Anthraquinone derivatives as an immune booster and their therapeutic option against COVID-19

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Abstract

Multiple Anthraquinolines derivatives are reported for their immune-boosting, anti-inflammatory and anti-viral efficacy. Hence, the present study dealt to investigate the reported anthraquinone derivatives as an immune booster molecule in COVID-19 infection and evaluate their binding affinity with three reported targets of novel coronavirus i.e. 3CLpro, PLpro and spike proteins. The reported anthraquinone derivatives were retrieved from an open-source database and filtered based on a positive druglikeness score. Further, the probably modulated gene by compounds with positive druglikeness score was evaluated for the modulation of proteins using DIGEP-Pred and the interaction of proteins was evaluated using STRING; associated pathways were recorded concerning the KEGG pathway database. Finally, docking was carried using autodock4; pose scoring minimum binding energy was chosen to visualize the ligand-protein interaction. Among 101 compounds, 36 scored positive druglikeness scores; modulating multiple pathways for immune-boosting as well as pathways involved in infectious and non-infectious diseases. Similarly, docking study revealed torososide B to have the highest binding affinity with PLpro and 3clpro and 1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)- β -D-xylopyranosyl-(1 $\rightarrow$ 2)- β -D-glucopyranoside with spike protein

1. Introduction

Coronaviruses are the group of an RNA virus and cause diseases to mammals and birds; presently, CoV Disease (COVID-19) has to lead to millions of death in the last six months [1]. Further, the risk of getting affected with COVID-19 is more in subjects with lower immunity. Hence, the special subjects like pediatrics/geriatrics and the subjects suffering from multiple communicable and non-communicable diseases are more risk of getting affected with this virus due to lower immunity [2]. Although approaches are being made to prevent this virus to get spread via social distancing and so on; boosting of immunity in subjects could play an important role in inhibiting the transmission of the virus and its invasion into the body. Investigations are undergoing to develop the vaccine against COVID-19, but it may still take time as the drug discovery process is much complicated. Hence, it is important to identify any alternative approach as prophylaxis against COVID-19 which can be implemented via the utilization of immune boosters from natural sources. Three targets 3C-like protease (3CLpro), papain-like protease (PLpro), and spike protein [3-6] are being targeted by multiple investigators to identify the new hit molecule for the management of COVID-19. Further, in COVID-19 infection severe inflammation and cell necrosis are contributing to the worsening of pathogenesis. Hence, it may be beneficial if a new drug molecule is identified as anti-viral with anti-inflammatory and immune-boosting properties.

Anthroquinolines are the group of compounds from multiple folk medicines like Senna species which are utilized in Ayurvedic system of medicines and Traditional Chinese Medicines for the management of various infectious and non-infectious diseases [7, 8]. Further, multiple anthraquinone derivatives are also reported as the anti-viral potency [9], anti-inflammatory [10] and also act as immune booster [11]. Since, in the COVID-19 infection, it may be beneficial if the bioactive is identified that acts as the immune booster, anti-inflammatory, and as well as act as anti-viral property which can be demonstrated via the concept of network pharmacology or polypharmacological approach.

Hence, based on the above theme, of anti-viral/anti-inflammatory/immune boosting reports of anthraquinones we attempted to screen the multiple anthraquinone derivatives as an immune booster and anti-viral efficacy using *in silico* and system biology tools.

2. Materials And Methods

2.1. Bioactives and their druglikeness score

The reported phytoconstituents under the phytochemistry of anthraquinone were retrieved from available literature/ChEBI records (<https://www.ebi.ac.uk/chebi/>). All the compounds were then subjected to a druglikeness score by querying the SMILES of each molecule in MolSoft (<https://molsoft.com/mprop/>).

2.2. Target prediction and their enrichment analysis

Anthraquinone derivatives with positive druglikeness scores were then queried in DIGEP-Pred [12] to query "*Proteins based targets*"; up-regulated/downregulated protein at the probable activity of 0.5. The list of regulated proteins was then queried in STRING [13] to identify the biological process, cellular function, and molecular processes of combined gene-set. Further, the probably modulated pathways were also identified concerning the KEGG pathway database. Network among the bioactives, their targets, and probably modulated targets were constructed using the Cytoscape [14] version 3.5.1. Any duplicates during the interconnection of two nodes were eliminated to avoid the appearance of a false hit. The whole network was then analyzed using the "*network analyzer*" tool based on node size and count representing the edge count as "*low values to small size*" and "*low values to bright colors*" respectively as explained previously [16].

2.3. Prediction of probable anti-viral activity

Anti-viral activity of each ligand molecule was predicted by querying the SMILES in PASS [17] at Probable activity (Pa) > probable inactivity (Pi); queried keyword "*anti-viral*" as its probable biological spectrum. Records were queried for their probable pharmacological spectrum against multiple viruses like Adenovirus, CMV, Hepatitis B, Hepatitis C, Hepatitis, Herpes, HIV, Influenza A, Influenza, Parainfluenza, Picornavirus, Poxvirus, Rhinovirus, and Trachoma.

2.4. *In silico* molecular docking

2.4.1 Preparation of ligand molecules

All the 3D.sdf format of ligand molecules with positive drug-likeness scores was retrieved from PubChem database or structures were drawn in ChemSketch (<https://www.acdlabs.com/resources/freeware/chemsketch/>) if not available. The ligand molecules were then converted into.pdb format using Discovery studio 2019 [18]. All the bioactives were then minimized using MMFF94 forcefield [19] using conjugate gradients as an optimization algorithm. After the minimization of energy, all ligand molecules were converted into.pdbqt format.

2.4.2 Preparation of macromolecules

Three target proteins of COVID-19 i.e. coronavirus main proteinase (3clpro); PDB: 6LU7, papain-like protease (PLpro); PDB: 4M0W and spike proteins (homology modeled target, accession number: AVP78042.1 as query sequence and PDB: 6VSB as a template using SWISS-MODEL [20] were selected. The retrieved proteins were in complex with other heteroatoms which were removed using Discovery studio 2019 and saved in.pdb format.

2.4.3 Ligand-protein docking

The ligand molecules were docked with protein molecules using autodock4 [21] by setting 8 exhaustiveness as default to obtain 10 different poses of ligand molecules. After docking the pose scoring lowest binding energy with the target was selected to visualize the ligand-protein interaction in Discovery Studio 2019 as explained previously [22,23].

3. Results

3.1. Bioactives and their druglikeness score

The complete datasheet of 101 compounds including their name, ChEBI ID, Molecular formula/weight, and synonym including phytochemistry for the retrieved compounds was summarized; *Table S1*. Among 101 different bioactives; 36 bioactives were identified with positive druglikeness scores. Among them laccaic acid A scored highest druglikeness score i.e. 0.85 with molecular weight 537.09, 12 hydrogen bond acceptor, 8 hydrogen bond donors and 2.88 MolLogP. The details of the druglikeness score of each compound are summarized in *Table 1*.

3.2. Target prediction and enrichment analysis

Among the compounds with positive druglikeness score, anthragallol was predicted to modulate the highest number of genes i.e. 25. Similarly, human carbonyl reductase 1 (CBR1) was targeted by the highest number of bioactives i.e. 33. Similarly, the enrichment analysis identified the modulation of 54 different pathways in which pathways in cancer was majorly modulated by regulating 12 genes (AR, CASP8, CDK4, CTNNB1, EPAS1, HMOX1, KLK3, MMP2, NFE2L2, NOS2, RAC1, RARA) under the background of 515 proteins at the false discovery rate of $7.71\text{E}-08$. *Table 2* summarizes the gene enrichment analysis of the modulated gene set along with modulated pathways with their respective gene codes. The protein-protein interaction of modulated proteins is presented in *Figure 1*. Similarly, the combined bioactives-proteins-pathways also reflected the anthragallol to target the highest number of proteins. Likewise, TNFRSF1A was majorly targeted by the highest number of bioactives modulating the pathways in the cancer pathway (*Figure 2*).

3.3. Prediction of probable anti-viral activity

The anthraquinones were found to be anti-viral agents against Adenovirus, CMV, Hepatitis B, Hepatitis C, Hepatitis, Herpes, HIV, Influenza A, Influenza, Parainfluenza, Picornavirus, Poxvirus, Rhinovirus, and Trachoma. Among them, the majority of the compounds were active against herpes virus i.e. 13.28%. The overall activity of compounds against multiple viruses is summarized in *Figure 3*.

3.4. *In silico* molecular docking

Torososide B was predicted to have the highest binding affinity (-8.7 kcal/mol) with Papain-Like Protease with 9 hydrogen bond interactions with THR302, ASP303, TYR274, TYR265, ARG167, TYR269, ASP165. Further, Torososide B was predicted to possess the highest binding affinity (-9.3 kcal/mol) with Coronavirus Main Proteinase with 14 hydrogen bond interaction with LEU287, TYR237, THR199, ARG131, LYS137, LYS5, GLU290, ILE281, LEU282, PHE3. Similarly, 1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)- β -D-xylopyranosyl-(1 $\rightarrow$ 2)- β -D-glucopyranoside was predicted to possess the highest binding affinity (-8.7 kcal/mol) with spike protein with the highest number of hydrogen bond interactions i.e. 6 with ASP820, ILE816, ASP815, GLN825, MET703. The binding

affinity of each compound with individual targets with hydrogen bond interactions and residues is summarized in *Table S2*. The interaction of Torososide B with Papain-Like Protease and Coronavirus Main Proteinase and 1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)- β -D-xylopyranosyl-(1 $\rightarrow$ 2)- β -D-glucopyranoside with spike protein is presented in *Figure 4*.

4. Discussion

During the COVID-19 infection, several necrosis and inflammation are leading to defect in the supply of necessary nutrients and oxygen into the cells which are more terrible in the subjects with compromised immunity. Hence, in the present study, we investigated multiple anthraquinone derivatives from various traditional medicines to act against COVID-19 targets i.e. 3CLpro, PLpro, and spike protein, and their combined immune-boosting efficacy. Initially, the majority of the plant-based medicines are utilized via the oral route we calculated their druglikeness score based on the "*Lipinski's Rule of Five*" [24] which identified 35 different bioactives with positive druglikeness score and were considered to get absorbed from the oral route. Hence, these thirty-six molecules (Table 1) were considered for further study.

The conventional drug discovery process utilizes the concept of "*single drug-single protein-single disease*" [25] which may not be applicable in the management of infectious diseases. This is due to the affinity of the pathogens (viruses/bacteria) to affect the homeostatic function of protein molecules. It means multiple proteins from the pathogens are involved to generate this effect. Hence, this can be managed via the utilization of modified drug development process "*multi compound-multi protein-disease*" interaction in which multiple bioactives regulate multiple proteins [15] which can also be taken as a basic key of boosting the immune system. Hence, in the present study, the combined synergistic phenomena of anthraquinones were investigated rather than a single bioactive molecule that identified the multiple pathways which are directly or indirectly involved in the immune system.

Gene set enrichment analysis identified multiple pathways like p53 signaling pathway [26], PI3K-Akt signaling pathway [27], Rap1 signaling pathway [28], NF-kappa B signaling pathway [29] which are directly involved in the boosting the immune system. Similarly, some other pathways like pathways in cancer, PPAR signaling pathway, colorectal cancer, chemical carcinogenesis, estrogen signaling pathway were also identified which reflects the potency of anthraquinones to be beneficial in the subjects which are suffering from these pathways associated diseases like cancer. Further, pathways like p53 signaling pathways, PI3K-AKT, Wnt signaling pathways are also associated diseases like compromised immune systems like diabetes and obesity; which could be beneficial in managing the diseases they are suffering and boosting the immune system. Additionally, the enrichment analysis identified the modulation of multiple pathways that are associated with a pathogenic infection like viral myocarditis and tuberculosis; reflects the potency to manage the infectious diseases. Further, previous reports also reflect the anti-viral potency of the multiple plants that are rich in anthraquinones against various viruses. Hence, we attempted to identify the probable anti-viral activity of the anthraquinones with positive druglikeness score which identified their efficacy against multiple viruses like the rhino, influenza, herpes trachoma, pox, and CMV.

3C-like protease or main protease of coronavirus (3CLpro) alters the ubiquitin system to incorporate the viral polypeptides; deregulates the homeostatic task of functional proteins [30]; which was majorly targeted by Torososide B. Further, PLpro alters the function of protein phosphatase 1A (pp1a) and protein phosphatase 1B (pp1b) into the replicase proteins to adjust viral life cycle [31]; majorly inhibited by Torososide B. Similarly, spike protein utilizes angiotensin-converting enzyme 2 (ACE-2) as a receptor to enter inside the host cell [32, 33] which was chiefly regulated by modulated by 1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)- β -D-xylopyranosyl-(1 $\rightarrow$ 2)- β -D-glucopyranoside. These results reflect the potency of the anthraquinone derivatives to act as the anti-viral

potency against COVID–19. However, as the time proceeds, it is to be understood that the binding affinity of probable lead hit molecules may get altered due to mutation in the possible protein targets and the inhibitory function may not occur as predicted.

5. Conclusion

The present study utilized in silico molecular docking tools to identify the binding affinity of previously recorded anthraquinones derivatives against 3clpro, PLpro, and spike protein which identified Torososide B and 1,3,6-trihydroxy–2-methyl–9,10-anthraquinone–3-O-(6'-O-acetyl)- β -D-xylopyranosyl-(1 $\rightarrow$ 2)- β -D-glucopyranoside as a lead hits. Similarly, the combined synergies of the network identified the modulation of multiple pathways involved in the immune system like p53, chemokine, and PI3K-Akt signaling pathways. Additionally, anthraquinone derivatives were also identified as the modulators of the disease pathways where the immune system is compromised like diabetes and obesity. All these results suggest the probable therapeutic option of utilization of anthraquinones as an immune booster and anti-viral against novel coronavirus by acting on the three targets as investigated. However, the present findings are only based on the computer simulations which need to be further validated using well designed experimental protocols.

Declarations

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Authors' contribution

Pukar Khanal developed the protocol and performed the work and drafted the manuscript. Prof. B.M. Patil reviewed and finalized the manuscript draft. Jagdish chand and Yasmin Naaz had equal contribution in mining the database and assisting in the work performance.

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Conflict of interest

There is no conflict of interest.

Ethical statement

This manuscript doesn't include any animal or human studies

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Tables

Table 1: Druglikeness of anthraquinone derivatives with positive score

Bioactives	Molecular Formula	Molecular Weight	NHBA	NHBD	MolLogP	MolPSA (Å ²)	MolVol (Å ³)	DLS
versicolorone tricyclic form	C ₂₀ H ₁₈ O ₈	386.1	8	5	1.67	122.82	375.84	0.09
(1'S,5'S)-5'-hydroxyaverantin	C ₂₀ H ₂₀ O ₈	388.12	8	6	1.9	125	371.14	0.17
(1'S,5'R)-5'-hydroxyaverantin	C ₂₀ H ₂₀ O ₈	388.12	8	6	1.9	125	371.14	0.17
chrysophanol 8-O-β-D-glucoside	C ₂₁ H ₂₀ O ₉	416.11	9	5	1.11	122.67	379.02	0.45
1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)-α-L-rhamnopyranosyl-(1->2)-β-D-glucopyranoside	C ₂₉ H ₃₂ O ₁₅	620.17	15	7	1.46	189.78	556.02	0.6
1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)-β-D-glucopyranoside	C ₂₃ H ₂₂ O ₁₁	474.12	11	5	2.07	143.34	436.83	0.45
1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-α-L-rhamnopyranosyl-(1->2)-β-D-glucopyranoside	C ₂₇ H ₃₀ O ₁₄	578.16	14	8	0.85	185.66	510.27	0.5
1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)-β-D-xylopyranosyl-(1->2)-β-D-glucopyranoside	C ₂₈ H ₃₀ O ₁₅	606.16	15	7	1.01	190.6	540.52	0.69
1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(3'-O-acetyl)-α-L-rhamnopyranosyl-(1->2)-β-D-glucopyranoside	C ₂₉ H ₃₂ O ₁₅	620.17	15	7	1.42	190.3	556.1	0.76
1-hydroxy-2-(β-D-glucosyloxy)-9,10-anthraquinone	C ₂₀ H ₁₈ O ₉	402.1	9	5	1.1	121.6	358.41	0.04
(2S)-versicolorone	C ₂₀ H ₁₈ O ₈	386.1	8	5	1.67	122.82	375.84	0.09
(S)-5'-oxoaverantin	C ₂₀ H ₁₈ O ₈	386.1	8	5	1.77	121.86	374.32	0.16
BDA-366	C ₂₄ H ₂₉ N ₃ O ₄	423.22	5	3	2.54	74.7	440.1	0.09
Emodin 8-glucoside	C ₂₁ H ₂₀ O ₁₀	432.11	10	6	0.61	140.29	389.71	0.74
anthragallol	C ₂₁ H ₂₀ O ₁₀	432.11	10	6	0.61	140.29	389.71	0.74

nogalonic acid	C ₂₀ H ₁₄ O ₈	382.07	8	3	2.33	113.17	370.61	0.44
mitoxantrone	C ₂₂ H ₂₈ N ₄ O ₆	444.2	8	8	-0.55	135.33	432.17	0.53
torososide B	C ₄₀ H ₅₂ O ₂₅	932.28	25	14	-4.46	320.41	789.36	0.63
versicolorone	C ₂₀ H ₁₆ O ₇	368.09	7	3	2.68	97.9	364.43	0.14
4'-O-demethylknipholone-4'-O-β-D-glucopyranoside	C ₂₉ H ₂₆ O ₁₃	582.14	13	8	1.91	184.8	529.73	0.61
aklanonic acid	C ₂₁ H ₁₆ O ₈	396.08	8	3	2.83	112.59	389.73	0.51
kermesic acid	C ₁₆ H ₁₀ O ₈	330.04	8	5	2.1	119.71	304.77	0.06
gaboroquinone A	C ₂₄ H ₁₈ O ₉	450.1	9	5	3.35	130.76	428.49	0.27
variecolorquinone A	C ₂₀ H ₁₈ O ₉	402.1	9	4	1.36	121.77	383.49	0.71
laccaic acid A	C ₂₆ H ₁₉ NO ₁₂	537.09	12	8	2.88	186.68	496.66	0.85
laccaic acid B	C ₂₄ H ₁₆ O ₁₂	496.06	12	8	3.19	178.91	446.68	0.65
laccaic acid C	C ₂₅ H ₁₇ NO ₁₃	539.07	14	10	0.68	211.46	476.5	0.68
carminic acid	C ₂₂ H ₂₀ O ₁₃	492.09	13	9	0.62	189.58	434.4	0.77
ekatetrone	C ₁₉ H ₁₃ NO ₇	367.07	7	4	2.12	112.76	348.31	0.5
4-bromo-1-hydroxyanthraquinone-2-carboxylic acid	C ₁₅ H ₇ BrO ₅	345.95	5	2	4.39	70.07	269.12	0.24
scutianthraquinone A	C ₃₉ H ₃₂ O ₁₃	708.18	13	4	7.75	165.64	711.03	0.45
scutianthraquinone B	C ₃₈ H ₃₀ O ₁₃	694.17	13	4	7.41	165.64	693.71	0.26
scutianthraquinone C	C ₃₄ H ₂₄ O ₁₂	624.13	12	5	5.88	161.07	618.22	0.31
1,3,6-trihydroxy-2-hydroxymethyl-9,10-anthraquinone-3-O-(6'-O-acetyl)-β-D-glucopyranoside	C ₂₃ H ₂₂ O ₁₂	490.11	12	6	0.93	160.45	445.01	0.41
rubianthraquinone	C ₁₆ H ₁₂ O ₅	284.07	5	2	3.26	68.06	281.86	0.07
5-hydroxyanthraquinone-1,3-dicarboxylic acid	C ₁₆ H ₈ O ₇	312.03	7	3	3.47	99.54	281.87	0.28

DLS: Druglikeness score

Table 2: Enrichment analysis of modulated proteins by the reported anthraquinone derivatives

#term ID	term description	observed gene count	background gene count	false discovery rate	matching proteins in your network (labels)
hsa05200	Pathways in cancer	12	515	7.71E-08	AR, CASP8, CDK4, CTNNB1, EPAS1, HMOX1, KLK3, MMP2, NFE2L2, NOS2, RAC1, RARA
hsa05418	Fluid shear stress and atherosclerosis	7	133	1.18E-06	CTNNB1, HMOX1, MMP2, NFE2L2, PLAT, RAC1, TNFRSF1A
hsa05167	Kaposi's sarcoma-associated herpesvirus infection	6	183	0.00012	CASP8, CD86, CDK4, CTNNB1, RAC1, TNFRSF1A
hsa05215	Prostate cancer	5	97	0.00012	AR, CTNNB1, KLK3, PLAT, PLAU
hsa05014	Amyotrophic lateral sclerosis (ALS)	4	50	0.00015	CAT, GPX1, RAC1, TNFRSF1A
hsa04932	Non-alcoholic fatty liver disease (NAFLD)	5	149	0.00044	ADIPOQ, CASP8, PPARA, RAC1, TNFRSF1A
hsa05202	Transcriptional misregulation in cancer	5	169	0.00068	CD86, FLT1, PLAT, PLAU, RARA
hsa04066	HIF-1 signaling pathway	4	98	0.0012	FLT1, HMOX1, NOS2, TIMP1
hsa00380	Tryptophan metabolism	3	40	0.0017	CAT, CYP1A1, CYP1A2
hsa04915	Estrogen signaling pathway	4	133	0.003	FKBP5, MMP2, PGR, RARA
hsa05416	Viral myocarditis	3	56	0.0037	CASP8, CD86, RAC1
hsa00980	Metabolism of xenobiotics by cytochrome P450	3	70	0.0053	CBR1, CYP1A1, CYP1A2
hsa04115	p53 signaling pathway	3	68	0.0053	CASP8, CDK4, CHEK1
hsa04920	Adipocytokine signaling pathway	3	69	0.0053	ADIPOQ, PPARA, TNFRSF1A
hsa05152	Tuberculosis	4	172	0.0053	CASP8, NOS2, TNFRSF1A, VDR
hsa05225	Hepatocellular carcinoma	4	163	0.0053	CDK4, CTNNB1, HMOX1, NFE2L2
hsa05203	Viral carcinogenesis	4	183	0.0056	CASP8, CDK4, CHEK1, RAC1
hsa05204	Chemical carcinogenesis	3	76	0.0056	CBR1, CYP1A1, CYP1A2
hsa05205	Proteoglycans in cancer	4	195	0.0065	CTNNB1, MMP2, PLAU, RAC1
hsa04933	AGE-RAGE signaling pathway in diabetic complications	3	98	0.0097	CDK4, MMP2, RAC1
hsa04620	Toll-like receptor	3	102	0.01	CASP8, CD86, RAC1

signaling pathway					
hsa05142	Chagas disease (American trypanosomiasis)	3	101	0.01	CASP8, NOS2, TNFRSF1A
hsa05145	Toxoplasmosis	3	109	0.0113	CASP8, NOS2, TNFRSF1A
hsa04670	Leukocyte transendothelial migration	3	112	0.0117	CTNNB1, MMP2, RAC1
hsa05166	HTLV-I infection	4	250	0.0121	CDK4, CHEK1, CTNNB1, TNFRSF1A
hsa04215	Apoptosis - multiple species	2	31	0.0131	CASP8, TNFRSF1A
hsa04216	Ferroptosis	2	40	0.0204	GSS, HMOX1
hsa04310	Wnt signaling pathway	3	143	0.0204	CTNNB1, MMP7, RAC1
hsa05219	Bladder cancer	2	41	0.0204	CDK4, MMP2
hsa05224	Breast cancer	3	147	0.0204	CDK4, CTNNB1, PGR
hsa05165	Human papillomavirus infection	4	317	0.0226	CASP8, CDK4, CTNNB1, TNFRSF1A
hsa00480	Glutathione metabolism	2	50	0.0261	GPX1, GSS
hsa04978	Mineral absorption	2	51	0.0263	HMOX1, VDR
hsa04151	PI3K-Akt signaling pathway	4	348	0.0285	CDK4, FLT1, GH1, RAC1
hsa00140	Steroid hormone biosynthesis	2	58	0.0316	CYP1A1, CYP1A2
hsa00590	Arachidonic acid metabolism	2	61	0.0337	CBR1, GPX1
hsa00830	Retinol metabolism	2	62	0.0338	CYP1A1, CYP1A2
hsa04024	cAMP signaling pathway	3	195	0.0339	PPARA, RAC1, TNNI3
hsa04510	Focal adhesion	3	197	0.034	CTNNB1, FLT1, RAC1
hsa04015	Rap1 signaling pathway	3	203	0.0359	CTNNB1, FLT1, RAC1
hsa05211	Renal cell carcinoma	2	68	0.0363	EPAS1, RAC1
hsa01524	Platinum drug resistance	2	70	0.0374	CASP8, TOP2A
hsa04520	Adherens junction	2	71	0.0375	CTNNB1, RAC1
hsa03320	PPAR signaling pathway	2	72	0.0376	ADIPOQ, PPARA
hsa05100	Bacterial invasion of epithelial cells	2	72	0.0376	CTNNB1, RAC1

hsa05212	Pancreatic cancer	2	74	0.0379	CDK4, RAC1
hsa04610	Complement and coagulation cascades	2	78	0.0409	PLAT, PLAU
hsa04146	Peroxisome	2	81	0.0429	CAT, NOS2
hsa05132	Salmonella infection	2	84	0.045	NOS2, RAC1
hsa05210	Colorectal cancer	2	85	0.045	CTNNB1, RAC1
hsa05323	Rheumatoid arthritis	2	84	0.045	CD86, FLT1
hsa04211	Longevity regulating pathway	2	88	0.0462	ADIPOQ, CAT
hsa05222	Small cell lung cancer	2	92	0.0492	CDK4, NOS2
hsa04064	NF-kappa B signaling pathway	2	93	0.0493	PLAU, TNFRSF1A

Figures

Figure 1

Protein-protein interaction of regulated proteins

Figure 2

Network interaction of anthraquinone derivatives with their proteins and regulated pathways

Figure 3

Predicted anti-viral activity of anthraquinone derivatives against multiple viruses

A **B** **C** **Figure 4**

Interaction of Torososide B with (A) Papain-Like Protease and (B) Coronavirus Main Proteinase and 1,3,6-trihydroxy-2-methyl-9,10-anthraquinone-3-O-(6'-O-acetyl)- β -D-xylopyranosyl-(1 $\rightarrow$ 2)- β -D-glucopyranoside (C) with spike protein

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [TableS1S2.xlsx](#)