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Developing Potential Drugs for COVID-19 Using Ligand Based Virtual Screening

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Abstract *Background and purpose:* Coronavirus disease 2019 (COVID-19) caused by Severe Acute Respiratory Syndrome- Coronavirus 2 (SARS-CoV2) is a highly contagious disease that has infected more than 2.4 million patients and led to more than 160, 000 deaths in less than five months. Chloroquine is very effective in management of COVID-19. Compounds similar to chloroquine may have the same biological activity and thus inhibit SARS-CoV2. *Methods:* SwissSimilarity tool was used to identify similar compounds to chloroquine in the ZINC database. Compounds which were more similar than hydroxychloroquine were selected and used to test molecular docking with quinone reductase 2 (a target for chloroquine). Pharmacokinetic and toxicity profiles of selected compounds were assessed using SwissADME and Protox Server respectively. *Results:* There were 49 drug-like compounds in the ZINC database having a higher similarity index to chloroquine compared to hydroxychloroquine. 17 of these had a better binding potential to quinone reductase 2 compared to chloroquine while two had similar binding potential to chloroquine and three had similar binding potential to hydroxychloroquine. Out of these 22 compounds, 18 had a higher predicted LD50 compared to chloroquine but lower when compared to hydroxychloroquine. *Conclusion:* Eighteen drug-like compounds in the ZINC database bind with high affinity to quinone reductase 2, are less toxic but similar to chloroquine. Therefore, they may have activity against SARS-CoV2. However, in vivo or in vitro study should be done since this is an in silico study.

Keywords: *COVID-19, SARS-CoV2, chloroquine, quinone reductase, ZINC database, ligand-based virtual screening*

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1. Introduction

Coronavirus disease 2019 (COVID -19), a pandemic declared by the World Health Organisation (WHO) on 11th March 2020, is highly infectious [1]. It is caused by Severe Acute Respiratory Syndrome- Coronavirus 2 (SARS-CoV2) and was initially reported in Wuhan, China in December 2019 [2,3,4]. According to an interactive web-tool created by Centre for Systems Science and Engineering at John Hopkins University, the confirmed cases of COVID-19 by 20th April 2020 were 2, 420, 439. The total deaths stood at 166, 205 [5]. Within less than five months, COVID-19 had already infected patients in 185 countries and/or regions [5].

Chloroquine has been shown to have activity against SARS-CoV2 and thus used for treatment of COVID-19. A number of clinical trials conducted in China verified the activity of chloroquine in management of COVID-19 [6,7]. A systematic review conducted by Cortegiani et al. involving 23 clinical trials, two national guidelines, an in vitro study, a narrative, an editorial and an expert consensus paper concluded that there was sufficient data for the use of chloroquine in management of COVID-19 [8]. Hydroxychloroquine, a similar drug to chloroquine, has also been used for the management of COVID-19 [9]. However, chloroquine has several side effects including but not limited to ocular toxicity which can lead to retinopathy, pruritus, nausea, vomiting and cardio-depressant effects [10,11].

Chloroquine is thought to act by several mechanisms. It can increase the pH of the lysosomes and thus affect the activity of viral enzymes [11,12]. It is also thought to inhibit quinone reductase 2 which is critical for biosynthesis of sialic acid, an important component for glycosylation of angiotensin converting enzyme 2 (ACE2) [11,13,14]. ACE2 facilitates SARS-CoV2 entry into target human cells and thus leads to development of COVID-19 [15].

Virtual screening involves screening large computer databases to identify potentially active compounds [16,17]. Ligand-based virtual screening involves screening large computer databases to identify compounds that are similar to a known ligand. Compounds similar to a known ligand are thought to have similar biological activity [18]. The ZINC database contains more than 120 million compounds which are drug-like, are available and can be purchased [19,20]. Therefore, screening compounds similar to chloroquine may yield other compounds which may be active against SARS-CoV2 and thus provide alternatives in the management of COVID-19.

2. Methods

In-silico drug analysis was done. The canonical smiles of chloroquine were obtained from PubChem (https://pubchem.ncbi.nlm.nih.gov/) and inserted into the SwissSimimilarity online tool to identify compounds similar to chloroquine. This online tool quantifies molecular similarity by describing molecular structures and properties [18]. The combined technique which involves molecular fingerprints, pharmacophore recognition and shape-based similarity was used to screen the ZINC database of drug-like compounds [19]. A similarity score was generated based on a combination of Tanimoto coefficient and Electro-shape 5D manhattan distance. The equation for the similarity scores was described by Zoete et al. (2016).

The compounds that had a higher similarity index compared to hydroxychloroquine (> 0.799) were selected and drawn on pubchem sketcher tool and the molfile downloaded and converted to their respective 3-D structures by Avogadro software [21]. By using the Avogadro software, the 3-D structures were optimised to the most stable conformation by using MMFF94s as the force field. With the help of Chimera software, hydrogen atoms and charge were added to the stable conformations of the selected compounds [22].

The Protein Databank (PDB) (https://www.rcsb.org/) was used to access the quinone reductase 2 target (PDB ID: 4FGK). It was downloaded and the non-standard residues removed by the Chimera software [22]. Surface - binding analysis was carried out between the selected compounds and the quinone reductase 2 enzyme using AutoDock vina feature in the Chimera software. This was also done for chloroquine to serve as a positive control. Ligand interactions with quinone reductase 2 were examined using Discovery Studio software.

The pharmacokinetic profiles of the selected compounds were predicted using SWISSADME online tool (http://www.swissadme.ch/) [23]. This online tool predicts and evaluates drug-likeness, pharmacokinetic properties and medicinal chemistry likeness. The canonical SMILES of chloroquine, hydroxychloroquine and selected compounds were inserted on the SWISSADME website and the potential absorption, distribution, metabolism and excretion properties predicted.

In addition, toxicity profiles of chloroquine, hydroxychloroquine and selected compounds were predicted using the ProTox server: http://tox.charite.de/protox_II/ [24]. ProTox server assists in prediction of oral toxicity, hepatotoxicity, immunotoxicity, carcinogenicity, cytotoxicity, mutagenicity, 15 toxicity targets based on Novartis in vitro safety panels and toxicological pathways (nuclear receptor signaling pathways – 7 models and stress response pathways – 5 models).

3. Results and Discussion

The similarity scores were based on the combination of Tanimoto coefficient which is important in assessing the chemical structure similarity and Electro-shape 5D manhattan distance which estimates 3-D similarity in a nonsuperpositional shape-based approach [18]. Hydroxychloroquine had a similarity score of 0.799 relative to chloroquine. There were 49 drug-like compounds in the ZINC database that had a higher similarity score compared to hydroxychloroquine as shown in Table 1. The similarity scores were above 0.799. All the selected compounds complied with rules of druglikeness proposed by Lipinski which stated that a compound with 10 or less hydrogen acceptors, molecular weight of less than or equal to 500, 5 or less hydrogen bonds and a logP of less than or equal to 5 would result in a compound being orally active [25]. These compounds also complied to Veber's rules on druglikeness which proposed that compounds with 10 or less rotatable bonds and polar surface area of less than or equal to 140 angstroms would make a compound orally active [26]. Therefore, it is plausible that all these compounds can be administered orally.

Table 1. Properties of selected compounds relative to Chloroquine.

COMPOUND	BINDING ENERGIES TO QUINONE REDUCTASE	SIMILARITY RELATIVE TO CHLOROQUINE	PREDICTED LD50 (MG/KG)
CHLOROQUINE	-8.6		311
HYDROXYCHLOROQUINE	-8.5	0.799	1240
ZINC38050614	-9.6	0.956	750
ZINC82133908	-9.6	0.940	750
ZINC95362918	-9.6	0.937	750
ZINC82133910	-9.5	0.947	750
ZINC38050615	-9.4	0.950	750
ZINC96331701	-9.3	0.988	750
ZINC78617776	-9.3	0.974	750
ZINC78758909	-9.2	0.800	770
ZINC01596768	-9.1	0.975	750
ZINC08579986	-9.1	0.869	311
ZINC01706629	-9.1	0.843	750
ZINC01683221	-9.0	0.957	234
ZINC04409428	-8.9	0.833	750
ZINC44514898	-8.8	0.904	695
ZINC33956413	-8.8	0.878	750
ZINC04335994	-8.8	0.833	770
ZINC20552561	-8.7	0.919	117
ZINC41719758	-8.6	0.828	695
ZINC83070730	-8.6	0.802	555
ZINC73738703	-8.5	0.946	750
ZINC37985880	-8.5	0.906	117
ZINC73738671	-8.5	0.899	750

The binding potential of chloroquine to quinone reductase 2 was -8.6 while that of hydroxychloroquine was -8.5. The compounds with a similar binding potential to hydroxychloroquine and chloroquine and those with a greater binding potential are also presented in Table 1. There were 17 compounds that had a better binding potential compared to chloroquine while two compounds had a similar binding potential to chloroquine and three had a similar binding potential to hydroxychloroquine. This indicated that 22 compounds in the ZINC database were similar to chloroquine and had the added benefit of having a high affinity to quinone reductase 2, an important enzyme for biosynthesis of sialic acid. Therefore, the 22 compounds can inhibit glycosylation of ACE2 and thus prevent entry of SARS-CoV2 to target cells. Out of these 22 compounds, 18 had a higher predicted LD50 compared to chloroquine but lower when compared to hydroxychloroquine. Therefore, these 18 compounds are less toxic compared to chloroquine and have a high affinity for quinone reductase 2 and can be used as alternatives to chloroquine for the management of COVID-19.

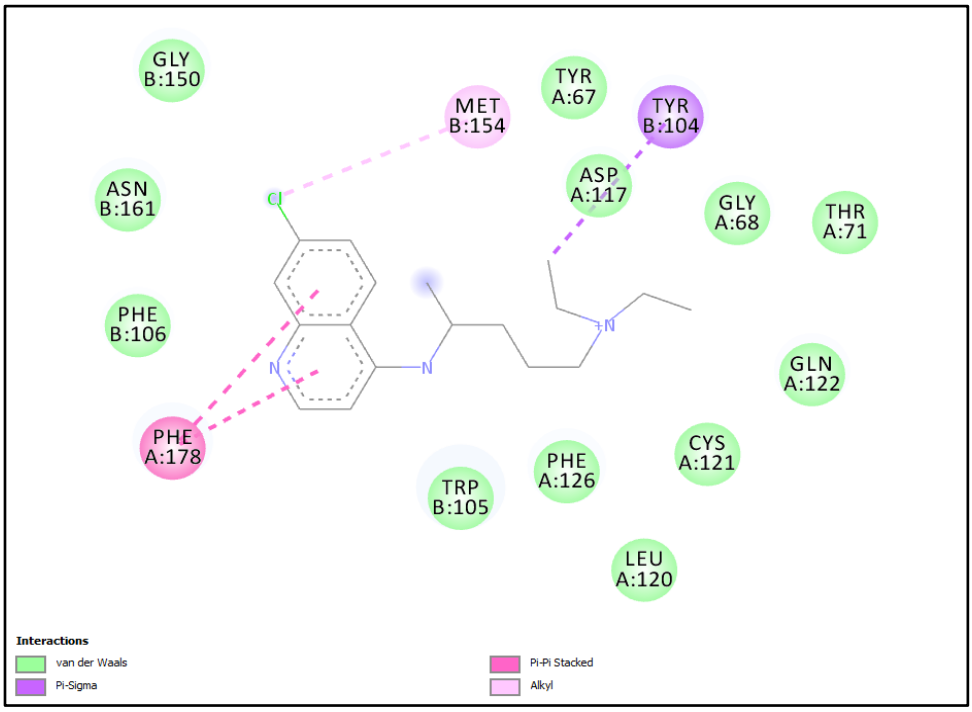

Figure 1. Interactions between chloroquine and quinone reductase 2

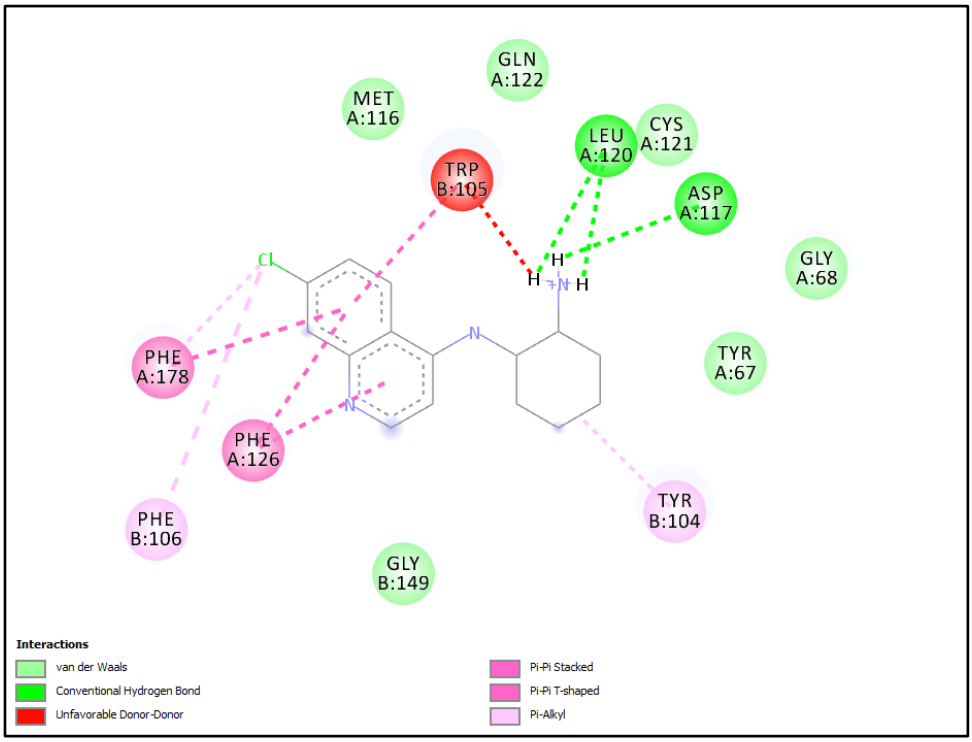

Figure 2. Interactions between ZINC38050614 and quinone reductase 2

The major interactions between chloroquine and quinone reductase 2 involved pi-pi stacked interactions with phenylalanine in chain A at position 178, pi-sigma interactions with tyrosine in chain B at position 104 and alkyl interactions with methionine in chain B at position 154 as shown in Figure 1. ZINC38050614 with the highest binding energy of -9.6 also interacted with phenylalanine in chain A at position 178 and tyrosine in chain B at position 104. However, it also had interactions involving conventional hydrogen bonds with aspartate at position 117 and leucine at position 120 in chain A, alkyl interactions with phenylalanine at position 106 in chain B and tryptophan at position 105 in chain B as shown in Figure 2. This indicates that they both act at the active site of quinone reductase 2 enzyme which was shown to involve the phenyalanine at position 178 [27]. However, ZINC38050614 and the compounds with a binding energy above -9.1 have more interactions with quinone reductase 2 and may have a higher affinity for the enzyme since they interact with tryptophan at position 105, phenylalanine at position 106 and 126 which make up the interior wall of the active site [27]. The compounds with a higher binding energy compared to chloroquine but less than -9.1 interacted with at least one of the amino acids forming the interior wall of the active site.

When predicted for immunotoxicity and mutagenicity, both chloroquine and hydroxychloroquine were active. Hydroxychloroquine also potentially bound to the toxicity target amine oxidase A. ZINC01706629, ZINC01683221, ZINC20552561, ZINC01596768 and ZINC37985880 were also active for both immunotoxicity and mutagenicity. In addition, ZINC01596768 and ZINC37985880 were active for cytotoxicity. ZINC38050614 and ZINC38050615 were active for mutagenicity but less active for immunotoxicity than chloroquine. They also affected the aryl hydrocarbon receptor. ZINC95362918, ZINC96331701, ZINC08579986, ZINC33956413 and ZINC73738671 were active for immunotoxicity but less active for mutagenicity. ZINC78617776, ZINC78758909 and ZINC04409428 were active for immunotoxicity but inactive for mutagenicity. ZINC82133908 and ZINC82133910 were less active for both immunotoxicity and mutagenicity than chloroquine. ZINC44514898, ZINC04335994, ZINC41719758, ZINC83070730 and ZINC73738703 were inactive for immunotoxicity but potentially less active for mutagenicity compared to chloroquine.

All the compounds were drug-like and had high gastrointestinal absorption. With the exception of ZINC41719758; chloroquine, hydroxychloroquine and the other 21 highly active compounds inhibit cytochrome p450 (CYP) isoform 1A2 and 2D6 may thus affect the metabolism of tricyclic antidepressants, selective serotonin reuptake inhibitors, codeine, propranolol, timolol, tizanidine, triamterene, quinidine, caffeine, ropivacaine, melatonin, clozapine, risperidone, olanzapine, lidocaine, tacrine, zolmitriptan, frovatriptan and many other drugs [28,29]. In addition, chloroquine, ZINC96331701, ZINC01706629 and ZINC08579986 also inhibit CYP3A4 and thus affect metabolism of many drugs including but not limited to clotrimazole, fluconazole, itraconazole, ketoconazole, budesonide, diazepam, disopyramide, nifedipine, erythromycin, salbutamol, terfenadine, clozapine, propofol, quinidine, procainamide, haloperidol and ibuprofen [30].

ZINC04335994, ZINC01683221, ZINC20552561, ZINC01596768, ZINC33956413, ZINC38050614, ZINC01706629 and ZINC38050615 also inhibit CYP2C19 and may affect the metabolism of barbiturates like mephobarbital & hexobarbital, proton pump inhibitors like omeprazole, pantoprazole & lansoprazole, moclobemide, diazepam, mephenytoin, and carisoprodol [31]. ZINC01706629 also inhibits CYP2C9 and thus affects metabolism of angiotensin receptor blockers like losartan & candesartan, some non-steroidal anti-inflammatory drugs, warfarin, zafirlukast, phenytoin, cyclophosphamide, tolbutamide, and other drugs [32]. Therefore, a drug with minimal drug-drug interactions like ZINC41719758 would be very beneficial for patients who have severe disease of COVID-19 and comorbidities. In addition, patients with comorbidities tended to have severe disease of COVID-19 [33,34].

P-glycoprotein substrates were ZINC73738703, ZINC83070730, ZINC41719758, ZINC04335994, ZINC44514898, ZINC04409428, ZINC95362918, ZINC73738671, ZINC82133908, ZINC82133910, ZINC38050614 and ZINC38050615. Chloroquine, hydroxychloroquine and the other 10 active compounds were not substrates of p-glycoprotein

4. Conclusion

There are 18 drug-like compounds in the ZINC database which bind with high affinity to quinone reductase 2, are less toxic but similar to chloroquine. They may thus have activity against SARS-CoV2. ZINC41719758 has minimal drug-drug interactions and can potentially be beneficial for patients with comorbidities.

5. Recommendations

In vitro and in vivo studies should be carried out on the 18 drug-like compounds identified from the ZINC database to assess antiviral activity against SARS-CoV2.

References

[1] Hageman JR. The Coronavirus Disease 2019 (COVID-19). Pediatr Ann 2020; 49: e99-100.

[2] Wu F, Zhao S, Yu B, Chen Y-M, Wang W, Song Z-G, et al. A new coronavirus associated with human respiratory disease in China. Nature 2020.

[3] Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al. A Novel Coronavirus from Patients with Pneumonia in China, 2019. N Engl J Med 2020:727-33.

[4] Wang C, Horby PW, Hayden FG, Gao GF. A novel coronavirus outbreak of global health concern. Lancet 2020; 395: 470-3.

[5] Dong E, Du H, Gardner L. An interactive web-based dashboard to track COVID-19 in real time. Lancet Infect Dis 2020; 3099: 19-20.

[6] Gao J, Tian Z, Yang X. Breakthrough: Chloroquine phosphate has shown apparent efficacy in treatment of COVID-19 associated pneumonia in clinical studies. Biosci Trends 2020; 14: 72-3.

[7] Touret F, de Lamballerie X. Of chloroquine and COVID-19. Antiviral Res 2020; 177: 104762.

[8] Cortegiani A, Ingoglia G, Ippolito M, Giarratano A, Einav S. A systematic review on the efficacy and safety of chloroquine for the treatment of COVID-19. J Crit Care 2020: 3-7.

[9] Gautret P, Lagier J-C, Parola P, Hoang VT, Meddeb L, Mailhe M, et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. Int J Antimicrob Agents 2020: 105949.

[10] Luzzi GA, Peto TEA. Adverse Effects of Antimalarials: An Update. Drug Saf 1993; 8: 295-311.

[11] Thomé R, Lopes SCP, Costa FTM, Verinaud L. Chloroquine: Modes of action of an undervalued drug. Immunol Lett 2013; 153: 50-7.

[12] Vincent MJ, Bergeron E, Benjannet S, Erickson BR, Rollin PE, Ksiazek TG, et al. Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. Virol J 2005; 2: 1-10.

[13] Devaux CA, Rolain J-M, Colson P, Raoult D. New insights on the antiviral effects of chloroquine against coronavirus: what to expect for COVID-19? Int J Antimicrob Agents 2020: 105938.

[14] Savarino A, Di Trani L, Donatelli I, Cauda R, Cassone A. New insights into the antiviral effects of chloroquine. Lancet Infect Dis 2006; 6: 67-9.

[15] Zhang H, Penninger JM, Li Y, Zhong N, Slutsky AS. Angiotensin-converting enzyme 2 (ACE2) as a SARS-CoV-2 receptor: molecular mechanisms and potential therapeutic target. Intensive Care Med 2020; 46: 586-90.

[16] Shoichet BK. Virtual screening of chemical libraries. Nature 2004; 432: 862-5.

[17] Patrick Walters W, Stahl MT, Murcko MA. Virtual screening - An overview. Drug Discov Today 1998; 3: 160-78.

[18] Zoete V, Daina A, Bovigny C, Michielin O. Swiss Similarity: A Web Tool for Low to Ultra High Throughput Ligand-Based Virtual Screening. J Chem Inf Model 2016; 56: 1399-404.

[19] Irwin JJ, Shoichet BK. ZINC - A free database of commercially available compounds for virtual screening. J Chem Inf Model 2005; 45: 177-82.

[20] Sterling T, Irwin JJ. ZINC 15 - Ligand Discovery for Everyone. J Chem Inf Model 2015; 55: 2324-37.

[21] Hanwell MD, Curtis DE, Lonie DC, Vandermeerschd T, Zurek E, Hutchison GR. Avogadro: An advanced semantic chemical editor, visualization, and analysis platform. J Cheminform 2012; 4.

[22] Yang Z, Lasker K, Schneidman-Duhovny D, Webb B, Huang CC, Pettersen EF, et al. UCSF Chimera, MODELLER, and IMP: An integrated modeling system. J Struct Biol 2012; 179: 269-78.

[23] Daina A, Michielin O, Zoete V. SwissADME: A free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Sci Rep 2017; 7: 1-13.

[24] Banerjee P, Eckert AO, Schrey AK, Preissner R. ProTox-II: A webserver for the prediction of toxicity of chemicals. Nucleic Acids Res 2018; 46: W257-63.

[25] Lipinski CA. Lead- and drug-like compounds: The rule-of-five revolution. Drug Discov Today Technol 2004; 1: 337-41.

[26] Veber DF, Johnson SR, Cheng HY, Smith BR, Ward KW, Kopple KD. Molecular properties that influence the oral bioavailability of drug candidates. J Med Chem 2002; 45: 2615-23.

[27] Leung KKK, Shilton BH. Chloroquine binding reveals flavin redox switch function of quinone reductase 2. J Biol Chem 2013; 288: 11242-51.

[28] Faber MS, Jetter A, Fuhr U. Assessment of CYP1A2 activity in clinical practice: Why, how, and when? Basic Clin Pharmacol Toxicol 2005; 97: 125-34.

[29] Ingelman-Sundberg M. Genetic polymorphisms of cytochrome P450 2D6 (CYP2D6): Clinical consequences, evolutionary aspects and functional diversity. Pharmacogenomics J 2005; 5: 6-13.

[30] Kenworthy KE, Bloomer JC, Clarke SE, Houston JB. CYP3A4 drug interactions: Correlation of 10 in vitro probe substrates. Br J Clin Pharmacol 1999; 48: 716-27.

[31] Wedlund PJ. The CYP2C19 enzyme polymorphism. Pharmacology 2000; 61: 174-83.

[32] Rettie AE, Jones JP. CLINICAL AND TOXICOLOGICAL RELEVANCE OF CYP2C9: Drug-Drug Interactions and Pharmacogenetics. Annu Rev Pharmacol Toxicol 2005; 45: 477-94.

[33] Yang J, Zheng Y, Gou X, Pu K, Chen Z, Guo Q, et al. Prevalence of comorbidities in the novel Wuhan coronavirus (COVID-19) infection: a systematic review and meta-analysis. Int J Infect Dis 2020.

[34] Wang T, Du Z, Zhu F, Cao Z, An Y, Gao Y, et al. Comorbidities and multi-organ injuries in the treatment of COVID-19. Lancet 2020; 395: e52.