# Pinpointing the P- Glycoprotein Modulatory Potential of Natural Flavones against Liver Cancer: A Computational Approach Integrating Physicochemical and Docking Analysis

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**Structured abstract**

**Background:** Multi-drug resistance is the major impediment to chemotherapy, and most of the drug resistance in cancer cells emerges from over expression of specific proteins, such as P-glycoprotein, which increases the efflux of chemotherapeutic agents from cancer cells especially from liver. Inhibition of certain types of proteins to eliminate the therapeutic inefficiency of chemotherapy has increased the need for a large number of attempts to develop effective P-gp inhibitors. **Objective:** The objective of the present study is to evaluate physicochemical properties, ADMEt parameters, and binding interaction of various flavones with P-gp (PDB Id. 3G5U) to get the potential Pg-P inhibitor. **Methods:** In this study, we investigated the

physicochemical properties, ADMEt profile, and binding affinity of flavones. Verapamil which is a first-generation P-gP inhibitor is considered as a positive control. Prediction of binding affinity and molecular interaction of flavones with the potential binding cavity of P-gP were made with Molegro Virtual Docker while physicochemical properties and ADMEt profile were evaluated by SWISS ADME and pkCSM web servers, respectively. **Results:** The docking results reveal that out of 45 flavones, only 10 flavones having the MolDock score higher than verapamil; however, the MolDock score of 6-Prenylapigenin (4.7) was the highest among all the tested compounds. **Conclusion:** The present study provides valuable information about the physicochemical properties, ADMAEt profile, and binding interaction of flavone; these compounds may serve as a potential lead to the development of new P-gP inhibitors to overcome multi-drug resistance in chemotherapy of liver cancer.

**Keywords**

Anticancer; Flavone; Molecular docking; ADMET; P-glycoprotein; P-gP inhibitors.

## 1. Introduction

Liver cancer is a deadly prevalent over other cancer, and chemotherapy is a preferred option for the treatment of cancer as well as microbial infections despite progressing resistance development[1-2]. Understanding of molecular mechanism involved in drug resistance is a meaningful effort since the chemotherapeutic agent develops resistance [3-6]. Complex molecular mechanism, including increased efflux of drugs[7], impaired drug influx[8], Alteration of drug target, Enhanced DNA damage repair[9], enhanced drug extrusion mediated by MDR efflux transporters[10], intrinsic genetic and epigenetic alteration in drug-metabolizing enzymes[11], cellular senescence escape[12], tumor heterogeneity[13], ATP and ATP-mediated drug resistance[14], P-gp based drug resistance[15], receptor kinase based drug resistance[16], as well as anti-apoptotic mechanisms are involved in the progression of drug resistance[17-18]. Over expression of certain ATP-binding cassettes which produce the transporter-mediated resistance has been recognized as one of the major mechanisms among these aforementioned complex molecular mechanisms[19-20]. There are 48 members of the ABC transporter super family, which effluxes out a diverse group of substances such as drugs, toxins, ions, salts, lipids, cholesterol, peptides, and bile salts[21]. Nowadays, MDR causes over 90% failure of

chemotherapy in metastatic cancer patients [22]. The most prevalent of these MDR transporters, is P-glycoprotein (P-gp), a member of the ATP Binding Cassette (ABC) Super family[23].P-gp is an ATP-dependent efflux pump with broad substrate specificity, discovered in 1971 by victor ling[24]. This protein was identified for the first time in multidrug-resistant in 1974[25]. Glycoproteins are important integral membrane proteins that contain oligosaccharides chain covalently attached to amino acids side chain[26]. Plasma- glycoprotein is a cell surface protein, which acts as a localized drug transport mechanism actively exporting drug out of the cell and evolved as a defense mechanism against foreign harmful substances in fungi, bacteria as well as in animals[27]. P-glycoprotein, also known as multidrug resistance protein 1(MDR 1) or ATP-binding cassette sub-family B member-1(ABC B1),which is well-characterized ABC- transporter in human and encoded by the ABCB1 gene [28-30]. P-glycoprotein acts as an energy-dependent drug efflux pump and is composed of 1280 amino acids (170 kDa) organized in two transmembrane domains, each one comprised of twelve highly hydrophobic α-helices and two intracellular nucleotide binding regions with ATPase activity, these two ATP binding regions separated by a flexible linker polypeptide region [31].Intracellularly, there are two ATP-binding domains, which are also known as nucleotide-binding domains (NBDs), which constitute the power units of P-glycoprotein[32]. The NBDs are located in the cytoplasm and transfer energy to transport the substrates across the membranes. The structure of P-gp represents a nucleotide-free inward-facing conformation arranged as two “halves” with pseudo-two-fold molecular symmetry spanning ∼136 Å perpendicular to and ∼70 Å in the plane of the bilayer[33]. The nucleotide-binding domains (NBDs) are separated by ∼30 Å. The inward-facing conformation, formed from two bundles of six helices, results in a large internal cavity open to both the cytoplasm and the inner leaflet[34].P-gp containing energy-dependent drug efflux pump encoded by the human MDR1 gene [29]. It is responsible for decreased drug accumulation in multidrug-resistant cells and often mediates the development of resistance to anticancer drugs[35]. This protein also functions as a transporter in the blood-brain barrier[36]. Efflux transporters such as P-glycoprotein play an important role in drug transport in many organs[37]. In the gut, P-glycoprotein pumps drugs back into the lumen, decreasing their absorption[38]. Drugs that induce P-glycoprotein, such as rifampicin, can reduce the bioavailability of some other drugs [37]. Inhibitors of P-glycoprotein, such as verapamil, increase the bioavailability of susceptible drugs[39]. P-glycoprotein is one of the drug transporters that determine the uptake and efflux of

a range of drugs[40]. This process affects their plasma and tissue concentrations and ultimately their final effects [32]. P-glycoprotein functions as a transmembrane efflux pump, pumping its substrates from inside to outside the cell. P-glycoprotein is a major barriers that can actively efflux out the variety of therapeutic agents and reduce the bioavailability of different drugs[41].MDR1 and MDR3 are two members of the P-gp gene family, which exist in humans, whereas MDR1A, MDR1B, and MDR2, are found in animals[42]. The human MDR1is widely distributed and is known to excrete a wide range of drugs across the cell membrane. However, MDR3 shows its highest expression in the canalicular membranes of hepatocytes[23]. The human MDR1 is ubiquitously expressed and is perhaps one of the most significant ABC transporters for drug disposal in humans and thus carries pharmacological importance. It has been identified as a primary cause of MDR. P-gp shows MDR by affecting the absorption, distribution, excretion, and metabolism of drugs that reduce the effectiveness of certain drugs like anticancer, antibiotic, antidepressant, antihypertensives, antiarrhythmic, calcium channel blockers, immunosuppressant, HIV protease inhibitors, and cardiac glycosides[43-45]. The overexpression of P-gp which pumps chemotherapeutic drugs outside the cell via ATP hydrolysis is the major mechanism of drug resistance[46]. By this process, P-gp restricts the intracellular retention and cytotoxicity of chemotherapeutic agents and manifests an MDR phenotype to the tumor. Hence inhibition of p-gp could play important role in cancer chemotherapy, treatment of HIV as well as antimicrobial therapy. The nature of interaction of a particular compound with a receptor or protein detects it either as a P-gp inhibitor or substrate or an inducer[47]. Based on their affinity, specificity, and toxicity, P-gp inhibitors are classified into three generations and the chemical structures are given in **Fig. 1**[31].

The ***First Generation inhibitors*** are- verapamil, cyclosporine A, vincristine, reserpine, tamoxifen, trifluoperazine, etc, and the ***Second Generation inhibitors*** are- dexniguldipine, elacridar, dofequidar, and cyclosporin D derivative i.evalspodar while inhibitors that belong to the ***third Generation are***- tariquidar, zosuquidar, laniquidar, annamycin. P-gp inhibitors/blockers are potential enhancers for the cellular bioavailability of several clinically important anticancer drugs such as, anthracyclines, taxanes, vinca alkaloids, and podophyllotoxins[48-50]. Besides several chemically synthesized P-gp inhibitors/ blockers, some naturally occurring compounds and plant extracts were reported for their modulation of multidrug resistance[51]. Inhibition of P-gp by herbal constituents is an innovative technique for reversing drug resistance in

chemotherapies [42, 52-53]. Therefore, many efforts are currently being done to find natural compounds from plant sources that inhibit P-gp, reverse the MDR phenotype and sensitize the target cells to conventional chemotherapy without undesirable toxicological effects[54]. The inhibitors of P-gp are obtained from various natural sources in the form of alkaloids, flavonoids, coumarins, resins, saponins, terpenoids, and miscellaneous other species [23, 55].

Flavonoids are the secondary metabolites of plant polyphenols having structural arrangements of two benzene rings (Ring A & B) attached with an oxygen-containing six-membered heterocyclic pyran ring (Ring C)[56-57]. According to the attachment pattern of ring B at the varying position of the pyran ring (ring C), the flavonoids are categorized into three main groups including-*flavonoid* (ring B is linked at C-2 of Pyran ring), *isoflavonoids* (ring B is linked at C-3 of Pyran ring) and *neoflavonoids* (ring B is linked at C-4 of Pyran ring)[58]. Based on unsaturation and varying hydroxy substitution of heterocyclic pyran ring (ring C), the flavonoid is further categorized into *flavone, flavonols, flavanone,* and *flavan compounds* **(Fig. 2)**. Flavone belongs to the flavonoid subgroup of polyphenols with a basic 15-carbon phenylpropanoid skeleton ($C_6$-$C_3$-$C_6$ core) containing a double bond between C-2 and C-3 with a carbonyl group at the C-4 position[59].Flavonoid compounds like flavones are nonessential dietary factors present in regular human diets are generally occurring in green vegetables, fruits, soybean oils, tea, and chocolate[60]. One significant link between the human diet and the prevention of chronic disorders such as cancer has been set up by these flavones. Flavones are already reported as an antiallergic[61], antioxidant[62], anti-inflammatory[63], hepato protective[64], antiviral[65], antiproliferative[66], anticarcinogenic activities[67-68], and estrogen-like activities[69]. In the present study, we analyzed the physicochemical properties, ADMEt parameters, and also performed molecular docking simulation of 45 flavones with P-gp (PDB Id. 3G5U), and compared all the results with verapamil, which is taken as a standard Pg-P inhibitor.

## 2. Material and methods

### 2.1. Ligand preparation

The chemical structures of 45 flavones were drawn by using Chem Draw Professional 15.0 these structures were further subjected to Chem 3D software for energy minimization. The energy minimization was done with the molecular mechanics (MM2) tool of Chem 3D software[70]. The root-mean-square (RMS) gradient value was set up at 0.0001 kcal/mol Å for the geometrical

optimization and the process was run until the RMS value reaches to lesser than 0.0001 kcal/mol Å. These energy-minimized ligands were imported into the Molegro Virtual Docker software workspace and further ligand preparation was done to assign bond order & hybridization and detect flexible torsions & create explicit hydrogen if missing[71].

### 2.2. Prediction of ADMEt score and physicochemical properties of flavones

Screening of physicochemical parameters of desired compound is important to get an idea of whether this leads to having a drug-like activity or not. The SWISS ADME (http://www.swissadme.ch/) [72] and pkCSM (http://biosig.unimelb.edu.au/pkcsm/prediction) [73]server was used to predict physicochemical properties and ADMEt profile of flavones respectively. To predict the physicochemical property and ADMEt profile of flavones the SMILES of all selected flavones were generated in Chem Draw Professional 15, furthermore, these SMILES were pasted in the respective webserver to find the desired results[74]. Various parameters that exhibit physicochemical properties of flavones were predicted by the SWISS ADME server and obtained parameters are molecular weight, Log P, hydrogen donor, hydrogen acceptor, total polar surface area, and the number of rotatable bonds. Assessment of ADMEt is a critical step to determine the absorption, distribution, metabolism, excretion, and toxicity profile of lead compounds in the early stage of drug discovery[75]. Different parameters related to ADMEt properties of lead compounds were evaluated by using pkCSM webserver and the obtained parameters are Caco2 permeability, intestinal absorption, skin permeability, VDss, BBB permeability CNS permeability, effect on cytochromes P450 and P-gp substrate, total clearance, renal OCT2 substrate, AMES toxicity, and hepatotoxicity were well correlated with a standard value.

### 2.3. Protein preparation and receptor grid generation

The x-ray crystal structure of P-glycoprotein (PDB Id. 3G5U) at a resolution of 3.80 Å was retrieved from RCSB protein data bank having a total structure weight of 286.14 kDa, atom count 18352, sequence length 1284, unique protein chain 1,and complex with Mercury $Hg^{2+}$ as a cofactor **Fig. 3**[33, 76]. The retrieved protein was imported into Molegro Virtual Docker software to repair the warning errors and preparation for docking. Maximum five cavities where ligand can bind were detected with the volume size of 1520.64 $Å^3$, 160.256 $Å^3$, 99.328 $Å^3$, 66.048 $Å^3$, and 70.656 $Å^3$. For the molecular docking study, the highest volume occupied cavity

(1520.64 $Å^3$) was selected out of the detected five cavities and the detail of all detected cavities are shown in **Table 1**. Then the receptor grid generation was done to specify the binding site of the protein where ligand can bind. The center coordinates of protein were set with a grid spacing of 0.30 Å, the grid size was 59.67×75.63×51.38 point and the binding site radius was 20Å.

### 2.4. Molecular docking study

Molegro Virtual Docker (MVD 20019.7.0.0) developed by: Molexus IVS, Denmark was used for molecular docking of flavones with Plasma-glycoprotein[71]. MVD provides an integrated environment for ligand and protein interaction by flexible ligand docking[77]. Verapamil as reference standard along with all the energy minimized ligand of flavone compounds were imported into the pre-saved workspace of MVD having optimized P-glycoprotein**.** Molecular docking simulation of ligand was done with most potential binding sites of the highest volume occupied cavity (cavity 1 with a volume size of 1520. 64 $Å^3$), the verapamil was used as a positive control to check the reliability & accuracy of docking simulations. The grid-based scoring function of Molecular docking simulation at 0.3 Å grid resolutions was done with MolDock SE (MolDock Simplex Evolution) search algorithm with 10 runs and 50 population size[71]. Other parameters such as maximum iterations, binding site radius, and RMSD were set to 1500, 20 Å, and 1.00 Å respectively[78]. Total 10 number of runs means ten times docking simulation was repeated for each ligand and returning to a single final pose. For the analysis of binding interaction with target P-glycoprotein (H-bond and steric interaction), the highest MolDock score was selected[79].

## 3. Result and Discussion

Multidrug resistance (MDR) represents a major challenge in cancer chemotherapy because it limits the effectiveness of many clinically important drugs[46, 80]. In humans, two closely related genes, MDR 1 and *MDR2* or *MDR3* (the so-called multidrug-resistance genes), encode highly homologous P-glycoproteins[81]. Only the *MDRl*gene has been linked to then multidrug-resistance phenomenon however[37]. Many cancers fail to respond to chemotherapy by acquiring MDR, to which has been attributed the failure of treatment in over 90% of patients with metastatic cancer[82]. Although MDR can have several causes, one major form of resistance to chemotherapy has been correlated with the presence of at least three molecular “pumps” that actively transport drugs out of the cell[26, 83]. The normal excretion of xenobiotic

back into the gut lumen by p-gp and reduce efficacy of some pharmaceutical substances, those substances effluxes by p-gp are called substrate for p-gp[84]. There are several p-gp substrate are- lipids[85], steroid[86], xenobiotic[87], peptide[88], bilirubin[89], cardiac glycoside[90], immunosuppressive agent[91], glucocorticoid[92] and various chemotherapeutic agents[93].According to some published reports, several isolated pure plant compounds have resistance modulating activity by inhibiting P-gp[94]. Natural molecules have structural diversity, which provide a valuable tool in the search of highly target specific P-gp inhibitors [50]. It has been observed that many P-gp inhibitors from natural sources are very non-specific, but less toxic in nature [41]. Therefore, due to their low toxicity level, the research on natural P-gp inhibitors is getting more and more attention nowadays[95]. The main challenges of using natural products instead of conventional inhibitors are structural diversity and non-specific target bindings [96]. Some of the plant-based bioactives are reported to involve non-specific P-gp inhibition, and the process could affect other proteins and enzymes[42]. Therefore, it is necessary to use some standard methods to evaluate all-natural molecules to avoid this contradiction through specific and targeted research [97]. Therefore, efforts to develop effective P-gp inhibitors with less toxicity, high specificity, and different mechanisms of action are completely logical [42].Several compounds of flavone category, including chrysin [98], oroxylin A [99], hesperidin [100], apigenin [101], baicalin [102], wogonin [103], sinensetin[99], and tangeritin[104], etc. have been reported to inhibit P-gp mediated efflux and enhance the accumulation and efficacy of anticancer drugs[105].

Therefore, we decided to select additional flavonoids to screen their interaction with the P-gP binding pocket, which can also have Pg-P inhibitory activity. In present study we have selected 45 flavone compounds for analysis purposes and compared the physicochemical properties, ADMEt profile, and molecular docking study results of all selected flavones with verapamil as a reference standard. The chemical structures of all selected flavones are given in **Fig. 4**.

### 3.1. Prediction of ADMEt score and physicochemical properties of flavones

The physicochemical property of chemical compounds influences the ability of a chemical compound to elicit a pharmacological and or therapeutic effect with physical properties and chemical properties[106]. Evaluation of physicochemical properties at the optimum level is an elemental segment to determine the solubility of drug-like compounds[107]. SWISS ADME

server predicts all the parameters required for Lipinski's rules[108] and Veber's rules [109]of drug-likeness. According to Lipinski's rules thelogP< = 5, Molecular weight < = 500, number of hydrogen bond acceptors < = 10, number of hydrogen bond donors < = 5, while Veber's rules is based on the total polar surface area (TPSA < = 140 Å2) and number of rotatable bonds (No. of rotatable bond < = 10) of compounds. For the prediction of ADMEt parameters the pkCSM web server was used which predicts all relevant parameters of absorption (Caco2 permeability, intestinal absorption, Skin permeability), distribution (VDss, BBB permeability, CNS permeability), Metabolism (Pg-P substrate, effect on cytochromes P450), excretion (total clearance, renal OCT2 substrate), and toxicity (AMES toxicity, hepatotoxicity) of 45 flavones including verapamil as reference P-gp inhibitor. According to the pkCSM web server, less than 30% intestinal absorption is considered a poorly absorb the drug, for skin permeability log Kp> -2.5 considered as relatively low skin permeable, and Caco2 permeability (log Papp > 0.9 is considered as high Caco2 permeable. The volume of distribution, BBB permeability, and CNS permeability has been considered as critical parameters to understand the distribution pattern of drugs[110]. The PkCSM give the predefined value of log VDss< -0.15 (VDss< 0.71 L/kg) and log VDss> 0.45 (VDss> 2.81 L/kg) is considered as drug with low volume of distribution and drug with high volume of distribution respectively. To understand the BBB permeability the pkCSM webserver considered the drug with logBB< -1 is poorly distributed to the brin while drug having logBB> 0.3 can readily cross the BBB. Moreover, drugs having logPS> -2 can easily penetrate the CNS while drugs with logPS< -3 are unable to penetrate CNS. The inhibitory effect of Cytochrome P450 and P-gP are important parameters to understand the metabolism pattern of drugs. To understand the excretion rate of the drug the PkCSM webserver measure the drug clearance by the proportional constant i.eCLtot which is given in log (ml/min/kg). The predicted result of physicochemical properties and ADMEt profile of 45 flavonoids including verapamil as a reference drug is tabulated in the **Table 2**. The obtained parameters of physicochemical properties result showed that among the 45 screened compounds, there were only 26 compounds, including 4.1. 4.2. 4.3, 4.4, 4.5, 4.6, 4.7, 4.8, 4.10, 4.12, 4.14, 4.17, 4.18, 4.20, 4.22, 4.23, 4.24, 4.27, 4.28, 4.29, 4.30, 4.31, 4.32, 4.41, 4.42, and 4.43, follow Veber's rule & Lipinski's rule of five.

### 3.2. Molecular Docking Study

All the flavones along with verapamil as a positive control were subjected to the molecular docking study with P-gp to predict the ligand-target binding interactions by using Molegro Virtual Docker.After successful ligand-protein docking, various outcomes such as Mol Dock score, Rerank Score, and H-bond score were obtained which are further used to understand the ligand-protein binding affinity. The MolDock score is the best plausible orientation of the ligand which applies piecewise linear potential (PLP) for the calculating of stearic energy while the weighted combination of the energy terms with MolDock score denoted by Re-rank score which represents the accuracy of molecular docking study. The strength of hydrogen bond interaction between ligand and the target protein is represented by the H-bond score. The result of molecular docking simulation of flavones with potential binding cavity (1) of P-gP (PDB Id. 3G5U) is tabulated in **Table**3, which contains MolDock score; Re-rank score and H-bond score. After the successful completion of the molecular docking study, the MolDock score and Rerank score of Verapamil with P-gp protein was found -79.6711 Kcal/mol and -38.3384 Kcal/mol respectively. The molecular docking study reveals that verapamil shows the steric interaction through the residues Asp382 (A) and Tyr461(B) while hydrogen bond interaction through the residue Tyr461(A) and Lys380(B) of P-gP. The obtained molecular docking results were compared with standard P-gP inhibitors, and it has found that out of 45 flavones only 10 flavones having the MolDock score higher than verapamil; however, the MolDock score of 6-Prenylapigenin (4.7) was highest among all the tested compounds and it shows the steric interaction through the residues His379(B), Pro381(B), Asp382(B), and Asn458(B) as well as hydrogen bond interaction through the residue Ser377(B) and Lys380(B) of P-gP. The 2D and 3D interaction diagrams of verapamil and flavones exhibiting highestMolDock score with target P-gP within the constraints of the highest volume occupied cavity 1 is pictured in **Fig 5** and **Fig 6** respectively.

Nobiletin (4.5) exhibits the MolDock score of -131.488 Kcal/mol and the Re-rank score of -67.9994 Kcal/mol. It shows H-bond interactions with Ser377(B), Lys380(B), Glu464(B), and steric interactions with Pro381(B), asn458(B), Tyr461(B), residues of the target. Entadanin (4.30) is the compound that exhibits the third-highest MolDock score of -102.911 Kcal/mol with the Re-rank score of -78.2184 Kcal/mol, it shows hydrogen bond interactions with Ser377(B), Gly378(B), Asp458(B), Arg460(B) the residues and steric interactions with Lys 376(B), His379(B), Asn458(B) residues of the target protein. Diosmetin (4.18) shows the hydrogen bond interactions with the Ser375(A), Ser377(A), Arg460(A), Glu464(A), Asp490(B), Glu495(B)

residues and steric interaction with Ser375(A), Lys376(A), Arg460(A) residues of P-gP with MolDock score of -100.986 Kcal/mol and Re-rank score of -60.2849 Kcal/mol.Vitexin (4.33) exhibits the MolDock score of -88.7793 Kcal/mol and Re-rank score of -79.3577 Kcal/mol. The hydrogen bond interaction of vitexin with P-gP target was seen at Ser377(B), Gly378(B), Lys380(B), Pro381(B),Lys546(A) residues while it shows stearic interactions with His379(B), Pro381(B), Ile384(A) residues.Licoflavone (4.27) exhibits the MolDock score of -88.5816 Kcal/mol and Re-rank score of -78.6427 Kcal/mol. It shows hydrogen bond interaction with His149(A), Arg460(A), Asp490(B), Thr492(B), Gln910(A) and stearic interaction with His149(A), Thr492(B), Glu909(A) residues of target protein. The MolDock score and Re-rank score of Apigenin(4.14) is -88.2421 Kcal/mol and -78.6427 Kcal/mol respectively and it shows stearic interaction with Lys376(A), Ser375(A),Glu495(B), Ala999(B), Val542(B) and hydrogen bond interactions with His149(A), Lys368(A), Glu495(B), Lys498(B) residues of P-gP pocket. The MolDock score of the compound 6-hydroxyflavone (4.8) was -88.2311 Kcal/mol and Re-rank score was -78.6339 Kcal/mol. It shows hydrogen bond interaction to Ser377(B), Glu464(B), Asp490(A) residues and stearic interactions to Asp382(B), Asn458(B), Arg460(B) residues of target protein. Wogonin (4.31) shows the H-bond interactions at Ser375(A), Lys376(A), Ser377(A), Lys380(A) residues and stearic bond interactions at Ser375(A), Lys376(A), Lys380(A),Asn458(A), Arg460(A) residues of the P-gP with the MolDock score of -87.7968 Kcal/mol and Re-rank score of -82.547 Kcal/mol. Amentoflavone (4.21) exhibits the MolDock score -80.6874 Kcal/mol and Re-rank score of -70.5523 Kcal/mol, which shows hydrogen bond interactions with Ser377(B), Lys380(B) and stearic interactions with Asp382(B), Asn458(B) residues of the target protein.

**Conclusion**

P-gp can expel a broad range of structurally different exogenous compounds out of the cells. For this reason, a very active P-gp transporter could potentially diminish drug delivery to the target organ and has been correlated to treatment resistance, despite peripheral drug concentrations that are within their therapeutic range. As a result, P-gp mediated drug efflux is recognized as a desirable target for therapeutic intervention in order to target and optimize the drug delivery of drugs to tumor cells [111]. Inhibition of P-gp leads to an increase in the permeability of some target organs[112]. This result could permit administering lower drugs oral doses, and it may help to decrease drug toxicity. Some plant-based molecules are also active against microbial

efflux systems and some are active in both humans and microbes, so there may be a probability those molecules that are active against microbial efflux systems may affect the efflux system in cancer cells. Further research is needed to prove this hypothesis and find new novel P-gp inhibitors. Finally, modern experimental methodologies and techniques, such as structure-activity relationships (SAR), quantitative structure-activity relationships(QSAR), 3-dimensional structure-activity relationships (3DQSAR), and pharmacophore studies should also be taken into consideration and should be regarded as an important guiding tool for the modern researchers in discovering very selective and potent P-gp inhibitors[113].This study provides a piece of valuable information about the physicochemical properties, ADMAEt profile, and flavone binding interaction with the bonding cavity of P-gP as a target protein; however, the result of this study indicate that these compounds can serve as potential lead to the development of new P-gP inhibitors to overcome multi-drug resistance in chemotherapy.

**Figure legends:**

**Figure 1:** Representation of classification of P-gp inhibitors along with their chemical structures.

**Figure 2:** Representation of chemical structure of basic flavonoid system with its varying subclasses.

**Figure 3:** Predicted binding cavity 1 (green) of P-gp (PDB Id. 3G5U).

**Figure 4:** Chemical structures of the natural flavone selected for screening in the present study.

**Figure 5:** The 2-D molecular docking view of verapamil and flavone exhibiting highest MolDock score; 6-Prenylapigenin (4.7) is shown in **(a)** and **(b)** respectively.

**Figure 6:** The 3-D molecular docking view of verapamil and flavone exhibiting highest MolDock score; 6-Prenylapigenin (4.7) is shown in **(a)** and **(b)** respectively.

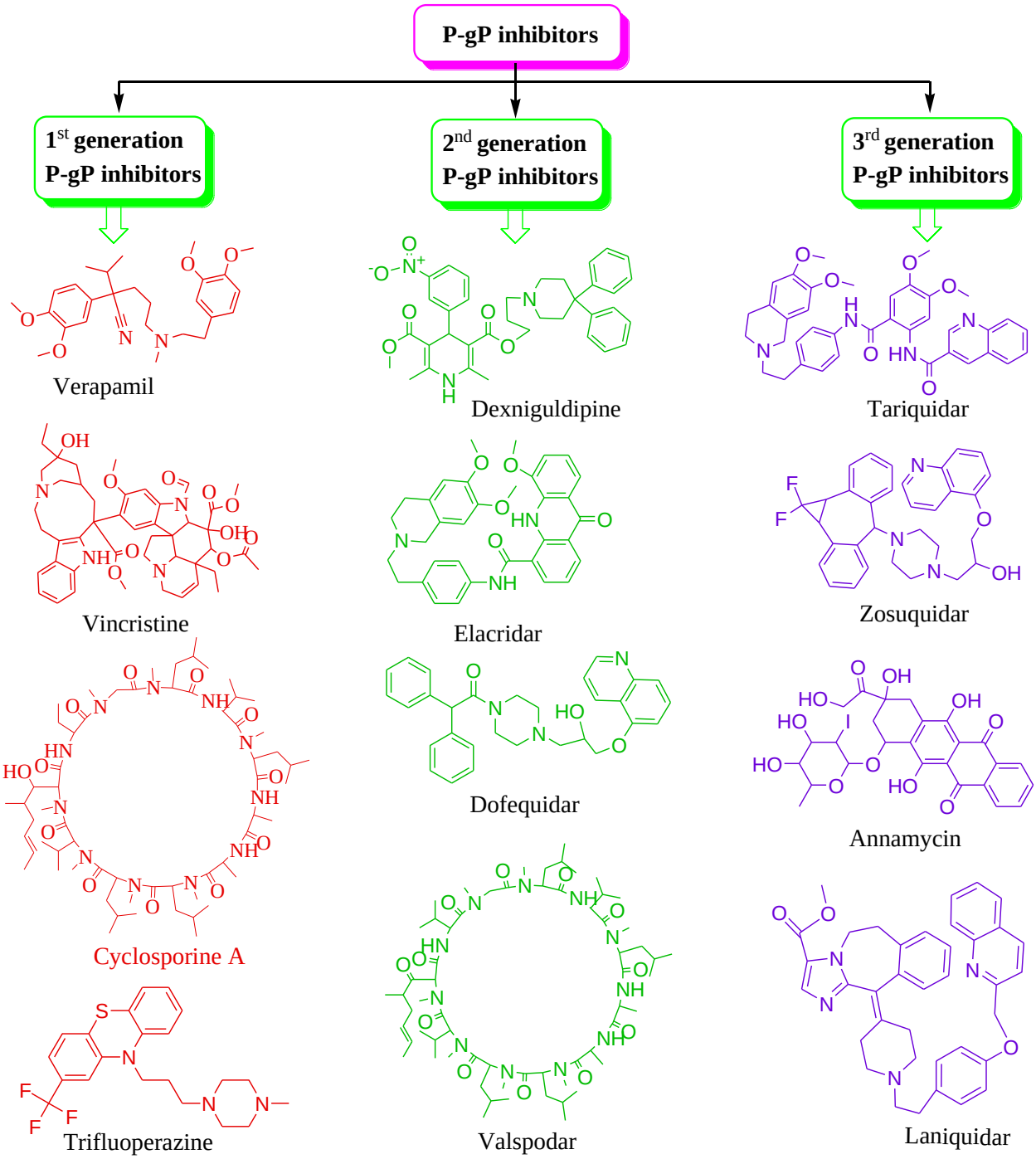

P-gP inhibitors
1st generation P-gP inhibitors
2nd generation P-gP inhibitors
3rd generation P-gP inhibitors
Verapamil
Dexniguldipine
Tariquidar
Vincristine
Elacridar
Zosuquidar
Dofequidar
Annamycin
Cyclosporine A
Valspodar
Trifluoperazine
Laniquidar

**Figure 1**

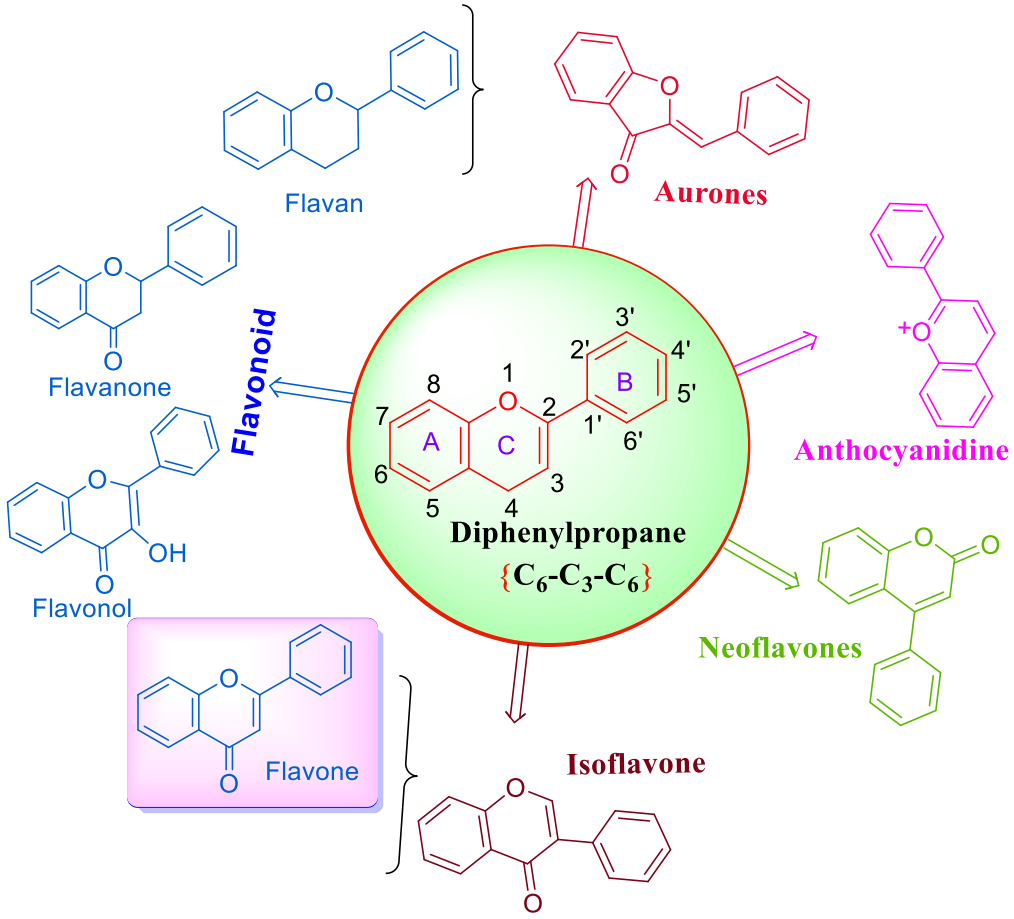

Flavan
Aurones
Flavanone
Flavonoid
Anthocyanidine
Flavonol
Diphenylpropane
{C6-C3-C6}
Neoflavones
Flavone
Isoflavone

**Figure 2**

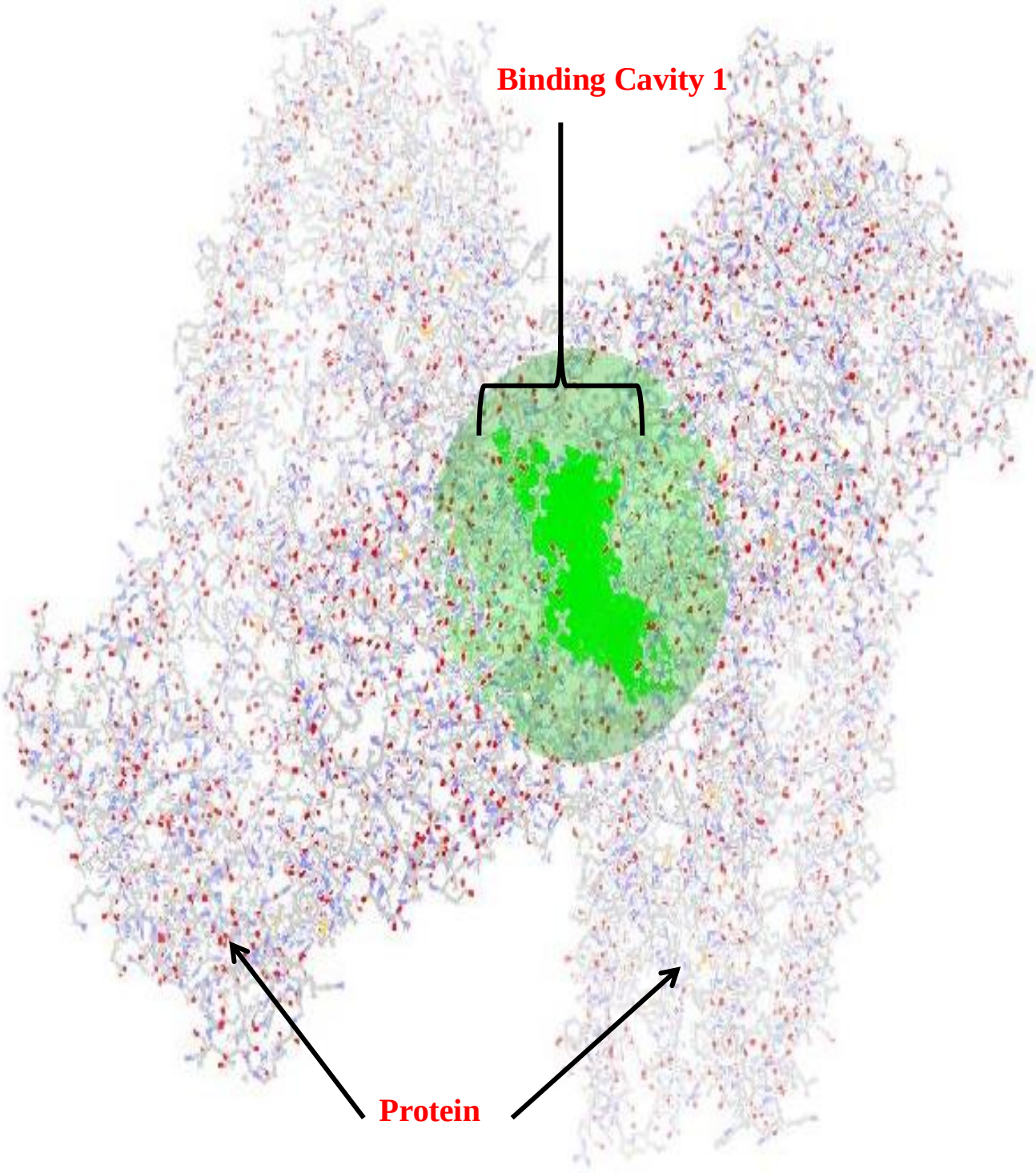
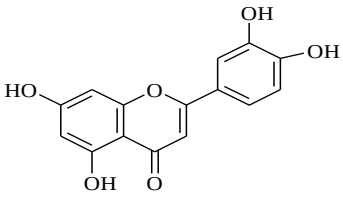
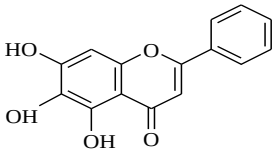


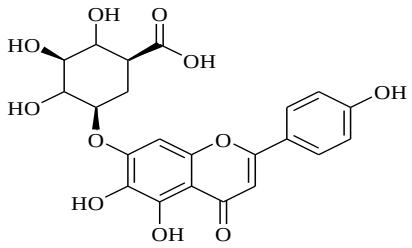

**Figure 3**



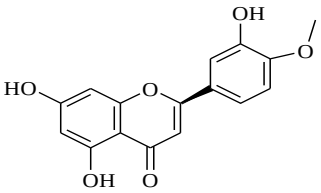

**Luteoline (4.1)**



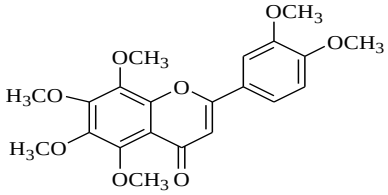

**Baicalin (4.2)**



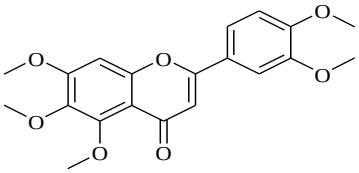

**Scutellarin (4.3)**



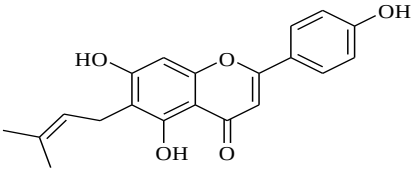

**Hesperetin (4.4)**



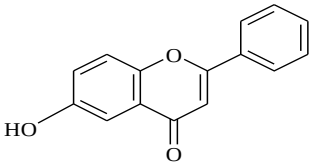

**Nobiletin (4.5)**



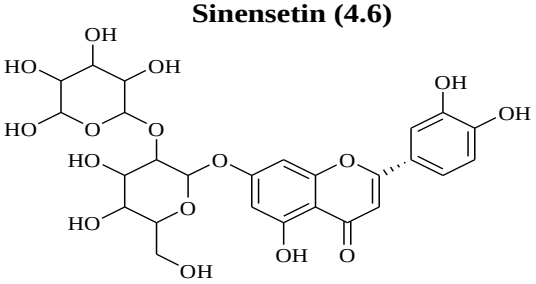

**Sinensetin (4.6)**



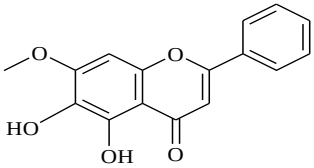

**6-Prenylapigenin (4.7)**



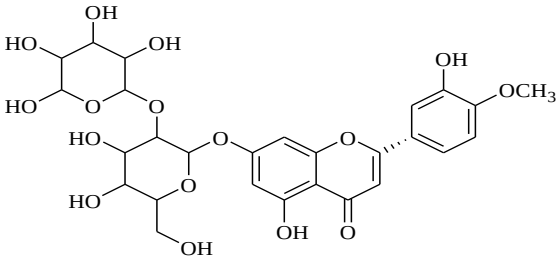

**6-Hydroxyflavone (4.8)**



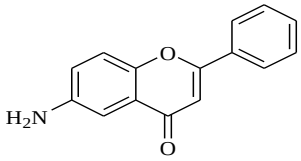

**Neoeriocitrin (4.9)**



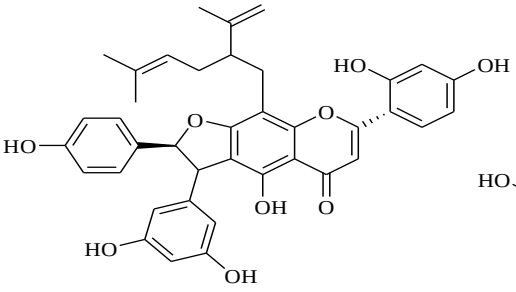

**Negletein(4.10)**



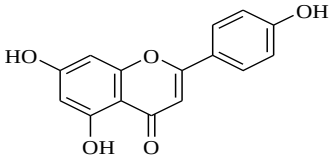

**Neohesperidin (4.11)**



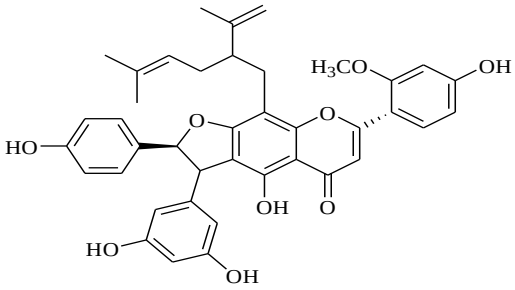

**6- Aminoflavone (4.12)**




**Alopecurone A (4.13)**

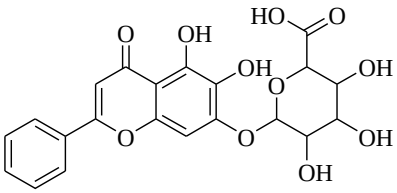



**Apigenin (4.14)**

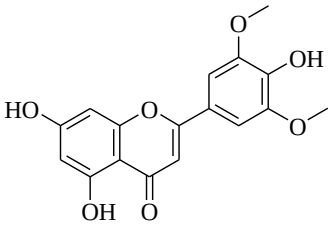



**Alopecurone D (4.15)**

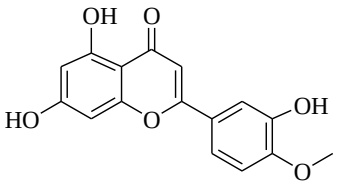
**Figure 4**



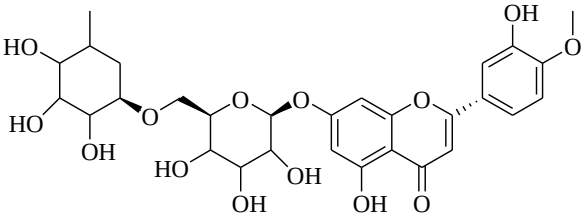
**Baicalin (4.16)**



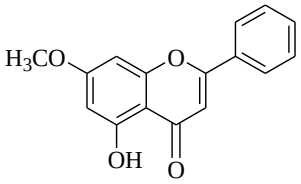
**Chrysin (4.17)**



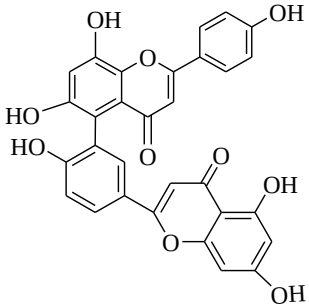
**Diosmetin (4.18)**



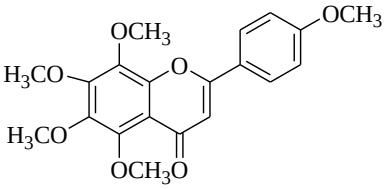
**Hesperidin (4.19)**



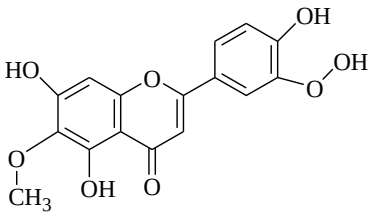
**Techtochrysin (4.20)**



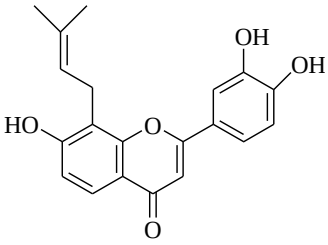
**Amentoflavone (4.21)**



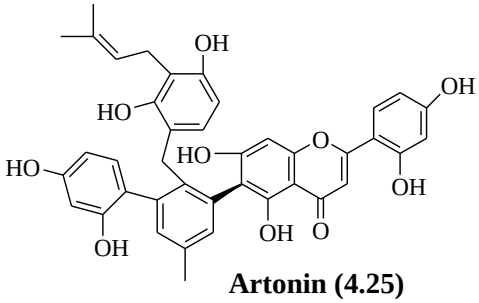
**Tangeritin (4.22)**



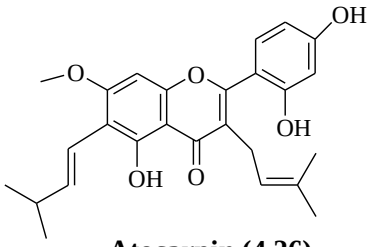
**Jaceosidin (4.23)**



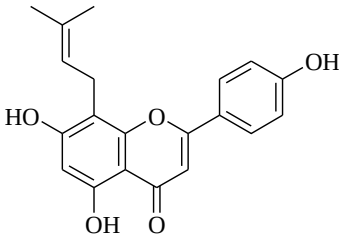
**Corylifol C (4.24)**



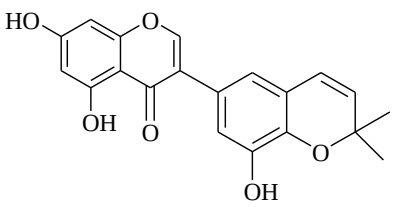
**Artonin (4.25)**



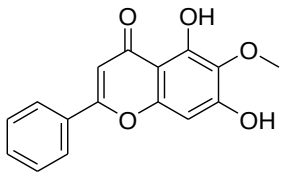
**Atocarpin (4.26)**



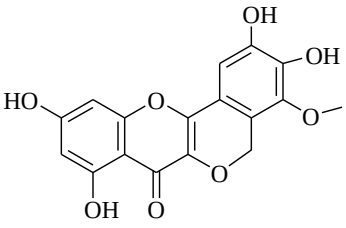
**Licoflavone (4.27**



**Semilicoisoflavone B (4.28)**



**Oroxylin A (4.29)**



**Entadanin (4.30)**

**Figure 4** Continued…..

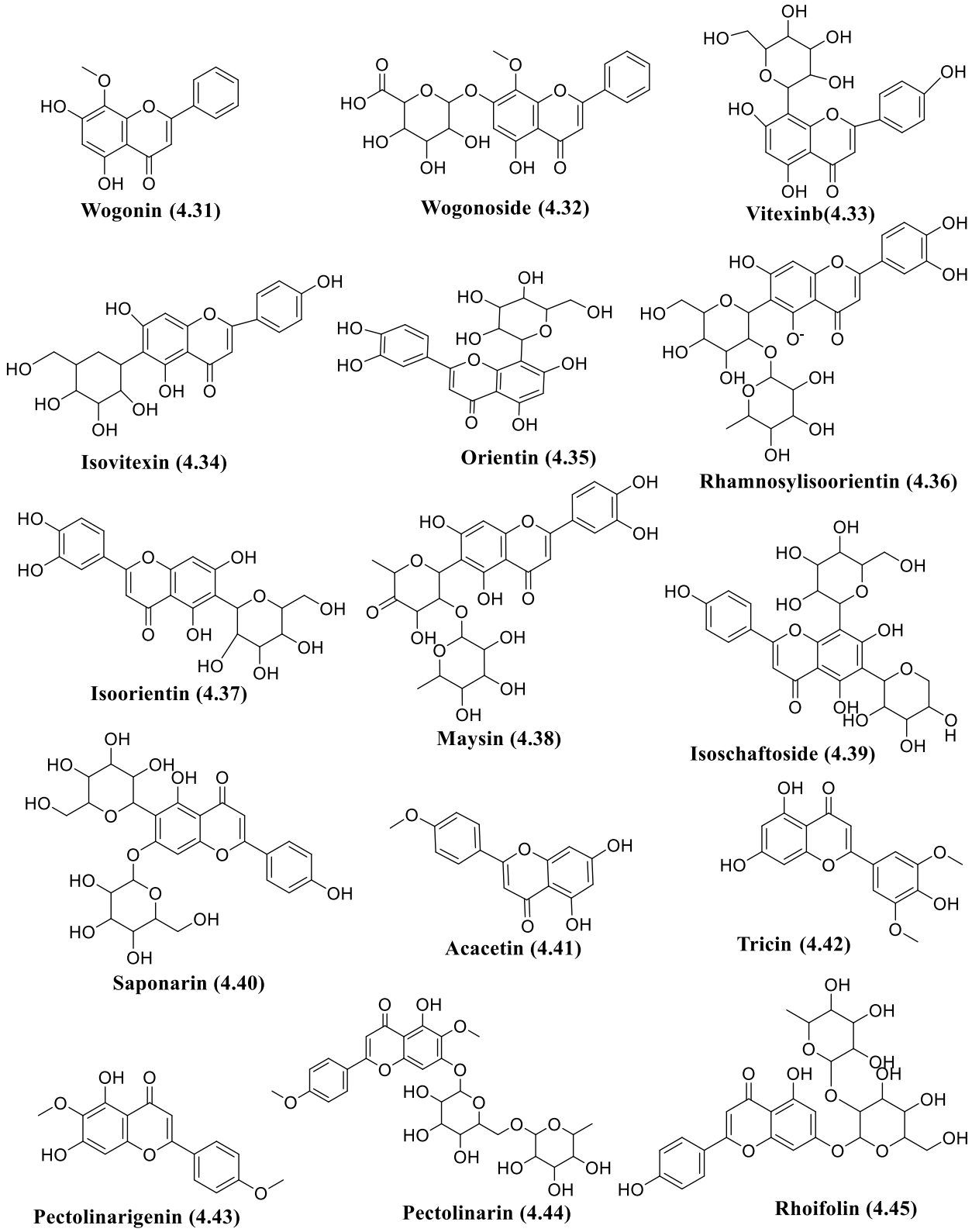



**Figure 4** Continued…..

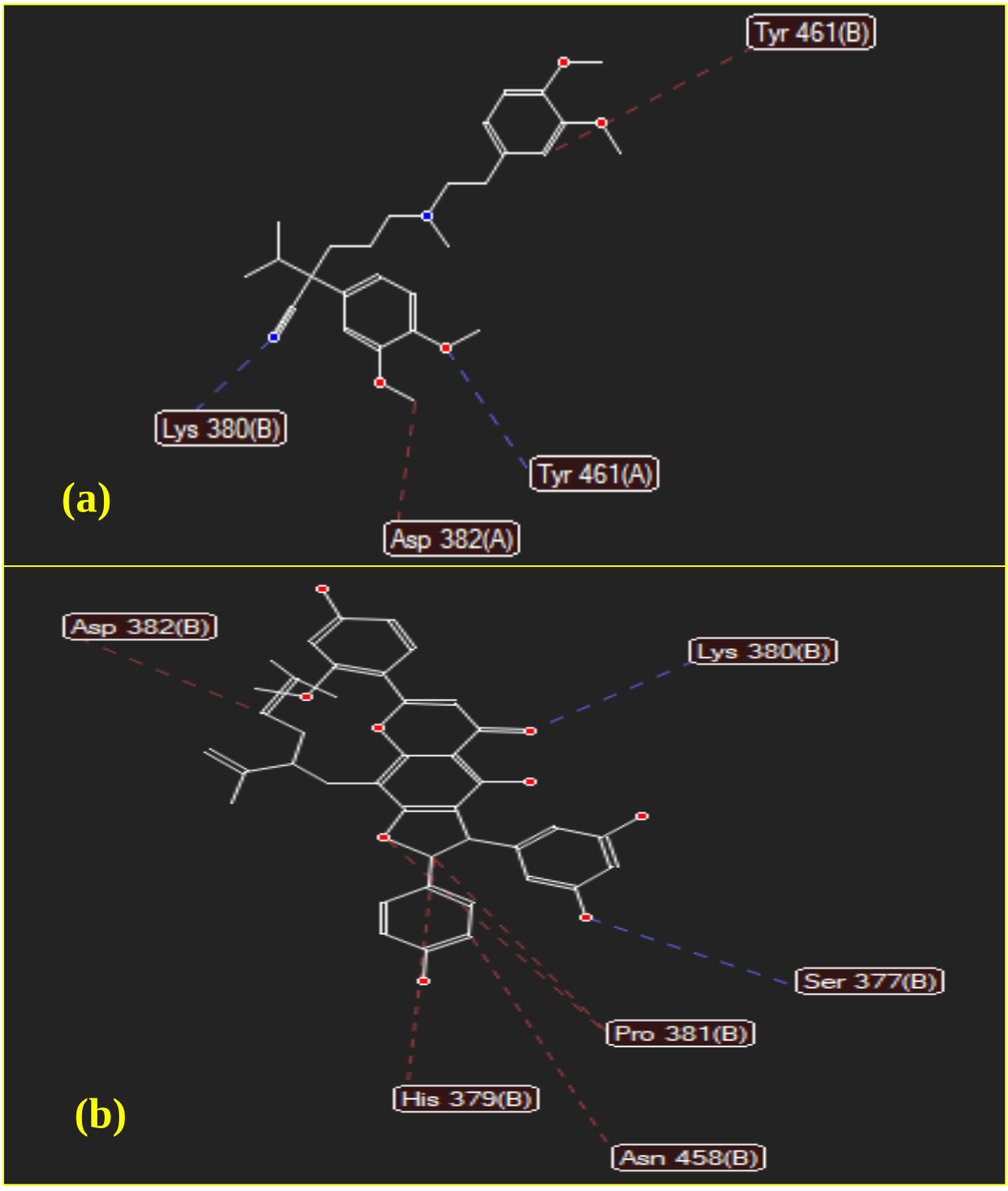

Tyr 461(B)
Lys 380(B)
Tyr 461(A)
Asp 382(A)
(a)
Asp 382(B)
Lys 380(B)
Ser 377(B)
Pro 381(B)
His 379(B)
Asn 458(B)
(b)

**Figure 5**

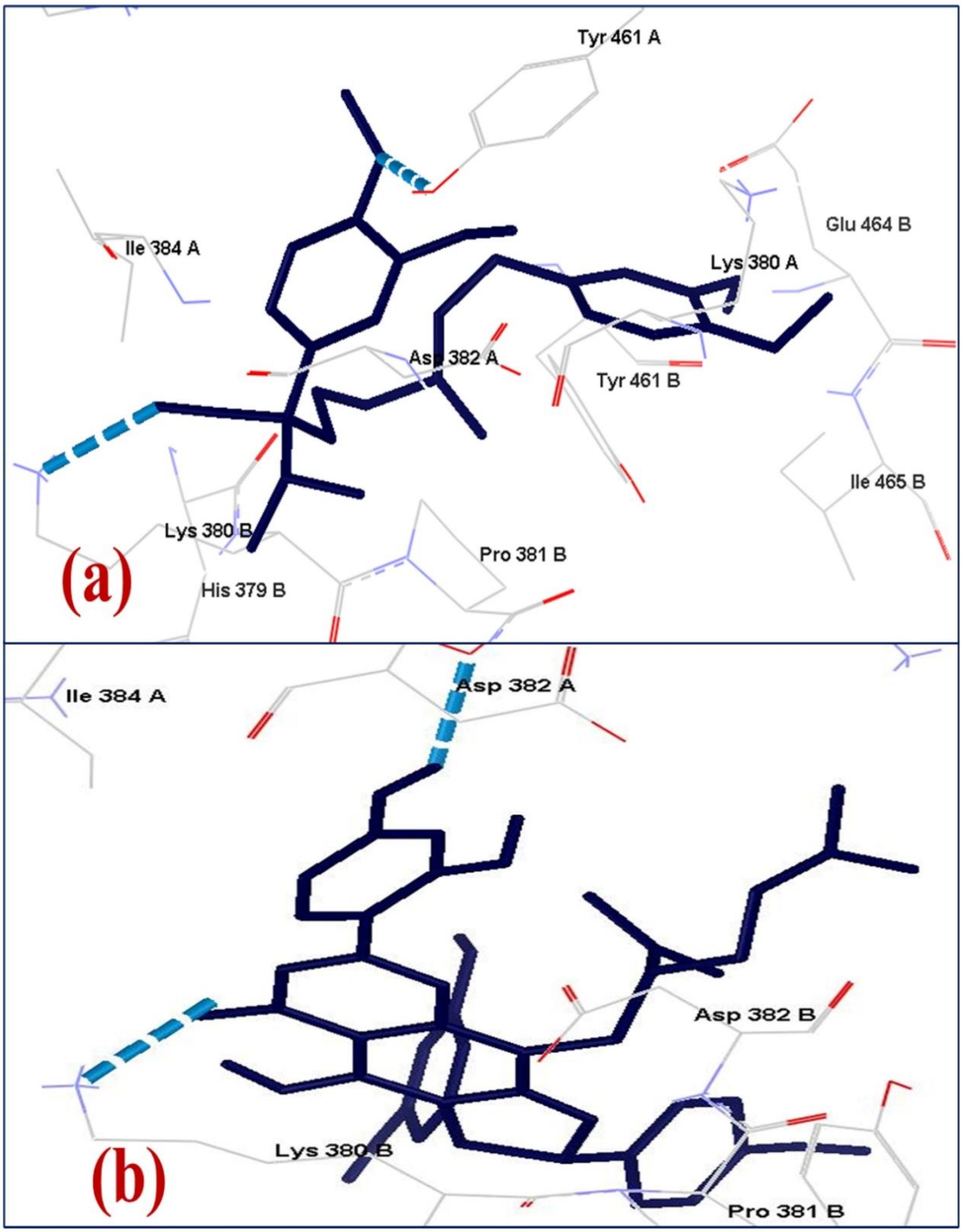

Tyr 461 A
Glu 464 B
Ile 384 A
Lys 380 A
Asp 382 A
Tyr 461 B
Ile 465 B
Lys 380 B
Pro 381 B
(a)
His 379 B
Ile 384 A
Asp 382 A
Asp 382 B
Lys 380 B
(b)
Pro 381 B

**Figure 6**

**Table 1**Predicted potential binding cavities (1–5) within P-gp (PDB Id.3G5U) along with their volume, surface area and position coordinates.

| Cavity | Volume Area (Å$^3$) | Surface Area (Å$^2$) | Position co-ordinates (Å) | | |
|---|---|---|---|---|---|
| | | | X | Y | Z |
| 1 | 1520.64 | 3333.12 | 61.5715 | 80.4877 | 50.8652 |
| 2 | 160.256 | 540.16 | 8.3139 | 57.8180 | 17.7671 |
| 3 | 99.328 | 386.56 | 11.9955 | 83.2312 | 62.7272 |
| 4 | 66.048 | 284.16 | 71.0084 | 23.2991 | 65.2342 |
| 5 | 70.656 | 208.64 | 10.1031 | 43.7275 | 7.9668 |

**Table 2** In silico predicted ADMET and physicochemical properties of the flavones implicated for the designing of lead-like P-gP inhibitors.

| Name of Flavones | Physiochemical Parameters | | | | | | ADMEt Profile | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | Absorption Parameters | | | Distribution Parameters | | | Metabolism Parameters | | Excretion | | Toxicity Profile | |
| | Mole. Wt. (g/mol) | Log P | Hydrogen donor | Hydrogen acceptor | Total polar surface area (Å2) | No. of rotatable bonds | Caco2 permeability | Intestinal absorption (human) | Skin Permeability | VDss (human) | BBB permeability | CNS permeability | Effect on cytochromes P450 | P-gP Substrate | Total Clearance | Renal OCT2 substrate | AMES toxicity | Hepatotoxicity |
| **Verapamil** | 454.6 | 5.0931 | 0 | 6 | 63.95 | 13 | 1.336 | 97.362 | -2.774 | 0.871 | -0.218 | -2.48 | CYP3A4 inhibitior | Yes | 1.023 | No | No | No |
| **Luteoline (4.1)** | 286.24 | 2.2824 | 4 | 6 | 111.13 | 1 | 0.218 | 78.901 | -2.735 | -0.218 | -1.326 | -2.45 | CYP1A2 inhibitior CYP3A4 inhibitior CYP2C9 inhibitior | Yes | 0.556 | No | Yes | No |
| **Baicalin (4.2)** | 270.24 | 2.5768 | 3 | 5 | 90.9 | 1 | 0.997 | 95.472 | -2.735 | -0.506 | -1.219 | -2.296 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor | Yes | 0.252 | No | No | No |
| **Scutellarin (4.3)** | 460.39 | 0.5114 | 7 | 11 | 198.12 | 4 | -0.47 | 32.747 | -2.735 | -0.114 | -1.988 | -4.651 | | Yes | 0.63 | No | No | No |
| **Hesperetin (4.4)** | 300.26 | 2.5854 | 3 | 6 | 100.13 | 2 | 0.713 | 83.152 | -2.899 | 0.263 | -1.027 | -2.322 | CYP1A2 inhibitior CYP2C9 inhibitior | Yes | 0.648 | No | No | No |
| **Nobiletin (4.5)** | 402.39 | 3.5116 | 0 | 8 | 85.59 | 7 | 1.505 | 100 | -2.738 | -0.452 | -1.16 | -2.992 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor CYP3A4 inhibitior | Yes | 0.897 | No | No | No |
| **Sinensetin (4.6)** | 372.37 | 3.503 | 0 | 7 | 76.36 | 6 | 1.407 | 100 | -2.74 | -0.12 | -0.952 | -2.935 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor CYP3A4 inhibitior | No | 0.886 | No | No | No |
| **6-Prenylapigenin (4.7)** | 338.35 | 4.0855 | 3 | 5 | 90.9 | 3 | 0.858 | 92.167 | -2.738 | 0.027 | -1.049 | -2.072 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor | Yes | 0.475 | No | No | No |
| **6-Hydroxyflavone (4.8)** | 238.24 | 3.1656 | 1 | 3 | 50.44 | 1 | 1.25 | 94.327 | -2.744 | -0.233 | 0.297 | -1.747 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor CYP2D6 inhibitior | Yes | 0.277 | Yes | Yes | No |

| | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Neoeriocitrin (4.9)** | 596.49 | -2.4628 | 10 | 16 | 269.43 | 6 | -0.488 | 5.168 | -2.735 | -0.18 | -2.387 | -5.898 | | Yes | 0.17 | No | Yes | No |
| **Negletein(4.10)** | 284.26 | 2.8798 | 2 | 5 | 79.9 | 2 | 1.009 | 94.206 | -2.784 | -0.352 | -0.298 | -2.132 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor CYP3A4 inhibitior | Yes | 0.311 | No | Yes | No |
| **Neohesperidin (4.11)** | 610.52 | -2.1598 | 9 | 16 | 258.43 | 7 | -0.245 | 9.68 | -2.735 | -0.241 | -2.216 | -5.791 | | Yes | 0.278 | No | No | No |
| **6-Aminoflavone (4.12)** | 237.25 | 3.0422 | 1 | 2 | 56.23 | 1 | 1.331 | 96.07 | -2.729 | -0.034 | 0.204 | -1.725 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor CYP2D6 inhibitio | Yes | 0.34 | Yes | Yes | No |
| **Alopecurone A (4.13)** | 648.7 | 8.0503 | 6 | 9 | 160.82 | 8 | 0.11 | 100 | -2.735 | -1.003 | -1.867 | -2.96 | | Yes | -1.071 | No | No | No |
| **Apigenin (4.14)** | 270.24 | 2.5768 | 3 | 5 | 90.9 | 1 | 1.118 | 91.433 | -2.737 | -0.308 | -1.069 | -2.185 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibito | Yes | 0.592 | No | No | No |
| **Alopecurone D (4.15)** | 662.72 | 8.3533 | 5 | 9 | 149.82 | 9 | -0.222 | 100 | -2.735 | -1.052 | -1.79 | -2.839 | | No | -0.907 | No | No | No |
| **Baicalin (4.16)** | 446.36 | 0.1422 | 6 | 11 | 187.12 | 4 | -0.612 | 30.333 | -2.735 | -0.695 | -2.016 | -4.727 | | Yes | 0.276 | No | No | No |
| **Chrysin (4.17)** | 330.29 | 2.594 | 3 | 7 | 109.36 | 3 | 0.369 | 77.89 | -2.736 | -0.053 | -1.317 | -3.261 | CYP1A2 inhibitior CYP3A4 inhibitior | Yes | 0.658 | No | No | No |
| **Diosmetin (4.18)** | 300.26 | 2.5854 | 3 | 6 | 100.13 | 2 | 0.995 | 77.107 | -2.736 | -0.039 | -1.147 | -2.421 | CYP1A2 inhibitior CYP2C9 inhibitior | Yes | 0.722 | No | No | No |
| **Hesperidin (4.19)** | 606.57 | -0.4261 | 8 | 14 | 228.97 | 7 | -0.342 | 27.877 | -2.735 | -0.2 | -1.89 | -5.223 | | Yes | 0.21 | No | Yes | No |
| **Techtochrysin (4.20)** | 59.67 | 3.1742 | 1 | 4 | 59.67 | 2 | 1.325 | 97.046 | -2.47 | 0.006 | 0.247 | -1.914 | CYP1A2 inhibitior CYP2C19 inhibito | Yes | 0.472 | Yes | No | No |
| **Amentoflavone (4.21)** | 538.46 | 5.134 | 6 | 10 | 181.8 | 3 | 0.077 | 82.666 | -2.735 | -1.202 | -2.181 | -3.241 | | Yes | 0.636 | No | No | No |
| **Tangeritin (4.22)** | 372.37 | 3.503 | 0 | 7 | 76.36 | 9 | 1.429 | 99.904 | -2.739 | -0.326 | -0.942 | -2.958 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibitor CYP3A4 inhibitior | No | 0.862 | No | No | No |
| **Jaceosidin (4.23)** | 332.26 | 2.4372 | 4 | 8 | 129.59 | 3 | 0.395 | 69.214 | -2.735 | -0.111 | -1.546 | -3.465 | CYP1A2 inhibitior | Yes | 0.631 | No | No | No |
| **Corylifol C (4.24)** | 338.35 | 4.0855 | 3 | 5 | 90.9 | 3 | 0.854 | 91.118 | -2.735 | -0.557 | -1.084 | -1.978 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibito | Yes | 0.312 | No | No | No |

| | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Artonin (4.25)** | 674.69 | 7.8467 | 8 | 10 | 192.05 | 7 | -0.282 | 96.854 | -2.735 | -0.499 | -2.359 | -3.241 | | Yes | 0.441 | No | No | No |
| **Atocarpin (4.26)** | 436.5 | 5.7633 | 3 | 6 | 100.13 | 6 | 0.813 | 93.766 | -2.735 | -0.662 | -1.234 | -1.953 | CYP2C9 inhibitior CYP2C19 inhibito | Yes | 0.474 | No | No | No |
| **Licoflavone (4.27** | 338.35 | 4.0855 | 3 | 5 | 90.9 | 3 | 0.958 | 90.112 | -2.735 | -0.632 | -1.128 | -1.959 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibito | Yes | 0.413 | Yes | No | No |
| **Semilicoisoflavone B (4.28)** | 352.34 | 3.761 | 3 | 6 | 100.13 | 1 | 0.71 | 95.782 | -2.789 | 0.297 | -1.06 | -2.022 | CYP1A2 inhibitior CYP2C9 inhibitior CYP2C19 inhibito CYP1A4 inhibitior | Yes | 0.215 | No | No | No |
| **Oroxylin A (4.29** | 284.26 | 2.8798 | 2 | 5 | 79.9 | 2 | 1.027 | 95.112 | -2.806 | -0.187 | 0.077 | -2.233 | CYP1A2 inhibitior CYP3A4 inhibitior CYP2C9 inhibitior CYP2C19 inhibito | Yes | 0.382 | No | No | No |
| **Entadanin (4.30)** | 344.27 | 2.1834 | 4 | 8 | 129.59 | 1 | 0.665 | 76.426 | -2.735 | 0.049 | -1.459 | -3.61 | CYP1A2 inhibitior | Yes | 0.518 | No | Yes | No |
| **Wogonin (4.31)** | 284.267 | 2.8798 | 2 | 5 | 79.9 | 2 | 0.92 | 94.964 | -2.762 | -0.216 | -0.041 | -2.298 | CYP1A2 inhibitior CYP3A4 inhibitior CYP2C9 inhibitior CYP2C19 inhibito | Yes | 0.429 | No | No | No |
| **wogonoside (4.32)** | 460.39 | 0.4452 | 5 | 10 | 176.12 | 5 | -0.408 | 36.482 | -2.735 | -0.934 | -1.883 | -4.763 | | Yes | 0.404 | No | No | No |
| **Vitexin (4.33)** | 432.38 | 0.0917 | 7 | 10 | 181.05 | 3 | -0.303 | 50.293 | -2.735 | -0.008 | -2.048 | -4.611 | | Yes | 0.662 | No | Yes | No |
| **Isovitexin (4.34)** | 430.4 | 0.7553 | 7 | 9 | 171.82 | 3 | -0.071 | 49.266 | -2.735 | 0.016 | -1.969 | -4.301 | | Yes | 0.617 | No | Yes | No |
| **Orientin (4.35)** | 448.38 | -0.2027 | 8 | 11 | 201.28 | 3 | -0.61 | 42.403 | -2.735 | 0.148 | -2.296 | -4.911 | | Yes | 0.594 | No | Yes | No |
| **Rhamnosylisoorientin (4.36)** | 593.51 | -1.9829 | 9 | 15 | 263.03 | 5 | -0.414 | 49.241 | -2.735 | 0.193 | -2.353 | -5.567 | | Yes | 0.235 | No | Yes | No |
| **Isoorientin (4.37)** | 448.38 | -0.2027 | 8 | 11 | 201.28 | 3 | -0.756 | 40.733 | -2.735 | 0.231 | -2.419 | -4.791 | | Yes | 0.593 | No | No | No |
| **Maysin (4.38)** | 576.5 | -0.1151 | 8 | 14 | 236.81 | 4 | -0.35 | 68.831 | -2.735 | 0.363 | -2.355 | -5.123 | | Yes | 0.049 | No | Yes | No |
| **Isoschaftoside (4.39)** | 564.49 | -1.7543 | 10 | 14 | 250.97 | 4 | -1.158 | 21.533 | -2.735 | 0.296 | -2.673 | -4.971 | | Yes | -0.01 | No | No | No |
| **Saponarin (4.40)** | 594.52 | -2.4352 | 10 | 15 | 260.2 | 6 | -0.232 | 18.37 | -2.735 | -0.033 | -2.825 | -6.073 | | Yes | 0.37 | No | Yes | No |
| **Acacetin (4.41)** | 284.26 | 2.8798 | 2 | 5 | 79.9 | 2 | 1.004 | 93.909 | -2.778 | -0.224 | -0.287 | -2.208 | CYP1A2 inhibitior CYP3A4 | Yes | 0.724 | No | No | No |

| | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | | | | | | inhibitior CYP2C9 inhibitior CYP2C19 inhibito | | | | | |
| **Tricin (4.42)** | 330.29 | 2.594 | 3 | 7 | 109.36 | 3 | 0.67 | 77.097 | -2.738 | 0.04 | -1.306 | -3.262 | CYP1A2 inhibitior CYP3A4 inhibitior | Yes | 0.687 | No | No | No |
| **Pectolinarigenin (4.43)** | 314.29 | 2.8884 | 2 | 6 | 89.13 | 3 | 1.224 | 94.3 | -2.78 | -0.279 | -0.443 | -2.374 | CYP1A2 inhibitior CYP3A4 inhibitior CYP2C9 inhibitior | Yes | 0.663 | No | No | No |
| **Pectolinarin (4.44)** | 622.57 | -0.7867 | 7 | 15 | 227.2 | 8 | -0.115 | 34.309 | 34.309 | -0.209 | -2.513 | -5.48 | | Yes | 0.536 | No | No | No |
| **Rhoifolin (4.45)** | 578.52 | -1.0983 | 8 | 14 | 228.97 | 6 | -0.167 | 30.44 | -2.735 | 0.03 | -2.327 | -5.327 | | Yes | 0.495 | No | No | No |

**Table 3** The Mol Dock, Re-rank and H-bond score of flavones with P-gP in Kcal/mol.

| Name of ligands | MolDock Score | Rerank Score | H Bond |
|---|---|---|---|
| Verapamil | -79.6711 | -38.3384 | 0 |
| Luteoline (4.1) | -66.8828 | -57.4208 | -7.78949 |
| Baicalin (4.2) | -61.6734 | -59.1988 | -2.5 |
| Scutellarin (4.3) | -60.1541 | -57.9984 | -11.2292 |
| Hesperetin (4.4) | -66.1779 | -53.4898 | 0 |
| Nobiletin (4.5) | -131.488 | -67.9994 | -6.65563 |
| Sinensetin (4.6) | -66.0729 | -55.8129 | -3.86065 |
| 6-Prenylapigenin (4.7) | -137.793 | -77.3561 | -2.20151 |
| 6-Hydroxyflavone (4.8) | -88.2311 | -78.6339 | -10.0051 |
| Neoeriocitrin (4.9) | -65.3198 | -55.7081 | -1.92013 |
| Negletein(4.10) | -66.8696 | -57.7667 | -4.22741 |
| Neohesperidin (4.11) | -60.2394 | -61.7761 | -14.1528 |
| 6- Aminoflavone (4.12) | -66.6084 | -57.6082 | -7.61855 |
| Alopecurone A (4.13) | -64.3409 | -55.7061 | -1.8177 |
| Apigenin (4.14) | -88.2421 | -72.0404 | -13.3487 |
| Alopecurone D (4.15) | -66.9175 | -60.1083 | 0 |
| Baicalin (4.16) | -76.0091 | -68.0697 | -6.23986 |
| Chrysin (4.17) | -75.6822 | -62.3566 | -6.20847 |
| Diosmetin (4.18) | -100.986 | -60.2849 | -3.5012 |
| Hesperidin (4.19) | -75.1363 | -46.0942 | -4.55382 |
| Techtochrysin (4.20) | -76.1145 | -62.4932 | -4.85912 |
| Amentoflavone (4.21) | -80.6874 | -70.5523 | -4.51726 |
| Tangeritin (4.22) | -67.1381 | -60.536 | -4.42621 |
| Jaceosidin (4.23) | -72.3416 | -50.694 | -10.858 |
| Corylifol C (4.24) | -61.5771 | -56.3872 | -4.72462 |
| Artonin (4.25) | -66.86 | -55.9319 | -5.7821 |
| Atocarpin (4.26) | -75.222 | -71.2961 | -5.86772 |
| Licoflavone (4.27) | -88.5816 | -78.6427 | -12.1892 |
| Semilicoisoflavone B (4.28) | -74.6254 | -67.0388 | -9.80673 |
| Oroxylin A (4.29 | -74.8297 | -66.9141 | -6.76787 |

| | | | |
|---|---|---|---|
| Entadanin (4.30) | -102.911 | -78.2184 | -10.5963 |
| Wogonin (4.31) | -87.7968 | -82.547 | -9.80104 |
| wogonoside (4.32) | -56.1185 | -53.9894 | -9.04169 |
| Vitexin (4.33) | -88.7793 | -79.3577 | -13.0655 |
| Isovitexin (4.34) | -74.8441 | -56.314 | -8.10262 |
| Orientin (4.35) | -74.7195 | -70.3868 | -9.43297 |
| Rhamnosylisoorientin (4.36) | -65.7847 | -56.2057 | -2.49568 |
| Isoorientin (4.37) | -71.4257 | -60.9508 | -5.49482 |
| Maysin (4.38) | -62.5691 | -54.1851 | 0 |
| Isoschaftoside (4.39) | -64.6923 | -70.1509 | -8.25904 |
| Saponarin (4.40) | -76.084 | -58.0397 | -10.1255 |
| Acacetin (4.41) | -32.0354 | -33.8327 | 0 |
| Tricin (4.42) | -52.6876 | -45.5791 | 0 |
| Pectolinarigenin (4.43) | -68.5282 | -54.6006 | -5.89783 |
| Pectolinarin (4.44) | -66.2112 | -53.8448 | 0 |
| Rhoifolin (4.45) | -71.9431 | -64.5007 | -7.92299 |

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**Availability of data and materials**

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**Author contribution:**

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# These author contributed equally and are designated as first authors
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LIST OF ABBREVIATIONS

| | |
|---|---|
| MDR | Multi-drug resistance |
| ADME | Absorption, Distribution, Metabolism, Excretion and Toxicity |
| MVD | Molegro Virtual Docker |
| NBDs | Nucleotide-binding domains |
| P-gp | Plasma-glycoprotein |
| BBB | Blood brain barrier |
| OCT2 | Organic cation transporter-2 |
| TPSA | Total polar surface area |
| VD | Volume of distribuion |
| Caco-2 | Cancer coli-2 |
| SAR | Structure-activity relationships |
| QSAR | Quantitative structure-activity relationships |
| ABC | ATP- binding cassette |
| BCRP | Breast cancer resistance protein |

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