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Research Article

# Study of the association of reduced level of adinopectin, vistafin and eslastase with increased risk of diabetes in a group of first degree relatives of middle-aged Pakistani people.

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**Abstract**

**A cross sectional study was designed to observed that the reduced level of plasma adiponectin and visfatin concentrations may be associated with decreased sensitization of insulin and increased risk of diabetes in first degree relatives of diabetics. We have studied 50 subjects (aged between 26-35 year-old) who had a family history of diabetes during 2010-2011. 20 age matched control subjects without a family history of diabetes were also taken. A questionnaire on demographics, history, clinical examination and laboratory tests was filled out for each patient. Plasma glucose was measured using the GOD-PAP enzymatic method. Level of insulin was measured by ELIZA technique. Raw volume of adinopectin, visgatin and elastase was estimated by 12% SDS polyacrylamide gel electrophoresis. Their BMI and insulin resistance was calculated. Results showed that mean age of first degree relatives was 29.67 year. Their BMI was non significantly greater than their controls. Level of fasting blood sugar, serum insulin and insulin resistance were increased as compared to their controls but significant difference was only observed in case of level of serum insulin. Raw volume of adinopectin, visfatin and elastase was non significantly decreased in FDR as compared to their controls. It is concluded that diabetes screening including estimation of visfatin, adinopectin and elastase is an effective means of detecting unknown cases of diabetes among FDR, improving glycemic control and mitigating CVD risk factors.**

**Keywords**: First degree relatives, diabetes, visfatin, elastase, adinopectin.

## INTRODUCTION

A family history of diabetes is a major risk factor for the disease of diabetes. First-degree relatives of NIDDM patients have an approximately 40% lifetime risk of developing diabetes. The prevalence of type II diabetes in people varies considerably depending on the country of habitat (Osei et al., 1993). First-degree relatives (FDRs) of patients with type2 diabetes develop diabetes at a greater rate than the normal population. (Warram et al. 1990) and have been used to uncover early abnormalities that may be important in the development of type 2 diabetes.

Researchers have developed a variety of simple tools to identify high-risk individuals for diabetes in populations. Diabetic individuals with BMI >25 may be at increased risk for underlying exocrine pancreatic insufficiency (Nunes et al. 2003). There seems to be a connection between the loss of exocrine function and the increasing body mass index. However, the BMI is an additional factor for lowered level of elastase, adinopectin and visfatin (Cnop et al. 2003;

Teichmann et al.2011). Evidence shows that the detection of impaired glucose metabolism in its early stages or prediabetes stage could lead to the delay or prevention of the disease and its complications Valdez 2009.

Insulin resistance is the best predictor of developing diabetes. However, insulin resistance is altered by many other factors, including age, diet, exercise, and medications (Perseghin et al. 1997). Insulin resistance, the central metabolic disturbance associated with fat accumulation, endocrine and pro-inflammatory alterations. During the last decade it is observed there may be an endocrine source of active modulators of insulin sensitivity such as adiponectin,visfatin, resistin and cytokines Paula FJ and Rosen CJ (2010).

Adiponectin, A 30-kDa protein is an adipocyte-derived polypeptide that has insulin sensitizing properties and is abundantly present in peripheral circulationWeyer et al. (2001); Chandran et al. (2003). It is inversely correlated with body mass index (BMI), intraabdominal fat, and indices of insulin resistanceArita et al (1999); Cnop et al. 2003. Low plasma adiponectin has been identified as a risk factor for type 2 diabetes. Results from animal study suggest that adiponectin is likely to play an important role in regulating insulin actionCnop et al. 2003. It is reportedHotta et al (2003), that a decline in adiponectin concentration coincides with the development of hyperinsulinemia and insulin resistance.

Visfatin a 52 Kda protein has been identified as a novel adipocytokine that is up-regulated in visceral fat in parallel with insulin resistanceAdeghate (2008). Visfatin was further hypothesized to bind directly to the insulin receptor and to exert insulin-like effects in vivo and in vitro. The effects of visfatin on adipogenesis and glucose metabolismFukuhara et al. (2005) are of particular interest with respect to a putative role in the pathogenesis of obesity and diabetes. Some subsequent clinical studies confirmed the association of visfatin and diabetesChen et al. (2006), whereas others did not find an associationJian et al (2006). Similarly, data regarding the relationship of visfatin with parameters of glucose metabolism and insulin resistance were contradictoryDogru et al. (2007); Berndt et al (2005), and over-all, there was no clear effect of visfatin on metabolism.

Elastase is a serine protease with a molecular weight of 25 Kda hydrolyzes amides and esters. It is produced in the pancreas as an inactive zymogen, and activated in the duodenum by trypsin. Despite the high functional reserve of the pancreas, it is recognized that a significant proportion of diabetic patients may also have a deficit of the exocrine functionNunes et al., 2003. It was observed that pancreatic volume was decreased in patients with low elastase-1 concentrationPhilippe et al. (2011). Decreased function of the exocrine pancreas is frequent in patients with diabetes and associated with low level of elastase and chymotrypsinLarger et al., (2012).

Diabetes screening is an effective tool for diagnosing patients who are unaware of their diabetes and for providing them with optimal treatmentAmini et al (2008). Glycemic control and screening for potential diabetic complications in patients can prevent microvascular complications of diabetesOhkubo et al.(1995). Existing evidence indicates that glycemic control can slow down the progression of micro vascular and macro vascular complications in diabetic patientsStettler et al., (2006).

We hypothesized that lower plasma adiponectin and visfatin concentrations would be associated with decreased sensitization of insulin and increased risk of diabetes in first degree relatives of diabetics. This study may provide a tool to evaluate the efficacy of diabetes screening.

## Patients and methods

We have studied 50 subjects (aged 35-55 year-old) who had a family history of diabetes during 2010-2011. Of these, mostly had one i.e. mother only or father with type 2 diabetes. In addition, we have studied 20 age matched control subjects without a family history of diabetes. . Exclusion criteria were as follows: age >55 yr; intake of orlistat or acarbose; and history of diarrhea, pancreatitis, GI surgery, immunodeficiency, or cancer. A questionnaire on demographics, history, clinical examination and laboratory tests was filled out for each patient. Informed consent was obtained from all participants.

All patients underwent the same study protocol, which included clinical evaluation, determination of level of fasting blood glucose, insulin, adinopectin, visfatin and elastase. Body mass index (BMI) of patients was calculated as weight (kg) divided by the square of height (m). Homeostasis model assessment (HOMA) index was calculated from the product of glucose (mmol/l) and insulin [(µUl/l)/22.5] and used as an indicator of insulin resistancePelikánová et al., (1994).Three milliliters of blood were collected from all participants to measure fasting plasma glucose, insulin, adiponectin, visfatin and elastase. Plasma glucose was measured using the GOD-PAP enzymatic method. Level of insulin was measured by ELIZA technique. Raw volume of adinopectin, visfatin and elastase was estimated by 12% SDS polyacrylamide gel electrophoresis.

Statistical Analysis:

Data were expressed as the mean (SD), unless stated otherwise. Statistical analysis was performed using the *t*-test for quantitative and chi-square test for qualitative variables using SPSS version 115. We considered p-values less than 0.05 to be statistically significant.

Figure1 : 12% SDS gel analysis showed the 52 Kda band of visfatin and 30Kda band of adinopectin

Mean age of first degree relatives was 29.67 year. Their BMI was non significantly greater than their controls. Level of fasting blood sugar, serum insulin and insulin resistance were increased as compared to their controls but significant difference (P<0.001) was only observed in case of level of serum insulin. Raw volume of adinopectin, visfatin and elastase was non significantly decreased in FDR as compared to their controls (Table 1 and Figure1).

**Table1.** Clinical characteristics of first-degree relatives of type 2 diabetes patients and controls.

| Characteristics | Subjects (50) | Controls (20) |
|---|---|---|
| Age (years) | 29.67±3.88 | 32.53±5.8 |
| BMI | 25.78±6.05 | 26.10±2.60 |
| Fasting blood sugar | 88.75±12.81 | 80.75±9.5 |
| Serum insulin (u IU/ml) | 14.16±13.79** | 5.19±5.66 |
| Insulin resistance | 3.1 | 1.03 |
| Raw volume of visfatin | 40641.86±17895.82 | 65374.31±23073.8 |
| Raw volume of Adiponectin | 137.64±135.33 | 155.04±132.22 |
| Raw volume of Elastase | 24984.80±17082.08 | 32937.39±6028.44 |

*p<0.05= Significant difference,
**p<0.001= Highly significant differenc

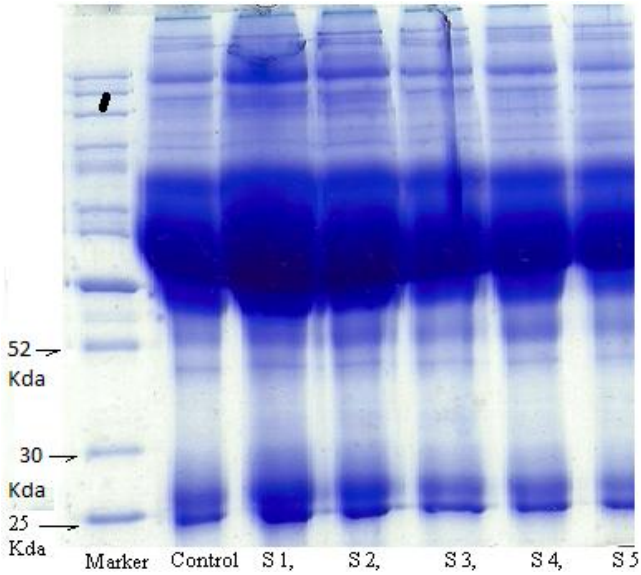


**Figure1.** 12%SDS polyacrylamide gel analysis showed peptide band of 52Kda Visfatin, 30Kda Adinopectin and 25Kda Elastase . (Legend S represent Patient's sample).

## DISCUSSION

Diabetes mellitus type II (insulin-nondependent diabetes) is a disease conditioned by a dysbalance between insulin secretion and effect; it has not been decided whether the cause is insulin resistance or impaired insulin secretion, although a defect of insulin secretion for the manifestation of the disease is generally acceptedBeaty et al., (1982).

Mean age of first degree relatives was 29.67 year. Their BMI was non significantly greater than their controls. According to a study the general risk factors, age andobesity, were important in predicting diabetes at initial visit,although the predicted risk curves were very different for malesand females, Amini and Janghorbani (2007); Vaag et al (2001). A study also reported that the BMI was slightly higher in subjects with positive family history as compared to

subject with no family history of diabetesNyholm et al., (2004). Another study stated that visceral obesity is closely linked to both insulin resistance and type 2 diabetes, Janghorbani and Amini (2009).

We observed that an impaired fasting blood sugar FDR as compared to their controls. It is found that impaired fasting glucose showed the progression rate of diabetes in participants are highAmoah et al., (2001). It is reported that FDR of people with type 2 diabetes are at higher risk of IGT and type 2 diabetes than the population at large, Vaag et al., (2001). A study stated that plasma glucose levels are directly implicated in the development of diabetic complications, Lindsay et al., (2002)

Present study observed that serum insulin and insulin resistance were increased as compared to their controls but significant difference ($P<0.001$) was only observed in case of level of serum insulin. A study demonstrates that hyperinsulinemia and a tendency to lower insulin sensitivity (insulin resistance), but not altered glucose effectiveness, are found in healthy non-diabetic, first-degree relatives of patients with type 2 diabetes as compared to healthy subjects. Study concludes that genetic factors could play a significant role in the development of type 2 diabetes, Spranger et al (2003). Another study found that Insulin resistance is present in non-diabetic relatives of Type 2 diabetic patients. The insulin resistance is independent of degree of obesity and is restricted solely to the pathway of non-oxidative glucose metabolismNyholm et al (2004).

We observed by SDS gel electrophoresis that raw volume of adinopectin and visfatin was non significantly decreased in FDR as compared to their controls. Our study in inline with studies who observed plasma adiponectin concentrations were statistically significantly reduced in patients with type 2 diabetes as compared with controls, Hotta et al (2003); Sandeep et al, (2007). This observation was corroborated by a group of workers, Fernandez-Real et al (2007), who suggest that adiponectin is likely to play a role in the pathogenesis of insulin resistance and type 2 diabetes.

Like adinopectin, the level of visfatin was also reduced in FDR as compared to their controls. A study does not support the hypothesis that visfatin is increased in visceral obesity in parallel with insulin resistance and impaired glucose tolerance Fukuhara et al (2005) although some studies did show associations with diabetesChen et al. (2006)Mancilla et al (2006).

Present study observed a non significant decrease of elastase in FDR of diabetics when compared with control. A study found that there was a significant association between low elastase levels and development of diabetes. Other study reported that low elastase level may be a good predictor of failure of pancreatic exocrine functionTeichmann et al., 2011.

In conclusion, our results reveal inconsistencies and limitations in the detection of serum visfatin, adinopectin and elastase levels by SDS analysis. However it is concluded that diabetes screening is an effective means of detecting unknown cases of diabetes, improving glycemic control and mitigating CVD risk factors.

**References**


Adeghate E(2008).Visfatin: structure, function and relation to diabetes mellitus and other dysfunctions. Curr Med Chem.15(18):1851-62.

Amini M, Janghorbani M (2007). Diabetes and Impaired Glucose Regulation in First-Degree Relatives of Patients with Type 2 Diabetes in Isfahan, Iran: Prevalence and Risk Factors Rev Diabet Stud, 4(3):169-176

Amini M, Timori A, Aminorroava A (2008). Quality of Care for First-Degree Relatives of Type 2 Diabetes Patients Diagnosed with Diabetes at a Screening Program One Year After DiagnosisRev Diabet Stud, 5(1):52-58

Amoah AG, Owusu SK, Ayittey OM, Schuster DP, Osei K (2001). Minimal model analyses of beta cell secretion, insulin sensitivity and glucose effectiveness in glucose tolerant, non-diabetic first-degree relatives of Ghanaian patients with type 2 diabetes and healthy control subjects. Ethn Dis.11(2):201-10

Arita Y, Kihara S, Ouchi N, Takashashi M, Maeda K, Miyagawa J, Hotta K, Shimoura I, Nakamura, T, Miyaoka K, Kuriyama H, Nishida M, Yamashita S, Okubo K, Matsabura K, Muraguchi M, Ohmoto Y, Funahashi T, Matsuzawa Y (1999). Paradoxical decrease of an adipose-specific protein, adiponectin,in obesity. Biochem Biophys Res Commun, 257:79 – 83

Beaty TH, Neel JV, Fajans SS (1982). Identifying risk factors for diabetes in first degree relatives of non-insulin dependent diabetic patients. Am J Epidemiol. Mar; 115(3):380–397.

Berndt J, Kloting N, Kralisch S, Kovacs P, Fasshauer M, Schon MR, Stumvoll M, Bluher M(2005). Plasma visfatin concentrations and fat depot-specific mRNA expression in humans. Diabetes,54:2911–2916

Chandran M, Phillips SA, Ciaraldi T, Henry RR (2003). Adiponectin: more than just another fat hormone? Diabetes Care, 26:2442–2450

Chen MP, Chung FM, Chang DM, Tsai JC, Huang HF, Shin SJ, Lee YJ (2006). Elevated plasma level of visfatin/pre-B cell colony-enhancing factor in patients with type 2 diabetes mellitus. J Clin Endocrinol Metab. 91:295–299

Cnop M, Havel PJ, Utzschneider KM, Carr DB, Sinha MK, Boyko EJ, Retzlaff BM, Knopp RH, Brunzell JD, Kahn SE (2003). Relationship of adiponectin to body fat distribution, insulin sensitivity and plasma lipoproteins: evidence for independent roles of age and sex. Diabetologia.46:459 – 469

Dogru T, Sonmez A, Tasci I, Bozoglu E, Yilmaz MI, Genc H et al. (2007). Plasma visfatin levels in patients with newly diagnosed and untreated type 2 diabetes mellitus and impaired glucose tolerance. Diabetes Res Clin Pract. 76:24 –29

Fernandez-Real JM, Moreno JM, Chico B, Lopez-Bermejo A, Ricart W (2007). Circulating visfatin is associated with parameters of iron metabolism in subjects with altered glucose tolerance. Diabetes Care. 30:616 – 621

Fukuhara A, Matsuda M, Nishizawa M, Segawa K, Tanaka M, Kishimoto K et al (2005). Visfatin: a protein secreted by visceral fat that mimics the effects of insulin. Science 307:426 – 430

Hotta K, Funahashi T, Bodkin NL, Ortmeyer HK, Arita Y, Hansen BC, Matsuzawa Y (2003).Circulating concentrations of the adipocyte protein adiponectin are decreased in parallel with reduced insulin sensitivity during the progression to type 2 diabetes in rhesus monkeys. Diabetes,50:1126 –1133

Janghorbani M, Amini M (2009). Progression to impaired glucose metabolism in first-degree relatives of patients with type 2 diabetes in Isfahan, Iran.Diabetes/metabolism research and reviews. 25(8):748-55.

Jian WX, Luo TH, Gu YY, Zhang HL, Zheng S, Dai M et al(2006). The visfatin gene is associated with glucose and lipid metabolism in a Chinese population. Diabet Med. 23: 967-973, 2006.

Larger E, Philippe MF, Barbot-Trystram L, Radu A, Rotariu M, Nobécourt E, Boitard C(2012). Pancreatic exocrine function in patients with diabetes. Diabet Med.Aug;29(8):1047-54.

Lindsay RS, Funahashi T, Hanson RL, Matsuzawa Y, Tanaka S, Tataranni PA, Knowler WC, Krakoff J (2002). Adiponectin and development of type 2 diabetes in the Pima Indian population. Lancet. 360:57–58

Maeda N, Takahashi M, Funahashi T, Kihara S, Nishizawa H, Kishida K (2001). PPAR- ligands increase expression and plasma concentrations of adiponectin, an adipose-derived protein. Diabetes,50:2094 –2099

Mancilla A C, Hurtado H C, Tobar A E, Orellana N I, Pineda B P, Castillo M I, Ledezma RR, Berger FZ(2006). [Pancreatic exocrine function in diabetes mellitus. Determination of fecal elastase]. Rev Med Chil.134(4):407-14.

Nunes AC, Pontes JM, Rosa A, Gomes L, Carvalheiro M, Freitas D(2003). Screening for pancreatic exocrine insufficiency in patients with diabetes mellitus. Am J Gastroenterol.98(12):2672-5.

Nyholm B, Nielsen MF, Kristensen K, Nielsen S, Østergård T, Pedersen SB (2004). Evidence of increased visceral obesity and reduced physical fitness in healthy insulin-resistant first-degree relatives of type 2 diabetic patients. . Eur J Endocrinol. Feb;150(2):207-14.

Ohkubo Y, Kishikawa H, Araki E, Miyata T, Isami S, Motoyoshi S, Kojima Y, Furuyoshi N, Shichiri M (1995).Intensive insulin therapy prevents the progression of diabetic microvascular complications in Japanese patients with non-insulin-dependent diabetes mellitus: a randomized prospective 6-year study. Diabetes Res. Clin. Prac. 28:103-117.

Osei K, Cottrell DA, Adenuwon CA, Ezenwaka EC, Akanji AO, O'Dorisio TM. 1993. Serum insulin and glucose concentrations in people at risk for type II diabetes. A comparative study of African Americans and Nigerians. Diabetes Care. 16(10):1367-75.

Paula FJ, Rosen CJ (2010). Obesity, diabetes mellitus and last but not least, osteoporosis. Arq Bras Endocrinol Metabol.54(2):150-7.

Pelikánová T, Válek J, Kazdová L, Saudek F, Karasová L (1994). [Insulin resistance and insulin secretion in type II non-obese diabetics]. Cas Lek Cesk.21;133(6):172-6.

Perseghin G, Ghosh S, Gerow K, Shulman GI (1997). Metabolic defects in lean nondiabetic offspring of NIDDM parents: a cross-sectional study. Diabetes. Jun., 46(6):1001-9.

Philippe MF, Benabadji S, Barbot-Trystram L, Vadrot D, Boitard C, Larger E (2011). Pancreatic volume and endocrine and exocrine functions in patients with diabetes. Pancreas. Apr;40(3):359-63.

Sandeep S, Velmurugan K, Deepa R, Mohan V (2007). Serum visfatin in relationto visceral fat, obesity, and type 2 diabetes mellitus in Asian Indians. Metabolism, 56:565–570

Spranger J, Kroke A, Mohlig M, Bergmann MM, Ristow M, Boeing H, Pfeiffer AFH(2003). Adiponectin and protection against type 2 diabetes mellitus. Lancet, 361:226 –228

Stettler C, Allemann S, Juni P, Cull CA, Holman RR, Egger M, Krahenbuhl S, Diem P(2006). Glycemic control and macrovascular disease in types 1 and 2 diabetes mellitus: meta-analysis of randomized trials. Am Heart J.152:27-38

Teichmann J, Riemann JF, Lange U(2011). Prevalence of exocrine pancreatic insufficiency in women with obesity syndrome: assessment by pancreatic fecal elastase 1. *ISRN Gastroenterol.* 2011:951686.

Vaag A, Lehtovirta M, Thye-Rönn P, Groop L (2001). European Group of Insulin Resistance. Metabolic impact of a family history of Type 2 diabetes. Results from a European multicentre study (EGIR). Diabet Med.18(7):533-40.

Valdez R (2009). Detecting Undiagnosed Type 2 Diabetes: Family History as a Risk Factor and Screening Tool. *J Diabetes Sci Technol,* 3(4):722-726

Warram JH, Martin BC, Krolewski AS, Soeldner JS, Kahn CR (1990). Slow glucose removal rate and hyperinsulinemia precede the development of type 2 diabetes in the offspring of diabetic patients. Ann Intern Med. 113:909–915.

Weyer C, Funahashi T, Tanaka S, Hotta K, Matsuzawa Y, Pratley RE, Tataranni PA (2001). Hypoadiponectinemia in obesity and type 2 diabetes: close association with insulin resistance and hyperinsulinemia. J. Clin Endocrinol Metab. 86:1930 –1935