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CLINICAL ANALYSIS, DRUG DESIGNING AND QSAR STUDIES ON RHEUMATOID ARTHRITIS

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ABSTRACT

The clinical data related to Rheumatoid Arthritis has shown that there is more number of males (60%) than females (40%) during January month at KGH, Visakhapatnam, India. The patients also have more number of Neutrophils (72%) and Lymphocytes (18%) and less number of Eosinophils (10%) and Monocytes (0%) suffering from Rheumatoid Arthritis. Based on docking results Leflunomide and **Desoxycorticosterone Pivalate** shown good results based on distance approach (43.5) against RA. Based on Emin values, **Auranofin** (-66.73) has shown good activity compared with other selected drugs. Based on QSAR analysis **Cyclosporine** and **Phenacetin** are related and also shown similar results with docking.

KEY WORDS

Rheumatoid Arthritis, QSAR.

INTRODUCTION

Rheumatology is a sub-specialty in internal medicine and pediatrics, devoted to the diagnosis and therapy of rheumatic diseases. In rheumatoid arthritis, the small joints of the hands, wrists, feet, and knees are typically inflamed in a symmetrical distribution (affecting both sides of the body). Studies involving various types of the connective tissue collagen are in progress and show encouraging signs of reducing rheumatoid disease activity. Finally, genetic research and engineering is likely to bring forth many new avenues for earlier diagnosis and accurate treatment in the near future [1, 2.]

Through the technique of in vitro molecular evolution, many kinds of peptide aptamers that bind to a target protein can be generated [3, 4, 5]. In most cases, binding sites of these peptides on the protein surface remain unknown. It takes much time and experimental costs to obtain such information including the binding mode (conformation and orientation of a peptide) by biophysical or biochemical experiments. Therefore, predicting the binding site and binding mode through theoretical calculation within a reasonable computational time is desirable.

Prediction of a protein–ligand complex structure is called the “molecular docking” problem [6]. In particular, docking without prior knowledge of the binding site is called the “blind docking” [7, 8]. There have been a lot of studies related to the docking problem with prior knowledge of the binding site [9, 10], and a number of programs have been developed such as DOCK [11] and AutoDock [12]. Warren et

al. examined the performance of many docking programs for small ligand molecules [6], however, few studies on the blind docking of flexible ligand molecules such as peptides [7].

Hetenyi and van der Spoel in 2002, [7] systematically tested the ability of the AutoDock program to perform the blind docking of peptides to proteins, and concluded that the program is able to select the correct complexes based on energy without prior knowledge of the binding site. The AutoDock program requires a lot of computational costs due to calculation at the atomic level.

In our present study, we have investigated on drugs, which are better acted against autoantigen of Rheumatoid Arthritis (RA). QSAR Properties of these drugs were also investigated in this study.

MATERIALS AND METHODS

In this work, we attempted to carry out the Analysis of protein and drug designing with the following infrastructure.

1. ***SYSTEM USED*** –Intel Pentium 4 GHz, 2GB RAM
2. ***OPERATING PLATFORM***- Microsoft Windows XP pro 2002 service pack
3. **PACKAGES** – Hyperchem v8.0, HEX 5.1, GEMDock Version 1.0
4. ***PROTEIN***- 1NWR.PDB.
5. ***Ligand structures from DrugBank***

Data related to Rheumatoid Arthritis (RA) is collected from patient reports from KGH, visited during January 2010.

QSAR Properties is conducted on Hyperchem package v8.0, which allows calculation and estimation of a variety of molecular descriptors commonly used in Quantitative Structure-Activity Relationship (QSAR) studies. Most of the methods were developed for and are primarily applicable to organic molecules. Here are some of the properties you can estimate using QSAR Properties:

- Atomic charges
- Van der Waals and solvent-accessible surface areas.
- Molecular volumes, bounded by Van der Waals or solvent-accessible surfaces.
- Hydration energy (for peptides and similar systems.
- Log P (the log of the octanol-water partition coefficient), a hydrophobicity indicator.
- Refractivity.
- Polarizability.
- Mass.

Hex is an interactive protein docking and molecular superposition program, written by Dave Ritchie. *Hex* understands protein and DNA structures in PDB format. Version 5.1 can also read small-molecule SDF files. Hex is an interactive molecular graphics program for calculating and displaying feasible docking modes of pairs of protein and DNA molecules. Hex can also calculate protein-ligand docking, assuming the ligand is rigid, and it can superpose pairs of molecules using only knowledge of their 3D shapes. The graphical nature of Hex came about largely because to visualise the results of such docking calculations in a natural and seamless way, without having to export unmanageably many (and usually quite big) coordinate files to one of the many existing molecular graphics programs. For this reason, the graphical capabilities in Hex are generally relatively primitive compared to professional molecular graphics packages.

Generic Evolutionary Method for Molecular Docking (GEMDOCK) is a program for computing a ligand confirmation and orientation relative to the active site of target Protein.

RESULTS

Based on the results shown in Fig. 1and 2, Table 1 and 2, following are the observations:

- The clinical data has shown that more number of males (60%) than females (40%).

- The patients also have more concentrations of Neutrophils (72%) and Lymphocytes (18%) and less number of Eosinophils (10%) and Monocytes (0%) in Rheumatoid patients.
- Other data is predicted to be normal (Lipid and serum profiles).
- Based on docking results Leflunomide and **Desoxycorticosterone Pivalate** shown good results against HCgp-39 (acts as an autoantigen in rheumatoid arthritis) based on distance approach (43.5).
- Based on Emin **Auranofin** (-66.73) has shown good activity.
- Based on QSAR analysis **Cyclosporine** and **Phenacetin** are related and also shown similar results with docking using hex. All the other ligand parameters are unrelated.

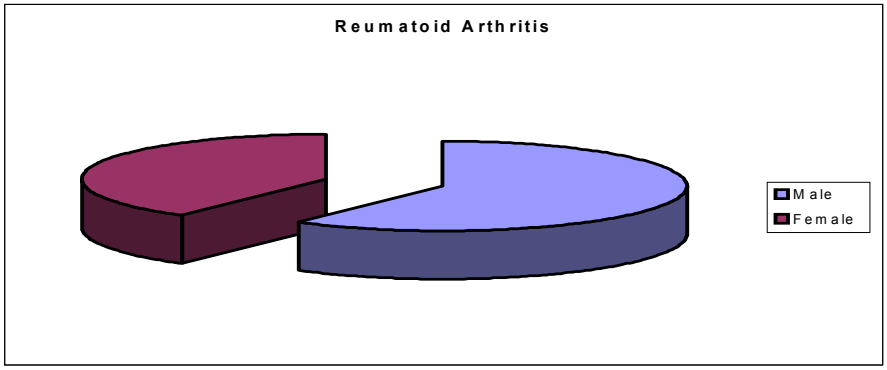

Figure 1
Male female ratio with RA

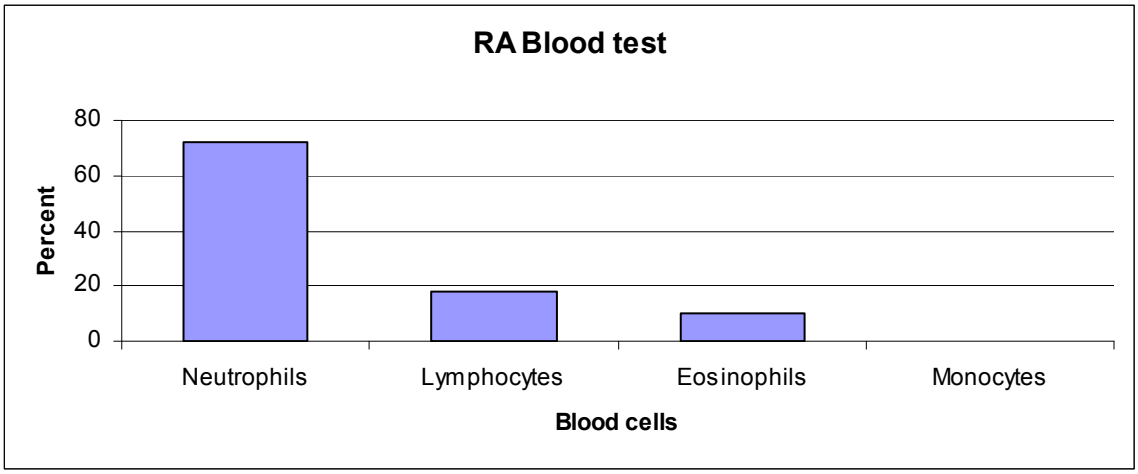

Figure 2
Graphical Representation of Blood test with RA

Table 1

Docking studies of drugs against HCgp-39 protein

LIGAND NAME	Hex Results				GEMDOCK Results			
	Emin	Emax	Etotal	R	Fitness	VDW	H Bond	Elec Bond
Mefenamic acid	-65.4	-24.0	-26.4	46.9	-61.2	-54.7	-6.5593	20.8889
Meclofenamic acid	42.2	-19.1	-34.2	48.8	-61.2	191.9	-52.5941	-14.8757
Naproxen	-42.2	-19.1	-33.0	48.8	-71.2	-52.7	-18.5042	21.9412
Ibuprofen	-47.9	-21.0	-30.4	49.9	-54.5	-47.5	-7	22.6
Tenoxicam	-26.4	-26.4	-27.1	51.8	-78.9	-48.8	-30.1314	18.8182
L-Cystine	-30.4	-10.9	-37.8	51.8	-72.4	-30.7	-38.36	3.28697
Phenyl butazone	-52.2	-20.4	-31.8	48.8	-75.1	-55.0	-20.144	18.2174
Cyclosporine	-44.6	-32.4	-44.6	51.8	-88.7	-65.4	-23.281	8.3647
Colchicine	-27.7	-18.5	-27.7	48.9	-82.9	-76.2	-6.64416	18.931
Temsirolimus	-55.1	-19.4	-70.8	43.9	251.5	-67.4	-33.0293	3.28697
Phenacetin	-48.7	-20.2	-33.9	49.9	-68.0	-48.3	-19.7614	23.1538
Hydrocortisone	-48.7	-20.2	-33.9	49.9	-81.0	-51.5	-29.5462	15.423
Niflumic acid	-54.3	-25.4	-25.4	48.8	-63.9	-57.9	-5.93301	18.75
Diclofenac	-55.1	-19.4	-21.9	49.5	-58.1	-36.1	19.9712	2.00317
L-Histidine	-31.2	-16.9	-31.2	54.8	-65.2	-37.1	-25.6198	-2.4269
Salicylate-magnesium	-56.2	-25.2	0	63.9	-88.9	-46.1	-38.523	4.2862
Ketoprofen	-27.7	-24.0	-27.6	49.9	-72.9	-55.5	-16.735	-0.572204
Fenoprofen	-43.5	-19.9	-27.4	48.8	-65.2	-35.1	-27.1254	-3.1088
Auranofin	-66.7	-17.0	-81.2	44.6	5595	-37.2	-35.6636	14.875
Indomethacin	-56.7	-27.2	-29.1	51.8	-80.0	-54.6	-25.365	17.52
Sulindac	-57.1	-23.1	-37.0	43.9	-77.9	-61.5	-15.8774	0.572254
Aspirin	-41.7	-22.2	-41.7	63.9	-65.2	-37.1	-25.6198	-42.69
N-Acetyl-D-glucosamine	-41.7	-22.2	-22.1	57.8	-81.8	-42.3	-39.5129	25.5333
Piroxicam	-25.9	-12.5	-31.4	46.9	-79.4	-48.4	-30.9738	20
Penicillamine	-54.9	-24.1	-54.9	63.9	-54.4	-21.2	-28.044	3.06145
Diflunisal	-48.2	-26.8	-32.2	55.6	-71.8	-49.8	-18.9306	-3.04822
Sulfinpyrazone	-46.9	-24.6	46.96	45.4	-76.7	-67.6	-9.11418	16.3667
Sulfasalazine	-35.5	13.1	-40.2	49.9	-80.7	-72.0	-8.75337	16
Cortisone acetate	-28.5	-25.3	-28.5	51.8	-86.0	-60.8	-22.1051	3.06147
Hydroxychloroquine	-27.3	-14.0	-27.3	49.9	-63.4	-45.9	-17.5061	0
Celecoxib	-23.0	-7.19	-43.8	48.8	-78.3	-51.0	-27.2574	14.9231
Desoxycorticosterone Pivalate	-45.8	-20.2	-40.1	43.5	-76.1	-64.6	-11.5287	16
Azathioprine	-42.2	-21.3	-25.6	49.9	-65.8	-42.8	-3.06145	18.6316
Etodolac	-60.0	-27.9	0	63.9	-76.1	-51.0	-17.5061	3.06147
Methotrexate	-65.1	-30.7	-31.8	49.9	-67.2	-51.0	-22.1051	14.9231
Oxaprozin	-50.9	-21.2	-36.8	45.8	-77.8	-60.6	-17.2394	18.2273
Levamisole	-16.5	-44.1	-44.1	43.9	-58.1	-37.8	-17.3002	3.06145
Nabumetone	-29.4	-10.8	-45.1	48.8	-71.0	-53.7	-17.2935	20.0417
Etoricoxib	-45.1	-10.8	-5.17	48.8	-81.5	-61.9	-19.6566	20.3158
Tolmetin	-50.1	-24.4	-34.5	49.9	-71.2	-66.8	-48.6959	-15.1091
Flurbiprofen	-47.7	-24.4	-44.4	43.9	-56.5	-8.54	0	15.7273
Chloroquine	-50.9	-21.2	-36.8	45.8	-77.8	-60.6	-17.2394	18.2273
Glucosamine	-48.4	-17.6	-17.9	49.9	-69.0	-33.8	-35.2016	23.669
Menthol	-37.6	-16.4	-30.6	48.8	-44.5	-38.5	23.36	0
Meloxicam	-49.9	-23	-25.6	48.8	-96.8	-61.4	-32.3124	21.04
Leflunomide	-46.1	-23.4	-44.7	43.5	140.2	-53.4	-22.0948	19.1053

Probenecid	-37.2	-15.9	-22.0	45.4	-73.8	-48.6	-25.76	19.57
Methylprednisolone	-51.0	-20.8	0	63.9	-78.9	-50.6	-28.287	19.3704
Vitamin E	-51.0	-19.4	-54.9	45.8	-75.3	-72.2	0	3.061

Table 2

QSAR studies of drugs using against HCgp-39 protein

LIGAND NAME	QSAR						
	Surface Area	**Volume**	**Hydration energy**	**LogP**	**Refractivity**	**Polarisability**	**Mass**
Mefenamic acid	513.48	939.41	-8.66	5.33	24.78	27.67	277.3
Meclofenamic acid	376.48	753.6	-8.36	6.27	29.12	29.69	308.16
Naproxen	390.09	697.67	-9.97	4.39	26.18	25.32	230.2
Ibuprofen	426.49	698.18	-5.87	5.4	36.33	24	206.28
Tenoxicam	365.76	799.67	13.01	6.75	38.4	29.84	337.57
L-Cystine	354.67	636.14	-23.52	2.93	48.7	21.93	240.29
Phenyl butazone	460.9	554.87	930.81	-3.23	38.94	35.94	308.38
Cyclosporine	384.45	746.94	460.98	4.82	46.52	39.29	356.78
Colchicine	563.35	1106.16	6.88	5.42	58.68	41.74	399.44
Temsirolimus	1098.71	22.83	-13.83	19.85	132.72	69.48	944.63
Phenacetin	383.86	603.09	-5.66	3.44	26.78	19.85	179.22
Hydrocortisone	44137	984.48	-9.41	5.57	38.1	38.1	362.47
Niflumic acid	365.17	706.62	-11.25	4.62	20.15	24.86	282.22
Diclofenac	382.87	762.17	10.78	6.01	29.42	29.69	296.15
L-Histidine	252.92	477.39	-16.79	0.73	21.67	15.8	155.16
Salicylate-magnesium	341.29	692.81	-13.31	6.33	17.22	20.85	288.85
Ketoprofen	393.96	372.32	-11.02	5.48	23.17	26.96	242.27
Fenoprofen	393.96	372.32	-11.02	5.48	23.17	26.96	242.27
Auranofin	-4552490	1573.97	-1196002	-0.53	101.84	45.86	679.49
Indomethacin	499.16	946.95	-11.68	7.6	38.62	36.7	357.79
Sulindac	505.6	956.52	-10.46	7.03	37.91	34.82	356.41
Aspirin	307.73	521.08	-10.74	4.96	17.26	17.39	180.16
N-Acetyl-D-glucosamine	342.38	620.62	-21.5	0.09	44.5	19.3	221.21
Piroxicam	403.86	835.89	-12.98	7.29	32.45	30.32	331.38
Penicillamine	290.17	455.77	-11.1	1.74	34.3	15.02	149.21
Diflunisal	339.89	640.14	-13.72	5.07	13.46	23.11	250.2
Sulfinpyrazone	709.89	1330.25	-8.78	5.45	102.98	49.53	416.64
Sulfasalazine	519.66	1101.46	-8.26	9.2	44.24	41.6	404.48
Cortisone acetate	496.64	1061.96	-5.19	9.6	85.7	41.31	402.49
Hydroxychloroquine	585.59	1029.25	-7.6	2.82	61.78	37.69	335.88
Celecoxib	514.09	959.86	-10.83	7.86	37.41	32.8	381.37
Desoxycorticosterone Pivalate	621.15	1209.48	8.677	11.18	100.06	46.09	414.59
Azathioprine	317.15	682.98	-13.07	0.81	29.08	25.74	277.26
Etodolac	391.56	843.34	-7.39	4.6	48.41	31.59	287.36
Methotrexate	533.87	684.34	-31.11	5.4	52.87	45.06	454.45
Oxaprozin	421.82	864.45	12.86	5.36	24.51	32.43	293.32
Levamisole	298.22	616.63	-2.5	1.99	32.7	23.35	204.99
Nabumetone	511.14	963.19	-6.01	6.89	34.38	35.07	358.84
Etoricoxib	430.82	768.26	-9.36	6.53	30.08	27.95	257.29
Tolmetin	381.56	716.72	-8.93	5.55	21.46	26.23	244.27
Flurbiprofen	571.37	1004.94	0.81	3.61	60.24	37.05	319.88
Chloroquine	421.82	864.45	12.86	5.36	24.51	32.43	293.32

Glucosamine	276.94	526.51	-23.5	-2.11	37.44	15.54	179.17
Menthol	344.33	579.76	1.4	2.78	47.44	18.99	156.27
Meloxicam	450.33	874.87	-11.46	7.33	43.3	31.68	351.21
Leflunomide	375.39	683.39	-9.88	4.39	23.44	23.21	270.21
Probenecid	523.68	834.03	-9.74	8.02	49.88	26.49	285.36
Methylprednisolone	467.92	1014.13	-9.86	5.92	79.8	39.75	374.48
Vitamin E	892.73	1477.37	2.76	9.07	122.31	53.14	430.71

DISCUSSION

Proteins start out life as a cluster of amino acids linked together to guide a protein into its three-dimensional structure, to send signals for interaction with other molecules, to set its stability and lifetime, and to determine its capability to function as an enzyme.

Comparative modeling is a flexible and powerful tool, which can be used in the prediction of the structure, properties, and functions of proteins, is very important in computational chemistry, chemo informatics, immuno informatics and bioinformatics.

An important area is the computational prediction of interactions between proteins (protein active sites) and ligands. The problem of computational prediction of interactions between ligands and proteins are also called molecular docking, and the knowledge about protein–ligand interactions and their prediction is important in pharmacology for the purpose of drug designing.

CONCLUSION

The research finally concludes that Rheumatoid arthritis is related to the changes in concentrations of cell blood counts. Docking studies provides good results for designing drugs against human diseases like RA.

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