



Bridging Drug Delivery and Molecular Biology: A Synergistic Alliance between Liposomes and Bioinformatics

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ABSTRACT

Liposomes, since their discovery have emerged as valuable nanocarriers in drug delivery and molecular biology, significantly enhancing the therapeutic index of many drugs. They were first coined in early 1960s by discovery by Bangham and co-workers. Beyond their conventional applications, a growing interest lies in integrating liposome technology with bioinformatics. Bioinformatic tools play an important role in predicting, analyzing and simulating complex biological processes. These tools offer deeper insights into liposomal interactions with biological systems. Thanks to this advanced computational modeling, scientists can now design liposomes with improved targeting abilities, optimized stability, and enhanced therapeutic potential. One such model i.e Quantitative Structure–Activity Relationship (QSAR) is used for the precise prediction of liposome–drug and liposome–cell interactions, ultimately contributing to higher drug efficacy and reduced adverse effects. Many studies conducted till date throw light upon the promising synergy between liposomes and bioinformatics, highlighting its potential to transform drug delivery. This review explores this integration, focusing on how computational approaches can advance liposomal design, precision, and efficiency in therapeutic applications.

Keywords Nanocarriers, Bioinformatics, Liposomes drug delivery, Computational modeling, Therapeutic applications

INTRODUCTION

There have been major revolutions in the field of Nanotechnology, especially in drug delivery, medicine, specifically nanomedicine. These revolutions counter a key impetus for the development of various types of drug-carrying nanomaterials including; liposomes, micelles, polymeric nanoparticles etc (Lu *et al.*, 2021). Liposomes; the concerned nanocarriers in this review, first coined by Alec D. Bangham in 1965 comprises of phospholipid bilayers, represents a pioneering class of therapeutic nanoparticles (NPs) specially approved for clinical use (Abbasi *et al.*, 2022; Hamid *et al.*, 2024). Since discovery, they have successfully embarked a significant portion of clinical stage nanotherapeutics, and still many more are in the pipeline. Thanks to their bioavailability, biocompatibility, non-toxicity, and biodegradability they have made way progressively in the market (Li *et al.*, 2019; Abbasi *et al.*, 2022). These nanocarriers have the potential to entrap hydrophobic and hydrophilic drugs in them. As discussed earlier they are comprised of the phospholipid bilayers, these bilayers help them in the protection and stabilization of encapsulated drugs, enhancing their pharmacological effectiveness. Additionally, surface modification of liposomes with polyethylene glycol (PEG) impart stability in them, increasing their shelf life directing

them toward the host's reticuloendothelial system (RES) (Najlah *et al.*, 2019). This PEGylation process, involving covalently linking PEG chains to liposomes, has emerged as a pivotal strategy for enhancing the pharmacodynamic properties of therapeutic agents (Kanamala *et al.*, 2019). PEGylated liposomes are frequently approved for clinical use due to their improved residence time and effective targeting capabilities (Mohamed *et al.*, 2019).

The combination of liposomes with Bioinformatics has given molecular biology research a new height. While traditional liposome development relies heavily on experimental trial-and-error, integration with bioinformatics allows for rapid, precise, and predictive design of liposomal formulations, optimizing drug encapsulation, targeting, and release profiles more efficiently than conventional approaches (Eugster *et al.*, 2024). Thanks to bioinformatic tools we are able to design and predict the combination of liposomes with drugs to be encapsulated in them. We can get a control over customized drug release towards target sites under favorable conditions

(Han *et al.*, 2023). Through these tools the drug encapsulation efficiencies can also be determined, and we can predict the influence of molecular interactions

on liposomal behavior (Aghanejad *et al.*, 2021). Such advancements are a triumph success in the overall enhancement of therapeutic outcomes (Patel and Ghosh, 2022).

Composition of liposomes: Liposomes vary in size, they are used to encapsulate hydrophilic and hydrophobic drugs (Sharma *et al.*, 2018; Nakhaei *et al.*, 2023). Due to the presence of phospholipid bilayers, liposomes are able to promote active cellular uptake by actively binding to the cell membrane (Gonda *et al.*, 2019). They can further be classified on the basis of their lamellarity, ranging from small uni-lamellar vesicles (ULVs) to large multi-lamellar vesicles (MLVs), with each of them varying in their size range, and key features (Liu *et al.*, 2022). When discussing about the composition of liposomes its important to know that they are biocompatible in nature, they reduce toxicity and make the encapsulated material bioavailable when taken. Furthermore, they are able to cross the biological barriers due to their lipid profiles, thereby facilitating their absorption. The traditional neutral liposomes tend to aggregate and assemble together due to their amphiphilic structure, as a result they yielded high polydisperse distribution, drug leakage, and inefficient drug delivery resultantly low encapsulation efficiency, as compared to modern liposomes which yield high encapsulation efficiency liposomes (Jia *et al.*, 2023).

Preparation and characterization of liposomes: Liposomes can be prepared through different means (Aslam *et al.*, 2022). The preparatory methods include; solvent injection method, thin film hydration method, rehydration- dehydration method, detergent removal, reverse phase evaporation and freeze thaw method (Nsairat *et al.*, 2022). Each method comprises of its

own properties and features (Lombardo, D., & Kiselev., 2022), resultantly producing liposomes with difference in their charge, particle size and surface hydration (Yan *et al.*, 2020), their characterization is based on their size, shape, distribution, lamellarity and encapsulation (Cuazzo *et al.*, 2021; Fan *et al.*, 2021). Different parameters can be used to characterize liposomes, for instance dynamic light scattering (DLS), atomic force microscopy (AFM), Zeta sizer, fluorescent based assays, scanning electron microscopy (SEM), transmission electron microscopy (TEM), UV-Vis-spectroscopy, flow cytometry, lipid mixing assay, nuclear magnetic resonance (NMR), HPLC, and immune-cytochemistry are more common in practice (Sharma *et al.*, 2018; Torres Flores *et al.*, 2020; , Nsairat *et al.*, 2022).

Liposomes as the Drug delivery vehicles: Different nanocarriers are available to cargo drug in the body, using different administrative routes. Liposomes are one of them, because of their biocompatible and bioavailable nature as shown in Figure 1. They have been extensively used to achieve targeted drug delivery with reduced toxicity and side effects. Many FDA approved liposomal formulations like Doxil®, AmBisome®, Mayocet®, Abelcet®, Mepact® etc (Bulbake *et al* 2017) are readily available in the market and still more are in the pipeline (Man *et al.*, 2019; Almeida *et al.*, 2020). Many fields like pharmaceutical, material, and biomedical sciences have obtained quite remarkable progress in the research area of drug delivery (El-Hammadi, 2019). Liposomes are becoming a choice now a days because of its ability to modify the conventional drugs to achieve targeted drug delivery (Pauli *et al.*, 2019; Gagliardi and Ashizawa, 2021).

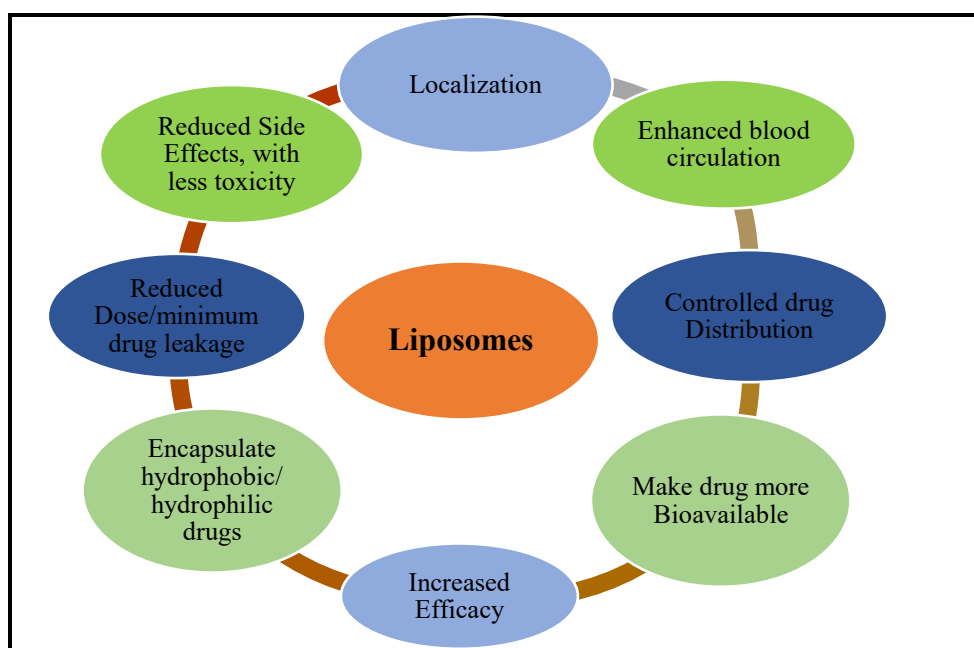


Figure 1: Benefits of liposomal drug delivery systems.

Drug loading pathways: The moment the drug is being loaded in liposomes, they aids the drug to move towards its target specifically with few traces of its side effects in the rest of the body (Tamam *et al.*, 2019). Two mechanisms of drug loading are used in liposomes either its active loading (remote loading) or passive loading (Pande, 2023). Liposomes are prepared first and then drug is being incorporated in them, this technique is active loading of drug which is used to achieve high encapsulation efficiency along with enhanced drug stability. The advantage of using this method is that it helps preventing the drug leakage. Moreover, active loading also known as the “remote loading” is an substitute strategy for drug encapsulation taking advantage of transmembrane chemical gradients involving buffer or ammonium sulphate gradients (Aslam *et al.*, 2022; Pisani *et al.*, 2023) to entrap amphipathic compounds from the surrounding environment into preformed liposomes, increasing their encapsulation efficiency (Maja *et al.*, 2020). Whereas there is another method also available in which the drug of choice is added during thin film preparation of liposomes, this method is termed as the passive loading, generally this method is not much appreciated because Passive loading relies on random drug encapsulation during the formation of liposomes, which leads to very poor reproducibility, low encapsulation efficiency and drug leakage especially for hydrophilic or amphipathic molecules (Allen and Cullis, 2013; Vakili-Ghartavol *et al.*, 2020).

Applications of liposomes: Liposomes have a wide range of applications so far. In drug delivery they are used to achieve target specificity (Guimarães *et al.*, 2021). They also help in improving the distribution of drug in body making it bioavailable towards the infectious site only (Farooque *et al.*, 2021). This helps in the promotion of very effective delivery of drug encapsulated compounds towards the targeted sites (Basu *et al.*, 2021). Liposomes as nanocarriers; currently holds a variety of applications in different domains of sciences including immunology, dermatology, vaccine adjuvant, brain targeting therapeutics, multiple eye disorders, tumor therapy and targeting infectious diseases (Kumbham *et al.*, 2023).

Challenges in Liposome-Based Drug Delivery Systems: Biological challenges: As soon as liposomes mark their presence in the body, they come across with the defense mechanisms including include the reticuloendothelial system (RES), immunogenicity, and opsonization, identify, neutralize, and eliminate them accordingly (Peng *et al.*, 2023). However, this rapid clearance can limit their effectiveness in drug delivery. To overcome these challenges other factors are opted by researchers like enhanced permeability and retention (EPR) effect which is phenomena where liposomes preferentially accumulate in infectious site due to leaky vasculature and poor lymphatic drainage. This natural targeting mechanism helps liposomes deliver drugs more efficiently despite immune clearance issues (Sawant & Torchilin, 2012).

Challenges in Pharmaceutical liposomal delivery: Although there are dozens of the liposomal medications present in the market, still the research and development in this area remains a significant challenge, requiring the simultaneous optimization of multiple factors to make a final safe and effective formulation of liposomes (Guimarães *et al.*, 2021).

In Vivo Challenges of Liposomal Fate: Liposomes, in comparison to free drugs, experience a more intricate biological process. However, the comprehensive understanding of their biological fate remains incomplete. Presently, there is limited insight into the dynamic interactions among the three primary elements; drug, carrier, and material, along with ligands, surfactants, and stabilizers serving as contributory factors (Li *et al.*, 2019). Fundamental questions about the precise timing, location, and mechanisms of drug release from liposomes during *in vivo* studies remain mainly unanswered. A thorough understanding of liposomal behavior is essential for optimizing their design and expediting their clinical translation. The FDA guidance document for liposome drug products stresses the importance of reporting both free and total drug quantities in pharmacokinetic studies (FDA, 2018).

However, there is a need to separate and quantify the free and encapsulated drugs. Traditional techniques, such as radiation and fluorescence labeling, are hindered by significant interference between signals from released drugs and the liposomal carriers (Chen *et al.*, 2019). Drug encapsulated Liposomes can be considered as "macro-prodrugs" that remain inactive on their release however when conditions become favorable, they are converted to free molecules. For example, liposomal doxorubicin requires entry into the nucleus to interact with DNA for its cytotoxic action (Liu *et al.*, 2022). Only the free drug in the nucleus is bioavailable. Despite this, the kinetic processes of drug release at tumor sites, within cells, and in the nucleus remain poorly understood, often described as a "black box." The plasma concentration of drugs encapsulated in liposomes does not reliably reflect their concentration towards the target, such as the nucleus. Thus, a better grasp of liposomal drug delivery is critical for rational design. However, separating free drugs from encapsulated drugs in tissues or target cells remains challenging, necessitating the development of advanced bioanalytical techniques (Smits *et al.*, 2019). Emerging strategies, including stealth coatings beyond PEG, ligand-mediated targeting, and stimuli-responsive liposomes, are being developed to prolong circulation, improve site-specific delivery, and achieve a controlled release. Highlighting these approaches would strengthen the discussion on overcoming pharmacokinetic limitations (Suk *et al.*, 2016).

Challenges in Liposome Preparation: Liposomes can be prepared through several methods present till date, with each one having its own pros and cons (Liu *et al.*, 2018; Shrimal *et al.*, 2020). Often, a combination

of these methods is used, resulting in multi-step procedures that complicate quality control across production batches. Ensuring consistent particle size and its distribution, surface charge, and release behavior of drug across batches presents quite a significant challenge. The scalability of these processes remains one of the major obstacles, as in order to meet the needs of liposomal products one has to customize the equipment used for their production involving specialized mechanical and engineering technologies (Worsham *et al.*, 2019).

Introduction to Bioinformatics: Bioinformatics made its way before DNA sequencing. The historical marks highlighted for its origin are the finding of DNA

structure by Watson and Crick back in 1953, which has been integrated into biotechnology yielding various applications with advancements including the database development for genomic and proteomics (Du *et al.*, 2023). It helps in the analyses of computational data for wet-lab, genome sequencing and identification of protein coding segments (Ghosh *et al.*, 2024). Due to its multidisciplinary character, bioinformatics can also be regarded as “the application of computational tools, used for the organization, understanding, visualization, analyses and information storage house for many applications (as shown in Figure 2) associated with biological macromolecules (Diniz *et al.*, 2017; Clark *et al.*, 2024).

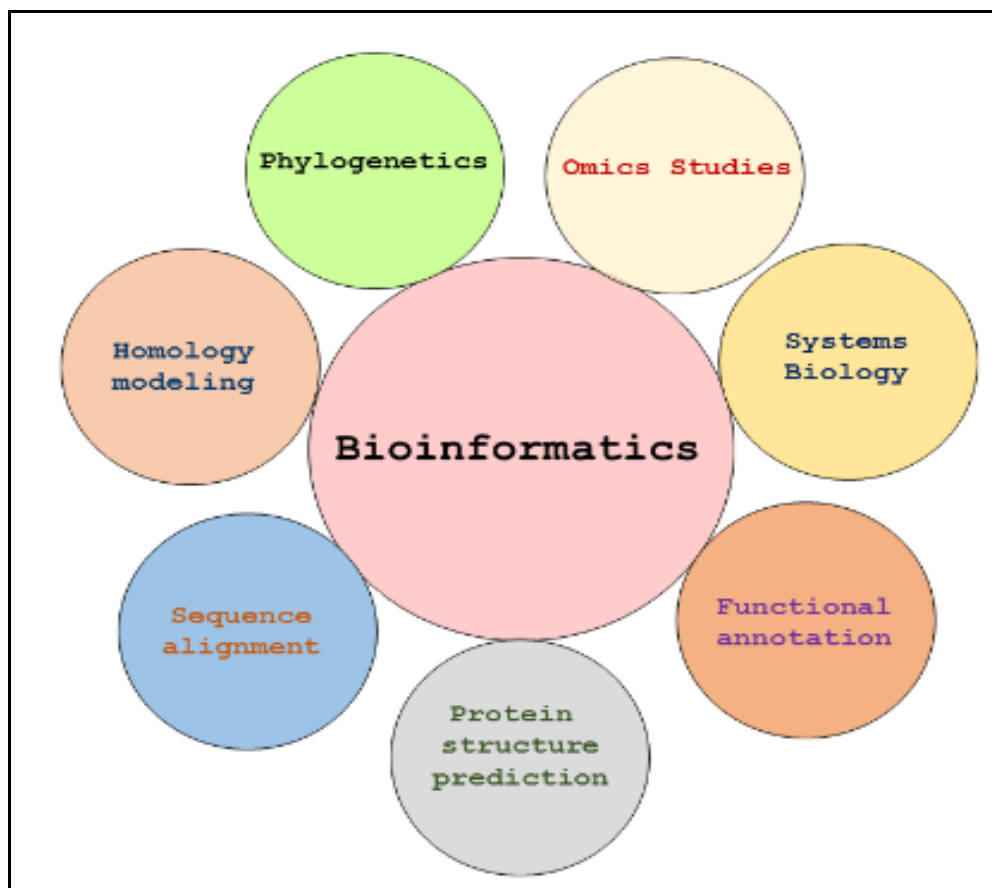


Figure 2: Different applications of Bioinformatics.

Bioinformatics with the aid of its tools helps to align and compare newly sequenced genomes with known genes, facilitating gene annotation and functional characterization, data retrieval and storage (Poetsch and Li, 2023). Bioinformatics is a relatively new field that connects the biology sciences and computer sciences. The enormous increase of biological sequence information necessitates the integration of these two fields. Thanks to the emerging technologies of Omics through which researchers are able to use different tools to examine the differences in proteins, DNA, RNA and other biological components among different species (Hasin *et al.*, 2017).

Bioinformatics Tools and Databases: Despite being a relatively young field, bioinformatics has made significant contributions to microbiology and biotechnology by developing innovative tools, algorithms, and discoveries that refine our understanding of microbial cell, its functions and behaviors. These bioinformatic tools shown in Figure 3 and databases shown in Figure 4 are vital sources for researchers as they aid in data retrieval, analysis and development of predictive models, These databases are also capable for managing, integrating and storing comprehensive information about gene’s location, function, annotation and expression analysis (Zhao *et al.*, 2023).

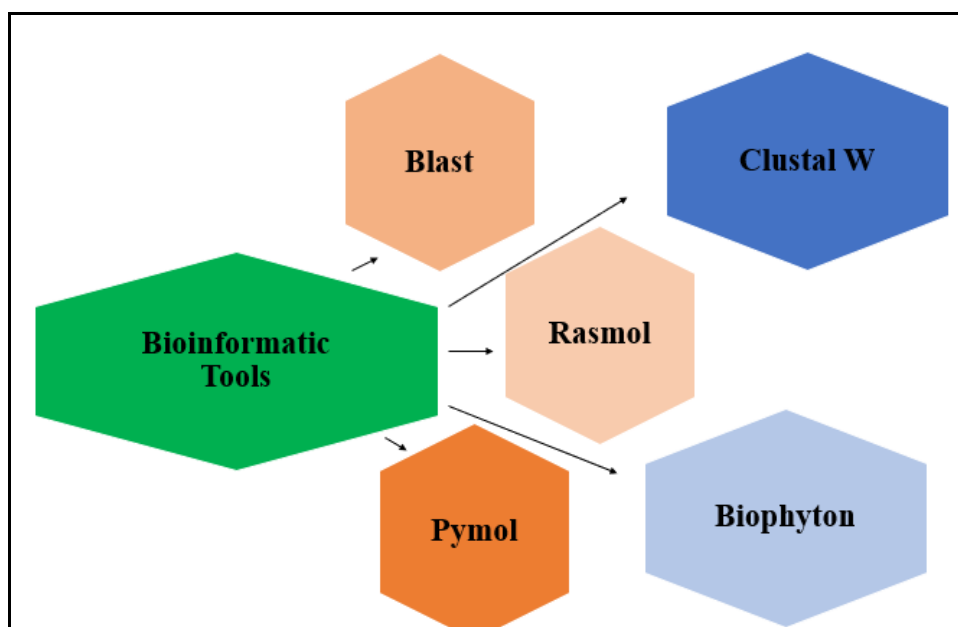


Figure 3: Overview of commonly used Bioinformatics tools.

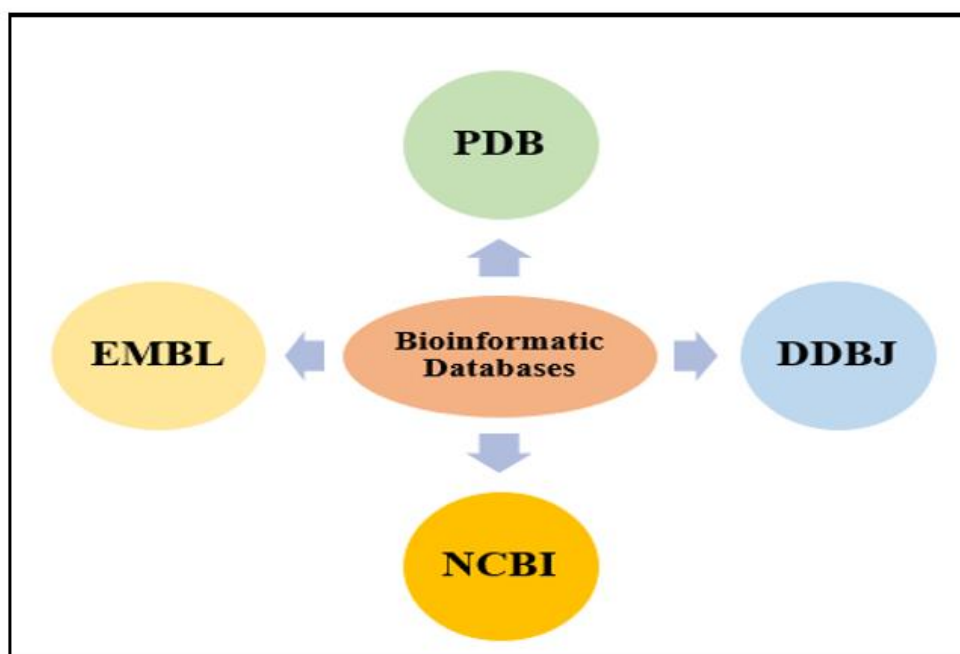


Figure 4. Some of the highly used Bioinformatic Data bases

Bioinformatics Integration in Drug Discovery: The convergence of bioinformatics and drug discovery has greatly transformed the pharmaceutical industry by enabling faster identification of promising drug candidates and optimizing their therapeutic potential (Mustafa *et al.*, 2023). Bioinformatics helps overcome a significant challenge in drug discovery by analyzing vast genomic and proteomic datasets to identify potential drug targets stealing the process of identification of suitable targets for therapeutic intervention (Xu *et al.*, 2023). Bioinformaticians can identify key molecules that are involved in disease

processes by analyzing gene expression patterns, protein-protein interactions, and metabolic pathways (Du *et al.*, 2023). This information is then used to design drugs, that specifically target these molecules, which boosts the likelihood of successful therapeutic outcomes (Paananen and Fortino 2020). Different steps are used to discover a particular drug using bioinformatics as shown in Figure 5. Another important aspect of drug discovery is the prediction of drug efficacy and safety (Reichel, A., & Lienau, 2016). Traditional *in vitro* or *in vivo* methods for drug testing are costly and time-consuming. Bioinformatics offers a

complementary approach, using computational models to predict the drug-target interactions. These models integrate a range of factors such as molecular structure, binding affinity, and pharmacokinetics to estimate the effectiveness and behavior of a drug candidate in the body. Furthermore, by prioritizing the top-performing

candidates, researchers can streamline the development processes for further testing, saving time, cost and resources (Malathi, K., & Ramaiah, S, 2018).

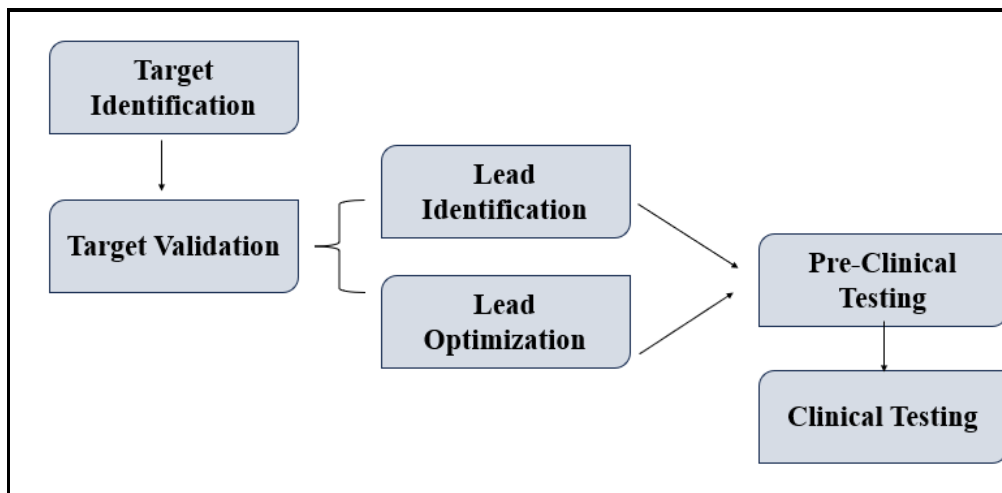


Figure 5. The crucial steps of drug discovery using bioinformatics approach.

Bioinformatic tools are essential for analyzing high-throughput screening data for drug discovery, shown in Figure 6. They help to identify and characterize various compounds required for drug discovery, using their metabolic profiles. The metabolic pathways to analyze repurposed drugs and the identification of potential off-target effects of present medications, leading to the discovery of new therapeutic applications are all driven by Bioinformatics reducing the financial barriers

(Tanoli, *et al.*, 2021; Amiri Ssouri *et al.*, 2023). To further relate the integration of bioinformatics in drug delivery one such case study reports the Molecular Dynamics for Lipid–Drug Interactions, which states that bioinformatics-based modeling can predict drug partitioning, membrane insertion, and stability, which guided optimization of lipid composition for improved encapsulation efficiency (Bunker *et al.*, 2021).

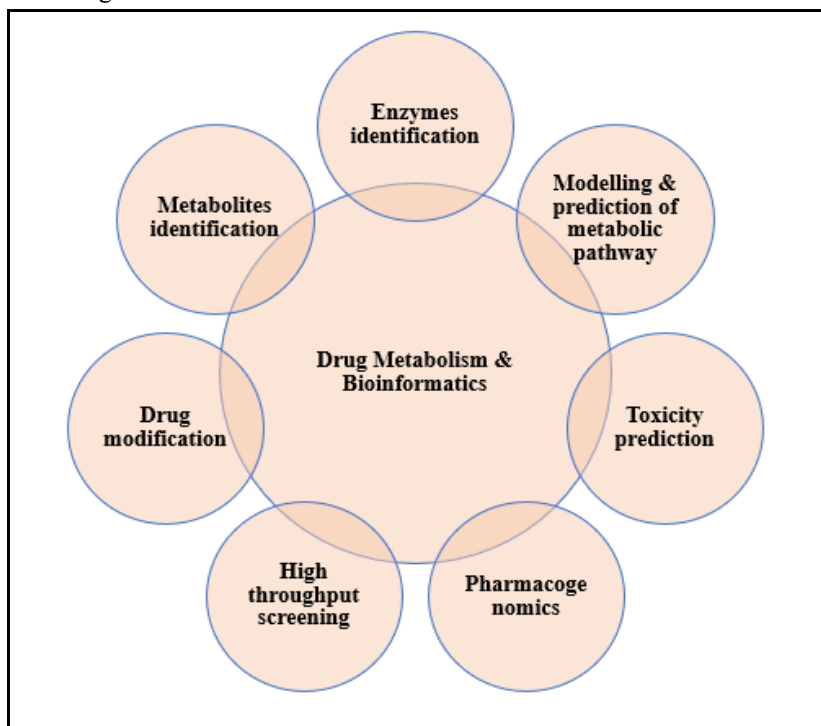


Figure 6. Drug metabolism and Bioinformatics

CONCLUSION AND FUTURE PROSPECTIVES

The integration of computational tools and algorithms with biological and chemical data has been fast-tracking the drug discovery process with high success rates and low cost. Drug effectiveness and safety can be optimized using the bioinformatic tools, this has given a tremendous increase in terms of drug discovery.

However, there still occurs many challenges, when dealing with drugs, which needs to be addressed at their earliest for the integration of bioinformatics into the drug discovery process. To overcome this issue there is a need to integrate and standardize the data in better ways using predictive models, for this purpose new data formats and ontologies must be developed for data integration and comparison following bioinformatic tools. Despite of the fact that machine learning has gained much importance till date for drug discovery, still improvement is required in the efficiency and reliability of the predictive models for more comprehensive datasets including; information on both positive and negative outcomes. Interdisciplinary collaboration remains another important factor found critical for the integration of bioinformatic tools in drug discovery. Drug discovery; a multidisciplinary field requires expertise from other fields including; chemistry, biology, computer science and pharmacology. To leverage the potential of bioinformatics in drug discovery, researchers from these different disciplines should come to one platform to work for its betterment. As a result of this cross-disciplinary collaboration, data is shared among expertise of all disciplines which leads to the development of new tools and methods tailored to meet the inimitable needs of drug discovery, accelerating innovation and progress in the field of drug delivery are developed. Furthermore, it can also be stated that frameworks such as the European Bioinformatics Institute (EMBL-EBI), the NIH's GenBank and PubChem, and integrative platforms like ELIXIR also play vital role in facilitating such collaboration by standardizing data formats and providing centralized access to genomic, chemical, and pharmacological datasets. These platforms enable cross-disciplinary knowledge sharing, foster the development of new predictive tools and methods, and ultimately bridge existing gaps, thereby accelerating innovation and advancing effective drug delivery systems.

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CONFLICT OF INTEREST

No conflict of interest among authors

REFERENCES

Abbasi, H., Kouchak, M., Mirveis, Z., Hajipour, F., Khodarahmi, M., & others. (2022). What we need

- to know about liposomes as drug nanocarriers: An updated review. *Advanced Pharmaceutical Bulletin*, 13(1), 7. <https://doi.org/10.34172/apb.2023.007>
- Hamid, M. S. S., Hatwar, P. R., Bakal, R. L., & Kohale, N. B. (2024). A comprehensive review on liposomes: As a novel drug delivery system. *GSC Biological and Pharmaceutical Sciences*, 27(1), 199–210. <https://doi.org/10.30574/gscbps.2024.27.1.0121>
- Li, M., Du, C., Guo, N., Teng, Y., Meng, X., Sun, H., & others. (2019). Composition design and medical application of liposomes. *European Journal of Medicinal Chemistry*, 164, 640–653. <https://doi.org/10.1016/j.ejmech.2019.01.007>
- Najlah, M., Said Suliman, A., Tolaymat, I., Kurusamy, S., Kannappan, V., Elhissi, A., & Wang, W. (2019). Development of injectable PEGylated liposome encapsulating disulfiram for colorectal cancer treatment. *Pharmaceutics*, 11(11), 594. <https://doi.org/10.3390/pharmaceutics11110610>
- Kanamala, M., Palmer, B. D., Jamieson, S. M., Wilson, W. R., & Wu, Z. (2019). Dual pH-sensitive liposomes with low pH-triggered sheddable PEG for enhanced tumor-targeted drug delivery. *Nanomedicine*, 14(15), 1971–1989. <https://doi.org/10.2217/nmm-2018-0510>
- Mohamed, M., Abu Lila, A. S., Shimizu, T., Alaaeldin, E., Hussein, A., Sarhan, H. A., & others. (2019). PEGylated liposomes: Immunological responses. *Science and Technology of Advanced Materials*, 20(1), 710–724. <https://doi.org/10.1080/14686996.2019.1627174>
- Gagliardi, M., & Ashizawa, A. T. (2021). The challenges and strategies of antisense oligonucleotide drug delivery. *Biomedicines*, 9(4), 433. <https://doi.org/10.3390/biomedicines9040433>
- Aghanejad, A., Omidian, H., & Omid, Y. (2021). Medical applications of multifunctional magnetic nanoparticles. In A. Ehrmann, T. A. Nguyen, M. Ahmadi, A. Farmani, & P. Nguyen-Tri (Eds.), *Magnetic nanoparticle-based hybrid materials* (pp. 447–462). Woodhead Publishing. <https://doi.org/10.1016/B978-0-12-823688-8.00001-6>
- Patel, S., & Ghosh, A. (2022). Integrating computational modeling with liposomal drug delivery systems. *Journal of Drug Targeting*, 30(4), 406–419. <https://doi.org/10.1080/1061186X.2021.1966691>
- Sharma, D., Ali, A. A. E., & Trivedi, L. R. (2018). An updated review on: Liposomes as drug delivery system. *PharmaTutor*, 6(2), 50–62. <https://doi.org/10.29161/PT.v6.i2.2018.50>
- Nakhaei, K., Bagheri-Hosseini, S., Sabbaghzade, N., Behmadi, J., & Boozari, M. (2023). Boswellic acid nanoparticles: Promising strategies for increasing therapeutic effects. *Revista Brasileira de*

- Farmacognosia*, 33(4), 713–723. <https://doi.org/10.1007/s43450-023-00405-7>
- Gonda, A., Zhao, N., Shah, J. V., Calvelli, H. R., Kantamneni, H., Francis, N. L., & Ganapathy, V. (2019). Engineering tumor-targeting nanoparticles as vehicles for precision nanomedicine. *Med One*, 4, e190021. <https://doi.org/10.20900/mo.20190021>
- Liu, P., Chen, G., & Zhang, J. (2022). A review of liposomes as a drug delivery system: Current status of approved products, regulatory environments, and future perspectives. *Molecules*, 27(4), 1372. <https://doi.org/10.3390/molecules27041372>
- Aslam, M., Iqtedar, M., Saeed, H., Abdullah, R., & Kaleem, A. (2022). Formulation and characterization of ciprofloxacin encapsulated liposomes: In vitro antimicrobial activity against multi-drug resistant *Salmonella typhi*. *Revista Mexicana de Ingeniería Química*, 21(2), Bio2734. <https://doi.org/10.24275/rmiq/Bio2734>
- Nsairat, H., Khater, D., Sayed, U., Odeh, F., Al Bawab, A., & Alshaer, W. (2022). Liposomes: Structure, composition, types, and clinical applications. *Heliyon*, 8(5), e09387. <https://doi.org/10.1016/j.heliyon.2022.e09394>
- Lombardo, D., & Kiselev, M. A. (2022). Methods of liposomes preparation: Formation and control factors of versatile nanocarriers for biomedical and nanomedicine application. *Pharmaceutics*, 14(3), 543. <https://doi.org/10.3390/pharmaceutics14030543>
- Yan, W., Leung, S. S., & To, K. K. (2020). Updates on the use of liposomes for active tumor targeting in cancer therapy. *Nanomedicine*, 15(3), 303–318. <https://doi.org/10.2217/nnm-2019-0308>
- Cauzzo, J., Jayakumar, N., Ahluwalia, B. S., Ahmad, A., & Škalko-Basnet, N. (2021). Characterization of liposomes using quantitative phase microscopy (QPM). *Pharmaceutics*, 13(5), 590. <https://doi.org/10.3390/pharmaceutics13050590>
- Fan, Y., Marioli, M., & Zhang, K. (2021). Analytical characterization of liposomes and other lipid nanoparticles for drug delivery. *Journal of Pharmaceutical and Biomedical Analysis*, 192, 113642. <https://doi.org/10.1016/j.jpba.2020.113642>
- Torres-Flores, G., Gonzalez-Horta, A., Vega-Cantu, Y. I., Rodriguez, C., & Rodriguez-Garcia, A. (2020). Preparation and characterization of liposomal everolimus by thin-film hydration technique. *Advances in Polymer Technology*, 2020(1), 5462949. <https://doi.org/10.1155/2020/5462949>
- Man, F., Gawne, P. J., & de Rosales, R. T. (2019). Nuclear imaging of liposomal drug delivery systems: A critical review of radiolabelling methods and applications in nanomedicine. *Advanced Drug Delivery Reviews*, 143, 134–160. <https://doi.org/10.1016/j.addr.2019.05.012>
- Almeida, B., Nag, O. K., Rogers, K. E., & Delehanty, J. B. (2020). Recent progress in bioconjugation strategies for liposome-mediated drug delivery. *Molecules*, 25(23), 5672. <https://doi.org/10.3390/molecules25235672>
- El-Hammadi, M. M., & Arias, J. L. (2019). An update on liposomes in drug delivery: A patent review (2014–2018). *Expert Opinion on Therapeutic Patents*, 29(11), 891–907. <https://doi.org/10.1080/13543776.2019.1679767>
- Pauli, G., Tang, W.-L., & Li, S.-D. (2019). Development and characterization of the solvent-assisted active loading technology (SALT) for liposomal loading of poorly water-soluble compounds. *Pharmaceutics*, 11(9), 465. <https://doi.org/10.3390/pharmaceutics11090465>
- Tamam, H., Park, J., Gadalla, H. H., Masters, A. R., Abdel-Aleem, J. A., Abdelrahman, S. I., et al. (2019). Development of liposomal gemcitabine with high drug loading capacity. *Molecular Pharmaceutics*, 16(7), 2858–2871. <https://doi.org/10.1021/acs.molpharmaceut.8b01284>
- Pande, S. (2023). Liposomes for drug delivery: Review of vesicular composition, factors affecting drug release and drug loading in liposomes. *Artificial Cells, Nanomedicine, and Biotechnology*, 51(1), 428–440. <https://doi.org/10.1080/21691401.2023.2247036>
- Vakili-Ghartavol, R., Rezayat, S. M., Faridi-Majidi, R., Sadri, K., & Jaafari, M. R. (2020). Optimization of docetaxel loading conditions in liposomes: Proposing potential products for metastatic breast carcinoma chemotherapy. *Scientific Reports*, 10(1), 5569. <https://doi.org/10.1038/s41598-020-62501-1>
- Guimarães, D., Cavaco-Paulo, A., & Nogueira, E. (2021). Design of liposomes as drug delivery system for therapeutic applications. *International Journal of Pharmaceutics*, 601, 120571. <https://doi.org/10.1016/j.ijpharm.2021.120571>
- Farooque, F., Wasi, M., & Mughees, M. M. (2021). Liposomes as drug delivery system: An updated review. *Journal of Drug Delivery and Therapeutics*, 11, 149–158. <https://doi.org/10.22270/jddt.v11i5-S.5063>
- Basu, B., Prajapati, B., Dutta, A., & Paliwal, H. (2024). Medical application of liposomes. *Journal of Exploratory Research in Pharmacology*, 9(1), 13–22. <https://doi.org/10.14218/JERP.2023.00002>
- Kumbham, S., Ajjarapu, S., Ghosh, B., & Biswas, S. (2023). Current trends in the development of liposomes for chemotherapeutic drug delivery. *Journal of Drug Delivery Science and Technology*, 87, 104854. <https://doi.org/10.1016/j.jddst.2023.104854>
- Peng, T., Xu, W., Li, Q., Ding, Y., & Huang, Y. (2023). Pharmaceutical liposomal delivery—Specific considerations of innovation and challenges.

- Biomaterials Science*, 11, 62–75.
<https://doi.org/10.1039/D2BM01252A>
- Sawant, R. R., & Torchilin, V. P. (2012). Challenges in development of targeted liposomal therapeutics. *AAPS Journal*, 14, 303–315.
<https://doi.org/10.1208/s12248-012-9330-0>
- Chen, T., He, B., Tao, J., He, Y., Deng, H., Wang, X., et al. (2019). Application of Förster resonance energy transfer (FRET) technique to elucidate intracellular and in vivo biofate of nanomedicines. *Advanced Drug Delivery Reviews*, 143, 177–205.
<https://doi.org/10.1016/j.addr.2019.04.009>
- Liu, D., Zhang, H., Fontana, F., Hirvonen, J. T., & Santos, H. A. (2018). Current developments and applications of microfluidic technology toward clinical translation of nanomedicines. *Advanced Drug Delivery Reviews*, 128, 54–83.
<https://doi.org/10.1016/j.addr.2017.08.003>
- Shrimal, P., Jadeja, G., & Patel, S. (2020). A review on novel methodologies for drug nanoparticle preparation: Microfluidic approach. *Chemical Engineering Research and Design*, 153, 728–756.
<https://doi.org/10.1016/j.cherd.2019.11.031>
- Du, Z., Comer, J., & Li, Y. (2023). Bioinformatics approaches to discovering food-derived bioactive peptides: Reviews and perspectives. *TrAC Trends in Analytical Chemistry*, 162, 117051.
<https://doi.org/10.1016/j.trac.2023.117051>
- Ghosh, R., Bera, R. D., & Chandra, S. (2024). An overview of bioinformatics and its application: A ray of hope towards microbial biotechnology. *International Journal of Multidisciplinary Research*, 6(1), 1–23.
<https://doi.org/10.36948/ijfmr.2024.v06i01.13726>
- Diniz, W. J. d. S., & Canduri, F. (2017). Bioinformatics: An overview and its applications. *Genetics and Molecular Research*, 16(1), 17.
<https://doi.org/10.4238/gmr16019645>
- Clark, A. J., & Lillard, J. W., Jr. (2024). A comprehensive review of bioinformatics tools for genomic biomarker discovery driving precision oncology. *Genes (Basel)*, 15(8), 1036.
<https://doi.org/10.3390/genes15081036>
- Poetsch, A., & Li, Y. (2023). Multi-Omics data types and underlying technology. In *Methodologies of Multi-Omics Data Integration and Data Mining: Techniques and Applications* (Vol. 19, pp. 33). Springer. <https://doi.org/10.1007/978-981-19-8210-1>
- Hasin, Y., Seldin, M., & Lusis, A. (2017). Multi-omics approaches to disease. *Genome Biology*, 18(1), 1–15. <https://doi.org/10.1186/s13059-017-1215-1>
- Zhao, X., Zhao, H., Liu, Y., & Guo, Z. (2023). Methods, bioinformatics tools and databases in ecDNA research: An overview. *Computers in Biology and Medicine*, 167, 107680.
<https://doi.org/10.1016/j.combiomed.2023.107680>
- Mustafa, M., Mohammed, A., Alzebair, A., & Makhawi, A. (2023). Integrating bioinformatics in drug discovery. *Preprints*.
<https://doi.org/10.20944/preprints202310.1465.v1>
- Xu, M., Xu, C., Chen, M., Xiao, Z., Wang, Y., Xu, Y., et al. (2023). Comparative analysis of commonly used bioinformatics software based on omics. *Gene Reports*, 32, 101800.
<https://doi.org/10.1016/j.genrep.2023.101800>
- Paananen, J., & Fortino, V. (2020). An omics perspective on drug target discovery platforms. *Briefings in Bioinformatics*, 21(6), 1937–1953.
<https://doi.org/10.1093/bib/bbz122>
- Reichel, A., & Lienau, P. (2016). Pharmacokinetics in drug discovery: An exposure-centred approach to optimising and predicting drug efficacy and safety. In *New Approaches to Drug Discovery* (pp. 235–260). Springer.
https://doi.org/10.1007/164_2015_26
- Malathi, K., & Ramaiah, S. (2018). Bioinformatics approaches for new drug discovery: A review. *Biotechnology and Genetic Engineering Reviews*, 34(2), 243–260.
<https://doi.org/10.1080/02648725.2018.1502984>
- Tanoli, Z., Seemab, U., Scherer, A., Wennerberg, K., Tang, J., & Vähä-Koskela, M. (2021). Exploration of databases and methods supporting drug repurposing: A comprehensive survey. *Briefings in Bioinformatics*, 22(2), 1656–1678.
<https://doi.org/10.1093/bib/bbaa003>
- Amiri Souri, E., Chenoweth, A., Karagiannis, S. N., & Tsoka, S. (2023). Drug repurposing and prediction of multiple interaction types via graph embedding. *BMC Bioinformatics*, 24(1), 202.
<https://doi.org/10.1186/s12859-023-05317-w>
- Eugster, R., Orsi, M., Buttitta, G., Serafini, N., Tiboni, M., Casertari, L., Reymond, J.-L., Aleandri, S., & Luciani, P. (2024). Leveraging machine learning to streamline the development of liposomal drug delivery systems. *Journal of Controlled Release*, 376, 1025–1038.
<https://doi.org/10.1016/j.jconrel.2024.10.065>
- Suk, J. S., Xu, Q., Kim, N., Hanes, J., & Ensign, L. M. (2016). PEGylation as a strategy for improving nanoparticle-based drug and gene delivery. *Advanced Drug Delivery Reviews*, 99(Pt A), 28–51. <https://doi.org/10.1016/j.addr.2015.09.012>
- Liu, P., Chen, G., & Zhang, J. (2022). A review of liposomes as a drug delivery system: Current status of approved products, regulatory environments, and future perspectives. *Molecules*, 27(4), 1372.
<https://doi.org/10.3390/molecules27041372>
- Smits, E. A., Soetekouw, J. A., Pieters, E. H., Smits, C. J., de Wijs-Rot, N., & Vromans, H. (2019). The availability of drug by liposomal drug delivery: Individual kinetics and tissue distribution of encapsulated and released drug in mice after administration of PEGylated liposomal prednisolone phosphate. *Investigational New Drugs*, 37(5), 890–901.
<https://doi.org/10.1007/s10637-019-00788-8>

- Bunker, A., Magarkar, A., & Viitala, T. (2021). Rational design of liposomal drug delivery systems using molecular dynamics simulations. *Physical Chemistry Chemical Physics*, 23(4), 2141–2157. <https://doi.org/10.1039/D0CP05901A>
- Worsham, R. D., Thomas, V., & Farid, S. S. (2019). Potential of continuous manufacturing for liposomal drug products. *Biotechnology Journal*, 14(1), 1700740. <https://doi.org/10.1002/biot.201700740>
- Han, R., Ye, Z., Zhang, Y., Cheng, Y., Zheng, Y., & Ouyang, D. (2023). Predicting liposome formulations by the integrated machine learning and molecular modeling approaches. *Asian Journal of Pharmaceutical Sciences*, 18(4), 100811. <https://doi.org/10.1016/j.ajps.2023.100811>
- Bulbake, U., Doppalapudi, S., Kommineni, N., & Khan, W. (2017). Liposomal formulations in clinical use: An updated review. *Pharmaceutics*, 9(2), 12. <https://doi.org/10.3390/pharmaceutics9020012>
- FDA. (2018). *Liposome drug products: Chemistry, manufacturing, and controls; human pharmacokinetics and bioavailability; and labeling documentation*. U.S. Food and Drug Administration. <https://www.fda.gov/media/70837/download>
- Jia, C., Zhang, Y., Wang, Y., Gao, J., Raza, A., Ogawa, T., ... Wang, J. Y. (2023). Positively charged and neutral drug-loaded liposomes as aerosolized formulations for the efficiency assessment of tumor metastases to lungs. *Journal of Drug Delivery Science and Technology*, 80, 104081. <https://doi.org/10.1016/j.jddst.2023.104081>
- Pisani, S., Di Martino, D., Cerri, S., Genta, I., Dorati, R., Bertino, G., Perteghella, S., Bruni, G., Modena, T., & Conti, B. (2023). Investigation and comparison of active and passive encapsulation methods for loading proteins into liposomes. *International Journal of Molecular Sciences*, 24(17), 13542. <https://doi.org/10.3390/ijms241713542>
- Aslam, M., Iqtadar, M., Saeed, H., Abdullah, R., & Kaleem, A. (2022). Formulation and characterization of ciprofloxacin encapsulated liposomes: In vitro antimicrobial activity against multidrug resistant *Salmonella typhi*. *Revista Mexicana de Ingeniería Química*, 21(2), Bio2734. <https://doi.org/10.24275/rmiq/Bio2734>
- Allen, T. M., & Cullis, P. R. (2013). Liposomal drug delivery systems: From concept to clinical applications. *Advanced Drug Delivery Reviews*, 65(1), 36–48. <https://doi.org/10.1016/j.addr.2012.09.037>
- Maja, L., Željko, K., & Mateja, P. (2020). Sustainable technologies for liposome preparation. *The Journal of Supercritical Fluids*, 165, 104984. <https://doi.org/10.1016/j.supflu.2020.104984>
- Lu, H., Zhang, S., Wang, J., & Chen, Q. (2021). A review on polymer and lipid-based nanocarriers and its application to nano-pharmaceutical and food-based systems. *Frontiers in Nutrition*, 8, Article 783831. <https://doi.org/10.3389/fnut.2021.783831>

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