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Pharmacophore modeling, virtual screening and molecular docking of ATPase inhibitors of HSP70

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ABSTRACT

Heat shock protein 70 is an effective anticancer target as it influences many signaling pathways. Hence the study investigated the important pharmacophore feature required for ATPase inhibitors of HSP70 by generating a ligand based pharmacophore model followed by virtual based screening and subsequent validation by molecular docking in Discovery studio V4.0. The most extrapolative pharmacophore model (hypotheses 8) consisted of four hydrogen bond acceptors. Further validation by external test set prediction identified 200 hits from Mini Maybridge, Drug Diverse, SCPDB compounds and Phytochemicals. Consequently, the screened compounds were refined by rule of five, ADMET and molecular docking to retain the best competitive hits. Finally Phytochemical compounds Muricatetrocin B, Diacetylphiladelphicalactone C, Eleutheroside B and 5-(3-{[1-(benzylsulfonyl)piperidin-4-yl]amino} phenyl)- 4-bromo-3-(carboxymethoxy)thiophene-2-carboxylic acid were obtained as leads to inhibit the ATPase activity of HSP70 in our findings and thus can be proposed for further *in vitro* and *in vivo* evaluation.

1. Introduction

HSP70, a stress inducible protein is an important member of the Chaperon Protein family. It is a 70 Kilo Dalton, ATP-dependent molecular Chaperon. HSP70 plays a major role in protein folding and re-folding misfolded client proteins, also in the clearance of aberrant client proteins and inhibition of programmed cell death (Clare and Saibil, 2013). The heat shock family members also contributes in malignancy by resistance induction to chemotherapy, apoptosis inhibition and in the stability of oncoproteins (Fewell et al., 2011). HSP 70 acts as co chaperons of heat shock protein 90 (HSP 90) and inhibits effectors of the apoptotic machinery, signaling pathways and caspase-activation (Mayer and Bukau, 2005).

Heat shock protein 70 is detectable at low concentrations in most unstressed normal cells and tissues and is over-expressed in tumor cells. In noncancerous cell lines, silencing of HSP 70 isoforms had not arrested cell growth thus acting as a prospective target for targeted therapy. The Chaperon activities of Hsp70 and Hsp90 depend highly on the hydrolyses of ATP energy and posses conserved ATPase domain (Craig et al., 2006; Kityk et al., 2012).

Also, cellular HSP 70 in complex with HSP 90 helps in reverse transcriptase activity of hepatitis, biosynthesis of envelope protein, helicase activity & transcriptional activation of Papillomavirus, initiation of replication herpes simplex virus (Brown et al., 2005; Chromy et al., 2006).

Recent investigations emphasize that targeting HSP70 in combination with HSP 90 is important for effective cancer treatment as they coregulate the molecular mechanisms involved in cancer cycle (Balaburski et al., 2013; Evans et al., 2010; French et al., 2013; Powers et al., 2010). So far, the explored inhibitors of HSP70 was found to act at different sites like substrate binding domain (PES, PES-CL); ATP binding site(VER-155008); Allosteric binding site near the ATP binding site (MKT-077) however the experimental evidences reveal only slight differences in their mode of action relating to their altered binding sites on HSP70 (Budina-Kolomets et al., 2014; Arakawa et al., 2011).

Pharmacophore based virtual screening of the ATPase inhibitors were carried out to identify new leads with high potency. The study generated pharmacophore models for ATP ase inhibitors present in the ATP binding domain of HSP70 through common feature

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pharmacophore generation process by utilizing a set of experimentally identified ATPase inhibitors. In addition the aim was attained by validation and confirmation by molecular docking of the leads with the protein structure.

2. Materials and methods

2.1. Data set

26 compounds were selected for the generation of pharmacophore models based on the ATPase inhibitory activity of HSP70 from the reported literatures (Koren et al., 2012; Li et al., 2013; Mamelak and Lingwood, 2001; Plowman, 1987; Jinwal et al., 2009; Rousaki et al., 2011; Wang et al., 2010; Kang et al., 2014; Tutar, 2017; Ozgur and Tutar, 2014). The compound selection in training set were deeply considered based on the already defined properties. The respective 2D structure of the compounds with their different magnitude of ATP dissociation constant used in the training set were represented in Fig. 1. The dataset was limited to 26 due to the lack of reported compounds on HSP70 inhibitory activity. Experiments with different training sets showed that the five training set compounds 15DSG, Apoptozole, MAL3-101, Myrecitin, NSC 630668-R1 were most suitable for ligand-based pharmacophore generation.

2.2. Pharmacophore model generation

The ligands were selected based on the two properties already defined, Principal and MaxOmitFeat. Common feature pharmacophore generation uses these values to determine which molecules should be considered when building the pharmacophore space and which molecules should map to all or some of the features in the final pharmacophores.

The Principal property is dependent on the activity level of the molecule; 2 for active in which a reference molecule ensures that all of the chemical features in the molecule are considered in building pharmacophore space; 1 for moderately active in which Conformations of this molecule are considered and 0 for inactive in which the molecule is not considered when placing pharmacophore features and is considered as an inactive molecule when optionally attempting to place excluded volumes.

The MaxOmitFeat property indicates the number of features required to miss each molecule and it is based on the value. Hence in our study the selected five ligands based on the *in vitro* HSP ATP ase activities were considered equally active and as reference ligands. The Common Feature Pharmacophore Mapping module of DS was performed to identify the intrinsic chemical features of the compounds in the training set. 5 features including HBA, HBD, Hydro, ring aromatic (Ring Atom) features, Positive ionization were selected in the generation of a quantitative pharmacophore model (Martin et al., 1993; Clark, 1997). Input parameters change of Conformation Generation was set to BEST; Maximum Conformations to 200; Energy Threshold to 10. Upto 200 conformations for each ligand were prepared and the conformations with 10 kcal mol-1 energy range were subjected for further analysis (Micheli et al., 2006; John et al., 2011) .Based on the fit value and heat map, the two best hypogen were selected from the generated ten hypotheses.

2.3. Validation

20 compounds of test set was employed to validate and to select the best pharmacophore model by both Test set and Decoy set Method using the Ligand Pharmacophore Mapping protocol. Validation was done automatically using the parameters under the Validation group.

2.3.1. Test-set method

10 compounds showing experimental ATPase activity were selected from the literature (Fig. 2) .Test set method was done to determine the predicting efficiency of the right class of compounds with their activity scale from the other training set of compounds by the pharmacophore models generated. Each pharmacophore was compared against a set of actives and inactives along with the ROC curves. Ligand Profiler maps a set of ligands against a set of pharmacophores and quickly determine the matches. The other paramaeters were kept as default except by changing Scale Fit Values to False and Save Aligned Ligands to True in the advanced parameter (Gupta et al., 2012).

2.3.2. Decoy-set method

1000 compounds comprising 10 active compounds were collected from the literatures already reported (Fig. 3). Database screening was carried out in DS using the Ligand Pharmacophore Mapping protocol. The parameters like total hits (Ht), enrichment factor (E), % yield of actives, % ratio of actives, false positives, false negatives and goodness of hit score (GH) and the Enrichment factor (EF) was calculated.

$$\frac{EF = \mathrm{TA/n}}{\mathrm{A/D}}$$

TA represents the number of active compounds screened; n represents the total number of active and decoy molecules screened; A is the total number of active compounds retained in the database; D represents the number of compounds in the validation database.

2.4. Virtual screening

The best Hypo gen model that fitted the “Maximum omitted Features” Option to zero was used as a query for screening potent molecules from the in built MiniMaybridge (2000 compounds) chemical database, Drug Diverse (5384), SCPDB compounds (5465) and Phytochemicals (2500). Hit compounds with the highest fit values were further subjected to drug-likeness properties using Lipinski rule of 5 and ADMET (Absorption, Distribution, Metabolism, Excretion and Toxicity).

2.5. Lipinski rule and ADMET

Based on the “Lipinski’s Rule of Five, drug likeness of the compounds was determined. ADME and Toxicity studies was performed by considering the parameters such as Atom based Log P98 (A LogP 98), polar surface area (PSA), Blood Brain Barrier (BBB), Cytochrome P450, Hepatotoxicity.

2.6. Molecular docking

The protein structure of HSP70 was retrieved from PDB (ID: 1S3X). The ligand and crystallographic water molecules were removed and loop refinement was carried out to screen the violations and determination of disallowed aminoacids in the protein. Ligand molecules were prepared and energy was minimized using CHARMm force field in DS. Active site was predicted followed by the binding site definition. The similarity analysis was performed for the best hit compounds using Pubchem search.

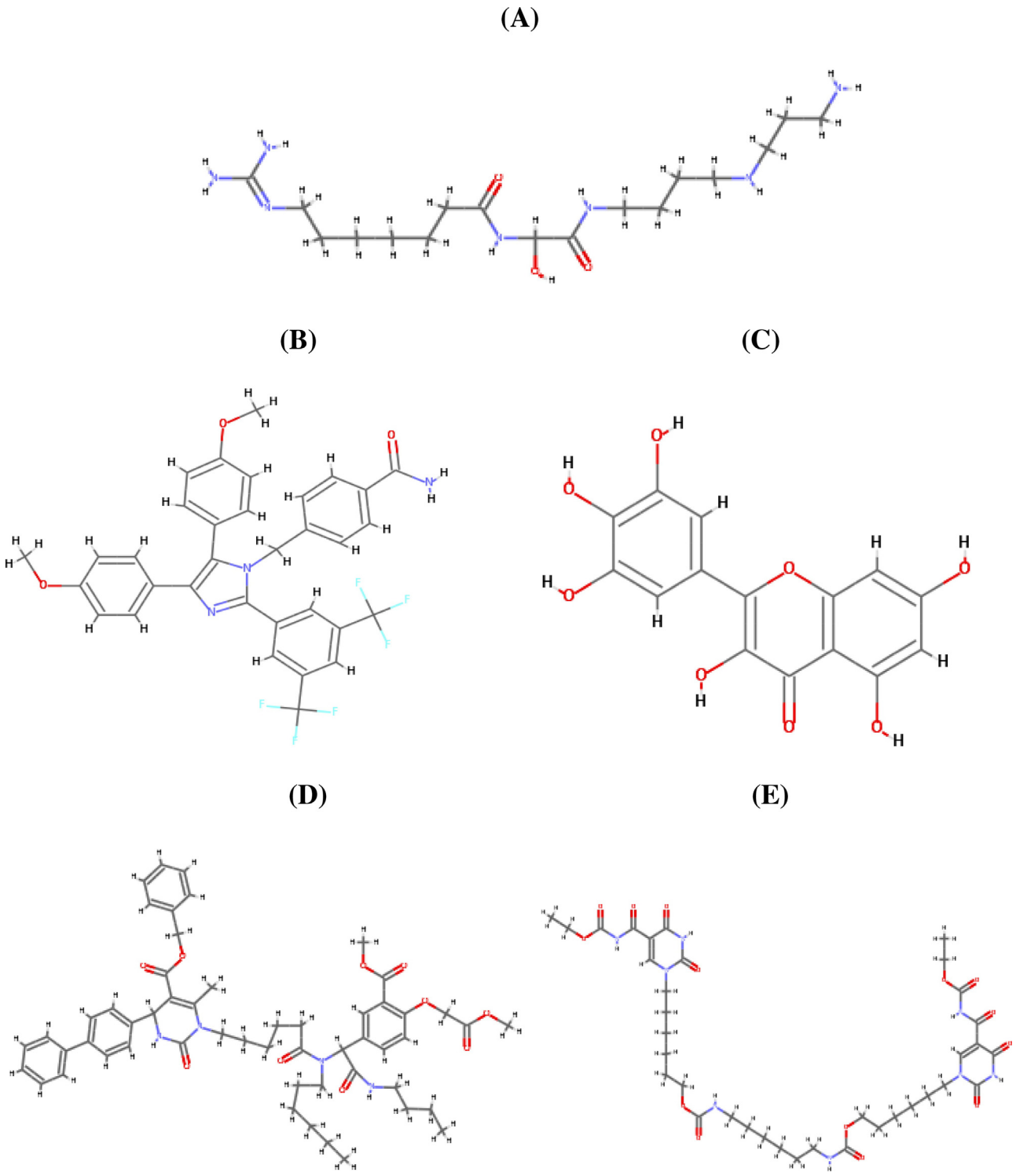

Fig. 1. Training Set of Compounds A) 15 DSG (K_D: 4 μM); B) Apoptozole(K_D: 0.4 μM); C) Myrecitin (K_D: 12 μM) D). MAL3-101 (K_D: 300 μM) (E) NSC 630668-R1 (K_D: 0.12 μM).

3. Result and discussion

3.1. Pharmacophore model generation

In this study, top ten hypotheses were generated based on the training set molecules using common feature Pharmacophore generation in DS. Hypogen 4 showed the combination of 2 chemical features like hydrogen bond acceptor (HBA) and hydrogen bond donor (HBD) whereas hypogen 8 shared only one common chemical feature like hydrogen bond acceptor with the highest fit value (Figs. 4–6). Among the 10 generated hypotheses, hypo 4 and 8 were chosen as a best qualitative Pharmacophore model on the basis of heat map, highest fit valueand chemical feature similarities (Table 1).

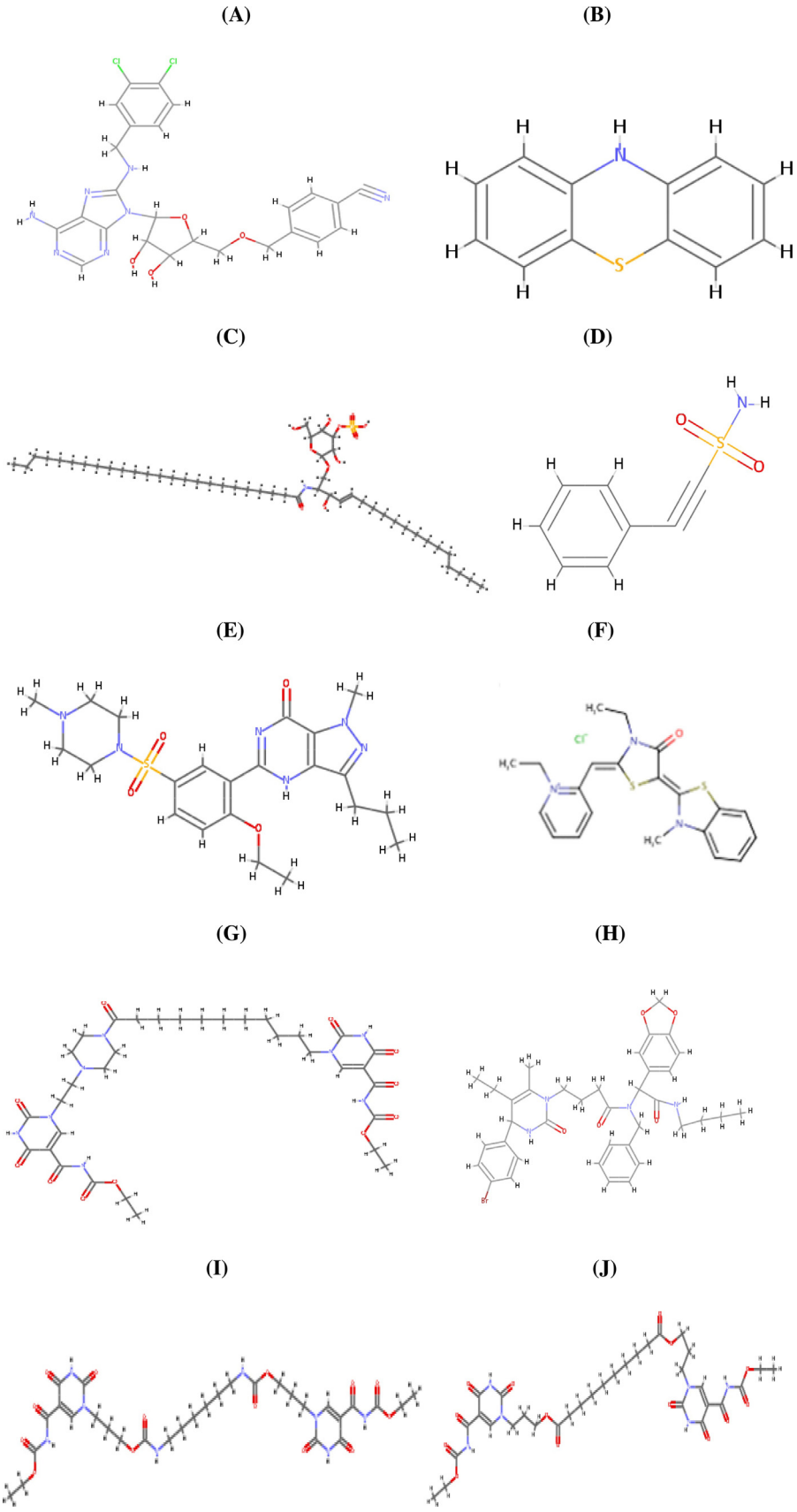

Fig. 2. Active Compounds from test set A)VER 155008 B) NSC625194C) 3SGC D) NSC625195 E) NSC625513 F) NSC625512 G) MKT–077 H)MAL3-90 I) Phenothiazine J) Sidenafil used for Pharmacophore validation.

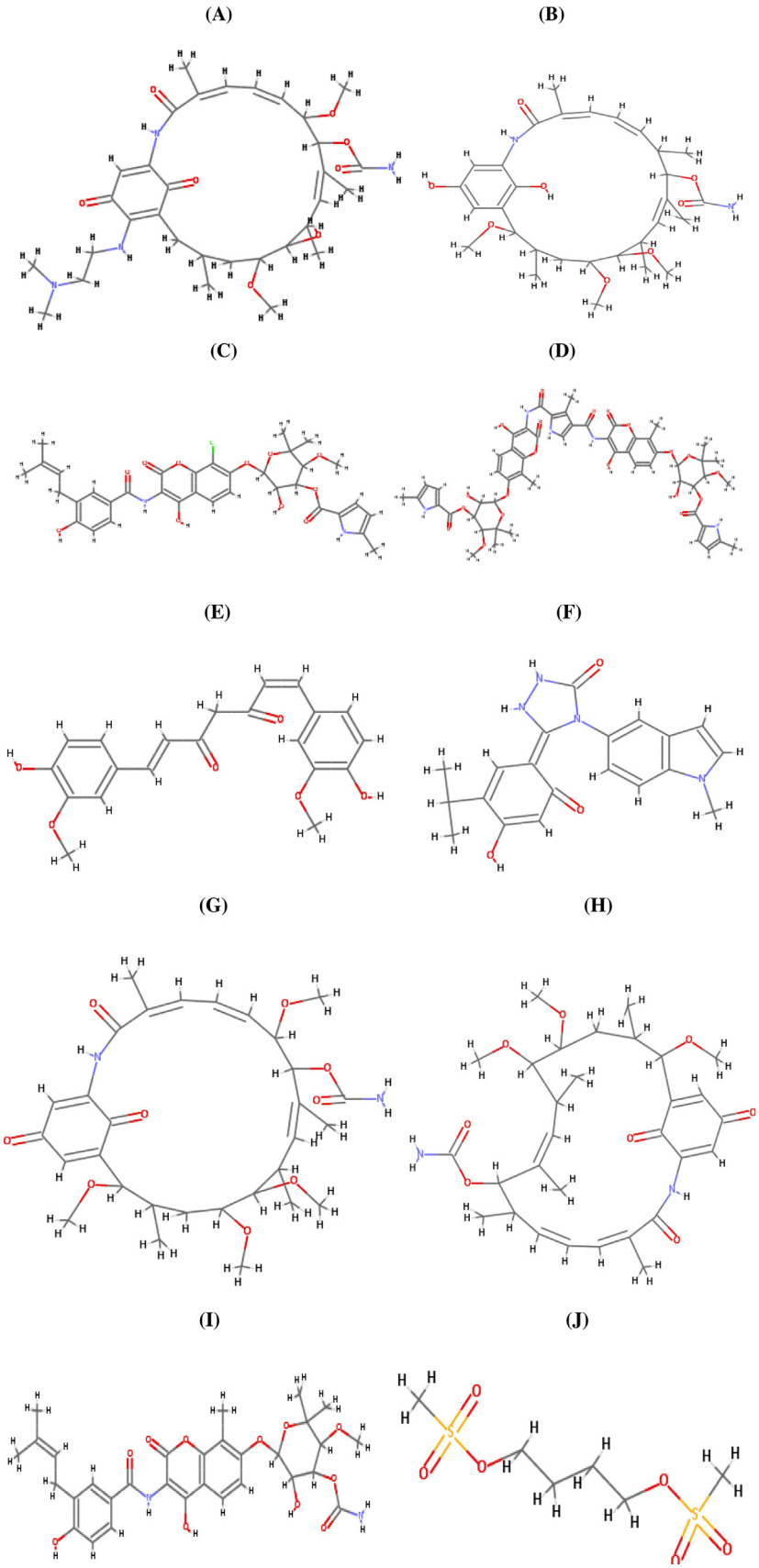

Fig. 3. Test Set of Compounds A) Alvepimycin B) Mabecin II C) Chlrobiocin D) Coumermycin E) Curcumin F) Ganetespib G) Nerbimycin A H) Mabecin I I) Busulfan J) Novobiocin.

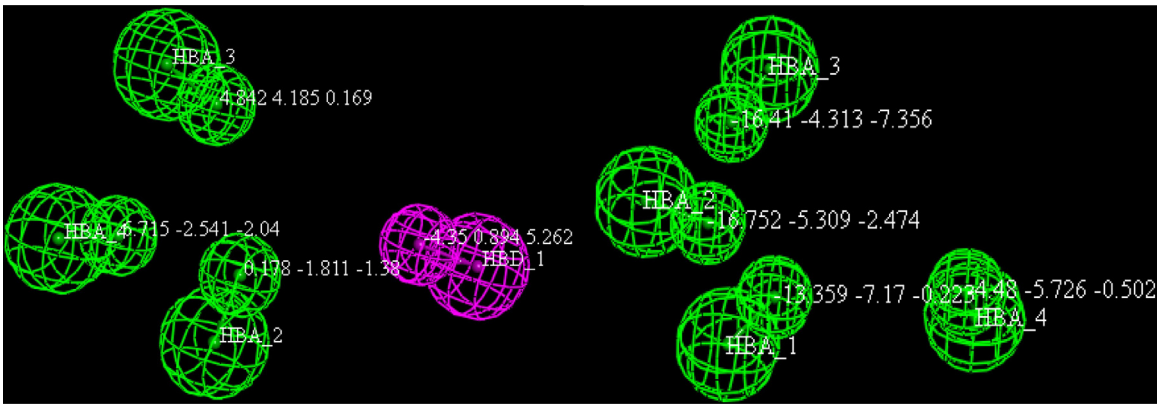

Fig. 4. Pharmacophoric features Identified from Best Hypotheses 4 and Hypotheses 8; Green represents the Hydrogen Bond Donor and Magenta represents the Hydrogen bond acceptor pharmacophoric features of generated models with its coordinates. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

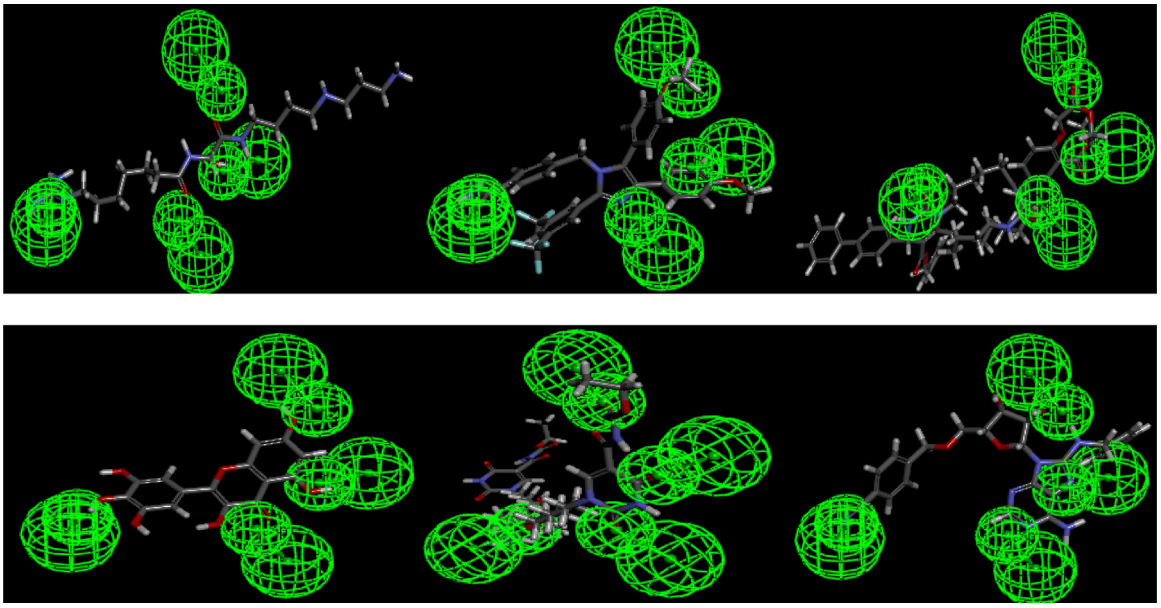

Fig. 5. Training set of Compounds (A) 15 DSG (B) Apoptozole (C) MAL3-101 (D) Myrecitin (E) NSC 630668-R1 (F) VER 155008 showing alignment with Hypogen 8. Green represents the Hydrogen Bond Donor and Magenta represents the Hydrogen bond acceptor pharmacophoric features of generated models with its coordinates. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

3.2. Validation of hypotheses

The best hypothesis were validated using 2 different methods in DS with the test set of about 10 active compounds containing structurally diverse properties with active, moderately active and low activity. Decoy set consisted of 10 known inhibitors and 990 decoy molecules of HSP70 inhibitors. The discriminating efficiency of the actives from inactives by the best hypotheses were used for validation. Hypo 8 and 4 showed the EF of 5 and 3.5 indicating the quality of pharmacophore as acceptable models. Hypo 8 and 4 identified 100% and 70% of active compounds respectively from decoy set. Results of hit list (H_t) number of active present ratio of activities in the hit list (%A) enrichment factor (E) and goodness of hit score (GH) are presented in Table 2. Based on the efficient discrimination of active from in activities, the Hypotheses 8 represented superior Pharmacophore.

3.3. Virtual screening, ADMET and molecular docking studies

Hypo 8 was used as 3D structural query to screen 15,349 compounds for the identification of leads respectively. Hypogen 8 was both highly qualitative and superior than hypogen 4 although hypogen 4 had more extendable pharmacophoric features. Pharmacophore model 8 signified the presence and role of Hydrogen bond acceptor as an essential characteristic feature for the development of ATPase inhibitor of HSP70 and showed best alignment with the test set of compounds. Hypo 8 retrieved 200 compounds and was further narrowed down by applying the maximum fit value to more than 2. Totally 18 final leads were selected with the highest fit value that qualified the Lipinski's rule of 5. The final leads with characteristic ADME and drug likeness properties are depicted in Table 3.

Furthermore Molecular docking was used as a confirmatory tool for the virtually selected compounds.Totally 18 leads showed best

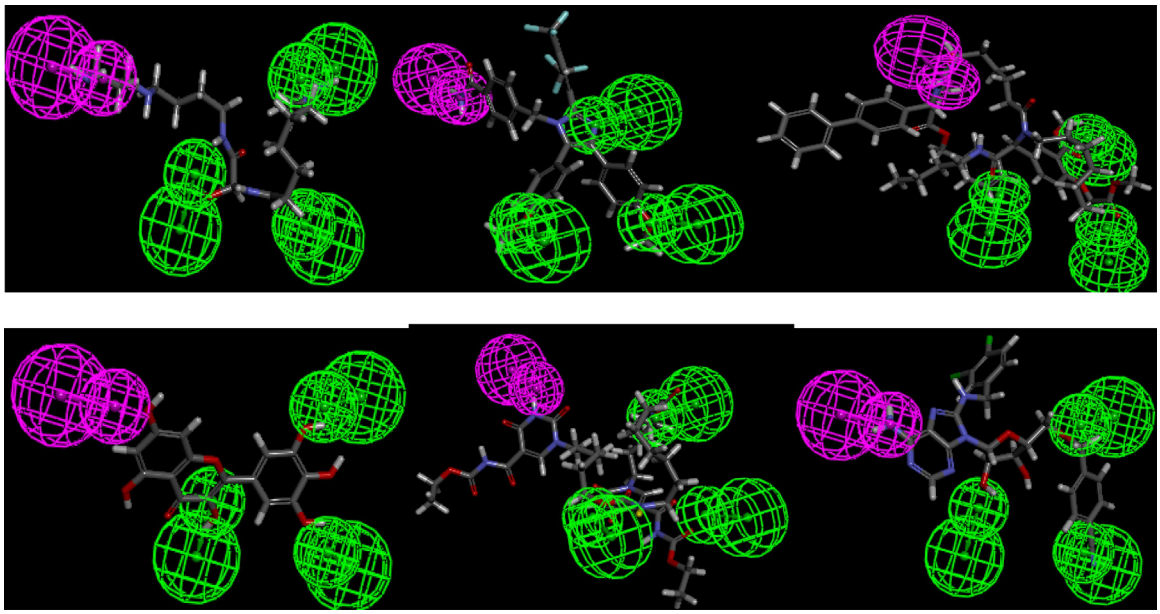

Fig. 6. Training set of Compounds (A) 15 DSG (B) Apoptozole (C) MAL3-101 (D) Myrecitin (E) NSC 630668-R1 (F) VER 155008 showing alignment with Hypogen 4. Green represents the Hydrogen Bond Donor and Magenta represents the Hydrogen bond acceptor pharmacophoric features of generated models with its coordinates. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 1
Fit value of the training set of compounds generated from best Hypogen 8.

S.no	Name	Fit value
1	15 DSG	2.13408
2	APOPTOZOLE	1.59723
3	MAL3-101	2.48344
4	MYRECITIN	2.45847
5	NSC 630668-R1	3.9994

interaction and high binding energy with HSP 70. Among the Compounds screened from Minimaybridge RJC 03785 & RDR 02049 showed the highest fit values of 2.6272 & 2.01395 which was equivalent to that of active training set compounds Mal3-101 and Myrecitin but lesser than for NSC 630668-R1. However the comparative docking scores of the compounds RJC 03785 & RDR 02049 was not that appreciable to the other final leads.

From the SCPDB, compound with Id 3116_JZX showed highest fit value of 3.46136 that was equivalent to the test compound NSC 630668-R1. Further validation of this compound against the target 1S3X ATPase showed higher binding energy of −80.99 and promising interaction at 4 sites Arg 76, Ala 223, Asp 206 with the distances less than 2.5. The similarity search revealed the compound as 5-(6-{[4-(methylsulfonyl)piperazin-1-yl]methyl}- 4-morpholin-4-ylthieno[3,2-*d*]pyrimidin-2- yl)pyridin-2-amine, a known potent mTOR Inhibitor (Sutherlin et al., 2010). Likewise the

Table 2
Statistical parameters of the Generated Hypogen from screened Decoy set.

Characters	Hypo 8	Hypo 4
Total number of molecules in database (D)	1000	1000
Total number of actives in Database(A)	10	10
Total number of hit molecules from the database(Ht)	19	25
Total number of active in hit list (Ha)	10	7
% of yield of actives(Ha/Ht*100)	52%	28%
% Ration of actives (Ha/A*100)	100%	70%
Enrichment Factor	5	3.5
False Negatives	0	3
False Positives	9	18

compound 2qbp_527 showed fit value of 3.26 and highest binding energy of −295.708 with hydrogen bond interaction observed at Arg 264, His 227, Glu 231. The compound search revealed it as 5-(3-{[1-(benzylsulfonyl)piperidin-4-yl]amino}phenyl)- 4-bromo-3-(carboxymethoxy) thiophene-2-carboxylic acid that has been reported to exhibit protein tyrosine phosphatase 1 B inhibitor. Similarly, the other screened compounds from SCPDB database showed good fit values and docking scores.

Likewise compounds screened from Drug Diverse Database showed highest fit values ranging from 2.8 to 3.12 and higher binding energies of −77.8086 to −56.556. Among those, ENA1218592 showed interaction with the target at the sites ASP10, Thr 204, Thr 13, Arg 72, Tyr 149, Lys 71. Screening of Phytochemicals resulted in the identification of muricatetrocin B, Diactylphiladelphilactone C, Eleutheroside B, Nicotanamine, Artelasticin as leads and scored the fit values from 2.4 to 2.6 which was equivalent to the test compounds in the training set. The interaction score of the other compounds are shown in Table 4. Although the above mentioned plant compounds has been reported for anticancer activity in various cancerous cell lines however their mode of action, in specific, against the HSP70 has not yet been reported. Thus this study reveals their common mode of interaction could be the inhibition of ATPase of HSP70 in anticancer activity. Also the docking studies reveals the presence of Hydroxy, 4β-hydroxy-2-en-1 one and the pyrimidine derivatives could better bind the Active site of ATP ase and enhance the anticancer activity. The screened 18 hit compounds were non hepatotoxic and expressed their value below 1 except the compounds Artelasticin, RJC03785, 3116_JZX and 3LQ8_88Z. Also the screened compounds were non inhibitor of CYP2D6 enzyme however Artelasticin was inhibitory and expressed the value to above 1.

Thus the Phytochemical compounds Muricatetrocin B, Diacetylphiladelphicalactone C, Eleutheroside B and 5-(3-{[1-(benzylsulfonyl)piperidin-4-yl]amino}phenyl)- 4-bromo-3-(carboxymethoxy)thiophene-2-carboxylic acid was identified as lead candidates against the target ATPase of HSP70 based on the

Table 3
ADMET and lipinski rule of 5 properties of the Virtually screened lead Molecules identified by the Best Hypogen Models.

S.NO	Name	FitValue	CYP2D6	AlogP98	HEPATOTOXIC	HBD	HBA
1	Muricatetrocin B	2.60864	−2.70459	4.221	−7.77625	4	7
2	Diacetylphiladel phicalactone C	2.59784	−8.31845	1.564	−4.68053	2	10
3	Eleutheroside B	2.57144	−8.77025	−0.512	−7.0903	5	9
4	Nicotianamine	2.5142	−3.01495	−10.268	−6.94511	6	9
5	Artelasticin	2.45693	1.50979	7.698	2.2568	4	6
6	RJC 03785	2.6272	−2.2116	3.967	1.93886	2	9
7	RDR 02049	1.71573	−9.46244	2.32	0.534099	4	8
8	HTS 02804	0.905687	−12.9185	−0.798	−6.30141	2	9
9	ENA793908	3.12998	−6.79566	1.578	−4.36543	1	10
10	ENA1218592	3.04051	−4.36353	0.86	0.534099	2	6
11	ENA195244	2.89353	−1.07295	2.64	−7.32336	2	6
12	UKR1120838	2.8756	−4.54239	3.386	−6.30141	0	8
13	UKR311976	2.80204	−3.21218	3.678	−1.94067	1	7
14	3l16_JZX	3.46136	−14.4895	−0.41	1.0028	3	10
15	2qbp_527	3.26389	−11.8198	1.041	−2.4965	1	9
16	2zva_1N1	3.24117	−9.74258	1.878	−1.94442	4	9
17	3lq8_88Z	3.19678	−6.89825	2.876	1.01786	3	10
18	3kry_3KR	3.17197	−9.84838	3.494	−0.597306	1	9

Table 4
Molecular Docking and Interaction profile of the lead compounds.

S.NO	Name	Cdocker energy	LibDockScore	Binding Energy	Hydrogen bond interaction	Distance Å
1.	Muricatetrocin B	−3.117	62.589	−146.579	ASN 151 N—H···O	2.64
					ARG 76 N—H···O	2.14
					GLN 154 N—H···O	2.58
					GLN 154 N—H···O	2.77
					ARG 76 N—H···O	1.97
					ARG 76 N—H···O	2.29
2.	Diacetylphiladelphicalactone C	−27.908	34.091	−138.923	ARG 264 C—H···O	2
					LYS 56 C—H···O	2.97
					LYS 56 C—H···O	1.84
					GLU 231 C—H···O	2.65
3.	Eleutheroside B	−13.472	28.294	−119.393	THR 14 C—H···O	2.02
					THR 13 C—H···O	2.87
					C—H···O TYR 15	2.68
					C—H···O TYR 15	2.8
					C—H···O ASP10	2.32
					C—H···O ASP10	2.56
					C—H···O ASP10	1.91
					C—H···O GLU175	2.31
					C—H···O GLU175	2.03
4.	Nicotianamine	1.771	46.621	−161.079	ARG 272 C—H···O	2.73
					TYR 15 N—H···O	2.02
					THR 14 C—H···O	1.94
					THR 14 C—H···O	2.68
					GLY 201 C—H···O	2.58
					C—H···O ASP 199	3.02
					C—H···O ASP 199	3.08
					N—H···O GLY 175	2.38
					N—H···O ASP 199	2.29
					N—H···O ASP 199	2.73
5.	Artelasticin	−38.129	63.772	−53.341	C—H···O THR 37	2.12
					THR 37 C—H···O	2.75
					ARG 36 C—H···O	2.77
6.	RJC 03785	−25.456	54.5	−37.1664	GLU 543 C—H···O	2.57
					GLY 407 C—H···O	2.66
7.	HTS 02804	22.571	38.204	−3.6805	ALA 406 C—H···O	2.65
					C—H···O GLU 404	2.42
					C—H···O GLU 543	2.8
					C—H···O GLU 404	2.61
					C—H···O GLU 404	2.17
					C—H···O LEU439	2.66
					C—H···O LEU439	2.78
8.	RDR 02049	22.854	36.578	−68.3264	LYS 526 C—H···O	2.43
					LYS 526 C—H···O	3.06
					C—H···O GLU 530	2.37

Table 4 (*Continued*)

S.NO	Name	Cdocker energy	LibDockScore	Binding Energy	Hydrogen bond interaction	Distance Å
					ARG 533 C—H···O	1.81
					C—H···O GLU 446	2.76
9.	RJC 03202	12.734	39.997	−69.4749	C—H···O GLU 530	2.1
					LYS 526 C—H···O	2.91
10.	ENA793908	−12.623	19.483	−56.556	THR 222 C—H···O	2.09
					C—H···O ASP 206	2.65
11.	ENA1218592	8.201	98.522	−123.626	C—H···O ASP 10	2.41
					THR 204 C—H···O	2.61
					C—H···O THR 13	3.08
					ARG 72 C—H···O	2.66
					TYR 149 C—H···O	2.55
						2.57
12.	ENA195244	40.543	95.804	−85.1856	C—H···O ASP 199	2.79
					C—H···O ASP 199	2.92
					GLY 338 C—H···O	2.44
					GLY 339 C—H···O	2.86
					ARG 272 C—H···O	2.99
					ARG 272 C—H···O	2.74
13.	UKR1120838	11.778	52.117	−59.5499	C—H···O TYR 41	2.4
					C—H···O GLU 268	2.85
					C—H···O GLU 268	2.7
					GLY 202 C—HvO	2.86
					LYS 271 C—H···O	2.86
					GLY 230 C—H···O	2.31
					THR 14 C—H···O	1.41
14.	UKR311976	17.365	33.437	−77.8086	C—H···O ASP 69	2.46
					C—H···O GLU 268	2.65
					C—H···O HIS 227	2.97
					ASN 235 N—H···O	2.8
					ASN 235 N—H···O	2.88
15.	3I16_JZX	−4.583	−46.149	−80.9971	ARG 76 C—H···O	2.11
					C—H···O TYR 149	3.25
					C—H···O ALA 223	2.1
					C—H···O ASP 206	2.33
					C—H···O ASP 206	2.51
16.	2qbp_527	29.087	44.417	−295.708	ARG 264 C—H···O	2.5
					ARG 264 C—H···O	2.44
					HIS227 C—H···O	2.81
					C—H···O GLU 231	1.33
					C—H···O GLU 231	2.62
17.	3lq8_88Z	−3.788	34.257	−79.9656	LYS 126 C—H···O	1.92
					N—H···O GLN 33	2.9
					GLN 33 C—H···O	2.22
					ASN 31 C—H···O	2.75
					C—H···O TYR 15	1.55
					C—H···O ASP53	2.39
					C—H···O ASP53	2.61
					C—H···O ASP53	3
					LYS 56 C—H···O	2.03
					C—H···O ASN 57	2.55
					C—H···O ASN 57	2.74
					C—H···O ASN 57	2.94
					C—H···O ASN 57	3
18.	3kry_3KR	7.528	5.712	−182.135	ASN 235 C—H···O	2.01
					C—H···O GLU 231	2.22
					C—H···O ASP 69	2.48
					C—H···O ASP 86	2.37
					C—H···O ASP 86	2.84
19.	Apoptozole	−6.622	4.633	−31.3553	N—H···O ASP 86	2.9
					C—H···O HIS 89	2.94
					ARG 264 C—H···N	2.71
					C—H···O THR 13	2.19
					C—H···O ASP69	2.34
					LYS 56 C—H···O	2.9
					C—H···O GLU268	2.47

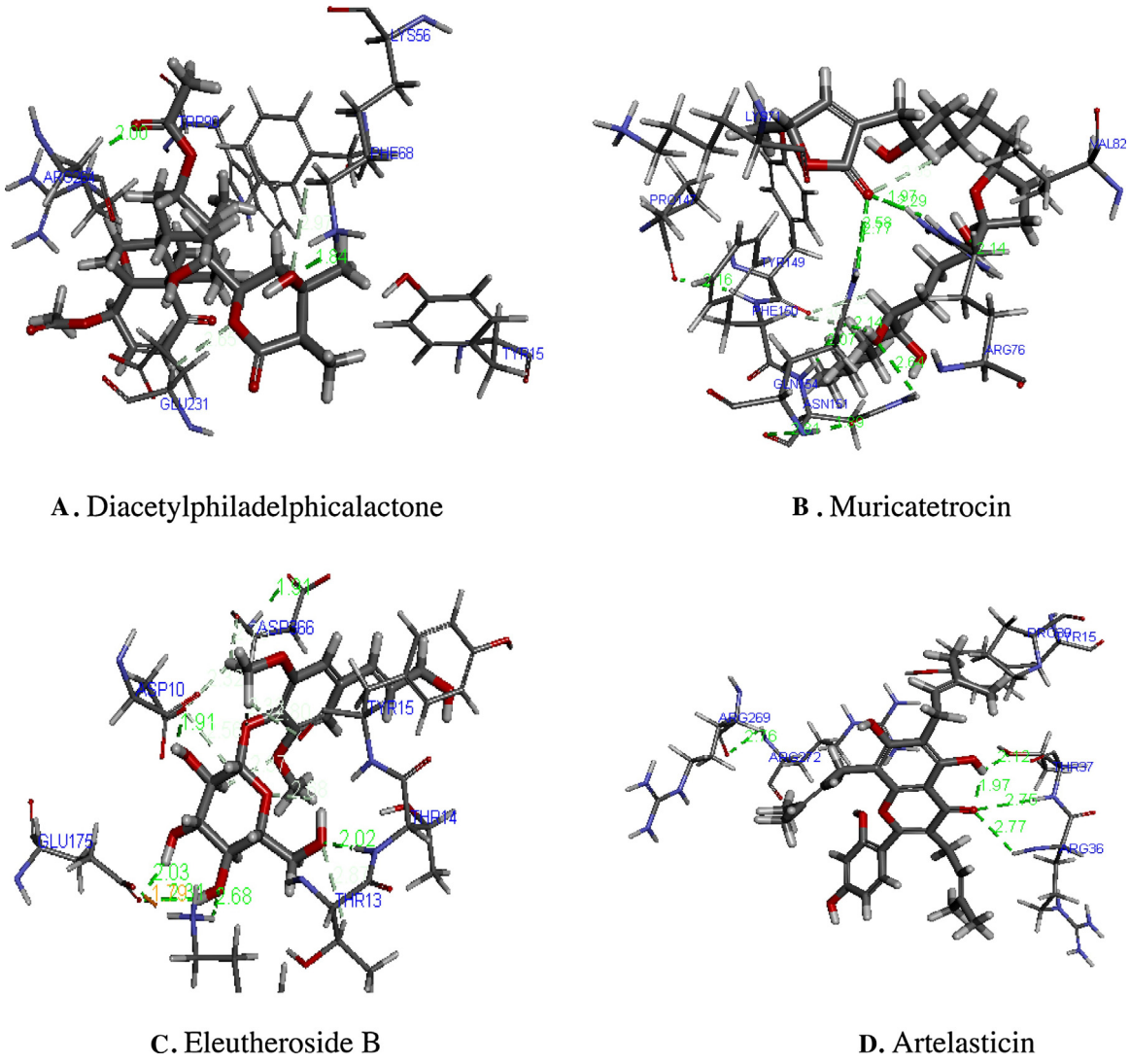

Fig. 7. Molecular Interaction of lead phytochemicals with ATPase Domain of HSP70. Green colour represents the hydrogen bond interaction between the target and ligand molecule. Active site residues involved in the interation are represented by Blue colour. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

higher fit values, binding energies, molecular interactions and desirable ADMET Properties (Fig. 7).

4. Conclusion

A computational approach was employed to identify new leads for ATPase inhibitors of HSP70. The best Pharmacophore consited of four hydrogen bond acceptor features and showed high fit value. Three phytochemical compounds and one from SCPDB were identified as two active compounds from the virtual screening which satisfied the Lipnski rule of 5 and ADMET properties and also showed strong hydrogen bond interaction with HSP70. Thus the screened compounds can be proposed as lead candidates and also the study warrants further *in vitro* and *in vivo* evaluation.

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