Research Article

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Potential PDE4B inhibitors as promising candidates against SARS-CoV-2 infection

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Abstract: Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) is an RNA virus belonging to the coronavirus family responsible for coronavirus disease 2019 (COVID-19). It primarily affects the pulmonary system, which is the target of chronic obstructive pulmonary disease (COPD), for which many new compounds have been developed. In this study, phosphodiesterase 4 (PDE4) inhibitors are being investigated. The inhibition of PDE4 enzyme produces anti-inflammatory and bronchodilator effects in the lung by inducing an increase in cAMP concentrations. Piclamilast and rolipram are known selective inhibitors of PDE4, which are unfortunately endowed with common side effects, such as nausea and emesis. The selective inhibition of the phosphodiesterase 4B (PDE4B) subtype may represent an intriguing technique for combating this highly contagious disease with fewer side effects. In this article, molecular docking studies for the selective inhibition of the PDE4B enzyme have been carried out on 21 in-house compounds. The compounds were docked into the pocket of the PDE4B catalytic site, and in most cases, they were almost completely superimposed onto piclamilast. Then, in order to enlarge our study, drug-likeness prediction studies were performed on the compounds under study.

Keywords: phosphodiesterases, SARS-CoV-2, molecular docking, COVID-19, PDE4B, piclamilast, rolipram, drug-likeness predictions

1 Introduction

Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) is a member of the Coronavirus family responsible for the worldwide pandemic of human respiratory illness coronavirus disease 2019 (COVID-19) [1]. Several studies are addressed to find new drug targets and vaccines to combat this disease [2–6]. Among the different involved targets, cyclic nucleotide phosphodiesterases (PDEs) are being investigated, a large superfamily of enzymes that play a major role in intracellular signaling by controlling tissue cAMP (cyclic 3′,5′-AMP) and cGMP (cyclic 3′,5′-GMP) levels in response to receptor activation. Among the 11 subtypes of the PDE family, PDE4 is the principal cAMP-metabolizing enzyme found in the immune and inflammatory cells. The inhibition of PDE4 may induce an increase in cAMP [7,8] and prolongation of the anti-inflammatory effect with a consequent bronchodilator effect. Thus, PDE4 inhibitors have been proven as anti-inflammatory agents against different pulmonary disorders by inhibiting the release of inflammatory signals and cytokines [9]. This inhibition has been addressed for diverse diseases [10], including inflammatory diseases [11], the topical treatment of psoriasis [12] and atopic dermatitis [13], cancer [14], inflammatory bowel diseases [15,16], neurological disorders [17], diabetic nephropathy [18], and pulmonary dysfunctions in COVID-19, such as chronic obstructive pulmonary disease (COPD) [19,20]. Several studies are being carried out on PDE4 inhibitors in order to understand their mechanism of action. Our study was focused on rolipram and piclamilast (RP 73401) (Figure 1) [21,22].

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Rolipram (4-(3-cyclopentyloxy-4-methoxyphenyl)-2-pyrrolidone, A, Figure 1) was designed as a COPD therapeutic target acting as a PDE4 inhibitor [23,24]. PDE4 inhibition by piclamilast (3-cyclopentyloxy-*N*-(3,5-dichloropyridin-4-yl)-4-methoxybenzamide, B, Figure 1) was effective in preventing blast-induced long-term potentiation deficits [25]. A possible mechanism of action responsible for the anti-inflammatory and immunomodulatory effects of this class of compounds has been recently suggested by Nguyen et al. [26] for tanimilast, a new potent and selective inhaled inhibitor of PDE4 in advanced clinical development for the treatment of COPD. The study was carried out *in vitro* on a model of dendritic cell activation by SARS-CoV-2 genomic ssRNA (SCV2-RNA). Tanimilast lowered the release of pro-inflammatory cytokines (TNF-α and IL-6), chemokines (CCL3, CXCL9, and CXCL10), and Th1-polarizing cytokines (IL-12, type I IFNs).

PDE4, which consists of different isoforms [27], comprises four subtypes, PDE4A, B, C, and D. Non-selective PDE4 inhibitors, which bind all four PDE4 subtypes simultaneously, produce many promising therapeutic benefits, accompanied, however, by undesired side effects, notably, nausea, diarrhea, and emesis [28]; they may also induce gastroparesis, as demonstrated in mice [29]. Thus, the selective inhibition of a single subtype may be addressed. In particular, our study was aimed at the PDE4B subtype, which has been related to diverse activities. PDE4B inhibitors have also been suggested as promising therapeutic targets for *Pseudomonas aeruginosa*, which is a frequent cause of hospital-acquired lung infections [30], and other molecules have been designed and studied as potential PDE4B inhibitors to reduce or block inflammatory processes of the respiratory tract [31–33]. Indeed, PDE4B regulates the pro-inflammatory toll receptor – tumor necrosis factor α pathway in monocytes, macrophages, and microglial cells. Interestingly, it has been demonstrated that PDE4B, which is the predominant form in the immune and respiratory systems, is involved in human respiratory illness COVID-19, the worldwide pandemic caused by SARS-CoV-2 [34,35]. Therefore, it represents a molecular target for anti-inflammatory and antiviral drugs. Unfortunately, to date, developing selective PDE4B inhibitors is not easy as the amino acid sequence of the PDE4 active site is identical in all PDE4 subtypes (PDE4A-D) [36]. However, recently, a modeling study was carried out on PDE4B and PDE4D active cavities [37]. Among the PDE4 selective inhibitors, roflumilast, which shows a higher affinity to PDE4B than PDE4A, C, and D, represents a potential and effective therapy for COVID-19 [38]. Moreover, a PDE4B selective inhibitor, BI 1015550, has been proposed as a clinical drug candidate for the oral treatment of idiopathic pulmonary fibrosis [39]. In this view, our work aimed at designing new selective inhibitors of PDE4B as potential therapeutic agents for COVID-19 disease. Specifically, 21 in-house molecules were examined and correlated with the structures of piclamilast and rolipram in order to seek greater possible interactions with the catalytic site of the A chain of PDE4 [40]. Recently, a modeling study was carried out on PDE4B and PDE4D active cavities [37]. Piclamilast and rolipram were chosen since, in the literature, they are among the major PDE4 inhibitors (IC_{50} = 1.1 and 1,200 nM for piclamilast and rolipram, respectively) [41]. The magnesium and zinc ions present at the PDE4B catalytic site were taken into account for the docking study using AutoDock Vina software, as they could be involved in PDE4B inhibitory activity. Drug-likeness predictions, i.e., physicochemical-pharmacokinetic and absorption, distribution, metabolism, and excretion (ADME) properties, of the studied compounds have also been performed. These studies will allow us to select the best compounds to be proposed for future in-depth *in vitro* and/or *in vivo* studies. We are sure that identifying potentially active molecules could be important for the discovery and development of new drugs for COVID-19 and at the same time useful for the study of the molecular mechanisms involved in this devastating disease.

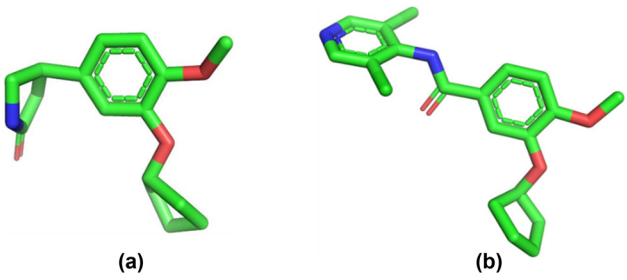


Figure 1: PyMOL version of rolipram (a) and piclamilast (b).

2 Methodology

2.1 Molecular modeling studies

A molecular docking study was performed on piclamilast and rolipram in the catalytic site of PDE4B, located mainly on the A chain of PDE4B (PDB code: 1XM4) [42]. AutoDock Vina was used for docking [43]. The magnesium and zinc ions present at the catalytic site of PDE4B were also considered. With this study, it is possible to predict the structure of the ligand–protein intermolecular complex and determine what is known as Pose or Binding Mode, which

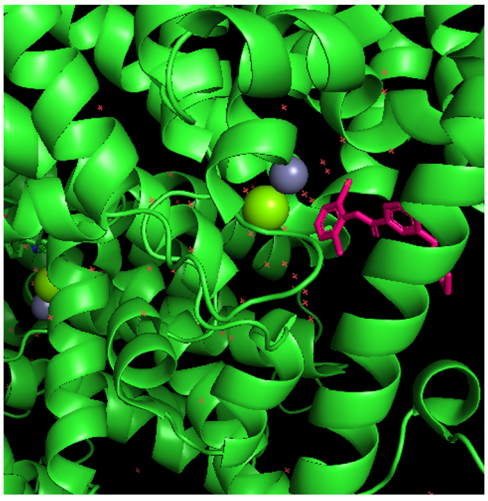


Figure 2: Catalytic domain of human PDE4B in complex with piclamilast (in magenta).

is the position, orientation, and conformation that each ligand assumes on the surface of the biological receptor macromolecule. All the side chains of the flexible amino acid residues selected have been included in the flex file, that is, those side chains present in the active site on the A chain of PDE4B in that could have a probable interaction with the ligand (Figure 2). In particular, the selected residues are ARG‘409, ASN‘235, ASN‘283, ASN‘395, ASP‘275, ASP‘346, ASP‘392, CME‘432, GLN‘284, GLN‘443, GLU‘304, HIS‘234, HIS‘238, HIS‘274, HIS‘278, HIS‘307, ILE‘410, ILE‘450, LEU‘303, LEU‘393, MET‘347, MET‘411, MET‘431, PHE‘414, PHE‘446, SER‘229, SER‘236, SER‘282, SER‘348, SER‘429, SER‘442, THR‘345, THR‘407, TYR‘233, TYR‘403, TYR‘449. Also, for the ligand, a file with the extension.pdbqt has been created, in which its atoms have been defined, also its potentials and its degrees of freedom. This last process was obviously repeated for all ligands tested.

Table 1: Docking study with piclamilast

Pose	Binding energy “affinity” (kcal/mol)	RMSDL.b.	RMSDU.b.
1	−10.1	0.000	0.000
2	−9.5	0.851	1.392
3	−9.4	0.807	2.110
4	−8.4	1.023	2.553
5	−8.4	1.480	2.512
6	−8.0	1.276	2.087
7	−7.8	1.624	4.096
8	−7.8	1.734	2.126
9	−7.7	1.520	2.351

The average RMSD of the poses are compared with pose 1.

Table 2: Docking study with rolipram

Pose	Binding energy “affinity” (kcal/mol)	RMSDL.b.	RMSDU.b.
1	−8.4	0.000	0.000
2	−8.4	1.124	2.156
3	−8.2	1.322	3.113
4	−8.2	1.291	2.763
5	−8.1	1.355	2.998
6	−8.1	1.602	2.875
7	−7.8	1.237	1.902
8	−7.7	1.354	2.961
9	−7.5	1.458	2.124

The average RMSD of the poses are compared with pose 1.

2.2 Drug-likeness predictions – physicochemical-pharmacokinetic/ADME properties

Drug-likeness is one of the qualitative ideas employed for predicting drug-like properties. It is designated as an intricate balance of diverse molecular and structural features that assesses qualitatively the chance for a molecule to become an oral drug with respect to bioavailability. The targeted molecules were evaluated for predicting drug-likeness based on five separate filters, namely, Egan, Ghose, Muegge, Veber, and Lipinski rules accompanying bioavailability and drug-likeness scores using the Molsoft software and Swiss ADME program (http://swissadme.ch) using the ChemAxon’s Marvin JS structure drawing tool [44–47]. Drug-likeness was established from structural or physicochemical inspections of development compounds advanced enough to be considered oral drug candidates.

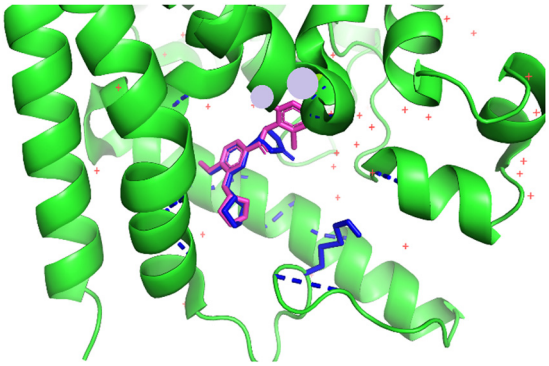
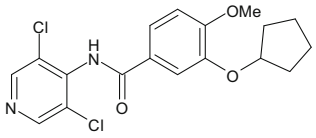
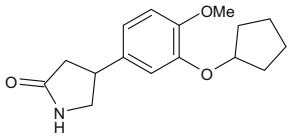
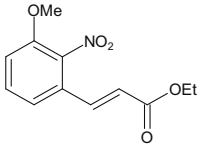
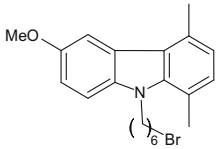
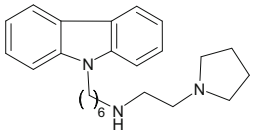
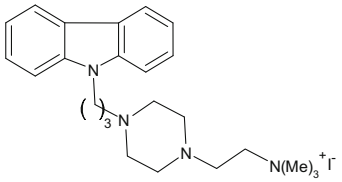
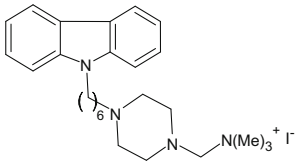
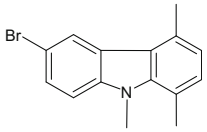
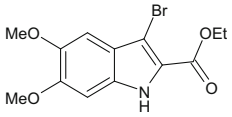
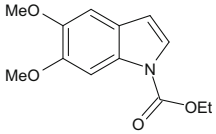
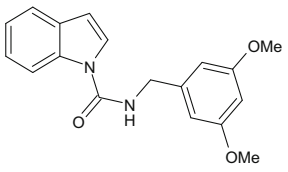


Figure 3: Comparison between the crystalline structure of piclamilast (in magenta) with pose 1 of the docking of rolipram (in blue). Part of the catalytic site of PDE4B with part of the flexible residues is visible in green.

Table 3: Energy levels of the first 5 poses out of a total of 9 of each molecule (whose name is indicated in the first column) with an exhaustiveness level of 16, obtained by molecular docking compared to piclamilast and rolipram with the program AutoDock Vina

Compound	Structure	1	2	3	4	5
Piclamilast		−10.1	−9.5	−9.4	−8.4	−8.4
Rolipram		−8.4	−8.4	−8.2	−8.2	−8.1
1		−6.8	−6.8	−6.8	−6.7	−6.6
2		−9.1	−8.6	−8.3	−8.3	−8.0
3		−9.5	−9.4	−9.3	−9.2	−9.0
4		−10.2	−10.0	−9.7	−9.6	−8.6
5		−10.1	−9.9	−9.8	−9.8	−9.6
6		−9.8	−9.5	−9.5	−9.1	−7.9
7		−7.5	−7.5	−7.3	−7.1	−6.9
8		−7.4	−7.1	−7.1	−7.1	−7.0
9		−9.1	−9.1	−8.8	−8.7	−8.6

(Continued)

Table 3: *Continued*

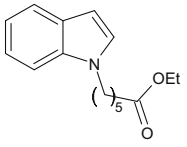
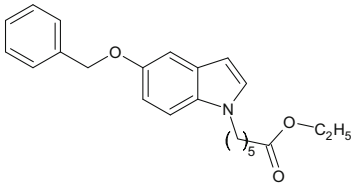
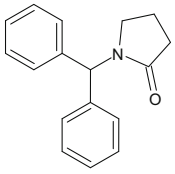
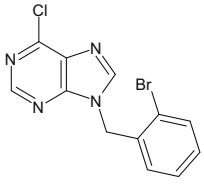
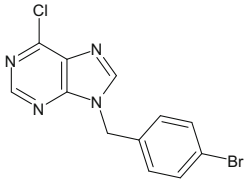
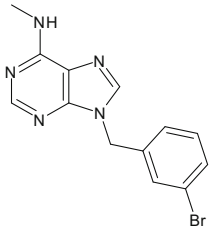
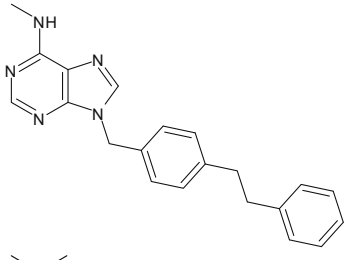
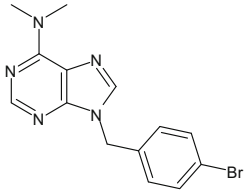
Compound	Structure	1	2	3	4	5
10		−7.5	−7.3	−7.3	−7.3	−7.1
11		−8.3	−8.3	−8.1	−8.0	−8.0
12		−10.0	−9.1	−8.9	−8.8	−8.6
13		−8.0	−7.6	−7.6	−7.2	−7.1
14		−7.2	−7.0	−6.9	−6.8	−6.2
15		−7.8	−7.7	−7.7	−7.6	−7.3
16		−10.0	−9.9	−9.5	−9.4	−9.3
17		−7.9	−7.9	−7.9	−7.4	−6.8

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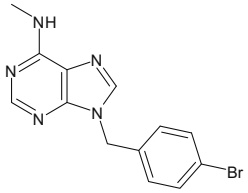
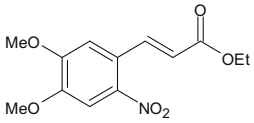
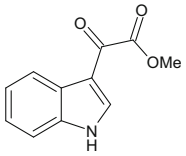
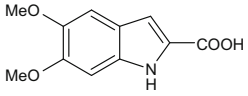
Table 3: *Continued*

Compound	Structure	1	2	3	4	5
18		−7.5	−7.2	−7.0	−6.7	−6.5
19		−7.0	−6.9	−6.7	−6.7	−6.6
20		−7.0	−6.5	−6.4	−6.1	−6.1
21		−7.6	−7.3	−7.3	−7.0	−7.0

3 Results and discussion

3.1 Piclamilast

Piclamilast is a selective second-generation PDE4 inhibitor with important anti-inflammatory effects [48]. It has been studied for its applications in treating conditions such as COPD, bronchopulmonary dysplasia, and asthma. It acts through the selective inhibition of the four PDE4 isoforms (PDE4A-D) while showing no inhibition of the other PDEs [49]. PDE4 isoforms are particularly important for inflammatory and immunomodulatory cells. They are the most common PDE in inflammatory cells such as mast cells, neutrophils, basophils, eosinophils, T lymphocytes, macrophages, and structural cells such as sensory nerves and epithelial cells. Inhibition of PDE4 blocks the hydrolysis of cAMP, thereby increasing the levels of cAMP within the cells. cAMP suppresses the activity of immune and inflammatory cells; thus, the inhibition of PDE4 in a mouse model of induced chronic lung disease demonstrated anti-inflammatory properties, determined the reduction of pulmonary fibrin deposition and alveolar vascular loss, and prolonged survival in hyperoxia-induced neonatal lung injury. Moreover, a PDE4 inhibition study in a mouse

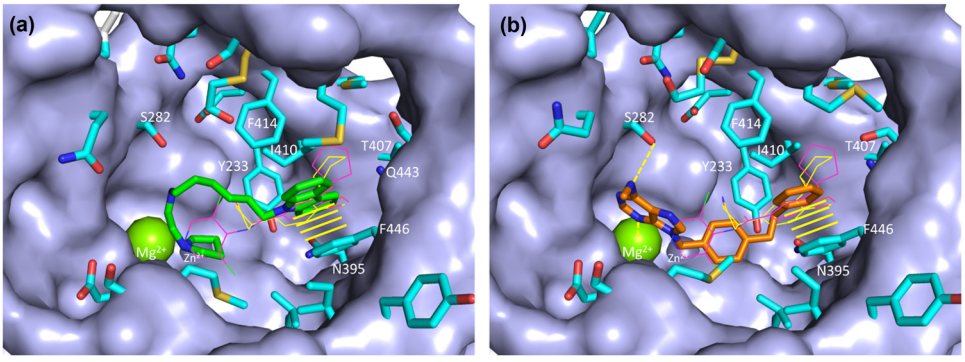


Figure 4: Docking of compounds **3** and **16** in the active site of PDE4B. Compound **3** (pose 3 in (a)) is shown with carbons in green, and compound **16** (pose 1 in (b)) with carbons in orange. The molecular surface of PDE4B is displayed in light blue, and the flexible side chains with carbons in magenta, of which the ones interacting with the ligands are labeled. Strong interactions and hydrogen bonds are indicated with dashed yellow lines and π–π interactions with transversal yellow lines. Rolipram and piclamilast are shown in lines with carbons in yellow and magenta, respectively.

Table 4: Drug-likeness predictions and physicochemical-pharmacokinetic/ADME properties of tested compounds

No	MW	Number of HBA[a]	Number of HBD[b]	Log $P_{o/w}$ (iLOGP)[c]	Log S[d]	TPSA[e]	BBB permeant[f]	GI absorption	Lipinski, ghose, veber, egan, and muegge violations	Bioavailability score	Drug-likeness model score
1	251.24	5	0	2.28	Soluble	81.35	No	High	0	0.55	−1.20
2	318.21	1	0	3.09	Poorly soluble	14.16	Yes	High	0	0.55	−1.11
3	**293.41**	**2**	**1**	**3.46**	**Moderately soluble**	**20.20**	**Yes**	**High**	**0**	**0.55**	**0.40**
4	320.45	2	0	0.00	Moderately soluble	11.41	Yes	High	0	0.55	0.09
5	461.38	2	0	0.00	Poorly soluble	11.41	No	High	0	0.55	0.40
6	288.18	0	0	3.02	Poorly soluble	4.93	Yes	High	0	0.55	−1.60
7	328.16	4	1	2.84	Moderately soluble	60.55	Yes	High	0	0.55	−0.46
8	249.26	4	0	3.00	Soluble	49.69	Yes	High	0	0.55	−0.73
9	310.35	3	1	3.04	Moderately soluble	52.49	Yes	High	0	0.55	0.09
10	259.34	2	0	3.07	Moderately soluble	31.23	Yes	High	0	0.55	−0.72
11	309.36	3	0	3.40	Moderately soluble	40.46	Yes	High	0	0.55	−0.77
12	251.32	1	0	2.68	Moderately soluble	20.31	Yes	High	0	0.55	−0.04
13	323.58	3	0	2.51	Moderately soluble	43.60	Yes	High	0	0.55	−0.55
14	323.58	3	0	2.61	Moderately soluble	43.60	Yes	High	0	0.55	−0.38
15	318.17	3	1	2.52	Moderately soluble	55.63	Yes	High	0	0.55	0.07
16	**343.42**	**3**	**1**	**3.31**	**Moderately soluble**	**55.63**	**Yes**	**High**	**0**	**0.55**	**0.37**
17	332.20	3	0	2.82	Moderately soluble	48.64	Yes	High	0	0.55	0.12
18	318.17	3	1	2.54	Moderately soluble	55.63	Yes	High	0	0.55	0.10
19	281.26	6	0	2.57	Soluble	90.58	No	High	0	0.55	−1.18
20	203.19	3	1	1.65	Soluble	59.16	Yes	High	0	0.55	−1.30
21	221.21	4	2	1.54	Soluble	71.55	Yes	High	0	0.55	−1.10

[a]Number of hydrogen bond acceptors; [b]number of hydrogen bond donors; [c]lipophilicity; [d]water solubility (SILICOS-IT [*S* = Soluble]); [e]topological polar surface area (Å^2); [f]blood–brain barrier permeant.

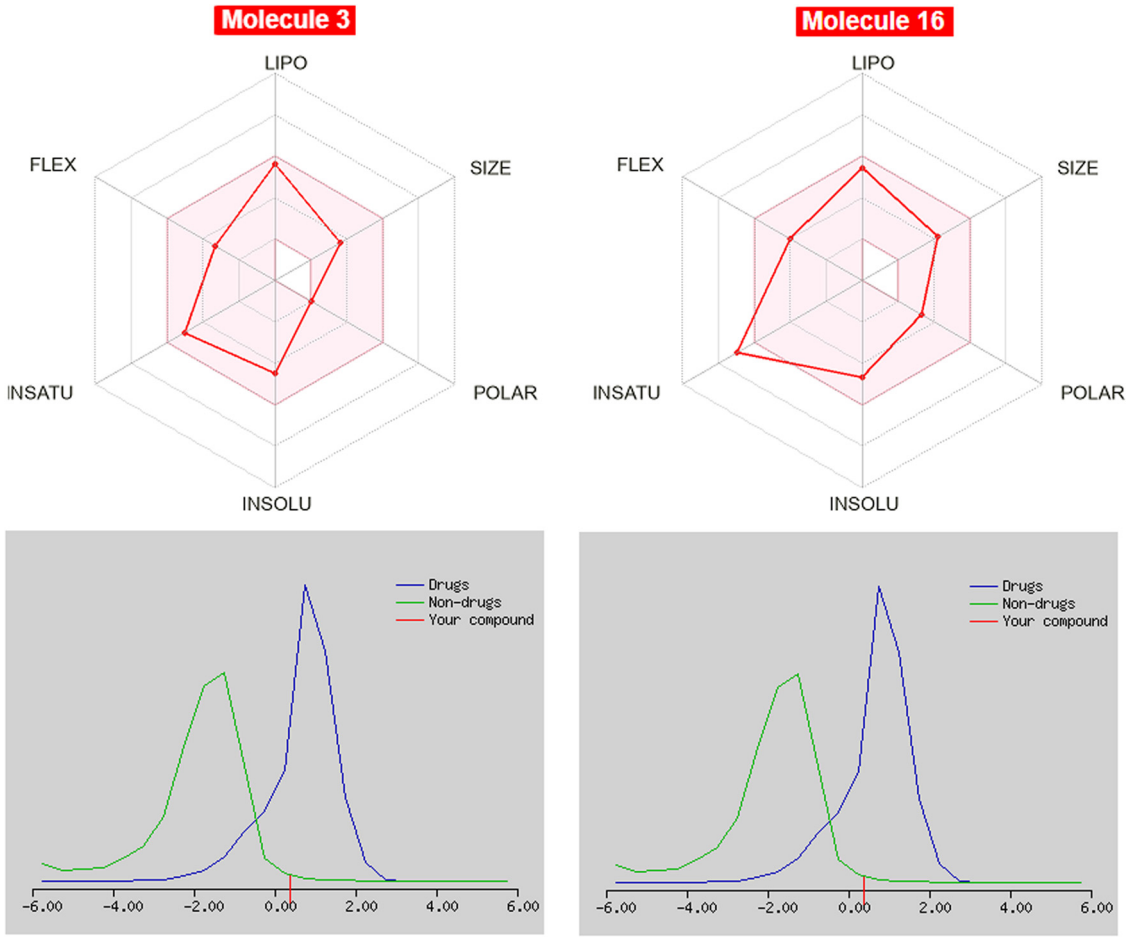


Figure 5: Bioavailability radar of the tested compounds. The pink area represents the optimal range for each property for oral bioavailability (lipophilicity [LIPO]: XLOGP3 between −0.7 and +5.0, molecular weight [SIZE]: MW between 150 and 500 g/mol, polarity [POLAR] TPSA between 20 and 130 $Å^2$, solubility (INSOLU): log S not higher than 6, saturation (INSATU): fraction of carbons in the sp3 hybridization not less than 0.25, and flexibility (FLEX): no more than nine rotatable bonds).

model of allergic asthma showed that piclamilast significantly improved lung function, airway inflammation, and goblet cell hyperplasia. Recently, the neuroprotective effect of piclamilast has been studied. It has been suggested that post-ischemia pharmacological treatment with piclamilast determines an improvement of cerebral ischemia–reperfusion injury in mice [50]. The docking study with piclamilast gave the following resulting poses (Table 1).

The docking results show a similar bonding energy for pose 2 and pose 3, equal to −9.5 and −9.4 kcal/mol, respectively. The nine poses obtained from the docking were compared to the pose of piclamilast obtained experimentally with PyMol.

3.2 Rolipram

Rolipram is a selective PDE4 inhibitor discovered and developed as a potential antidepressant drug in the early 1990s. It has been used as a prototype molecule for drug discovery and pharmaceutical research development by several companies. The study on rolipram was stopped after clinical trials showed that its therapeutic window was too narrow; it could not be used at levels high enough to be effective without causing significant gastrointestinal (GI) side effects. Nevertheless, rolipram has several activities that make it a continuing target for research [51–54]. Rolipram has been used to study whether PDE4 inhibition could be useful in autoimmune diseases, Alzheimer's disease, cognitive enhancement, spinal cord injury, and respiratory diseases such as asthma and COPD [55]. In this study, rolipram was used to better understand the exact level of exhaustiveness to be used to set the data when starting molecular docking. The data obtained with an exhaustiveness of 16 are satisfactory and comparable to those obtained with piclamilast. The results of the docking study with rolipram gave the following poses (Table 2).

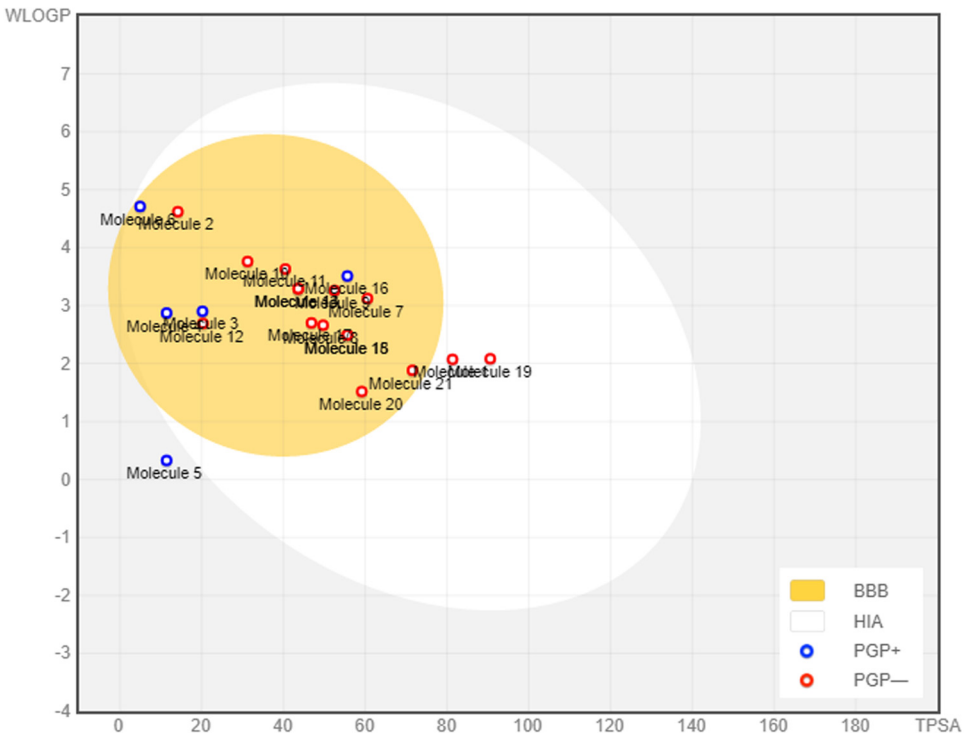


Figure 6: BOILED-Egg diagram of the selected compounds **1–21**.

The docking results show the same binding energy for pose 1 and pose 2, equal to −8.5 kcal/mol. The nine poses obtained from docking were compared with the data obtained experimentally by PyMol. The conformation that overlaps almost perfectly is pose 1 (Figure 3).

From the data obtained with different degrees of exhaustiveness (4, 8, 12, 14, 16, 20) for piclamilast and rolipram, we concluded that level 16 was the most favorable, which gave the results most similar to the experimentally obtained structure, and it was therefore chosen for the docking of all the synthetic compounds.

3.3 Other synthetic compounds

Docking studies were subsequently carried out on 21 compounds designed in our laboratories and with structural similarity with both rolipram and piclamilast, with an exhaustiveness level of 16 for all the molecules under examination [56,57]. Some of them are described in the literature for their synthesis and biological activities [58–65]. Docking studies will help us verify the possible inhibitory activity of our synthetic molecules and predict the possible interaction on the catalytic site of PDE4 *in vitro*. The different energy levels of molecules under study, along with their molecular structure, are represented in Table 3.

Both compounds **3** (pose 3) and **16** (pose 1) interact with PDE4B mainly through hydrophobic interactions (Figure 4a and b). In the docking solutions, the two compounds are deeply buried in the active site of PDE4B, which covers about 90% of their accessible surface area. They interact mostly with the same PDE4B residues and the two divalent cations. It is especially noteworthy that aromatic rings of both **3** and **16** form π–π interactions with F446, which together with Q443 bind cAMP as part of the active site. Compound **16** makes an additional hydrogen bond with S282 and a very strong bond with Mg^{2+} (Figure 4b). Compared to piclamilast and rolipram, which both form two hydrogen bonds with Q443 and otherwise bind many of the same residues and ions as compounds **3** and **16**, these new compounds interact with many more residues in the active site, probably due to the potential of their slightly larger size.

3.4 Drug-likeness predictions – physicochemical-pharmacokinetic/ ADME properties

3.4.1 Physicochemical-Pharmacokinetic properties

Drug-likeness is examined as an important part that provides the base for the molecules to be powerful oral drug

Table 5: Main pharmacokinetic descriptors studied on pkCSM predictive models

No	Absorption				Distribution		Metabolism	
	Wsol	Caco2	HIA	P-glycoprotein substrate	BBB	CNS	Cyp2d6s	CYP3A4s
1	−3.366	1.002	96.865	No	−0.581	−2.517	No	Yes
2	−6.939	1.072	96.683	Yes	0.895	−1.470	No	Yes
3	−4.182	0.977	92.785	Yes	0.857	−1.937	Yes	Yes
4	−3.913	1.380	94.520	Yes	0.928	−1.617	Yes	Yes
5	−4.419	0.971	92.957	Yes	0.982	−1.561	Yes	Yes
6	−5.837	1.177	96.071	Yes	0.712	−1.321	No	Yes
7	−4.098	1.293	94.354	No	0.003	−2.916	No	No
8	−3.293	1.375	97.884	No	−0.011	−2.881	No	No
9	−3.929	1.394	95.590	Yes	0.259	−2.067	No	Yes
10	−3.471	1.368	95.921	No	0.338	−2.159	Yes	Yes
11	−5.694	1.085	95.294	No	0.038	−1.524	No	Yes
12	−3.790	1.730	93.813	No	0.445	−1.020	Yes	Yes
13	−2.650	1.362	92.431	No	−0.345	−1.700	No	Yes
14	−2.688	1.374	92.736	No	0.336	−1.711	No	Yes
15	−2.710	1.315	91.839	No	0.162	−2.738	No	Yes
16	−2.496	0.028	91.873	Yes	0.068	−2.122	No	Yes
17	−2.680	1.326	94.734	No	0.277	−1.743	No	Yes
18	−2.672	1.312	91.268	No	0.166	−2.781	No	Yes
19	−3.844	1.174	97.077	No	−0.815	−2.660	No	Yes
20	−2.012	1.206	94.222	No	−0.169	−2.429	No	No
21	−2.888	1.173	93.615	Yes	−0.320	−3.027	No	No

WSol, water solubility in 25°C (mg/L); Caco2, permeability of Caco2 cell line (Papp in $\times 10^{-6}$ cm/s) high permeability of Caco2 would translate in values >0.90; HIA, human intestinal absorption (% absorbed); BBB, represents the BBB permeability as logBB (the logarithmic ratio of brain to plasma concentrations) LogBB > 0.3 cross the brain, while logBB < −1 is poorly distributed to the brain; CNS, blood–brain permeability-surface area product as (logPS) compounds with logPS > −2 are considered to penetrate the CNS; CYP2D6s, substrate for CYP450 isoform 2D6; CYP3A4s, substrate for CYP450 isoform 3A4.

candidates. Various rules, namely, Lipinski, Ghose, Veber, Egan, and Muegge, were considered to measure drug-likeness of the candidate compounds to find out whether they can be bioactive oral drug candidates according to some acute criterion like molecular weight, LogP, number of hydrogen bond acceptors, and donors. The number of violations of the above-disclosed rules, along with bioavailability and drug-likeness scores, are given in Table 4.

The results revealed that none of the compounds violated any rule, and their bioavailability score was around 0.55. All the tested molecules could pass the blood–brain barrier (BBB) except compounds **1**, **5**, and **19**. All compounds exhibited moderate to good drug-likeness scores ranging from −1.60 to 0.40. The bioavailability radar of best-predicted compounds is displayed in Figure 5. Compounds **3** and **16** appeared to be the most promising in the *in silico* predictions, with a drug-likeness score of 0.40 and 0.37, respectively, without any rule violation.

Moreover, all the tested compounds displayed high GI absorption, and most of them are P-gp (p-glycoprotein) non-inhibitors. The predictions for the passive BBB permeation, HIA (human GI absorption), and P-gp substrates are displayed together in the BOILED-Egg diagram, as shown in Figure 6.

3.4.2 ADME properties

The early evaluation of ADMET properties of drug candidates plays an important role in the research and development of new drugs. Taking into account that the existing methods for evaluating ADME-Tox properties are expensive and time-consuming and usually require extensive animal testing the computer modeling techniques for ADME-Tox prediction are more preferable method in early drug discovery. Thus, “drug-like” molecules were evaluated *in silico* for their ADME properties (Table 5) in order to rapidly screen multiple properties [40]. Compounds that have been predicted to exhibit a high BBB, low water solubility, and poor Caco2-permeability were excluded from potential hits. The server pkCSM [45] was used for this purpose. pkCSM relies on graph-based signatures. These encode distance patterns between atoms to represent the small molecule and to train predictive models.

4 Conclusions

In order to identify new molecules to be investigated in the fight against COVID-19, a docking study, through the AutoDock Vina program, was carried out. The possible interactions of 21 in-house compounds with the catalytic site of the A chain of PDE4B were investigated, identifying the poses of the molecules with high similarity to piclamilast and rolipram in the most favorable spatial conformation of interaction with the catalytic site of PDE4B. These compounds were inserted into the pocket of the catalytic site of the PDE4B, and in many cases, they overlapped piclamilast almost completely. The obtained results indicated that the designed compounds might represent promising ligands for PDE4B receptors, and therefore, they deserve further in-depth *in vitro* studies. Moreover, drug-likeness was carried out on these compounds. The most interesting compounds in the *in silico* predictions were **3** and **16,** showing drug-likeness scores of 0.40 and 0.37, respectively, without any rule violation.

5 Future perspectives

This interesting study requires further investigations of the selected compounds in terms of *in vitro* cytotoxicity and *in vivo* studies. The promising *in silico* and ADME results obtained urge us to proceed with inhibition assays, which will be carried out on isolated PDE4B enzymes in the not-too-distant future. We are confident that these results may lead to recognizing molecules active and selective as PDE4B inhibitors to be developed as new agents active at the pulmonary level for COVID-19 patients.

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Conflict of interest: Authors state no conflict of interest.

Data availability statement: The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

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