Designing Visfatin inhibitors to limit its Insulin Mimcy and Type II Diabetes.

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Keywords: Visfatin, Type II Diabetes, Inflammation

Visfatin, otherwise known as Nicotinamide phosphoribosyltransferase (NAmPRTase or Nampt), is an adipocytokine that promotes B cell maturation and inhibits neutrophil apoptosis as well as promoting the condensation of nicotinamide. Visfatin plays an important role in promoting insulin resistance by binding to insulin receptor (IR) at a site distinct from insulin exerting a variety of insulin-mimetic effects, thereby playing a role in the development of obesity-associated insulin resistance and Type II diabetes. This research sought to understand binding interaction of pharmaceuticals to Visfatin. 11 crystal structures of the Visfatin were docked using IGEMDock to FDA, Alkaloids, Lactams, Lactones, Flavinoids, Sulfanilamide, Cyclic Imides, and NSAIDs drugs to determine structural correlation for the most effective binders. Structural similarities were determined with IGEMDock and vROCS and partition coefficient was determined using DRAGON program. This data found a cluster of potential inhibitors to Visfatin which are possible targets for Type II diabetes treatments. This research will be used in the engineering of improved Visfatin inhibitors.

Introduction

This project was designed around structural understanding and pharmaceutical engineering of the Visfatin inhibitors. Visfatin being a fat produced insulin mimic may play a part in the initiation of Type 2 diabetes. Additionally, its interaction as an adipocytokine may be a factor in inflammatory damage in obese individuals. The understanding and limitation of Visfatin may help limit both Type 2 diabetes and inflammatory damage caused by Visfatin.

Specific and Overall Goal

The overall goal of this research is to investigate the interaction of multiple drug candidates to find the best drug candidates for targeted inhibition of Visfatin. This research will first determine the binding and chemical properties of the Visfatin active site molecules as a control group. Secondly, a group of select drug candidates whose properties are more effective at binding to the active site versus the control molecules will be chosen. Drug classification analysis will indicate preferences to improved active site binding. Finally, quantitative structure and activity relationship (QSAR) analysis will be done on both the control and experimental molecules to identify similar trends and values. Once found, similarities between these drugs will allow for a better understanding of the inhibition of Visfatin.

Experimental Methodology

1. The selection of 11 isoforms of Visfatin.
2. The screening and analysis of multiple drug candidates using IGemDock.
3. Similarity calculations were done to determine if molecular functionalities showed any preference to increased binding.
4. Selected candidates from IGemDock based on binding energy were tested using Dragon to determine structural similarities.

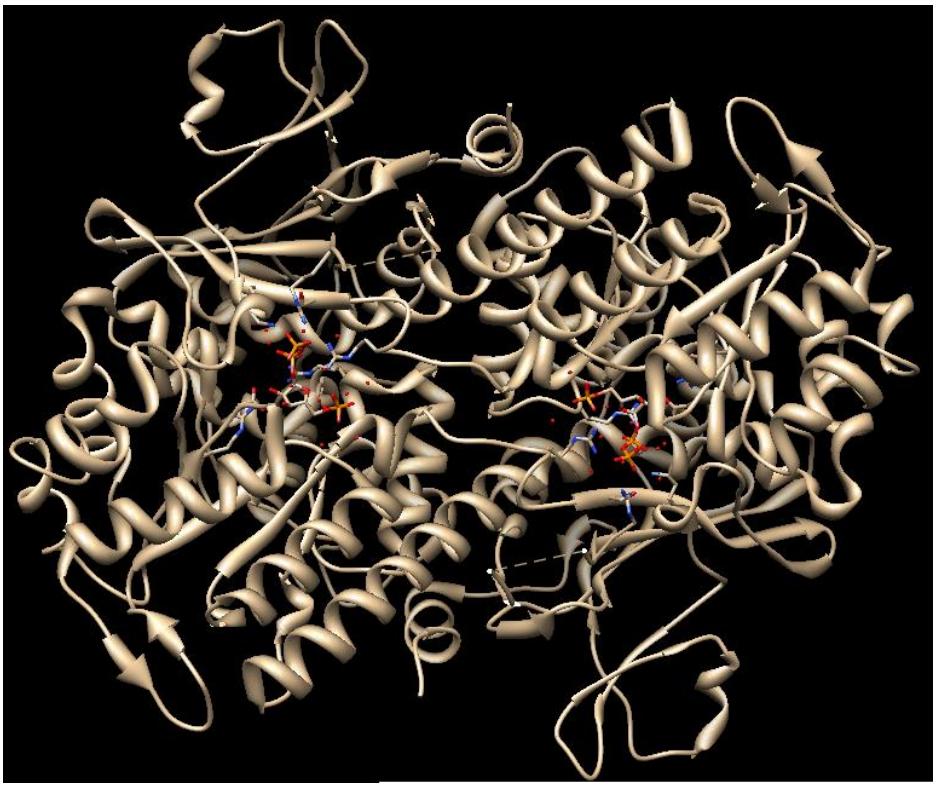
Methods and Materials

1172 structures were chosen which included, 715 FDA approved, 197 Alkaloids, 73 Imides, 40 Lactams, 36 Lactones, 50 NSAIDs, 25 Sulfanilamide and 37 Flavonoids pharmaceuticals were selected as ligands and computationally bound to 11 isoforms of the Visfatin protein using IGEMDock. IGEMDock used two independent docking with the average of both binding to factor in binding selectivity. An ANOVA was done to determine if any discrepancies in binding were seen between proteins. Additionally, grouping of the molecular functionalities were determine to find differences. The best 10 drug candidates on binding energies were selected and structural data such as molecular weight and partition coefficient was collected using Dragon.

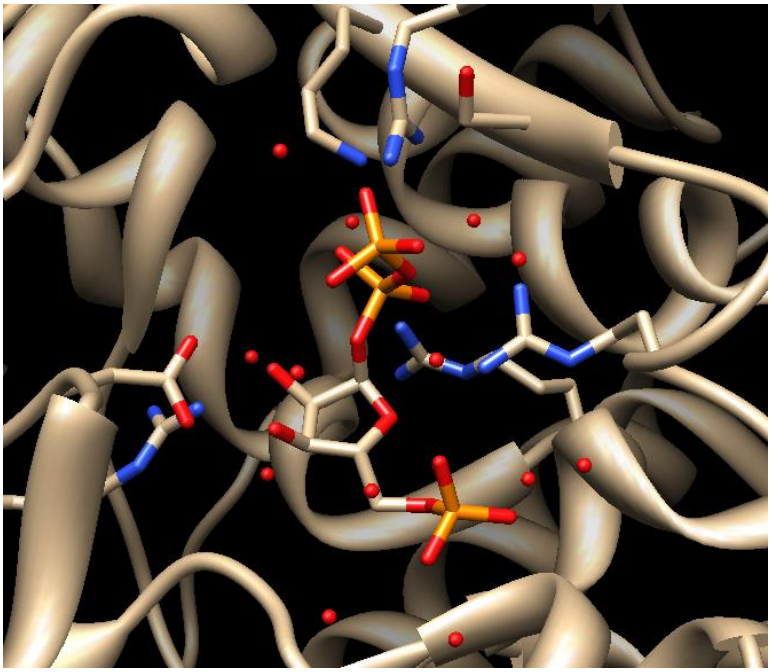
Visfatin Crystal Structures from Protein Databank

#	PDB	Title
1	2E5C	Crystal structure of Human NMPRTase complexed with 5'-phosphoribosyl-1'-pyrophosphate
2	2E5D	Crystal structure of Human NMPRTase complexed with nicotinamide
3	2G96	Crystal Structure of Visfatin In Complex with Niconamide Mononucleotide
4	2G97	Crystal Structure of Visfatin In Complex with the Specific Inhibitor FK-866
5	2GVG	Crystal Structure of human NMPRTase and its complex with NMN
6	2GVL	Crystal Structure of Murine NMPRTase
7	2H3B	Crystal Structure of Mouse Visfatin
8	3DGR	Crystal structure of human NAMPT complexed with ADP analogue
9	3DHD	Crystal structure of human NAMPT complexed with nicotinamide mononucleotide and pyrophosphate
10	3DKJ	Crystal structure of human NAMPT complexed with benzamide and phosphoribosyl pyrophosphate
11	3DKL	Crystal structure of human NAMPT complexed with benzamide and phosphoribosyl pyrophosphate

Crystal Structure of 2E5C (UCSF Chimera)



Active Site of 2E5C (UCSF Chimera)



Summary of 1172 Drug Candidates vs Proteins (IGEMDock Data).

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
2E5C	2344	-187874	-80.1508	6019.472
2E5D	2344	-172874	-73.7518	5691.071
2G96	2344	-184175	-78.5729	5881.788
2G97	2344	-176668	-75.3704	5409.499
2GVG	2344	-196070	-83.6476	6144.705
2GVL	2344	-115955	-49.4689	7256.643
2H3B	2344	-156773	-66.8826	5304.75
3DGR	2344	-184425	-78.6798	4800.213
3DHD	2344	-186579	-79.5986	6195.604
3DKJ	2344	-191112	-81.5322	6340.178
3DKL	2344	-152832	-65.2012	5911.7

ANOVA of 1172 drug candidates (IGEMDock Data).

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	2328693	10	232869.3	39.43557	6.27E-78	1.83107
Within Groups	1.52E+08	25773	5905.057			
Total	1.55E+08	25783				

Breakdown of drug candidates by types (IGEMDock Data).

Classification	Average	St. Dev
Alkaloids	-88.31890148	12.78578401
Flavonoids	-92.04583914	15.64317212
Imides	-89.20714988	10.78330885
Lactams	-86.50791125	13.02275967
Lactones	-80.13076378	67.6932637
NSAIDs	-91.34529884	10.11470257
Sulf	-84.68074545	8.485802076

Drug candidates by types (IGEMDock Data).

# of Drugs	Drug Title	Energy
1	FDA - 99-1	-136.84
	FDA - 99-0	-135.57
2	FDA - 570-0	-146.57
	FDA - 570-1	-145.65
3	FDA - 266-0	-135.91
	FDA - 266-1	-135.77
4	FDA - 446-1	-153.41
	FDA - 446-0	-151.65
5	FDA - 182-1	-152.83
	FDA - 182-0	-149.23
6	FDA - 533-1	-130.52
	FDA - 533-0	-129.69
7	FDA - 288-0	-128.29
	FDA - 288-1	-128.02
8	Vincristine-1-1	-122.65
	Vincristine-1-0	-116.12
9	Etoposide-1-0	-122.01
	Etoposide-1-1	-120.41
10	Alkaloids - 504 -0-1	-117.91
	Alkaloids - 504 -0-0	-114.93

Summary of Control Drugs (IGEMDock Data).

Visfatin Control	
Low Value	-37.9203
High Value	470.4448
Average	9.625877
Standard Deviation	104.82

Dragon Data of Visfatin Control Molecules.

NAME	MW	MLOGP	MLOGP2
3G8E_IS1_A_501	486.3	2.456	6.031
3DKL_UNU_A_504	114.1	1.308	1.711
3DKL_PRP_B_503	377	-0.664	0.441
3DKJ_UNU_A_502	114.1	1.308	1.711
3DKJ_PRP_A_501	377	-0.664	0.441
3DHD_NMN_A_503	319.1	-0.067	0.004
3DGR_A12_A_501	408.1	-0.57	0.325
2H3D_NMN_A_3819	319.1	-0.067	0.004
2H3B_SO4_A_1000	96.07	-2.399	5.755
2GVJ_DGB_A_502	362.3	3.333	11.108
2GVG_NMN_A_501	319.1	-0.067	0.004
2GVG_NMN_C_503	319.1	-0.067	0.004
2GVG_NMN_E_505	319.1	-0.067	0.004
2G97_DGB_A_1001	362.3	3.333	11.108
2G96_NMN_A_1001	319.1	-0.067	0.004
2E5D_NCA_A_1501	116.1	0.119	0.014
2E5C_PRP_A_902	377	-0.664	0.441

Dragon Data of Visfatin drug candidates.

NAME	MW	MLOGP	MLOGP2
FDA 2 - 446	359.04	0.364	0.133
FDA 2 - 570	312.211	-0.009	0
FDA 2 - 266	254.15	0.372	0.139
FDA 2 - 182	516.61	0.391	0.153
FDA 2 - 533	265.13	3.174	10.076
FDA 2 - 99	504.72	2.029	4.117

Discussion

Multiple compounds were identified as effective based upon their interactions with each protein. Specifically an average energy of -133.699 was found for the drug candidates compared to 9.625877 for the control molecules. There were 4 control molecule which seemed to be an outliers with values of 39.8249, 87.6701, 99.1493, and 470.4448. Without these outliers the average decreases to -23.3034. An ANOVA to determine differences between the

11 proteins analyzed indicated major differences with a F value of 39.43557 compared to F critical value of 1.831. A search of the data indicates that 3DKL shows the greatest differences. Grouping analysis will be used to understand differences in protein active sites. 10 drugs were chosen due to their low binding energies (for both binding interactions). Structural analysis found that many of these molecules are small with similar partition coefficient (-0.009 to 3.174) of the top binders similar to that of the control molecules.

Conclusion

By using the computational techniques we were able to identify several molecule that show improved binding efficacy over currently used Visfatin inhibitors. These Visfatin drug candidates indicated a diverse pool of Visfatin binders with improved efficacy. This work can be used to engineer these motifs into novel Visfatin inhibitors for improved drug efficacy.

Acknowledgments

This Project was supported by The College of Natural Science and Mathematics at The Richard Stockton College of New Jersey.

References

1. Cinar, N.; Gurlek, A., Association between novel adipocytokines adiponectin, vaspin, visfatin, and thyroid: An experimental and clinical update. *Endocr Connect* 2, (4), R30-8.
2. Shrestha, C.; He, H.; Liu, Y.; Zhu, S.; Xiong, J.; Mo, Z., Changes in Adipokines following Laparoscopic Roux-en-Y Gastric Bypass Surgery in Chinese Individuals with Type 2 Diabetes Mellitus and BMI of 22-30 kg.m(-2.). *Int J Endocrinol* 2013, 240971.
3. Al-Suhaimi, E. A.; Shehzad, A., Leptin, resistin and visfatin: the missing link between endocrine metabolic disorders and immunity. *Eur J Med Res* 18, 12.
4. Lu, H.; Gauvreau, D.; Tom, F. Q.; Lapointe, M.; Luo, X. P.; Cianflone, K., Inflammatory markers and adipokines alter adipocyte-derived ASP production through direct and indirect immune interaction. *Exp Clin Endocrinol Diabetes* 121, (4), 194-200.
5. Mazaherioun, M.; Hosseinzadeh-Attar, M. J.; Janani, L.; Vasheghani Farahani, A.; Rezvan, N.; Karbaschian, Z.; Hossein-Nezhad, A., Elevated serum visfatin levels in patients with acute myocardial infarction. *Arch Iran Med* 15, (11), 688-92.
6. Mannino, G. C.; Sesti, G., Individualized therapy for type 2 diabetes: clinical implications of pharmacogenetic data. *Mol Diagn Ther* 16, (5), 285-302.
7. Skvarca, A.; Tomazic, M.; Krhin, B.; Blagus, R.; Janez, A., Adipocytokines and insulin resistance across various degrees of glucose tolerance in pregnancy. *J Int Med Res* 40, (2), 583-9.
8. Kang, Y. S.; Cha, D. R., The role of visfatin in diabetic nephropathy. *Chonnam Med J* 47, (3), 139-43.
9. Derosa, G.; Cicero, A. F.; D'Angelo, A.; Fogari, E.; Maffioli, P., Effects of 1-year orlistat treatment compared to placebo on insulin resistance parameters in patients with type 2

diabetes. *J Clin Pharm Ther* 37, (2), 187-95.
10. Sonoli, S. S.; Shivprasad, S.; Prasad, C. V.; Patil, A. B.; Desai, P. B.; Somannavar, M. S., Visfatin--a review. *Eur Rev Med Pharmacol Sci* 15, (1), 9-14.