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# UNLOCKING POTENTIAL: EXPLORING DRUG REPURPOSING STRATEGIES FOR THERAPEUTIC INNOVATION

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**Abstract:** Drug repositioning or repurposing is playing a significant role in advancing new therapeutic innovations, complementing traditional drug discovery approaches to enhance therapeutic efficacy. The repurposing or repositioning of drugs involves considerations related to the drugs which are not in use or have shown more than one or other therapeutic effects, marking the commencement of a new era for pharmaceuticals and exploring new market opportunities and value. It has given a reoccurrence or a new opportunity to all the drugs which are currently undergoing clinical trials or those that have been withdrawn from the market. This approach aims to overcome the limitations of traditional methods, resources, and time. Despite economic and regulatory challenges, drug repositioning has played a crucial role in various pathological fields, public health benefits, and commercial value. Notable examples of drugs that have undergone repositioning include sildenafil, dimethylfumarate, acetylsalicylic acid, and thalidomide. The history of repositioned drugs

has provided solutions for various pathologies. Drug repositioning is an emerging approach that has gained significant interest. Nowadays, it has become a more rational method for identifying new drug candidates for conditions such as type 2 diabetes, cancer, and rheumatoid arthritis and many more.

**Keywords:** T2DM , Drug repositioning , innovations , pathologies , Repurposing

## Introduction

The conventional process of drug development requires a significant amount of time and resources before a molecule can be brought to the market. Even with substantial investments, the likelihood of a lead molecule making it to the market is frequently low..(2) The drug companies and researchers have been disheartened by the prolonged time and increased costs associated with finding an appropriate drug or novel drug delivery system. This has led to a loss of hope in their endeavors. Furthermore, only a few numbers of drug design projects are completed, and most of them fail during different rigorous development phases(3).

The concept of drug repurposing or repositioning has emerged as a novel strategy that expedites the drug discovery process, garnering significant interest from researchers across various scientific domains. This innovative approach has propelled the drug development trajectory, enabling scientists to explore new therapeutic possibilities for existing drugs. (1). This method provides significant benefits to the entire medical industry, facilitating the development of new drug approaches across various pharmaceutical sectors. It streamlines the process and enhances efficiency in the field of medicine, making it easier for professionals to innovate and create novel solutions for different health issues

The objectives of drug repurposing vary depending on their intended application. In the case of a pharmaceutical company, a particular objective could involve the discovery of a highly effective drug that revolutionizes the standard treatment for a widespread condition like hypertension, thereby benefiting millions of individuals. (4). Repurposing refers to a strategic approach that involves utilizing the untapped potential of a drug or substance for purposes other than its original intended use. This process involves identifying alternative applications or benefits that can be derived from an underutilized drug, thereby maximizing its value and potential impact. By repurposing a drug, it can be effectively redirected towards serving the

needs and objectives of other therapeutic purposes, ultimately enhancing its overall value and utility.

Drug repurposing, also referred to as drug repositioning or drug rediscovery, involves the identification and exploration of existing drugs for alternative therapeutic purposes that go beyond their initial indications. Rather than creating entirely new drugs from the beginning, drug repurposing seeks to utilize the existing understanding and safety profiles of approved or investigational drugs to address medical needs that have not been met or to explore innovative treatment options for different diseases. By repurposing drugs, researchers can potentially save time and resources while still providing potential benefits to patients.

The exploration of alternative applications for pre-existing drugs, known as drug repurposing or drug repositioning, involves identifying new uses for medications that are already available. Although this endeavor presents various challenges, it holds immense potential in terms of reducing the expenses associated with drug development and enhancing its safety measures. (6). Several instances of serendipity-based drug repurposing can be observed in the field of medicine. One such example is thalidomide, which was initially created to alleviate morning sickness in pregnant women but is now utilized in the treatment of multiple myeloma. Another instance is sildenafil, which was originally intended for the management of angina and hypertension but is now commonly prescribed for erectile dysfunction. Additionally, amantadine, an antiviral medication initially indicated for influenza, has found application in the treatment of Parkinson's disease. These examples highlight the unexpected discoveries and benefits that can arise from repurposing drugs for different medical conditions.The primary benefit of drug repurposing lies in its ability to accelerate the traditional drug discovery process by identifying alternative clinical uses for drugs that have already been deemed safe and effective in humans, and have obtained regulatory approval for different indications. (7). Repurposing has several implications in the drug regulatory setting as well as in the scientific setting, especially if it occurs during a public health emergency.

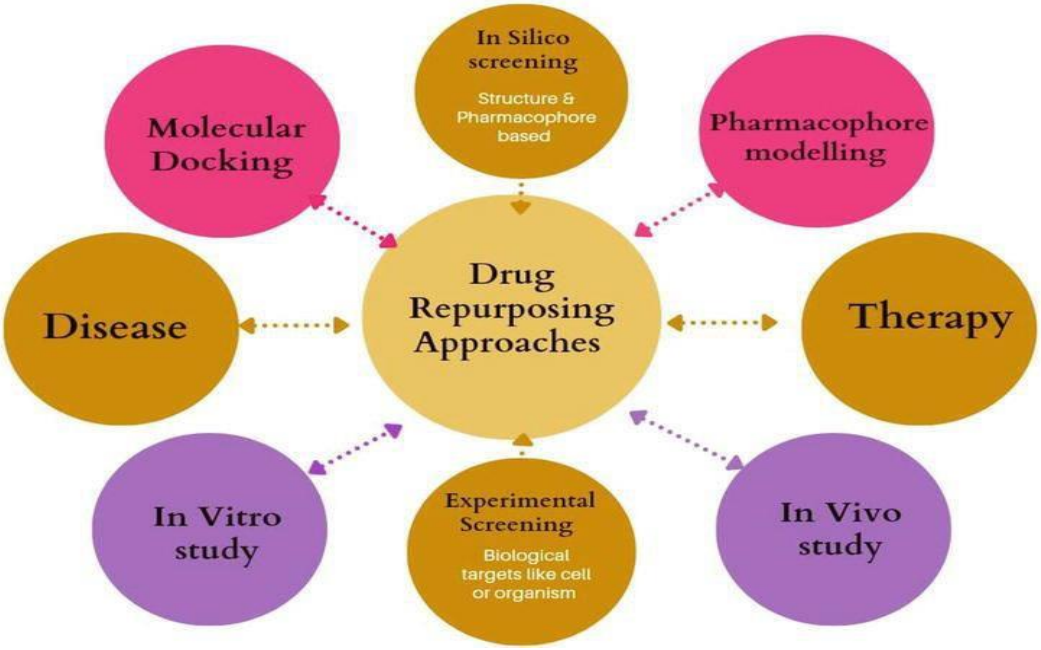


Figure 1: Different approaches in drug repurposing

**Drug repurposing as a new approach**

Drug repurposing approach puts the drug discovery process in fast track, and has been gaining attention from the researchers in wide range of scientific fields(8). The utilization of in-vitro and in-vivo screening data, comprehensive chemical optimization, toxicity studies, bulk manufacturing, formulation development, and pharmacokinetic profiles of FDA-approved drugs has led to the reduction of drug development cycles by allowing the bypassing of these essential steps. Drug repurposing involves the identification of new applications for existing marketed drugs or well-characterized compounds, even if they have previously failed in clinical trials.. A drug discovery program has been developed that offers a faster and safer method for creating medications to combat diseases and disorders that currently have no potential treatment options. In recent years, approximately 30% of newly FDA approved drugs and vaccines have been successfully developed through the drug repurposing approach. This approach has garnered significant interest from pharmaceutical companies due to its ability to bypass the lengthy initial development period of six to 10 years typically required for new drugs. Furthermore, many phases of de novo drug discovery and development can be skipped, as the re-purposed candidates have already undergone clinical and pre-clinical studies for their original indications. Consequently, this approach effectively minimizes the duration and expenses required to penetrate the market, while simultaneously mitigating the inherent risks associated with research and development initiatives. Additionally, the likelihood of encountering clinical

setbacks is significantly diminished. Furthermore, this strategy presents an advantageous opportunity to expand the market reach and extend the duration of a drug's patent protection, thereby maximizing its profitability (9-10 )

Few examples of the repurposed drugs on different backgrounds are mentioned below:-

- A medication that was initially approved for a particular purpose, but was later discovered to have more effective therapeutic benefits for a different condition, is a prime example of serendipitous drug discovery. One such instance is the case of Sildenafil, which was originally formulated to treat hypertension and angina pectoris, but is now widely used for the management of erectile dysfunction, leading to its significant success in the market.
- The investigation involves the evaluation of FDA-approved medications for their potential off-label applications. Itraconazole, originally authorized as an antifungal treatment, has been discovered to exhibit an extra attribute of inhibiting angiogenesis, the formation of new blood vessels..
- Implication of failed investigational drugs for new therapeutic areas. Saracatinib, a failure drug developed by AstraZeneca as an anticancer agent exhibited substantial reversal of symptoms in Alzheimer's Disease (AD) mice mode.

**Approaches for Drug -repurposing**

The drug repurposing strategy involves the utilization of drug-related information such as drug targets, chemical structures, pathways, adverse effects, and more. Through this approach, models are constructed to predict unknown targets, biomarkers, or mechanisms for diseases. This strategy encompasses target-based, pathway-based, and target mechanism-based drug repurposing methods. (10 ). Repurpose is a compendium of drugs (small molecules and biotech or protein drugs) and their associated primary and secondary diseases in which the compound was indicated as effective. Analyzing these datasets through enrichment analysis provided valuable insights into essential biological pathways, functional mechanisms, physicochemical characteristics, and potential side effects linked to effectively repurposed medications. This knowledge can be instrumental in enhancing the design of future drug repositioning studies. (11 )

**Strategies for drug repurposing :**

- **Knowledge-based repurposing:**

In this approach to repurposing drugs, data related to drugs such as drug targets, chemical structures, pathways, adverse effects, and more is leveraged to develop models that can forecast unidentified targets, biomarkers, or disease mechanisms. (12 ).This strategy includes target-based, pathway-based, and target mechanism based drug-repurposing(13).

- **Target-based drug-repurposing :**

Target-based drug repurposing involves the utilization of proteins or biomarkers of interest to conduct high-throughput or high-content screening (HTS/HCS) of drug compounds. This approach aims to identify potential therapeutic agents by assessing their interaction with specific targets or biomarkers, which may lead to the repurposing of existing drugs for new indications or the discovery of novel drug candidates. By employing HTS/HCS techniques, researchers can efficiently screen a large number of drug compounds against the desired targets, enabling the identification of promising candidates for further development and evaluation. This strategy offers a cost-effective and time-efficient approach to drug discovery and development, potentially accelerating the availability of new treatments for various diseases (14 ) In silico screening of drug compounds from drug libraries, such as ligand-based screening or docking, is followed by. However, target-based repurposing, unlike blinded search or screening, directly connects targets with disease mechanisms. This approach utilizes biological and pharmacological information during screening, leading to a higher probability of drug discovery (15) .

The advantage of the target-based approach lies in its ability to screen nearly all drug compounds with known chemical structure. However, target-based methods cannot identify unknown mechanisms beyond the targets already known.

- **Pathway-based drug-repurposing:**

  The strategy of pathway-based drug repurposing relies on the integration of metabolic pathways, signaling pathways, and protein-interaction networks to ascertain the relationship or likeness between a disease and a drug. By leveraging omics data derived from either human patients or animals, it becomes possible to reconstruct disease-specific pathways, which can then be utilized as potential targets for repurposing existing drugs

- **Target mechanism-based drug-repurposing:**

The process of drug repurposing through target mechanism-based approaches involves the integration of signaling pathway information, treatment omics data, and protein interaction networks. This comprehensive approach enables the identification of novel mechanisms of action for existing drugs.

The growing significance of precision medicine has further emphasized the need for such repurposing strategies. One notable advantage of these approaches is their ability to uncover mechanisms that are not only associated with diseases or drugs but also with the specific treatment of certain diseases.(15 ).

**Methods in drug repouposing –**

- **In-silico Screening** :Computational algorithms and databases are employed to forecast potential interactions between drugs and targets. Techniques utilized encompass virtual screening, molecular docking, and similarity-based strategies..
- **Computational Approaches** :- Data structures known as networks are versatile and uncomplicated, enabling the identification of various connections using statistical and computational methods. The utilization of biological networks has become prevalent in illustrating molecular relationships and simulating interactions among biological components. A thorough review of literature was carried out across multiple databases such as ABI/Informa, Academic Search Premier, Business Source Complete, Cochrane Library, EconLit, Google Scholar, Ovid Embase, Ovid Medline, Pubmed, Scopus, and Web of Science Core Collection to locate pertinent article.
- **Pubmed :** PubMed, the leading research database for the health sciences, is a product of the National Center for Biotechnology Information (NCBI) situated at the National Library of Medicine (16 ). Health care researchers, practitioners, and students worldwide utilize PubMed as a valuable resource. In the beginning of 2020, the new PubMed website was launched, providing users with access to a range of information sources including MEDLINE, PubMed Central, and a selection of freely available e-books on various health sciences subjects. (17). The updated version of PubMed places a strong emphasis on conducting searches with minimal planning or search construction beforehand. The training resources for the new PubMed platform advise against utilizing special characters or Boolean operators when formulating the initial search query..

The basic search function on PubMed promotes the use of automated term mapping and presents results based on the best match, making the search process more akin to that of

popular general search engines. The resources offered by NCBI, including PubMed, MEDLINE, and MeSH, are invaluable tools for both educational and research purposes in the health sciences field, offering users the opportunity for more intricate exploration of information.

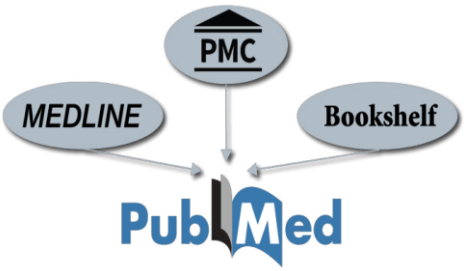


Figure 2: Pubmed Software

**Docking:**

Molecular Docking is a computational technique used to predict the optimal positioning of a ligand in relation to a receptor (typically a protein) to form a stable complex. This preferred positioning is crucial in estimating the strength of interaction or binding affinity between the ligand and the protein, which is achieved through the utilization of scoring functions. The process of docking is commonly employed to forecast the binding orientation of potential drug molecules with protein targets, enabling the prediction of drug affinity and activity. (18).

Protein-ligand or protein-protein docking is a crucial aspect of contemporary drug discovery, as it aids in the prediction of the ligand's orientation upon binding to a protein receptor or enzyme. This prediction is based on shape and electrostatic interactions, which are used to quantify the binding process. In addition to Coulombic interactions and the formation of hydrogen bonds, van der Waals interactions also contribute significantly to this process. The cumulative effect of all these interactions is estimated through a docking score, which serves as an indicator of the binding potential. (19).

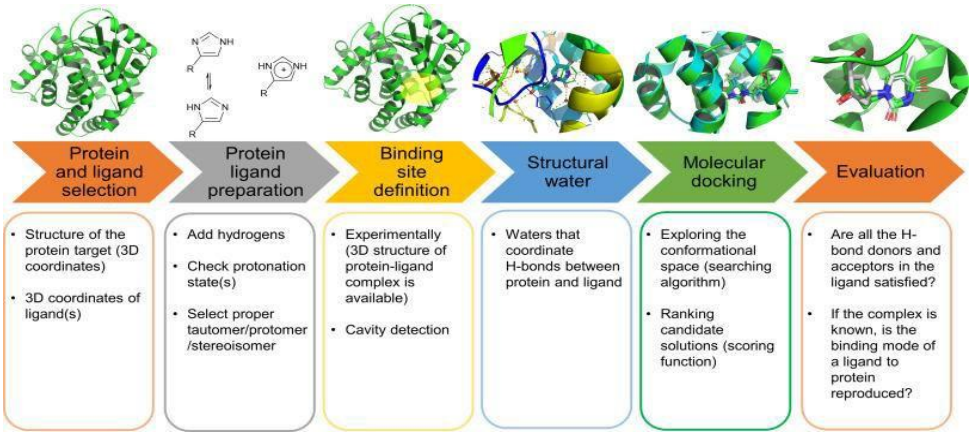


**Figure 3: Different Stages In Drug Repourpposig Approach**

## Protein data bank :

The Protein Data Bank (PDB) serves as the primary worldwide database housing structural information on essential biological molecules. Its inception in 1971 aimed to collect experimental findings from the emerging field of macromolecular crystallography, which unveiled intricate three-dimensional structures of vital macromolecules such as proteins, DNA, and RNA at the atomic level (20). The PDB archive is managed jointly by theWorldwide Protein Data Bank partnership (wwPDB;wwpdb.org),consisting of the RCSB Protein DataBankProtein Data Bank Japan (PDBj),theProtein Data Bank in Europe (PDBe),and BioMa-gResBank(BMRB)(21 ).

### Autodock vina :

Molecular docking has become a powerful tool for lead discovery and optimization process( 22 ). Docking programs typically rely on two main components: a scoring function and an exploration method. The scoring function is responsible for estimating the free energy of the modeled system, while the exploration method is used to sample the positional and conformational space. One example of a docking program is AutoDock Vina, which was developed by Trott and Olson at the Scripps Research Institute in La Jolla, California. AutoDock Vina is a successor to previous versions of AutoDock and is just one of many protein-ligand docking programs available. It was initially published in 1997 and has since undergone improvements. AutoDock Vina is released under a free software license, specifically the GNU GPL since 2007. The Vina scoring function is entirely empirical and incorporates various elements such as Gaussian steric interaction terms, a finite repulsion term, piecewise linear hydrophobic and hydrogen-bond interaction terms, and an entropic term that is proportional to the number of rotatable bonds.( 23 ). Furthermore, AutoDock Vina introduces a novel search algorithm and a hybrid scoring function that merges empirical and knowledge-based scoring functions. The program's capacity to utilize multiple cores, coupled with its high performance and improved accuracy, along with its user-friendly interface and open accessibility, have all contributed to its rapid adoption within the docking community, as evidenced by the substantial number of citations in the original publication. (24).Various methods can be employed for docking procedures, with the Lamarckian genetic algorithm (LGA) being recognized as one of the most effective techniques. Genetic algorithms and simulated annealing are considered traditional approaches to docking. In practice,

AutoDock is executed multiple times to generate several docked conformations, and the analysis of predicted energy values and result consistency is crucial in determining the optimal solution.AutoDock Tools encompass of different techniques for analyzing the results of docking simulations, comprising tools for clustering results by conformational similarity,

visualizing confirmations, ligand protein interactions, and affinity potentials created by AutoGrid.

**Uses:-**

- Identification of aromatic rings.

- Used to explore the conformational states of a flexible ligand, using the maps generated by AutoGrid to evaluate the ligand-protein interaction at each point in the docking simulation.

**High throughput screening :**

High throughput screening is commonly defined as automatic testing of potential drug candidates at a rate in excess of 10,000 compounds per week( 25 ). The objective of high throughput drug discovery involves screening extensive compound libraries to identify potentially active compounds (referred to as 'hits'), which can then be further developed into compounds for pre-clinical testing (known as 'leads'). Additionally, high-throughput screening techniques are utilized to analyze metabolic, pharmacokinetic, and toxicological information related to novel drugs.

| No. | Programs | Application | Accessibility | Reference |
|---|---|---|---|---|
| 1. | AutoDock | It is employed in molecular docking. It predicts the binding capacity of a tiny chemical and assigns a target protein to a 3D structure | https://autodock.scripps.edu/ | [74] |
| 2. | LPCCSU | Based on a comprehensive investigation of interatomic interactions and interface complementarity | https://oca.weizmann.ac.il/oca-bin/lpccsu | [75] |
| 3. | PatchDock | The method performs rigid docking, with surface variability | https://bioinfo3d.cs.tau.ac.il/PatchDock/php.php | [76] |
| 4. | Hex | For docking studies | http://hex.loria.fr/ | [77] |
| 5. | Glide Schrodinger | Comprehensive molecular modeling and computer-aided drug development (CADD) tool | https://www.schrodinger.com/ | [78] |
| 6. | Molecular operating environment | Comprehensive molecular modeling and computer-aided drug development (CADD) tool | https://www.chemcomp.com/ | [79] |
| 7. | DockingServer | A user-friendly web-based interface that manages all elements of molecular docking. | https://www.dockingserver.com/web | [80] |
| 8. | SwissDock | A web service for predicting a protein's association with a small molecule ligand. | http://www.swissdock.ch/ | [81] |

The process of developing a new drug molecule from its initial concept to the creation of a final product is a complex undertaking that may span 12 to 15 years and require an investment of over $1 billion. Typically, drug discovery entails the exploration and evaluation of potential drug candidates against specific therapeutic targets that have been predetermined. (26). To expedite the process of drug discovery, a target-centric library is employed. This library comprises a range of compounds specifically designed to interact with a particular protein target or a group of related targets, such as kinases, voltage-gated ion channels, and serine/cysteine proteases.

This approach proves to be beneficial in accelerating the discovery of new drugs. (27). Generally, HTS involves an automated operationplatform, modern robotics, highly sensitive detection methods, sophisticated control software, specific

screening model (in vitro), an abundant components library, advanced liquid handling and a data acquisition and processing system(28)

A strong increase in the number of chemical compounds for testing and the concomitant increase in the number of molecular targets for lead finding can be accommodated only via substantial miniaturization of HTS assays(29 ). Enzymes, including kinases, proteases, phosphatases, oxidoreductases, phosphodiesterases, and transferases, are the primary focus of current lead discovery efforts in biochemistry. In terms of cell-based targets, certain G-protein-coupled receptors (GPCRs), nuclear hormone receptors, and specific types of voltage- and ligand-gated ion channels (such as Ca2+ channels) are particularly suitable for screening extensive compound collections using modern screening technologies. Despite the vast number of human genes (over 25,000) and the even larger number of gene variants and proteins (over 100,000), the number of approved drugs targeting these molecular targets remains relatively limited, with only 324 targets having approved drugs.. The explanation for this discrepancy can be manifold: some targets might not be feasible at all for modulation via low molecular weight compounds.Others, however, might simply be not approachable by current technologies and therefore constitute not only a great challenge(30).

Hit and lead generation are key processes involved in the creation of successful new medicinal entities, and it is the quality of information content imparted through their exploration and refinement that largely determines their fate in the later stages of clinical development.

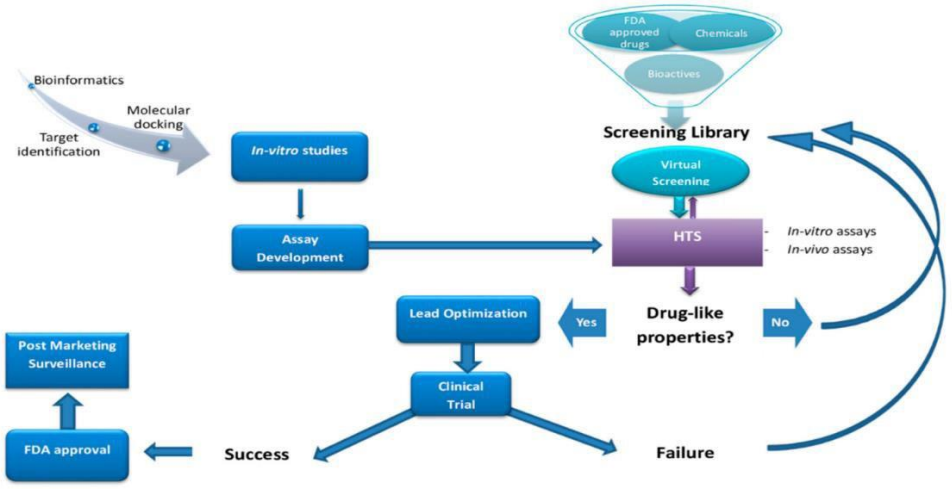


Figure 4: Different Stages In Drug Reporposing of Studies for a new Compound

**Molegro :-**

Molecular docking serves as a computational method utilized for the purpose of predicting the binding mode and affinity of small molecules, including potential drug candidates, to target proteins or biomolecules. The MolDock technique is founded upon a novel heuristic search algorithm that effectively combines differential evolution with a cavity prediction algorithm.

In order to enhance the precision of docking, a re-ranking scoring function is introduced, which aids in identifying the most promising docking solution among the solutions generated by the docking algorithm. The accuracy of MolDock has been thoroughlyassessed by docking flexible ligands to a total of 77 protein targets. Remarkably, MolDock successfully identified the correct binding mode in 87% of the complexes. ( 31). Molegro Virtual Docker (MVD) is a software tool designed for molecular docking simulations in the field of computational chemistry and drug discovery

MolDock Score:- MolDock Score [Grid]‟ is a grid based version of the MolDock Score function. It precalculates potential-energy values on an evenly spaced cubic grid inorder to speed up calculations. he energy potential is Optimizer) used in MVD is based on an evolutionary algorithm.

**Uses** :-

a) MVD facilitates virtual screening campaigns by docking large libraries of compounds against target proteins to identify potential drug candidates.
b) MVD aids in lead optimization efforts by analyzing and refining the binding interactions between lead compounds and target proteins.
c) MVD supports structure-based drug design approaches by utilizing the three-dimensional structure of target proteins to guide the rational design of new drug molecules.
d) MVD can be used for protein-protein docking simulations, where it predicts the three-dimensional structure of protein complexes based on the structures of individual protein components.

**GOLD (Genetic Optimization for Ligand Docking)**

GOLD (Genetic Optimization for Ligand Docking) is a genetic algorithm designed for the docking of flexible ligands and proteins containing flexible hydroxyl groups. The software employs a scoring function that incorporates favorable conformations identified in the Cambridge Structural Database, as well as empirical data on weak chemical interactions.

Various genetic algorithm parameters are adjusted to manage the trade-off between the speed of GOLD and the accuracy of its predictions (32). It gives reliable results and correct atom typing for both protein and ligand.

GOLD Suite software comprises three software components: GOLD, Hermes, and GoldMine. GOLD is responsible for facilitating the docking of ligands into protein binding sites using prepared input files. Hermes, on the other hand, serves as a tool for preparing input files for docking with GOLD, visualizing docking results, and calculating descriptors. It obtains input files from Hermes, which includes tasks such as adding hydrogen atoms and defining the correct ionization and tautomeric states of protein residues. (33 ).

The Hermes visualizer is also employed for interactive docking setup such as for defining the binding site and the setting of constraints. Gold Mine is a tool for the analysis and post-processing of docking results. GOLD will likely be used in conjunction with a modeling program to create and edit starting models.

**Benefits** :-

a) It is used for Protein-Ligand Docking by using Genetic Algorithm.
b) Forbinding mode predictions.
c) GOLD is employed in structural biology research to study protein-ligand interactions, protein conformational changes, and protein-ligand recognition mechanisms
d) GOLD can be used in fragment-based drug design (FBDD) to identify small molecular fragments that bind to specific regions of the target protein.

**BioSuite:-**

BioSuite combines the capabilities of macromolecular sequence and structural analysis, chemo informatics, and algorithms to assist in the process of drug discovery. It is comprised of four main modules, which consist of a total of 79 distinct programs. This makes BioSuite one of the few comprehensive suites that cover a significant portion of the bioinformatics applications spectrum. The four major modules, namely Genome and Proteome Sequence Analysis, 3D Modeling and Structural Analysis, Molecular Dynamics Simulations, and Drug Design, are conveniently accessible through a user-friendly graphical interface. Additionally, BioSuite provides ample documentation and tutorials to support users in effectively utilizing its functionalities.

The BioSuite's Genome and Proteome Sequence Analysis module focuses on various applications related to the analysis of nucleic acid and protein sequences.

It not only deals with individual molecules but also encompasses complete genome and proteome sequences. This module provides the capability to annotate genomes, predict protein secondary structures, establish phylogenetic relationships among organisms, and compare gene or protein similarities between two genomes. ( 34 ).

The 3D modeling and analysis module has capabilities to build, analyze and predict three dimensional structures of macromolecules and macromolecular complexes. The 'Simulations' module essentially simulates the behavior of a molecule, in terms of its three dimensional structure(35).

The Drug Design module provides the following functionalities:

a. Prediction of biological activities of unknown chemical entities using QSAR.
b. Identification of pharmacophores in biologically active molecules.
c. Superimposition of a set of molecules in 3D space by alignment.
d. Identification of the ligand poses in 3D space when it binds to a target using Docking.

**Uses :-**

1. Genome analyzing and sequence analyzing.
2. 3D modeling, simulation, structural changes, drug design, pathway modeling, SNP analysis and comparative genomics.
3. BioSuite provides a range of bioinformatics tools for analyzing and annotating protein sequences, predicting protein structure and function, and comparing protein structures across different species or homologous proteins.
4. BioSuite offers virtual screening tools for in silico screening of compound libraries against protein targets of interest.
5. BioSuite includes modules for molecular dynamics simulation, allowing researchers to simulate the dynamic behavior of biomolecular systems over time.

**GRAMM (global range molecular matching):-**

The GRAMM software serves as a protein docking tool, employed to predict the structure by utilizing the atomic coordinates of the two molecules under consideration. It produces a compilation of ligand positions with high scores, indicating low-energy configurations, which can be directly utilized or further improved using alternative techniques. Moreover, GRAMM offers the flexibility to incorporate user-defined constraints during the docking process. These constraints can include distance restraints or specific interactions between particular residues. This characteristic empowers researchers to customize the docking process according to their specific requirements and hypotheses.

Uses:-

a) It is used for protein-protein docking and protein-ligand docking.
b) GRAMM can be used to model the binding interactions between proteins and DNA molecules.
c) GRAMM provides tools for docking small molecule ligands to protein targets.
d) GRAMM docking software can aid in the study of protein structure and function by predicting the interactions between biomolecules.
e) GRAMM docking tools can be applied in virtual screening campaigns to identify lead compounds with potential pharmacological activity against specific protein targets.
f) GRAMM docking software enables structure-based drug design approaches, where the three-dimensional structure of the target protein is used to guide the rational design of new drug molecules.

**FlexX**:-

This allows FlexX to accurately predict the binding affinity and specificity of ligands to their target proteins, making it a powerful tool for drug discovery and design. By utilizing flexible ligands and rigid proteins, FlexX is able to explore a wide range of conformational space and identify optimal binding poses for potential drug candidates. The incorporation of the MIMUMBA torsion angle database further enhances the accuracy of the predictions by capturing the subtle nuances of molecular interactions. Overall, FlexX's fragment-based approach, coupled with the MIMUMBA database, provides a robust platform for rational drug design and lead optimization.

***In-vitro* studies in drug repurposing**

in vitro, ex vivo, or phenotypic assays are defined as those that model some aspect of disease biology in cells or tissues derived from an experimental species or humans, and that provide a reason to believe that activity has translatable relevance. In vitro assays have the following benefits when used to identify repositioning activity:

a) Direct knowledge is gained in relation to a potential new disease setting.
b) multiple compounds with different modes of action can be tested and, depending on the throughput of the assay, over a full concentration–effect range.
c) It allows for serendipitous, or hypothesis-free, assessment of compounds
d) It provides data-driven choices for subsequent evaluation in more complex phenotypic or in vivo tests.

Example -zebrafish in drug repositioning include studies of pancreatic disease(37 ), tobacco dependence. and hearing impairment. developed transgenic zebrafish in which pancreatic β-cell differentiation as(38)well as Notch signaling, a critical pathway in pancreatic development(39) could be observed;.

| Drug | Original indication | Reposition |
|---|---|---|
| Amantadine | Influenza | Parkinson's disease |
| Amphotericin | Antifungal | Leishmaniasis |
| Aspirin | Inflammation, pain | Antiplatelet |
| Bromocriptine | Parkinson's disease | Diabetes mellitus |
| Bupropion | Depression | Smoking cessation |
| Colchicine | Gout | Recurrent pericarditis |
| Finasteride | Benign prostatic hyperplasia | Male pattern baldness |
| Gabapentin | Epilepsy | Neuropathic pain |
| Methotrexate | Cancer | Psoriasis, rheumatoid arthritis |
| Miltefosine | Cancer | Visceral leishmaniasis |
| Minoxidil | Hypertension | Male pattern baldness |
| Propranolol | Hypertension | Migraine prophylaxis |
| Sildenafil | Angina | Erectile dysfunction, pulmonary hypertension |
| Thalidomide | Morning sickness | Erythema nodosum leprosum |
| Zidovudine | Cancer | HIV/AIDS |

**Figure 5: some examples of repourposed drugs**

**Some examples of repurposing**

Introducing approved drugs, such as those developed to treat diabetes (Metformin) or inflammation (Thalidomide), identified to have cytostatic activity, can enhance chemotherapy or even replace more cytotoxic drugs.

**Doxorubicin:-**

Doxorubicin is classified as an anthracycline, a specific type of antibiotics that were originally derived from Streptomyces peucetius. Its medical approval dates back to 1974, however, it was not until two decades later that its significance in the treatment of breast cancer was fully understood and established (40-41 ). The drug functions by intercalating with DNA, causing breaks in the DNA strands, and by interfering with the topoisomerase-II-mediated DNA repair process, thereby hindering DNA. It replication is frequently employed as a chemotherapeutic treatment for breast cancer and various other types of malignancies, even though it exhibits variability in tumor response based on individual patients and is linked to significant levels of toxicity.

( 42 ). Indeed, cardiotoxicity is one of the most important parameters to consider when administering doxorubicin, as several studies suggest that the degree of toxicity is influenced by the genetic variability of the patients.

**Cyclophosphamide:-**

Cyclophosphamide, an alkylating agent, has demonstrated its effectiveness in modulating the immune system by inhibiting suppressive regulatory T cells and promoting the activity of effector T cells within the tumor microenvironment. This medication has been repurposed for the treatment of breast cancer and various other malignancies.

The drug exhibits varying functions based on its dosage, whereby lower concentrations regulate the immune system, whereas higher concentrations function as an alkylating agent, leading to cancer development and the death of lymphoid cells. ( 43 ). The efficacy of this chemotherapeutic agent is contingent upon the process of bioactivation facilitated by cytochrome P450 (CYP) enzymes. Among these enzymes, CYP2B6 and CYP2C19, encoded by various genes, exhibit polymorphic characteristics that can impact their functionality significantly. Genetic variants within these genes, such as CYP2B6 (rs12721655, rs3745274, *1, *6) and CYP2C19 (rs4244285, *1/*17/*2), have been identified as potential influencers of the response to therapy, particularly in the context of cyclophosphamide treatment for breast cancer.

**Everolimus :-**

Everolimus, a drug that has undergone repurposing, received its initial approval in 2009 for the treatment of renal cancer. Subsequently, in 2010, it was granted approval for its use in suppressing the immune system during renal transplants. Finally, in 2011, Everolimus was approved for the treatment of pancreatic cancer.

Everolimus exhibits its anti-neoplastic properties by exerting its influence on the mTOR pathway. Currently, it is being utilized in the treatment of metastatic breast cancer that is resistant to anastrozole and letrozole. In 2012, the Food and Drug Administration (FDA) granted its approval for the use of Everolimus in this specific type of cancer. Furthermore, Everolimus has also been included in the phase III clinical trial known as the 'Breast Cancer Trial of 14 Oral Everolimus-2 (BOLERO-2)'. This trial investigates the combination of Everolimus with exemestane for the treatment of HR+, HER2 advanced metastatic cancers that demonstrate resistance to letrozole or anastrozole.( 44 ). Like many other drugs, everolimus is metabolized by cytochrome enzymes. Some of the genes involved in its metabolism, such as the CYP3A4 (rs35599367), have been linked to a lower drug metabolic rates. The pharmacokinetic characteristics of everolimus are subject to variation based on the genetic variations of certain genes, specifically those associated with the PI3K/AKT/mTOR pathway, as well as the gene responsible for encoding its protein transporter (ABCB1).

Several studies have suggested a connection between the toxicity of everolimus and specific single nucleotide polymorphisms (SNPs) found in genes related to the PI3K/AKT/mTOR pathway (45 ) as well as to ABCB1 (rs1045642), associated with the development of mucositis, ABCB1 (rs2032582), associated with lymphopenia, PIK3R1 (rs10515074),associated with hyperglycemia and leucopenia, and RAPTOR (rs9906827), associated withpneumonitis(46 ).

**Tamoxifen:-**

Tamoxifen, initially developed as a selective estrogen receptor modulator, was originally intended for the treatment of Albright syndrome and to stimulate ovulation in women. By acting as a competitive inhibitor of estradiol, it has the ability to bind to estrogen receptors and exhibit anti-neoplastic properties specifically in mammary tissue. (47-49 ). The major challenge when using tamoxifen is the tumour resistance to the therapy and its adverse effects on the liver. Drug resistance may arise due to pharmacological factors, with tamoxifen undergoing conversion into its primary active metabolite, endoxifen, via the enzymatic activity of cytochrome P450 family genes, particularly CYP2D6, which exhibits numerous genetic variations linked to its function. ( 50-51 ). A lot of studies have been developed to assess the influence of the genetic variants of CYP2D6 in tumour resistance. , the association between SNPs in CYP2D6 and the mechanisms of drug resistance and side effects observed with the treatment with tamoxifen must be clarified to link the genotyping of this gene with the disease outcomes.

**Anastrazole:-**

anastrazole is a repurposed drug that was originally used for ovary stimulation and the induction of ovulation in infertile females or women with polycystic ovary syndrome. This drug functions as an aromatase inhibitor, reducing estrogen production and is therefore used in ER+ breast cancer treatment (52 ). However, recent findings warn of the eventuality of this drug becoming a ligand for estrogen receptors, binding to ERα and activating the downstream pathway of malignant growth. Consequently, it is of great importance to perform further studies to clarify the mechanisms of anastrozole, to provide clear evidence when this drug becomes a ligand of estrogen receptor, allowing to design adequate treatments and improve clinical outcomes. Some reports defend that a genetic variant in CSMD1, associated with increased expression of CSMD1 and CYP19 proteins, sensitizes malignant cells to be responders of anastrozole. Indeed, it has been reported that patients who carry rs4646 SNP in CYP19A1 have a good response to anastrozole.

Thus, predicting which tumours are more sensitive to this therapeutic strategy is mandatory for adequate oncologic treatment. According to the previous studies, CSMD1 and CYP19A1 SNPs could, in fact, be used as predictive markers of anastrozole response in breast cancer.

**Paclitaxel:-**

Paclitaxel is another repurposed drug whose initial use was addressed to arterial restenosis. It was isolated for the first time in 1971 from pacific yew. In cancer, its first use was in ovarian cancer, in 1992( 53 ) Currently, paclitaxel is used as a neoadjuvant or adjuvant treatment of breast cancer, as well as ovarian cancer. Its anti-tumoral mechanism is by the inhibition of mitosis of the cells, decreasing the proliferation rate of cancer cells [29,31]. The tumour response rate to paclitaxel has a largely associated dispersion, attributed to the genetic variation of the patients. In silico studies have reported some SNPs as potential predictive biomarkers of tumour insensitivity to paclitaxel, located on LPHN2, ROBO1, SNTG1, and GRIK1.(54 )

**Aspirin :-**

Another well-known drug repurposed to treat triple-negative breast cancer is aspirin. Aspirin is an antiplatelet agent that was initially used for cardiovascular diseases, but its clinical use was further expanded with the discovery of its anti-tumoral effect. Some studies refer that aspirin regular use is linked to a decrease in breast cancer risk ( 55 ). Importantly, polymorphisms in PIK3CA have been reported as being related to different effects of aspirin in breast cancer treatment. This is supported by the fact that aspirin can suppress the growth in breast cancer cells that have a mutation in this gene, through the activation of AMPK and mTORC1 inhibition signalling pathways (56). Therefore, Henry et al. suggest that breast cancer can be potentially treated using a combinatory ther- apy between aspirin and PI3K inhibitors. Nevertheless, Henry and colleagues defend that for this to be effective, it is essential to previously stratify these patients according to their genetic profile regarding PI3K gene, to find potential responders to this therapy( 57).

**Metformin:-**

Metformin was first synthesized in 1922,It has become the first-line treatment for type 2 diabetes(58).The first cancer trials arose from reports that diabetics taking metformin daily had lower rates of breast and other cancers, augmented by studies showing cells from metformin-treated diabetics do not grow well in culture( 59 ).

The cytostatic effects reported led to introducing metformin as an adjunct therapy for different types of cancers, some of which have reported remarkable success. Metformin also improved PFS and OS in advanced, previously untreated non-small cell lung cancer (NSCLC)( 60) , In two phase II studies, the overall impact of metformin, when administered alongside platinum-based chemotherapy with or without the anti-VEGF inhibiting antibody, bevacizumab (Avastin), would result in a reduction in tumor cell proliferation and metastasis. This effect is achieved through the specific deprivation of tumor cells, which exhibit an increased metabolic demand for glucose, due to the lowered concentration of circulating glucose caused by metformin.

**Thalidomide and other derivatives** :-

Thalidomide analogues are utilized in a range of applications within contemporary cancer treatment. Originally intended for alleviating morning sickness, Thalidomide was initially marketed to expectant mothers in Germany during the 1950s, with suggested dosages falling within the same range as aspirin treatments (300–500 mg). ( 61 ). The approval of the drug in the USA was halted due to its side effects, such as peripheral neuropathy. Thalidomide was globally removed in 1962 following its association with significant birth defects. Despite this discontinuation, thalidomide continued to be utilized in clinical settings for the treatment of Hansen's disease (leprosy). The primary application of thalidomide and its analogs for an extended period was in immunomodulation. ( 62 ). The earliest introduction of thalidomide derivatives to cancer therapy was to control inflammation. Thalidomide and its more potent structural relatives, lenalidomide and pomalidomide, are used to treat multiple myeloma, mantle cell lymphoma, (63 ) and myelodysplastic syndromes associated with the deletion 5q abnormality. Another reason for thalidomide-related compounds' action in cancer cells: their ability to bind cereblon,33 a protein involved in limb outgrowth. Early results indicated that thalidomide induced specific degradation of repressors in T-cells that can lead to activation and increased IL-2 secretion, ( 64 ) thus providing another way to stimulate the immune system to fight cancer cells.

**Sodium–glucose cotransporter 2 (SGLT2) :-**

Sodium-glucose cotransporters (SGLTs) are a group of membrane proteins responsible for facilitating the movement of glucose, amino acids, vitamins, ions, and osmolytes through the brush-border membrane of proximal renal tubules and the intestinal epithelium. These transporters belong to the SLC5 gene family.

SGLT2 inhibitors have been proven for renoprotective effects. In the early stage of diabetic nephropathy SGLT2 inhibition causes decrease in blood glucose level and also prevents albuminuria and inflammatory processes.

An animal study with empagliflozin in type 1 diabetic mice reported that inhibition of SGLT2 causes decreased glomerular filtration rate (GFR) and defended diabetic glomerular hyperfiltration. Empagliflozin has also shown to improve diabetic nephropathy and pancreatic damage by reducing proteinuria and pancreatic fatty acid infiltration.

**SGLT-2 inhibitors in cardioprotection**:-

The positive effects of SGLT2 inhibitors on blood pressure have been demonstrated in early clinical studies. Both dapagliflozin and canagliflozin have been proven to reduce systolic blood pressure in patients with type 2 diabetes who have either low or normal blood pressure levels. These inhibitors have also been reported to improve other cardiovascular risk factors such as HbA1c, body weight, and lipid profile. Additionally, a recent study with dapagliflozin showed a lower rate of cardiovascular death or hospitalization for heart failure in type 2 diabetes patients who are at risk for atherosclerotic cardiovascular diseases. In randomized, placebo-controlled trials, SGLT2 inhibitors have demonstrated an 11% decrease in major adverse cardiovascular events, a 23% decrease in the risk of cardiovascular death or hospitalization for heart failure, and a decrease in death from any other cause.

**SGLT-2 inhibitors in cancer :-**

Recent research studies have demonstrated the potential anti-cancer properties of certain SGLT2 inhibitors. Specifically, Canagliflozin has been found to inhibit the growth of liver cancer and angiogenesis by blocking glucose uptake. Additionally, Canagliflozin has shown anticancer activity by inhibiting mitochondrial complex-I supported respiration. Another SGLT2 inhibitor, Dapagliflozin, has also been reported to possess anticancer activity, particularly for colon cancer cell populations that do not express UGT1A9 (UDP Glucuronosyltransferase 1 family, Polypeptidase A9). Both Canagliflozin and Dapagliflozin have demonstrated anticancer effects for pancreatic and prostate cancer as well. Furthermore, a recent study has indicated that Canagliflozin could potentially be repurposed to treat lung cancer that is resistant to epidermal growth factor tyrosine kinase inhibitors. This observed effect is likely due to the enhanced inhibition of the estimated glomerular filtration rate.

**Result and discussion :**

Drug repurposing, also known as drug repositioning, has gained significant importance in recent years within the realm of pharmaceutical research. This is largely attributed to various advantages associated with this approach, including the ability to reduce the duration of clinical trials, the revitalization of existing medications through identification of novel therapeutic targets, and the unveiling of unforeseen connections between seemingly disparate

Medical conditions.Computational based approaches has made it easy to overcome various difficulties which are being faced at the time of the traditional approaches. Drug repurposing has given a new boom to the pharma sectors as well as to the research industries resulting in giving a new opportunity to drugs which there exist more than one pourposesas well as for the drugs which have been removed from the market due to their side effects , adverse drug reactions , cost , better alternate of them. Drug repurposing or repositioning approach has set a new platform for many drugs and has provided as an alternate route for the drugs which posses more than one therapeutic properties among them.

Many drugs like sidldenafil which was shown as a therapeutic drug for the treatment of hypertension and angina possesed its therapeutic action today as for treatment of erctile dysfunction . similarly many such drugs which have gone thorugh the phase of drug repurposing or repositioning have shoen more than one therapeutic effect or actions . for e.g – a well known medicine which is used as the anti – platelet aggregation is known founf to have posses ita action in the treatment a severe disease cancer.Aspirin has possesed to show its therapeutic action in the treatment of triple – negative breast cancer.

Aspirin can suppress the growth in breast cancer cells through the activation of AMPK and mTORC1 inhibition signalling pathways. Another example of drug repurposing is Sodium Glucose – CoTransporter which have shown properties other than for they were derived . Sodium Glucose – Co Transporter are a members of protein family that are involved in the transport of glucose, amino acids, vitamins, ions and osmolytes across the brush-border

membrane of proximal renal tubules as well as the intestinal epithelium. SGLT2 inhibitors have been demonstrated their beneficial effects on blood pressure. Dapagliflozin and canagliflozin both have confirmed to reduce systolic blood pressure in hypotensive and normotensive patients with type- 2diabetes. Thalidomide derivatives have a variety of uses in modern cancer therapy earlier they were used Thalid to treat morning sickness and sold over the counter to pregnant women in Germany in the 1950s

**Conclusion**

Repurposing drugs is an encouraging approach to accelerate the creation of novel therapies, providing advantages in terms of financial savings, time efficiency, and safety enhancements. Through utilizing current information and assets, pharmaceutical firms and scientists can more effectively tackle unmet healthcare requirements. Repurposing drugs can also offer safety advantages, as the side effects and potential risks of a drug are already well-known. This can help to streamline the regulatory approval process, as regulators can have more confidence in the safety profile of a repurposed drug compared to a completely new compound. This can ultimately lead to faster access to new treatments for patients in need.

**References**:-

– Vineela Parvathaneni, Vivek Gupta(2020): Utilizing drug repurposing against COVID-19 Efficacy, limitations, and challenge, life sciences 2020, 118275(https://doi.org/10.1016/j.lfs.2020.118275 )

2. Hema Sree GNSa , Saraswathy GRa , Manikanta Muraharia, Mamatha Krishnamurthy(2019): An update on Drug Repurposing: Re-written saga of the drug's fate, biomdeicine and pharmacotherapy 2019, Pages 700-719,(https://doi.org/10.1016/j.biopha.2018.11.127)
3. Yosef Masoudi-Sobhanzadeha , Yadollah Omidib , Massoud Amanlouc , Ali Masoudi-Nejad(2020): Drug databases and their contributions to drug repurposing, genomics 2020,1087-1095(https://doi.org/10.1016/j.ygeno.2019.06.021 )

4. James Schuler, Zackary Falls, William Mangione, Matthew L. Hudson, Liana Bruggemann, Ram Samudrala(2022): Evaluating the performance of drug-repurposing technologies, drug discovery today January 2022, Volume 27, Number 1,49-64,( https://doi.org/10.1016/j.drudis.2021.08.002)
5. Kyungsoo Park(2019): A review of computational drug repurposing, Translational and Clinical Pharmacology2019 ,Jun;27(2),pages 59-63(https://doi.org/10.12793/tcp.2019.27.2.59 )
6. Francisco Javier Somolinos , Carlos León , and Sara Guerrero-Aspizua(2021):biological processes and systems **2021**, *9*(6), 1057, 7-14 (https://doi.org/10.3390/pr9061057 )
7. Janet Sultan, Salvatore Crisafulli, Flic Gabbay, Elizabeth Lynn, Saad Shakir and Gianluca Trifirò(2020): Challenges for Drug Repurposing in the COVID-19 Pandemic Era, Front. Pharmacol,06 November 2020,volume-11( https://doi.org/10.3389/fphar.2020.588654)
8. Vineela Parvathaneni, Vivek Gupta(2020): Utilizing drug repurposing against COVID-19 Efficacy, limitations, and challenges, life sciences 2020,volume-259,( https://doi.org/10.1016/j.lfs.2020.118275 )
9. Thanigaimalai Pillaiyar, Sangeetha Meenakshisundaram, Manoj Manickam,Murugesan Sankaranarayanan(2020): A medicinal chemistry perspective of drug repositioning: Recent advances and challenges in drug discovery, europeanjournal of medical chemistry,June 2020,volume-259,(https://doi.org/10.1016/j.ejmech.2020.112275)
10. Dorothea Emig, Alexander Ivliev, Olga Pustovalova, Lee Lancashire, Svetlana Bureeva, Yuri Nikolsky, Marina Bessarabova(2013): Drug Target Prediction and Repositioning Using an Integrated Network-Based Approach, institute for research in biomedicine ,April 2013,volume 8( https://doi.org/10.1371/journal.pone.0060618 )
11. Nithya Krishnamurthy, Alyssa A.Grimshaw, Sydney A. Axson , Sung Hee Choe and Jennifer E. Miller(2022): Drug repurposing: a systematic review on root causes, barriers and facilitators, BMC health services research,July 2022,volume22 (https://doi.org/10.1186/s12913-022-08272-z)

12. Maryam Lotfi Shahreza, Nasser Ghadiri, Sayed Rasoul Mousavi, Jaleh Varshosaz and James R. Green(2017): A review of network-based approaches to drug repositioning, briefing in bioinformatics,27 February 2017, Pages 878–892 ,( https://doi.org/10.1093/bib/bbx017 )
13. Dorothea Emig, Alexander Ivliev, Olga Pustovalova, Lee Lancashire, Svetlana Bureeva, Yuri Nikolsky, Marina Bessarabova(2013): Drug Target Prediction and Repositioning Using an Integrated Network-Based Approach, open access ,April 4, 2013,volume 8( doi:10.1371/journal.pone.0060618 )
14. S. Joshua Swamidass(2011): Mining small-molecule screens to repurpose drugs,briefings in bioinformatics,March 2011, Pages 327–335 ( https://doi.org/10.1093/bib/bbr028 )
15. Kamal Kumar Chaudhary and Nidhi Mishra(2016): A Review on Molecular Docking: Novel Tool for Drug Discovery,scimed central,August 2016
16. Jacob White(2020): PubMed 2.0, MEDICAL REFERENCE SERVICES QUARTERLY,Oct 2020, Pages 382-387 (https://doi.org/10.1080/02763869.2020.1826228 )
17. Zhiyong Lu(2011): PubMed and beyond: a survey of web tools for searching biomedical literature,the journal of biological databases and curation,january 2011,volume 2011(https://doi.org/10.1093/database/baq036)
18. Nataraj S. Pagadala1 & Khajamohiddin Syed& Jack Tuszynski(2016): Software for molecular docking: a review, International Union for Pure and Applied Biophysics (IUPAB) and Springer-Verlag Berlin Heidelberg, December 2016, pages 91–102 ,(10.1007/s12551-016-0247-1)
19. Christopher Markosian, Luigi Di Costanzo, Monica Sekharan , Chenghua Shao, Stephen K. Burley & Christine Zardeck(2018): Analysis of impact metrics for the Protein Data Bank, scientific data ,October 2018,(DOI: 10.1038/sdata.2018.212)

20. PAYAM BEHZADi and MARI O GAJD ACS(2021): Worldwide Protein Data Bank (wwPDB): A virtual treasure for research in biotechnology, european journal of micrbiology and immunology, December 15, 2021,pages 77-86,(https://doi.org/10.1556/1886.2021.00020)
21. Stephen K. Burley, Helen M. Berman, Cole Christie,Jose M. Duarte, Zukang Feng, John Westbrook,Jasmine Young,and Christine Zardecki(2017): RCSB Protein Data Bank: Sustaining a living digital data resource that enables breakthroughs in scientific research and biomedical education, protein science , 25 October 2017,pages 316-330,(https://doi.org/10.1002/pro.3331 )
22. Mario S. Valdés-Tresanco1, Mario E. Valdés-Tresanco, Pedro A. Valiente and Ernesto Moreno(2020): AMDock: a versatile graphical tool for assisting molecular docking with Autodock Vina and Autodock,biology direct, 16 September 2020,volume 15(https://doi.org/10.1186/s13062-020-00267-2 )
23. Thomas Gaillard(2018): Evaluation of AutoDock and AutoDock Vina on the CASF-2013 benchmark, Journal of Chemical Information and Modeling, July 10, 2018, 1697–1706 (https://doi.org/10.1021/acs.jcim.8b00312 )
24. Tatiana F. Vieira and Sérgio F. Sousa(2019): Comparing AutoDock and Vina in Ligand/Decoy Discrimination for Virtual Screening,applied biosciences and bioengineering, 25 october 2019,(https://doi.org/10.3390/app9214538 )
25. K.P. Mishra, L. Ganju, M. Sairam, P.K. Banerjee, R.C. Sawhney(2008): A review of high throughput technology for the screening of natural products,biomedicine and pharmacotherapy, 26 June 2008, Pages 94-98 ,(doi:10.1016/j.biopha.2007.06.012)
26. C. J. ZHENG, L. Y. HAN, C. W. YAP, Z. L. JI, Z. W. CAO, AND Y. Z. CHEN(2006): Therapeutic Targets: Progress of Their Exploration and Investigation of Their Characteristics,researchgate,july 2006, 259-279 (http://dx.doi.org/10.1124/pr.58.2.4)

27. C. John Harris, Richard D. Hill , David W. Sheppard,Martin J. Slater and Pieter F.W. Stouten(2011): The Design and Application of Target-Focused Compound Libraries,researchgate,april 2011, 521-531 ( http://dx.doi.org/10.2174/138620711795767802 )
28. DA Pereira and JA Williams(2007): Origin and evolution of high throughput screening,british journal of pharmacology ,september 2007,pages53-61,( https://doi.org/10.1038/sj.bjp.0707373 )
29. John P. Overington, Bissan Al-Lazikani and Andrew L. Hopkins(2006): How many drug targets are there?,nature reviews,december 2006, pages993–996,( 10.1038/nrd2199).
30. Wilson Z. Shou(2020): Current status and future directions of high-throughput ADME screening in drug discovery, journal of pharmaceutical analysis, 14 May 2020,201-208 ( https://doi.org/10.1016/j.jpha.2020.05.004 )
31. Lisina K.V, Shanmughavel Piramanayagam(2014): An insilico study on anti inflammatory compounds from marine system using Molegro virtual docker , 2014 ,world journal of pharmacveutical sciences 259-421,( https://www.wjpsonline.com/index.php/wjps/issue/view/10 ).
32. http://www.ccdc.cam.ac.uk/Solutions/GoldSuite/Pages/GOLD.aspx (accessed 20. 12.16).
33. Elizabeth Yurieva, Mark Agostinoa and Paul A. Ramsland(2010): Challenges and advances in computational docking: 2009 in review, journal of molecular recognition ,21 July 2010,149-164,( DOI:10.1002/jmr.1077 )
34. http://www.serc.iisc.ernet.in/facilities/ComputingFacilities/software/biosuite. html (accessed 25.12.16).
35. Prasad G. Jamkhandea, , Mahavir H. Ghantea , Balaji R. Ajgunde: Software based approaches for drug designing and development(2017): A systematic review on commonly used software and its applications, bulletin of faculty of pharmacy,15 October 2017,203-210,( http://dx.doi.org/10.1016/j.bfopcu.2017.10.001 ).

36. Vincent Zoete , Aurélien Grosdidier , Olivier Michielin(2009): Docking, virtual high throughput screening and in silicofragment-based drug design,journal of cellular and molecular biology , January 11, 2009,238-248,( https://doi.org/10.1111/j.1582-4934.2008.00665.x )
37. Meritxell Roviraa,Wei Huanga, Shamila Yusuffa, Joong Sup Shimb, Anthony A. Ferrantec, Jun O. Liub,and Michael J. Parsons(2011): Chemical screen identifies FDA-approved drugs andtarget pathways that induce precocious pancreaticendocrine differentiation,PNAS nexus , October 18, 2011,19264-19269,(https://doi.org/10.1073/pnas.1113081108 )
38. Cousin, M. A, Ebbert, J. O,Wiinamaki, A. R.; et al(2014):Larval Zebrafish Model for FDA-Approved Drug Repositioning for Tobacco Dependence Treatment,march 21,2014,PLoS ONE,volume 9,(https://doi.org/10.1371/journal.pone.0090467 )
39. Parsons, M. J, Pisharath, H. Yusuff, S,et al.,(2009):NotchResponsive Cells Initiate the Secondary Transition in Larval Zebrafish Pancreas, MECHANISMS OF DEVELOPMENT ,10 July 2009,898-912,(https://doi.org/10.1016/j.mod.2009.07.002)
40. Aggarwal, S,Verma, S.S, Aggarwal, S, Gupta, S.C.(2021) :-Drug repurposing for breast cancer therapy: Old weapon for new battle. Semin. Cancer Biol. 2021, 68, 8–20,( https://doi.org/10.1016/j.semcancer.2019.09.012).
41. Antoszczak, M., Markowska, A., Markowska, J., Huczynski,(2020):- Old wine in new bottles: Drug repurposing in oncology. Eur. J. Pharmacol. 2020,volume 866,(https://doi.org/10.1016/j.ejphar.2019.172784).
42. Todorova, V.K., Makhoul, I.,Dhakal, I.,Wei, J., Stone, A.,Carter, W., Owen, A.,Klimberg, V.S (2017):-Polymorphic Variations Associated With Doxorubicin-Induced Cardiotoxicity in Breast Cancer Patients. Oncol Res 2017, 25, 1223–1229,( https://doi.org/10.3727/096504017X14876245096439 ).

43. de Jonge, M.E,HuitemaA.D, Rodenhuis S,BeijnenJ.H(2005):- Clinical pharmacokinetics of cyclophosphamide,Clinical Pharmacokinetics, 2005, 1135–1164,( https://doi.org/10.2165/00003088-197904050-00004 )
44. Royce M.E,Osman D,(2015): Everolimus in the Treatment of Metastatic Breast Cancer. Breast Cancer,sage journal,2015,73–79,( https://doi.org/10.4137/BCBCR.S29268).
45. Baselga J,CamponeM,Piccart M,Burris H.A,RugoH.S, Sahmoud T,Noguchi S, Gnant M,Pritchard K.I, Lebrun F, et al (2011):-Everolimus in Postmenopausal Hormone-Receptor–Positive Advanced Breast Cancer. N. Engl. J. Med. 2011, 366, 520–529,(DOI: 10.1056/NEJMoa110965 ).
46. Hoskins J.M, Carey L.A, McLeod H.L,(2009):-CYP2D6 and tamoxifen: DNA matters in breast cancer. Nat. Rev. Cancer 2009, 9, 576–586,( 10.1038/nrc2683 )
47. Bezerra L.S,Santos-Veloso, M.A.O, Bezerra Junior, N.D.S Fonseca, L.C.D Sales, W.L.A(2018) :-Impacts of Cytochrome P450 2D6 (CYP2D6) Genetic Polymorphism in Tamoxifen Therapy for Breast Cancer. Rev. Bras. Ginecol. Obstet. 2018, 40, 794–799,(DOI: 10.1055/s-0038-1676303 )
48. Wickramage I,Tennekoon K.H,Ariyaratne M.A, Hewage A.S,Sundralingam (2017):-T. CYP2D6 polymorphisms may predict occurrence of adverse effects to tamoxifen: A preliminary retrospective study. Breast Cancer 2017, 9, 111–120,(https://doi.org/10.2147/BCTT.S126557 ).
49. Hertz D.L, Kidwell K.M,Hilsenbeck S.G, Oesterreich S, Osborne C.K, Philips S, Chenault C, Hartmaier R.J,Skaar T.C, Sikora M.J.,et al (2017) : CYP2D6 genotype is not associated with survival in breast cancer patients treated with tamoxifen: Results from a population-based study. Breast Cancer Res. Treat. 2017, 166, 277–287,(DOI 10.1007/s10549-017-4400-8 ).
50. Nowell S.A, Ahn J,Rae J.M,Scheys J.O,Trovato A, Sweeney C,MacLeod S.L,Kadlubar F.F, Ambrosone C.B. ( 2005):Association of genetic variation in tamoxifen-metabolizing enzymes with overall survival and recurrence of disease in breast cancer patients. Breast Cancer Res. Treat. 2005, 91, 249–258,( DOI 10.1007/s10549-004-7751-x ).

51. Wegman P, Vainikka L,Stål O, Nordenskjöld B,Skoog L, Rutqvist L.E, Wingren S,(2005):-Genotype of metabolic enzymes and the benefit of tamoxifen in postmenopausal breast cancer patients. Breast Cancer Res. 2005, 7, 284-90,( https://doi.org/10.1186/bcr993).
52. Ingle J.N.,Cairns J, Suman V.J, Shepherd L.E, Fasching P.A,Hoskin T.L, Singh R.J,Desta Z,Kalari K.R.; Ellis, M.J.; et al(2020):-. Anastrozole has an Association between Degree of Estrogen Suppression and Outcomes in Early Breast Cancer and is a Ligand for Estrogen Receptor α. Clin. Cancer Res. 2020, 26, 2986–2996,( https://doi.org/10.1158/1078-0432.CCR-19-3091 ).
53. Cairns, J, Ingle J.N,Dudenkov T.M, Kalari K.R,Carlson E.E, Na J, Buzdar A.U, Robson M.E, Ellis M.J, Goss P,et al.,(2020):- Pharmacogenomics of aromatase inhibitors in postmenopausal breast cancer and additional mechanisms of anastrozole action. JCI Insight 2020, 5, e137571,(https://doi.org/10.1172/jci.insight.137571 ).
54. Perez-Ortiz A.C,Ramirez I,Cruz-López J.C, Villarreal-Garza C,Luna-Angulo A,Lira E, Jiménez-Chaidez S, Díaz-Chavez J, Matus-Santos J.A, Sánchez-Chapul L,et al (2017):-Pharmacogenetics of response to neoadjuvant paclitaxel treatment for locally advanced breast cancer. Oncotarget 2017, 8, 106454–106467,( https://doi.org/10.18632/oncotarget.22461 ).
55. Ma, S, Guo C, Sun C, Han T, Zhang H,Qu G, Jiang Y, Zhou Q,Sun Y,(2019) :-Aspirin Use and Risk of Breast Cancer: A Meta-analysis of Observational Studies from 1989 to 2019. Clin. Breast Cancer 2021, 21, 552–565,( https://doi.org/10.1016/j.clbc.2021.02.005).
56. Zhou Y ,Simmons J,Jordan C.D, Sonbol M.B,Maihle N, Tang S.-C,(2019):- Aspirin Treatment Effect and Association with PIK3CA Mutation in Breast Cancer: A Biomarker Analysis. Clin. Breast Cancer 2019, 19, 354–362,( https://doi.org/10.1016/j.clbc.2019.05.004 ).

57. Henry W.S , Laszewski T ,Tsang T ,Beca F , Beck A.H , McAllister S.S, Toker, A (2017):-Aspirin Suppresses Growth in PI3K-Mutant Breast Cancer by Activating AMPK and Inhibiting mTORC1 Signaling. Cancer Res. 2017, 77, 790–801,( https://doi.org/10.1158/0008-5472.CAN-16-2400 ).
58. Decensi A, Puntoni M, Goodwin P, et al,(2010) :- Metformin and cancer risk in diabetic patients: a systematic review and meta-analysis. Cancer Prev Res 2010;3:1451–61,( https://doi.org/10.1158/1940-6207.CAPR-10-0157 ).
59. Palazzolo G, Mollica H, Lusi V, et al,(2020):- Modulating the distant spreading of patient-derived colorectal cancer cells via aspirin and metformin. Transl Oncol 2020;volume 13:100760,( https://doi.org/10.1016/j.tranon.2020.100760 ).
60. Arrieta O, Barron F, Padilla MS, et al,(2019):- Effect of metformin plus tyrosine kinase inhibitors compared with tyrosine kinase inhibitors alone in patients with epidermal growth factor receptor-mutated lung adenocarcinoma: a phase 2 randomized clinical trial. JAMA Oncol 2019;e192553,(https://doi.org/10.1001/jamaoncol.2019.2553 ).
61. Schein CH(2020):- Repurposing approved drugs on the pathway to novel therapies. Med Res Rev 2020;40: 586–605,( https://doi.org/10.1002/med.21627 )
62. Zhu YX, Kortuem KM, Stewart AK (2012):- Molecular mechanism of action of immune-modulatory drugs thalidomide, lenalidomide and pomalidomide in multiple myeloma. Leuk Lymphoma 2012;54:683–687,( https://doi.org/10.3109/10428194.2012.728597 )
63. Corral LG, Haslett PA, Muller GW, et al,(1999):- Differential cytokine modulation and T cell activation by two distinct classes of thalidomide analogues that are potent inhibitors of TNF-alpha. J Immunol 1999;163: 380–386,(https://doi.org/10.4049/jimmunol.163.1.380 ).
64. Gandhi AK, Kang J, Havens CG, et al,(2014):- Immunomodulatory agents lenalidomide and pomalidomide costimulate T cells by inducing degradation of T cell repressors Ikaros and Aiolos via modulation of the E3 ubiquitin ligase complex CRL4(CRBN).Br J Haematol 2014;164:811–821,( https://doi.org/10.1111/bjh.12708)