Synthesis of new 3-(hydroxymethyl)-2-phenyl-2,3 dihydroquinolinolone and in-silico evaluation of COVID-19 main protease inhibitor

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Abstract
An exclusive approach towards the synthesis of novel 3-(hydroxymethyl)-2-phenyl-2,3 dihydroquinolinolone (1H)-one and its in-silico evaluation as inhibitor of COVID-19 main protease. The one-pot synthesis of an established procedure Claisen ester condensation reaction was sodium hydride mediated with intramolecular cyclization with solvent free conditions. The structures of the synthesized compound were confirmed by IR, 1H,13C NMR, and EI-MS spectral studies. Chemo-informatics study showed that the compound obeyed the Lipinski’s rule, PASS, Swiss ADME. Computational docking analysis was performed using PyRx, AutoDock Vina option based on scoring functions. In-silico molecular docking study results demonstrated Greater binding energy and affinity to the active pocket the N3 binding site of the Coronavirus primary protease.

Keywords. Methylantharanate, 2-Phenylquinoline, Claisen condensation, molecular docking, RNA polymerase, protease Covid-19.

1. INTRODUCTION
Coronavirus disease stated December 2019 in Wuhan, China.[1] The epidemic was isolated to a new type strain of coronavirus, the name of which world health organization (WHO) 2019-nCoV or COVID-19, later renamed the international committee on virus taxonomy to SARS-CoV-2, which was confirmed as the pathogenic source of virus severe acute respiratory syndrome.[2] The virus disease is extremely spreadable in air, and breathing problem of some cases affected with COVID-19, were rapidly fatal is public health emergency of global fear. The pandemic disease has brought about travel restrictions and countrywide lockdowns in many countries, as of global death-to-case ratio is 2.2% (1,792,251/82,051,958) as of 30th December 2020.[3]

Coronavirus are RNA source into four types; α-coronavirus and β-coronavirus,[4,5] Covid-19 diseases were connected to be beta type of coronavirus that causes severe acute respiratory syndrome (SARS)[6] similar to SARS-CoV-1. COVID-19 virus enters human lungs cells through the angiotensin converting-enzyme (ACE2) receptor. Coronavirus contain a positive-sense with single-stranded RNA genome[7] The genome size of coronaviruses ranges from 26.4 to 31.7 kilobases genome size is one of significant between RNA viruses. The genome has 5-methylated cover and 3-polyadenylated tail. A genome group of four encodes for a coronavirus is 5’-leader-UTR-replicas (ORF1ab) -spike glycoprotein (S), little envelope protein (E)-membrane, matrix glycoprotein (M), nucleocapsid protein (N), 3’UTR-poly (A) tail.[8]

3CL-pro main protease presented in four matured structural genes is vital for coronavirus life cycle by making it a target of anti-corona viral drug by sequence alignment 3CLpro-2. The crystal structure of COVID-19 (PDB ID: 6LU7)[9] contains 9 alpha-helices and 13 beta-strands that make up three distinct domains. 3CLpro-2 Nucleocapsid protein (NP), an essential RNA-binding viral protein existed in a human coronavirus (COVID-19) infected cells, is required for the replication, and transcription of viral
RNA. Recent studies suggested that human COVID-19 is a valid target for antiviral drug development based on this aspect, a structure-based virtual screening approach to target the nucleocapsid protein (NP) are made to show good chemical starting points for medicinal chemistry.

Quinoline compounds are greatest known for their various biological activities, however they have also successfully treated many diseases such as anticancer and antimalarial. While many quinoline alkaloid and derivatives used for anti-mycobacterial drugs, anticonvulsant, anti-inflammatory, cardiovascular activities quinoline constructed by the products assessed their potential as β-glucuronidase and α-glucosidase inhibitors therefore chloroquinoline even displays anti-leishmanial properties. In recently, used chloroquine and hydroxychloroquine drugs are people’s medical treatment with COVID-19. Utmost prescribed medication in the American country with over five million prescriptions. The hypothetical use of hydroxychloroquine for COVID-19 portends its availability for people with established suggestions. Quinoline alkaloids type of compounds medication by are used to treatment of malaria fever infection is effective in decreasing viral replication in other infections, including the SARS associated coronavirus (CoV) and MERS-CoV (WHO) model list of fundamental prescription medicines. Synthesized quinoline hetero-cyclic compounds are synthesis has become a powerful for making new molecules for drug discovery and development and other hands, quinoline derivatives owe their popularity since quinoline ring is quickly available synthetic approaches have been reported. Although modern methods of Conrad-Limpach, Niementowski, Doebner-Miller, Vilsmieer-Haak reactions Claisen condensation reaction of amines with carboxyl derivative followed by cyclization to produce the desired quinolines molecule. The classical multi-step reaction operations, not only results in an increase in efficiency and but also reduces the generating large quantities of waste. Organic synthesis by one-pot, tandem, Claisen condensation reactions become an interesting field of research in organic chemistry, consequently accelerating drug discovery programs. The Claisen condensation reaction is one among the classical and valuable multi-component reactions for the assembly of pharmacologically active groups of quinoline and quinoline derivatives from commercially accessible precursors.

In the present study we have design and synthesis of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one drug discovery processes the capability of a unique compound. 2-phenylquinoline is a valuable precursor for the synthesis of many biologically active compounds. Nevertheless, there was just one reported investigation using this precursor for preparing of quinoline derivatives. In this research paper, has sodium hydride as catalyst, solvent-free reaction method, given the apparent biological importance of these compounds and in continuation of our interest within the synthesis of biologically active heterocyclic compounds, the synthesis of 3-(Hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one was performed via three-component one-pot condensation of ethyl benzoate, ethyl acetoacetate and methyl 2-aminobenzoate within the occurrence of sodium hydride as catalyst (scheme 1). The synthesized compounds has been evaluated for their potential anti-COVID-19 activity and explored through in-silico molecular docking studies.

2. MATERIALS AND METHODS
2.1. Chemistry

2.1.1. General

All chemicals and solvents were purchased from Fine and Merk Chemicals, India. Melting points are uncorrected. Perkin-Elmer Paragon 1000 FT-IR spectrometers as potassium bromide discs unless otherwise indicated scanning 32 times from 4000 to 400 cm⁻¹ at 4 cm⁻¹ resolutions. ¹H and ¹³C NMR spectra were obtained in the CDCl₃ solvent on a Bruker (500 MHz) instrument. Perkin Elmer, Mass Spectrometer Clarus 600 (EI), Clarus 680 GC was used in the analysis. All compounds were routinely checked by thin layer chromatography (TLC) with Merck silica gel 60F254 glass plates. In column chromatography, Merck silica gel 60-120 mesh, Petroleum ether and ethyl acetate, as eluents, were used. Solvents and reagents were purified by literature methods. Petroleum ether refers to the hydrocarbon fraction of boiling range 60-80 °C. Compounds were detected by short and long ultraviolet light and with iodine vapor.

2.1.2. General Procedure for the synthesis of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4

An equimolar mixture of ethyl benzoate 1 (3.06 mL, 0.2 mol), ethyl 3-oxobutanoate (Ethylacetooacetate) 2 (2.60 mL, 0.2 mol) and methyl 2-aminobenzoate 3 (3.02 mL, 0.1 mol) and a catalytic amount of NaH was added. The reaction mixture was the reflux at 140 °C for 6 hours, A yellow solid was obtained and monitoring the progress of the reaction by TLC. It
was cooled to room temperature then transferred into 500 mL ice water, neutralized with 10 % HCl. The precipitate was filtered off and purified by column chromatography followed by recrystallization from ethanol to yield yellow crystalline solid, 75 % yield; FT-IR (KBr, \( \nu_{\text{max}} \), cm\(^{-1} \)): 3263, 2949, 2843, 1693, 1668, 1589, 1533, 1431; \( ^1 \)H NMR (400 MHz, CDCl\(_3 \)): 611.93 (br s, 1N-H), 8.84 (dd, \( J = 8.4,1.1 \)), 7.96-7.94 (m, 3H), 7.74-7.72 (m, 3H), 7.50-7.38 (m, 1H), 7.01-6.97 (m, 1H), 6.57-6.51 (m, 1H), 5.75 (s, 1H), 3.86-3.83 (m, 2H), 3.74 (s, 1H). \( ^{13} \)C NMR (100 MHz, CDCl\(_3 \)) \( \delta = 169.05, 165.67, 150.51, 141.89, 134.89, 134.80, 134.06, 131.95, 130.96, 128.82, 122.60, 120.44, 116.68, 116.21, 115.15, 52.47, 51.47. HRMS (ESI, m/z), C\(_{16}\)H\(_{15}\)NO\(_2 \) [M+H] = 255.1968

2.2. Pharmacological/biological assays

2.2.1. Prediction of activity spectra for substances (PASS)

We have used PASS (Prediction of Activity Spectra for Synthesis of compound 4, available host at http://www.way2drug.com/PASSOnline/predict.php and [https://proteins.plus/])\(^{[3]}\) for computational screening of conceivable properties which are biological activities like bi-corona virus along with other relevant activities i.e., nicotinic alpha-6-beta-3-beta-4-alpha-5-receptor, gluconate-2-dehydrogenase (acceptor) inhibitor, Ubiquinol-cytochrome-c reductase inhibitor, anti-hypoxic, phosphatase inhibitor activities, etc. These servers program designed a tool for evaluating the typical biological potential of an organic drug-like aspiran.\(^{[3]}\) PASS predictions of a range that is wide of Activity based on the structure of organic compounds Thus, PASS may be used to evaluate the biological activity outline for virtual molecules, This tool of methods provided quantitative structure–activity relationships (SAR) because of the disintegration of chemical structures using 2D and/or 3D descriptors, closely by the generation of models got from bioactive ligands. The PASS activity was estimated concerning structures with Pa (probable activity) higher and Pi (probable inactivity) considered for a pharmacological activity.\(^{[3]}\)

2.2.2. ADME analysis

The success of a drug entrant is resolute not only by its most readily useful potential, but in addition to a satisfactory analysis that is ADME that predict of some important capacity to properties in-silico, and it is valuable for analysis regarding the excellent qualities of the molecules. Which evaluates necessary Lipinski's rule the important pharmacokinetic parameters such as for instance ADME. The rule is helpful in drug design and development of a potential drug molecule.\(^{[3]}\)

ADME analysis was carried out using the SWISS ADME and molinsperation predictor for the current investigation.\(^{[3]}\) This web servers as a tool to evaluate the ADMET properties, such as molecular weight< 500, number of <10 Rotatable bonds, hydrogen bond acceptor, hydrogen bond donor (HBDs), molar refractivity, topological polar surface area (TPSA), water solubility (logS), blood brain-barrier, skin permeability (logKP), synthetic accessibility score (SA), percentage absorption, pharmacokinetics, drug-likeness and medicinal chemistry friendliness properties of drug molecules. The lipophilicity of molecules by integrating analysis results obtained from several log P prediction programs such as iLOGP, XLOGP3, WLOGP, MLOGP and SILICOS-IT. A portion of lipophilicity of a molecule is the log of the ratio of the concentration of a drug substance in two solvents in a unionized form, lower the log P value the stronger the lipophilicity is good. The aqueous solubility Log S of a compound are important of absorption and distribution properties, low water solubility often leads to bad absorption and therefore general aim is to avoid poorly soluble compounds. The distribution of log S between -1 and -4 will be improved for better absorption and distribution of drugs in the form.\(^{[3]}\)

2.3. Molecular docking

Molecular docking for the total most part used approach in structure-based drug design. The synthesized compound of ligand was drawn with Chem Draw Ultra version 16.0 (Cambridge Software) followed by resulting molecular mechanics (MM2) energy minization of ligands using ChemBio-3D Ultra version 12.0 with GAMESSS Interface by assuring connection error in the bonds. These energy-minimized ligands (structures) MOL, SDF format of that ligands had been converted to mol2 file using open babel and Discovery studio tool and ligand preparation was done using the Chimera software ended up being used in molecular docking study.\(^{[4]}\) Crystal structure of 3CLpro-2 (PDB code: 6LU7)\(^{[4]}\) was obtained through the Protein Data Bank (http://www.pdb.org) and any heteroatoms, water molecules were eliminated for molecular docking studies. Molecular docking was performed utilizing Autodock Tools 1.5.4 package (http://mgltools.scripps.edu)\(^{[4]}\) and Autodock Vina (version 4.2 docking programs) and SAMSON
(extended docking programs) to grasp the drug molecule interaction with protein, the potential binding mode and energy and analysis the binding affinity of COVID-19, molecular docking analysis ended up being carried out making use of Autodock 4.2.18. Autodock Vina Wizard approach. The grid box parameters values in VINA search space (X = 10.711, Y = 12.411 and Z = 68.83) had been adjusted using the default exhaustiveness value = 8 to maximize the binding conformational analysis. We have the adjusted sufficient grid box size on binding pocket residues to allowing the ligand to move freely into the search space.\textsuperscript{[43]} The synthesized ligand was docked individually against the target protein (6LU7). In docked complexes, the ligand conformational poses were keenly observed to obtain the useful docking results. The generated docked complexes were evaluated based on the lowest binding energy (kcal/mol) values and structure activity relationship (SAR) analyses the clear presence of hydrogen bond, hydrophobic interaction between ligand compound and good control to each focused-on the receptor. The 3D and 2D graphical depictions of all the docked complexes were accomplished by UCSF Chimera 1.10.1, Discovery Studio (2.1.0), PyMOL software and (https://proteins.plus/) online web server.\textsuperscript{[44]}

The torsions of this ligand were groups of detecting the roots in Autodock Vina 1.1.2. Ligand preparation had been finished with adding Gasteiger charges, polar hydrogen. Water molecules and ligand were removed. Protein and grid preparation were done Autodock tools and auto dock Vina 1.1.2 was utilized to perform molecular docking. The results of docking by using AutoDock4 were first converted to *.pdbqt. Docking was done with a Lamarckian genetic algorithm and default parameters. The grid box size was set at 40 Å for x, y and z respectively, and the grid center was set to -11.183, 10.406 and 68.139 for x, y and z respectively, which covered all the amino acid residues in the thought about dynamic pocket. Docking software AutoDock 4.2 Program supplied with AutoGrid 4.0 and AutoDock 4.0 was used to produce grid maps. The spacing between grid points was 0.514 angstroms. The Lamarckian Genetic Algorithm (LGA) was chosen to search for the best conformers. The first ten top-ranked docking poses were saved for each docking run. To approve the molecular docking protocol, the corresponding reference ligands were initially docked into the crystal structure. The Ramachandran plot and characteristics were gotten to from PDB.\textsuperscript{[45,46]}

The protein building and geometric rate assessments of receptor proteins, a-helices, b-sheets, coils and turns were anticipated from online server VADAR 1.8.\textsuperscript{[47]}

3. RESULTS AND DISCUSSION

3.1. Chemistry

3.1.1. Synthesis of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4

The planned 2-phenylquinoline compound was synthesized through single step novel protocol of Multi-component Assembly Process (MCAP) where three or more compounds react to form a single product described in Scheme 1. Novel type reaction of three ketone reactants, (ethyl benzoate, ethyl acetoacetate, methyl 2-aminobenzoate) within the sight of sodium hydride cyclic and followed by hydrolysis in yield suggests value close to 100 % of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4 were prepared by thermal cyclization at relative high temperature (160 °C) with solvent less reaction environments. The resulting yellow is solution was stirred for a half hour between 0 °C and room temperature. After normal aqueous workup, the condensed residue was purified by silica-gel column chromatography to give the light-yellow needle crystal solid as 80-95 % yields. Structure of the compound was well established by gas Chromatography, IR, GC-MS, \textsuperscript{1}H NMR and \textsuperscript{13}C NMR spectral data with elemental analysis. The compound 4 found to be in 90 % good yield. The identity of the reported compound was confirmed by comparing it’s melting point 176-180 °C and molecular masses from GC–MS with the reported and calculated values, respectively. Its molecular formula, C\textsubscript{18}H\textsubscript{15}NO\textsubscript{2}, was identified by CHN analysis data and molecular mass was justified by the molecular ion peak in its EI-MS at m/z 255. For main fragmentation ions were at m/z 244.0531, 196.1694, 179.1977, 146.0727, 119.1200, 105.9482, 90.1190 and 76.5125, FT-IR spectroscopy to insist various functionalities.

The prominent absorption bands in IR spectrum appeared at 3488 cm\textsuperscript{-1} which is due to the (-OH stretching), 3269 cm\textsuperscript{-1} (N-H stretching), 2949 cm\textsuperscript{-1} (-CH\textsubscript{2}-stretching), 1589 cm\textsuperscript{-1} (C=H of aromatic ring), 1668 (C=C stretching of aromatic ring), 1693cm\textsuperscript{-1} due to stretching frequency of ketone functional group respectively, bond at 1598 cm\textsuperscript{-1} is due to C-N group further confirms the structure.

In \textsuperscript{1}H NMR spectrum of 4 showed a downfield board singlet at δ 11.93 is due to quinoline N-H protons, doublet at δ 8.84 is due to quinoline C=H protons. Aromatic protons appeared as multiplet at δ 7.96-7.94, 7.50-7.38, 7.18-7.08, 7.01-6.97, 6.57-6.51 and doublet δ 7.74-7.72 are due to phenyl ring and quinoline C\textsubscript{6-8}-H, appeared as a triplet at δ 6.52 is due
to quinoline C2-H, and similarly to multiplets at δ 3.86 -3.83 due C3-H. Board singlet peak appeared at δ 5.75 and 3.744 are due to ethyl-OH and -CH2-protons respectively, 13C NMR spectra unambiguously matched with the structure of their corresponding 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4.

Scheme 1: Synthetic pathway of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4.

Reagents and conditions: (i) NaH, 160 °C, rt, 4-6 hrs, 80-96 % yields

A possible mechanism for the formation of selected product 4 is Claisen ester condensation by the sodium hydride base to give the enolate anion which is stabilized by resonance. The next step is the nucleophilic addition of the enolate anion, to the ketonic group of amine group, followed by the proton transfer and subsequent elimination of acetic acid and water can take place, leading to an intermediate involving the alkaline mediated hydrogen peroxide oxidation to give product 4.

Scheme 2: The possible reaction mechanism is outlined for synthesis of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4

SMILES:

[H]OC([H])([H])1([H])C(=O)C2=C(N([H])\[C@\]1([H])C([H])=C([H])=C([H])=C1[H]) C([H])=C([H])C([H])=C2[H].

3.2. Pharmacology

3.2.1. Biological activity spectrum PASS analysis

The synthesized compounds were determined by online server of PASS. In the pass analysis of compound 4 the pa is showed (0.614) highest Pa value for Antiviral activities (Picornavirus) (table 2) refer to http://www.way2drug.com/PASSOnline/predict.php.

3.2.2. ADME Analysis

The predicted chemo-informatic properties had been evaluated by computational tool-aided in-silico studies demonstrably suggesting that the compounds had drug-like prospect properties. It absolutely was interesting to see that the results associated with the SWISS ADME and Molinspiration predictor values of log P, molar refractivity with the total polar surface area in these molecules were in excellent agreement with the most important rules of drug-likeness. The predicted chemo-informatics properties were evaluated by computational tools. Results exposed that compound 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one, has better predicted value of molecular weight 253.30 (g/mol), was within the range value (< 500 g/mol) which greater molecular weight compound compared to borderline value. ADME parameters of H-bond acceptor and H-donor, polar surface area and molar volume. Though this compound exhibited a good hydrophilicity-lipophilicity stability therefore the predicted bioavailability, high lipophilicity was expected to show decent GI absorption. Another key property that is related to drug bioavailability. Thus, pass analysis is demonstrated to have low oral bioavailability, the molecular polar surface area (PSA) is a very useful parameter for drug transport properties, molecule is well-defined as the surface sum over all polar atoms, primarily oxygens, nitrogen and attached hydrogen atoms. This parameter has been shown to correlate very finely with the human intestinal absorption, Caco-2 monolayers, permeability and blood-brain barrier penetration. The PSA parameter is commonly used for a drug optimization ability to permeate cells. Prior research data showed the standard value of PSA (< 89 A2).
The Lipinski’s rule RO5 of result 0 violation showed that compound 4, possess good molecular weight (g/mol), two Hydrogen Bond Accepted and Hydrogen Bond Donor values two log P 2.45 which are significantly justified if their drug likeness behaviour were justifiable with the standard values. The RO5 deviation (Mol. Wt ≥ 500 g/mol; HBD ≥ 5; HBA > 10; and log P ≥ 5), drug-likeness score is an amalgam of complex balance of several molecular properties and structure features which determine the behaviour of molecules drug. The drug score values 0.55 good, which depicted their drug like to be suitable against COVID-19. The results got from the Swiss ADME and Mol inspiration search engine are listed in table 1.

Table 1: Physicochemical descriptors and ADME parameters

| Property               | Value     |
|------------------------|-----------|
| M.W, g/mol             | 253.30    |
| RB                     | 2         |
| H-A                    | 2         |
| H-D                    | 2         |
| TPSA                   | 49.33 Å²  |
| MR                     | 77.22     |
| WlogP (lipophilicity)  | 2.25      |
| ESOL logS              | -3.52     |
| BBB Permeant           | Yes       |
| log Kp cm⁻¹            | -5.84     |
| Lipinski violations    | 0         |
| PAINS alerts           | 0         |
| GI absorption          | High      |
| Synthesis Accessibility| 2.79      |

*R bond = Rotatable bond, H-A = Hydrogen bond acceptor, H-D = hydrogen bond donor, TPSA = topological polar surface area, BBB = blood brain-barrier, log P = lipophilicity, log S = water solubility, log Kp = permeability coefficient, PAINS = pan-assay interference structure.

The boiled-egg diagram analysis shows that the compound strong within the permissible range of standard drugs, blue dot indicates cannot be affected by the P-glycoprotein of the CNS system by P-glycoprotein, point locate in Boiled Eggs yolk is a molecule passively permeate through the blood-brain barrier (BBB). In the current study, the synthesized ligand and its complexes was initiate d to be in a good pact with the criteria and can be said to possess good bioavailability.

3.3. Molecular docking

Molecular docking studies technique in medicinal chemistry has led to advances in drug discovery and design. This technique explores the binding mode and affinity of a small molecule within the binding site of the receptor target protein. The docked ligands were ranked according to their binding affinity in a ligand-receptor (figures 3 and 4a-f). Molecular docking was performed on compound 4, against the 3CLpro-2 (6LU7) main protease to identify the ligand-protein interactions.

Figure 1: Bioavailability radar graph of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one, (pink area reflects the allowed values of drug likeness properties of the molecule)

Figure 2: ADME properties of compound 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one by graphical representation (boiled-egg)

The docking scores is -7.39 due to the tested ligand determined that the prepared mixtures possessed potential for interaction with one or more amino acids in the active site of the receptor. The docking result showed that six hydrogen bonds, one electrostatic bond and one hydrophobic interaction were observed in synthesized docked complex. Both oxygen molecules of amine groups in 4 formed six conventional strong hydrogen bonds with Gly143:HN, Ser144:HN, Ser 144: HG, Cys145:HN, Leu141:O and Lig-H:O with bond lengths of 2.30, 2.38, 2.79, 2.37, 2.21 and 2.56 Å (COVID-19 Main protease) respectively. Similarly, a Pi-Alkyl hydrophilic interaction was observed between
quinoline phenyl group and Cys145 having bond distance of 4.87 Å. Another positive Pi-Cation electrostatic bond was observed between quinoline ring and His41 having bond length of 4.87 Å. Hydrogen bonds play an important role in molecular docking because they help to stabilize and strengthen the docked enzyme–inhibitor complex.

The shorter distances (< 3.0-3.5 Å) give greater stability to ligand-protein docking complexes compared to larger ones having distance > 4.0 Å. However, for hydrophobic interactions, distances vary up to 5 Å. Synthesized compounds showed practical of great docking scores and binding energy to the selected protein targets ranged from SWISS docking -7.39 dG kcal mol⁻¹ with full fitness -1202.74 kcal mol⁻¹, autodock result of binding energy -7.52 kcal mol⁻¹ and reference RMSD 64.07 shows RMSD (~2 Å) values of the inhibitor compound were found reliable. Autodock vina According to the got results, it was decided that the docked ligand form a stable complex with COVID-19 inhibitor. For 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one nine representative binding modes have

Table 2: The activity spectrum of 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one, pa represents probability to active and pi represents probability to be inactive

| S. No | Activity name                          | pa    | pi     |
|-------|----------------------------------------|-------|--------|
| 1     | Antiviral (Picornavirus)               | 0.614 | 0.016  |
| 2     | 5 Hydroxytryptamine release stimulant  | 0.613 | 0.037  |
| 3     | Pterin deaminase inhibitor             | 0.594 | 0.028  |
| 4     | Saccharopepsin inhibitor               | 0.626 | 0.066  |
| 5     | Acrocylinropepsin inhibitor            | 0.626 | 0.066  |

Figure 3: (A) The docked ligand 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4 at the same catalytic site receptor, (B) The docking poses of (3-hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one 4 in the binding site of the COVID-19 receptor PDB ID: 6LU7 (C) interaction with the ligand 4 the H-bond surfaces of the receptor COVID-19 inhibitor, (D) 3D Docking poses of compound 4 into the N3 binding site of the COVID-19 main protease
been detected with binding affinities ranging from -6.2 to -5.3 kcal/mol (table 3). As it can be seen somewhat higher binding affinity has been calculated in the case of 3-hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one while this molecule can be considered as a lead compound for the development of new COVID-19 drugs.

Table 3: The binding affinity values of different poses of compound (3-hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one predicted by autodock Vina

| Mode | Affinity (kcal/mol) | Distance from the best mode RMSD 1.b. | RMSD u.b. |
|------|---------------------|--------------------------------------|-----------|
| 1    | -6.7                | 0.000                                | 0.000     |
| 2    | -6.5                | 2.007                                | 3.670     |
| 3    | -6.5                | 2.410                                | 5.861     |
| 4    | -6.4                | 2.825                                | 6.025     |
| 5    | -6.3                | 1.402                                | 5.214     |
| 6    | -6.3                | 3.759                                | 4.871     |
| 7    | -6.3                | 2.006                                | 2.596     |
| 8    | -6.0                | 2.981                                | 3.936     |
| 9    | -6.0                | 3.742                                | 5.091     |

The docking model was also estimated for structural and stereochemical quality. The colouring/shading on the plot represents the different regions were as described Ramachandran phi-psi plot for compound 4 (Fig. 6A) revealed that 91 % of residues 281 lay in the phi-psi core region (Red), another 23 residues 7 % were in the allowed region (Yellow), 0 % of residues were in the generous region (Green) and only 0 % lay residues in the outside region (Gray). The analysis of predicted structure afford solid evidence that the predicted 3D structure of synthesized compound 4 is of excellent quality.

Figure 4: (a) 2D interaction with the ligand 4 the H-bond surfaces of the receptor COVID-19 inhibitor, (b) 2D Docking poses of compound 4 into the N3 binding site of the COVID-19 main protease.

The Prediction of the accessible surface areas (ASA) of each amino acid in the sequence along with the fractional residual volume available for the amino acids in the main and in the side chains, an area accessible to water molecules on the protein structure, which was measured in square angstroms or as fractional ASA ranging from 0 to 1 (figure 6B). The hydrophilic residues have engaged a significant fraction of ASA, with hydrophobic residues making only a minor fraction. The common residues in a docking compound 4, have ASA scores less than 0.8, the representative tight folding normally leaves residues inaccessible to water molecules. We predictable accessible surface areas of wholly residues of the active site quality of the model with regard to the stereo chemical packaging and 3D profile quality were also predicted by using the VADAR free web server (figure 7).

4. CONCLUSION

In summary, an efficient 3-(hydroxymethyl)-2-phenyl-2,3-dihydroquinolin-4(1H)-one was synthesized using sodium hydride, an effective, base and reusable catalyst, and performing a solvent-free condition. Structurally diverse small molecules useful for medicinal chemistry and drug discovery. The in-silico inhibition studies of synthesized compounds binding to the (6LU7) main protease showed that the compounds, and exhibited high binding affinities (-7.52 kcal/mol by AutoDock 4 and -6.70 by AutoDock Vina). These results are, under fact, that configuration can assume in the binding site a favourable orientation. The in-silico ADME reporting toxicity, drug likeness, drug scoring results, PASS analysis and in vitro anti-COVID-19 suggested that...
Figure 6: (A) Ramachandran plot of the compound 4 showing 91% of amino acid residues in the core region into the N3 binding site of the COVID-19 main protease (B) accessible surface area of compound 4 VADAR online servers for model the COVID-19 main protease.

Figure 7: (a) Fractional accessible surface area of compound 4 VADAR online servers for model the COVID-19 main protease, (b) 3D profile quality of compound 4 VADAR online servers for model the COVID-19 main protease.

the compound is a promising lead for the development of selective, safe and potent COVID-19.

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REFERENCES

1. N. Zhu, D. Zhang, W. Wang, X. Li, B. Yang, J. Song, X. Zhao, B. Huang, W. Shi, R. Lu, P. Niu, F. Zhan, X. Ma, D. Wang, W. Xu, G. Wu, G. F. Gao, W. A. Tan. Novel coronavirus from patients with pneumonia in China, N. Engl. J. Med., 2019, 382(8), 727-733.

2. A. Wu, Y. Peng, B. Huang, X. Ding, X. Wang, P. Niu, J. Meng, Z. Zhu, Z. Zhang, J. Wang, J. Sheng, L. Quan, Z. Xia, W. Tan, G. Cheng, T. Jiang. Genome Composition and Divergence of the novel coronavirus (2019-nCoV) originating in China, Cell Host. Microbe., 2020, 27(3), 325-328.

3. ArcGIS. COVID-19 Dashboard by the Centre for 2011, Systems Science and Engineering (CSSE), Johns Hopkins University (JHU), 2020.

4. N. Decaro. Alphacoronavirus Coronaviridae, The Springer Index of Viruses, 2011, 371-383.

5. N. Decaro. Betacoronavirus Coronaviridae, The Springer Index of Viruses, 2011, 385-401.

6. M. Hoffmann, H. Kleine-Weber, S. Schroeder, N. Krüger, T. Herrler, S. Erichsen, T. S. Schiergens, G. Herrler, N. Wu, A. Nitsche, M. A. Müller, C. Drosten, S. Pöhlmann. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor, Cell, 2020, 181, 271-280.

7. A. R. Fehr, S. Perlman. Coronaviruses: An overview of their replication and pathogenesis, Methods. Mol. Biol., 2015, 1282, 1-23.

8. A. D. Reddy, S. B. Suh, R. Ghaffari, N. J. Singh, D. J. Kim, J. H. Han, K. S. Kim. Bioinformatics analysis of SARS proteins and molecular dynamics simulated structure of an alpha-helix motif, Bull. Korean Chem. Soc., 2003, 24, 899-900.
9. Z. Jin, X. Du, Y. Xu, Y. Deng, M. Liu, Y. Zhao, B. Zhang, X. Li, L. Zhang, C. Peng, Y. Duan, J. Yu, L. Wang, K. Yang, K. Liu, R. Jiang, X. Yang, T. You, X. Liu, X. Yang, F. Bai, H. Liu, X. Liu, L. W. Guddat, W. Xu, G. Xiao, C. Qin, Z. Shi, H. Jiang, Z. Rao, H. Yang. Structure of M^2 from SARS-CoV-2 and its discovery of inhibitors, *Nature*, **2020**, 582, 289-293.

10. C. H. Wu, S. H. Yeh, Y. G. Tsay, Y. H. Shieh, C. L. Kao, Y. S. Chen, S. H. Wang, T. J. Kuo, D. S. Chen, P. J. Chen. Glycogen synthase kinase-3 regulates the phosphorylation of severe acute respiratory syndrome coronavirus nucleocapsid protein and viral replication, *J. Biol. Chem.*, **2009**, 284, 5229-5239.

11. P. Zhou, X. Yang, X. G. Wang, B. Hu, L. Zhang, W. Zhang, H. R. Si, Y. Zhu, B. Li, C. L. Huang, H. D. Chen, J. Chen, Y. Luo, H. Guo, R. D. Jiang, M. Q. Liu, Y. Chen, X. R. Shen, X. Wang, X. S. Zheng, K. Zhao, Q. J. Chen, F. Deng, L. L. Liu, B. Yan, F. X. Zhan, Y. Y. Wang, G. F. Xiao, Z. L. Shi. Discovery of a novel coronavirus associated with the recent pneumonia outbreak in humans and its potential bat origin, *Nature*, **2020**, 579, 270-273. doi.org/10.1038/s41586-020-2012-7.

12. A. Marella, O. P. Tanwar, R. Saha, M. R. Ali, S. Srivastava, M. Akhter, M. Shaququzzaman, M. M. Alam. Quinolone: A versatile heterocyclic, *Saudi Pharm. J.*, **2013**, 21, 1-12.

13. X. Y. Jin, H. Chen, D. Li, A. Li, W. Y. Wang, N. Gu. Design, synthesis, and anticancer evaluation of novel quinoline derivatives of ursoic acid with hydrazide, oxadiazole, and thiadiazole moieties as potent MEK inhibitors, *J. Enzyme. Inhib. Med. Chem.*, **2019**, 34, 1, 955-972.

14. X. Ngqor, N. Tobeka. B. A. Aderibigbe. Quinoline based hybrid compounds with antimalarial activity, *Molecules*, **2017**, 22, 2268.

15. D. Insuasty, O. Vidal, O. Bernal, E. Marquez, J. Guzman, B. Insuasty, J. Quiroga, L. Svetaz, S. Zaccchino, G. Puerto, R. Abonia. Antimicrobial activity of quinoline-based hydroxymidazolium hybrids, *Antibiotics*, **2019**, 8, 239.

16. N. Muruganantham, R. Sivakumar, N. Anbalagan, V. Gunasekaran, J. T. Leonard. Synthesis, anticonvulsant and antihypertensive activities of 8-substituted quinoline derivatives, *Biol. Pharm. Bull.*, **2004**, 27, 1683-1687.

17. S. K. Gupta, A. Mishra. Synthesis, Characterization and screening for anti-inflammatory and analgesic activity of quinoline derivatives bearing azetidinone scaffolds, *Anti. Inflamm. Antiallergy Agents. Med. Chem.*, **2016**, 15, 31-43.

18. S. Kumar, G. Himanshu. Biological activities of quinoline derivatives, *Mini-Rev. Med. Chem.*, **2009**, 9, 1648.

19. H. Nikookar, M. Mohammadi-Khanaposhani, S. Imanparast, M. A. Faramarzi, P. R. Ranjarb, M. Mahdavi, B. Larijani. Design, synthesis and in vitro α-glucosidase inhibition of novel dihydropyran[3,2-c]quinoline derivatives as potential anti-diabetic agents, *Bioorg. Chem.*, **2018**, 77, 280-286.

20. A. Upadhyay, P. Kushwaha, S. Gupta R. P. Dodda, K. Ramalingam, R. Kant, N. Goyal, K. V. Sashidhara. Synthesis and evaluation of novel triazolyl quinoline derivatives as potential antileishmanial agents, *Eur. J. Med. Chem.*, **2018**, 154, 172-181.

21. P. Colson, J. M. Rolain, D. Raout. Chloroquine for the 2019 novel coronavirus SARS-CoV-2, *Int. J. Antimicrob. Agents.*, **2020**, 55, 105923.

22. A. Cortegiani, G. Ingoglia, M. IPPolito, A. Giarratano, S. A. Eina. Systematic review on the efficacy and safety of chloroquine for the treatment of COVID-19, *J. Crit. Care.*, **2020**, 57, 279-283.

23. J. Gao, Z. Tian, X. Yang. Breakthrough: Chloroquine phosphate has shown apparent efficacy in treatment of COVID-19 associated pneumonia in clinical studies, *Biosci. Trends.*, **2020**, 14, 72-73.

24. F. Mojab. Antimalarial natural products: a review. *Avicenna J. PhytoMed.*, **2012**, 2, 52-62.

25. R. Shetty, A. Ghosh, S. G. Honavar, P. Khamar, S. Sethu. Therapeutic opportunities to manage COVID-19/SARS-CoV-2 infection: Present and future, *Indian. J. Ophthalmol.*, **2020**, 68, 693-702.

26. S. Jain, V. Chandra, P. K. Jain, K. Pathak, D. Pathak, A. Vaidya. Comprehensive review on current developments of quinoline-based anticancer agents, *Arab. J. Chem.*, **2019**, 12, 4920-4946.

27. A. Nepolraj, P. Pitchai, P. Mani. One-pot Synthesis of Pyrano [2,3-a] Quinoline via the Tandem Cyclization of Algar-Flyn-Oyamanda Reactions, *Organic Chemistry Research*, **2019**, 5, 167-173.

28. R. A. Mekheimer, M. A. Al-Sheikh, H. Y. Medrasi, K. U. Sadek. Advancements in the synthesis of fused tetracyclic quinoline derivatives, *RSC. Adv.*, **2020**, 10, 19867.

29. S. Madapa, Z. Tusi, S. Batra. Advances in the Syntheses of Quinoline and Quinoline-Annulated Ring Systems, *Curr. Org. Chem.*, **2008**, 12, 1116-1183.

30. N. D. Heindel, I. S. Bechara, P. D. Kennewell, J. Molnar, C. J. Ohnmacht, S. M. Lemke, T. F. Lemke. Antihypertensive 2-amino-4(3H)-quinazolinones, *J. Med. Chem.*, **1968**, 11, 1218-1221.

31. Y. M. Poronik, J. Klajn, W. Borzęcka, D. T. Gryko. The Niementowski reaction of anthranilic acid with ethyl acetooacetate revisited: a new access to pyrano[3,2-c]quinoline-2,5-dione, *Arkivoc.*, **2017**, 2, 7-11.

32. Y. C. Wu, L. Liu, H. J. Li. Skraup-Doebner-Von Miller Quinoline Synthesis Revisited: Reversal of the Regiochemistry for γ-Aryl-β-γ-unsaturated α-Ketoesters, *J. Org. Chem.*, **2006**, 17, 6592-6595.

33. M. A. Alonso, J. I. Úbeda, C. Avendaño, C. Menéndez, M. Villacampa. New findings on the Vilsmeier-Haack approach to quinoline derivatives, *Tetrahedron*, **1993**, 49, 10997-11008.

Q. Lv, L. Fang, P. Wang. A simple one-pot synthesis of quinoline-4-carboxylic acid derivatives by
Andrographolide as a potential inhibitor of SARS-CoV-2 main protease: An in-silico approach, J. Biomol. Struct. Dyn., 2020, doi: 10.1080/07391102.2020.1760136.

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