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Computer-aided drug repurposing to tackle antibiotic resistance based on topological data analysis

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ABSTRACT

The progressive emergence of antimicrobial resistance has become a global health problem in need of rapid solution. Research into new antimicrobial drugs is imperative. Drug repositioning, together with computational mathematical prediction models, could be a fast and efficient method of searching for new antibiotics. The aim of this study was to identify compounds with potential antimicrobial capacity against *Escherichia coli* from US Food and Drug Administration-approved drugs, and the similarity between known drug targets and *E. coli* proteins using a topological structure-activity data analysis model. This model has been shown to identify molecules with known antibiotic capacity, such as carbapenems and cephalosporins, as well as new molecules that could act as antimicrobials. Topological similarities were also found between *E. coli* proteins and proteins from different bacterial species such as *Mycobacterium tuberculosis*, *Pseudomonas aeruginosa* and *Salmonella* Typhimurium, which could imply that the selected molecules have a broader spectrum than expected. These molecules include antitumor drugs, antihistamines, lipid-lowering agents, hypoglycemic agents, antidepressants, nucleotides, and nucleosides, among others. The results presented in this study prove the ability of computational mathematical prediction models to predict molecules with potential antimicrobial capacity and/or possible new pharmacological targets of interest in the design of new antibiotics and in the better understanding of antimicrobial resistance.

1. Introduction

Currently, antimicrobial resistance (AMR) is one of the ten main threats for the public health worldwide [1]. According to the World Health Organization (WHO), this health problem could cause up to 10 million deaths yearly by 2050, surpassing other death causes such as cancer, cardiovascular diseases, or traffic accidents [2]. A study carried out by Murray et al. states that the global burden associated to drug-resistant infections in 2019 was of 4.95 million deaths of which 1.27 million were directly attributable to AMR [3]. In Europe, this burden is associated to 541,000 deaths, of which 133,000 were directly related to antibiotic resistance [4]. On an economic level, the World Bank calculates that, by 2050, up to 3.8% of gross domestic product could be lost due to AMR [5]. These data show that there is a great need to invest in R&D to search of new antimicrobial agents. In May 2015, the 68th session of the World Health Assembly adopted a worldwide action plan to support the urgent need to reinforce the knowledge and empiric basis of AMR via research to tackle this issue [6].

Several interesting initiatives focused on the search of alternative therapies to antibiotics have appeared recently [7]. Examples include antimicrobial peptides (AMP), phagotherapy, and antivirulence therapy. However, these strategies are not capable of completely replacing traditional antimicrobial therapies, which would remain as adjuvants in infection treatment [8–10]. This is due to drawbacks, such as disadvantages in treatment efficacy and security, a lack of studies of mechanism of action, or high production costs [11,12].

Since the beginning of the 20th century, the pharmaceutical industry has been involved in the search for and discovery of new drugs with activity against bacterial infections [13]. The design of lead compounds and computational design were techniques that contributed to reach the

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so-called Golden Age of antibiotics [14]. Unfortunately, AMR appears and progresses as soon as an antibiotic is released to market, which has resulted in a decrease in the interest of the pharmaceutical industry in investing in the R&D of these treatments [13]. Most of the antibiotics currently used to treat bacterial infections were discovered more than 30 years ago and the "new" antibiotics are usually based on previously known molecules or pre-existing antimicrobial agents [15].

The WHO established four criteria for antibiotic innovation, which include (1) new target, (2) new mechanism of action, (3) new family, and (4) absence of known cross resistance [16,17]. Of the 11 new antibiotics approved by the US Food and Drug Administration (FDA) and the European Medicines Agency (EMA), only two meet at least one of the four criteria. Being aware that a number of approved antibiotics are not good options for the treatment of deep infections or those caused by extremely resistant Gram-negative microorganisms, it is still imperative to promote the research to increase the available antimicrobial arsenal [18]. Therefore, the objective of this study is the identification of compounds with potential antimicrobial capacity from drugs already approved by the FDA through the application of a model based on topological data analysis (TDA). This model has been called structure-activity topological data analysis (SA-TDA). SA-TDA is defined as a submodel of TDA capable of identifying, from protein and drug databases, molecules with the potential to present a specific pharmacological activity.

2. Background

The *de novo* development of new antimicrobials is a slow process due to the progressive increase of unsatisfactory clinical data, such as pharmacokinetic parameters, bad stability and permeability, or lack of activity *in vivo* [19]. Historical data show that the success rate of a drug in clinical development is low and that only one fifth of the molecules studied are approved for stage 1 clinical trials [20]. These difficulties have a direct economic effect, as releasing a drug to market costs between 200 and 300 million dollars in a process that takes between 13 and 15 years. More specifically, an antibiotic can take up to 20 years in development and cost between 568 and 700 million dollars [21,22]. Additionally, among the antibiotics in development belonging to existing families, only 1 in 15 will be approved and reach patients. In the case of new antibacterial families, only 1 in 30 is successful [23]. This is mainly because the development of new antimicrobials is intrinsically limited by the design of the process. To slow down and control the appearance of antibiotic resistance, new drugs should have their use limited to extreme cases. While securing the efficacy of antibiotics contributes to combat antibiotic resistance, it also hinders the recovery of the outlay of the pharmaceutical companies that invest in the R&D of said drugs [24]. This, in turn, decreases the interest of the industry in these drugs as it struggles to find a return on investment. The decrease in benefits along with the efficacy problems of antimicrobials and the increasing development of AMR has led many pharmaceutical companies to abandon their antibiotic research lines or to bankruptcy. Consequently, all the innovation relies on small research institutes and universities [25].

In this context, drug repurposing appears a useful method to accelerate the search of new antimicrobial agents as it is based on the idea of finding new clinical opportunities for already existing drugs [26]. As these molecules have already been tested for their pharmacokinetic, toxicological, and pharmacological properties, development phases can be accelerated considerably. This translates in savings in time, costs, and risks when compared to developing a new drug *de novo* [27]. Among the candidates for drug repurposing are molecules that failed at one stage or another in the process of developing a drug for a different disease and the successful repurposing of these would allow the recovery of part of the money originally invested in their development [21]. This methodology offers an opportunity to accelerate the discovery of molecules with antimicrobial activity and to improve our knowledge of the mechanisms of bacterial resistance through the identification of pharmacological targets that have yet to be identified or studied.

Mathematical computational prediction models constitute another important tool in the discovery of new drugs, capable of characterizing structural biomolecular properties, physical properties, and chemical properties. In the current era of Big Data, where biomedical data is practically infinite, these models have become key in the success of machine learning (ML) for the design and repurposing of drugs [28]. One of the emerging techniques in the search of new antimicrobial drugs is TDA. This encompasses a series of visualization, exploration, and data analysis tools based on topology, a branch of mathematics that studies abstract notions of form and continuity [29]. It can be said that two objects are topologically equivalent if one can be transformed into the other through twisting and stretching, but not tearing, cutting, or gluing. The ability to process and analyze molecular data presented by a TDA model allows the exploration of proteins characteristics, including their 3D structure, flexibility, compressibility, and folding. This could accelerate drug repurposing by allowing the comparison of bacterial proteins to existing protein drug targets for already known drugs so that, if the similarity between the two proteins is very high, the drugs have a higher probability of interacting with bacterial proteins [30,31]. These new interactions could provide information on new drug targets and new antibacterial molecules. TDA models have the unique ability to concisely summarize structures in a way that often detects features that other methods overlook [29]. It is also capable of reducing structures to essential topological relations. In addition, topological properties do not depend on any choice of coordinate system and are not affected by continuous reshaping such as scaling or translation. This avoids certain problems related to noise and reproducibility: if two databases collect the same information about a protein structure, the representation of this information may differ slightly even if the data describes the same object (it can be rotated, moved, or viewed from a different angle as if it were another) [32]. These types of changes are not a problem for this model because it is not based on a coordinate system.

An important feature of this method is that it uses persistent homology (PH) for the comparison of bacterial proteins of interest and known target proteins of approved drugs. PH uses theoretical abstract algebra tools to detect topologically shared features in the proteins to be compared [32]. This method is useful for generating a topological fingerprint for each of the proteins to be studied. This fingerprint incorporates the identification or classification of proteins, the quantitative analysis of flexibility and stiffness, and the structural characteristics that occur throughout protein folding [33].

3. Methods

3.1. Bacteria selection

For the search for new drugs with potential antimicrobial activity through a SA-TDA-based model, the Gram-negative species *Escherichia coli* was selected as a candidate microorganism to test in the model due to the high number of proteins registered in the Protein Data Bank (PDB) and because it is classified by the WHO as a critical priority in the search for new antimicrobials [34].

3.2. Structure-activity topological data analysis

3.2.1. TDA pre-processing

Once *E. coli* was selected as a target microorganism, the mathematical prediction model was developed using TDA-persistent homology (TDA-PH) for protein analysis (Fig. 1). The procedure carried out for the elaboration of the model consisted, first, in obtaining the data of all the drugs approved by the FDA. These data were obtained from a query in the DB database, where the known target proteins of these drugs were also identified from the same data [https://go.drugbank.com/releases/5-1-9, accessed on February 25, 2022]. To do this, the database

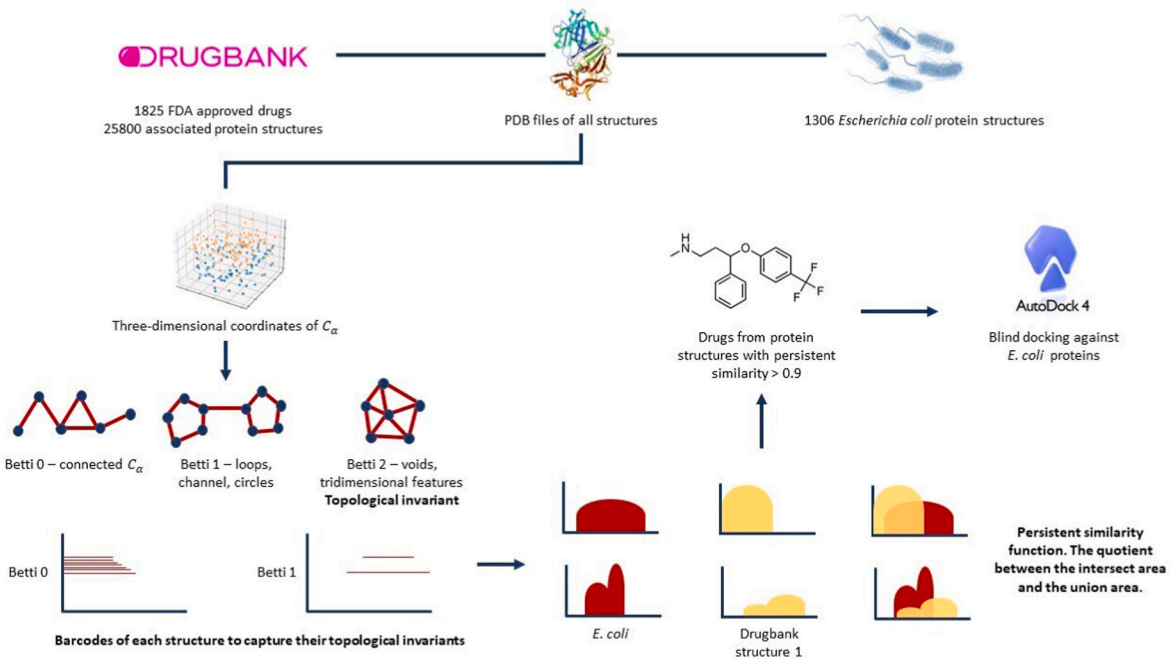

Fig. 1. Diagram of all the processes performed in the SA-TDA methodology to observe the similarity between *E. coli* proteins and drug targets of FDA-approved drugs.

was downloaded in XML format and the relevant information was extracted using the dbparser package and customized R code [35]. The name and UniProt identifier of FDA-approved drugs was obtained to map these IDs with the corresponding PDB structure using the Retrieve/ID mapping tool available in UniProt. All PDB structures that targeted FDA-approved drugs were downloaded in PDB format. The 3D structures of *E. coli* proteins were also obtained from PDB.

3.2.2. SA-TDA

This paper uses an adaptation of a TDA-based methodology that combines concepts and results from algebraic topology to compare three-dimensional protein structures [36,37]. This model, called SA-TDA, encompasses a set of tools for visualizing, exploring, and analyzing data based on the topology of TDA models [29], the application of PH for the comparison/similarity of known DNA proteins of approved drugs and bacterial proteins [32], and the combination with other computational models such as molecular docking for the identification of biologically active drug-target interactions (DTIs) from their binding energy [38–40]. All protein structures in PDB format were loaded into the R environment using the Bio3d package [41]. Then, each structure was represented in coarse grain, generating a 3D representation of the atomic coordinates of the alpha carbons of amino acids [42]. We worked with this representation because the complete structure of each protein presents a high degree of detail that could mask the general structure of the protein and, consequently, useful information. The capacity of the R environment allows these representations to be performed on proteins with a number of amino acids equal to or less than 1000. Therefore, those proteins with an amount greater than 1000 amino acids were discarded from the assay.

Taking as a "template" the arrangement of the alpha carbons of each protein, persistence diagrams were generated using the R TDAstats package, which internally uses the Ripser C++ library, a fast software optimized for Vietoris-Rips computing and persistence diagram construction [43]. If a protein did not present any of the three dimensions, a pseudo-persistence diagram was added to each dimension.

Once the persistence diagrams were generated, the similarity test was carried out. This operation was based on the principle of homology, where each query (query = *E. coli* protein) results in a comparison by similarity with all known drug targets from the DB database.

Using the information contained in the persistent diagram, we construct a set of three functions in two variables. The first function, denoted by f_1, represents the structure of the position of the individual coordinates of the alpha carbons of amino acids, the second function f_2, corresponds to the non-intersecting segments about these positions and, finally, the third function f_3 corresponds to the non-intersecting triangles constructed around these segments. These three functions are called the persistent Betti functions (PBFs) and they allow us to characterize the representation of a protein's tertiary structure.

Therefore, we computed the PBFs using PDB structures from DB. To compare the shape of both structures, one given by the PBF $\{f_i\}_{i=1}^{i=3}$ of each structure from DB, against the PBF of each query proteins $\{f_i^{query}\}_{i=1}^{i=3}$ we construct the persistent similarity measure (PSM), which is defined as

$$PSM_i = \frac{\int min(f_i(x), f_i^{query}(x))dx}{\int \max\,(f_i(x), f_i^{query}(x))dx} \text{ for } i = 0, 1, 2.. \quad (1)$$

Then, we calculate the mean of the PSMs:

$$\overline{PSM} = \frac{1}{3}(PSM_1 + PSM_2 + PSM_3) \quad (2)$$

for each protein comparison. A strict similarity threshold $\overline{PSM} \geq 0.9$ was selected to consider two protein structures as similar. This average similarity (Av. Sim) was calculated by the mean of the similarities of the primary (PSM_1 = Sim. 1), secondary (PSM_2 = Sim. 2) and tertiary (PSM_3 = Sim. 3) structures of each of the proteins present in the study.

3.2.3. Molecular docking

Once the similarity study was completed, an *in silico* coupling screening using AutoDock 4.2, a molecular coupling software developed by the Scripps Research Institute, was added to the model. This screening allowed the addition to the model of information on the binding energies presented by the DTIs resulting from the trial.

For this, the ligands were prepared obtaining the SDF format of the molecules in DB approved by the FDA. A customized R code and Open Babel v.3.0.0 were used to transform SDF to mol2 format [44–47]. The MGLTools v.1.5.7 toolkit was used to add polar hydrogens and protonation at pH 7.4. Next, the drug structures in mol2 were converted to

PDBQT format and their stereochemical properties were calculated using AutoDock 4.2 [48]. So, a virtual screening library was built using these pre-processed drugs. Drugs containing atoms other than H, C, N, O, F, Mg, P, S, Cl, Ca, Mn, Fe, Zn, Br and I were discarded for the next steps because AutoDock does not include the values of the atomic force field and is therefore unable to dock on them. Polar hydrogens were also added to *E. coli* proteins extracted from PDB, which were also transformed into PDBQT format. A grid box spanning the whole protein structure was set to perform blind docking. AutoDock was configured following the manual recommendations [49]. This screening allowed the exclusion of those DTI that presented binding energies greater than −7 kcal/mol, since the literature considers these interactions as inactive at the biological level [38–40].

3.3. Interspecies protein similarities

Since some of the pharmacological targets of the drugs approved by the FDA include proteins from bacterial species other than *E. coli,* a query was performed to find similarities between *E. coli* proteins present in the model results and proteins from different bacterial species present in the database of drug targets present in the same assay. This analysis met the same criteria as the main trial: a strict similarity threshold of 0.9 and binding energy less than −7 kcal/mol. This assay was performed to provide information on molecules with potential broad-spectrum antimicrobial activity.

4. Results

4.1. Structure-activity topological data analysis (SA-TDA)

The search of the DrugBank (DB) database resulted in 1,825 drugs being approved by the FDA (Fig. 1). These molecules were associated with 1,821 known protein targets, for which 27,839 three-dimensional structures were found in the PDB database. Computational limitations prevented the reading of 2,039 structures, so the persistence diagrams of 25,800 protein structures were generated.

In a second search in PDB, 1,306 *Escherichia coli* proteins were collected, and their persistence diagrams were generated following the same procedure as for the known target proteins. We detected that 33 of the proteins collected corresponded to essential genes for *E. coli.*

4.2. Similarity test

Once the similarity assay had been performed using TDA to compare the persistence diagrams of bacterial proteins and target proteins, 146 *E. coli* proteins were found with an average similarity greater than 90% with the known targets of 529 drugs registered by the FDA. No average similarity exceeded 98% similarity between *E. coli* proteins and targets of drugs registered by the FDA. These bacterial proteins presented 891 DTIs with the studied molecules.

Among the 529 resulting molecules, drugs already known for their antimicrobial activity were observed (Table 1). These antibiotics are found within the families of cephalosporins of different generations, carbapenems, and semisynthetic penicillins. Some of these antibiotics had interactions with the proteins 6G9F and 1GER, proteins from essential genes of the bacterial species. Experimental antibiotics such as platensimycin, β-lactamase inhibitors such as relebactam and avibactam, other antibiotic adjuvants such as adapalene, as well as anti-infective agents such as sulfacetamide and nitrofural, and antiseptics such as hexachlorophene were also found.

4.3. Molecular docking

A subsequent screening of the binding energies of the DTI found in the similarity test showed that, of these interactions, 361 presented binding energies greater than −7 kcal/mol, which corresponded to 97 bacterial proteins and 254 drugs, including drugs already established as antimicrobials (Table 1).

4.4. Interspecies protein similarities

Within the pharmacological targets of FDA-approved drugs, bacterial

Table 1
FDA-recognized antimicrobial molecules found in the SA-TDA model.

DTI (*E. coli* protein-DrugBank ID)	*E. coli* essential genes	Name	Description	Sim. 0	Sim. 1	Sim. 2	Av. Sim.	Energy (kcal/mol)
1B3N-DB08407	No	Platensimycin	Antibiotic in preclinical phase	0.999	0.962	0.909	0.957	−9.7
6G9F-DB00303	Yes	Ertapenem	Carbapenem antibiotic drug	0.998	0.981	0.916	0.965	−9.4
6G9F-DB00948	Yes	Mezlocillin	Semisynthetic ampicillin-derived acylureido penicillin	0.998	0.981	0.916	0.965	−9
6G9F-DB01329	Yes	Cefoperazone	Semisynthetic broad-spectrum third-generation cephalosporin	0.998	0.981	0.916	0.965	−9
6G9F- DB01328	Yes	Cefonicid	Second-generation cephalosporin	0.998	0.981	0.916	0.965	−8.7
3NBX-DB00210	No	Adapalene	Retinoid. Antibiotic adjuvant in the treatment of acne	0.997	0.856	0.858	0.904	−8.6
4J8L- DB01147	No	Cloxacillin	A semisynthetic antibiotic that is a chlorinated derivative of oxacillin	0.994	0.932	0.786	0.904	−8.5
6G9F-DB09050	Yes	Ceftolozane	Semisynthetic broad-spectrum fifth generation cephalosporin	0.998	0.981	0.916	0.965	−8.1
6G9F-DB01415	Yes	Ceftibuten	Third-generation cephalosporin antibiotic	0.998	0.981	0.916	0.965	−7.9
1BDH-DB01150	No	Cefprozil	Cephalosporin antibiotic	0.985	0.919	0.816	0.907	−7.8
6G9F-DB06211	Yes	Doripenem	Carbapenem antibiotic drug	0.998	0.981	0.916	0.965	−7.7
6G9F-DB01327	Yes	Cefazolin	Semisynthetic cephalosporin analog	0.998	0.981	0.916	0.965	−7.5
1GER-DB00336	Yes	Nitrofural	A topical anti-infective agent	0.999	0.984	0.906	0.963	−7.4
6G9F-DB01413	Yes	Cefepime	Fourth-generation cephalosporin antibiotic	0.998	0.981	0.916	0.965	−7.3
6G9F-DB01598	Yes	Imipenem	Semisynthetic thienamycin	0.998	0.981	0.916	0.965	−7.3
1B6R-DB01150	No	Cefprozil	Cephalosporin antibiotic	0.979	0.889	0.859	0.909	−7.2
2QZS-DB00634	No	Sulfacetamide	Anti-infective agent	0.928	0.934	0.841	0.901	−7.1
4J8L-DB00456	No	Cephalotin	Cephalosporin antibiotic	0.994	0.932	0.786	0.904	−6.9
1DPE-DB00634	No	Sulfacetamide	Anti-infective agent	0.989	0.921	0.794	0.901	−6.7
5V0I-DB12377	No	Relebactam	Diazabicyclooctane β-lactamase inhibitor	0.984	0.923	0.795	0.901	−6.6
5V0I-DB09060	No	Avibactam	Non- β-lactam β-lactamase inhibitor	0.984	0.923	0.795	0.901	−6.5
1F5V-DB00756	No	Hexachlorophene	A chlorinated bisphenol antiseptic (bacteriostatic)	0.956	0.893	0.851	0.900	−6.4
1C3B-DB00161	No	Valine	Branched-chain essential amino acid. Precursor in the penicillin biosynthetic pathway	0.999	0.929	0.788	0.905	−4.6

DTI: drug-target interaction.

proteins of bacterial species other than *E. coli* were observed with an average topological similarity greater than 90%. Both Gram-positive and Gram-negative bacteria and mycobacteria were found. Some of these are also listed by the WHO as priorities in the search for new antibacterials [34]. Table 2 shows the most important bacterial species that have appeared in the results of the similarity study along with proteins that have been shown to have a high degree of similarity with several *E. coli* proteins. Similarities were found with proteins present in the cell membrane as Penicillin-binding protein (PbP), in the periplasm as those related to the N-acetyl-anhydromuramoyl-L-alanine amidase activity and at the cytoplasm level as the malate synthase G (Table 2).

4.5. Candidates

The drugs selected by the model as molecules with potential antimicrobial capacity belong to several families. Among these, some stand out for presenting molecules with greater binding energies and similarities: antitumoral drugs, nucleotides, nucleosides, antipsychotics, antidepressants, retinoids, antimalarial agents, hypoglycemics, antihistamines, antitussives, and neurotransmitters (Table 3). Nucleotides have been the family most present in this trial, followed by antidepressants. Several molecules, such as the antipsychotic aripiprazole, the antidepressants vortioxetine and duloxetine, the cough suppressant dextromethorphan, and the antihistamine chlorpheniramine, have coincided in presenting high binding energies against the same two *E. coli* proteins, 1HTT and 1F6D. The antidepressant desvenlafaxine and the neurotransmitter serotonin also targeted the 1HTT protein, becoming the most frequent protein in this study, followed by the 1F6D protein. Among the DTIs identified were some corresponding to targets from essential genes. All nucleotides except uridine-5′-diphosphate showed one or more interactions with proteins codified by essential genes, in addition to the molecules 7-methylguanosine, pyridoxine phosphate, tosyl-D-proline, and 3-indolebutyric acid.

5. Discussion

TDA has proved to be an important tool for examining dynamic processes such as protein folding [50]. One of the advantages of TDA is that it does not require a group of inactive compounds for the model to carry out its learning process. This is very favorable considering that negative activity results are not normally published, greatly complicating access to molecular structures that are known to be inactive for the construction and validation of models [51]. This has contributed to understanding other dynamic properties of macromolecules, such as ligand binding, and to show the potential of this mathematical approach for identifying molecules of interest from their topological invariants

Table 2
Bacterial species classified by the WHO as priorities, and their proteins with high similarity to *E. coli* proteins resulting from the SA-TDA.

E. coli Protein	Similar protein	Protein name	Species (priority)	General function	Cellular location	Sim. 0	Sim. 1	Sim. 2	Av. Sim
1K75	1MV8	GDP-mannose 6-dehydrogenase	*Pseudomonas aeruginosa* (critical)	Udp-glucose 6-dehydrogenase activity	NI[a]	0.987	0.954	0.815	0.919
1BJN	1RCQ	Alanine racemase, catabolic		Pyridoxal phosphate binding	NI[a]	0.992	0.909	0.803	0.902
2RJG						0.993	0.943	0.813	0.916
4PTY						0.975	0.881	0.845	0.9
5WAN	4K2F	Acyl-homoserine lactone acylase PvdQ		N-acetyl-anhydromuramoyl-l-alanine amidase activity	Periplasm	0.957	0.931	0.819	0.902
1USG						0.967	0.909	0.853	0.909
1DC3						0.925	0.948	0.842	0.905
1BRM						0.994	0.928	0.796	0.906
6LPI	1KBZ	dTDP-4-dehydrorhamnose reductase	*Salmonella* Typhimurium (critical)	Metal ion binding	NI[a]	0.963	0.938	0.825	0.909
3AWI	2P2M	Acetyl-coenzyme A synthetase		Metal ion binding	NI[a]	0.981	0.92	0.816	0.906
1HTT	1LC7	Threonine-phosphate decarboxylase		Threonine-phosphate decarboxylase activity	NI[a]	0.988	0.914	0.804	0.902
6XGY						0.987	0.902	0.843	0.911
3TCF	1B3L	Periplasmic oligopeptide-binding protein		NI[a]	Periplasm	0.993	0.97	0.84	0.935
6IYK						0.996	0.918	0.822	0.912
3O9P						0.976	0.946	0.826	0.916
1GLG	3GBP	d-galactose-binding periplasmic protein		Metal ion binding	Periplasm	0.985	0.959	0.796	0.913
4GD3	2JJH	Probable l-lysine-epsilon aminotransferase	*Mycobacterium tuberculosis* (critical)	NI[a]	NI[a]	0.984	0.945	0.832	0.921
3UQY				NI[a]	NI[a]	0.971	0.943	0.816	0.91
2DGK				NI[a]	NI[a]	0.995	0.943	0.816	0.918
3C3J	1TED	Alpha-pyrone synthesis polyketide synthase-like Pks18		Involved in the biosynthesis of tri- and tetraketide alpha-pyrones. Pks18 catalyzes the extension of medium- and long-chain aliphatic acyl-CoA substrates by using malonyl-CoA as an extender molecule to synthesize polyketide products	NI[a]	0.979	0.929	0.801	0.903
1D8C	6DNP	Malate synthase G		Involved in glycolate utilization. Catalyzes the condensation and subsequent hydrolysis of acetyl-coenzyme A (acetyl-CoA) and glyoxylate to form malate and CoA	Cytoplasm	0.987	0.943	0.872	0.934
1BJN						0.992	0.926	0.815	0.911
1DS7	1KQD	Oxygen-insensitive NAD (P)H nitroreductase	*Enterobacter cloacae* (critical)	Oxidoreductase activity	NI[a]	0.994	0.944	0.763	0.901
1BDH	5OJ1	Penicillin-binding protein 2x	*Streptococcus pneumoniae* (medium)	Penicillin binding	Cell membrane	0.985	0.889	0.832	0.902
1B6R						0.973	0.935	0.829	0.913
1BDH	1RP5	Penicillin-binding protein 2x		Penicillin binding	Cell membrane	0.985	0.919	0.816	0.907
1B6R						0.979	0.889	0.859	0.908
2J5T	1LXK	Hyaluronate lyase		Hyaluronate lyase activity	Secreted	0.991	0.912	0.805	0.903
1BRM	1PR3	Aspartate-semialdehyde dehydrogenase	*Haemophilus influenzae* (medium)	Nadp binding	NI[a]	0.999	0.962	0.813	0.925
5WAN					NI[a]	0.956	0.95	0.816	0.907

[a] NI: not identified.

Table 3
Molecules registered in the DB database with higher binding energies.

Molecule	Drug class	*E. coli* target	General function	Cellular location	Energies (kcal/mol)
Enzastaurin	Antitumor	2WIU	Bacterial persistence mechanism regulator	Cytosol; DNA-complex	−10.8
Ezetimibe	Lipid-lowering	2DQ6	Aminepeptidase N activity	Plasma membrane	−9.9
Nicotinamide adenine Dinucleotide phosphate	Nucleotide	5AED	Glucoside-hydrolase activity	NI[b]	−9.9
		1OG6	Aldo-keto reductase activity	Cytosol	−9.8
		1BSV	GDP-fucose synthetase activity	Cytoplasm	−9.7
		1L5J	Aconitase B activity	Cytoplasm	−9.2
		2XHY	Hydrolase activity	Cytosol	−9.2
		3DMY	NI[b]	Cytosol; integral component of membrane; succinate-CoA ligase complex	−9
		1BRM[a]	Aspartate beta-semialdehyde dehydrogenase activity	Cytosol	−8.4
		5WAN[a]	Oxidoreductase activity	NI[b]	−8.4
Uridine-5′-diphosphate	Nucleotide	1AA6	Formate dehydrogenase H activity	Cytosol; plasma membrane respiratory chain complex I	−9.5
		1A9Y	UDP-galactose 4-epimerase activity	Cytoplasm; cytosol	−8.8
Histidyl-adenosine monophosphate	Nucleotide	1Q12[a]	Transport protein	ATP-binding cassette (ABC) transporter complex; extrinsic component of cytoplasmic side of plasma membrane; maltose transport complex	−9.3
		1C3B[a]	AmpC beta-lactamase activity	Outer membrane-bounded periplasmic space	−9.1
		6XGY[a]	Lipid transport function	Cytosol; protein-containing complex	−7.7
Cytidine-5′-triphosphate	Nucleotide	1H3M[a]	4-diphosphocytidyl-2C-methyl-D-erythritol synthetase	Cytosol	−8.8
2′-deoxyguanosine-5′-monophosphate	Nucleotide	1AIQ[a]	Thimidylate synthase activity	Cytosol	−8.4
Deoxyuridine monophosphate	Nucleotide	1AIQ[a]	Thimidylate synthase activity	Cytosol	−8.2
Thymidine monophosphate	Nucleotide	1CY0[a]	DNA topoisomerase type I activity	Chromosome; cytosol	−7.1
7-Methylguanosine	Nucleoside	1Q12[a]	Transport protein	ATP-binding cassette (ABC) transporter complex; extrinsic component of cytoplasmic side of plasma membrane; maltose transport complex	−8.1
Aripiprazole	Antipsychotic	1F6D	UDP-N-acetylglucosamine 2-epimerase activity	Cytosol	−9.6
		1HTT	Histidyl-tRNA synthetase activity	Cytosol	−8.6
Vortioxetine	Antidepressant	1HTT	Histidyl-tRNA synthetase activity	Cytosol	−9.1
		1F6D	UDP-N-acetylglucosamine 2-epimerase activity	Cytosol	−8
Desvenlafaxine	Antidepressant	1HTT	Histidyl-tRNA synthetase activity	Cytosol	−8.1
Duloxetine	Antidepressant	1HTT	Histidyl-tRNA synthetase activity	Cytosol	−8.3
		1F6D	UDP-N-acetylglucosamine 2-epimerase activity	Cytosol	−7.2
Tamibarotene	Retinoid	3NBX	Regulatory ATPase activity	Cytoplasm; cytosol	−8
Artenimol	Antimalarial agent	2XHY	Hydrolase activity	Cytosol	−8.5
		1K75	L-histidinol dehydrogenase activity	Cytoplasm; cytosol	−7.9
		1YNF	N-succinylarginine dihydrolase activity	NI[b]	−7.8
		4GD3	Oxidoreductase activity and electron transport	[Ni–Fe] hydrogenase complex; integral component of plasma membrane	−7.2
		3UQY	Oxidoreductase activity	[Ni–Fe] hydrogenase complex; outer membrane-bounded periplasmic space; ferredoxin hydrogenase complex; integral component of membrane; intrinsic component of periplasmic side of plasma membrane	−7
Acarbose	Hypoglycemic	1DPE	Dipeptide-binding protein	ATP-binding cassette (ABC) transporter complex; outer membrane-bounded periplasmic space	−8.5
		3UQY	Oxidoreductase activity	[Ni–Fe] hydrogenase complex; outer membrane-bounded periplasmic space; ferredoxin hydrogenase complex; integral component of membrane; intrinsic component of periplasmic side of plasma membrane	−7
Pyridoxine phosphate	Pyridoxine	1HO4[a]	Pyridoxine 5′-phosphate synthase	Cytosol	−8.2
Dextromethorphan	Cough suppressant	1F6D	UDP-N-acetylglucosamine 2-epimerase activity	Cytosol	−8

(*continued on next page*)

Table 3 (*continued*)

Molecule	Drug class	*E. coli* target	General function	Cellular location	Energies (kcal/mol)
		1HTT	Histidyl-tRNA synthetase activity	Cytosol	−7.6
Serotonin	Neurotransmitter	1HTT	Histidyl-tRNA synthetase activity	Cytosol	−7.7
Deoxycholic acid	Cytolytic agent	3FSL[a]	Tyrosine aminotransferase activity	Cytoplasm; cytosol	−7.7
		1DEA	Glucosamine 6-phosphate deaminase activity	Cytoplasm; cytosol	−7.5
Tosyl-d-proline	Toluen	1AIQ[a]	Thimidylate synthase activity	Cytosol	−7,5
Chlorpheniramine	Antihistamine	1F6D	UDP-N-acetylglucosamine 2-epimerase activity	Cytosol	−7.4
		1HHT	Histidyl-tRNA synthetase activity	Cytosol	−7.2
3-Indolebutyric acid	Heterocyclic compound, fused ring	3FSL[a]	Tyrosine aminotransferase activity	Cytoplasm; cytosol	−7,3

[a] *E. coli* proteins codified by essential genes.
[b] NI: not identified.

[52]. Being a newer tool compared to other techniques, there are few studies in the literature where this model is applied in terms of the search for new antimicrobials. In 2021 Hernández-Ochoa et al. applied a TDA strategy to verify the possible antimicrobial activity of 55 new compounds against *Helicobacter pylori* [53]. In this study, interactions were discovered with the enzyme glucose-6-phosphate dehydrogenase. After the results obtained, the compounds represent new candidates for promising drugs against this infection. However, it is important to remember that we are talking about *in silico* methods and results. Such methodologies have two key points to consider. The first point is the importance of having any information resulting from these tests checked and revised by the corresponding *in vitro* tests to confirm that the computational models meet their predictive capacity. By performing this order of testing (*in silico* -> *in vitro*), the second key point is to improve the prediction models. These improvements can be carried out by applying the model to new molecule databases to obtain more predictions, which can be experimentally validated to adjust the model. One of the databases that could support the specificity of the model could be DRESIS. DRESIS is a comprehensive database which contains drug resistance information [54]. It can systematically provide all existing types of molecular mechanisms underlying drug resistance and describe the clinically/experimentally verified resistance data for the largest number of drugs. The results presented in this work are the result of a first trial with the SA-TDA-based model, which does not include the DRESIS database. This database could improve the information provided by the topological data of the proteins and drugs present in the results. It would be interesting to add to the model databases related to resistance mechanisms and drugs associated with them to try to create a version of the SA-TDA model more specific to the search for new antimicrobials effective against resistant microorganisms.

This work aims to identify molecules with potential antimicrobial capacity through an SA-TDA model, which uses the methodology of TDA and PH to identify molecules with potential antimicrobial activity from the topological similarities between proteins registered as pharmacological targets of FDA-approved drugs and *E. coli* proteins. The combination of models that use TDA together with ML tools results in a methodology with impressive predictive accuracy, preserving the performance and interpretability of the TDA, compared to models that use a single approach [55,56]. The presence of carbapenems, cephalosporins of different generations, and semisynthetic penicillins, along with other agents involved as antibiotic, disinfectant, or antiseptic adjuvants among the drugs selected by the model, confirms its predictive capacity to find molecules with antimicrobial activity (Table 1). In addition, carbapenems and some cephalosporins present in the assay have DTI with proteins from essential genes. These results confirm the model's ability to detect molecules with antibiotic capacity. However, the possibility of detecting molecules with antivirulent properties using the same model should not be ruled out, since this class of drugs act against virulence factors, which is an advantage as the growth and selective pressure of resistant bacteria is not influenced [57]. It is important to note that some of the molecules present in the table, such as cephalotin or avibactam, have binding energies below the corresponding criterion with the literature consulted to define a DTI as having biological activity [38–40]. However, the properties of these two drugs have been broadly defined in the literature, being molecules that are used in current therapeutics [58–61]. It is possible that the SA-TDA model does not contemplate other parameters of interest for the search for potential molecules with the desired pharmacological activity. Therefore, it would be interesting to carry out more analysis on those molecules that are in these margins or to combine other computational approaches that allow the screening of other data of interest on DTI to expand the search for molecules with potential antimicrobial activity.

Regarding the shape analysis of *E. coli* proteins with other bacterial species, high degrees of similarity have been observed in certain proteins of *M. tuberculosis* and *S. pneumoniae.* Proteomic similarities between *E. coli* and *M. tuberculosis* have been obtained in several studies [62–64]. However, despite coinciding in some mechanisms of infection, no studies have been found that speak to the structural similarities of *E. coli* and *S. pneumoniae* proteins [65,66]. Most of the bacteria present in Table 2 belong to the class of Gammaproteobacteria. Among them we find *S.* Typhimurium, which belongs to the *Enterobacterales* order, like *E. coli.* The model's ability to find similarities between species could provide information on the potential spectra of active molecules found in future similarity tests. These spectra could range from antibiotics for *Enterobacterales* or other Gram-negative microorganisms to broad-spectrum antimicrobials that also exhibit activity against Gram-positive and mycobacteria. It is important to highlight that one of the main challenges in drug discovery is the search for active molecules that cross the cell wall of Gram-negative bacteria, due to their lipid bilayer membranes, porins and efflux pumps [16,67]. As can be seen in Table 2, our model presents DTI with bacterial targets found in the cytoplasm, so it would be interesting to perform in *vitro* tests with these molecules to check if this interaction occurs, since antimicrobial molecules capable of crossing the cell wall of Gram-negative bacteria could be found.

Regarding the drugs present in the results, we observed a wide variety of molecules with different structures (Figs. 2–5). Enzastaurin, an investigational antitumor drug for the treatment of several cancers, including breast cancer [68,69], has a high binding energy against one of the *E. coli* proteins (Table 3). 2WIU, known as HipA, is a protein kinase that, when bound to HipB DNA, forms a complex that regulates the persistence of *E. coli*, inducing a latency period during which it is

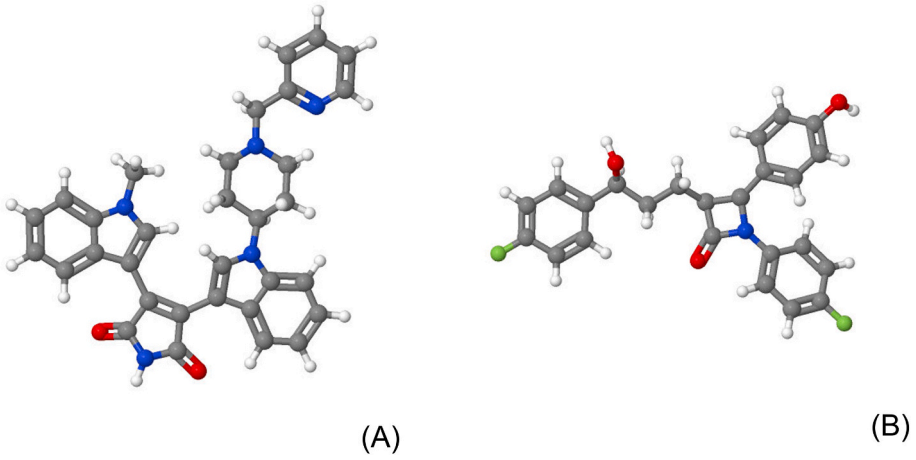

Fig. 2. (A). Structure of the antitumor enzastaurin; (B). Structure of the lipid-lowering ezetimibe.

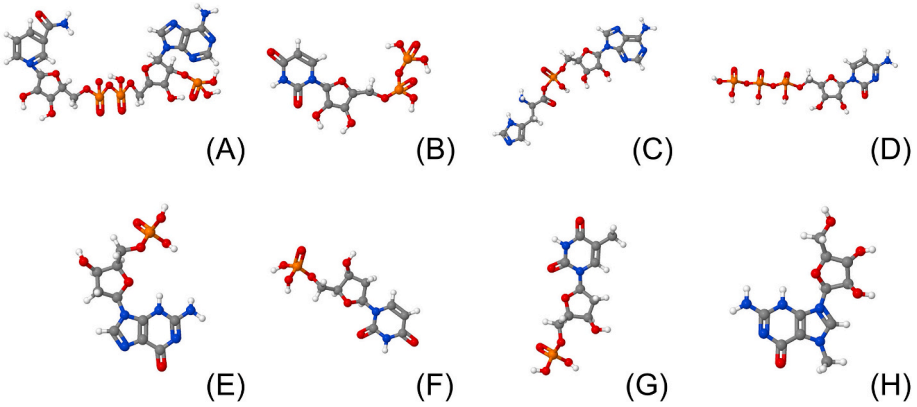

Fig. 3. (A). Structure of the nucleotide nicotinamide adenine dinucleotide phosphate (NADPH); (B). Structure of the nucleotide uridine-5′-diphosphate (UDP); (C). Structure of the histidyl-adenosine monophosphate nucleotide (H-AMP); (D). Cytidine-5′-triphosphate (CTP) nucleotide structure; (E). Nucleotide structure 2′-deoxyguanosine-5′-monophosphate (dGMP); (F). Structure of the nucleotide deoxyuridine monophosphate (dUMP); (G). Structure of the nucleotide thymidine monophosphate (TMP); (H). Structure of the nucleoside 7-methylguanosine.

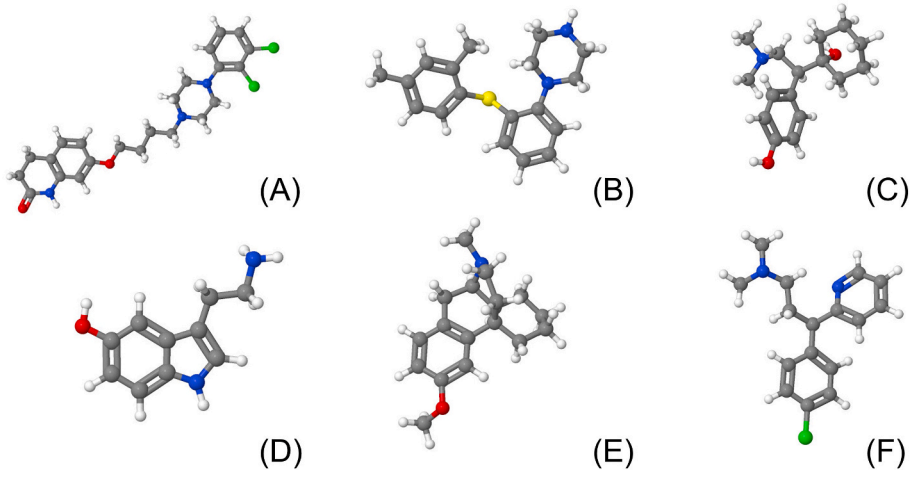

Fig. 4. (A). Structure of the antipsychotic aripiprazole; (B). Structure of the antidepressant vortioxetine; (C). Structure of the antidepressant desvenlafaxine; (D). Structure of the neurotransmitter serotonin; (E). Structure of the antihistamine chlorpheniramine; (F). Structure of the antitussive dextromethorphan.

protected from the effects of antibiotics [70]. Therefore DTI HipA-enzastaurin could alter persistence activity allowing antibiotic treatments to be effective. No studies have been found that describe any non-human pharmacological target. There were also no targets from any organism other than humans in the DB database. However, literature exists on the use of antibiotics as antitumor drugs, sometimes as the main method to treat malignancies [71]. This is due to the high structural similarities that exist between bacterial and human proteins. These

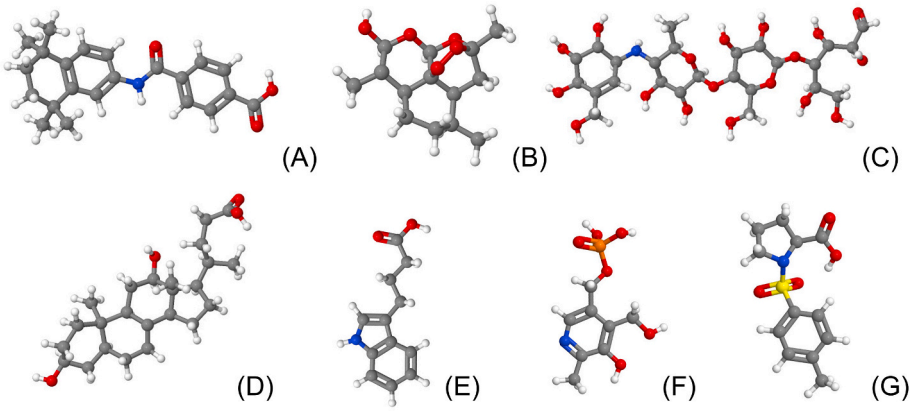

Fig. 5. (A). Structure of the retinoid tamibarotene; (B). Structure of the antimalarial artenimol; (C). Structure of the hypoglycemic acarbose; (D). Structure of the cytolytic deoxycholic acid; (E). Structure of the heterocyclic compound 3-indolebutyric acid; (F). Structure of the pyridoxine phosphate molecule; (G). Structure of the molecule tosyl-D-proline.

similarities, which are also reflected in the results of this model, could be the mathematical prediction results needed to design new antibiotics targeting these proteins and/or to reposition known drugs that recognize these targets as potential antimicrobials [72]. These results could be of interest to "reverse" the process of repositioning antitumor drugs for the search for new antibacterials. Ezetimibe is a drug belonging to the group of non-antibiotic betalactams that has been widely used as a lipid-lowering agent in disorders such as primary hyperlipidemia and familial cholesterolemia [73]. In this study ezetimibe had only one DTI with the protein 2DQ6, the major aminopeptidase in E. coli [74]. It is involved in ATP-dependent downstream processing during cytosolic protein degradation and possess significant physiological functions as a receptor [75]. No work has reflected any DTI of ezetimibe with *E. coli* proteins. However, due to its topological properties and the DTI observed in the results, ezetimibe could be a molecule of interest for the search for new betalactam antibiotics, acting as a lead compound.

The dinucleotide nicotinamide adenine dinucleotide phosphate (NADPH) is present in metabolic processes of many living organisms, including bacteria such as *E. coli* and *H. influenzae* [76,77]. According to the model, the two *E. coli* proteins similar to the *H. influenzae* protein 1PR3 could interact with the presence of NADPH with high biological activity (Tables 2 and 3). There are studies that show that the alteration of NADPH in the metabolism of Gram-negative bacteria and mycobacteria allows a greater sensitivity to different antibiotics [78–80]. A study by Lyons et al., explains the role of NADPH as an adjuvant to F-Box Stress-induced 1 (FBsl), an N-hydroxylated monooxygenase involved in the biosynthesis of fimsbactin A, the main siderophore produced by *Acinetobacter baumannii* [81]. In this study, NADPH had eight DTI with the proteins 5AED, 1OG6, 1BSV, 1L5J, 2XHY, 3DMY, 1BRM and 5WAN. 5AED, or YihQ is a sulfoquinovosidase with hydrolisis activity against alpha-glucosyl fluoride [82]. This activity allows the stability in the protein synthesis. 1BSV is a GDP-fucose synthetase with dual function in the sugar metabolism [83]. Defects in GDP-fucose biosynthesis have been shown to affect nodulation in bacteria. 1L5J is the major bifuncional aconitase of *E. coli* (AcnB), and represents a large, distinct group of Gram-negative bacterial aconitases that have an altered domain organization relative to other aconitases [84]. AcnB serves as either an enzymic catalyst or a mRNA-binding post-transcriptional regulator. 2XHY or BglA is a cytoplasmic enzyme and part of the glycosal hydrolase family that is able to hydrolase aromatic β-glucoside phosphates [85]. 1BRM is a aspartate β-semialdehyde dehydrogenase (ASADH) which lies at the first branch point in an essential aspartic biosynthetic pathway found in bacteria and other organisms [86]. ASADH may be an effective target for antibacterial agents because mutations in the asd gene encoding for ASADH produce an enzyme that is inactive and lethal to the organism. 5WAN or flavoenzyme RutA catalyze, along with the flavin reductase RutF, the initial uracil ring opening by an unprecedent "oxidative" hydrolysis reaction to give 3-ureidoacrylate [87]. Many of the proteins that interact with NADPH are involved in metabolic processes, some are even essential for *E. coli*. It would therefore be interesting to investigate these targets further to see if they can be applied in the design of future antimicrobial therapies.

Another nucleotide present in our results was uridine-5′-diphosphate (UDP), which had two pharmacological targets of *E. coli* registered in the DB database, although it did not specify what its function was in them. A work by Šudomová et al. reveals the efficacy of UDP as an inhibitor of UDP-galactopyranose mutase, an enzyme vital in the cell wall biosynthesis of *M. tuberculosis* [88]. Our model has not identified these targets in the similarities between *E. coli* and *M. tuberculosis*. However, the molecules presented in this work could have other *M. tuberculosis* targets with functions similar to those of UDP. Histidyl-adenosine monophosphate (H-AMP) presents a single target of *E. coli* in the DB database, specifically a histidine-tRNA ligase. This model has identified three new possible targets of *E. coli* for H-AMP that come from essential genes. One of them, the 1C3B protein, is a AmpC β-lactamase, and the othe one protein, 6XGY, has topological similarities with the threonine-phosphate decarboxylase of *S.* Typhimurium (Tables 2 and 3). However, no literature has been found related to antibacterial activity against either microorganism. These results expose the possibility of finding new molecules with activity against bacteria of *Enterobacterales* from the topological properties of H-AMP. Cytidine-5′-triphosphate (CTP) is a nucleotide that presents *E. coli* and *S.* Typhimurium targets in DB, although no mechanism of action is explained. The model has identified a single protein from essential *E. coli* genes but, in contrast to H-AMP, no topological similarities were found between of *E. coli* and *S.* Typhimurium targets (Table 2), nor literature that speaks of antibiotic actions against any microorganism. However, the protein 1H3M is an essential enzyme in the mevalonate-independent pathway of isoprenoid biosynthesis, whose structure suggest that the enzyme is a suitable target for a structure-based approach to the development of novel broad-spectrum antibiotics [89]. These results could be the beginning of a search for new antimicrobials that present these proteins as pharmacological targets. 2′-deoxyguanosine-5′-monophosphate (dGMP) and deoxyuridine monophosphate (dUMP) have presented a single DTI both in our model and in the DB database. This is thymidylate synthase, an enzyme from the essential thyA gene that participates in DNA synthesis in the case of *E. coli*, but also appears in other microorganisms with other vital functions related to the folate pathway [90,91]. There are no bibliographic records on the activity of dGMP in this target or any other as a molecule with antimicrobial properties. In the case of dUMP, it is a

molecule that interacts directly with the enzyme whereby the alteration of the latter can encourage the appearance of resistance in species such as *M. tuberculosis* [90], so it would be interesting to carry out studies on similar molecules or those with the same target to find drugs with antituberculous activity. The nucleotide thymidine monophosphate (TMP) also has interactions with *M. tuberculosis* enzymes, in addition to *E. coli* and *P. aeruginosa*, according to the DB database. In our study, such coincidences were not found, since only a single protein from the essential *topA* gene was observed with a DTI of high binding energy, which, according to PDB, corresponds to the enzyme DNA Topoisomerase I of *E. coli* (Table 3). However, several studies describe the importance of this molecule in the search for nucleotide analogs to identify new antituberculous drugs, so that, with the data provided by our model, we could expand the antimicrobial spectrum of known molecules [92–96]. For 7-methylguanosine nucleoside, no target information was found for *E. coli* or any other bacteria in the DB database or in the literature. However, the model identified a DTI with a protein from essential genes. These results for nucleotides and nucleosides highlight the importance of looking for new antimicrobials from the DTIs involving these molecules or their participation with enzymes of other microorganisms of interest, such as most of those present in Table 2, in addition to *A. baumannii*, listed by the WHO as a critical priority in the search for new antibiotics [34]. These results also highlight the importance of searching for new antimicrobial drugs from nucleotide and nucleoside analogs.

One of the most relevant results of our model relates to the presence of psychotropic drugs (Fig. 3) such as aripiprazole, vortioxetine, desvenlafaxine, and duloxetine (Table 3). Recent studies show the influence of these drugs on the composition of the intestinal microbiota, whose role in the regulation of gut-brain interactions is being increasingly demonstrated [97]. In our study, these molecules presented DTI with two proteins with similar binding energies, becoming the two most frequent bacterial proteins in the model. These proteins are 1HTT and 1F6D (Table 2). 1HTT is a histidyl-tRNA synthetase belongs to the class II of aminoacyl-tRNA synthetases [98]. 1F6D is a bacterial epimerase which provides bacteria with UDP-N-acetylmannosamine (UDP-ManNAc), the activated donor of ManNAc residues [99]. ManNAc is critical for several processes in bacteria, including formation of the antiphagocytic capsular polysaccharide of pathogens such as *S. pneumoniae*. These proteins also present DTI with chlorpheniramine and dextromethorphan, which will be considered further below. The neurotransmitter serotonin also interacts with the protein 1HTT, and there is a very extensive literature on the role of this molecule in altering microbial diversity in the gut microbiota of different animal species [100,101]. This is because gut microorganisms influence brain functions by acting through the vagus nerve or by altering the production of short-chain fatty acids or the amino acid tryptophan, the building block of serotonin [102]. In addition, there are microorganisms capable of producing or regulating serotonin metabolism [103]. However, this neurotransmitter does not present any target against *E. coli* registered in the DB database. Nor does aripiprazole, a molecule with antipsychotic pharmacological function, present any known DTI with *E. coli* in DB, but there are studies that demonstrate its role as a modulator of the intestinal and cutaneous microbiota [99,104]. These data show that aripiprazole, or molecules with similar characteristics, may merit future research related to microbiota therapy. Vortioxetine, desvenlafaxine, and duloxetine are antidepressant modulators and stimulators of neurotransmitters such as serotonin and norepinephrine. No DTI with bacterial proteins were found in DB. In the case of vortioxetine and desvenlafaxine, no literature was found relating to the targets that appeared in our study, nor a pharmacological role beyond that of antidepressant. In the case of duloxetine, literature has been found regarding its role in the manipulation of the intestinal microbiota, specifically in the acceleration of the emergence of antibiotic resistance due to the combination of both classes of drugs in therapy [105,106]. Several studies explain how the antidepressants duloxetine, fluoxetine, sertraline, quetiapine, and bupropion promote conjugative transfer of multi-antibiotic resistance genes [105,107]. Another more recent study by Wang et al. revealed by means of a mathematical model and phenotypic and genotypic analyzes the increase of species with greater resistance and persistence to antimicrobial treatments [108]. These results expose the importance of the use of antidepressants in antimicrobial therapies, where new measures should be proposed for a reasonable use of these drugs and their combinations. In addition, due to the lack of information on vortioxetine and desvenlafaxine, it would be interesting to carry out trials to test their possible repositioning as antibiotics to prove the effectiveness of the model in the search for new antimicrobials, as well as studies that expose the possible synergy between these molecules and different antibiotics, along with their role in the manipulation of the intestinal microbiota and the possible risk of increasing AMR, as seen in other antidepressants. Overall, this model could provide new insights into bacterial responses to antidepressants and improve understanding of the effects of antidepressant treatment.

On a different note, as mentioned previously, both dextromethorphan and chlorpheniramine coincide with the targets of the four psychotropic resulting from our model (Table 3). Like vortioxetine and duloxetine, no records on DTI with bacterial proteins are found in the DB database. A study by Kirkwood et al. described the *in vitro* efficacy of chlorpheniramine as a growth inhibitor of *Mycobacterium abscessus* [109]. Given the lack of literature on these molecules in antimicrobial therapeutics, and the results obtained in this *in silico* trial, it would be interesting to carry out more studies to show their possible antimicrobial activity and, in the case of chlorpheniramine, its antimicrobial spectrum.

Tamibarotene is a new synthetic retinoid that is approved in Japan and in clinical trials in the US for the treatment of acute promyelocytic leukemia [110]. This molecule presents, according to our model, the same target as adapalene, a third-generation topical retinoid (Tables 1 and 3). Adapalene is used as an antibiotic adjuvant in the treatment of *acne vulgaris*, a disorder of the pilosebaceous unit caused by *Cutibacterium acnes* (*Propionibacterium acnes)* [111–113]. Due to the similarity of binding energies against the same target, it seems possible that tamibarotene has the same adjuvant properties as adapalene (Table 1 and 3). However, no literature has been found in this regard, nor pharmacological targets of microbial origin that interact with this molecule in the DB database, which makes it of interest for future trials to verify its possible antimicrobial or antibiotic adjuvant effect.

Artenimol (also known as dihydroartemisinin) is an antimalarial agent used in combination with piperaquine for uncomplicated *Plasmodium falciparum* infections [114]. However, there are also studies that expose its efficacy as an adjuvant in chemodynamic and photodynamic antibacterial therapies [115,116]. Additionally, studies such as those of Huang et al. and Kalani et al. exhibit the efficacy of artenimol in combination with antibiotics such as cefuroxime and ampicillin against *E. coli* infections, and in combination with rifampicin against *M. tuberculosis* [117,118].

Acarbose, an oral hypoglycemic agent used for glycemic control in patients with type 2 diabetes mellitus, has also demonstrated efficacy as a potential antimicrobial drug in several *in silico* and *in vitro* trials [119]. It has even been used as a control molecule to evaluate other α-glucosidase inhibitors as potential bacterial growth inhibitors [120–122]. The mechanism of acarbose as an inhibitor of bacterial growth is due to the similarities of human and bacterial α-glucosidase enzymes. In fact, there are known cases of intestinal and oral human microbiota that present enzymes that phosphorylate acarbose, inactivating it [123]. Therefore, as we have been able to observe with psychotropic drugs, it is important to make reasonable use of this drug to avoid possible AMRs.

Deoxycholic acid is a cytolytic agent that has demonstrated its efficacy in regulating primary bile acids by altering the intestinal microbiota, and as an antibiotic adjuvant together with polyamines for facial infections [124,125]. This association with the intestinal or cutaneous microbiota favors the prevention and treatment of bacterial infections,

alone or combined with antibiotics, which it potentiates.

Finally, 3-indolebutyric acid, which, according to the model, could interact with the protein from essential genes 3FLS, presents a single study where it exposes its antimicrobial efficacy in the so-called nanoparticle antimicrobial therapy, so it would be interesting to carry out more *in vitro* tests to demonstrate this efficacy as an antibiotic [126]. Pyridoxine phosphate and tosyl-D-proline both present, in our model and in the DB database, a pharmacological target against *E. coli* from essential genes (Table 3). However, no literature has been found on DTI with antimicrobial effect against any pathogen, so it would be interesting to carry out more studies to verify the possible efficacy as an antibiotic or antivirulent adjuvant of these two molecules.

All these results prove the specificity of the model to predict molecules with potential antimicrobial activity or antibiotic adjuvants, also helping to predict possible DTI that could favor the appearance of AMR.

6. Conclusion

The present *in silico* assay has shown that the SA-TDA-based model has been able to identify of antibiotic drugs such as cephalosporins, carbapenems and semisynthetic penicillins, in addition to other molecules also known for their use as antibiotic adjuvants or disinfectant agents. These results confirm the model's ability to predict, from the topological similarities of known target proteins of FDA-approved drugs and *E. coli* proteins, molecules with potential antimicrobial properties. Within the known targets, proteins of other pathogens of interest in the search for new antibiotics have been found, so the model could be detecting new broad-spectrum antibiotics, being able to deal with Gram-positive, Gram-negative and mycobacteria. The molecules detected in the model have been very diverse in terms of their pharmacological function. Among them, nucleotides stand out as they are the most frequent family in the study, followed by antidepressants. Nucleotides have an extensive literature as antibiotic adjuvants. In addition, our model has presented new targets that could be of interest for future antimicrobial therapies, so it would be interesting to carry out more tests aimed at finding antibiotic molecules with topological properties similar to those of nucleotides. Antidepressants also stood out for concurring in the same two targets in *E. coli*, so it would be interesting to carry out more studies to confirm the antibacterial activity of antidepressants with these targets, in addition to other molecules that, as in this study, show DTI with high binding energies to these proteins. Although the results provided by the model are promising, there is still a need for *in vitro* assays to confirm the reliability and specificity of the model, as well as to observe its limitations, thus improving the search for molecules with potential antimicrobial capacity by using mathematical prediction models, topological data analysis and drug repurposing.

Author contributions

Conception and design of the study: A. T-P., B. S-G., J. F-M, A. F. and M-T. P-G.; adquisition of data: J. F-M.; analysis and interpretation of data: A. T-P., B. S-G., M-T. P-G. and A. F.; draft the article: A. T-P., B. S-G., A. F. and M-T. P-G.; revision: M-T. P-G. and A. F. All authors have read and agreed to the published version of the manuscript.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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References

[1] World Health Organization, Ten Threats to Global Health in 2019, World Health Organization, Geneva, 2019. Available online: https://www.who.int/news-room/spotlight/ten-threats-to-global-health-in-2019. (Accessed 4 January 2023).
[2] World Health Organization, Global Antimicrobial Resistance and Use Surveillance System (GLASS) Report 2022. Geneva, 2022. License: CC BY-NC-SA 3.0 IGO.
[3] Antimicrobial Resistance Collaborators, Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis, Lancet 12 (399) (2022) 629–655, https://doi.org/10.1016/S0140-6736(21)02724-0, 10325.
[4] European Antimicrobial Resistance Collaborators, The burden of bacterial antimicrobial resistance in the WHO European region in 2019: a cross-country systematic analysis, Lancet Public Health 7 (11) (2022) E897–E913, https://doi.org/10.1016/S2468-2667(22)00225-0.
[5] Drug-resistant Infections: a Threat to Our Economic Future, World Bank, Washington, D.C., 2017. License: Creative Commons Attribution CC BY 3.0 IGO. Available online: http://documents.worldbank.org/curated/en/323311493396993758/final-report. (Accessed 30 January 2023).
[6] World Health Organization, Global Action Plan on Antimicrobial Resistance. Geneva, 2015. Available online: https://www.who.int/publications/i/item/9789241509763. (Accessed 30 January 2023).
[7] A. Tarín-Pelló, B. Suay-Garcia, M.T. Perez-Gracia, Antibiotic resistant bacteria: current situation and treatment options to accelerate the development of a new antimicrobial arsenal, Expert Rev. Anti. Infect. Ther. (2022) 1–14, https://doi.org/10.1080/14787210.2022.2078308.
[8] O. Pacios, L. Blasco, I. Bleriot, L. Fernandez-Garcia, M. González Bardanca, To Ambroa, M. Lopez, G. Bou, M. Tomás, Strategies to combat multidrug-resistant and persistent infectious diseases, Antibiotics (Basel) 6 (9) (2020), 65, https://doi.org/10.3390/antibiotics9020065, 2.
[9] T. Mirski, M. Lydia, T. Nakonieczna, R. Gryko, Bacteriophages, phage endolysins, and antimicrobial peptides - the possibilities for their common use to combat infections and in the design of new drugs, Ann. Agric. Environ. Med. 26 (2) (2019) 203–209, https://doi.org/10.26444/aaem/105390.
[10] L. Rodrigues, P. Cravo, M. Viveiros, Efflux pump inhibitors as a promising adjunct therapy against drug resistant tuberculosis: a new strategy to revisit mycobacterial targets and repurpose old drugs, Expert Rev. Anti Infect. Ther. 18 (8) (2020) 741–757, https://doi.org/10.1080/14787210.2020.1760845.
[11] Z. Shang, S.Y. Chan, Q. Song, P. Li, W. Huang, The strategies of pathogen-oriented therapy on circumventing antimicrobial resistance, Research 28 (2020), https://doi.org/10.34133/2020/2016201.
[12] WHO Antibacterial Agents in Clinical and Preclinical Development: an Overview and Analysis. Geneva, 2021. Available online: https://www.who.int/publications/i/item/9789240021303. (Accessed 30 January 2023).
[13] B. Aslam, W. Wang, M.I. Arshad, M. Khurshid, S. Muzammil, M.H. Rasool, M. A. Nisar, R.F. Alvi, M.A. Aslam, M.U. Qamar, et al., Antibiotic resistance: a rundown of a global crisis, Infect. Drug Resist. 11 (2018) 1645–1658, 10.2147/IDR. S173867.
[14] K.I. Mohr, History of antibiotics research, Curr. Top. Microbiol. Immunol. 398 (2016) 237–272, https://doi.org/10.1007/82_2016_499.
[15] J.A. Al-Tawfiq, H. Momattin, A.Y. Al-Ali, K. Eljaaly, R. Tirupathi, M. B. Haradwala, S. Areti, S. Alhumaid, A.A. Rabaan, To Al Mutair, et al., Antibiotics in the pipeline: a literature review (2017-2020), Infection 50 (3) (2022) 553–564, https://doi.org/10.1007/s15010-021-01709-3.
[16] U. Theuretzbacher, S. Gottwalt, P. Beyer, M. Butler, L. Czaplewski, C. Lienhardt, L. Moja, M. Paul, S. Paulin, J.H. Rex, et al., Analysis of the clinical antibacterial and antituberculosis pipeline, Lancet Infect. Dis. 19 (2019) E40–E50, https://doi.org/10.1016/S1473-3099(18)30513-9.
[17] WHO Antibacterial Agents in Clinical Development: an Analysis of the Antibacterial Clinical Development Pipeline. Geneva, 2019. Available online: https://www.who.int/publications/i/item/9789240000193. (Accessed 31 January 2023).
[18] E.B. Chahine, J.A. Dougherty, K.A. Thornby, E.H. Guirguis, Antibiotic approvals in the last decade: are we keeping up with resistance? Ann. Pharmacother. 56 (4) (2022) 441–462, https://doi.org/10.1177/10600280211031390.
[19] R. Knoblauch, C.D. Geddes, Carbon nanodots in photodynamic antimicrobial therapy: a review, Materials 13 (18) (2020), 4004, https://doi.org/10.3390/MA13184004.
[20] M. Hay, D.W. Thomas, J.L. Craighead, C. Economics, J. Rosenthal, Clinical development success rates for investigational drugs, Nat. Biotechnol. 32 (1) (2014) 40–51, https://doi.org/10.1038/NBT.2786.
[21] N.K. Boyd, C. Teng, C.R. Frei, Brief overview of approaches and challenges in new antibiotic development: a focus on drug repurposing, Front. Cell. Infect. Microbiol. 17 (2021), 684515, https://doi.org/10.3389/fcimb.2021.684515, 11.
[22] J.W. Scannell, To Blanckley, H. Boldon, B. Warrington, Diagnosing the decline in pharmaceutical R&D efficiency, Nat. Rev. Drug Discov. 11 (3) (2012) 191–200, https://doi.org/10.1038/NRD3681.
[23] PhRMA. Fact, Sheet on Challenges with AMR, 2021. Available online: https://phrma.org/-/media/Project/PhRMA/PhRMA-Org/PhRMA-Org/PDF/A-C/AMR-Ecosystem-Challenges-Backgrounder_PhRMA.pdf. (Accessed 31 January 2023).
[24] K. Iskandar, J. Murugaiyan, D. Hammoudi Halat, S.E. Hage, V. Chibabhai, S. Adukkadukkam, C. Roques, L. Molinier, P. Salameh, M. Van Dongen, Antibiotic

discovery and resistance: the chase and the race, Antibiotics (Basel) 11 (2) (2022), 182, https://doi.org/10.3390/antibiotics11020182.
[25] N. Dheman, N. Mahoney, E.M. Cox, J.J. Farley, T. Amini, M.L. Lanthier, An analysis of antibacterial drug development trends in the United States, 1980-2019, Clin. Infect. Dis. 73 (11) (2021) E4444–E4450, https://doi.org/10.1093/cid/ciaa859.
[26] B. Suay-Garcia, To Falcó, J.I. Bueso-Bordils, G.M. Anton-Fos, M.T. Perez-Gracia, P.A. Alemán-López, Tree-based QSAR model for drug repurposing in the discovery of new antibacterial compounds against *Escherichia coli*, Pharmaceuticals 13 (2020), 431, https://doi.org/10.3390/ph13120431.
[27] R. Gupta, D. Srivastava, M. Sahu, S. Tiwari, R.K. Ambasta, P. Kumar, Artificial intelligence to deep learning: machine intelligence approach for drug discovery, Mol. Divers. 25 (3) (2021) 1315–1360, https://doi.org/10.1007/s11030-021-10217-3.
[28] S. Kim, J. Chen, T. Cheng, To Gindulyte, J. He, S. He, Q. Li, B.A. Shoemaker, P. A. Thiessen, B. Yu, et al., PubChem in 2021: new data content and improved web interfaces, Nucleic Acids Res. 49 (D1) (2021) D1388–D1395, https://doi.org/10.1093/nar/gkaa971.
[29] Y. Skaf, R. Laubenbacher, Topological data analysis in biomedicine: a review, J. Biomed. Inf. 130 (2022), 104082. https://doiDOIg/10.1016/j.jbi.2022.104082.
[30] Z. Meng, K. Xia, Persistent spectral based machine learning (PerSpect ML) for drug design, Sci. Adv. 7 (19) (2021), eabc5329, https://doi.org/10.1126/sciadv.abc5329.
[31] R. Wang, S. Li, L. Cheng, M.H. Wong, K.S. Leung, Predicting associations among drugs, targets and diseases by tensor decomposition for drug repositioning, BMC Bioinf. 20 (26) (2019) 628, https://doi.org/10.1186/s12859-019-3283-6.
[32] K. Xia, G.W. Wei, Persistent homology analysis of protein structure, flexibility, and folding, Int. J. Numer Method Biomed. Eng. 30 (8) (2014) 814–844. https://doiDOIg/10.1002/cnm.2655.
[33] K. Tamal, M. Dey, M. Sayan, Protein classification with improved topological data analysis, in: L. Parida, E. Ukkonen (Eds.), 18th International Workshop on Algorithms in Bioinformatics (WABI 2018), vol. 113, Schloss Dagstuhl-Leibniz-Zentrum fuer Informatik, Dagstuhl, Germany, 2018, https://doi.org/10.4230/LIPIcs.WABI.2018.6, 6:1–6:13.
[34] WHO, WHO Publishes the List of Bacteria for Which New Antibiotics Are Urgently Needed. Geneva, 2017. Available online:.
[35] M. Ali, A. Ezzat, DrugBank Database XML Parser. Dainanahan, R Package Version 1.2.0, 2020. Available online: https://CRAN.R-project.org/package=dbparser. (Accessed 1 February 2023).
[36] V. Robins, Towards computing homology from finite approximations, Topol. Proc. 24 (1999) 503–532.
[37] R. Pérez-Moraga, J. Forés-Martos, B. Suay-Garcia, J.L. Duval, To Falcó, J. Climent, A COVID-19 drug repurposing strategy through quantitative homological similarities using a topological data analysis-based framework, Pharmaceutics 13 (4) (2021), 488, https://doi.org/10.3390/pharmaceutics13040488.
[38] S. Nistorescu, G. Gradisteanu Pircalabioru, A.-M. Udrea, To Simon, M.L. Pascu, M.-C. Chifiriuc, Laser-irradiated chlorpromazine as a potent anti-biofilm agent for coating of biomedical devices, Coatings 10 (12) (2020), 1230, https://doi.org/10.3390/coatings10121230.
[39] M. Dwivedi, S. Mukhopadhyay, S. Yadav, K.D. Dubey, A multidrug efflux protein in Mycobacterium tuberculosis; tap as a potential drug target for drug repurposing, Comput. Biol. Med. 146 (2022), 105607, https://doi.org/10.1016/j.compbiomed.2022.105607.
[40] To Kumar, To Alam, S. Grover, S. Pandey, D. Tripathi, M. Kumari, M. Rani, To Singh, Y. Akhter, N.Z. Ehtesham, S.E. Hasnain, Peptidyl-prolyl isomerase-B is involved in *Mycobacterium tuberculosis* biofilm formation and a generic target for drug repurposing-based intervention, NPJ. Biofilms Microbiomes 5 (1) (2019) 3, https://doi.org/10.1038/s41522-018-0075-0.
[41] B.J. Grant, A.P.C. Rodrigues, K.M. El Sawy, J.A. Mc Cammon, L.S.D. Caves, Bio3d: an R package for the comparative analysis of protein structures, Bioinformatics 22 (21) (2006) 2695–2696, https://doi.org/10.1093/bioinformatics/btl461.
[42] Z. Cang, L. Mu, K. Wu, K. Opron, K. Xia, G.W. Wei, A topological approach for protein classification, Mol. Base. Math. Biol. 3 (2015) 140–162. Available online, https://hdl.handle.net/10356/82112. (Accessed 1 February 2023).
[43] U. Bauer, Ripser: Efficient Computation of Vietoris-Rips Persistence Persistence Diagrams, *arXiv*, 2019. arXiv:1908.02518.
[44] B.J. Jain, M. Lappe, Joining softassign and dynamic programming for the contact map overlap problem, For. Bird 4414 (2007) 410–423, https://doi.org/10.1007/978-3-540-71233-6_32.
[45] G. Lancia, R. Carr, B. Walenz, S. Istrail, 101 optimal pdb structure alignments: a branch-and-cut algorithm for the maximum contact map overlap problem, in: Proceedings of the Fifth Annual International Conference on Computational Biology, Canada, Montreal, QC, 22–25 April 2001, pp. 193–202.
[46] N.K. Fox, S.E. Brenner, J.M. Chandonia, SCOPe: structural Classification of Proteins–extended, integrating SCOP and ASTRAL data and classification of new structures, Nucleic Acids Res. 42 (2014) D304–D309, https://doi.org/10.1093/nar/gkt1240.
[47] N.M. O'Boyle, M. Banck, C.A. James, C. Morley, T. Vandermeersch, G. R. Hutchison, Open Babel: an open chemical toolbox, J. Cheminf. 3 (2011), 33, https://doi.org/10.1186/1758-2946-3-33.
[48] G.M. Morris, R. Huey, W. Lindstrom, M.F. Sanner, R.K. Belew, D.S. Goodsell, A. Olson, AutoDock4 and AutoDockTools4: automated docking with selective receiver flexibility, J. Comput. Chem. 30 (2009) 2785–2791, https://doi.org/10.1002/jcc.21256.
[49] S. Forli, R. Huey, M. Pique, M. Sanner, D. Goodsell, A. Olson, Computational protein-ligand docking and virtual drug screening with the AutoDock suite, Nat. Protoc. 11 (2016) 905–919, https://doi.org/10.1038/nprot.2016.051.
[50] V. Kovacev-Nikolic, P. Bubenik, D. Nikolić, G. Heo, Using persistent homology and dynamical distances to analyze protein binding, Stat. Appl. Genet. Mol. Biol. 15 (1) (2016) 19–38, https://doi.org/10.1515/sagmb-2015-0057.
[51] S.J.Y. Macalino, J.B. Billones, V.G. Organo, M.C.O. Carrillo, *In silico* strategies in tuberculosis drug discovery, Molecules 25 (3) (2020), 665, https://doi.org/10.3390/molecules25030665.
[52] T. Ichinomiya, I. Obayashi, Y. Hiraoka, Protein-folding analysis using features obtained by persistent homology, Biophys. J. 118 (12) (2020) 2926–2937, https://doi.org/10.1016/j.bpj.2020.04.032.
[53] B. Hernández-Ochoa, G. Navarrete-Vázquez, R. Aguayo-Ortiz, P. Ortiz-Ramírez, L. Morales-Luna, V. Martínez-Rosas, A. González-Valdez, F. Gómez-Chávez, S. Enríquez-Flores, C. Wong-Baeza, et al., Identification and *in silico* characterization of novel *Helicobacter pylori* glucose-6-phosphate dehydrogenase inhibitors, Molecules 26 (16) (2021), 4955, https://doi.org/10.3390/molecules26164955.
[54] X. Sun, Y. Zhang, H. Li, Y. Zhou, S. Shi, Z. Chen, X. He, H. Zhang, F. Li, J. Yin, et al., DRESIS: the first comprehensive landscape of drug resistance information, Nucleic Acids Res. 6 (51) (2023) D1263–D1275, https://doi.org/10.1093/nar/gkac812. D1.
[55] G. Naitzat, N. Lokare, J. Silva, I. Kaynar-Kabul, M-Boost: profiling and refining deep neural networks with topological data analysis: extended abstract, Article 4, in: J.B. Sartor, T. D'Hondt, W. De Meuter (Eds.), Workshop on Interactive Data Exploration and Analytics, ACM, New York, USA, 2018, p. 9, 10.475/123_4.
[56] R. Brüel-Gabrielsson, B.J. Nelson, To Dwaraknath, P. Skraba, L.J. Guibas, G. Carlsson, A topology layer for machine learning, in: 23rd International Conference on Artificial Intelligence and Statistics (AISTATS), vol. 108, PMLR, Palermo, Italy, 2020, https://doi.org/10.48550/arXiv.1905.12200.
[57] R. Dehbanipour, Z. Ghalavand, Anti-virulence therapeutic strategies against bacterial infections: recent advances, Germs 12 (2) (2022) 262–275, https://doi.org/10.18683/germs.2022.1328.
[58] E.J. Kingston, S.L. Zagora, R.J. Symes, P. Raman, P.J. McCluskey, J.A. Lusthaus, Infective necrotizing scleritis after XEN gel stent with mitomycin-C, J. Glaucoma 31 (2) (2022) 129–132, https://doi.org/10.1097/IJG.0000000000001959.
[59] T. Kuroda, Y. Minamijima, H. Niwa, H. Mita, N. Tamura, K. Fukuda, To Kuwano, F. Sato, Concentration of cephalothin in body fluids and tissue samples of Thoroughbred horses, J. Equine Sci. 33 (3) (2022) 51–54, https://doi.org/10.1294/jes.33.51.
[60] F. Sanz Herrero, Ceftazidime-avibactam, Rev. Española Quimioter. 35 (1) (2022) 40–42, https://doi.org/10.37201/REQ/S01.09.2022. Suppl 1.
[61] Y. Chen, H.B. Huang, J.M. Peng, L. Weng, B. Du, Efficacy and safety of ceftazidime-avibactam for the treatment of carbapenem-resistant *Enterobacterales* bloodstream infection: a systematic review and meta-analysis, Microbiol. Spectr. 10 (2) (2022), E0260321, https://doi.org/10.1128/spectrum.02603-21.
[62] J.E. Cronan, The *Escherichia coli* FadR transcription factor: too much of a good thing? Mol. Microbiol. 115 (6) (2021) 1080–1085, https://doi.org/10.1111/MMI.14663.
[63] S. Yousuf, R.K. Angara, To Roy, S.K. Gupta, R. Misra, A. Ranjan, Mce2R/Rv0586 of *Mycobacterium tuberculosis* is the functional homologue of FadR*E. coli*, Microbiology (Read.) 164 (9) (2018) 1133–1145, https://doi.org/10.1099/mic.0.000686.
[64] C. Datta, R.K. Jha, S. Ganguly, V. Nagaraja, NapA (Rv0430), a novel nucleoid-associated protein that regulates a virulence operon in *mycobacterium tuberculosis* in a supercoiling-dependent manner, J. Mol. Biol. 431 (8) (2019) 1576–1591, https://doi.org/10.1016/j.jmb.2019.02.029.
[65] V. Khemici, M. Prudhomme, P. Polard, Tight interplay between replication stress and competence induction in *Streptococcus pneumoniae*, Cells 10 (8) (2021), 1938, https://doi.org/10.3390/cells10081938.
[66] K.P. Rathbun, A.M. Bourgault, M.L. Sole, Oral microbes in hospital-acquired pneumonia: practice and research implications, Crit. Care Nurse 42 (3) (2022) 47–54, https://doi.org/10.4037/CCN2022672.
[67] R.E. Impey, D.A. Hawkins, J.M. Sutton, T.P. Soares da Costa, Overcoming intrinsic and acquired resistance mechanisms associated with the cell wall of gram-negative bacteria, Antibiotics 9 (9) (2020), 623, https://doi.org/10.3390/antibiotics9090623.
[68] To Polisety, G. Misra, J. Rajawat, To Katiyar, H. Singh, A.N. Bhatt, Therapeutic natural compounds Enzastaurin and Palbociclib inhibit MASTL kinase activity preventing breast cancer cell proliferation, Med. Oncol. 39 (5) (2022) 100, https://doi.org/10.1007/s12032-022-01701-3.
[69] G.S. Nowakowski, J. Zhu, Q. Zhang, J. Brody, X. Sun, J. Maly, Y. Song, S. Rizvi, Y. Song, F. Lansigan, et al., ENGINE: a Phase III randomized placebo controlled study of enzastaurin/R-CHOP as frontline therapy in high-risk diffuse large B-cell lymphoma patients with the genomic biomarker DGM1, Future Oncol. 16 (15) (2020) 991–999, https://doi.org/10.2217/fon-2020-0176.
[70] A. Evdokimov, I. Voznesensky, K. Fennell, M. Anderson, J.F. Smith, D.A. Fisher, New kinase regulation mechanism found in HipBA: a bacterial persistence switch, Acta Crystallogr. D Biol. Crystallogr. 65 (8) (2009) 875–879, https://doi.org/10.1107/S0907444909018800.
[71] O.Y. Selyutina, A.V. Mastova, N.E. Polyakov, The interaction of anthracycline based quinone-chelators with model lipid membranes: 1H NMR and MD study, Membranes 13 (1) (2023), 61, https://doi.org/10.3390/membranes13010061.
[72] F. Farouk, A.A. Elmaaty, To Elkamhawy, H.O. Tawfik, R. Alnajjar, M.A. S. Abourehab, M.A. Saleh, W.M. Eldehna, A.A. Al-Karmalawy, Investigating the potential anticancer activities of antibiotics as topoisomerase II inhibitors and

DNA intercalators: *in vitro*, molecular docking, molecular dynamics, and SAR studies, J. Enzym. Inhib. Med. Chem. 38 (1) (2023), 2171029, https://doi.org/10.1080/14756366.2023.2171029.

[73] U. Laufs, A.L. Catapano, R. De Caterina, F. Schiele, To Sionis, To Zaman, J. W. Jukema, The effect of the 2019 ESC/EAS dyslipidaemia guidelines on low-density lipoprotein cholesterol goal achievement in patients with acute coronary syndromes: the ACS EuroPath IV project, Vasc. Pharmacol. 148 (2023), 107141, https://doi.org/10.1016/j.vph.2023.107141.

[74] K. Ito, Y. Nakajima, Y. Onohara, M. Takeo, K. Nakashima, F. Matsubara, T. Ito, T. Yoshimoto, Crystal structure of aminopeptidase N (proteobacteria alanyl aminopeptidase) from Escherichia coli and conformational change of methionine 260 involved in substrate recognition, J. Biol. Chem. 3 (281) (2006) 33664–33676, https://doi.org/10.1074/jbc.M605203200, 44.

[75] D. Chandu, D. Nandi, PepN is the major aminopeptidase in Escherichia coli: insights on substrate specificity and role during sodium-salicylate-induced stress, Microbiology (Read.) 149 (12) (2003) 3437–3447, https://doi.org/10.1099/mic.0.26518-0.

[76] J.P. Overington, B. Al-Lazikani, A.L. Hopkins, How many drug targets are there? Nat. Rev. Drug Discov. 5 (12) (2006) 993–996, https://doi.org/10.1038/NRD2199.

[77] P. Imming, C. Sinning, A. Meyer, Drugs, their targets and the nature and number of drug targets, Nat. Rev. Drug Discov. 5 (10) (2006) 821–834, https://doi.org/10.1038/NRD2132. Erratum in: Nat Rev Drug Discov. 2007, 6(2), 126.

[78] T.A. Seregina, I.Y. Petrushanko, R.S. Shakulov, P.I. Zaripov, A.A. Makarov, V. A. Mitkevich, A.S. Mironov, The inactivation of LPS biosynthesis genes in *E. coli* cells leads to oxidative stress, Cells 11 (17) (2022), 2667, https://doi.org/10.3390/cells11172667.

[79] J.D. Peace, N. Denise de Moura Sperotto, A.S. Ramos, K. Pissinate, V. da Silva Rodrigues Junior, B.L. Abbadi, A.F. Borsoi, R.S. Rambo, A.C. Corso Minotto, To da Silva Dadda, et al., Novel 4-aminoquinolines: synthesis, inhibition of the *Mycobacterium tuberculosis* enoyl-acyl carrier protein reductase, antitubercular activity, SAR, and preclinical evaluation, Eur. J. Med. Chem. 245 (Pt 1) (2023), 114908, https://doi.org/10.1016/j.ejmech.2022.114908.

[80] D. Zhang, J. Chen, Z. Wang, C. Wang, Integrated metabolomic and network analysis to explore the potential mechanism of three chemical elicitors in rapamycin overproduction, Microorganisms 10 (11) (2022), 2205, https://doi.org/10.3390/microorganisms10112205.

[81] N.S. Lyons, A.N. Bogner, J.J. Tanner, P. Sobrado, Kinetic and structural characterization of a flavin-dependent putrescine N-hydroxylase from *Acinetobacter baumannii*, Biochemistry 61 (22) (2022) 2607–2620, https://doi.org/10.1021/acs.biochem.2c00493.

[82] G. Speciale, Y. Jin, G.J. Davies, S.J. Williams, E.D. Goddard-Borger, YihQ is a sulfoquinovosidase that cleaves sulfoquinovosyl diacylglyceride sulfolipids, Nat. Chem. Biol. 12 (4) (2016 Apr) 215–217, https://doi.org/10.1038/nchembio.2023.

[83] W.S. Somers, M.L. Stahl, F.X. Sullivan, GDP-fucose synthetase from Escherichia coli: structure of a unique member of the short-chain dehydrogenase/reductase family that catalyzes two distinct reactions at the same active site, Structure 15 (6) (1998) 1601–1612, https://doi.org/10.1016/s0969-2126(98)00157-9, 12.

[84] C.H. Williams, T.J. Stillman, V.V. Barynin, S.E. Sedelnikova, Y. Tang, J. Green, J. R. Guest, P.J.E. Artymiuk, *coli* aconitase B structure reveals a HEAT-like domain with implications for protein-protein recognition, Nat. Struct. Biol. 9 (6) (2002) 447–452, https://doi.org/10.1038/nsb801.

[85] M. Totir, N. Echols, M. Nanao, C.L. Gee, A. Moskaleva, S. Gradia, A.T. Iavarone, J. M. Berger, A.P. May, C. Zubieta, T. Alber, Macro-to-micro structural proteomics: native source proteins for high-throughput crystallization, PLoS One 7 (2) (2012), e32498, https://doi.org/10.1371/journal.pone.0032498.

[86] A. Hadfield, G. Kryger, J. Ouyang, G.A. Petsko, D. Ringe, R. Viola, Structure of aspartate-beta-semialdehyde dehydrogenase from Escherichia coli, a key enzyme in the aspartate family of amino acid biosynthesis, J. Mol. Biol. 18 (289) (1999) 991–1002, https://doi.org/10.1006/jmbi.1999.2828, 4.

[87] T. Mukherjee, Y. Zhang, S. Abdelwahed, S.E. Ealick, T.P. Begley, Catalysis of a flavoenzyme-mediated amide hydrolysis, J. Am. Chem. Soc. 28 (132) (2010) 5550–5551, https://doi.org/10.1021/ja9107676, 16.

[88] M. Šudomová, M.A. Shariati, J. Echeverría, I. Berindan-Neagoe, S.M. Nabavi, S.T. S. Hassan, A Microbiological, Toxicological, and Biochemical Study of the Effects of Fucoxanthin, a Marine Carotenoid, on *Mycobacterium tuberculosis* and the enzymes implicated in its cell wall: a link between mycobacterial infection and autoimmune diseases, Mar. Drugs 17 (11) (2019), 641, https://doi.org/10.3390/MD17110641.

[89] L.E. Kemp, C.S. Bond, W.N. Hunter, Structure of a tetragonal crystal form of Escherichia coli 2-C-methyl-D-erythritol 4-phosphate cytidylyltransferase, Acta Crystallogr. D Biol. Crystallogr. 59 (3) (2003) 607–610, https://doi.org/10.1107/s090744490202365x.

[90] B. Pandey, S. Grover, J. Kaur, A. Grover, Analysis of mutations leading to para-aminosalicylic acid resistance in Mycobacterium tuberculosis, Sci. Rep. 9 (1) (2019), 13617, https://doi.org/10.1038/s41598-019-48940-5.

[91] A.A. Lopez-Zavala, Guevara-Hernandez, L.H. Vazquez-Lujan, To Sanchez-Paz, K. D. Garcia-Orozco, C.A. Contreras-Vergara, G. Lopez-Leal, A.A. Arvizu-Flores, To Ochoa-Leyva, R.R. Sotelo-Mundo, A novel thymidylate synthase from the *Vibrionales*, Alteromonadales, *Aeromonadales*, and *Pasteurellales* (VAAP) clade with altered nucleotide and folate binding sites, PeerJ 6 (2018), e5023, https://doi.org/10.7717/peerj.5023.

[92] L.A. Alexandrova, A.L. Khandazhinskaya, E.S. Matyugina, D.A. Makarov, S. N. Kochetkov, Analogues of pyrimidine nucleosides as mycobacteria growth inhibitors, Microorganisms 10 (7) (2022), 1299, https://doi.org/10.3390/microorganisms10071299.

[93] N.G. Biteau, V. Roy, C. Nicolas, H.F. Becker, J.C. Lambry, H. Myllykallio, L. A. Agrofoglio, Synthesis and structure-activity relationship studies of pyrido [1,2-e]Purine-2,4(*1H,3H*)-Dione derivatives targeting *flavin-dependent thymidylate synthase* in *Mycobacterium tuberculosis*, Molecules 27 (19) (2022), 6216, https://doi.org/10.3390/molecules27196216.

[94] M.S. El-Shoukrofy, To Atta, S. Fahmy, D. Sriram, M.A. Mahran, I.M. Labouta, New tetrahydropyrimidine-1,2,3-triazole clubbed compounds: antitubercular activity and Thymidine Monophosphate Kinase (TMPKmt) inhibition, Bioorg. Chem. 131 (2023), 106312, https://doi.org/10.1016/j.bioorg.2022.106312.

[95] Y. Jian, R. Merceron, S. De Munck, H.E. Forbes, F. Hulpia, M.D.P. Risseeuw, K. Van Hecke, S.N. Savvides, H. Munier-Lehmann, H.I.M. Boshoff, S. Van Calenbergh, Endeavors towards transformation of M. *tuberculosis* thymidylate kinase (MtbTMPK) inhibitors into potential antimycobacterial agents, Eur. J. Med. Chem. 206 (2020), 112659, https://doi.org/10.1016/j.ejmech.2020.112659.

[96] K. Suthagar, W. Jiao, H. Munier-Lehmann, A.J. Fairbanks, Synthesis of sulfamide analogues of deoxythymidine monophosphate as potential inhibitors of mycobacterial cell wall biosynthesis, Carbohydr. Res. 457 (2018) 32–40, https://doi.org/10.1016/j.carres.2018.01.001.

[97] S. Cussotto, C.R. Strain, F. Fouhy, R.G. Strain, V.L. Peterson, G. Clarke, C. Stanton, T.G. Dinan, J.F. Cryan, Differential effects of psychotropic drugs on microbiome composition and gastrointestinal function, Psychopharmacology (Berl) 236 (5) (2019) 1671–1685, https://doi.org/10.1007/s00213-018-5006-5.

[98] J.G. Arnez, D.C. Harris, A. Mitschler, B. Rees, C.S. Francklyn, D. Moras, Crystal structure of histidyl-tRNA synthetase from *Escherichia coli* complexed with histidyl-adenylate, EMBO J. 14 (17) (1995) 4143–4155, https://doi.org/10.1002/j.1460-2075.1995.tb00088.x.

[99] R.E. Campbell, S.C. Mosimann, M.E. Tanner, N.C. Strynadka, The structure of UDP-N-acetylglucosamine 2-epimerase reveals homology to phosphoglycosyl transferases, Biochemistry 39 (49) (2000) 14993–15001, https://doi.org/10.1021/bi001627x.

[100] J. Ou, P. Elizalde, H.B. Guo, H. Qin, B.T.D. Tobe, J.S. Choy, TCA and SSRI antidepressants exert selection pressure for efflux-dependent antibiotic resistance mechanisms in *Escherichia coli*, mBio 13 (6) (2022), E0219122, https://doi.org/10.1128/mbio.02191-22.

[101] V.V. Jadhav, J. Han, Y. Fasina, S.H. Harrison, Connecting gut microbiomes and short chain fatty acids with the serotonergic system and behavior in *Gallus gallus* and other avian species, Front. Physiol. 13 (2022 Nov 2), 1035538, https://doi.org/10.3389/fphys.2022.1035538.

[102] K. Dinan, T. Dinan, Antibiotics and mental health: the good, the bad and the ugly, J. Intern. Med. 292 (6) (2022) 858–869, https://doi.org/10.1111/joim.13543.

[103] Y. Gu, C. Wang, X. Qin, B. Zhou, X. Liu, T. Liu, R. Xie, J. Liu, B. Wang, H. Cao, *Saccharomyces boulardii*, a yeast probiotic, inhibits gut motility through upregulating intestinal serotonin transporter and modulating gut microbiota, Pharmacol. Res. 181 (2022), 106291, https://doi.org/10.1016/j.phrs.2022.106291.

[104] F.J. Navarro-Triviño, P. Jaime Ruiz, To Porras Segovia, V. Garrido Torres-Puchol, Oral Isotretinoin for the treatment of Aripiprazole-induced acneiform rash, Dermatol. Ther. 31 (4) (2018), E12637, https://doi.org/10.1111/dth.12637.

[105] P. Ding, J. Lu, Y. Wang, M.A. Schembri, J. Guo, Antidepressants promote the spread of antibiotic resistance via horizontally conjugative gene transfer, Environ. Microbiol. 24 (11) (2022) 5261–5276, https://doi.org/10.1111/1462-2920.16165.

[106] D. Shi, H. Hao, Z. Wei, D. Yang, J. Yin, H. Li, Z. Chen, Z. Yang, T. Chen, S. Zhou, H. Wu, J. Li, M. Jin, Combined exposure to non-antibiotic pharmaceutics and antibiotics in the gut synergistically promote the development of multi-drug-resistance in *Escherichia coli*, Gut Microb. 14 (1) (2022), 2018901, https://doi.org/10.1080/19490976.2021.2018901.

[107] Y. Kyono, L. Ellezian, Y. Hu, K. Eliadis, J. Moy, E.B. Hirsch, M.J. Federle, S. A. Flowers, The atypical antipsychotic quetiapine promotes multiple antibiotic resistance in *Escherichia coli*, J. Bacteriol. 204 (5) (2022), E0010222, https://doi.org/10.1128/jb.00102-22.

[108] Y. Wang, Z. Yu, P. Ding, J. Lu, L. Mao, L. Ngiam, Z. Yuan, J. Engelstädter, M. A. Schembri, J. Guo, Antidepressants can induce mutation and enhance persistence toward multiple antibiotics, Proc. Natl. Acad. Sci. U.S.A. 120 (5) (2023), E2208344120, https://doi.org/10.1073/PNAS.2208344120.

[109] Z.I. Kirkwood, B.C. Millar, D.G. Downey, J.E. Moore, Antimycobacterial activity of nonantibiotics associated with the polypharmacy of cystic fibrosis (CF) against *Mycobacterium abscessus*, Int. J. Mycobacteriol. 7 (4) (2018) 358–360, https://doi.org/10.4103/ijmy.ijmy_142_18.

[110] I. Amino, T. Maeda, M. Aoki, H. Kinoshita, To Muroya, Y. Taji, K. Kobayashi, D. Okamura, Y. Murayama, N. Asou, et al., Therapy-related acute promyelocytic leukemia developing during chemotherapy for thymic carcinoma, Rinsho Ketsueki 61 (8) (2020) 874–878, https://doi.org/10.11406/rinketsu.61.874. Japanese.

[111] G. Ullah, F. Ali, M.M. Paracha, S.M. Noor, J. Naeem, Comparison of efficacy of topical adapalene plus oral azithromycin and topical adapalene plus oral doxycycline in treatment of *acne vulgaris*, J. Ayub Med. Coll. Abbottabad 34 (4) (2022) 782–785, https://doi.org/10.55519/JAMC-04-9568.

[112] N. Auffret, J.P. Claudel, M.T. Leccia, F. Ballanger, B. Dreno, Novel and emerging treatment options for *acne vulgaris*, Eur. J. Dermatol. 32 (4) (2022) 451–458, https://doi.org/10.1684/ejd.2022.4306. English.

[113] S. Shailaja, N. Harshitha, To Fasim, S.S. More, S. Das Mitra, Identification of a potential inhibitor for New Delhi metallo- â-lactamase 1 (NDM-1) from FDA

approved chemical library- a drug repurposing approach to combat carbapenem resistance, J. Biomol. Struct. Dyn. (2022) 1–12, https://doi.org/10.1080/07391102.2022.2123402.

[114] J. Hernandez Maldonado, O. Grundmann, Drug-drug interactions of artemisinin-based combination therapies in malaria treatment: a narrative review of the literature, J. Clin. Pharmacol. 62 (10) (2022) 1197–1205, https://doi.org/10.1002/jcph.2073.

[115] Y. Xu, L. Xiao, J. Chen, Q. Wu, W. Yu, W. Zeng, Y. Shi, Y. Lu, Y. Liu, á-Fe 2O3 based nanotherapeutics for near-infrared/dihydroartemisinin dual-augmented chemodynamic antibacterial therapy, Acta Biomater. 150 (2022) 367–379, https://doi.org/10.1016/j.actbio.2022.07.047.

[116] C. Ding, F. Zhang, Y. Gao, Y. Li, D. Cheng, J. Wang, L. Mao, Antibacterial photodynamic treatment of *Porphyromonas gingivalis* with toluidine blue O and a NonLaser red light source enhanced by dihydroartemisinin, Photochem. Photobiol. 97 (2) (2021) 377–384, https://doi.org/10.1111/php.13333.

[117] M. Huang, J. Luo, J.Y. Shen, Synergistic antibacterial effect and mechanisms of dihydroartemisinin and cefuroxime incombination, Zhongguo Zhongyao Zazhi 45 (12) (2020) 2975–2981, https://doi.org/10.19540/j.cnki.cjcmm.20200225.402. Chinese.

[118] K. Kalani, V. Chaturvedi, P. Trivedi, S. Tondon, S.K. Srivastava, Dihydroartemisinin and its analogs: a new class of antitubercular agents, Curr. Top. Med. Chem. 19 (8) (2019) 594–599, https://doi.org/10.2174/1568026619666190304142802.

[119] A.K. Singh, K.S. Prajapati, S. Kumar, Acarbose potentially binds to the type I peptide deformylase catalytic site and inhibits bacterial growth: an *in silico* and *in vitro* study, Curr. Pharmaceut. Des. 28 (35) (2022) 2890–2900, https://doi.org/10.2174/1381612828666220922100556.

[120] J.N. Nyemb, R.T. Tchuenguem, To Venditti, A.T. Tchinda, C. Henoumont, E. Size, S. Laurent, J. Iqbal, Antimicrobial and α-glucosidase inhibitory activities of chemical constituents from *Gardenia aqualla* (Rubiaceae), Nat. Prod. Res. 36 (24) (2022) 6369–6374, https://doi.org/10.1080/14786419.2022.2031187.

[121] S.B. Liu, L. Zeng, Q.L. Xu, Y.L. Chen, T. Lou, S.X. Zhang, J.W. Tan, Polycyclic phenol derivatives from the leaves of *Spermacoce latifolia* and their antibacterial and α-glucosidase inhibitory activity, Molecules 27 (10) (2022), 3334, https://doi.org/10.3390/molecules27103334.

[122] M.S.M. Abd El Hafez, M.A.E. Aziz Okbah, H.A.H. Ibrahim, A.A.E.R. Hussein, N.A. A. El Moneim, A. Ata, First report of steroid derivatives isolated from starfish *Acanthaster planci* with anti-bacterial, anti-cancer and anti-diabetic activities, Nat. Prod. Res. 36 (21) (2022) 5545–5552, https://doi.org/10.1080/14786419.2021.2021200.

[123] J. Balaich, M. Star, G. Wu, P.D. Jeffrey, To Biswas, L. Zhao, To Korennykh, M. S. Donia, The human microbiome encodes resistance to the antidiabetic drug acarbose, Nature 600 (7887) (2021) 110–115, https://doi.org/10.1038/s41586-021-04091-0.

[124] D. Fang, T. Xu, J. Sun, J. Shi, F. Li, Y. Yin, Z. Wang, Y. Liu, Nicotinamide mononucleotide ameliorates sleep deprivation-induced gut microbiota dysbiosis and restores colonization resistance against intestinal infections, Adv. Sci. (2023), E2207170, https://doi.org/10.1002/advs.202207170.

[125] L. Zhang, Y. Fan, L. Galantini, K. Schillén, To Del Giudice, G. Du, Y. Wang, Noncovalent bile acid oligomers as facial amphiphilic antimicrobials, Langmuir 39 (1) (2023) 495–506, https://doi.org/10.1021/acs.langmuir.2c02787.

[126] R. Singh, D. Jha, H.K. Gautam, P. Kumar, Supramolecular self-assemblies of engineered polyethylenimines as multifunctional nanostructures for DNA transportation with excellent antimicrobial activity, Bioorg. Chem. 106 (2021), 104463, https://doi.org/10.1016/j.bioorg.2020.1.