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Research Article

VIRTUAL SCREENING OF POTENTIAL DRUG-LIKE INHIBITORS FROM MEDICINAL PLANTS AGAINST HBXIP OF HEPATOCELLULAR CARCINOMA

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ABSTRACT

Objective: Medicinal plants are resources of new drugs. Many of the modern medicines are produced indirectly from plants. Medicinal plants like *Anethum graveolens, Apium graveolens, Camellia sinensis, Citrus sinensis, Daucus carota, Glycine max, Lycopersicon esculentum, Vitis vinifera, Zea mays, Zingiber officinale,* were taken for the study against hepatocellular carcinoma. Hepatitis B Virus X interacting protein is an oncogenic protein and a binding partner of HBx leading to HCC. In present study, HBXIP as target treated with medicinal plants compounds showing anticancer and antiviral activity. The main objective of the study is the inhibition of HBXIP from the phytochemicals present in medicinal plants.

Methods: The three dimensional structure of HBXIP was retrieved from Protein Databank. The anticancer, antiviral activity phytochemicals were screened for ADMET and its Drug likeness activity, and the binding energy of the phytochemicals to the target was studied using docking through Accelerys Discovery studio. Also, the docked phytochemicals were compared with the present drugs for HCC.

Results: Comparative docking results showed that medicinal plant compounds showed a high affinity and dock score than the synthetic drugs available for HCC.

Conclusion: This study is useful for an invitro development of lead molecules from these medicinal plants.

Keywords: HBXIP, ADMET, Druglikeness, Accelerys Discovery studio, Medicinal plants

INTRODUCTION

Medicinal plants defined medicines are widely used in traditional culture all over world and they are becoming increasingly popular in modern society as naturally alternative to synthetic chemicals. The most majority of people on earth still early on their traditional material Medicine for every day health care minerals. "A medicinal plant is any plant which in one or more of its organs containing substances that can be used for therapeutic properties or which are precious for synthesis of useful drugs" [1-4].

Anethum graveolens, dill is an annual, erect, 50-150cm tall and glabrous herb belongs to family Apiaceae. A. graveolens exhibited significant anti-stress, antioxidant [5], antibacterial [6], cardio protective agent [7].

Apium graveolens, Celery, is a biennial with stems 0.3-2.4 m, were erect and branching belonging to the family Apiaceae. The major components of *Apium graveolens* are alkaloids, glycosides, terpenoids, flavonoids, tannin, polyphenols [8, 9]. The plant has anticancer [9], antimicrobial [10], hepatoprotective activity [11].

Camellia sinensis, the tea plant, is a member of the Theaceae family. It is an evergreen shrub or tree can grow to heights of 30 feet appear in clusters or singly. The polyphenols present in tea have demonstrated significant antioxidant, anticarcinogenic, anti-inflammatory, thermogenic, probiotic, and antimicrobial activity [12, 13].

Citrus sinensis, Sweet Orange, a hesperidium belongs to the Rutaceae family [14]. One of the mechanisms by which these phytochemicals exerts their beneficial effects in human health has been related to their antioxidant activity. Phenolics in fruits and vegetables, as well as vitamin C, are said to be effective antioxidants. It was shown that vitamin C contributes in 100% to the total antioxidant activity [15].

Daucus carota, Carrot, is a member of Apiaceae. It has the highest carotenoid content with protective effects against cancer and other chronic diseases [16]. The polyacetylenes (falcarinol-type) found in carrot has anti-inflammatory, antiplatelet-aggregatory, and antitumor activity as well as its activity against bacteria and mycoplasma [17].

Glycine max, Soybean, is a legume belongs to the Fabaceae family. It is an important oil crop worldwide. It ia natural source of Vitamin E, which includes a group of structurally related compounds, alpha, beta, gamma, tocopherols [18] which have anti-inflammatory and anticancer activity [19][20].

Lycopersicon esculentum, tomato, belongs to the Solanaceae family. It is among the most widely consumed vegetables worldwide and an important source of certain antioxidants including lycopene, beta-carotene and vitamin C [21].

Vitis vinifera, grape belongs to the Vitaceae family. Grapes are a rich source of monomeric phenolic compounds, such as Epicatechin, Epicatechin-3-o-gallate and dimeric, trimeric and tetrameric procyanidins and these compounds act as antimuagenic and antiviral agents [22].

Zea mays, corn, belongs to the Poaceae family. The major components of corn are flavonoids [23] It is well known to exhibit wide range of biological effects including anti-inflammatory, antibacterial, antiviral, anti-allergic[24], antioesteoporotic [25] and posses antioxidative and antitumor activities [26]

Zingiber officinale, ginger belongs to family Zingiberaceae. It is consumed worldwide as a spice and flavoring agent and is attributed to have many medicinal properties. It possess antioxidant properties [27].

Hepatitis B X-interacting protein (HBXIP), a cellular 18 KD protein, interacts with the C-terminus of hepatitis B virus (HBV) X protein (HBx). It has been reported that HBXIP may form a complex with survivin, an antiapoptotic protein, resulting in the suppression of cell apoptosis through the mitochondrial/cytochrome pathway. HBXIP can also bind to hSuv3p [29]. In addition, as a regulator of centrosome dynamics and cytokinesis, HBXIP is necessary for bipolar spindle formation in human HeLa carcinoma cells [29].

The present study has mainly focused on the phytochemicals present in the medicinal plants targeting the oncogenic protein, HBXIP. The study helps to develop a prodrug against synthetic drugs.

MATERIALS AND METHODS

Protein Databank

Protein Databank is a structural database contains information about experimentally-determined structures of proteins, nucleic acids, and complex assemblies

Dr.Duke's phytochemical and ethanobotanical Database

This database contains information on the activity of chemicals in plants, and ethanobotanical uses for plants. Databases are searchable by plant (scientific or common name), chemical (e.g., ascorbic acid), or activity (e.g., antiviral).

Chemspider

ChemSpider is a free chemical structure database providing fast text and structure search access to over 28 million structures from hundreds of data sources.

Isis Draw

Isis Draw is a chemical structure 2D drawing program supports chemical fileformats. It is available free for academic and personal use. The mol files of the phytochemicals were generated using this tool.

Accelerys Discovery Studio

Accelerys Discovery Studio software provides comprehensive modeling and simulation capabilities for computational chemists, computational biologists, and other scientists engaged in small molecule and biotherapeutics based research.

Admet of phytochemicals

Open the phytochemical compound. Go to protocol ADMET. Select ADMET distributors click run. After job completed double click on it and view the results. Protocol ADMET. To predict Toxicity ADMET TOPKAT (Toxicity prediction Komputer Analysis Tool) is used. TopKat is performed by Protocol ADMET TOPKAT menu. Choose the models and Change the detailed report ass true and then run. After job completed double click on it and view the results (pdf form)

Druglikeness activity of phytochemicals

The druglikeness activity of phytochemicals is studied through the compounds satisfying Lipinski's rule of five, partition co-effiecient, ADMET properties. This process is analysed through the results of the ADMET from Accelerys Discovery studio.

Energy calculation and minimisation of protein

Open the PDB structure and add hydrogen bonds. Click tools receptor ligand interaction. Select force field and apply force field. Go to protocol simulation and then select calculate energy – run. After job completed double click on it and view the results. Open the energy calculated structure. Then Go to Protocol simulation and then minimization. Change max steps to 3000 and run. After job completed double click on it and view the results. Save the minimized structure.

Activesite prediction of protein

Open the minimized structure and then go to tools, define and editing binding site. Select the protein then tools, select define selected molecule as receptor and then click on select find sites from receptor cavity. Sites will be displayed in hierarchy window. Select the site. Go to tools, select sphere from site then tools, select show/hide residues and then select show/hide site residues and then select the structure then copy it. Go to file new molecule then paste and save it.

Docking-Ligandfit

Open the compound. Open the minimized structure of protein and then select define selected molecule as receptor and then find sites from receptor cavities. Then select the first site and click define sphere form selection. Then go to tools – receptor ligand interaction and click Ligandfit. Run the molecule and view the results.

RESULTS AND DISCUSSION

Database screening for phytochemicals

The phytochemicals from *Anethum graveolens, Apium graveolens, Camellia sinensis, Citrus sinensis, Daucus carota, Glycine max, Lycopersicon esculentum, Vitis vinifera, Zea mays, Zingiber officinale* showing anticancer and antiviral activity were screened from Dr.Duke's phytochemical and ethanobotanical databases. The screening of phytochemicals from the database in specific to antiviral activity and anticancer activity is done because HBXIP is an oncogenic and Hepatitis B virus interacting in nature. There were about many repeated compounds present in these plants. At grand total, there were about 85 compounds were screened for anticancer and antiviral activity.

Structure generation of phytochemicals

The structures of these compounds were obtained from ChemSpider database. The mol files of the phytochemicals were generated using ISIS Draw.

ADMET and Druglikeness of phytochemicals

The phytochemicals were further preceded for the ADME (Absorption, Distribution, Metabolism, and Excretion) and Toxicity study using Accelerys Discovery Studio-ADMET and TOPKAT. Among the 85 compounds, 38 phytochemicals satisfied Lipinski rule of five and Drug likeness activity.

Docking-Ligandfit

The Protein HBXIP (3MSH), functional residues were found to be Asp25, Ser26, Ile45, Val47,Leu48,Ala49, Gln50, Gln51, Ala52, Lys54, Asp61, Ile62, Pro63, Val64,Val65, Ile76, Gln77, Lys78, Gly81, Ile82, Thr83, Val84. The Ligandfit program evaluates candidate ligand poses and prioritized to the Dockscore function. It explains PLP (Piecewise Linear Potential) to correlate with protein ligand binding affinities. It is measured in arbitrary units. Higher PLP scores indicate stronger receptor ligand binding. PMF was computed by summing pairwise interaction terms over all inter atomic pairs of the receptor ligand complex. Almost each phytochemical targeted the HBXIP and its binding energy were studied (Table 1).

Based upon the PLP score, the compounds Apigenin, Chrysin, Luteolin, Myricetin, Naringenin, Scutallerin (Figure 1), were found to have optimal PLP scores and binding energy, less toxicity. Synthetic drugs have poor PLP scores and dock score compared to the phytochemicals. The interaction of the compounds was shown in Figure 2.

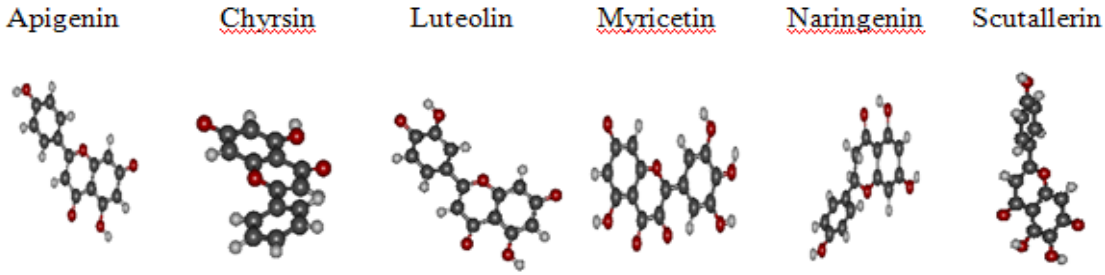


Fig. 1: It shows structures of phytochemicals with high piecewise linear potential (PLP) score

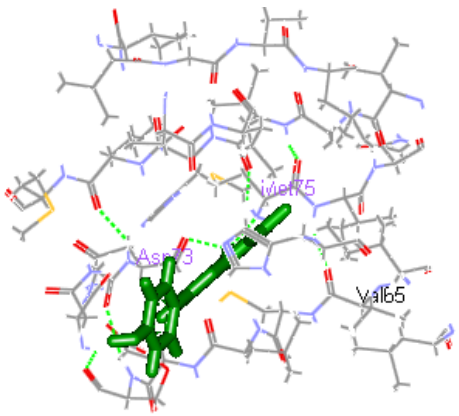
Table 1: It shows docking results of phytochemicals

Compound	LigS1	LigS2	-PLP1	-PLP2	Jain	PMF	Dockscore
Adenine	1.84	2.66	15.32	14.66	-0.74	26.98	72.956
Ascorbic acid	3.13	3.94	46.93	44.43	-0.15	39.99	23.959
Caffeic acid	3.2	3.71	38.96	43.44	0.43	52.05	26.889
Cinnamaldehyde	0.76	3.33	41.01	38.57	0.17	66.69	26.339
Citral	0.86	3.48	37.7	35.1	-0.43	45.01	25.301
Chlorine	-0.1	1.94	5.78	5.52	-0.15	2.98	5.372
Chlorogenic acid	3.53	4.38	66.11	66.19	-0.95	112.14	29.211
Chrysin	2.51	4.39	71.34	58.21	0.69	88.27	35.086
Dipentine	1.15	3.9	48.95	48.3	-0.49	42.6	33.172
Epicatechin	3.31	4.49	81.96	77.43	1.8	76.28	37.144
Eugenol	1.91	4.14	52.09	47.05	0.68	70.28	31.499
Ferulic acid	2.45	3.98	50.89	49.8	0.29	64.18	26.686
Formaldehyde	-0.09	2.14	6.1	3.59	-0.45	8.48	6.512
Gentisic acid	1.72	3.59	46.1	43.77	-0.09	49.75	22.514
Iodine	0.25	2.7	10.84	5.16	-1.68	0	12.863
Isoborneol	1.2	3.52	31.26	29.99	-0.23	81.94	18.827
Kaempferol	0.7	1.06	12.12	17.06	-0.43	47.86	31.772
Lauric acid	1.5	3.78	44.42	42.3	-1.02	74.16	26.98
Limonene	0.79	3.31	36.83	35.35	0.46	58.32	25.766
Lignin	0.86	3.48	37.7	35.1	-0.43	45.01	25.301
Linalool	1.65	3.54	31.64	31.72	-0.64	54.15	24.629
Luteolin	2.1	4.28	78.54	66.53	0.47	86.65	23.604
Myricetin	3.9	4.99	82.02	68.38	-0.35	88.98	26.174
Naringenin	2.6	4.52	75.04	64.85	0.64	85.9	39.448
Neryl acetate	1.61	3.88	38.34	32.71	-0.24	90.32	24.089
P-cymene	0.82	3.35	37.19	36.66	0.52	48.48	26.045
Pelargonidin	3.58	4.5	68.16	64.42	2.55	69.63	39.415
Phenol	0.88	3.37	28.06	27.15	-0.02	37.39	23.243
Protocatechic acid	2.25	4.27	45.85	43.76	-1.06	52.63	26.779
Psoralen	1.88	3.81	54.61	46.94	1.32	78.12	31.953
Quercetin	4.22	4.9	84.31	72.12	0.7	75.52	16.295
Reservatol	1.57	3.87	60.96	56.23	0.21	71.81	33.03
Rosmarinic acid	2.93	4.96	68.25	62.14	-0.19	144.06	32.406
Scutallerin	2.87	4.61	79.81	66.94	0.14	84.52	35.695
Shikimic acid	2.55	4.05	47.43	38.66	-0.34	55.85	24.833
Subaphyllin	1.65	4.53	54.48	52.82	-1.11	91.91	42.672
Theophyllin	1.18	3.75	54.02	45.29	1.53	49.07	28.889
Vanillin	2.34	4.1	53.72	42.92	0.98	54.58	29.42

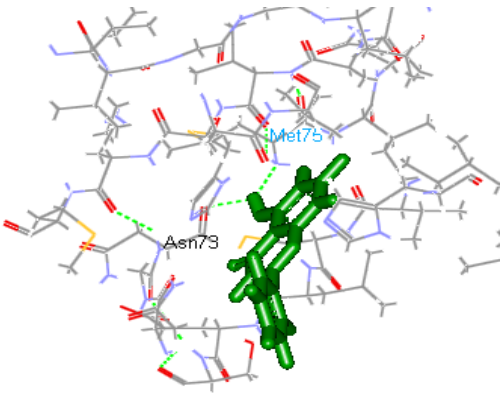
Also, the docking of HBXIP with FDA approved drugs was performed in comparison with phytochemicals (Table 2).

Table 2: It shows docking results of synthetic drugs.

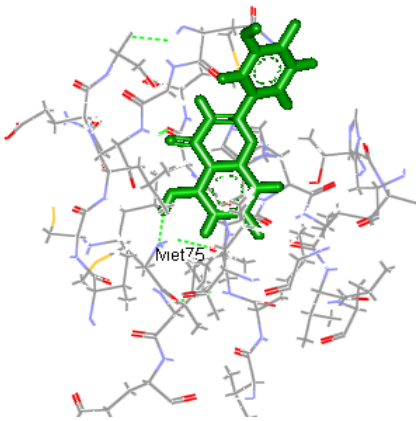
Compound	Ligs1	Ligs2	-PLP1	-PLP2	JAIN	-PMF	Dockscore
Capecitabine	3.3	2.96	20.53	24.06	-1.52	-1.52	21.907
Cisplatin	0.08	2.28	20.09	9.65	-0.77	-3.57	8.023
Gemcitabine	-0.2	1.31	5.16	7.22	-2.27	32.78	10.031
Gemox	1.8	1.41	22.68	19.16	-0.92	39.21	3.944
Irofluven	1.54	3.3	26.5	26.02	-1.39	119.66	17.85
Thalidomide	0.16	2.25	31.03	34.13	-0.57	26.80	15.773
Zalcitabine	0.56	2.93	28.45	18.74	-2.06	33.44	13.227



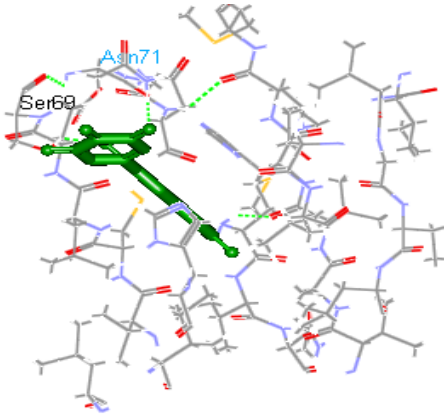
a)Apigenin with HBXIP



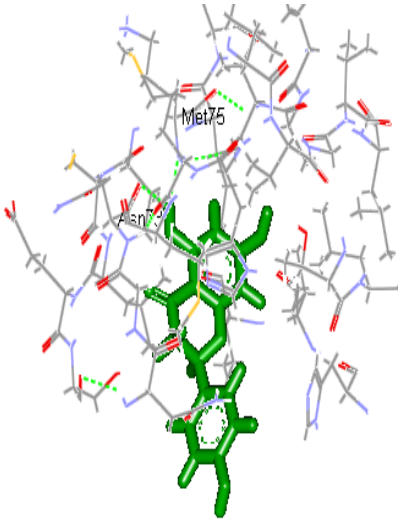
b)Chrysin with HBXIP



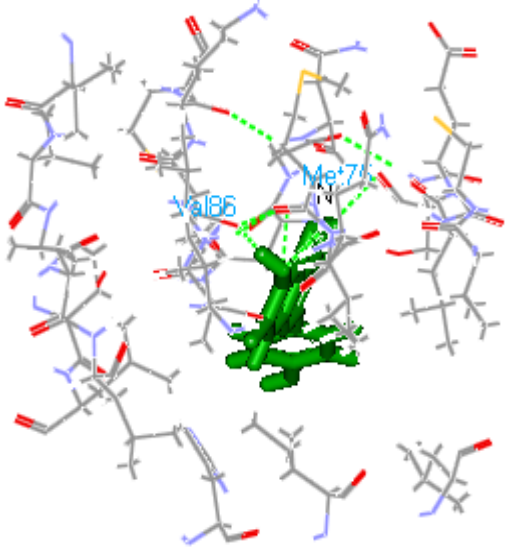
c)Luteolin with HBXIP



d)Myricetin with HBXIP



e)Naringenin with HBXIP



f)Scutallerin with HBXIP

Fig. 2: It shows interaction of phytochemicals with HBXIP

CONCLUSION

Medicinal Plants are potential and powerful to deliver new drugs. The selected medicinal plant contains phytochemicals with various activities. Here we focused mainly on anticancer and antiviral activity because HBXIP is both viral and oncogenic in nature. HBXIP binds to HBX and leads to advance HCC. The inhibition of HBXIP by the compounds can inhibit the binding of HBXIP to HBx. The phytochemicals present in these plants are well-targeted to HBXIP than synthetic drugs. Also, synthetic drugs for HCC have several side effects. Most of all the phytochemicals targeted the protein and bound to the active site with high PLP score and had good affinity. Based on the PLP score and binding affinity to the active site, the phytochemicals are screened for the invitro preparation. Hence the Phytochemicals Apigenin, Chrysin, Luteolin, Myricetin, Naringenin, Scutallerin, can be further proceeded for wet lab preparations to deliver a new drug for Hepatocellular carcinoma.

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