

C948T POLYMORPHISM IN PRE-B CELL COLONY-ENHANCING FACTOR IN TYPE 2 DIABETES: FUNCTIONAL ANALYSIS BY GENE ONTOLOGY

Viroj Wiwanitkit

*Key words: pre-B cell colony-enhancing factor;
gene polymorphism*

SUMMARY

Visfatin or pre-B cell colony-enhancing factor (PBEF) is a pro-inflammatory adipokine expressed predominantly in visceral fat. C948T polymorphism in the PBEF is believed to be associated with the risk of type 2 diabetes. However, this correlation is still a matter of controversy. It is difficult to study functional aberration in the human wild and mutated types of PBEF in the pathogenesis of type 2 diabetes. The author used a new gene ontology technology to predict the molecular function of the human wild and mutated types of PBEF. There was no functional difference between the wild and mutated types of PBEF. This appears to support the null effect of this polymorphism in patient clinical findings. This polymorphism might not be an important factor, having a null effect in the pathogenesis of diabetes mellitus.

*Corresponding author: Viroj Wiwanitkit, MD, Department of Laboratory Medicine, Faculty of Medicine, Chulalongkorn University, Bangkok Thailand 10330
E-mail: wviroj@pioneer.netserv.chula.ac.th*

INTRODUCTION

Visfatin (also known as pre-B cell colony-enhancing factor, or PBEF; NM_005746) is a pro-inflammatory adipokine expressed predominantly in visceral fat (1,2). It is generally accepted that the C948T polymorphism in PBEF might be associated with an increased risk of type 2 diabetes. Increasing concentrations of PBEF are independently and significantly associated with type 2 diabetes (3). However, studies on the association between the polymorphism and clinical findings have yielded some inconsistent findings (4). Bottcher *et al.* found that the C948T variant was associated with 2-h plasma glucose and fasting insulin concentrations. They have suggested that genetic variation in the PBEF gene may have a minor effect on visceral and sc PBEF mRNA expression profiles (4).

However, the role of the C948T change needs a systematic theoretical explanation. To study the functional difference due to gene polymorphism is hard. Fortunately, the new development in bioinformatics can be applied in nanoscale genomics and proteomics research. In this study, the author used a new gene ontology technology to predict the molecular function of human PBEF. Gene ontology is a fundamental knowledge that provides not only standards for annotating and indexing biological

information, but also the basis for implementing functional classification and interpretation models (5). It can be applied to study the expression of biological molecules (5). The aim of this study was to investigate the effect of C948T polymorphism in PBEF on phenotypic expression.

MATERIALS AND METHODS

Getting the sequence and mutation assignment

The Pubmed database was used for data mining of the amino acid sequence for human PBEF. Then the assigned C948T mutation was performed.

Prediction of molecular function and biological process

The author performed prediction of the molecular function and biological process of PBEF in the wild and mutated types using a novel gene ontology prediction tool, GoFigure. GoFigure is a computational algorithm tool which has been recently developed in gene ontology. The tool is available online at <http://udgenome.ags.udel.edu/gofigure/>. The tool accepts an input DNA or protein sequence, and uses BLAST to identify homologous sequences in gene ontology annotated databases. The approach is to use BLAST search to identify homologs in public databases that have been annotated with gene ontology terms. These include SwissProt, Flybase (Drosophila), the Saccharomyces Genome Database (SGD), Mouse Genome Informatics (MGI) and Wormbase (nematode). The contents of the results will show results for molecular function as well as biological process of the studied protein (6).

RESULTS

Sequence of human PBEF

From searching the database, the sequence of human PBEF was derived and the G948T mutation was performed.

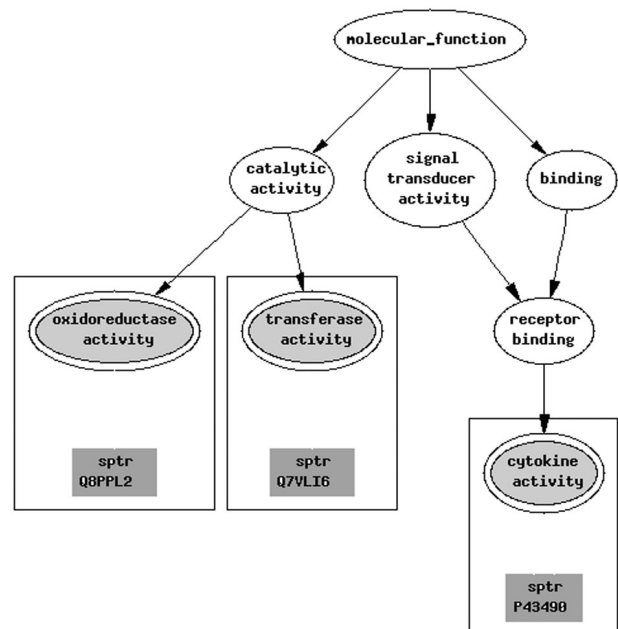
Prediction of molecular function

Using the GoFigure server, molecular functions of the wild and mutated types of PBEF were predicted. The molecular function of the human wild and mutated types of PBEF was found to be the same and is presented in Figure 1. A summary on the molecular function of the human wild and mutated types of PBEF is presented in Table 1.

Table 1. Summary on the molecular function and biological process of human wild and mutated types of pre-B cell colony-enhancing factor

	Wild type	Mutated type
Molecular function	Oxidoreductase activity, transferase activity, cytokine activity	Oxidoreductase activity, transferase activity, cytokine activity
Biological process	Cell-cell signaling, signal transduction, positive regulation of cell proliferation, metabolism	Cell-cell signaling, signal transduction, positive regulation of cell proliferation, metabolism

Figure 1. Expected molecular function of wild and mutated types of pre-B cell colony-enhancing factor.



DISCUSSION

Visceral and subcutaneous adipose tissue displays important metabolic differences that underlie the association of visceral obesity with obesity-related cardiovascular and metabolic alterations (7). PBEF is identified as an adipokine, which is predominantly secreted from visceral adipose tissue in humans (7). PBEF is a cytokine that is highly expressed in visceral fat and its blood levels correlate with obesity (1). This cytokine exhibits insulin-mimetic effects and correlates strongly with visceral obesity (8). It is originally isolated as a secreted factor that promotes the growth of B cell precursors and has recently been found to act as an insulin analog on the insulin receptor; its pathophysiological role in humans remains largely unknown (1,2). Bailey *et al.* have proposed that the polymorphism in the PBEF gene could influence plasma insulin levels (8). C948T polymorphism in the PBEF is believed to be associated with the risk of type 2 diabetes (1). However, there is still a controversy in this correlation. Recently, Pagano *et al.* noted that BCEF was not related to insulin resistance either as assessed by homeostasis model assessment or during lipid infusion (9). Clarification of the exact effect of this polymorphism is needed.

Based on the recent advances in the genomics technology, current microarray technologies permit the examination of gene expression patterns of tens of thousands of genes. One challenge facing the biologist interpreting such data is recognizing the function of many of the hits identified in a single experiment (6). While one can check the literature, a rapid means to get some idea of potential function of a gene product is to obtain the ontology terms that describe the gene (6). The gene ontology is developed for this specific purpose. At present, many gene ontology tools are constructed and launched. In this study, the author used a gene ontology tool to predict the function of the human wild and mutated types of PBEF.

According to this study, there is no functional difference between the wild and mutated types of PBEF. Therefore, there should be no difference in the effect of the wild and mutated types of PBEF in type 2 diabetes. This can support the null effect of this polymorphism on patient clinical findings. This polymorphism might not be an important factor in the pathogenesis of diabetes mellitus. Conclusively, the C948T polymorphism in PBEF in type 2 diabetes might bring no aberration in phenotypic expression.

REFERENCES

1. Zhang YY, Gottardo L, Thompson R, Powers C, Nolan D, Duffy J, Maescotti MC, Avogaro A, Doria A. A visfatin promoter polymorphism is associated with low-grade inflammation and type 2 diabetes. *Obesity (Silver Spring)* 2006;14:2119-2126.
2. Chen MP, Chung FM, Chang DM, Tsai JC, Huang HF, Shin SJ, Lee YJ. Elevated plasma level of visfatin/pre-B cell colony-enhancing factor in patients with type 2 diabetes mellitus. *J Clin Endocrinol Metab* 2006;91:295-299.
3. Sethi JK, Vidal-Puig A. Visfatin: the missing link between intra-abdominal obesity and diabetes? *Trends Mol Med* 2005;11:344-347.
4. Bottcher Y, Teupser D, Enigk B, Berndt J, Kloting N, Schon MR, Thiery J, Bluher M, Stumvoll M, Kovacs P. Genetic variation in the visfatin gene (PBEF1) and its relation to glucose metabolism and fat-depot-specific messenger ribonucleic acid expression in humans. *J Clin Endocrinol Metab* 2006;91:2725-2731.
5. Azuaje F, Al-Shahrour F, Dopazo J. Ontology-driven approaches to analyzing data in functional genomics. *Methods Mol Biol* 2006;316:67-86.
6. Khan S, Situ G, Decker K, Schmidt CJ. GoFigure: automated Gene Ontology annotation. *Bioinformatics* 2003;19:2484-2485.

7. Berndt J, Kloting N, Kralisch S, Kovacs P, Fasshauer M, Schon MR, Stumvoll M, Bluher M. Plasma visfatin concentrations and fat depot-specific mRNA expression in humans. *Diabetes* 2005;54:2911-2916.
8. Bailey SD, Loredó-Osti JC, Lepage P, Faith J, Fontaine J, Desbiens KM, Hudson TJ, Bouchard C, Gaudet D, Perusse L, Vohl MC, Engert JC. Common polymorphisms in the promoter of the visfatin gene (PBEF1) influence plasma insulin levels in a French-Canadian population. *Diabetes* 2006;55:2896-2902.
9. Pagano C, Pilon C, Olivieri M, Mason P, Fabris R, Serra R, Milan G, Rossato M, Federspil G, Vettor R. Reduced plasma visfatin/pre-B cell colony-enhancing factor in obesity is not related to insulin resistance in humans. *J Clin Endocrinol Metab* 2006;91:3165-3170.