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Insilico Study of Surfactants Used in Formulation Development as Permeation Glycoprotein Inhibitor Potential

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Abstract
INTRODUCTION: Microbial multidrug resistance (MDR) has become one of the key treatments in many medication regimens throughout the past few decades. The pharmaceutical industry, the animal husbandry industry, and the agriculture industry have all been somewhat impacted as a result of this phenomenon.
MATERIAL AND METHODS: The molecular docking studies with specified ligands were carried out using the Schrodinger Maestro 9.1 software programme. Protein preparation wizard was used to prepare the selected receptors.
RESULTS: The docking simulations revealed the unusual importance of numerous elements in the protein ligand interaction profile, such as hydrogen bonds, lipophilic contacts, metal interactions, pi-pi interaction, and pi-cation interaction. Scoring functions are quick approximation mathematical algorithms used in computational chemistry and molecular modeling to predict the intensity of non-covalent contact between two molecules after they have been docked.
CONCLUSION: The findings of this study may aid in understanding the molecular mechanism of these excipients' possible P-gp inhibitory activity. The current findings will be validated further by formulation development with any P-gp substrate drug molecule, as well as in vitro and in vivo studies for ultimate confirmation.

1. Introduction

Colorectal cancer is a disease characterized by abnormal division of cells in the colon and rectal region causing an uncharacteristic change in bowel habits[1].In many Arab countries including Saudi Arabia, colorectal cancer is considered as one of the major causes of mortality among cancer patients[2]. Individuals of age 35-65 years are at higher risk to confront the colorectal cancer. Risk factors of the said disease rang from sedentary lifestyle to exposure to chemical fumes emerging from factories[3]. Cytokines on the other hand are also considered to contribute for the pathophysiological conditions of the patients. Cytokines are proteins secreted by cells and that are responsible for the growthand activity of other cells in the immune system. Pro-inflammatory cytokines are produced predominantly by activated macrophages and are involved in the up regulation of inflammatory reactions. Evidence shows that certain pro-inflammatory cytokines are involved in the process of pathological pain [4-6]. Interleukins and transforming growth factor B stimulate cancer cells proliferation and inversion [7]. Cytokine's receptors activation and intercellular sign allying accelerate tumor progression

A permeability glycoprotein is known as P-gp, which stands for multidrug resistance protein 1. P-gp is a major membrane transporter that plays a significant role in the metabolism of foreign particles and the subsequent efflux of these particles out of the cell. P-gp is responsible for the removal of foreign particles once they have been metabolized. P-gp's substrate-dependent efflux action can only occur in the presence of adenosine triphosphate, also known as ATP. It is believed that this protein plays a part in the process of defence against foreign organisms or chemicals. It has been discovered in a wide variety of microorganisms, including fungi, mammals, and bacteria, and its presence lends credence to this theory. P-gp is widely distributed throughout the body and plays a key part in the efflux processes of the gut, bile ducts and liver cells, kidney cells (such as proximal tubules), and capillary endothelium, which are essentially endothelial cells and include the blood-testis barrier and the blood-brain barrier. It has been discovered not just in the colon but also in other organs such as the pancreas and the adrenal glands. P-gp is typically overexpressed in cancer cells, which blocks the entry of a large number of anticancer medications and reduces the efficacy of treatment for cancer. However,

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it is produced in the lumen of the digestive system by the bile ducts, which means that it shields tissues from potentially dangerous noxious components and also helps in the elimination of metabolites. P-gp expression has been found to a significant extent in the vast majority of cancer cases, irrespective of the kind of malignancy. Chemotherapy treatments for breast cancer frequently fail because of drug resistance [7,8]. The epithelial lining of the ducts (which accounts for 85% of breast cancer cases) or the lobules, which accounts for 15% of breast cancer cases, is the location of cancer development in the majority of breast cancer instances. When cancer first begins to form, it is usually contained (also known as "in situ") within a duct or lobule, and it only rarely produces symptoms or spreads to other areas of the body. These in situ tumours have the potential to metastasis over time, eventually penetrating the breast tissue of nearby neighbours and spreading to lymph nodes in the region as well as further afield. The death of a woman from breast cancer is invariably the result of the cancer having progressed throughout the body [9,10]. The expression of proteins was investigated with immunohistochemistry in a number of different labs. Protein expression levels of Pgp, Ki-67, and p53 were significantly higher in locally advanced breast cancers, in contrast to the majority of breast tumours that could be surgically removed that were reported in the various trials [11,12]. Numerous excipients substances, such as HPMC, Polysorbate 20, Vitamin E- TPGS, Polysorbate 80, Rhamnolipids, Tetrglycerol Monooleate, Glyceryl Mono Stearate, Glyceryl Monoleate, Pentaethylene Glycol, Decussate sodium, Nanoxynol, and others, have been reported.

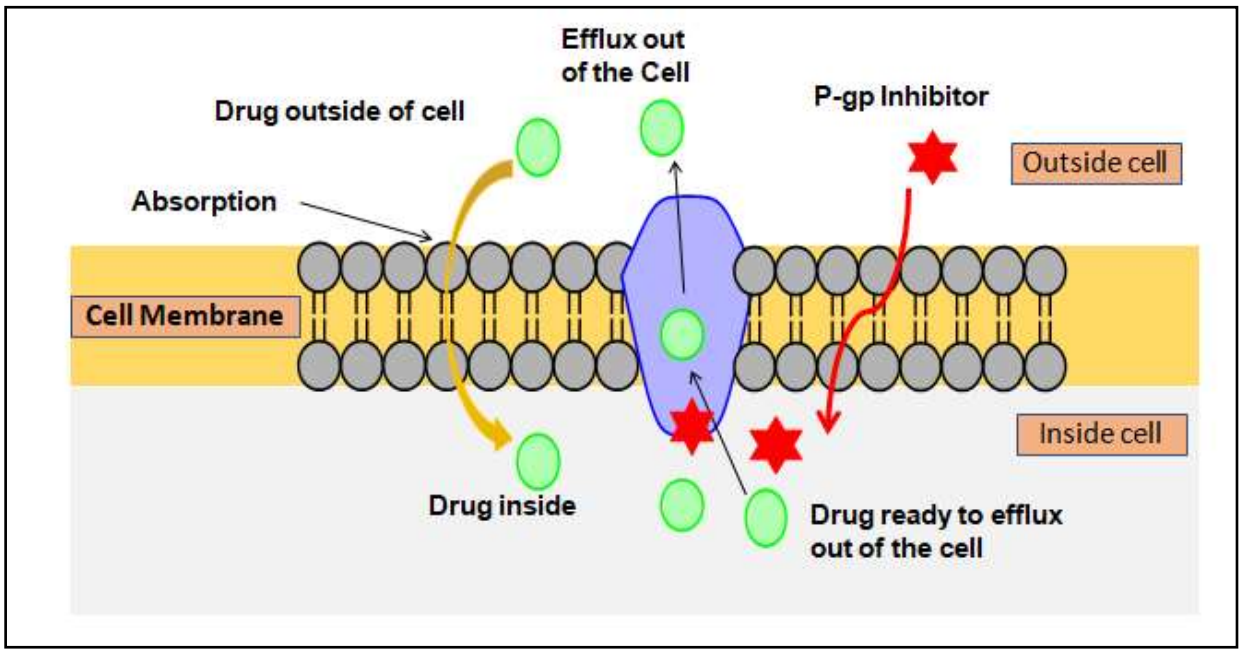

Figure 1: Molecular mechanism of permeation glycoprotein in the efflux mechanism and role of efflux pump inhibition (created using ChemBioDraw Ultra 14.0 software from PerkinElmer, Waltham, Massachusetts, USA.)

Molecular docking is the technique that is used the most frequently for in silico drug design and drug discovery. It is a form of structure-based virtual screening that seeks to identify new compounds that have an affinity for a specific target protein. In light of this background information, the purpose of the current work is to identify compounds derived from a variety of natural sources that contain excipients that have the potential to inhibit P-gp activity [13-15].

2. Materials and Methods

Ligand selection: A formulation cannot be constructed without the use of excipients. Investigations on the P-gp inhibiting ability of these substances have ushered in a new era in which these substances can be used as P-gp inhibitors. P-gp inhibitors include polymers, surfactants, lipids, and many more substances. In the domain of excipients, the function of specific P-gp inhibitors for enhanced drug administration has been investigated and discussed by a great number of research organizations. The several excipients that will be used in the p-gp

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interaction investigation were chosen by us based on the results of a survey of the relevant literature (Table 1).

Table 1: Primary selection of the excipients on the basis of literature survey

Sr. No.	Surfactants Name	Sr. No	Surfactants Name	Sr. No	Surfactants Name
1	Arginine	12	Chitosan	23	HPMC (Hydroxypropyl methyl cellulose)
2	Oleylamine	13	SLS (sodium lauryl sulphate)	24	Proline
3	polyethylene glycol Monooleate	14	B-Octylglucoside	25	Tween 80 (polysorbate 80)
4	Benzalkonium Chloride	15	Sodium stearate	26	PVP (Polyvinyl Pyrrolidone)
5	polyethylene lauryl ether	16	Docusate sodium	27	PVA (Polyvinyl Alcohol)
6	Sodium Alginate	17	cyclodextrin	28	TPGS (Tocopherol Polyethylene Glycol Succinate)
7	Ammonium Bromide	18	Dendrimer	29	PEG (Polyethylene Glycol)
8	Nanoxynol	19	Gaur Gum	30	Poloxamers (188, 407)
9	Rhamnolipid	20	Miglyol	31	Triton x-100 (Polyoxyethylene glycol octylphenol ethers)
10	Xanthine	21	Cetostearyl alccohol	32	Tween-20, 80(polysorbate 20, 80)
11	Sorbitan tristearate	22	Polysorbates (20, 80)	33	Cremophor EL (Polyethoxylated Caster oil)

Protein Selection: From the National Protein Data bank (PDB), permeation glycoprotein (P-gp) proteins with the ID 3G60 were chosen. A permeability glycoprotein is known as P-gp, which stands for multidrug resistance protein 1. P-gp is a major membrane transporter that plays a significant role in the metabolism of foreign particles and the subsequent efflux of these particles out of the cell. P-gp is responsible for the removal of foreign particles once they have been metabolised. P-gp's substrate-dependent efflux action can only occur in the presence of adenosine triphosphate, also known as ATP.

Molecular Docking: For the purpose of carrying out the molecular docking tests on the ligands of our choosing, we made use of the Schrodinger Maestro 9.1 software programme. The Protein preparation wizard was utilised in order to get the selected receptors ready. Proteins with the P-gp domain (PDB ID: 3G60) were used to retrieve the crystal structure of Protein Data Bank. The structure of the protein was preprocessed by protonation, which involved the removal of water molecules with the exception of those that were found near the active site. The ChemDraw software was used to diagram the structural components of the compounds. The application known as the Ligand preparation wizard

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was utilised in the production of the ligands. The OPLS-2005 force field was utilised in order to reduce the complexity of the geometries of the compounds and the protein structures. The development of the receptor grid was necessary for the process of ligand docking. For ligand docking experiments into the P-gp (PDB ID: 3G60) binding pocket, the XP mode of the Glide software developed by Schrodinger Inc. in the United States of America was utilised.

ADME properties: The ADME qualities of a potent lead molecule play a significant role in the conversion of that molecule to excipients. In order to use the lead molecule effectively in humans, it is vital to have a solid understanding of the absorption, distribution, metabolism, and excretion properties of the molecule. The QikProp application of Schrodinger Maestro 9.1 was used, as a result, to make predictions regarding the ADME properties of several compounds. It was possible to make predictions for properties such as total CNS activity, permeability of MDCK and Caco-2 cells, log BB and log Khsa for human serum albumin binding.

The ADME profile of the chosen excipients is included in the following table 2 for your reference.

Table 2: ADME **Properties of selected excipients**

Sr. No.	Compound Name	Mass	Log P	H-Bond Donor	H-Bond Acceptor
1.	HPMC	1261.4369	-2.3208	30	8
2.	Polysorbate 20	522.6684	1.9984	10	3
3.	Vitamin E- TPGS	574.8300	8.3557	6	1
4.	Polysorbate 80	604.8117	4.4626	10	3
5.	Rhamnolipids	650.7940	2.5485	13	6
6.	Tetraglycerol Monooleate	504.6961	3.6753	8	4
7.	Glyceryl Mono Stearate	358.5551	5.1443	4	2
8.	Glyceryl Monooleate	356.5392	4.9203	4	2
9.	Pentaethylene Glycol	238.2779	-0.9626	6	2
10.	Decussate sodium	444.5601	4.8903	7	0
11.	Nanoxynol	264.4023	4.3508	2	1

3. Results and Discussion

The docking simulations brought to light the particular impact that numerous elements have in the protein ligand interaction profile. Some of these factors include hydrogen bonds, lipophilic contacts, metal interactions, pi-pi interactions, and pi–cation interactions. Scoring functions are fast approximation mathematical approaches that are used in the field of computational chemistry and molecular modeling to forecast the intensity of a non-covalent connection between two molecules after the molecules have been

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docked. The docking score is an estimate of the energy that the posture takes up while it is contained within the binding site. It is based on physics and molecular dynamics. It takes into account the effect of the solvent, conformational changes in proteins and ligands, internal rotations, association energy of ligand and receptor to create a single complex, and free energy owing to variations in vibrational mode. It has been determined which molecules were picked based on the docking score of the protein. Excipients with a docking score were taken into consideration for further discussion (Table 3), along with their interaction docking scores with various amino acids found on the receptor [16-18].

The inability to produce new drug candidates that have reasonable chemical and biological features of the substances being studied is the limitation that prevents further leads optimization. Because of this, it is essential to have an accurate prediction of the pharmacokinetic profile of drugs [19-22]. On the basis of molecular docking, we chose only 11 of the 33 compounds to move on with for the subsequent phase (Table 3).

Table 3: Compilation of selected excipients docking result

Sr. No.	Excipients	Docking score
1.	HPMC	-14.16
2.	Polysorbate 20	-11.0
3.	Vitamin E- TPGS	-11.0
4.	Polysorbate 80	-9.0
5.	Rhamnolipids	-11.0
6.	Tetraglycerol Monooleate	-9.0
7.	Glyceryl Mono Stearate	-8.29
8.	Glyceryl Monooleate	-7.19
9.	Pentaethylene Glycol	-7.0
10.	Decussate sodium	-6.0
11.	Nanoxynol	-6.0

Additionally, in silico research was done on the pharmacokinetic characteristics of the chosen compounds with the corresponding receptors. Understanding a molecule's pharmacokinetic characteristics is crucial for turning it into a more potent lead in drug research. The permissible ranges for these attributes have been determined. The range for molecular weight is 130-725 kDa, and volume is defined as the expected number of hydrogen bonds that the solute from water will accept. There should be 5 and 10 hydrogen bond acceptors and donors, respectively. The octanol/gas projected partition coefficient is represented by the symbol Q P log Poct (8.0-35.0). Q P log Pw, the projected octanol/water partition coefficient log p (range -2.0 to 6.5), P log Po/w, and Q log S, the predicted aqueous solubility and S in mol/L (acceptable range -6.5 to 0.5) are all examples of predicted partition coefficients for water and gas [23-25].

All of the excipients demonstrated acceptable pharmacokinetics, with all of the previously

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mentioned attributes being within the bounds of acceptable standards. Compounds that demonstrated hydrogen bond donar and hydrogen bond acceptor values that were above the acceptable range should have their pharmacokinetic profiles changed in accordance with this. The molecular weight is within an acceptable range, as is the expected octanol/water partition coefficient, Q log S, predicted aqueous solubility, and S in mol/L. The expected octanol/gas partition coefficient is likewise below acceptable levels [26-28]

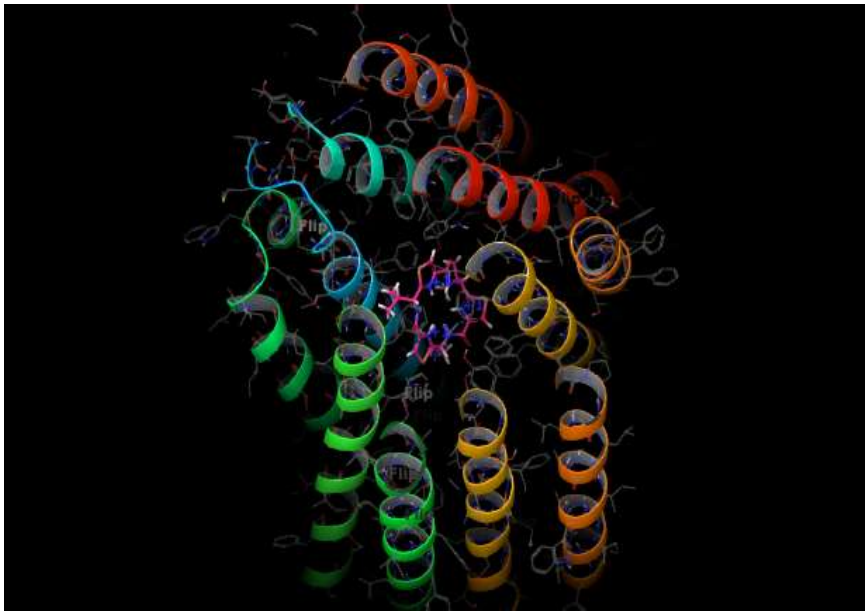

Figure 2: Excipients the hydrogen bond is shown as a yellow dotted line in the 3D ligand interaction, and the polar interaction is shown as a blue dotted line

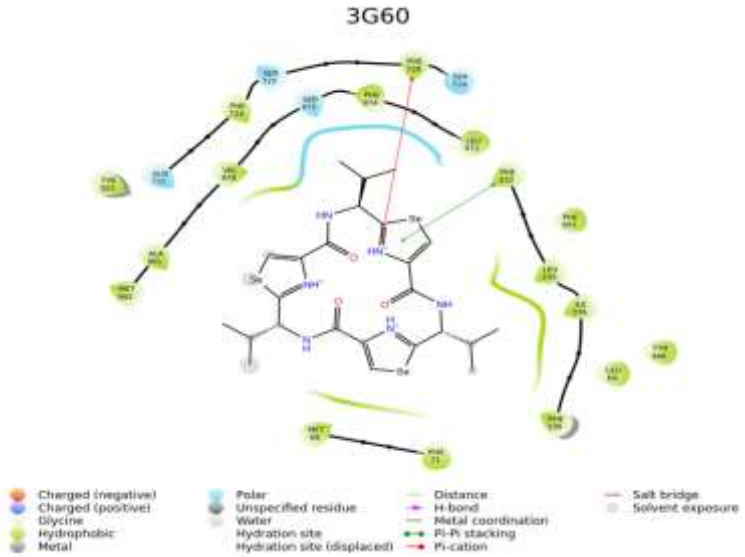

Figure 3: 2D view of the ligand-P-gp interaction (PDB ID: 3G60) obtained using the Schrodinger Glide SP programme; essential amino acid residues at the binding site are circled. The purple (+ve) and brown (-ve) circles represent electrostatic amino acids, the green circle represents hydrophobic amino acids, the blue circle represents polar interactions, and the purple arrow represents the hydrogen bond.

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4. Conclusions

The findings of this study may be useful in understanding the molecular mechanism of these excipients as prospective leads for P-gp inhibitors if they are found to be effective. The present findings will be further validated by the development of a formulation using any P-gp substrate medicinal molecule, as well as through in vitro and in vivo experiments that will be carried out.

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AUTHOR'S CONTRIBUTIONS

Both authors contributed equally to the study's conception, data collection, analysis, and interpretation, as well as to the writing and composing of the manuscript. The final draught was read and authorized by both authors.

ETHICAL APPROVAL

This study does not involve any animals or human subjects.

CONFLICT OF INTEREST

The authors declare no conflict of interests for this manuscript.

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