



INTERNATIONAL JOURNAL OF ADVANCE RESEARCH, IDEAS AND INNOVATIONS IN TECHNOLOGY

ISSN: 2454-132X

Impact factor: 4.295

(Volume 5, Issue 3)

Available online at: www.ijariit.com

Association of herbal plant in Rheumatoid Arthritis studies using in silico approach

Aparna Hiren Patil Kose
aparna.patil@gnkhalsa.edu.in
Guru Nanak Khalsa College,
Mumbai, Maharashtra

Priyanka Patel
priyankap8433@gmail.com
Guru Nanak Khalsa College,
Mumbai, Maharashtra

Samiksha Wani
nakshatra948@gmail.com
Guru Nanak Khalsa College,
Mumbai, Maharashtra

ABSTRACT

Arthritis is inflammation or degeneration of joints. There are more than 100 different types of arthritis most common types of inflammatory arthritis include Juvenile rheumatoid arthritis (in children's), Rheumatoid Arthritis (RA) (in adults), Psoriatic arthritis, gout, osteoarthritis, gonococcal Arthritis, etc. Rheumatoid Arthritis (RA) is an autoimmune disease. It is also a systemic disease which means it affects the whole body. People with Rheumatoid Arthritis (RA) have a higher risk of some other conditions such as heart disease, obesity, depression, anxiety, high blood pressure, diabetes, etc. No one knows the definite cause of this disease, but there is researcher's evidence that genetics, environmental and stochastic factors are involved. There are various drugs therapies and treatments are available to suppress Rheumatoid Arthritis (RA) and there is a need for an alternative approach to avoid unnecessary side effects and high cost. Therefore, phytochemicals are used which include anti-inflammatory properties. Thus, the present study is aimed for finding phytochemical which is Stigmasterol showing higher binding affinity towards the target receptors those are 4JJ7, 6E28, 5GJH, 4FKL, 4EK4, 3CZF, 5N19, 4RI5, 5BXU and 5N86 using molecular docking approach to find out potential drug molecule against protein causes inflammation. Tools used for molecular docking were GROMACS 5.1.1 for protein optimization, AutoDock 4.2.6 software for docking & Discovery Studio for visualization. Stigmasterol was ranked top among various docking conformations showing highest binding affinity with the target. Therefore it could be an alternative treatment method for rheumatic disorders. Plants like Glycine max (soybean), Physostigma venenosum (calabar bean), Brassica napus (rapeseed) comprise of this phytochemical. Further studies can confirm the usage of these phytochemical and plants for suppressing inflammation via inflammation.

Keywords— Rheumatoid Arthritis, TNF cascade, Stigmasterol, Anti-inflammatory, Docking

1. INTRODUCTION

The immune system is a collection of special cells and chemicals that fight infection-causing agents such as bacteria and viruses. An autoimmune disease occurs when a person's immune system mistakenly attacks their own body tissues. Autoimmune diseases have registered an alarming increase worldwide since the end of the Second World War. This pandemic includes more than 80 autoimmune disorder and an increase in both the incidence and prevalence of autoimmune disorder [1].

Autoimmunity develops over time, and preclinical autoimmunity precedes clinical disease by many years and can be detected in the peripheral blood in the form of circulating auto antibodies [2]. Some autoimmune diseases such as lupus, celiac disease, diabetes mellitus type 1, Graves' disease, inflammatory bowel disease, multiple sclerosis, psoriasis, rheumatoid arthritis, and systemic lupus erythematosus, etc.

Arthritis is though common is not well understood. Actually, "arthritis" is not a single disease; it is an informal way of referring to joint pain or joint disease. There are more than 100 different types of arthritis and related conditions. People of all ages, sexes and races can and do have arthritis, and it is the leading cause of disability in America. It is most common among women and occurs more frequently as people get older [3]. There are various categories of arthritis and they are a follow:

- Degenerative arthritis: E.g.: Osteoarthritis
- Infectious arthritis: E.g.: Gonococcal arthritis
- Metabolic arthritis: E.g.: Gout
- Inflammatory arthritis: E.g.: Rheumatoid Arthritis (RA), Psoriatic arthritis, Juvenile arthritis, Reactive arthritis

Rheumatoid Arthritis (RA) is the most common inflammatory arthritis and is a major cause of disability and is a long-term autoimmune disorder that primarily affects joints. It typically results in warm, swollen, and painful joints. Pain and stiffness often

worsen following rest. Most commonly, the wrist and hands are involved, with the same joints typically involved on both sides of the body.

Nearly seven million people in India suffer from a form of arthritis rheumatoid arthritis - that initially affects the small joints. Yet due to lack of awareness even among the medical fraternity has led to a situation where these patients are being treated like an osteoarthritis patient [4].

Treatment depends on the type and severity of the condition. There is no cure for rheumatoid arthritis. But clinical studies indicate that remission of symptoms is more likely when treatment begins early with medications known as Disease-Modifying Antirheumatic Drugs (DMARDs), Nonsteroidal Anti-Inflammatory Drugs (NSAIDs), Biological agents, ayurvedic, homoeopathy, immunosuppressants are often used [5].

These drugs can target parts of the immune system that trigger inflammation that causes joint and tissue damage. People with rheumatoid arthritis, higher doses of drugs can increase the risk of nausea, anxiety, migraine, ulcer, blood clots, stomach irritation, heart problems, kidney damage, etc.

The use of phytochemical has been considered for the treatment of human diseases since ancient times. Therefore, it is necessary to evaluate the rich heritage of traditional medicines; however, only a few numbers of plant species have been thoroughly studied for therapeutic properties. So, phytochemical can be a novel anti-inflammatory treatment for Rheumatoid Arthritis (RA) [6].

In this present study, molecular docking used to treat inflammation by targeting the 4jj7, 6e28, 5gjh, 4fkl, 4ek4, 3czf, 5ni9, 4ri5, 5bxu, and 5n86 protein receptors which causes inflammation due to increased level of TNF (Tumor necrosis factor alpha) protein inside the joints with Phytosterols such as stigmasterol; Lignan such as arctigenin; Phytochemicals such as coumarins, steroidal saponin, hydroxytyrosol & thymoquinone were seen.

Stigmasterol is a phytosterol and obtained from plant sources like *Glycine max* (soybean), *Physostigma venenosum* (calabar bean), *Brassica napus* (rape seeds) etc which suppress TNF protein that involves in inflammation with no side effects so, it is remarkable to observe that it has attained such good binding energy. Therefore, these phytochemical can be considered as potent medicine against Rheumatoid Arthritis (RA).

2. METHODOLOGY

Since ancient time India uses herbal medicines in the officially alternative systems of health such as Ayurveda, Unani, Sidha, Homeopathy, and Naturopathy [7]. In India, there are more than 2500 plants species which are currently used as herbal medicaments. Thus, from the knowledge of traditional plants, one might be able to discover new effective and cheaper drugs [8]. Thus the study has tried to cover all the strategies that are followed for the treatment of Rheumatoid Arthritis without any possible side effects. The future treatment of Rheumatoid Arthritis (RA) should provide more effective.

2.1 Selection of proteins

Total 10 RA proteins such as 4jj7, 6e28, 5gjh, 4fkl, 4ek4, 3czf, 5ni9, 4ri5, 5bxu, and 5n86 were selected as a target. The functions, mechanisms & pathways of these proteins were analyzed by UniProt database then; three-dimensional structures of RA proteins were taken from the PDB database on the basis of their resolution value & experimental methods.

2.2 Selection of ligands

Total 6 phytochemicals as ligands namely Phytosterols such as stigmasterol; Lignan such as arctigenin; Phytochemicals such as coumarins, steroidal saponin, hydroxytyrosol & thymoquinone were checked for the Lipinski's rule of 5 in PubChem (NCBI). Download 3D conformer of ligands, then clean 2D & 3D structures by MarvinSketch tool.

2.3 Energy minimization

Proteins are macromolecules & need to be a stable structure for molecular docking. Energy minimization helps to stabilize protein structure by GROMACS 5.1.1 software.

2.4 Docking studies

Energy minimized proteins were docked with selected phytochemicals to find a potent drug for RA disease which acts as an anti-inflammatory agent by using AutoDock 4.2.6 software. AutoDock is an automated procedure for predicting the interaction of ligands with biomacromolecule targets.

2.5 Visualization

Interaction between ligands (phytochemicals) & proteins was visualized by Discovery Studio 19.1 Visualizer. Discovery Studio is a comprehensive software suite for analyzing and modelling molecular structures, sequences, protein-ligand interactions and other data of relevance to life science researchers.

3. RESULTS

3.1 Docking results

Molecular docking approach was used to determine potent phytochemicals which will show the best binding energy with the selected RA proteins. Total 6 ligands (namely stigmasterol, arctigenin, coumarins, steroidal saponin, hydroxytyrosol & thymoquinone) were docked against each of 10 proteins (namely 4JJ7, 6E28, 5GJH, 4FKL, 4EK4, 3CZF, 5NI9, 4RI5, 5BXU and 5N86) using AutoDock

tool where coordinates for each docked conformation to the docking log file is been created, along with information on clustering and interaction energies and provides options for analyzing the information stored in the docking log file. With this study; it was found that stigmasterol docking score with all 10 proteins were showing the highest binding affinity as binding energy obtained in negative values indicates that the scores obtained are highly efficient. The following table shows top docking poses ranked according to their binding energy.

Table 1: Top docking conformations showing binding energy of docked structures

S no.	PDB ID	Phytochemical	Binding energy (Kcal/mol)
1	4JJ7	Stigmasterol	-8.26
2	6E28		-7.28
3	5GJH		-7.37
4	5NI9		-7.58
5	5N86		-8.51
6	3CZF		-10.13
7	5BXU		-7.35
8	4FKL		-7.51
9	4EK4		-7.91
10	4RI5		-8.78

3.2 Visualization results

Since docking was performed by AutoDock software; protein-ligand interactions of the docked structure were interested to be observed and performed using Discovery Studio 19.1 Visualizer.

Following figures shows protein-ligand interactions viewed in Discovery Studio 19.1 Visualizer where amino acid residues are labelled in green and ligand is coloured in green. Top-ranked phytochemical (Table 1) were selected to show interaction with the Rheumatoid Arthritis (RA) proteins.

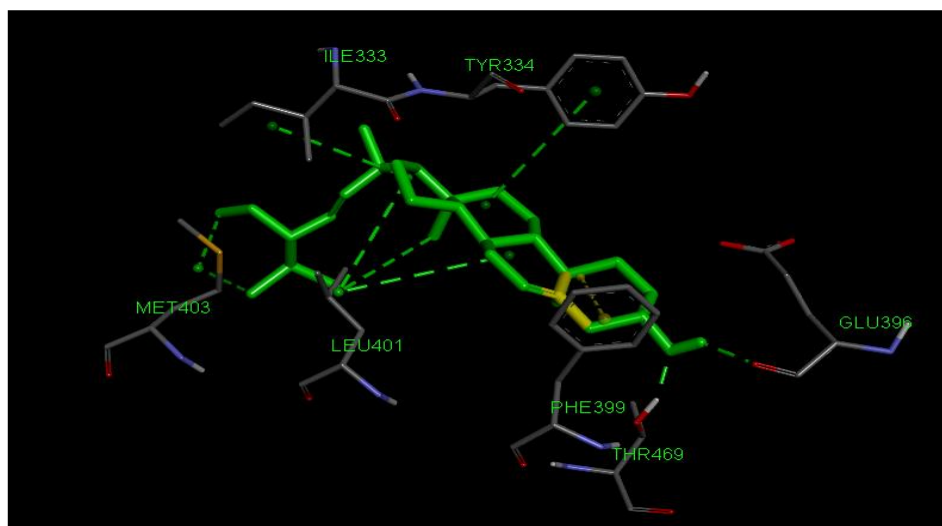


Fig. 1: 4jj7 docked with Stigmasterol showing various interaction with amino acids – ILE333, TYR334, PHE399, GLU396, LEU401, MET403 and THR469 (Refer table 1)

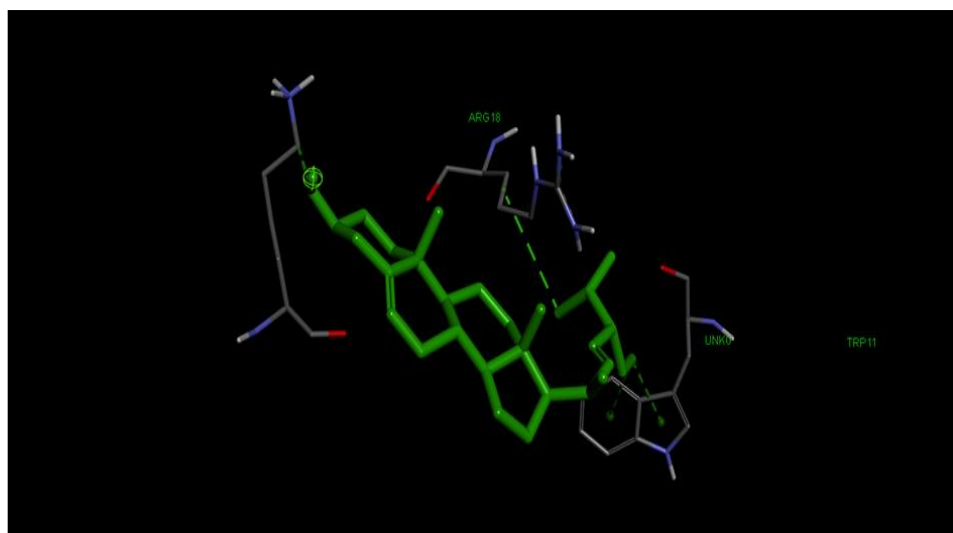


Fig. 2: 6e28 docked with Stigmasterol showing various interactions with amino acids- UNK0, TRP11 and ARG18 (Refer table 1)

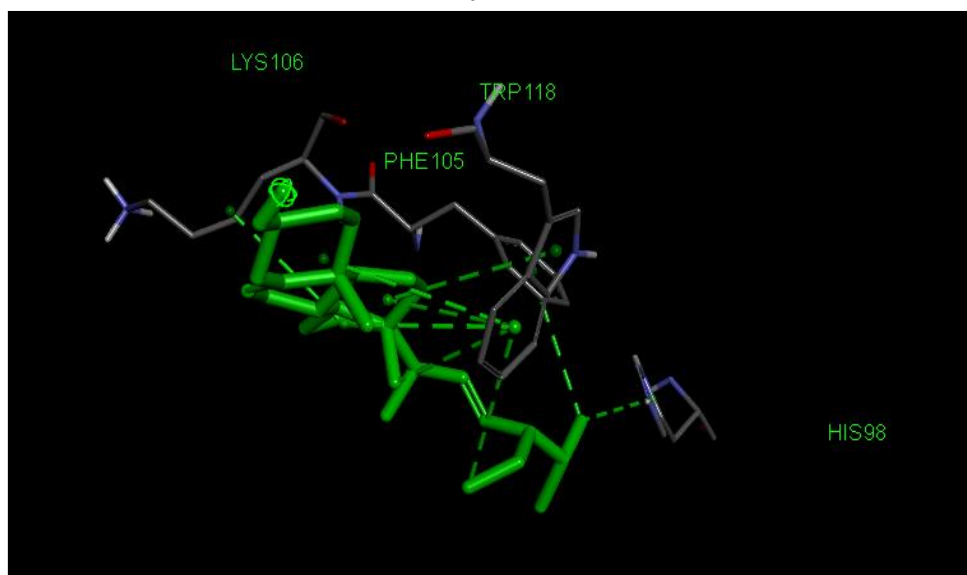


Fig. 3: 5gjh docked with Stigmasterol showing various interactions with amino acids- HIS98, PHE105, LYS106 and TRP118 (Refer table 1)

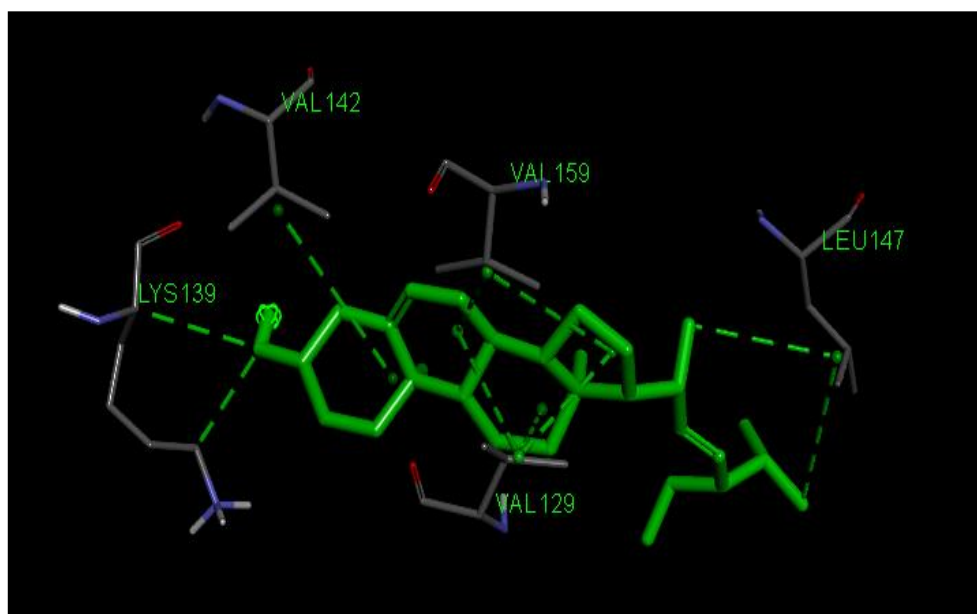


Fig. 4: 5ni9 docked with Stigmasterol showing various interactions with amino acids- VAL129, LYS139, VAL142, LEU147 and VAL159 (Refer table 1)

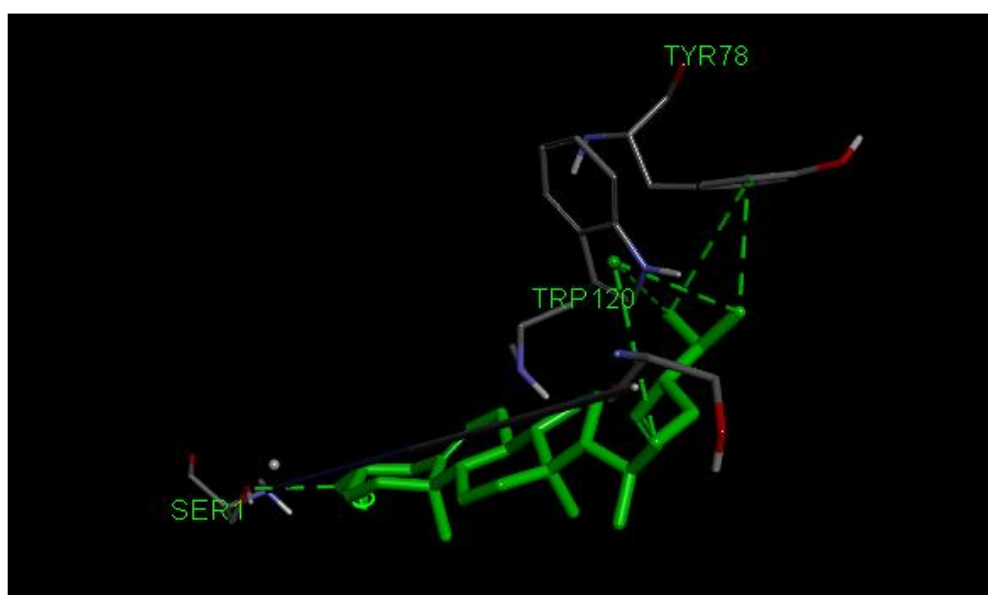


Fig. 5: 5n86 docked with Stigmasterol showing various interactions with amino acids- SER1, TYR78 and TRP120 (Refer table 1)

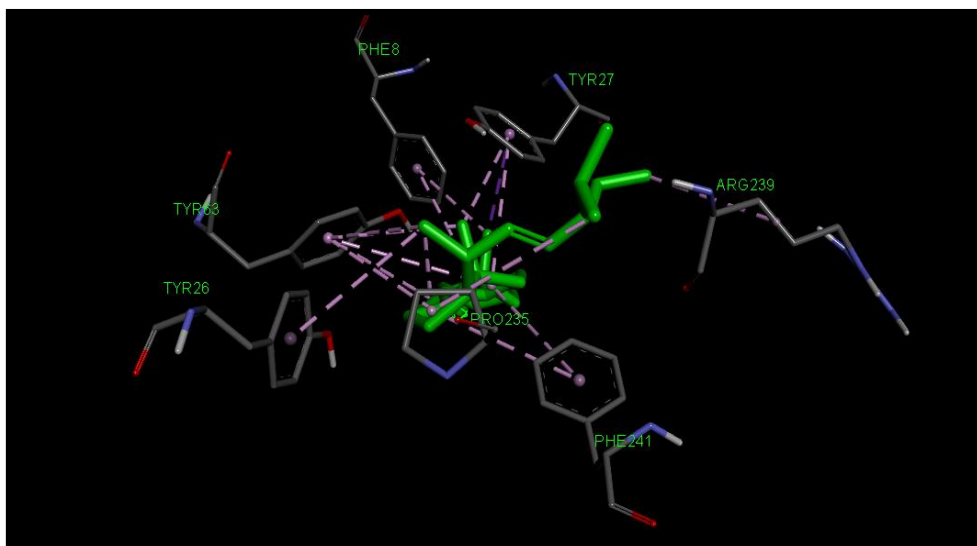


Fig. 6: 3czf docked with Stigmasterol showing various interactions with amino acids- PHE8, TYR26, TYR27, TYR63, PRO235, ARG239 and PHE241 (Refer table 1)

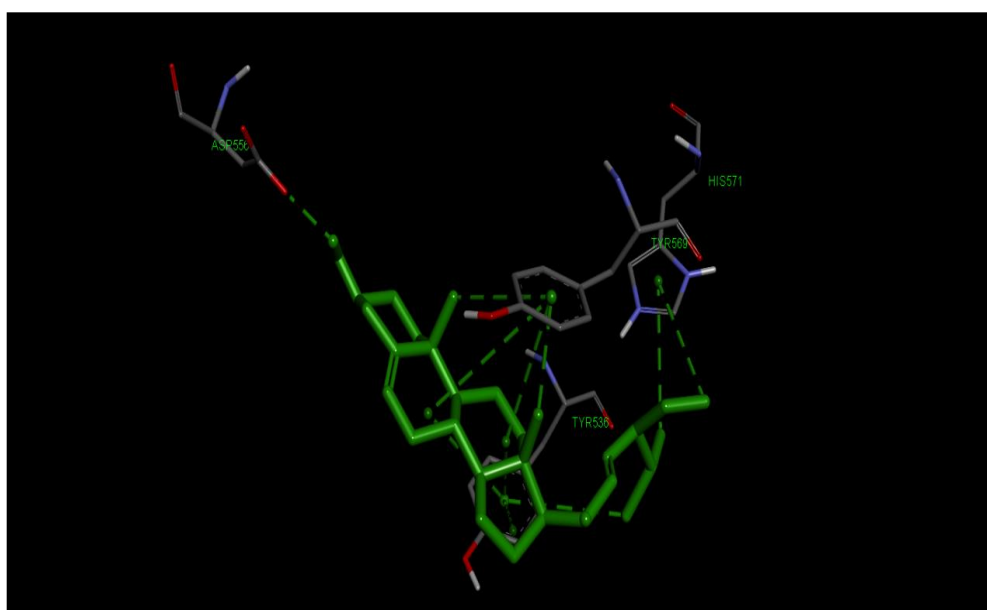


Fig. 7: 5bxu docked with Stigmasterol showing various interactions with amino acids- TYR536, ASP556, TYR569 and HIS571 (Refer table 1)

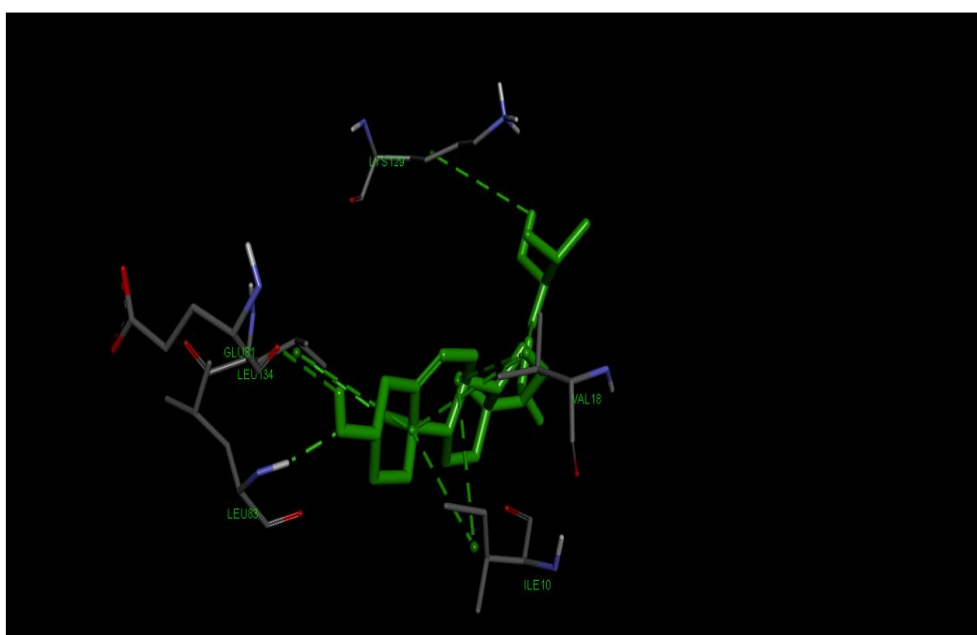


Fig. 8: 4fkl docked with Stigmasterol showing various interactions with amino acids- ILE10, VAL18, GLU81, LEU83, LYS129 and LEU134 (Refer table 1)

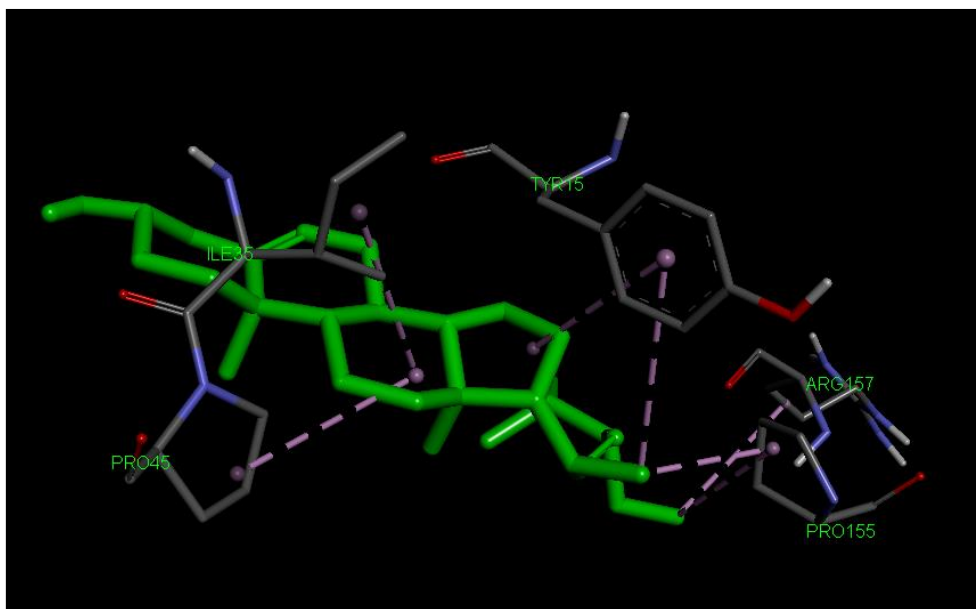


Fig. 9: 4ek4 docked with Stigmasterol showing various interactions with amino acids- TYR15, ILE35, PRO45, PRO155 and ARG157 (Refer table 1)

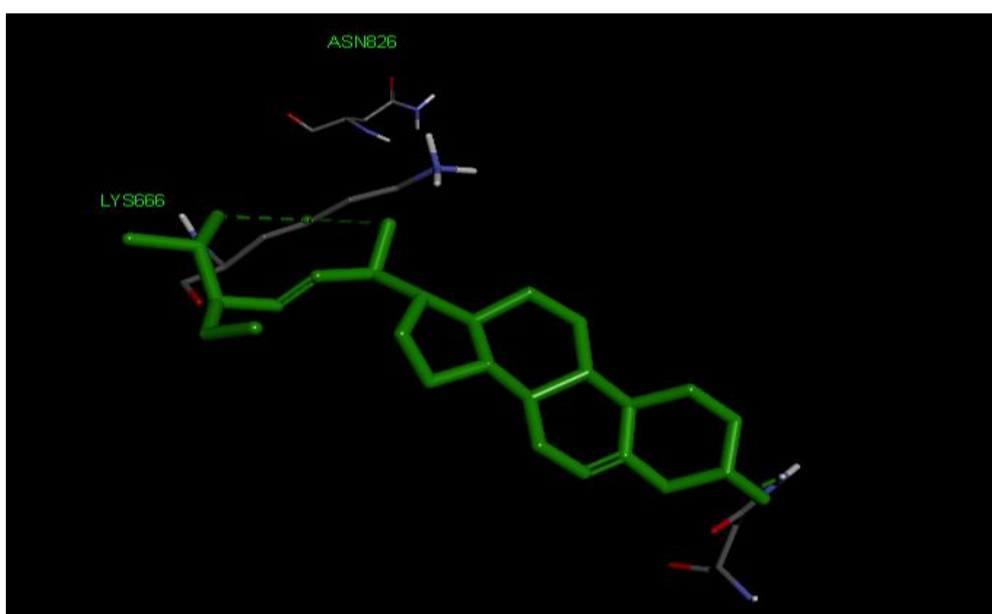


Fig. 10: 4ri5 docked with Stigmasterol showing various interactions with amino acids- LYS666 and ASN826 (Refer table 1)

3.3 Interpretation

Molecular docking studies are used to determine the interaction of two molecules and to find the best orientation of ligand which would form a complex with overall minimum energy. The small molecule known as ligand usually fits within the protein's cavity which is predicted by the search algorithm. These protein cavities become active when they come in contact with any external compound and thus called as active sites.

Docking is frequently used to predict the binding orientation of small molecule drug candidates to their protein targets in order to predict the affinity and cavity of the small molecule. Hence docking plays an important role in the rational drug design. Binding is directly proportional to the stability of the docked structure.

4. DISCUSSION

In this present study, molecular docking used to treat inflammation by targeting the 4jj7, 6e28, 5gjh, 4fkl, 4ek4, 3czf, 5ni9, 4ri5, 5bxu, and 5n86 protein receptors which causes inflammation due to increased level of TNF (Tumor necrosis factor alpha) protein inside the joints with Phytosterols such as stigmasterol; Lignan such as arctigenin; Phytochemicals such as coumarins, steroidal saponin, hydroxytyrosol l& thymoquinone were seen. From these the top binding energy with the target is only observed in stigmasterol ranging from -7.00Kcal/mol to -10.13 Kcal/mol (table 1).

Stigmasterol is a phytosterol and obtained from plant sources like *Glycine max* (soybean), *Physostigma venenosum* (calabar bean), *Brassica napus* (rape seeds) etc which suppress TNF protein that involves in an inflammation with no side effects so, it is remarkable to observe that it has attained such a good binding energy up to 10.13Kcal/mol. Therefore, these phytochemical can be considered as potent medicine against Rheumatoid Arthritis (RA).

Thus, very few *in silico* studies have been performed on Rheumatoid Arthritis (RA) proteins with respect to phytochemicals. On the basis of negative scores of binding energy, comparing current results with literature; we can conclude that the binding affinity obtained in the top docking poses is good. Various interactions with residues were visualized between protein and ligand. These interactions are assumed to be possible binding sites using Discovery Studio 19.1 Visualizer.

5. CONCLUSION

Modern medicine still needs the help of alternative and complementary medicine, which is most often based on phytotherapy. Therefore, the identification and characterization of plant extracts and their major bioactive phytochemicals (e.g. steroids, phytosterol, and vitamins) are highly valuable used as a potent drug for Rheumatoid Arthritis (RA).

Molecular docking approach was used to determine phytochemicals showing good binding affinity with Rheumatoid Arthritis (RA) proteins by AutoDock tool. Majorly, stigmasterol which belongs to phytosterol family of steroid provides high docking score. Stigmasterol was obtained from plant sources like *Glycine max* (soybean), *Physostigma venenosum* (calabar bean), *Brassica napus* (rape seeds) etc. and these interactions of proteins with phytochemicals perceived by using Discovery Studio 19.1 Visualizer. Hence, stigmasterol can be used as a potential drug for anti-inflammatory activity in rheumatoid arthritis which can suppress or inhibit the activity of RA proteins like 4jj7, 6e28, 5gjh, 4fkl, 4ek4, 5ni9, 5n86, 4ri5, 3czf and 5bxu can give relief to patients.

Stigmasterol is a phytosterol which suppresses TNF protein that involves inflammation with no side effects so, it is remarkable to observe that it has attained such good binding energy. Pharmacogenetic and pharmacogenomic studies, which help to determine the genetic profile of individual patients, may bring us closer to personalized medicine for Rheumatoid Arthritis (RA).

6. ACKNOWLEDGEMENT

The authors would like to show their gratitude to the GNIRD, Guru Nanak Khalsa College, the University of Mumbai for sharing their pearls of wisdom with us during the course of this research. The authors also want to thanks Rishi Biotech Company for providing the necessary information.

7. REFERENCES

- [1] Chandrasekar, r., & chandrasekar, s. (2017). Natural herbal treatment for rheumatoid arthritis-a review. International journal of pharmaceutical sciences and Research, 8(2), 368.
- [2] Wadekar, J. B., Sawant, R. L., & Patel, U. B. (2015). Rheumatoid arthritis and herbal drugs: A review. J Psychopharmacol, 4, 311-8.
- [3] Campbell, A. W. (2014). Autoimmunity and the gut. Autoimmune diseases, 2014.
- [4] R. (2001, October 18, Thursday). Early detection is crucial to treat rheumatoid arthritis. Retrieved June 10, 2019, from <https://www.thehindu.com/thehindu/2001/10/18/stories/08180003.htm>
- [5] Klak, A., Paradowska-Gorycka, A., Kwiatkowska, B., & Raciborski, F. (2016). Personalized medicine in rheumatology. Reumatologia, 54(4), 177.
- [6] Altemimi, A., Lakhssassi, N., Baharlouei, A., Watson, D., & Lightfoot, D. (2017). Phytochemicals: Extraction, isolation, and identification of bioactive compounds from plant extracts. Plants, 6(4), 42.
- [7] Ravishankar, B., & Shukla, V. J. (2007). Indian systems of medicine: a brief profile. African Journal of Traditional, Complementary and Alternative Medicines, 4(3), 319-337.
- [8] Pan, S. Y., Zhou, S. F., Gao, S. H., Yu, Z. L., Zhang, S. F., Tang, M. K., ... & Ko, K. M. (2013). New perspectives on how to discover drugs from herbal medicines: CAM's outstanding contribution to modern therapeutics. Evidence-Based Complementary and Alternative Medicine, 2013.