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In silico study of the admet properties of potential inhibitors of new Delhi methalo-ß-lactamase-1 (NDM-1)

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ABSTRACT
Both NDM-1 enzyme and its variants show multi-resistance to different antibiotics for the treatment of infectious diseases. The present work shows an in-silico study of some ADMET properties of potential inhibitors of New Delhi Metallo-β-lactamase-1 enzyme. The study started from a total of 56 compounds reported in the literature and the reference drugs meropenem and imipinem, which were drawn with the MedChemDesigner 5.5 program and from which the SMILES were obtained. Taking into account some values obtained with the online platform ADMETlab 2.0, of absorption, distribution, metabolism, excretion and toxicity, a filtering was performed, from which 22 compounds were generated, finding that the molecules with the best oral bioavailability and toxicity profile were M3, M4, M10.2, M17, M18, M23, M29 and M37.

Keywords: NDM-1; SwissADME ; Open Babel; ADMETlab 2.0.

1 INTRODUCTION

The treatment of infectious diseases is currently a challenge for humanity, due to the latent risk of a pandemic worldwide due to the emergence of drug-resistant bacteria, which would put the public health systems of the various countries in crisis (Haque, et al., 2022). This is largely due to the indiscriminate use of antibiotics (in some countries) which has caused microorganisms in their processes of mutation and therefore evolution, have created resistance mechanisms among which is more frequently the production of enzymes ß-lactamases that hydrolyze the ß-lactam ring of antibiotics for conventional use, inhibiting its action as bactericidal or bacteriostatic (Suárez et al., 2009; Yacoby et al., 2007; Cruz et al., 2010; Yang et al., 2015; Liu et al., 2015). When patients have infections with resistant bacteria, broad-spectrum antibiotics such as carbapenems are used as a last resort (Ariza et al., 2013; Deshpande et al., 2010), the drawback is that these drugs are not able to exert their therapeutic action, due to the appearance of carbapenemase-type enzymes such as NDM (New Delhi Metallobetalactamase) in microorganisms that specialize in the inactivation of drugs, causing the blockage, in some way, of their pharmacological function (Morales et al., 2014). Because of this, efforts have been focused on finding new inhibitors that circumvent these defense mechanisms and

that show a good profile of oral bioavailability and toxicity (Alcaide et al., 2004; Haque, et al., 2022; Valencia., 2014).

In recent years, approximately 90% of failures in pharmacological developments were due to poor pharmacokinetic profiles, lack of clinical efficacy (40-50%), uncontrolled toxicity (30%) and pharmacological properties outside the expected values (10-15%), however, the main problem may arise in the failures in the developments already advanced in the study of the molecule (Dulsat J, et al, 2023). Due to this potentially daunting prospect, scientific communities are continuously working on improving the drug discovery process by evaluating ADMET properties in the early stages of development and thus increasing the success rate in the later assessment stages (Dulsat J, et al, 2023).

In-silico studies continue to be one of the forms of analysis commonly performed in drug discovery and development, in order to reduce costs and obtain results in less time (Etruri et al., 2021). Through in-silico assays, it is possible to predict values or estimates of descriptors or properties of interest, through approximations and probabilities, by modeling with values already published in the literature, and by analyzing the structure of the molecule (Etruri, et al, 2021). These data give us an idea of the bioavailability and biosafety profile in a rational way, and discarding those compounds that may show undesirable values, although it is necessary to be very intuitive in the filtering of these compounds since many molecules can have values within the optimal ranges and, however, are inactive orally, while others are active despite deviating in some of these physicochemical parameters (Xiong, et al., 2021).

Pharmacokinetic investigations have determined that there is no ideal behavior that a candidate drug molecule should have, since it is usually related to the pharmacological target (Dulsat J, et al, 2023). As an example, we can mention a drug for the treatment of occasional pain which should have a high absorption for a rapid effect and therefore a rapid metabolism, thus avoiding the accumulation of the drug, while a drug designed for a chronic disease should ideally have a long elimination half-life (t1/2) and thus reduce doses (Dulsat J, et al, 2023).

Some key parameters that describe the pharmacokinetic behavior of a drug are:

ADMET	PARAMETER
Physicochemical properties	LogP, Log S, pKa
Absorption	caco-2, HIA, HOB, Pgp
Distribution:	BBB, PPB
Metabolism	CYP 450
Elimination	CL, t1/2
Toxicity	**AMES, carcinogenicity, acute toxicity**

LogP (log octanol-water partition coefficient), log S (solubility log), pKa (negative log of acidity constant), caco-2 (human intestinal permeability), HIA (human intestinal absorption), HOB (human oral bioavailability), Pgp (permeability glycoprotein), BBB (blood-brain barrier penetration), PPB (plasma protein binding), CYP450 (inhibition of cytochromes P-450), CL (clearance), t/2 (half-life), AMES (mutagenicity).

To analyze the absorption of a compound in the human body, its values of lipophilicity, LogP, water solubility LogS, intestinal human absorption (HIA) as well as its human oral bioavailability (HOB) must be evaluated (Etruri, et al, 2021). The caco-2 cell model is used to analyze how a molecule would permeate the gastrointestinal system. HOB is associated with part of an orally administered drug that reaches the systemic circulation and site of therapeutic action. On the other hand, molecules that induce P-glycoprotein (Pgp) are substrates that can reduce the bioavailability of other drugs, while inhibitors increase bioavailability (Etruri, et al, 2021). Regarding the distribution of the possible drug, parameters such as plasma protein binding (PPB) are evaluated, as well as its blood-brain penetration barrier (BBB). BBB is related to the drug's ability to passively cross the blood-brain barrier, influencing the central nervous system (Etruri, et al, 2021). Members of the CYP superfamily, cytochromes P-450 are responsible for metabolizing 90% of currently available drugs, being responsible for several drug interactions, since, if the drug is not activated, it cannot act properly in the body (Etruri, et al, 2021).

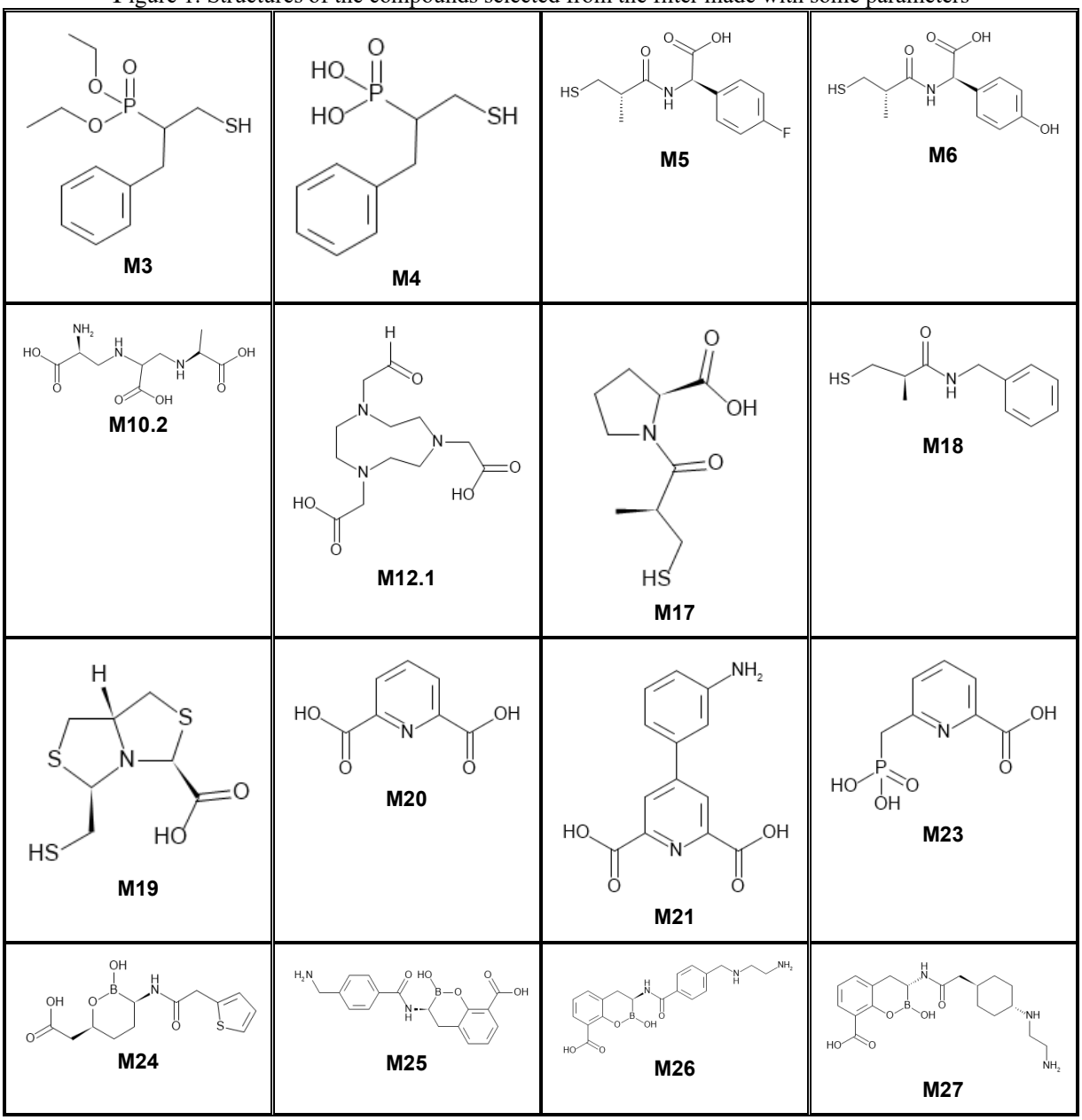
Regarding the elimination of the drug, the parameters mean life time (t1/2) and clearance (CL) are considered. The t1/2 is the time needed for the concentration of the drug in the body to reach half the dose. CL clearance is related to the ability of the kidneys or liver to remove metabolites derived from the drug compound from the bloodstream (Etruri et al, 2021). The evaluation of in-silicon toxicity is usually carried out and predicts some values, providing security to perform the subsequent steps in-vivo, so it is a good previous alternative. Parameters such as H-HT (human hepatotoxicity), DILI (drug-induced liver injury) and LD50 (lethal dose 50), among others, are usually worked on in in-silico assays (Etruri, et al, 2021).

In this research were evaluated, by in-sílico assays, a group of molecules reported in the literature with inhibitory activity of the NDM-1 enzyme, among which were derivatives of thiophene carboxylic acid, mercaptocarboxylic acid, chromones, substituted diaryl azolylthioacetamides, thiol esters of mercaptoacetic acid, ethylenediamines, cysteine derivatives, bioactive substances such as

magnolol, among others, generating a total of 56 starting compounds. The molecules were drawn with the MedChemDesigner 5.5 program, from which the SMILES nomenclature was also obtained. The computational tests, aim to evaluate the ADMET parameters of each compound and study its potential pharmacotherapeutic profile, these tests were carried out with the online platform ADMETlab 2.0, allowing a first filter, obtaining 22 compounds for subsequent analysis.

2 RESULTS AND DISCUSSION

Figure 1. Structures of the compounds selected from the filter made with some parameters



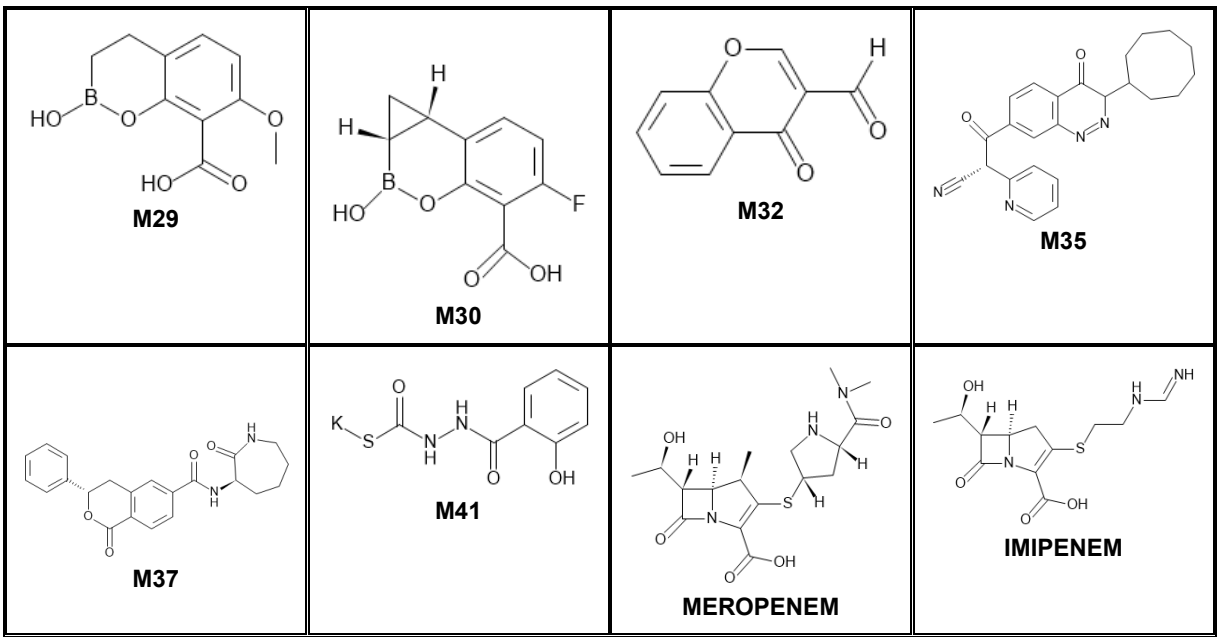


Figure 1 presents 22 compounds that obtained the best results for the ADMET parameters evaluated out of an initial total of 56, as well as two reference drugs. The compounds can be framed according to the mode of inhibition of the NDM-1 enzyme, in inhibitors that act on the zinc ions of the active site, which, being electrophilic, coordinate with the nucleophilic substituents of the NDM-1 inhibitors through ion-dipole interactions to produce the inhibitory activity of NDM-1, as well as inhibitors that act simultaneously on both the active site of NDM-1 and key catalytic amino acids. For the first case, that is, inhibitors that act on zinc ions, in Figure 1 we have the derivatives of ethylenediamine such as compound M10.2 (IC50 value of 4.0 ± 1.0 mM) and derivatives of compounds N,N',N',N'-triacetate-1,4,7-triazacyclononan (M12.1). For the second case, inhibitors that act both at the active site of NDM-1 and key catalytic amino acids, there are diversity of structures such as derivatives mercapto phosphonic acid ester (M3, IC50 1.8μM) and derivatives mercaptto phosphonic acid (M4, 2.5 μM), derivatives of acetic acid (((S)-3-mercapto-2-methylpropanamide)) (M5 (5.59 μM) and M6 (3.57 μM)), derivatives of sulfur-containing carboxylic acid compounds (M17 (6.4 μM), M18 (1.5 μM), M19 (23 ± 2 μM)), dipicholinic acid derivatives (M20 (0.52 μM), M21 (0.080 ± 0.002 μM), M23 (IC50 which may be between 0.3 to 7.2 μM)), cyclic borate derivatives (M24, M25, M26, M27, M29, M30 with estimated IC50 values around 0.01 μM), chromone derivatives (M32), natural compounds and their derivatives (M35, M37), and thioamides derivatives (M41 (IC50 estimated between 0.38 to 15.25 μM)).

Table 1. SMILES codes obtained with MedChemDesigner 5.5.

COMPOUND	SMILES
M3	O=P(OCC)(OCC)C(Cc1ccccc1)CS

M4	O=P(C(Cc1ccccc1)CS)(O)O
M5	C(=O)([C@@H](CS)C)N[C@@H](C(=O)O)c1ccc(F)cc1
M6	C(=O)([C@@H](CS)C)N[C@@H](C(=O)O)c1ccc(O)cc1
M10.2	C(=O)(C(NC[C@@H](C(=O)O)N)CNC(C(=O)O)C)O
M12.1	[H] [C] (=O) CN1CCN(CC(=O)O)CCN(CC(=O)O)CC1
M17	C(=O)(N1[C@H](C(=O)O)CCC1)[C@@H](CS)C
M18	C(=O)([C@H](CS)C)NCc1ccccc1
M19	C(=O)(O)[C@H]1N2[C@H](SC[C@@H]2CS1)CS
M20	C(=O)(c1nc(C(=O)O)ccc1)O
M21	C(=O)(c1nc(C(=O)O)cc(-c2cc(ccc2)N)c1)O
M23	C(=O)(c1nc(ccc1)CP(=O)(O)O)O
M24	C(=O)(N[C@@H]1B(O[C@H](CC(=O)O)CC1)O)Cc2sccc2
M25	C(=O)(c1ccc(cc1)CN)N[C@@H]2B(Oc3c(C(=O)O)cccc3C2)O
M26	C(=O)(c1ccc(cc1)CNCCN)N[C@@H]2B(Oc3c(C(=O)O)cccc3C2)O
M27	NCCN[C@@H]1CC[C@@H](CC(=O)N[C@@H]2Cc3cccc(C(=O)O)c3OB2O)CC1
M29	C(=O)(c1c(OC)ccc2c1OB(O)CC2)O
M30	OB1Oc2c(ccc(F)c2C(=O)O)[C@H]3C[C@@H]13
M32	[H] [C] (=O) C1C(=O)c2c(OC=1)cccc2
M35	C(#N)[C@H](C(=O)c1cc2c(C(=O)C(N=N2)C3CCCCCCC3)cc1)c4ncccc4
M37	C(=O)(c1cc2c(C(=O)O[C@H](c3ccccc3)C2)cc1)N[C@H]4C(=O)NCCCC4
M41	C(=O)(S[K])NNC(=O)c1c(O)cccc1
MEROPENEM	C(N(C)C)(=O)[C@H]1NC[C@@H](SC2=C(C(=O)O)N3C(=O)[C@H]([C@H](O)C)[C@H]3[C@H]2C)C1

IMIPENEM	C(=O)(C1=C(SCCNC=N)C[C@H]2N1C(=O)[C@@H]2[C@H](O)C)O

Source: Authors.

The compounds used (Figure 1) are a compilation of possible inhibitors of the NDM-1 enzyme, which have shown action against the pharmacological target alone or in combinations with some antibiotics such as meropenem, generating a synergistic antibacterial action (Shen et al., 2013). The SMILES codes (simplified online molecular data entry system), see Table 1, were obtained through the MedChem Designer 5.5. software, these codes are a chemical notation that allows the representation of chemical structures for use in computational analysis (Reyes et al., 2020); for this, five rules are followed that detail the representation of atoms, relationships, simple chains, chain trees and ring structure (Cardenas et al., 2019). All in-silico predictions of ADMET properties were carried out with the same SMILES code, both for the molecules under study, and for the carbapenems Meropenem and Imipenem, which are the reference compounds to make possible comparisons of the calculated properties.

Table 2. In-silico values of the physicochemical properties obtained with ADMETlab 2.0.

COMPOUND	Molecular weight	logP	logS	logD	TPSA	Nha	nHD	nRing	Nos
M3	288.09	2.43	-2.328	1.471	35.53	3	0	1	8
M4	232.03	0.826	-1.224	0.5	54.37	3	1	1	4
M5	257.05	1.789	-2.006	1.34	66.4	4	2	1	6
M6	255.06	1.038	-2.129	0.912	86.63	5	3	1	6
M10.2	263.11	-4.913	-2.056	-0.665	161.98	9	7	0	9
M12.1	287.15	-3.745	-1.177	-0.122	101.39	8	2	1	6
M17	217.08	0.21	-0.019	-0.254	57.61	4	1	1	4
M18	209.09	1.952	-1.541	1.955	29.1	2	1	1	5
M19	237.0	0.599	-1.132	0.569	40.54	3	1	2	2
M20	167.02	0.475	-1.439	1.252	87.49	5	2	1	2

M21	258.06	1.388	-2.818	0.592	113.51	6	4	2	3
M23	217.01	-0.487	-0.236	-0.452	104.56	6	2	1	3
M24	297.08	-0.286	0.086	0.346	95.86	6	3	2	6
M25	340.12	0.318	-2.355	0.022	121.88	7	5	3	5
M26	383.17	-1.221	-2.359	-0.129	133.91	8	6	3	8
M27	389.21	-1.259	-2.008	0.086	133.91	8	6	3	8
M29	222.07	0.891	-1.997	0.428	75.99	5	2	2	2
M30	222.05	0.928	-2.307	0.61	66.76	4	2	3	1
M32	174.03	1.838	-3.175	1.838	47.28	3	0	2	1
M35	400.19	5.876	-8.387	3.964	95.54	6	0	4	4
M37	378.16	3.407	-4.37	2.936	84.5	6	2	4	4
M41	249.98	1.727	-2.142	1.203	78.43	5	3	1	5
MEROPENEM	383.15	-1.08	-1.295	-0.065	110.18	8	3	3	6
IMIPINEM	299.09	-0.223	-1.429	0.0	113.72	7	4	2	7

Source: Authors.

The in-silico determination of physicochemical parameters of the compounds (Table 2), are shown within acceptable values, that is, molecular weights, nHA (H-bond acceptors) and nHD (H-bond donors). The ranges of LogP and LogS, related to the lipophilicity of the drug, were within the accepted ranges which could lead to infer an optimal oral bioavailability, indicating that they could be developed for eventual oral administration. It should be noted that for molecules M10.2 and M12.1, they have the lowest LogP values, that is, they are highly hydrophilic, which would hinder their transit through the cell membrane, a case that is equally evident in the drugs meropenem and imipinem and M35, presents the highest value, which would perhaps hinder its distribution in intracellular or aqueous media. Both the values of TPSA (total topological surface area) (optimal between 0 and 140) and nRot

(number of rotable bonds) (optimal from 0 to 10), related to Veber's rule (Khan, et al, 2021), are within appropriate ranges, except for the compound M10.2 that would not meet it. It has been observed that TPSA and nRot, can help optimize filters among compounds that could be more active orally (Khan, et al, 2021).

Table 3. In-silico values of Drug-likeness properties obtained with ADMETlab 2.0

COMPOUND	IS	NP score	Lipinski's rule	Pfizer Rule	GSK Rule	Golden Triangle	Chelator Rule
M3	0.584	-0.119	Approved	Approved	Approved	Approved	0 alerts
M4	0.615	-0.061	Approved	Approved	Approved	Approved	0 alerts
M5	0.713	-0.645	Approved	Approved	Approved	Approved	0 alerts
M6	0.605	0.091	Approved	Approved	Approved	Approved	0 alerts
M10.2	0.264	0.392	Approved	Approved	Approved	Approved	0 alerts
M12.1	0.556	-0.299	Approved	Approved	Approved	Approved	1 alerts
M17	0.682	0.113	Approved	Approved	Approved	Approved	0 alerts
M18	0.728	-0.754	Approved	Approved	Approved	Approved	0 alerts
M19	0.696	0.295	Approved	Approved	Approved	Approved	0 alerts
M20	0.668	-0.438	Approved	Approved	Approved	Rejected	1 alerts
M21	0.722	-0.357	Approved	Approved	Approved	Approved	1 alerts
M23	0.719	-0.657	Approved	Approved	Approved	Approved	1 alerts
M24	0.686	-0.788	Approved	Approved	Approved	Approved	0 alerts
M25	0.605	-0.193	Approved	Approved	Approved	Approved	0 alerts
M26	0.34	-0.369	Approved	Approved	Approved	Approved	0 alerts
M27	0.424	-0.253	Approved	Approved	Approved	Approved	0 alerts
M29	0.724	0.42	Approved	Approved	Approved	Approved	0 alerts
M30	0.701	0.017	Approved	Approved	Approved	Approved	0 alerts

M32	0.617	0.184	Approved	Approved	Approved	Rejected	0 alerts
M35	0.626	-0.732	Approved	Approved	Rejected	Approved	0 alerts
M37	0.804	0.003	Approved	Approved	Approved	Approved	0 alerts
M41	0.503	-0.927	Approved	Approved	Approved	Approved	0 alerts
MEROPENEM	0.559	0.782	Approved	Approved	Approved	Approved	0 alerts
IMIPENEM	0.224	0.891	Approved	Approved	Approved	Approved	0 alerts

Source: Authors.

Table 3 shows the in-silico values of the pharmacological properties obtained with ADMETlab 2.0, various parameters were calculated, such as the quantitative estimation of drug-likeness (QED), the NP score (similarity score with a natural product), the Lipinski rule, the Pfizer rule, the GSK rule, the Golden Triangle rule and the Chelator rule. The QED is a qualitative indicator of drug similarity that was introduced in 2012, it is based on a multicriteria optimization based on physicochemical and structural properties, where the quality of a compound is quantified by applying the concept of how "desired" it is (Serer, 2015; Haque et al., 2023). The range of values goes from zero (all unfavorable properties) to one (all favorable properties) (Serer, 2015). Taking into account the values obtained for QED, it can be evidenced that they are in a range between 0.264 for M10.2 and 0.804 for M37, which indicates that all the molecules evaluated have favorable drug-likeness properties, compared to the reference antibiotics used (Meropenem QED = 0.559, Imipenem QED = 0.224). As for the NP values, these are in a range between -5 and 5, if the value obtained is positive, there is a probability that the molecule analyzed has a greater similarity with a natural product (Haque et al., 2023). The compounds under study presented values between -0.927 for M41 and 0.42 for M29, which allows us to infer that they are acceptable data for this parameter compared to the reference drugs (Meropenem NP = 0.782, Imipenem NP = 0.891). For the Lipinski Rule, Pfizer Rule and GSK Rule, all possible inhibitors were accepted, except M35 which was rejected in the GSK Rule. Compounds that satisfy the Golden Triangle rule may have a more favorable ADMET profile (Haque et al., 2023), 20 of the compounds meet this rule, while M20 and M32 were rejected. The molecules M12.1, M20, M21, M23, present an alert as possible chelating agents, which suggests that these compounds can generate reactions with the metals present in the catalytic site of the enzyme, possibly causing a favorable aspect for its action as bactericidal or bacteriostatic, since these compounds have resistance to biotransformations before

binding to the metal, ability to form non-toxic or low-toxicity compounds from toxic metals; The other substances did not present any alert for this property.

Table 4. In-silico values of the absorption parameters obtained with ADMETlab 2.0.

COMPOUND	HIA	Caco-2 Permeability	MDCK Permeability	P-gp inhibitor	Substrate P-gp	F20%	F30%
M3	0.006	-4-476	2E-05	0.07	0.0	0.048	0.052
M4	0.449	-5.256	6E-06	0.001	0.0	0.992	0.995
M5	0.012	-5.277	1e-05	0.0	0.013	0.002	0.006
M6	0.017	-5.819	5E-06	0.0	0.014	0.722	0.011
M10.2	0.165	-6.612	0.000965	0.0	0.14	0.001	0.006
M12.1	0.999	-5.962	1.35E-04	0.0	0.007	0.714	0.963
M17	0.157	-5.928	2.2e-05	0.001	0.025	0.089	0.025
M18	0.013	-4.702	1.2E-05	0.0	0.001	0.472	0.041
M19	0.42	-6.049	3E-06	0.0	0.005	0.027	0.037
M20	0.007	-5.674	6E-06	0.0	0.001	0.011	0.394
M21	0.006	-5.758	5E-06	0.0	0.302	0.002	0.983
M23	0.216	-5.794	1e-05	0.0	0.001	0.689	0.995
M24	0.884	-5.783	0.000156	0.0	0.999	0.999	1
M25	0.936	-6.048	6E-06	0.001	0.999	0.997	0.999
M26	0.949	-6.144	4E-06	0.001	1	0.999	1
M27	0.852	-6.326	6E-06	0.001	1	0.994	0.998
M29	0.56	-5.634	1.4e-05	0.001	0.976	0.974	0.986
M30	0.649	-5.778	1.3e-05	0.001	0.994	0.958	0.992
M32	0.011	-4.513	1.7E-05	0.0	0.593	0.004	0.359

M35	0.011	-4.727	3E-05	0.577	0.001	0.805	0.044
M37	0.009	-4.907	2.7E-05	0.615	0.529	0.681	0.991
M41	0.011	-4.717	1.6E-05	0.0	0.001	0.008	0.255
MEROPENEM	0.982	-6.182	5E-06	0.001	0.754	0.652	0.991
IMIPENEM	0.996	-6.125	3E-06	0.0	0.877	0.985	0.997

Source: Authors.

In Table 4, for the values of HIA (human intestinal absorption), it is established that 14 of the molecules can be absorbed passively in the intestine, finding data between the range of 0.6% and 44.9%, while M12.1, M19, M24, M25, M26, M27, M29 and M30, may present difficulty in permeating the gastrointestinal system passively. The cell model of the human colon epithelial cancer cell line, known as Caco-2, is used to determine whether a substance is suitable for oral administration, predict intestinal permeability, and investigate drug efflux (Haque et al., 2023). The Caco-2 permeability is optimal when the value is greater than -5.15 logarithmic units, of the compounds analyzed, only M3, M18, M32, M35, M37 and M41, are in optimal values for this parameter, the others, do not satisfy this calculation. The reference carbapenems are also in unfavorable values for this calculation, a fact that is to be expected, since the route of administration for these drugs is intravenous or intramuscular.

The MDCK cell line, established in the 50s, has been used for the study of drug transport and permeability, for this the type II subclone is used, which under standard culture conditions form polarized monolayers and develop tight junctions similar to intestinal ones, which would allow its use as an accurate predictor of the permeability of the blood-brain barrier (Haque et al., 2023; Herrera et al., 2012). Taking into account the compounds under study, it can be established that none has low permeability for this parameter (less than 2x10-6cm / s), M3, M4, M6, M19, M20, M21, M25, M26, M27 and reference antibiotics, have a medium permeability (between 2x10-6 to 2x10-5cm / s), the remaining 13, have a high passive permeability (greater than 2x10-5cm / s).

P-glycoprotein is a transmembrane protein, which acts as a pump that actively exports drugs to the outside of the cell. The drug can act as a substrate, reducing bioavailability or inhibitor, increasing bioavailability, by interacting at some allosteric site, causing a conformational change that blocks the channel (Ruiz et al., 2002; Etruri et al., 2021). Taking into account the data obtained, the molecules that present a profile as a possible inhibitor of PGP, and that could favor their bioavailability, are M35 with a value of 57.7% and M37 with a value of 61.5%, although to consider a good union with the glycoprotein, a result above 90% is suggested (Etruri et al., 2021). On the other hand, M24, M25, M26,

M27, M29, M30, M32, M37, and carbapenems, have values between 52.9%-100% as possible substrates for PGP, which disfavor the bioavailability of these compounds. For the parameter of oral bioavailability in humans, HOB, it was found that M4, M12.1, M23, M24, M25, M26, M27, M29, M30 and the reference drugs, have high bioavailability of 20% and 30% for this calculation, the molecules M6 and M35, have a bioavailability of up to 20% and the compounds M21 and M37 have bioavailability of up to 30%, a fact that would allow these compounds to reach the systemic circulation and possibly the site of therapeutic action (Etruri et al., 2021).

Table 5. In-silico values of distribution and metabolism profiles obtained with ADMETlab 2.0.

COMPOUND	DISTRIBUTION				METABOLISM									
	PPB %	VD	BBB penetration	Fu %	CYP 1A2 Inhibitor	CYP 1A2 Substrate	CYP 2C19 Inhibitor	CYP 2C19 Substrate	CYP 2C9 Inhibitor	CYP 2C9 Substrate	CYP 2D6 Inhibitor	CYP 2D6 Substrate	CYP 3A4 Inhibitor	CYP3 A4 Substrate
M3	87.22%	1.336	0.3	8.086%	0.372	0.74	0.694	0.732	0.25	0.817	0.005	0.764	0.071	0.34
M4	54.05%	0.753	0.987	31.83%	0.02	0.087	0.026	0.059	0.028	0.591	0.003	0.177	0.005	0.17
M5	80.21%	0.203	0.027	20.01%	0.011	0.062	0.032	0.06	0.023	0.922	0.011	0.172	0.016	0.054
M6	63.48%	0.231	0.039	40.03%	0.006	0.044	0.027	0.053	0.028	0.959	0.006	0.149	0.023	0.028
M10.2	12.27%	0.714	0.223	87.90%	0.0	0.007	0.044	0.03	0.006	0.074	0.002	0.127	0.004	0.0
M12.1	26.49%	0.945	0.047	88.55%	0.003	0.029	0.05	0.058	0.006	0.704	0.004	0.457	0.002	0.01
M17	21.87%	0.376	0.295	73.31%	0.008	0.07	0.027	0.06	0.009	0.841	0.012	0.164	0.009	0.016
M18	82.85%	0.614	0.764	13.01%	0.457	0.109	0.17	0.786	0.025	0.135	0.14	0.576	0.061	0.188
M19	67.18%	0.702	0.009	35.57%	0.017	0.081	0.026	0.056	0.012	0.883	0.023	0.293	0.006	0.022
M20	41.07%	0.298	0.217	60.21%	0.036	0.051	0.023	0.033	0.007	0.055	0.028	0.051	0.015	0.013

M21	66.08%	0.328	0.047	45.67%	0.145	0.039	0.043	0.032	0.096	0.042	0.035	0.072	0.049	0.017
M23	29.36%	0.302	0.576	70.11%	0.009	0.047	0.039	0.035	0.003	0.166	0.031	0.08	0.006	0.013
M24	30.30%	0.204	0.05	68.12%	0.017	0.074	0.062	0.057	0.019	0.973	0.04	0.215	0.015	0.111
M25	43.62%	0.33	0.221	39.97%	0.194	0.044	0.147	0.045	0.284	0.112	0.063	0.161	0.042	0.076
M26	23.69%	1.161	0.027	61.64%	0.156	0.052	0.096	0.046	0.065	0.081	0.106	0.203	0.016	0.08
M27	10.67%	0.91	0.07	78.86%	0.037	0.057	0.092	0.051	0.018	0.065	0.042	0.18	0.014	0.079
M29	83.96%	0.241	0.375	10.84%	0.083	0.209	0.034	0.055	0.043	0.268	0.022	0.151	0.018	0.037
M30	86.09%	0.211	0.289	8.415%	0.046	0.172	0.028	0.059	0.079	0.805	0.013	0.11	0.017	0.096
M32	85.74%	0.846	0.32	17.57%	0.958	0.15	0.248	0.089	0.069	0.622	0.012	0.479	0.013	0.141
M35	98.43%	1.151	0.15	1.221%	0.246	0.719	0.898	0.317	0.927	0.929	0.503	0.087	0.792	0.414
M37	84.96%	0.533	0.185	5.185%	0.34	0.068	0.707	0.065	0.742	0.69	0.323	0.302	0.877	0.141
M41	53.70%	0.374	0.998	48.86%	0.491	0.081	0.408	0.06	0.339	0.931	0.021	0.141	0.041	0.128
MEROPENEM	13.51%	0.697	0.022	73.71%	0.012	0.067	0.03	0.102	0.027	0.107	0.008	0.14	0.009	0.308
IMIPINEM	18.10%	0.454	0.027	78.76%	0.025	0.073	0.036	0.062	0.029	0.585	0.007	0.147	0.011	0.097

Source: Authors.

The determined results of distribution and metabolism of the molecules are shown in Table 5. Studies have determined that drugs with high plasma protein binding (PPB) may have a lower therapeutic index. In the case of the selected compounds, it is observed that the molecule M35, presents

the highest value (98.43%). Regarding the volume of distribution (DV; optimal 0.04-20 L/kg), all molecules presented values that are within the acceptable limit range. None of the molecules showed a potential for penetration of the blood-brain barrier (BBB). However, it should be noted that the closer its value to 1, the greater the probability of penetration. In this sense, attention should be paid to the values of molecules M4 (0.987) and M41 (0.998), whose BBB values are the highest and closest to 1. The parameter %Fu (fraction not bound in plasma) is decisive in pharmacokinetic and pharmacodynamic studies, since it is important in the efficacy of the drug, because in general, only the unbound drug, that is, free, is able to interact with target proteins such as receptors, channels and enzymes, and can diffuse between plasma and tissues (Watanabe, R et al, 2018). Table 5 shows that 15 molecules have values above 20%, which is considered high. Special emphasis is placed on molecules M10.2, M12.1, M17, M23 and M27, which have %Fu values above 70%, as well as the reference drugs meropenem and imipinem. It is recommended to make more detailed predictions of %Fu, especially in molecules with values considered low, during the initial stages of drug development (Watanabe, R et al, 2018). For the review of the metabolism profile of the molecules analyzed, substrate and inhibitor values of some enzymes of the cytochrome-450 (CYP) family were determined. Within the families of CYP enzymes related to the metabolism of xenobiotics, there are mainly CYP1, CYP2, CYP3 and CYP4, with the CYP3A subfamily being the most abundant (Quiñones, et al., 2008). Cytochrome enzymes play an important role in drug metabolism; therefore, participation as a substrate or inhibitor contributes to drug action (Haque, et al., 2022). As shown in Table 5, none of the selected molecules presented values as inhibitory potential or substrate of CYP (0=no substrate or non-inhibitor, 1=substrate or inhibitor).

Table 6. In-silico values of excretion and toxicity profiles obtained with ADMETlab 2.0.

	EXCRETION		**TOXICITY**									
COMP.	**CL**	**T1/2**	**H-HT**	**DILI**	**AMES toxicity**	**Acute toxicity Rat Oral**	**FDAMDD**	**Skin Sensitization**	**Carcinogenicidad**	**Eye corrosion**	**Eye irritation**	**Respiratory toxicity**
M3	5.942	0.823	0.066	0.025	0.18	0.013	0.932	0.317	0.758	0.257	0.959	0.211
M4	4.059	0.831	0.146	0.232	0.136	0.031	0.931	0.688	0.302	0.778	0.944	0.43
M5	4.134	0.772	0.799	0.933	0.475	0.166	0.018	0.211	0.102	0.005	0.042	0.9

M6	6.963	0.899	0.481	0.857	0.323	0.034	0.005	0.263	0.075	0.006	0.097	0.909
M10.2	2.269	0.656	0.058	0.015	0.014	0.009	0.004	0.294	0.052	0.004	0.113	0.279
M12.1	4.42	0.503	0.011	0.012	0.141	0.029	0.013	0.706	0.285	0.287	0.83	0.671
M17	12.292	0.804	0.872	0.898	0.141	0.046	0.04	0.541	0.077	0.163	0.393	0.937
M18	11.302	0.87	0.156	0.149	0.895	0.288	0.028	0.444	0.065	0.005	0.209	0.615
M19	9.021	0.715	0.989	0.99	0.598	0.241	0.032	0.831	0.881	0.024	0.086	0.794
M20	1.326	0.81	0.422	0.922	0.017	0.25	0.004	0.163	0.012	0.004	0.99	0.929
M21	0.994	0.705	0.515	0.979	0.015	0.111	0.019	0.117	0.011	0.003	0.123	0.948
M23	1.318	0.792	0.541	0.947	0.015	0.002	0.851	0.329	0.036	0.008	0.981	0.635
M24	3.716	0.805	0.989	0.98	0.968	0.002	0.919	0.738	0.956	0.006	0.062	0.452
M25	2.455	0.673	0.974	0.979	0.039	0.023	0.219	0.133	0.952	0.003	0.012	0.035
M26	3.926	0.599	0.99	0.978	0.049	0.019	0.771	0.225	0.375	0.003	0.009	0.426
M27	4.344	0.54	0.985	0.887	0.209	0.015	0.935	0.569	0.937	0.003	0.012	0.753
M29	3.959	0.749	0.903	0.979	0.026	0.813	0.115	0.134	0.937	0.004	0.412	0.147
M30	1.601	0.634	0.987	0.989	0.147	0.981	0.413	0.14	0.977	0.004	0.33	0.768
M32	2.486	0.657	0.069	0.3	0.549	0.81	0.036	0.473	0.877	0.957	0.991	0.955
M35	1.43	0.06	0.946	0.972	0.887	0.348	0.966	0.22	0.069	0.003	0.016	0.983

	6											
M37	3.339	0.187	0.215	0.853	0.022	0.708	0.688	0.157	0.126	0.003	0.012	0.057
M41	9.145	0.616	0.079	0.954	0.954	0.745	0.011	0.89	0.449	0.055	0.908	0.974
MEROPENEM	2.586	0.932	0.951	0.979	0.004	0.116	0.127	0.784	0.574	0.004	0.013	0.617
IMIPENEM	2.484	0.944	0.908	0.981	0.012	0.022	0.004	0.831	0.204	0.003	0.019	0.031

Source: Authors.

Table 6 shows the predicted excretion and toxicity values for the molecules under study. In order to evaluate excretion, renal clearance (LC) and elimination half-life (T1/2) were used. CL is considered high, when values are greater than 15mg/mL/Kg, moderate when it is between 15 and 5mg/mL/Kg and low when it is less than 5mg/mL/Kg (Etruri et al., 2021; Haque et al., 2023). The data obtained for this parameter show that none of the compounds analyzed are within the high range for the elimination by the bloodstream of possible metabolites that can be formed in the liver. On the other hand, M3, M6, M17, M18, M19 and M41, present a moderate elimination, allowing the compounds to remain, possibly, a little longer in the body; the other molecules, including Meropenem and Imipenem, yielded data that could suggest a low renal elimination of the possible metabolites formed and, consequently, a longer retention time in the body, both in blood and tissues. For the measurement of T1/2, the following range is taken into account: if it is greater than 8 hours it is considered high, between 8 and 3 hours is moderate, and less than 3 hours is low (Etruri et al., 2021). For the values of T1 / 2 obtained, it is established that all molecules, including antibiotics worked, have a value for T1 / 2 below 3 hours, which allows inferring that the permanence of the compounds is not within an optimal range to be retained within the body, causing rapid elimination, possibly greater than 50% of the dose initially given. M6 has the longest half-life of all the molecules analyzed, with a value of T1/2=0.899, except for the reference drugs and M35 the shortest, with a value of T1/2=0.06. The two parameters have an inverse correlation generally, that is, the higher the clearance, the half-life of the drug will be lower, because for each volume that the organ processes, all the drug contained in that volume will be eliminated. Conversely, if the clearance is low, the half-life of the drug will be longer (Etruri et al., 2021).

For the evaluation of toxicity, calculations were made with different parameters, such as H-HT (human hepatotoxicity), DILI (drug-induced liver injury), AMES test, which is used to evaluate the mutagenic potential that a chemical compound may present, eye irritation, respiratory toxicity, among others. The values obtained of H-HT, show that the compounds M3, M4, M10.2, M12.1, M18, M32 and M41, are in a negative category, that is, it is likely that there are no problems of liver toxicity, the other compounds, including carbapenems, could present hepatotoxicity. For DILI values, a range was established between 0.012 for M10.2 and 0.99 for M19, where the compounds that would not generate possible drug-induced liver damage would be M3, M4, M10.2, M12.1, M18 and M32. For the AMES test, it was found that the molecules that may not show carcinogenic potential are: M3, M4, M10.2, M12.1, M17, M20, M21, M23, M25, M26, M27, M29, M30, M37 and the reference antibiotics, where M10.2 yielded the lowest value (0.014), for this parameter among the molecules under study. Regarding the values obtained for acute toxicity (Oral Rat), it could be established that the substances most likely to present toxicity for this category would be: M29, M30, M32, M37 and M41, on the other hand, the molecules that presented lower value and, therefore, lower risk of acute toxicity, were M23 and M24 with a value of 0.002 for both. For the carcinogenicity values, it was evidenced that the compounds that could induce malignant neoplasms will be M3, M19, M24, M25, M27, M29, M30 and M32. Regarding the respiratory toxicity parameter, the compounds that may not present affections with the respiratory system would be M3, M10.2, M25, M29 and M37.

Table 7. In-silico values of the environmental toxicity profile obtained with ADMETlab 2.0.

COMPOUND	Bioconcentration factors	IGC50	LC50FM	LC50DM
M3	1.603	3.646	4.225	7.57
M4	0.747	3.258	3.629	5.365
M5	0.634	3.063	3.672	6.084
M6	0.552	3.254	3.733	4.565
M10.2	-0.192	2.726	3.94	3.422
M12.1	-0.143	2.296	3.535	3.149
M17	0.461	2.888	3.564	3.762
M18	0.712	3.13	4.422	6.081
M19	0.387	3.459	3.808	5.163

M20	-0.035	2.367	2.652	2.834
M21	-0.006	2.473	2.682	3.598
M23	0.234	2.36	3.362	3.377
M24	0.059	2.266	2.611	2.253
M25	0.166	2.522	2.903	3.053
M26	0.125	2.508	2.758	3.336
M27	-0.056	2.53	2.384	2.989
M29	0.235	2.394	2.678	2.866
M30	0.316	2.719	3.343	3.292
M32	0.583	3.486	4.503	4.895
M35	1.797	5.391	8.861	7.04
M36	1.601	4.218	4.093	5.239
M37	0.468	4.171	5.368	5.302
M41	0.534	4.208	5.26	5.646
MEROPENEM	0.012	2.973	3.681	3.905
IMIPINEM	0.114	2.43	3.316	3.873

Source: Authors.

In Table 7, the in-silico values are presented of the environmental toxicity profile for the selected molecules, using the expression −log10[(mg/L)/(1000*MW)]. Bioconcentration factors are used to consider the potential for secondary poisoning and assess risks to human health throughout the food chain. IGC50 (concentration of a substance that inhibits 50% of the growth of the test population Tetrahymena pyriformis), IC50FM (concentration of a substance that causes the death of 50% of big-headed minnows after 96 hours) and IC50DM (concentration of a compound that causes the death of 50% of the population of Daphnia magna after 48 hours). The environmental toxicity profile of the molecules is optimal and within the acceptable range, however, the highest values would be related to the M35 molecule.

3 FINAL CONSIDERATIONS

The molecules evaluated in this study, showed favorable parameters in terms of their pharmacological values (Drug-likeness), absorption, excretion and toxicity, among which there must be a balance, so that there is no possible accumulation or deficiency of the compound and thus, can exert its pharmacological action at the site of interest. The compounds that presented better data for the descriptors under study were: M10.2, M17, M18, M19, M21, M29, M32 and M37 for Drug-likeness, M4, M23, M35 and M37 for absorption, M3, M6, M17, and M18 for excretion and finally, M3, M4, M10.2, M12.1, M23, M25, M29 and M37 for toxicity. In this way, the results found will allow to continue with the search for possible inhibitors of the NDM enzyme and its variants, which present a good profile of bioavailability and low toxicity, which can circumvent, in a better way, the different defense mechanisms that have the bacteria that are carriers of this metallocarbapenemase, since, at present, no drugs with relevant clinical effects are available.

On the other hand, it was determined that those compounds that have in their structure presence of groups such as derivatives mercapto phosphonic acid ester (M3 and M4), derivatives of ethylenediamine (M10.2), derivatives of carboxylic acid compounds containing sulfur (M17 and M18), derivatives of dipicholinic acid (M23), derivatives of cyclic borate (M29) and natural compounds and their derivatives (M37), would be potential inhibitors, because they were the molecules that presented a better balance in the parameters of Drug-likeness, absorption, excretion and toxicity in the present investigation.

Of the 22 structures of the compounds selected from the ADMET filters made, it was found that all are within the acceptable ranges. As for the LogP results, the M35 molecule presented the most positive value, which, in an eventual development, could hinder its dissolution in the gastrointestinal tract and bloodstream. Regarding the pharmacokinetic profile of distribution, it was found that the molecules M10.2, 12.1, M17, M23 and M27 have the best relationships %PPB (low) and %Fu (high), crucial in the efficacy of a drug. With the results obtained and studies of coupling and molecular dynamics that provide more information on the relationship ligands-receptors, it could be inferred that the compounds mentioned have potential to be main compounds or heads of series in the development of inhibitors of NDM-1.

AUTHORS' CONTRIBUTION

Eduvan Valencia and Wilson Olarte: Supervision and conceptualization. Eduvan Valencia, Wilson Olarte and Luisa Sastoque: revision and writing.

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