Anti-Neoplastic Drug-Biomembrane Interaction Studies through Bio Membrane Models and Their Utility in Drug Design and Development

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Abstract: *Drug molecule experiences variety of interactions with bio membranes once it is administered in the body. Bio membranes too experiences changes in the strategic parameters like permeability, charge transfer and fluidity on interactions with the drug molecule. Pharmacokinetics as well as Pharmacodynamics gets affected. Understanding of these interactions is very important as it may lead into the development of new antineoplastic drugs or can improve upon the efficacy profile of existing Drug molecule. However studying drug bio membrane interactions in live membranes may cause damage to live membrane. Artificial in vitro and in silico membrane systems, help in understanding of various factors and barriers involved in drug transport and uptake in to cells, by exactly mimicking the Bio membrane*

Keywords: *Drug, Bio membrane, in vitro, in silico, transport*

I. Introduction

Drug molecule encounters many of the bio membranes after its administration in the body, from macrophage cells to vessel endothelium to blood brain or blood retinal barriers. Time and concentration plays the most important role in drug distribution inside the body. Plethora of interactions are experienced by drug molecule as well as by bio membrane. These interactions can have reciprocal results, on one hand molecules have the capability to alter the structure and function of bio membrane in terms of permeability charge transfer, fluidity and so on, bio membranes can also affect the structure of bio molecules in terms of stereo specificity and other properties. Understanding of these changes are very crucial as proper understanding may result into the synthesis of better drug molecule, existing drug species can also be improved by improving upon the existing structure. However studying these interactions in live membranes may harm the membrane . Artificial model membrane systems, help in the understanding of various factors and barriers involved in drug transport and uptake into cells, by exactly mimicking the natural bio membranes.

II. Bio Membrane Models

Bio membrane models provide the perfect simulating body environment in vitro to drug molecule by mimicking the arrangement of lipids in natural cell membrane by lipid organizations artificially. Supported lipid bilayers, lipid monolayers, and liposomes tops the list of bio membrane models.

Francis Szokaand Demetriospapahadjopoulos [1]reported the formation of Large unilamellar and oligolamellar vesicles by introducing an aqueous buffer in to a mixture of phospholipid and organic solvent. Organic solvent is then removed by evaporation under reduced pressure

Thor D.Osborn and Paul Yager [2] has prepared Planar Solvent –Free phospholipid Bilayers by transferring Langmuir –Blodgett monolayers to micro machined apertures in Silicon, they used mass bilayer producible support device ,which consisted of an array of cavities in silicon substrate sealed on the back side by an anodically bounded piece of pyrex glass bearing Ag/AgCl electrodes to prepare reproducible bilayers .

Brezesinski G, Mohwald H. [3] used Landmuir monolayers to study interactions at model membrane surfaces. They studied interactions at interfaces beyond classical DLVO description.they explained that activity depends upon membrane structure. Reaction product may lead to structural changes in the monolayer leading to termination of reaction, indicating a subtle case of product inhibition via the membrane. They reported the possibility to manipulate the organization of polyelectrolytes at interfaces via lipid charge density and ionic strength.

Hubert Bader , Klaus Dorn, Bernd Hupfer, Helmut Ringsdorf [4] emphasized that Cell-cell recognition is of vital importance in immunological reactions . polymeric vesicles carrying sugar head groups can easily mimickbiomembranes . They made attempts to unite biological specifications of natural cells and toughness of polymerized membranes via cell vesicle fusion.

A technique for the rapid production of large unilamellar vesicles by repeated extrusion under moderate pressures of multilamellar vesicles through polycarbonate filters (100 nm pore size) is demonstrated by M.J.Hope,M.B.Bally, G.Webb and P.R.Cullis [5].The advantage of generating LUVET's' is the complete

absence of organic solvents and a very fast vesicle formation rate.The procedure is friendly with wide variety of lipid species and mixtures

Storage and stability of Bio membrane Models

*Li*posomes are used as common bio membrane models, their aqueous formulations are susceptible to chemical degradation .Hernandez –Caselles, Villalain J and Gomez Fernandez JC and other workers [6, 35] studied the effect of lipid composition of Liposomes on their storage for up to one year under different environmental conditions using 5,6- carboxyfluorescein as a model drug and reported that Liposome stability is enhanced when stored at 4 degree c or in an O2-free atmosphere.

D.J.A.Crommelin, Gal.J.Fransen and P.J.M.[7] reported three important conepts of aqueous dispersion, freeze drying and removal of free water via evaporation for the enhanced stability and storage of Liposome. They further underlined the causes of unstability , which could be aggregation or fusion processes.

Atomic Force microscopy and Surface force apparatus is used as a tool to study the topography of phospholipid bilayers. Marcel Benz, Thomas Gutsmann, Nianhuan, RafelTadmor and Jacob Israelachvili [8] described that the amount of irregularities in the topography of membrane could be studied by the hemifusion in the SFA, which is an indicator of defective bilayer. Langmuir Blodgett deposition was used for the preparation of bio membrane and then transferring at different surface pressures. They convincingly reported that defects appearing in the form of thick holes in monolayer and bilayer were energetically favorable over an evenly depleted bilayer

Drug Bio Membrane Interactions/in vitro studies

During its interaction with Bio membrane Drug molecule may suffer changes in terms of stereochemistry, molecular conformation, time of onset, and biological activity. The forces underlying these both kinds of interactions i.e Pharmacokinetics and Pharmacodynamics are same, that is, polar and hydrophobic chemical interactions.

Determination of monolayer structures and interaction between amphiphillic monolayers at the soft air/liquid interface and molecules dissolved in the sub phase are very important in material and life Sciences.Stefaniu C, Brezesinki G, Mohwald H [9] utilized Langmuir monolayers as models to study processes at membrane surfaces. Monolayer structure changes are followed in situ by X Ray diffraction studies and Infra red reflection absorption spectroscopy ,interactions can be studied by following the structural changes in the membrane, changes in the secondary structure of peptides and proteins and observing the yield of enzymatic reactions.The influence of these variations on the structural properties of bio molecules can also be determined, clearly defining the influence of modified nanoparticle on bio membrane model

Seydel JK, Coats EA, Cordes HP and Wiese M.and many other authors[10,11,21,22] discussed important aspects of important drug membrane interactions. They also discussed the influence of these interactions on drug transport, accumulation , efficacy and resistance. Interactions macroscopically bring changes in the physical statistics and thermodynamics of pure membranes or bilayers. The interactions may bring changes into gel to liquid crystalline phase, thus bringing a change in the permeability,cell fusion, cell resistance and may also lead in to conformational changes in the embedded receptor proteins. Specific lipid – Drug interactions may lead into the accumulation of Drug in the membrane providing very high concentration of Drug at the active site, than in the surrounding water phase.Team also predicted the conformational changes in the Drug structure. Bouquetteof all these interactions finally help in designing new improved drug molecule with improved efficacy profiles.

Cinzia A Ventura and many others have reported [12,23, 36-41] the use of Differential Scanning calorimetry as simple invasive technique to study the interaction of moxifloxacin with Bio membrane.They reported the free permeation of drug through liposomes, however decrease in ΔH value and loss of cooperativityof the main transition peak suggest the interaction of drug molecules with the hydrophobic zone of bilayer

Drug release from an insulin based hydrogel and its interaction with bio membrane is studied by Francesco Castelli, Maria GraziaSarpietro and team[13] and other workers [17] using Differntial Scanning Calorimetry as technique, it was observed after studying the release profile of characterized hydrogel that release not only depend on the drug loading , but depends enormously on the pH of the external environment.

Francesco Casterlli, Giovanni Puglisi, Rosario Pignatello and Salvatore Gurrieri [14]observed main transition peak T_m values when different molar amounts of 4-biphenylacetic acid (BPAA) or β- cyclodextrin-4-biphenylacetic acid(BPAA-β Cyd) are adhift in the ded to phospholipidic dispersions through calorimetric studies.DSC was used for these studies. Increased shift in the T_m values was interpreted in terms of increasing membrane fluidity. The study emphasized the possible interaction of BPAA with phospholipid in cell membranes, the potential effects of such interactions is in the modulation of membrane and in the regulation of fluidity of membrane.

A.AVdeef,K.J.Box, J.E.A Comer, C.Hibbert and K.Y.Tam[15],reported Ph-metric technique for the determination of Liposomal membrane-water partition coefficients and lipophilicity profile of ionizable drugs. The partition coefficients were determined for 8 drugs and found out to be in ranges of approximately 4.5 to 2.4 and 2.6 to 0.8 logarithmic units, which is in tune with other alternative methods like ultra-filtration and dialysis.

Chromatography was used to study Drug partitioning into Lipid bilayers by FaridehBeigi,Ingo Gottschalk, Christine LagerquistHagglund, Lars Haneskog et.al.[16]Chromatography was performed on liposomes and bio membranes immobilized in gel beads by freeze thawing.The drug retention factor was calculated and expressed as a capacity factor K_s for both liposomes and vesicles.

Atomic force microscopy has been used as premier technique to view dynamic interactions of proteins and a model lipid membrane. Quinn A S, Rand JH, Wu XX and Taaties D J [18] has conveniently used the technique with more than one type of lipid study. The AFM imaging protocols have been supplemented by procedures of ellipsometry, standardized western blotting and dot blots to verify appropriate purity and activity of all experimental molecular components.

Understanding the chemical composition ,structure and dynamics of membrane is of utmost importance Molecular biologists, Chemists and Medicinal Chemistry practitionersDifferent researchers have given myriad of techniques for studying the structure of Bio membranes which include labeling techniques as well as novel label free techniques.[19]

The antineoplastic drugs have proved to be a boon in the cancer management and treatment therapy, drug- bio membrane interactions again play an important role for drug transport, accumulation and activity. Investigation ofthese interactions are very difficult because of the complexity of biological membranes. Membranes can sequester drug molecules, or conversely may facilitate free diffusion, thereby modulating the accumulation of drugs into cells. Efflux pump is also affected. Antineoplastic drugs can alter the structure and properties of bio membranes.

Alicia Jacquemet,CristelleMeriadec, LoicLemiegre et.al [20]. performed SXAS Studies of two synthetic diastereomericarchaeal lipids bearing two lactosyl polar head groups at opposite ends revealed different lyotropicbehaviours.Thecis isomer led to Lc-Lα-QII transitions where as the trans isomer retained an Lα phase from 20 to 100℃.The cause of difference is attributed to conformational equilibrium(pseudorotation)of 1,3-disubstituted cyclopentanes.This pseudorotation exhibits quite similar orientation in trans isomers where as several orientations are expected in cis isomer

Antineoplastic drugs prevents or inhibits the maturation and proliferation of neoplasm. Different anticancer drugs clinically approved by international regulatory organizations present poor water solubility and low stability after systemic injection. The interactions of anticancer drugs with cell membranes are of primary importance for drug transport, accumulation and activity. Cell membranes are complex dynamic systems whose structures can be affected by drug molecules and in turn can affect the pharmacological properties of the drugs being administered. Cisplatin is an effective antineoplastic drug, which provides a perfect example of how small changes in the molecular structure of drug can lead to profound differences in biological activity [24, 25].Cisplatin loses two chloride ions inside the cell, creating a reactive species that form bonds with DNA Bases.

Boer D.R., Canals A, Coll M.[26] reported different unprecedented ways of targeting peculiar DNA structures such as junctions, quadruplexes and duplex DNAs.They reported that there are DNA binding agents which influence the field of DNA targeting.Classical binding modes of small molecular weight compounds to DNA, i.e groove binding, intercalation and covalent addition are discussed.

Wilmer Villarreal,LegnaColina-Vegas,Clayton Rodrigues de Oliveira et. al.[27] reported Chirality as an important Characteristic of drug which acts as a DNA Binding agent .A series of chiral platinum(II) complexes featuring phosphine and chloroquine ligands were prepared. Modification of quinolinic ligands and combination of NMR and XRD experimentsatropisomeric phenomenon exhibited by complex is demonstrated, identifying the necessary presence of an additional asymmetric center to generate diastereomers. The compounds were efficient with regard to inhibiting colony formation, as well as the proliferation and migration of MDA-MB-231 tumor cells.

Platinum complex are clinically used as potential drug molecules against Cancer. Cisplatin is used as a drug against cancer, but its biochemical mechanism is still not clear. Depending on cell type, concentration , dose, interference with DNA and many other factors, cisplatin induces cytotoxicity in non tumor cells while damaging tumor cells through apoptosis or necrosis [28,29]

III. In Silico Studies

Poor pharmacokinetics and toxicity are the most important causes of costly late stage failures in drug development process. Achievement of an early data on absorption,distribution, metabolism and excretion of the potential drug candidate is very useful. In Silico studies further increase our ability to predict and model the most relevant pharmacokinetic, metabolic and toxicity end points,there by increasing the drug discovery process

[30,31] .The rate of non facilitated permeation of solutes across lipid bilayers is very important for drug design and discovery.These rates can be assessed by using molecular dynamics simulations combined with inhomogeneous solubility diffusion models, requiring calculations of the potential of mean force and position dependent diffusivity of solute alongthetransmembrane axis.Cristopher T. Lee, Jeffrey Comer, Conner Herndon, Nelson Leung et al.[34] assessed the efficiency and accuracy of several methods for the calculation of permeability of a model DMPC bilayer through different simulative methods. QSAR studies relates quantitative chemical structure attributes to biological activity. QSAR studies have become an important technique in drug discovery and development as their application saves substantial timeand human resource. Linear or Non linear behavior of data set can be checked by applying different statistical techniques, where as feature selection techniques are applied to decrease the model complexity. QSAR helps in the designing of new compounds with improved potency and helps in synthesizing compounds with desirable activity [32]

IV. Conclusion

Drug Biomembrane interactions can be studied through biomembrane models, prepared in the lab.In vitro and in silico studies can performed.the results obtained thereof could be utilized for the design and development of new drug molecule. In silico studies save costly financial, time as well as human resource,as with these studies it is possible to avoid late stage failures in the tedious laboratory procedures.

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