXCL8, MAPK3, MAPK1, IL4, MAPK8, CAPS3 and STAT1 | 13 |
| 34 | Zhen-Zhen Wang et al., 2021 [47] | IL1b and IL6 | 2 |
| 35 | Zhen-Zhen Wang et al., 2021 [47] | VEGFA, TNF, IL-6, CXCL8, IL-10, CCL2, IL1B, TLR4, ICAM1, MMP9 | 10 |
| 36 | Yi-Wei Zhu et al., 2020 [45] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 13 |
| 37 | Z. Bao et al., 2021 [46] | NFkBIA, BUB3, EIF2S3, GADD45A, MET, MCL1, SOCS3 | 7 |
| 38 | Mostafa Rezaei-Tavirani et al., 2021 [42] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 13 |
| 39 | Mostafa Rezaei-Tavirani et al., 2021 [42] | VEGFA, TNF, IL-6, CXCL8, IL-10, CCL2, IL1B, TLR4, ICAM1, MMP9 | 10 |
| 40 | Z. Bao et al., 2021 [46] | NFkBIA, BUB3, EIF2S3, GADD45A, MET, MCL1, SOCS3 | 7 |
| 41 | Z. Bao et al., 2021 [46] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 13 |
| 42 | Z. Bao et al., 2021 [46] | NFkBIA, BUB3, EIF2S3, GADD45A, MET, MCL1, SOCS3 | 7 |
| 43 | Z. Bao et al., 2021 [46] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 13 |
| 44 | Z. Bao et al., 2021 [46] | NFkBIA, BUB3, EIF2S3, GADD45A, MET, MCL1, SOCS3 | 7 |
| 45 | Z. Bao et al., 2021 [46] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, TP53, MAPK1 | 13 |
| 46 | Z. Bao et al., 2021 [46] | NFkBIA, BUB3, EIF2S3, GADD45A, MET, MCL1, SOCS3 | 7 |
| 47 | Z. Bao et al., 2021 [46] | IL6, TNF, IL10, MAPK8, MAPK3, CXCL8, CASP3, PTGS2, GAPDH, CCL2, NFkBIA, MMP9, MMP2, CCND1, MCL1, MAPK1, MYC, CXCL8, JUN, CASP8, PPARG, IL1B | 24 |

**Common genes in at least 5 articles**
- CXCL8, IL6, TNF, TP53, IL1B, MMP9, NFkBIA, PTGS2, ICAM1, STAT1, CCL2
| SL | Articles & Datasets | Hub-Genes/Proteins | Number of Proteins |
|----|---------------------|--------------------|--------------------|
|    | Common genes in at least 6 articles | CXCL8, IL6, TNF, TP53, IL1B, MMP9 | 6 |
|    | Common genes in at least 7 articles | CXCL8, IL6, TNF, TP53, IL1B | 5 |
|    | Common genes in at least 9 articles | CXCL8, IL6, TNF | 3 |
|    | Common genes in at least 11 articles | CXCL8, IL6 | 2 |

2.1.2. Collection of Drug Agents

Among the 3410 FDA-approved antiviral drugs, the top listed SARS-CoV-2 3CL protease-guided 88 drugs that were collected from Beck et al., 2020 [52] were considered as drug Set A. We have also collected a total of 87 published drugs from our reviewed articles (Table 1) and consider them as Set B. Then, we have taken common and uncommon drugs from Set A and B, (AUB) = 145 drugs (Supplementary File S1). These drugs would be evaluated against the potential drug targets by molecular docking simulation study.

2.2. Methods

The integrated bioinformatics and system biology analysis approaches were utilized as described below. This widely used analytical approach has been utilized in transcriptome guided network-based analysis to identify leading biomolecules [50,53,54].

2.2.1. Protein–Protein Interaction (PPI) Network Analysis of Hub-DEGs

The PPI network analyses of DEGs are now widely being used to explore key proteins. The PPI network that is based on all sets of Hub-DEGs encoded proteins was constructed using the STRING database [55] to detect hHub-DEGs. The NetworkAnalyst and Cytoscape 3.7.2 (Boston, MA, USA) [53] were utilized to perform topological analyses of PPI network and visualization.

2.2.2. Functional and Pathway Enrichment Analysis of hHub-DEGs

Functional enrichment, annotation, and over-representation analysis [54,56–58], namely, biological processes (BP), molecular functions (MF), and cellular components (CC) and the Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways were retrieved from the hHub-DEGs. The functional and pathway enrichment analysis was performed by using g:GOSt software that was implanted in the g:Profiler web server to reveal the significant GO terms of BP, MF, CC, and KEGG pathways of hHub-DEGs for SARS-CoV-2 infections. Statistical significance was defined by the adjusted p-values < 0.05 and the Benjamini and Hochberg [59] procedure was considered for FDR controlling.

2.2.3. Regulatory Network Analysis of hHub-DEGs

To explore Hub-TFs of hHub-DEGs, we constructed the interaction network between TFs and hHub-DEGs based on the JASPAR database [60]. Similarly, the miRNA and hHub-DEGs interaction network was constructed based on the TarBase V8.0 [61] database. These key regulatory biomolecules were selected based on the highest topological matrices (degree of connectivity and betweenness) that were applied on the interaction network by using the NetworkAnalyst tool [53].

2.2.4. Association of hHub-DEGs with Comorbidities

The prognostic performance of hHub-DEGs based on lung cancer patient data was observed by SurvExpress [62], an online biomarker validation tool. The SurvExpress utilizes the log rank statistic to test the significance.
2.2.5. Drug Repurposing by Molecular Docking Simulation

Basically, the molecular docking simulation interprets the potential drug components based on computational binding affinity with the drug target molecules. In this study, the docking analysis was performed among the drug target key receptors biomolecule (i.e., hHub-DEGs, TFs) and 145 drug agents or small compounds (Supplementary File S1). The molecular docking simulation requires 3-Dimensional (3D) structures of both receptor proteins and candidate drugs.

The 3D structures of receptors were downloaded from the Protein Data Bank (PDB) [63] and SWISS-MODEL [64]. The PubChem database [65] was used to retrieve the 3D structures of 145 meta-drug agents. The “Discovery Studio Visualizer” was used to visualize the 3D structures of the protein interfaces [66]. The receptor proteins were preprocessed by removing solvent molecules (water) and all hetero atoms, and adding charges using AutoDock tools and also a grid box was generated over the entire surface of the receptors [67]. The drug agents were minimized energy through the Avogadro using the mmff94 force field along with the steepest descent optimization algorithm and a total number of 200 minimization steps [68] and pre-processed by AutoDock tools which convert each ligand into an acceptable format PDBQT [67], respectively. Subsequently, molecular docking between the receptors and drug agents were performed to calculate their binding affinities (kcal/mol) by using AutoDock Vina [69]. The receptor-ligand complexes with binding affinities that were less that \(-7.0\) (kcal/mol) were considered as the significant complexes. The number of iterations per pocket was used with the default value of 8. The PyMol [70] was used to analyze the docked complexes for surface complexes, types, and distances of non-covalent bonds.

Then, we validated our proposed drug agents by a molecular docking study with the top listed 11 previously published receptor proteins (CXCL8, IL6, TNF, TP53, IL1B, MMP9, NFKBIA, PTGS2, ICAM1, STAT1, CCL2) for novel SARS-CoV-2 infections (Table 1). Each of these 11 receptors was detected as a hub-gene/protein in at least 5 articles. There was no other independent receptor that is published in more than 5 articles.

3. Results
3.1. Basic Characteristics of the Selected Studies

In this meta-data analysis, we have collected mainly the hub-DEGs information of the published articles that were related to the COVID-19. To identify the dysregulated genes, most of the articles analyzed the RNA-Seq and/or microarray transcriptomics data of COVID-19 from Gene Expression Omnibus (GEO) and some were collected from various published sources (i.e., different databases, other published sources). One article used the SNP data to identify the significant target genes for COVID-19. The LIMMA, DESeq2, classical t-test, and EdgeR were the most used statistical tests to identify the DEGs from the gene expression datasets. Additionally, some articles analyzed other datasets of different comorbidities and compared the findings with COVID-19 (Supplementary Table S1). The drug information for COVID-19 treatments were also reported in some published articles. We have found 531 hub-DEGs in total (i.e., 370 unique genes) from the reviewed articles in this study that were used in PPI network.

3.2. Identification of Hub of Hub-Proteins (hHub-Proteins)

To identify the hHub-proteins, the PPI network analysis was utilized by using all collected the hub-DEGs from the selected articles. The PPI network of the hub-DEGs revealed the key hHub-proteins that were chosen based on the degree of connectivity, closeness, and betweenness of the nodes in the network. The top 14 hHub-proteins are AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNF, TP53, UBA52, UBC, and VEGFA (Figure 2) that were found by PPI network. These top hHub-proteins would be focused for the pre-clinical potential drug target molecule that may open a new era of therapeutic targets.
the SNP data to identify the significant target genes for COVID-19. The LIMMA, DESeq2, classical t-test, and EdgeR were the most used statistical tests to identify the DEGs from the gene expression datasets. Additionally, so me articles analyzed other datasets of different comorbidities and compared the findings with COVID-19 (Supplementary Table S1). The drug information for COVID-19 treatments were also reported in some published articles. We have found 531 hub-DEGs in total (i.e., 370 unique genes) from the reviewed articles in this study that were used in PPI network.

3.2. Identification of Hub of Hub-Proteins (hHub-Proteins)

To identify the hHub-proteins, the PPI network analysis was utilized by using all collected the hub-DEGs from the selected articles. The PPI network of the hub-DEGs revealed the key hHub-proteins that were chosen based on the degree of connectivity, closeness, and betweenness of the nodes in the network. The top 14 hHub-proteins are AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNF, TP53, UBA52, UBC, and VEGFA (Figure 2) that were found by PPI network. These top hHub-proteins would be focused for the pre-clinical potential drug target molecule that may open a new era of therapeutic targets.

Figure 2. Protein–protein interaction (PPI) network analysis of Hub-DEGs-detected hHub-DEGs. The nodes in octagon shape with a blue color indicate the hHub-DEGs and a small ellipse indicates hub DEGs.

3.3. Functional and Pathway Enrichment Analysis of hHub-DEGs

The GO functional enrichment analysis revealed that our proposed hHub-DEGs were significantly enriched with abundant number of biological processes (BPs), molecular functions (MFs), and cellular components (CCs) (Table 2). Table 2 shows top 10 significantly enriched GO-terms for each of three categories (MFs, BPs, and CCs). These functions and pathways are highly connected with the COVID-19-related biological functional pathways in the host which are crucial for developing therapeutic targets. The major GO molecular functions (MF) are namely, enzyme binding, identical protein binding, phosphatase binding, cytokine receptor binding; the major GO biological process (BP) showed that the positive regulation of cellular biosynthetic process, regulation of DNA-binding transcription factor activity, positive regulation of protein phosphorylation, positive regulation of protein modification process, response to endogenous stimulus, and cellular response to organic substance. Moreover, the intracellular organelle lumen, membrane-enclosed lumen, membrane endosome, endoplasmic reticulum, and cell periphery are the significant cellular areas for the hHub-DEGs showed in the functional enrichment analysis (Table 2).
Table 2. The top 10 significantly enriched GO terms for each of BPs, MFs, and CCs based on the hHub-DEGs-set enrichment analysis, where hHub-DEGs-set consisted of 14 genes.

| Source | GO Term ID | Description | P_adj-Value | Gene Count | Enriched Genes |
|--------|------------|-------------|-------------|------------|----------------|
|        | GO:0019899 | enzyme binding | 0.00000000 | 11 | AKT1, APP, EGFR, INS, JUN, MAPK1, STAT3, TNF, TP53, UBA2, UBC |
|        | GO:0098772 | molecular function regulator activity | 0.00000000 | 10 | AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, TNE, TP53, VEGFA |
|        | GO:0042802 | identical protein binding | 0.00000000 | 10 | AKT1, APP, EGFR, INS, JUN, MAPK1, STAT3, TNE, TP53, VEGFA |
| GO:MF  | GO:0005102 | signaling receptor binding | 0.00000000 | 9 | APP, CXCL8, EGFR, IL6, INS, STAT3, TNE, TP53, VEGFA |
|        | GO:0019602 | phosphatase binding | 0.00000003 | 5 | AKT1, EGFR, MAPK1, STAT3, TP53 |
|        | GO:0030546 | signaling receptor activator activity | 0.00000003 | 6 | APP, CXCL8, IL6, INS, TNE, VEGFA |
|        | GO:0005515 | protein binding | 0.00000008 | 14 | AKT1, JUN, MAPK1, STAT3, TP53, UBA2, UBC, VEGFA |
|        | GO:0005126 | cytokine receptor binding | 0.00000010 | 5 | CXCL8, IL6, STAT3, TNE, VEGFA |
|        | GO:0044389 | ubiquitin protein ligase binding | 0.00000015 | 5 | EGFR, JUN, TP53, UBA2, UBC |
|        | GO:0005768 | organelle lumen | 0.00000000 | 12 | AKT1, APP, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNE, TP53, UBA2, UBC |
|        | GO:0005783 | endoplasmic reticulum | 0.00000003 | 12 | AKT1, APP, CXCL8, EGFR, IL6, INS, MAPK1, STAT3, TNE, TP53, UBA2, UBC |
| GO:BP  | GO:0010562 | positive regulation of cell proliferation | 0.00000000 | 12 | AKT1, APP, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNE, TP53, UBA2, UBC |
|        | GO:0045937 | positive regulation of phosphorus metabolic process | 0.00000000 | 12 | AKT1, APP, EGFR, IL6, INS, MAPK1, STAT3, TNE, TP53, UBA2, UBC |
|        | GO:0031401 | positive regulation of protein modification process | 0.00000000 | 12 | AKT1, APP, EGFR, IL6, INS, MAPK1, STAT3, TNE, TP53, UBA2, UBC |
|        | GO:0009719 | response to endogenous stimulus | 0.00000000 | 13 | AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNE, TP53, UBA2, UBC |
|        | GO:0071310 | cellular response to organic substance | 0.00000000 | 14 | AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNE, TP53, UBA2, UBC |

On the other hand, the hHub-DEGs shared significant KEGG pathways, noticeably Kaposi sarcoma-associated herpesvirus infection, AGE-RAGE signaling pathway in diabetic complications, Human cytomegalovirus infection, Shigellosis, Hepatitis B, Lipid and atherosclerosis, Coronavirus disease-COVID-19, HIF-1 signaling pathway, and pathways in cancer (Figure 3). Among the top significant KEGG pathways, “the Human cytomegalovirus infection”, “Shigellosis”, and “Coronavirus disease-COVID-19” (Figure 3) are the most important pathways which are crucial for mortality due to COVID-19 infection.
cytomegalovirus infection”, “Shigellosis”, and “Coronavirus disease-COVID-19” (Figure 3) are the most important pathways which are crucial for mortality due to COVID-19 infection.

Figure 3. The KEGG pathways enriched by the proposed hHub-DEGs.

3.4. Transcriptional and Post-Transcriptional Regulatory Factors

The interaction network of regulatory TFs-target hHub-DEGs and the miRNA-hHub-DEGs show the substantial TFs and the miRNAs that regulate the hHub-DEGs. The transcription factors GATA2, FOXC1, TFAP2A, NFIC, and YY1 (Figure 4) and the miRNAs namely, miR-106a-5p, miR-17-5p, miR-20a-5p, miR-106b-5p, and miR-93-5p (Figure 5) were found from the network as key signaling and regulatory factors that were associated with SARS-CoV-2 infection in humans which play the key regulatory roles for hHub-DEGs (Figure 5).

Figure 4. TFs versus hHub-DEGs interaction network that was detected the key regulatory TFs of hHub-DEGs. Here, hHub-DEGs were marked as a blue color with octagon shape in both A and B. The key TFs proteins were marked as a red color with a hexagonal shape and small ellipses represents other TFs.
10 receptor proteins (Figure 6a). Therefore, we considered the top 10 lead compounds. The key TFs proteins were marked as a red color with a hexagonal shape and small ellipses represents other miRNAs.

3.5. Drug Repurposing by Molecular Docking

We selected 14 hub receptor proteins corresponding to our proposed 14 hHub-DEGs and 5 TFs proteins corresponding to the key regulators of those hHub-DEGs, as the $m = 19$ drug targets and 145 drug agents (Supplementary File S1). We downloaded the 3D structure of 15 drug target proteins (MAPK1, EGFR, CXCL8, STAT3, UBC7, IL6, AKT1, GATA2, TNF, YY1, UBA52, VEGFA, JUN, INS, and APP) from PDB [63] with source codes 6g54, 3ika, 4xdx, 6njs, 3h8k, 1l16, 2uzr, 5o9b, 1tnf, 1ubd, 2hth, 2vpf, 1jun, 6ver, and 4pwq, respectively, as well as the 3D structure of remaining four drug target TFs protein (TP53, FOXC1, NFIC, and TFAP2A) that were collected from SWISS-MODEL [64] using UniProt [71] ID of P04637, Q12948, P08651, and P05549, respectively. The 3D structures of 145 drugs were downloaded from the PubChem database [65] as mentioned previously.

Molecular docking analysis between our proposed receptors and meta-drug agents were performed to calculate the binding affinity scores. Then, for the selection of drug agents as candidate drugs, we ordered the target proteins according to the row sums of the binding affinity matrix $A = (A_{ij})$ and the column sums of the drug agents. Figure 6a shows the image of the docking score matrix $A = (A_{ni})$ corresponding to the ordered target proteins on the $Y$-axis and $n = 30$ top ranked drug agents on the $X$-axis. We observed that first two top lead compounds (lead1: Digoxin and lead2: Avermectin) produced binding affinities that were negatively larger than $-8.0$ kcal/mol with 15 receptor proteins out of 19. The next (3–10)th top lead compounds (lead3: Simprevir, lead4: Linifanib, lead5: Nelfinavir Mesylate, lead6: Atazanavir, lead7: Withaferin, lead8: Proscillaridin, lead9: Hesperidin, and lead10: Amuvatinib) produced binding affinities that were negatively larger than $-7.0$ kcal/mol with our proposed 12 receptor proteins out of 19. The remaining 20 lead compounds also produced good binding affinities with top ordered proposed 10 receptor proteins (Figure 6a). Therefore, we considered the top 10 lead compounds (Digoxin, Avermectin, Simprevir, Linifanib, Nelfinavir Mesylate, Atazanavir, Withaferin, Proscillaridin, Hesperidin, and Amuvatinib) as the most possible candidate drug agents.

Figure 5. The miRNAs versus hHub-DEGs interaction network that were detected the key regulatory miRNAs of hHub-DEGs. Here, hHub-DEGs were marked as a blue color with octagon shape in both A and B. The key miRNAs were marked as a red color with a hexagonal shape and small ellipses.
for SARS-CoV-2 infections and the 2D structures of these drug agents were provided in the Supplementary Table S2a as well as their pairwise ligand similarities scores in Supplementary Table S2b, were calculated by using the Tanimoto Coefficients (TC) [72,73].

Figure 6. Binding affinity scores that were calculated by Autodock-vina. The color bar indicates the score levels, where deeper and lighter reds indicated the strong and weak binding, respectively. (a) The binding affinity scores based on the top-ranked 30 drug agents out of 145 on the X-axis and ordered 19 target proteins (proposed) on the Y-axis (Supplementary File S2). (b) The binding affinity scores that were based on the top-ranked 30 drug agents out of 145 on the X-axis and top-ranked 11 target proteins (previously published) on the Y-axis; see details in (Supplementary File S3). The receptors with blue color indicate the common receptors between the proposed and previously published top-ranked receptors.

To investigate the binding performance of the proposed candidate drugs against the state-of-the-art alternative independent receptors, we considered SARS-CoV-2 infection causing top-ranked 11 hub-DEGs (CXCL8, IL6, TNF, TP53, IL1B, MMP9, NFKBIA, PTGS2, ICAM1, STAT1, CCL2) that were previously published in at least five articles (Table 1). The 3D structures of these 11 proteins were collected from the Protein Data Bank (PDB) with codes 2vpf, 1il6, 1tnf, 4mzi, 9ilb, 1gkc, 1nfi, 1cx2, 1p53, 1bf5, and 3ifd, respectively. Then, molecular docking interactions of the top drug agents with published 11 KGs were performed. Their binding affinities (kcal/mol) were visualized in Figure 6b. Then we ordered the publicly available receptor proteins in descending order of row sums of the binding affinity matrix $A = (A_{ij})$ and drug agents according to the column sums, in the same manner as before. Then, we observed that first ten top compounds that produced binding affinities that were negatively larger than \(-7.0\) kcal/mol with all published independent receptor proteins. Then, we selected common nine lead compounds (Digoxin,
Avermectin, Simeprevir, Nelfinavir Mesylate, Proscillaridin, Linifanib, Withaferin, Amuvatinib, Atazanavir) as the candidate drug agents from the top-ranked 10 lead compounds as displayed in Figure 6a,b for SARS-CoV-2 infections.

For example, Table 3 shows the summary results of the interacting properties of two potential target proteins (MAPK1 and EGFR) with the top three lead compounds (Digoxin, Avermectin, Simeprevir) that achieved the highest binding affinity scores. The interacting complex (3D), schematic diagram (2D), and the interaction neighbor residues (with 4 Å radius of drug) are represented in Table 3. The complex MAPK1-Digoxin was formed with two conventional hydrogen bonds at ARG191 and TRP192, five alkyl bonds at ALA52, LEU170, VAL39, LUE156, and ILE84. The complex between EGFR and Avermectin was formed with one conventional hydrogen bond at VAL876; one carbon hydrogen bond at ASP855; five alkyl bonds at VAL726, LYS745, ILE878, LYS879, and ARG858; and only one pi-alkyl bond at PHE723. The complex MAPK1-Simeprevir was formed two conventional hydrogen bonds at LYS151 and SER153; two pi-sigma bonds at ILE31 and LEU156; and five alkyl bonds at ALA52, VAL39, LYS54, LEU107, and LEU156. The docking poses of the proposed 10 ligands with the proposed top ordered six targets (MAPK1, EGFR, CXCL8, STAT3, UBC7, TP53) were displayed in Supplementary Figure S2. We observed that our suggested top-ranked 10 ligands significantly bind to the same pockets of the top-ranked two targets MAPK1 and EGFR (Supplementary Figure S1a,b). Few ligands bind to the different pockets for the (3–6)th-ranked targets (UBC7, CXCL8, STAT3, TP53) (Supplementary Figure S1c–f). Similarly, we also observed that previously published the top-ranked four receptors (uncommon) NFKBIA, CXCL8, MX1, and IRF7 are significantly targeted by our suggested ligands (Supplementary Figure S2a–d).
Table 3. Interacting properties of top-ranked three drug-target complexes. The fourth, fifth, and 6th columns displayed the surface view, binding poses, interacting mode, respectively. The 7th, 8th, and 9th columns showed the interacting residues, bonding types, and distances, respectively.

| Potential Targets | Structure of Ligand | Binding Affinity (kCal/mol) | Surface View of Complex | Pose View of Complex | Target Ligand Interaction | Interacting Amino Acid | Bond Type | Distance (Å) |
|------------------|--------------------|-----------------------------|-------------------------|---------------------|--------------------------|------------------------|-----------|-------------|
| MAPK1            | Digoxin            | −11.0                       |                         |                     |                          | ARG191                 | CH        | 2.55        |
| EGFR             | Avermectin         | −10.8                       |                         |                     |                          | VAL876                 | CH        | 2.068       |
| MAPK1            | Simprevir          | −10.3                       |                         |                     |                          | LYS151                 | CH        | 2.176       |
4. Discussion

Among the selected 39 published studies, it was commonly suggested the drug target proteins, called hub-proteins, encoded from the hub-DEGs for COVID-19. We have observed that the reported hub-DEGs were not consistent from the COVID-19 perspective. In total, 370 unique hub-DEGs were found from 41 articles in this study. The reported hub-DEGs were not consistent among the articles due to the different conditions of data collection (Table 1). The crucial key biomarkers play the vital role of biological functions in all the living organs. The deeper understanding of the molecular mechanism of these potential biomarkers is essential for all diseases. The recent transcriptional data analysis-based research is being utilized to focus on the pre-clinical prominent drug candidate biomarkers [4–7]. Due to reporting inconsistency among the published drug target hub-DEGs of COVID-19 host cell, the current study focused on identifying global key hub-DEGs (hHub-DEGs) using the metadata of published articles through a multi-omics data integration framework (Figure 1). The prominent substantial hHub-proteins and their associated regulatory TFs and miRNAs were identified which are related to SARS-CoV-2 infection and proliferation. The metadata analysis identified 14 key hHub-DEGs, 5 TFs, and 5 miRNAs as the top regulatory factors using an integrated bioinformatics framework analysis.

The hHub-DEGs were identified using the PPI network analysis of the selected published 370 unique hub-DEGs which revealed 14 key hHub-DEGs namely AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNF, TP53, UBA52, UBC, and VEGFA. The AKT1 gene encoded protein-kinase supports cell growth and proliferation, cell differentiation, and cell survival. This protein also helps normal development and function of the nervous system and also has the potential to cause normal cells to become cancerous when it mutates [74–77]. Recent studies show that the AKT1 gene is one potential drug target for COVID-19 treatment and also has a greater involvement in SARS-CoV-2 viral infection related complexity [78–80]. Amyloid Protein Precursor (APP) gene duplication is associated with Cerebral Amyloid Angiopathy which leads to intracerebral hemorrhagic stroke. This triggers the critical condition for COVID-19-infected persons that have Alzheimer’s, Down syndrome, and cardiovascular disease [81,82]. CXCL8 (also known as IL-8) is a major inflammatory mediator and its receptors are highly connected with the development of various colorectal cancer and its liver metastases [83] as well as it enhances the major human neutrophils conscription [84]. The cell growth process and lung cancer development is influenced by the EGFR gene [85]. Studies have suggested that the EGFR protein incorporates the novel coronaviruses by making bonds with S protein which facilitates the endocytosis process into the host cell [86,87] by SARS-CoV-2. The EGFR protein also triggers the MAPK pathways after binding with the spike protein of SARS-CoV-2 virus which is also important for viral entry [88]. The MAPK family proteins are also associated with the cytokine signaling after COVID-19 infection [89,90]. The MAPK1 gene is related with the regulation of immunity and inflammation [15] when the TNF is linked with the immune regulatory, pro-inflammatory [70,91], and anti-viral functions [92]. One of the most important hHub-DEGs, IL6, is related to the interleukin (IL) regulatory pathway genes which are critical for the significant pathophysiological functional mechanisms, namely systemic inflammation and cytokine release syndrome [91,93,94] which plays the major role in the cytokine storm [95,96]. The IL-6 can also be used as an early warning diagnosis and treatment index of COVID-19 [97,98] and also treated as the molecule for assessing the severity of infection and judge prognosis [99,100] and the response to treatment [101,102]. The c-Jun type gene, namely JUN is one of the important host proteins involving in HCoV infectious bronchitis virus [28,103]. The TP53 and UBC hHub-DEGs are involved in cell deaths that occurs from protein misfolding and aggregation [38]. The IFN-γ-mediated signaling function, apoptosis, and proteasomal degradation of CD4+ T-cells are mediated by the vital proteins namely, TP53, and CASP3, UBA52, and UBC, respectively [104]. The study revealed that the cancer growth is suppressed and the radio sensitivity is amplified by the activities of ubiquitin C (UBC) in NSCLC cells [105] The above discussions and the importance of the hHub-DEGs clearly demonstrated that these are the most important pro-
teins for COVID-19 infection, diagnosis, and proliferation which shows them as significant drug target proteins for COVID-19 treatment.

The functional annotation and enrichment analysis of the hHub-DEGs showed significant enrichment in various biological, molecular, and cellular functions into the host cells. Most of the hHub-DEGs showed involvement in different functional pathways. The top significant molecular functions demonstrated that the hHub-DEGs are associated with different binding activities which included proteins, enzymes, phosphatase, and signaling receptors (Table 2). The binding activities indicates the regulatory and functionality of the hHub-DEGs. Most importantly, the hHub-DEGs enriched the cytokine receptor binding and ubiquitin protein ligase binding molecular functions (MF) which are signifying the COVID-19-induced cytokine storm in the affected patients. The top Gene Ontology (GO) terms of biological processes (BP) are suggesting that the positive regulation of cellular biosynthetic process, regulation of DNA-binding transcription factor activity, and the protein phosphorylation-associated regulation and metabolic functions are the most significant (Table 2). The other top significant BPs, namely the positive regulation of protein modification process, response to endogenous stimulus, and the cellular response to organic substance is also enriched by our identified hHub-DEGs. The cellular presence of the proposed hHub-DEGs reveal that the cellular lumen, membrane, endosome, endoplasmic reticulum, cell periphery, extracellular areas, are significantly enriched.

The KEGG pathway enrichment analysis of the hHub-DEGs represents the top significant pathways that are shared by the hHub-DEGs are namely, Kaposi sarcoma-associated herpesvirus infection, AGE-RAGE signaling pathway in diabetic complications, Human cytomegalovirus infection, Shigellosis, Hepatitis B, Lipid and atherosclerosis, Coronavirus disease-COVID-19, HIF-1 signaling pathway, and pathways in cancer (Figure 3). Most of the hHub-DEGs are involved in the top 10 significant KEGG pathways. Among the enriched significant pathways, the Coronavirus disease-COVID-19-related pathways is the most significant pathway which involved most of the hHub-DEGs notably, CXCL8, EGFR, IL6, JUN, MAPK1, STAT3, TNF, and UBA52. The functions and pathway enrichment analyses are representing the significance of the hHub-DEGs in that they are very crucial to COVID-19 infection, diagnosis, and treatment.

The TFs vs. hHub-DEGs interaction network revealed that the GATA2, FOXC1, TFAP2A, NFIC, and YY1 are prominent and important TFs that are associated with the hHub-DEGs (Figure 5). Among the TFs, the Alzheimer’s disease, basal-like breast cancer (BLBC), and tissue invasion are highly related with FOXC1 TF [106,107]. The GATA2 TF is related to kidney and breast cancer-related pathway when the higher expression pattern of YY1 TFs increases the tumour size and higher TNM stage [108–110]. The contribution of TFAP2A was found for the malignant progression of lung cancer [111]. The NFIC TFs have greater involvement with the tumor genesis of breast cancer, gastric cancer, and glioma [112–114]. Also, the identified TFs have a significant involvement in various biological functions and pathways [4–7,115]. The interaction network among the miRNAs and hHub-DEGs fetched the five key miRNAs (miR-106a-5p, miR-17-5p, miR-20a-5p, miR-106b-5p and miR-93-5p) that were linked with the hHub-DEGs-encoded proteins. The miR-106a-5p miRNA is highly interconnected with the CD4+ T-cells regulation and works as a tumor suppressor by regulating the VEGFA gene [116,117]. The miR-106b-5p and miR-17-5p miRNAs are related to different cancers and bone formation pathways (i.e., cervical and breast cancer) when the miR-20a-5p, miR-106b-5p, and miR-93-5p are related to cervical cancer [118–121]. The survival analysis showed that the prognostic power of the hHub-DEGs clearly discriminate between the low and high-risk groups (Supplementary Figure S3). The survival curve was based on lung cancer data which indicates that the patients with lung-related comorbidities have a greater risk to be infected and higher mortality rate compared to others.

Based on the molecular docking binding scores, the top ranked nine drugs (Digoxin, Avermectin, Simeprevir, Nelfinavir Mesylate, Proscillaridin, Linifanib, Withaferin, Amuvatinib, Atazanavir) were selected as potential drug candidates for the preventing of SARS-
CoV-2 infection, where the first three drugs (Digoxin, Avermectin/Ivermectin, Simeprevir) showed the strongest binding affinities with maximum target proteins (Figure 6a,b). Studies suggested that COVID-19-affected patients had a greater risk of occurrence of congestive heart failure (CHF), atrial fibrillation or flutter, and certain cardiac arrhythmias which increases the morbidity and mortality significantly [122,123]. In these cases, the Digoxin has been injected to reduce the heart disease risk, entrance of coronavirus into the cells, and the suppression of a cytokine storm [124], although it has toxicity which causes the adverse events for instance vomiting, abdominal pain, dizziness, and delirium [122,125]. In spite of a lack of deeper knowledge about the activities of Avermectin/Ivermectin against SARS-CoV-2 virus, it has been practiced to treat COVID-19-affected patients [126] during the pandemic situation of the last couple of years. Ivermectin has been widely used for the treatment of virus infection [127,128] and many researchers have suggests that it has potential therapeutic power for COVID-19 also [129,130]. On the other hand, Simeprevir, a protease inhibitor has been used for hepatitis C virus (HCV) infection treatment [131,132]. The literature supports that Simeprevir inhibits SARS-CoV-2 viral replication by targeting the important viral polyproteins and cleaves them in vitro condition [133,134]. On the other hand, molecular docking analysis for the two top ranked proteins, MAPK1 and EGFR, showed maximum binding affinity with the top ranked drug components (Figure 6). The MAPK1 and the EGFR are very important proteins that are associated with the COVID-19 infection as discussed above. Therefore, the most importantly proposed three drug candidates might play a significant role for therapeutic development against SARS-CoV-2 variants. Moreover, further diverse research and experimental wet lab validations are being emphasized to investigate the effectiveness of the proposed target proteins and the drugs in clinical trials.

5. Conclusions

The global crisis due to the COVID-19 pandemic has caused millions of deaths and great economic losses. The analyses revealed the drug target proteins in host cells which were not conserved across the studies. Therefore, the current study focused on accumulating the published information about the drug target proteins to decode the more comprehensive drug target molecules in host human cells using an integrated bioinformatics approach. We identified 14 hHubGs (AKT1, APP, CXCL8, EGFR, IL6, INS, JUN, MAPK1, STAT3, TNF, TP53, UBA52, UBC, VEGFA) that were related to COVID-19 in host cells along with 5 TFs (GATA2, FOXC1, TFAP2A, and YY1) as the transcriptional and 5 miRNAs (miR-106a-5p, miR-17-5p, miR-20a-5p, miR-106b-5p, and miR-93-5p) as the post-transcriptional regulators. The GO functional and KEGG pathway enrichment analysis showed that the hHub-DEGs are significantly associated with infections. The molecular docking simulation analysis showed the significant drugs based on their binding affinities with the drug target hHub-DEGs. Finally, hHub-DEGs guided the top ranked nine drugs (Digoxin, Avermectin, Simeprevir, Nelfinavir Mesylate, Proscillaridin, Linifanib, Withaferin, Amuvatinib, Atazanavir) which were identified by molecular docking simulation and cross-validation. These drugs need more attention for their experimental and clinical validation to be used as therapeutics for COVID-19 treatment.
Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/vaccines10081248/s1, Supplementary Table S1: Basic characteristics/information of the selected articles. Supplementary Table S2: (a) 2D structures of the proposed drug agents/molecules. (b) Tanimoto’s drug/chemical similarity coefficients.; Supplementary File S1: Total 145 drugs/drug chemical list that was used in this study for molecular docking.; Supplementary File S2: Molecular docking scores of top 14 proposed hHub-DEGs and the 145 drug elements.; Supplementary File S3: Molecular docking scores of top 11 published hub-DEGs and the 145 drug elements.; Supplementary Figure S1: Docking poses of the proposed 10 ligands with the proposed top ordered 6 targets (a) MAPK1, (b) EGFR, (c) CXCL8, (d) STAT3, € UBC7, and (f) TP53.; Supplementary Figure S2: Docking poses of the proposed 10 ligands with the previously published top ordered 4 targets (uncommon) (a) NFKBIA, (b) IRF7, (c) MX1, and (d) CASP3.; Supplementary Figure S3: Survival curve analysis of the proposed hHub-DEGs.

Author Contributions: Conceptualization: M.N.H.M. and M.P.M. Data curation: M.P.M. and M.A.I. Formal analysis: M.P.M., M.B.H. and M.K.K. Investigation: M.P.M., M.S.R and M.N.H.M. Methodology: M.P.M., M.B.H. and M.K.K. Visualization: M.P.M., M.K.K. and M.B.H. Project administration: M.N.H.M. Supervision: M.N.H.M. Writing—original draft: M.P.M., M.K.K. and M.S.R. Writing—review & editing: M.N.H.M., R.A.M. K.A. and J.G. All authors have read and agreed to the published version of the manuscript.

Funding: This work formed part of the first author’s PhD research at the University of Southern Queensland, Australia which has been supported by funding from the Australian Government Research Training Program Scholarship. Additional funding was received from the Ministry of Science and Technology (MOST) research project, Government of the People’s Republic of Bangladesh, for the fiscal year 2020-2021 (Funding Number: 489-ID).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: All data analyzed in this study was provided in Tables 1 and S1.

Conflicts of Interest: The authors have declared no competing interests.

References
1. Zhu, R.F.; Gao, Y.L.; Robert, S.H.; Gao, J.P.; Yang, S.G.; Zhu, C.T. Systematic Review of the Registered Clinical Trials for Coronavirus Disease 2019 (COVID-19). J. Transl. Med. 2020, 18, 274. [CrossRef] [PubMed]
2. Treatments and Vaccines for COVID-19|European Medicines Agency. Available online: https://www.ema.europa.eu/en/human-regulatory/overview/public-health-threats/coronavirus-disease-covid-19/treatments-vaccines-covid-19#authorised-medicines-section (accessed on 11 January 2021).
3. Santiago, J.A.; Bottero, V.; Potashkin, J.A. Dissecting the Molecular Mechanisms of Neurodegenerative Diseases through Network Biology. Front. Aging Neurosci. 2017, 9, 166. [CrossRef] [PubMed]
4. Rahman, M.R.; Islam, T.; Turanli, B.; Zaman, T.; Faruquee, H.M.; Rahman, M.M.; Mollah, M.N.H.; Nanda, R.K.; Arga, K.Y.; Gov, E.; et al. Network-Based Approach to Identify Molecular Signatures and Therapeutic Agents in Alzheimer’s Disease. Comput. Biol. Chem. 2019, 78, 431–439. [CrossRef] [PubMed]
5. Islam, T.; Rahman, R.; Gov, E.; Turanli, B.; Gulidian, G.; Haque, A.; Arga, K.Y.; Haque Mollah, N. Drug Targeting and Biomarkers in Head and Neck Cancers: Insights from Systems Biology Analyses. Omi. A J. Integr. Biol. 2018, 22. [CrossRef] [PubMed]
6. Shahjaman, M.; Rezanur Rahman, M.; Shahinul Islam, S.M.; Nurul Haque Mollah, M. A Robust Approach for Identification of Cancer Biomarkers and Candidate Drugs. Medicina 2019, 55, 269. [CrossRef] [PubMed]
7. Moni, M.A.; Islam, M.B.; Rahman, M.R.; Rashed-Al-Mahfuz, M.; Awal, M.A.; Islam, S.M.S.; Mollah, M.N.H.; Quinn, J.M.W. Network-Based Computational Approach to Identify Delineating Common Cell Pathways Influencing Type 2 Diabetes and Diseases of Bone and Joints. IEEE Access 2020, 8, 1486–1497. [CrossRef]
8. Xie, T.A.; Han, M.Y.; Su, X.R.; Li, H.H.; Chen, J.C.; Guo, X.G. Identification of Hub Genes Associated with Infection of Three Lung Cell Lines by SARS-CoV-2 with Integrated Bioinformatics Analysis. J. Cell. Mol. Med. 2020, 24, 12225–12230. [CrossRef] [PubMed]
9. Oh, J.H.; Tannenbaum, A.; Deasy, J.O. Identification of Biological Correlates Associated with Respiratory Failure in COVID-19. BMC Med. Genom. 2020, 13, 186. [CrossRef]
10. Ge, C.; He, Y. In Silico Prediction of Molecular Targets of Astragaloside IV for Alleviation of COVID-19 Hyperinflammation by Systems Network Pharmacology and Bioinformatic Gene Expression Analysis. Front. Pharmacol. 2020, 11. [CrossRef]
11. Aishwarya, S.; Gunasekaran, K.; Margret, A.A. Computational Gene Expression Profiling in the Exploration of Biomarkers, Non-Coding Functional RNAs and Drug Perturbagens for COVID-19. J. Biomol. Struct. Dyn. 2022, 40, 3681–3696. [CrossRef] [PubMed]
12. Saxena, A.; Chaudhary, U.; Bharadwaj, A.; Wahi, N.; Kalli, J.R.; Gupta, S.; Kumar, S.; Gupta, S.; Raj, U. A Lung Transcriptomic Analysis for Exploring Host Response in COVID-19. *J. Pure Appl. Microbiol.* **2020**, *14*, 1077–1081. [CrossRef] [PubMed]

13. Tao, Q.; Du, J.; Li, X.; Zeng, J.; Tan, B.; Xu, J.; Lin, W.; Chen, X. lin Network Pharmacology and Molecular Docking Analysis on Molecular Targets and Mechanisms of Huashi Baidu Formula in the Treatment of COVID-19. *Drug Dev. Ind. Pharm.* **2020**, *46*, 1345–1353. [CrossRef] [PubMed]

14. Zhang, N.; Zhao, Y.D.; Wang, X.M. CXCL10 an Important Chemokine Associated with Cytokine Storm in COVID-19 Infected Patients. *Eur. Rev. Med. Pharmacol. Sci.* **2020**, *24*, 7497–7505. [CrossRef]

15. Han, L.; Wei, X.X.; Zheng, J.Y.; Zhang, L.L.; Wang, X.M.; Yang, H.Y.; Ma, X.; Zhao, L.H.; Tong, X.L. Potential Mechanism Prediction of Cold-Damp Plague Formula against COVID-19 via Network Pharmacology Analysis and Molecular Docking. *Clin. Med.* **2020**, *15*, 78. [CrossRef]

16. Wang, Z.; Jiang, C.; Zhang, X.; Zhang, Y.; Ren, Y. Identification of Key Genes and Pathways in SARS-CoV-2 Infection Using Bioinformatics Analysis. *Res. Sq.* **2020**, [CrossRef]

17. Gu, H.; Yuan, G. Identification of Potential Key Genes for SARS-CoV-2 Infected Human Bronchial Organoids Based on Bioinformatics Analysis. *bioRxiv* **2020**, [CrossRef]

18. Soon Nan, K.; Karuppanan, K.; Kumar, S.; Alam, S. Identification of Common Key Genes and Pathways between COVID-19 and Lung Cancer by Using Protein-Protein Interaction Network Analysis. *bioRxiv* **2021**, [CrossRef]

19. Sardar, R.; Satish, D.; Gupta, D. Identification of Novel SARS-CoV-2 Drug Targets by Host MicroRNAs and Transcription Factors Co-Regulatory Interaction Network Analysis. *Front. Genet.* **2020**, *11*. [CrossRef] [PubMed]

20. Vastrad, B.; Vastrad, C.; Tengli, A. Identification of Potential MRNA Panels for Severe Acute Respiratory Syndrome Coronavirus 2 (COVID-19) Diagnosis and Treatment Using Microarray Dataset and Bioinformatics Methods. *3 Biotech* **2020**, *10*, 422. [CrossRef] [PubMed]

21. Chong, X.; Peng, R.; Sun, Y.; Zhang, L.; Zhang, Z. Identification of Key Genes in Gastric Cancer by Bioinformatics Analysis. *Biomed Res. Int.* **2020**, *2020*, 7658230. [CrossRef] [PubMed]

22. Prasad, K.; Khatoon, F.; Rashid, S.; Ali, N.; AlAsmari, A.F.; Ahmed, M.Z.; Alqahtani, A.S.; Alqahtani, M.S.; Kumar, V. Targeting Hub Genes and Pathways of Innate Immune Response in COVID-19: A Network Biology Perspective. *Int. J. Biol. Macromol.* **2020**, *163*, 1–8. [CrossRef] [PubMed]

23. Satu, S.; Khan, I.; Rahman, R.; Howlader, K.C.; Roy, S.; Roy, S.S.; Quinn, J.M.W.; Moni, M.A. Diseasome and Comorbidities Complexities of SARS-CoV-2 Infection with Common Malignant Diseases. *Brief. Bioinform.* **2021**, *22*, 1415–1429. [CrossRef] [PubMed]

24. Taz, T.A.; Ahmed, K.; Paul, B.K.; Kawser, M.; Akhtar, N.; Mahmud, S.M.H.; Moni, M.A. Network-Based Identification Genetic Effect of SARS-CoV-2 Infections to Idiopathic Pulmonary Fibrosis (IPF) Patients. *Brief. Bioinform.* **2021**, *22*, 1254–1266. [CrossRef] [PubMed]

25. Moni, M.A.; Quinn, J.M.W.; Sinnaz, N.; Summers, M.A. Gene Expression Profiling of SARS-CoV-2 Infections Reveal Distinct Primary Lung Cell and Systemic Immune Infection Responses That Identify Pathways Relevant in COVID-19 Disease. *Brief. Bioinform.* **2020**, *2020*, 1324–1337. [CrossRef]

26. Islam, T.; Rahman, M.R.; Aydin, B.; Beklen, H.; Arga, K.Y.; Shahjaman, M. Integrative Transcriptomics Analysis of Lung Epithelial Cells and Identification of Repurposable Drug Candidates for COVID-19. *Eur. J. Pharmacol.* **2020**, *887*, 173594. [CrossRef] [PubMed]

27. Zhou, Y.; Hou, Y.; Shen, J.; Huang, Y.; Martin, W.; Cheng, F. Network-Based Drug Repurposing for Novel Coronavirus 2019-NCoV/SARS-CoV-2. *Cell Discov.* **2020**, *6*, 14. [CrossRef]

28. Caradonna, A.; Patel, T.; Toleska, M.; Alabad, S.; Chang, S.L. Meta-Analysis of APP Expression Modulated by SARS-CoV-2 Infection via the ACE2 Receptor. *Int. J. Mol. Sci.* **2022**, *23*, 1182. [CrossRef] [PubMed]

29. Gu, H.; Yuan, G. Identification of Key Genes in SARS-CoV-2 Patients on Bioinformatics Analysis. *bioRxiv* **2020**, [CrossRef]

30. Gu, H.; Yuan, G. Identification of Key Genes and Pathways in the HPSC-Derived Lungs Infected by the SARS-CoV-2. *Res. Sq.* **2020**, [CrossRef] [PubMed]

31. Selvaraj, G.; Kaliamurthi, S.; Peslherbe, G.H.; Wei, D.Q. Identifying Potential Drug Targets and Candidate Drugs for COVID-19: Biological Networks and Structural Modeling Approaches. *F1000Research* **2021**, *10*, 127. [CrossRef] [PubMed]

32. Tian, J.; Sun, D.; Xie, Y.; Liu, K.; Ma, Y. Network Pharmacology-Based Study of the Molecular Mechanisms of Qixuekang in Treating COVID-19 during the Recovery Period. *Int. J. Clin. Exp. Pathol.* **2020**, *13*, 2677–2690.

33. Jha, P.K.; Vijay, A.; Halu, A.; Uchida, S.; Aikawa, M. Gene Expression Profiling Reveals the Shared and Distinct Transcriptional Signatures in Human Lung Epithelial Cells Infected With SARS-CoV-2, MERS-CoV, or SARS-CoV: Potential Implications in Cardiovascular Complications of COVID-19. *Front. Cardiovasc. Med.* **2021**, *7*, 1–15. [CrossRef]

34. Ramesh, P.; Veerappapillai, S.; Karuppasamy, S. Gene Expression Profiling of Corona Virus Microarray Datasets to Identify Crucial Targets in COVID-19 Patients. *Gene Rep.* **2021**, *22*, 100980. [CrossRef] [PubMed]

35. Li, Z.; Yang, L. Underlying Mechanisms and Candidate Drugs for COVID-19 Based on the Connectivity Map Database. *Front. Genet.* **2020**, *11*. [CrossRef] [PubMed]

36. Li, G.; He, X.; Zhang, L.; Ran, Q.; Wang, J.; Xiong, A.; Wu, D.; Chen, F.; Sun, J.; Chang, C. Assessing ACE2 Expression Patterns in Lung Tissues in the Pathogenesis of COVID-19. *J. Autoimmun.* **2020**, *112*, 102463. [CrossRef] [PubMed]

37. Prasad, K.; AlOmar, S.Y.; Alqahtani, S.A.M.; Malik, M.Z.; Kumar, V. Brain Disease Network Analysis to Elucidate the Neurological Manifestations of COVID-19. *Mol. Neurobiol.* **2021**, *58*, 1875–1893. [CrossRef] [PubMed]
38. Liu, F.; Li, Y.; Yang, Y.; Li, M.; Du, Y.; Zhang, Y.; Wang, J.; Shi, Y. Study on Mechanism of Matrine in Treatment of COVID-19combined with Liver Injury by Network Pharmacology and Molecular Docking Technology. Drug Deliv. 2021, 28, 325. [CrossRef] [PubMed]

39. Nain, Z.; Rana, H.K.; Lio, P.; Islam, S.M.S.; Summers, M.A.; Moni, M.A. Pathogenetic Profiling of COVID-19 and SARS-like Viruses. Brief. Bioinform. 2021, 22, 1175–1196. [CrossRef]

40. Fang, K.Y.; Cao, W.C.; Xie, T.A.; Ly, J.; Chen, J.X.; Cao, X.J.; Li, Z.W.; Deng, S.T.; Guo, X.G. Exploration and Validation of Related Hub Gene Expression during SARS-CoV-2 Infection of Human Bronchial Organoids. Hum. Genom. 2021, 15, 1–13. [CrossRef] [PubMed]

41. Rezaei-Tavirani, M.; Nejad, M.R.; Arjmand, B.; Tavirani, S.R.; Razaghi, M.; Mansouri, V. Fibrinogen Dysregulation Is a Prominent Process in Fatal Conditions of COVID-19 Infection; A Proteomic Analysis. Arch. Acad. Emerg. Med. 2021, 9, 1–5. [CrossRef]

42. Li, S.; Wang, W.; Li, T.; Han, X.; Hu, C.; Wang, Y.; Shen, M.; Du, L.; Nai, Y.; Wang, J.; et al. Immune Characteristics Analysis Reveals Two Key Inflammatory Factors Correlated to the Expressions of SARS-CoV-2 S1-Specific Antibodies. Genes Dis. 2022, 9, 522. [CrossRef] [PubMed]

43. Kumar, S. COVID-19: A Drug Repurposing and Biomarker Identification by Using Comprehensive Gene-Disease Associations through Protein-Protein Interaction Network Analysis. Preprints 2020. [CrossRef]

44. Zhu, Y.W.; Yan, X.F.; Ye, T.J.; Hu, J.; Wang, X.L.; Qiu, F.J.; Liu, C.H.; Hu, X.D. Analyzing the Potential Therapeutic Mechanism of Huashi Baidu Decoction on Severe COVID-19 through Integrating Network Pharmacological Methods. J. Tradit. Complement. Med. 2021, 11, 180–187. [CrossRef] [PubMed]

45. Bao, Z.; Wang, J.; He, K.; Lin, X.; Yu, T.; Li, J.; Gong, J. Xiang High Expression of Ace2 in the Human Lung Leads to the Release of Il6 by Suppressing Cellular Immunity: Il6 Plays a Key Role in COVID-19. Eur. Rev. Med. Pharmacol. Sci. 2021, 25, 527–540. [CrossRef] [PubMed]

46. Wang, Z.Z.; Li, K.; Maskey, A.A.; Huang, W.; Toutov, A.A.; Yang, N.; Srivastava, K.; Geliebter, J.; Tiwari, R.; Miao, M.; et al. A Small Molecule Compound Berberine as an Orally Active Therapeutic Candidate against COVID-19 and SARS: A Computational and Mechanistic Study. EASEB J. 2021, 35, 1–19. [CrossRef]

47. Auwul, M.R.; Rahman, M.R.; Gov, E.; Shahjaman, M.; Moni, M.A. Bioinformatics and Machine Learning Approach Identifies Potential Drug Targets and Pathways in COVID-19. Brief. Bioinform. 2021, 22, bbab120. [CrossRef] [PubMed]

48. Mosharaf, M.P.; Reza, M.S.; Kibria, M.K.; Ahmed, F.E.; Kabir, M.H.; Hasan, S.; Mollah, M.N.H. Computational Identification of Host Genomic Biomarkers Highlighting Their Functions, Pathways and Regulators That Influence SARS-CoV-2 Infections and Drug Repurposing. Sci. Rep. 2022, 12, 1–22. [CrossRef]

49. Lee, H.; Park, J.; Im, H.J.; Na, K.J.; Choi, H. Discovery of Potential Imaging and Therapeutic Targets for Severe Inflammation in COVID-19 Patients. Sci. Rep. 2021, 11, 1–9. [CrossRef]

50. Alanazi, K.M.; Farah, M.A.; Hor, Y.Y. Multi-Targeted Approaches and Drug Repurposing Reveal Possible SARS-CoV-2 Inhibitors. Vaccines 2022, 10, 24. [CrossRef]

51. Beck, B.R.; Shin, B.; Choi, Y.; Park, S.; Kang, K. Predicting Commercially Available Antiviral Drugs That May Act on the Novel Coronavirus (SARS-CoV-2) through a Drug-Target Interaction Deep Learning Model. Comput. Struct. Biotechnol. J. 2020, 18, 784–790. [CrossRef]

52. Xia, J.; Gill, E.E.; Hancock, R.E.W. NetworkAnalyst for Statistical, Visual and Network-Based Meta-Analysis of Gene Expression Data. Nat. Protoc. 2015, 10, 823–844. [CrossRef] [PubMed]

53. Boyle, E.L.; Weng, S.; Gollub, J.; Jin, H.; Botstein, D.; Cherry, J.M.; Sherlock, G. GO: TermFinder-Open Source Software for Accessing Gene Ontology Information and Finding Significantly Enriched Gene Ontology Terms Associated with a List of Genes. Bioinform. Appl. NOTE 2004, 20, 3710–3715. [CrossRef] [PubMed]

54. Szklarczyk, D.; Morris, J.H.; Cook, H.; Kuhn, M.; Wyder, S.; Simonovic, M.; Santos, A.; Doncheva, N.T.; Roth, A.; Bork, P.; et al. The STRING Database in 2017: Quality-Controlled Protein-Protein Association Networks, Made Broadly Accessible. Nucleic Acids Res. 2017, 45, D362–D368. [CrossRef] [PubMed]

55. Kanehisa, M.; Furumichi, M.; Tanabe, M.; Sato, Y.; Morishima, K. KEGG: New Perspectives on Genomes, Pathways, Diseases and Drugs. Nucleic Acids Res. 2017, 45, D353–D361. [CrossRef] [PubMed]

56. Kanehisa, M.; Goto, S. KEGG: Kyoto Encyclopedia of Genes and Genomes. Nucleic Acids Res. 2000, 28, 27–30. [CrossRef] [PubMed]

57. Benjamini, Y.; Hochberg, Y. Controlling the False Discovery Rate: A Practical and Powerful Approach to Multiple Testing. J. R. Stat. Soc. Ser. B 1995, 57, 289–300. [CrossRef]

58. Khan, A.; Fornes, O.; Stiglami, A.; Gheorghe, M.; Castro-Mondragon, J.A.; Van Der Lee, R.; Bessy, A.; Chêneby, J.; Kulkarni, S.R.; Tan, G.; et al. JASPAR 2018: Update of the Open-Access Database of Transcription Factor Binding Profiles and Its Web Framework. Nucleic Acids Res. 2018, 46, D250–D266. [CrossRef] [PubMed]

59. Karagkouni, D.; Paraskevopoulou, M.D.; Chatzopoulos, S.; Vlachos, I.S.; Tatsoglou, S.; Kanellos, I.; Papadimitriou, D.; Kavakiotis, I.; Maniou, S.; Skoufos, G.; et al. DIANA-TarBase v8: A Decade-Long Collection of Experimentally Supported MiRNA-Gene Interactions. Nucleic Acids Res. 2018, 46, D239–D245. [CrossRef] [PubMed]
113. Brun, M.; Coles, J.E.; Monckton, E.A.; Glubrecht, D.D.; Bisgrove, D.; Godbout, R. Nuclear Factor I Regulates Brain Fatty Acid-Binding Protein and Glial Fibrillary Acidic Protein Gene Expression in Malignant Glioma Cell Lines. *J. Mol. Biol.* **2009**, *391*, 282–300. [CrossRef] [PubMed]

114. Rahman, M.R.; Islam, T.; Goy, E.; Turanli, B.; Gulfidan, G.; Shahjaman, M.; Banu, N.A.; Mollah, M.N.H.; Arga, K.Y.; Moni, M.A. Identification of Prognostic Biomarker Signatures and Candidate Drugs in Colorectal Cancer: Insights from Systems Biology Analysis. *Medicina* **2019**, *55*, 20. [CrossRef] [PubMed]

115. Rahman, M.B.; Islam, T.; Bari, M.R.; et al. Discovery and Development of Simeprevir (TMC435), a HCV NS3/4A Protease Inhibitor. *De Kock, H.; Nilsson, M.; et al. Discovery and Development of Simeprevir (TMC435), a HCV NS3/4A Protease Inhibitor.* *J. Mol. Biol.* 2009, *391*, 282–300. [CrossRef] [PubMed]

116. Majd, M.; Hosseini, A.; Ghaedi, K.; et al. MiRNA-106b-5p in Human Cancers: Diverse Functions and Promising Biomarker. *Expert Rev. Ant. Infect. Ther.* **2020**, *18*, 1079–1094. [CrossRef] [PubMed]

117. Fang, T.; Wu, Q.; Zhou, L.; et al. MiR-106b-5p and MiR-17-5p Suppress Osteogenic Differentiation by Targeting Smad5 and Inhibit Bone Formation. *Exp. Cell Res.* **2016**, *347*, 74–82. [CrossRef] [PubMed]

118. Saimi, T.; Kim, H.; Song, Y.; Cho, E.; Lee, A. MiR-106b-5p and MiR-17-5p Could Predict Recurrence and Progression in Breast Ductal Carcinoma in Situ Based on the Transforming Growth Factor-Beta Pathway. *Breast Cancer Res. Treat.* **2019**, *176*, 119–130. [CrossRef] [PubMed]

119. Heidary, F.; Gharebaghi, R. Ivermectin: A Systematic Review from Antiviral Effects to COVID-19 Complementary Regimen. *Int. J. Antimicrob. Agents* **2020**, *55*, 218–226. [CrossRef] [PubMed]

120. Yang, C.; Dou, R.; Yin, T.; Ding, J. MiRNA-106b-5p Functions as a Tumor Suppressor by Targeting VEGFA in Renal Cell Carcinoma. *Dis. Markers* **2020**, *2020*, 8837941. [CrossRef] [PubMed]

121. Majd, M.; Hosseini, A.; Ghaedi, K.; et al. MiR-106a-5p Functions as a Tumor Suppressor by Targeting VEGFA in Renal Cell Carcinoma. *Dis. Markers* **2020**, *2020*, 8837941. [CrossRef] [PubMed]

122. Peltzer, B.; Lerman, B.B.; Goyal, P.; Cheung, J.W. Role for Digoxin in Patients Hospitalized with COVID-19 and Atrial Arrhythmias. *J. Cardiovasc. Electrophysiol.* **2020**, *11*, 1127–1131. [CrossRef] [PubMed]

123. Siniorakis, E.; Arvanitakis, S.; Katsianis, A.; Elkouris, M. Atrial Fibrillation and Flutter in Patients Hospitalized for COVID-19: The Challenging Role of Digoxin. *J. Cardiovasc. Electrophysiol.* **2021**, *32*, 876. [CrossRef] [PubMed]

124. Mezaal, M.H.; Farhan, H.A.; Dakhi, Z.A. COVID-19 Pandemic Impact on Physicians' Decision-Making: Digoxin Toxicity in View of Combination of Hydroxychloroquine and Azithromycin: A Case Report. *Open Access Maced. J. Med. Sci.* **2020**, *8*, 150–153. [CrossRef] [PubMed]

125. Pandey, S.; Pathak, S.K.; Pandey, A.; Salunke, A.A.; Chawla, J.; Ortho, M.S.; Sharma, A.; Sharma, S.; Thivari, P.; Ratna, H.V. Ivermectin in COVID-19: What Do We Know? *Diabetes Metab. Syndr. Clin. Res. Rev.* **2020**, *14*, 2021. [CrossRef] [PubMed]

126. Heidary, F.; Ghearebaghi, R. Ivermectin: A Systematic Review from Antiviral Effects to COVID-19 Complementary Regimen. *J. Antimicrob. Agents* **2020**, *73*, 593–602. [CrossRef] [PubMed]

127. Gupta, D.; Sahoo, A.K.; Singh, A. Ivermectin: Potential Candidate for the Treatment of COVID-19. *Braz. J. Infect. Dis.* **2020**, *24*, 369–371. [CrossRef] [PubMed]

128. Siniorakis, E.; Arvanitakis, S.; Katsianis, A.; Elkouris, M. Atrial Fibrillation and Flutter in Patients Hospitalized for COVID-19: The Challenging Role of Digoxin. *J. Cardiovasc. Electrophysiol.* **2021**, *32*, 876. [CrossRef] [PubMed]

129. Fang, T.; Wu, Q.; Zhou, L.; et al. MiRNA-106b-5p in Human Cancers: Diverse Functions and Promising Biomarker. *Biomed. Pharmacother.* **2020**, *127*, 110211. [CrossRef] [PubMed]

130. Liao, W.; He, J.; Disoma, C.; Hu, Y.; Li, J.; Chen, G.; Sheng, Y.; Cai, X.; Li, C.; Cheng, K.; et al. Hsa_circ_0107593 Suppresses the Progression of Cervical Cancer via Sponging Hsa-MiR-20a-5p/93-5p/106b-5p. *Front. Oncol.* **2021**, *10*, 590627. [CrossRef] [PubMed]

131. Gurung, A.B.; Ali, M.A.; Lee, J.; Farah, M.A.; Al-Anazi, K.M. The Potential of Paritaprevir and Emetine as Inhibitors of SARS-CoV-2 RdRp. *Saudi J. Biol. Sci.* **2020**, *28*, 28–34. [CrossRef] [PubMed]

132. Gurung, A.B. In Silico Structure Modelling of SARS-CoV-2 Nsp13 Helicase and Nsp14 and Repurposing of FDA Approved Antiviral Drugs as Dual Inhibitors. *Gene Rep.* **2020**, *21*, 100860. [CrossRef] [PubMed]