Insilico Methodolgy - Resistin Involved In Diabesity

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Citation

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Abstract

In the present study, the role of several genes/proteins involved in the evolution of diabesity have been studied by several bioinformatics tools and software. Sequence and Phylogenetic Analysis was done using SDSC Biology workbench, a web based bioinformatics toolkit. Our bioinformatics analysis reports resistin gene as ominous link with diabesity. This bioinformatics study will be useful for future studies towards therapeutic inventions of diabesity. A multiple sequence alignment approach is used followed by construction of phylogenetic tree to compare the homology of functional proteins in one species and between other species. Sequences are analyzed to find mutations, evolutionary relationships, predicting structure, function and its abnormalities.

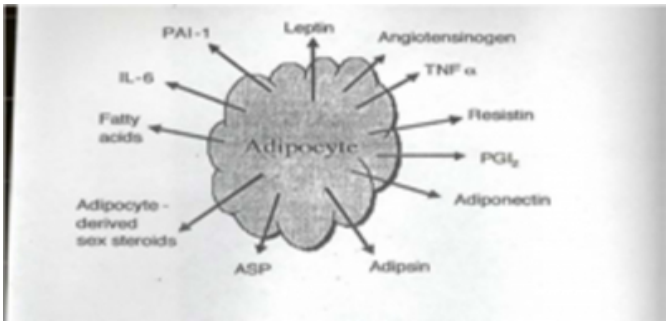
INTRODUCTION

Diabetes associated with obesity (diabesity) is common and is a life-long disease. In view of this, it calls for effective co-ordination among doctors, nurses, dietitians that requires reliable documentation and exchange of information[1]. The managementof patients with type 1 diabetes mellitus requires regular insulin injections and monitoring of the metabolic status of the patient Diabetes mellitus is a chronic disease[2]. In this article, we briefly reviewed the computer assisted case interventions and current state of bioinformatics in the understanding of diabetes associated with obesity. The diabesity epidemic-i) Prevalence of diabetes

135 million people in 1995[3]. ii) Projected to be over 300 million by 2025[6}. iii)Over 80% of type 2 diabetic patients are overweight .The adipocyte is a metabolically active source of multiple proteins and cytokines that act via autocrine, paracrine and endocrine means- The adipocyte, gut and brain communicate regarding the body's state of energy balance and set the "satiety" thermostat [4,5]

Figure 1

Fig 1



The Industrialization of computational biology research has resulted in an explosion of Bioinformatics data (like protein sequences, protein structures and its domain). Experimental methods capable of generating sets of co-regulated genes have become commonplace, however, recognizing remains difficult. As a result, computational detection of inhibitors in such data sets has been an active area of research. These existing methods have varying degrees of success depending on the strength and length of the amino acids and the number of available sequences [7-15]. We present a deterministic result for the Resistin protein among other functional proteins. Unlike other methods, sequences in the entire genome and the query sequence are taken into account in order to discriminate against commonly occurring signals and produce patterns. Recent studies revealed that the incidence of diabetes mellitus is assuming epidemic proportions both in the developing and developed world. This has been attributed largely to Westernized life style

pattern. In view of this increasing incidence of diabetes mellitus, it is imperative that more sophisticated, fast, reliable and robust methods need to be devised to develop the best use of information science and technology in relation to diabetes, decision support and clinical management [16-22]

Over the years a wide range of comparative modeling-based methods have been developed for predicting the structure of a protein (target) from its amino acid sequence. The central idea behind these techniques is to align the sequence of the target protein to one or more template proteins and then construct the target's structure from the structure of the template(s) using the alignment(s) as reference. The overall performance of comparative modeling approaches depends on how well the tree, constructed by considering sequence and sequence-derived information, agrees with the structure between the target and the template proteins. This can be quite challenging, as two proteins can have high structural similarity even though there exists very little sequence identity between them [23-26]. This led to the development of sophisticated profile-based methods and scoring functions1 that allowed high-quality alignments between protein pairs whose sequence identities are as low as 20%. However, these profilebased methods become less effective for protein pairs with lower similarities. As a result, researchers are increasingly relying on scoring methods that also incorporate various predicted structural information such as secondary structure, backbone angles, and protein blocks.[7,12,13,20]

Recently developed methods [21] that can accurately estimate the various parameters of resistin protein w.r.t to other proteins.For the first time, we're starting to understand why it is that being obese, diabetic and insulin resistant increases risk of atherosclerotic disease two-to-five-fold. Patients with diabesity have increased risk of coronary artery disease, stroke, and peripheral vascular disease [27-33]. Advances in scientific knowledge have provided the medical community with information and disease management strategies designed to tackle diabetes and its related complications. These strategies include behavioral and pharmaceutical methods for maintaining tight glycemic control, reducing hypertension, and reducing plasma lipids, which can help to prevent complications of the disease [34-39]. Because patient behavioral regimens can have a salient impact on the course of disease in diabetes, a patient-centered approach is particularly important. Accordingly, there is evidence to suggest that assessment of quality of life can be can be used in a clinical setting to improve communication among physician, diabetologist and patient to ensure that the clinical encounter focuses on topics that enhance well-being, that include effective symptom management, referral to relevant sub-specialty clinician , and good diet control and effective medication [40-42].

Through our research, we will look for the mechanisms involved. We hope that the knowledge we gain will help in preventing and treat Diabesity is classed as a metabolism disorder. '

This is an exciting area of research for researchers to pinpoint relationship between obesity-associated diabetes and heart disease The rise in diabesity, now at epidemic levels in the United States, a deadly combination that increases heart disease risk by two to five times. Research has shown an association between obesity and diabetes (diabesity) [43-48]

MATERIALS AND METHODS

Analysis encompasses a wide variety of methods used to find recurrent trends in data. In Bioinformatics the predominant applications are sequence analyses and micro array data analyses

For the functional protein i;e resistin (receptor) for diabesity

Sequence analyses – BLASTP

Basic Local Alignment Search Tool---BLAST is one of the most widely used bioinformatics programs. Blast enables to compare a query sequence with a library or database of sequences. It identifies library sequences resembling the query sequence above a certain threshold which is a heuristic method .Performs local alignments through searches of high scoring segment pairs (HSP's before fast algorithms such as BLAST and FASTA were developed Dynamic programming was used. Database searches for the protein or a nucleic sequence was very time consuming by using a full alignment program like dynamic programming. BLAST is about 50 times faster than the dynamic programming. BLAST is more time efficient than FASTA by searching only for the more significant patterns in the sequences, but with comparative sensitivity When we want to analyze a sequence, the best thing to do is to search for homologous sequences in a database. Sequence alignment is an arrangement of primary sequence of DNA, RNA, or protein. Identification of regions of similarity. Consequence of functional, structural, or evolutionary relationships between the sequences.BLAST

can perform both Global and Local alignment. Tips to improve BLAST searches Don't always use the default parameters View reports

Sequence analyses is done using BLASTP for query sequence i;e Resistin protein with the following parameters for given organisms for Resistin is a recently discovered signal molecule, which could help elucidation of the pathophysiology of the insulin resistance and its correlation with obesity [49-51] As little information was available about resistin determination in venous blood at the time of our study, we focused on the question whether any correlation exists between persons with type 2 diabetes mellitus, with systemic inflammation, healthy persons and resistin concentrations and laboratory markers of inflammation, peptone, BMI. Differences of resistin values in these types of volunteers were studied as well.[52-55]

In this paper we focus mainly on tree representation of the family history of set of sequences that share a common ancestor is called a PhylogeneticTree. A phylogeny tree shows the connection among various organisms and weight of the branches in the tree indicates time between evolutions of different organisms.Uses of Phylogenetic Tree

- Determining the relatives of the organisms and interested.
- Identify the functionality of a gene
- Trace the origin of a gene

1.Retrieving Required Sequence (Protein/DNA) from Major Databases.

1.1. Retrieving Protein sequence using major Protein databases.

Protein Information Resource(PIR)

Example to retrieve required Protein sequence from Swiss-Port

1.1.1.Open the link at expasy

1.1.2 Present required protein/gene i:e

Resistin sequence in the Gene name

window , and Submit .

1.1.3.Select the required gene from given result

1.1.4.Obtain FASTA format of it .

1.1.5.Save the Sequence as in fig a

1.2. Retrieving DNA sequence using major Nucleotide Sequence databases

Major Nucleotide Sequence databases are

- European Molecular Biology Laboratory(EMBL)
- GenBank
- DNA databank of Japan (DDBJ)

2.Using BLAST to Compare query sequence (Protein/DNA) to other Sequences

2.1. Open the link at NCBI/BLAST

2.2. Click the Standard Nucleotide-nucleotide BLAST[blastp]

2.4. Paste the saved FASTA-formatted

sequence into the BLAST Searching window as shown in fig b

2.5. Deselect the Do CD-Search box.

2.6. If you use Protein Sequence don’t change the Choose Database setting, because the nr (for non

redundant) is the default protein database

2.7. Click the BLAST! Button

2.8. Click the Format button

2.9. When the results page appear, scroll down the page until you reach along list of sequences and save all these sequences

3.Preparing our session

To compute your multiple sequence alignment, any of the following can be used

- ClustalW :
- Dialign :
- Biology Workbench:

In my work,

Steps to produce multiple sequence using Biology Workbench server

3.1. Open the link at the Biology Workbench

3.2. Start a new session from session tools and run. Give session description

3.3. Session gets created

3.4 Resume session and run

4.Computing the Tree.

Steps involved in a phylogenetic tree

4.1.Open the link at the Biology Workbench

4.2. In the protein tools of our session add on our protein sequences as lisited in the table 1.1

4.3. Select all protein sequences and run ClustalW multiple sequence alignment with required parameters ,then submit which results in phylogenetic tree.

4.4. Click the Run Button and view the tree.

Shown in fig g

Figure 2

Table1.1

1	*ADIPOQ*	AAH54496	244aa
2	*CETP*	AAB59388	425 aa
3	*HTR2C*	CAI41335	458 aa
4	*IAPP*	CAA39504	89 aa
5	*ICAM1*	AAH15969	532 aa
6	*IL6*	CAG29292	212 aa
7	*LEPR*	AAI31780	232 aa
8	*LMNA*	CAI15523	614 aa
9	*MAPK8*	AAI30571	427 aa
10	*PPARG*	AAH06811	477 aa
11	*PPARGC1A*	NP_037393	798 aa
12	*RETN*	AAI01561	108 aa
13	*SELE*	CAI19360	484 aa
14	*SLC2A4*	AAH34387	415 aa
15	*SOCS3*	CAG46495	225 aa
16	*UCP2*	AAC51336	309 aa
17	*RBP4*	CAH72328	201 aa

Figure 3

Fig a

Figure 4

Fig b

Figure 5

Fig c

Figure 6

Fig d

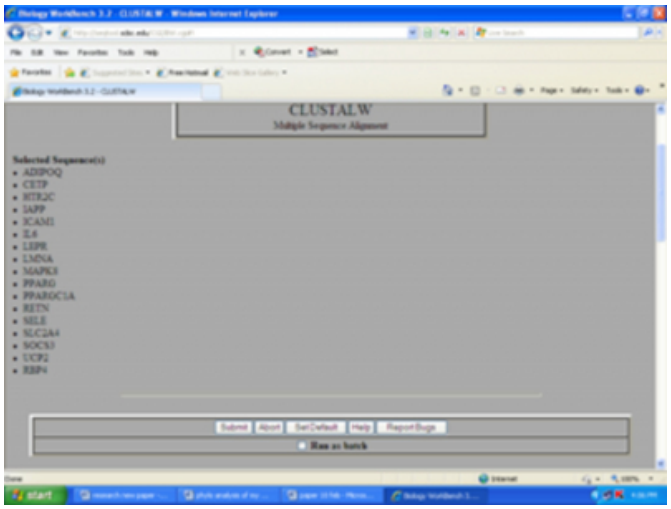


Figure 7

Fig e

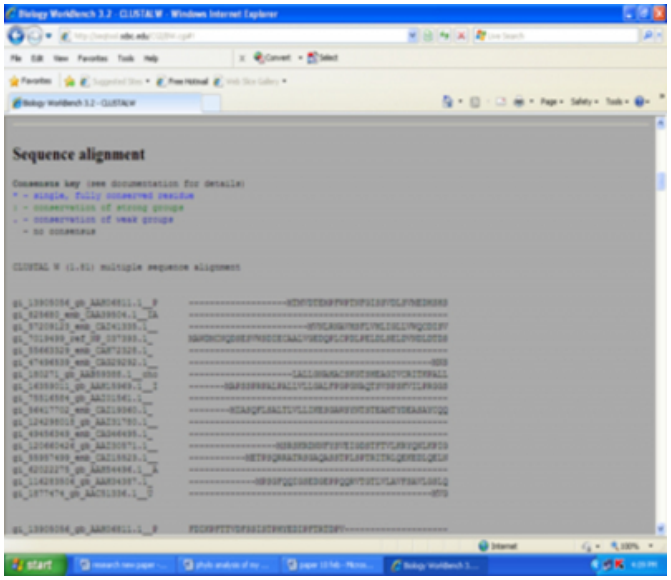


Figure 8

Fig f

Figure 9

Fig g

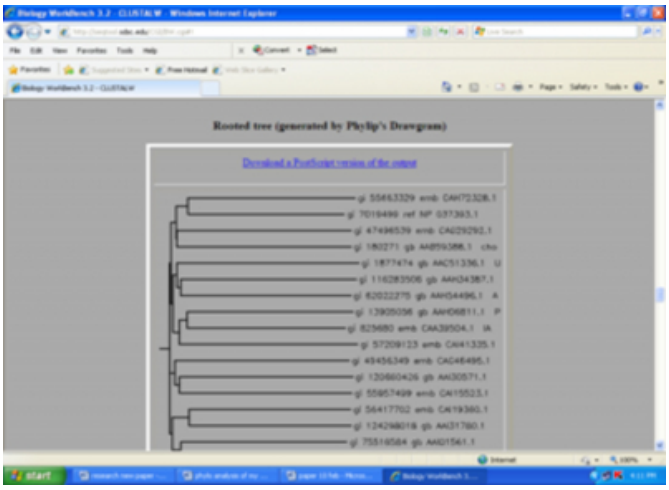


Figure 10

Fig h

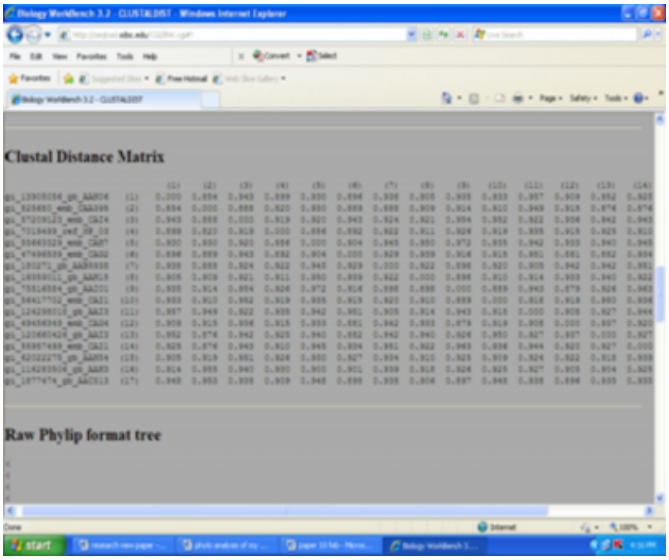


Figure 11

Fig i

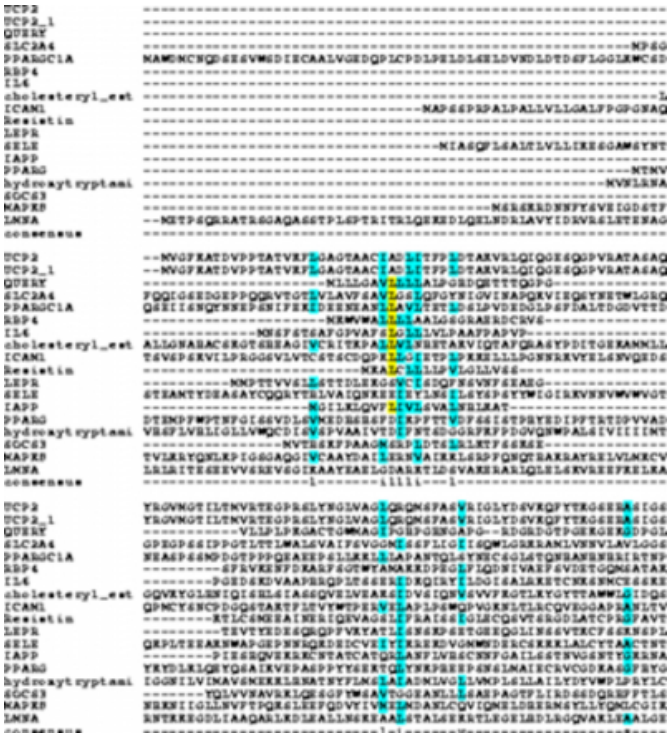


Figure 12

Fig j

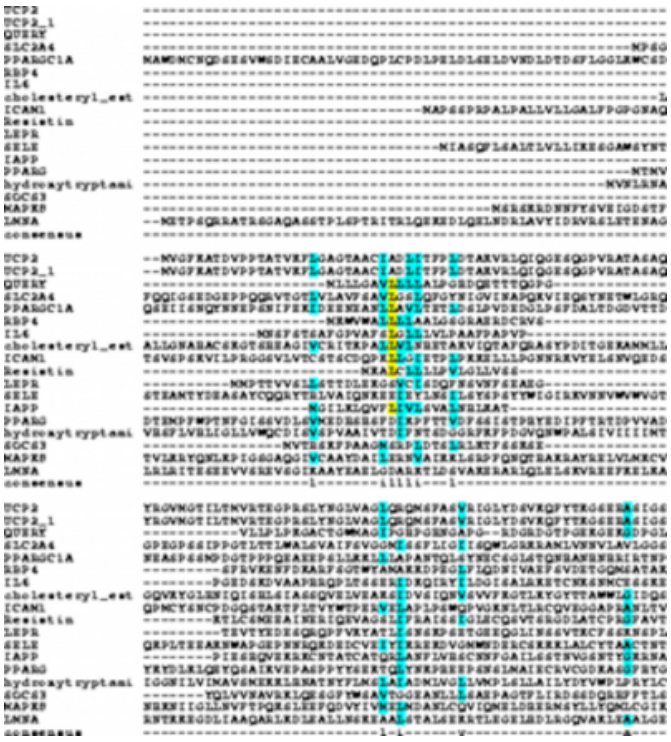


Figure 13

Fig k

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CLUSTAL W (1.81) Multiple Sequence Alignments

Sequence type explicitly set to Protein
Sequence format is Pearson
Sequence 1: gi_55663329_emb_CAH72328.1_ 201 aa
Sequence 2: gi_1877474_gb_AAC51336.1__U 309 aa
Sequence 3: gi_49456349_emb_CAG46495.1_ 225 aa
Sequence 4: gi_116283506_gb_AAH34387.1_ 415 aa
Sequence 5: gi_56417702_emb_CAI19360.1_ 484 aa
Sequence 6: gi_75516584_gb_AAI01561.1__ 108 aa
Sequence 7: gi_7019499_ref_NP_037393.1_ 770 aa
Sequence 8: gi_13905056_gb_AAH06811.1__P477 aa
Sequence 9: gi_120660426_gb_AAI30571.1__427 aa
Sequence 10: gi_55957499_emb_CAI15523.1__614 aa
Sequence 11: gi_124298018_gb_AAI31780.1__232 aa
Sequence 12: gi_47496539_emb_CAG29292.1__ 212 aa
Sequence 13: gi_16359011_gb_AAH15969.1__I 532 aa
Sequence 14: gi_825680_emb_CAA39504.1__IA 89 aa
Sequence 15: gi_57209123_emb_CAI41335.1__ 458 aa
Sequence 16: gi_180271_gb_AAB59388.1__cho 425 aa
Sequence 17: gi_62022275_gb_AAH54496.1__A 244 aa
```

RESULTS AND DISCUSSION

In this paper ,work projected the phylogenteic tree along with the distance matrix result screenshot in fig as well as boxshade results in fig I ,fig j .These results are helpful for further studies of this particular protein analysis in diabesity which aid in drug discovery for treating the patients after conducting the experiments i:e

Invitro methodology

CONCLUSION

For phylogenetic tree construction, the essential performance parameters have been discussed here in detail to provide guidance to computational biologists. In particular, the stepwise discussion strategy is a very realistic and attractive goal, and this methodology, which, if appropriately used, can solve several problems and constitutes a powerful tool in the hands of researchers. The aim of this article, with application to comparison of functional proteins in species and further drug discovery. Phylogenetic tree construction of Resistin has methodology, which, if appropriately used can contribute to a further research towards the Research towards therapeutical aspects.

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References

1. The diabetes control and complications trial research group. The effect of intensive treatment of diabetes on development and progression of long term-complications in insulin dependent mellitus, N Eng Journal of Medical 329 (1993) 977-986.

2. P.H wang, J Lav and T.C Chalmers, meta analysis of effects of intensive blood glucose –control on complications of diabetes, Lancet 341 (1993) 1306-1309.
3. WHO report. Diabetes care and research . The saint Vincent Declation, Diabetic Med.7 (1990),360.
4. E.D lehmann, The diabetes control and complications trail (DCCT)the role of computers in patient education? Diabetic. Natr. Metab. 7(1994) 308-316
5. Kasiska BL, Kalil RS, Majz, Liavm, Keame WF, Effect on antihepertensive therapy on the kidney in patients with diabetes:A meta regression analysis. Ann intern med 118 (1993) 129-138.
6. Carolyn Schwartz, weltchgarry, Kelly Ps Rita Bude, Xiaowusun: computerized adaptive testing im pact in an active clinical population : Qual.lif.Res 15 (2006)1503-1518.
7. Humphreys; individual and social benefits of mutual aid self help groups, Social policy spring(1997)12-19.
8. E.J madara The mutational aid self help on line revolution. Social policy , spring (1997) ,20-26
9. G.A Bogat, Sullivan LA grober J Application of social Supportto preventive interventions ; promoting health and mental health in children (205-232)network ,Springes.
10. K.F Burnet, C.B tailor, W.S Agras Ambulatory computer assisted behavior therapy for obesity . An empirical model for examining behavioral correlates of treatment out come, Computers in human behavior; 8 (1992)239-248
11. Gacken bach J. Psychology and internet ;Interpersonal and trans personal implications (1998) Sandigo CA Acadamic Press.
12. RL street, Gold jr, nannings T Health promotion and interactive technology : Theoritical applications and future directions London, Erbuam
13. King Sa, Moregg D. Internet theraphy and self –help groups- The Pros and Cons(1998)P.p 77-109. Sandigo CA Acadamic press
14. Barrere M.Jr Social Support research in community psychology In Journal report E Seidmen (Eds) Hand book of community Psychology p.p (215-245) Newyork. Plenum.
15. Gottilieb B.H ; Support interventions A typical agenda for research . In S.W dutch ; Hand booh of personnal relation ships p.p 519-541 Newyork Wiley.
16. Hellar K. price RH, Hugg J R The role of Social support in community and clinical interventions ; Social support an international view. Pp 482-507, Network wiley.
17. Hum Phrays K. Individual and social benefits of mutual aid – self helf groups. Social pully spring 12-19
18. D.Z Olds; Kitzman H. Review research on homo visiting of pregnant women and parents of young children ; the future of children 3 (1993)53-92.
19. Finn J Computerbased self help groups online recovery for addictions: Computers in human services 3 (1996) 21-41
20. N.Weinberg; Schmale.J UkenJ ,K.Wessel; computer aided support groups. Social work with groups 17 (1995) 43-54.
21. D.A Salem, G.A Bugat , C.Ried. Mutual help goes online Journal of community Psychology. 25 (1997) 189-207
22. Wein bergn, Schmahe J. Uken J, Wessel K Online help Patients participated in computer mediated support group; Health and social Work 21(1996) 24-29.
23. Winfebery A. The analysis of an Electronic support group for individuals with eating disorders; Computers in human behavior 13 (1997) 393-407.
24. Cohen S psychological models of the role of social support in the etiology physical disease. Health Psychology 7 (1988) 269-297.
25. Seshiah. V., Ganeshan, V.S. Handbook on Diabetes mellitus, (1977); first edition, 1-21, 119-121.
26. King, H., Albert, RE., Herman, W.H., Global burden of diabetes, 1995-2025-prevalence, numerical estimates and projections; Diabetes care, (1998), 21: 1414-31.
27. Allam Appa Rao, S. Vizianand Row, K.Mohini; Analysis of Genes Causing Hypertension, Cardio Vascular and Diabetes Diseases Using Composition Alignment Method; The International Journal of Bio Informatics, Vol 1, Num 2006, (1)pp.78-100;
28. R.Bhramaramba, Allam Appa Rao and GR Sridhar, 2007, 'Principal Component Analysis of protein variates among species affected by Diabetes: An Exploratory Study', International Journal of Diabetes in Developing Countries, ISSN 0973-3930–accepted for publication
29. Appa Rao Allam, Gumpeny Ramachandra Sridhar, Hanuman Thota, Changalasetty Suresh Babu, Akula Siva Prasad and Ch Divakar Alzheimer's disease and Type 2 diabetes mellitus: the cholinesterase connection; Lip. Health. Dise. 2006, 5:28 American Diabetes Association.. Standards of Medical Care in Diabetes. Diabetes Care, 2005, (28)(Suppl 1), S4-36.
30. Allam Appa Rao, Gumpeny R Sridhar, Bolem Srinivas and Undurti N Das, 2007, 'Bioinformatics analysis of functional protein sequences reveals a role for brain-derived neurotrophic factor in obesity and type 2 diabetes mellitus-medical Hypothesis-published online on June 4th 2007
31. Allam A. Rao, Gumpeny R. Sreedhar and Undurti N. Das, 2007, 'Elevated butyryl cholinesterase and acetyl cholinesterase may predict the development of type 2 diabetes mellitus and Alzheimer's disease-Medical Hypothesis published online on June 4th 2007
32. Cassman M (2005) Barriers to progress in systems biology. Nature 438:1079
33. Kitano H (2002) Systems biology: a brief overview. Science 295:1662–1664
34. Gata. B.A. Database Similarity Searching Using BLAST and FastA, Australasian Biotechnology, 5, 282-290, 1995.
35. S. F. Altschul, L. T. Madden, A. A. Schffer, J. Zhang, Z. Zhang, W. Miller, and D. J. Lipman. Gapped blast and psi-blast: a new generation of protein database search programs. Nucleic Acids Research, 25(17):3389–402, 1997.
36. M. Cline, R. Hughey, and K. Karplus. Predicting reliable regions in protein sequence alignments. Bioinformatics,18:306–314, 2002.
37. C. B. Do, S. S. Gross, and S. Batzoglou. Contralign: Discriminative training for protein sequence alignment. InProceedings of the Tenth Annual International Conference on Computational Molecular Biology (RECOMB),2006.
38. C. B. Do, M. S. P. Mahabashyam, and S. Batzoglou. Probcons: Probabilistic consistency-based multiple sequencealignment. Genome Research, 15:330–340, 2005.
39. R. Edgar and K. Sjolander. A comparison of scoring functions for protein sequence profile alignment. BIOINFORMATICS,20(8):1301–1308, 2004.
40. A. Elofsson. A study on protein sequence alignment quality. PROTEINS:Structure, Function and Genetics,46:330–339, 2002.
41. Barceló-Batllori S, Kalko SG, Esteban Y, Moreno S, Carmona MC, Gomis R., Integration of DIGE and bioinformatics analyses reveals a role of the anti-obesity agent tungstate in redox and energy homeostasis pathways in brown adipose tissue., Mol Cell Proteomics. 2007 (in press)
42. Bonadonna RC, Groop L, Kraemer N, Ferrannini E, Del Prato S, DeFronzo RA . Obesity and insulin resistance in humans: a dose-response study. Metabolism1990; 39:452-9.
43. Clara C. Elbers, N. Charlotte Onland-Moret, Lude Franke, Anne G. Niehoff, Yvonne T. van der Schouw and Cisca Wijmenga., A strategy to search for common obesity and type 2diabetesgenes Trends in Endocrinology &

Metabolism, Volume 18, Issue 1, January-February 2007, Pages 19-26
44. Conneely KN, K. Silander, L. J. Scott, K. L. Mohlke, K. N. Lazaridis, T. T. Valle, J. Tuomilehto, R. N. Bergman, R. M. Watanabe, T. A. Buchanan, F. S. Collins, M. Boehnke. Variation in the resistin gene is associated with obesity and insulin-related phenotypes in Finnish subjects. Diabetologia.2004 October; 47(10):1782-1788.
45. DeFronzo RA, Bonadonna RC, Ferrannini E. Pathogenesis of NIDDM. A balanced overview. Diabetes Care 1992; 15:318-68.
46. Fassshauer M, Paschke R. Regulation of adipocytokines and insulin resistance. Diabetologia (2003) 46:1594-1603.
47. Lee JH, Bullen JW Jr, Stoyneva VL, Mantzoros CS (2004) Circulating resistin in lean, obese and insulin-resistant mouse models: lack of association with insulinemia and glycemia. Am J Physiol Endocrinol Metab 288(3):E625-632.
48. List EO, Berryman DE, Palmer AJ, Qiu L, Sankaran S, Kohn DT, Kelder B, Okada S, Kopchick JJ., Analysis of mouse skin reveals proteins that are altered in a diet-induced diabetic state: a new method for detection of type 2 diabetes.,Proteomics.(2007) 7:1140-9.
49. Ochi M, Osawa H, Onuma H, et al. The absence of evidence for major effects of the frequent SNP +299G>A in the resistin gene on susceptibility to insulin resistance syndrome associated with Japanese type 2 diabetes. Diabetes Res Clin Pract 2003; 61:191-198.
50. Rajala MW, Lin Y, Ranalletta M et al. (2002) Cell type-specific expression and co regulation of murine resistin and resistin-like molecule-alpha in adipose tissue. Mol Endocrinol 16:1920-1930.
51. Sangeeta R Kashyap, Ralph A Defronzo. The insulin resistance syndrome: physiological considerations. Diabetes Vasc Dis Res 2007; 4:13-19.
52. Smith SR, Bai F, Charbonneau C, Janderova L, Argyropoulos G. A promoter genotype . and oxidative stress potentially link resistin to human insulin resistance. Diabetes 2003; 52:1611-1618.
53. Steppan CM, Bailey ST, Bhat S, Brown Ej, Banerjee RR, Wright CM et al. The hormone resistin links obesity to diabetes. Nature 2001; 409:307-312.
54. Steppan CM, Brown Ej, Wright CM, Bhat S, Banerjee RR, Dai CY et al. A family of tissue-specific resistin-like molecules. PNAS 2001 98 502-506.
55. Ukkola O. Resistin-a mediator of obesity associated insulin resistance or an innocent bystander? Eur J Endocrinol 2002; 147:571-574.

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