

# **Adiponectin Gene Polymorphism SNP +45 T/G in Type 2 Diabetic Patients in Assam, India**

Morteza Kordafshari<sup>1\*</sup>, Marzieh Kordafsharin<sup>2</sup>, Manab Deka<sup>3</sup>, Majid Khandouzi<sup>4</sup>, Priyanka Kashyap<sup>5</sup>, Namrata Kumari<sup>6</sup>

<sup>1</sup> Corresponding Author: ResearchScholar in Dept of Bioengineering and Technology, Guahati University, Guwahati, Assam.

<sup>2</sup> Master of Science in Biochemistry, Osmania University, Hyderabad, Telangana, India.

<sup>3</sup> Professor & Head, Dept of Bioengineering and Technology, Gauhati University.

<sup>4,5,6</sup> ResearchScholar Scholar, Dept of Bioengineering and Technology, Gauhati University, Guwahati.

**Abstract— Background:** Adiponectin SNPs gene have been associated with BMI, insulin sensitivity and type 2 diabetes, reportedly. In this Case study we investigate Adiponectin SNP gene +45\*T/G (rs2242766) in type 2 diabetes and Healthy control in Northeast India. **Methods:** Sample collected from Diabetic patients that visited the diabetes clinic. healthy control group were Volunteers students of Gauhati University. PCR-RFLP method. Is used to analysis the Adiponectin gene polymorphism. obese diabetic patients (n: 80), non-obese diabetic patients (n:72). Healthy controls group (n:70). **Results:** Frequency of wild genotype (TT) was 62.5% in non-obese diabetic Cases and 78% in control group, that was significant (P=0.02). The allele T and G frequencies were significant in non-obese diabetic. The frequency of allele G in non-obese diabetic Cases was increased (20.1%) in compared to controls (12%) (P=0.04) **Conclusion:** This Cases study we found TG and GG alleles of Adiponectin SNP +45\*T/G are risk factors for development of diabetes mellitus. This effect is independent from BMI and obesity.

**Keywords—** SNP, Adiponectin, Diabetes mellitus, PCR-RFLP, Frequency

## **I. INTRODUCTION**

Adiponectin is an adipocyte-derived hormone and known to form several species of multimer, however, the precise components of each multimer have not been fully determined[1]. The adipose tissue derived protein adiponectin exerts anti-diabetic, anti-inflammatory and anti-atherosclerotic effects[2]. Adiponectin gene polymorphisms are associated with serum levels of adiponectin, body mass index (bmi), insulin sensitivity and type 2 diabetes. In a prospective study over a 3-year follow-up period in subjects with normal glucose levels shown that there is a relationship between adiponectin gene polymorphism and susceptibility to have hyperglycemia [3]. This study has taken place to investigate the prevalence of adiponectin gene polymorphism at position 45 t / g in patients with type 2 diabetes in an Indian population and analysis of the allele frequency and genotype polymorphism in type 2 diabetic patients with normal bmi in compared with enhanced bmi.

## **II. METHODOLOGY**

non-diabetic controls were selected from Guwahati, which has an average age of 25-64 years (n = 80). Patients with type 2 diabetes from the Diabetes Clinic were selected based on the following criteria: presence of glucose 126 mg / dl <FBS or plasma glucose> 200, which is confirmed in two separate experiments, and the insulin dependent diabetics number patients were 168. BMI was calculated based on height and weight. The subjects blood samples in tubes containing EDTA at a rate of 5 L was taken and DNA was extracted. From all patients and healthy subjects, written consent was obtained for sampling. Molecular analysis of the adiponectin gene : To determine the polymorphism+45 T/G in exon 2 with PCR - RFLP the following primers were used[4]. Forward primer (5'-GCAGCTCCTAGAAGTAGACTCTGCTG-3 ') and Reverse primer :(5'-GCAGGTCTGTGATGAAAGAGGCC-3 '). The product was 372-bp fragment that was amplified by PCR. Then product was digested for 16 hours in the presence of enzyme SmaI. After the incubation period, the products were electrophoresed on 3% agarose gel. bands was observed included: Individuals genotype TT (wild form) had a band 372bp, three bands (163bp, 209bp and 372bp) that indicating heterozygous TG and two bands (163bp and 209 bp) represents homozygous GG.

## **III. STATISTICAL ANALYSIS**

Significant differences between groups for different sets of alleles or genotypes of adiponectin and their ratio (or) and confidence interval (CI) 95% was estimated. Levels of significance with Chi-square test and Fisher calculated. all statistical methods were

## International Journal for Research in Applied Science & Engineering Technology (IJRASET)

performed using SPSS software .

### IV. RESULTS

In this study the frequency of polymorphism +45 T / G adiponectin in 70 obese and 72 normal subjects and 80 patients with diabetic non-obese diabetic patients were studied. Clinical characteristics of the study population are shown in Table 1. Statistical analysis to examine the relationship between genotype TT vs. GG + TG showed that 62.5% of the diabetic population and 78% of controls had TT genotype., the difference was statistically significant. (TT vs TG + GG, P = 0.02).allele T (79.9%) versus G (20.1%) in the diabetic group compared to control group T (87.9%) and G (12.1%) was statistically significant . (p = 0.04) .There was no significant difference in the genotype and allele between patients groups (obese and diabetic obese) and control group (Table 2).

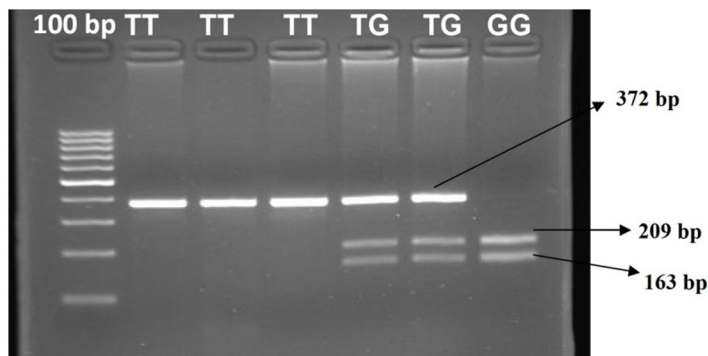
Table 1 - Clinical characteristics of type 2 diabetic patients and control subjects

	Control	Non-obese diabetic	Obese diabetic
Number (female / male)	80(45/55)	88(49/39)	80(66/14)
Age (years)	35±11	52±11	51±10
BMI (kg/m <sup>2</sup> )	24.8±3	26±3	35±4
Height (cm)	164.1 ± 7.5	159± 3.2	158± 6.2
Weight (kg)	50.1± 8.6	49.1± 3.1	77.4± 14.0

Table 2 - Genotype and allele frequencies of the adiponectin gene in type 2 diabetic patients and healthy subjects

	Control (N=70)	Non-obese diabetic (N=72)	Obese diabetic (N=80)
<b>Genotype</b>			
TT	55 (78.6%)	45 (62.5%)*	55 (68.8%)
TG	13 (18.6%)	25 (34.7%)	23 (28.9%)
GG	2 (2.9%)	2 (2.8%)	2 (2.5%)
<b>allele (2N)</b>			
T	123 (87.9%)**	115 (79.9%)**	133 (38.1%)
G	17 (12.1%)	29 (20.1%)	27 (16.9%)

\*(TT VS TG +GG) , p= 0.02) , \*\*P= 0.04 , OR =1.8 , CI (0.9 – 3.7)



**Figure 1:** Adiponectin polymorphisms +45T/G: SmaI restriction fragments. Lanes no. 1 & 2 & 3 were homozygous wild type (genotyped TT) (372 bp), lanes no. 4 & 5 were heterozygous mutant (TG) (372 bp, 209 bp, 163 bp) and lanes no. 6 was homozygous mutant (GG) (209 bp, 163 bp). 100 bp DNA ladder.

### V. DISCUSSION

The purpose of this study was to investigate the genotype SNP +45 \* T / G adiponectin in Assam , India. In studies conducted in different populations, Relation between adiponectin polymorphisms with type 2 diabetes, obesity, and insulin resistance and

## International Journal for Research in Applied Science & Engineering Technology (IJRASET)

cardiovascular is shown. In the Study conducted in Sweden, Ma J et al, The role of polymorphisms +45 \* T / G is shown in type 1 diabetes [4]. In another study in Japan [5], France [6], Italy [7], Sweden [8] found similar results in relation to adiponectin polymorphisms and diabetes. But the Inconsistent results showed that there is no strong correlation. This study showed that TG and GG versus TT genotype increase the chances of developing diabetes independent of obesity with high BMI. To determine the precise correlation of the role of adiponectin polymorphisms in type 2 diabetes studies with the larger sample size needs to be done.

### REFERENCES

- [1] Hada Y, Yamauchi T, Waki H, Tsuchida A, Hara K, Yago H et al. Selective purification and characterization of adiponectin multimer species from human plasma. *Biochemical and Biophysical Research Communications*. 2007;356(2):487-493.
- [2] Heiker J, Klötting N, Blüher M, Beck-Sickingher A. Access to gram scale amounts of functional globular adiponectin from E. coli inclusion bodies by alkaline-shock solubilization. *Biochemical and Biophysical Research Communications*. 2010;398(1):32-37
- [3] Fumeron F, Aubert R, Siddiq A, Betoulle D, Pean F, Hadjadj S et al. Adiponectin Gene Polymorphisms and Adiponectin Levels Are Independently Associated With the Development of Hyperglycemia During a 3-Year Period: The Epidemiologic Data on the Insulin Resistance Syndrome Prospective Study. *Diabetes*. 2004;53(4):1150-1157.
- [4] Schaffler A, Barth N, Palitzsch K, Drobnik W, Scholmerich J, Schmitz G. Mutation analysis of the human adipocyte-specific apM-1 gene. *Eur J Clin Invest*. 2000;30(10):879-887.
- [5] Ma J, Möllsten A, Falhammar H, Brismar K, Dahlquist G, Efendic S et al. Genetic association analysis of the adiponectin polymorphisms in type 1 diabetes with and without diabetic nephropathy. *Journal of Diabetes and its Complications*. 2007;21(1):28-33.
- [6] Hara K, Boutin P, Mori Y, Tobe K, Dina C, Yasuda K et al. Genetic Variation in the Gene Encoding Adiponectin Is Associated With an Increased Risk of Type 2 Diabetes in the Japanese Population. *Diabetes*. 2002;51(2):536-540.
- [7] Gibson F, Froguel P. Genetics of the APM1 Locus and Its Contribution to Type 2 Diabetes Susceptibility in French Caucasians. *Diabetes*. 2004;53(11):2977-2983.
- [8] Menzaghi C. Multigenic control of serum adiponectin levels: evidence for a role of the APM1 gene and a locus on 14q13. *Physiological Genomics*. 2004;19(2):170-174.
- [9] Gu H, Abulaiti A, Ostenson C, Humphreys K, Wahlestedt C, Brookes A et al. Single Nucleotide Polymorphisms in the Proximal Promoter Region of the Adiponectin (APM1) Gene Are Associated With Type 2 Diabetes in Swedish Caucasians. *Diabetes*. 2004;53(Supplement 1):S31-S35.
- [10] Schäffler A, Herfarth H, Paul G, Ehling A, Müller-Ladner U, Schölmerich J et al. Identification of Influencing Variables on Adiponectin Serum Levels in Diabetes Mellitus Type 1 and Type 2. *Exp Clin Endocrinol Diabetes*. 2004;112(7):383-389.
- [11] Kadowaki T. Adiponectin and adiponectin receptors in insulin resistance, diabetes, and the metabolic syndrome. *Journal of Clinical Investigation*. 2006;116(7):1784-1792.
- [12] Weyer C, Funahashi T, Tanaka S, Hotta K, Matsuzawa Y, Pratley R et al. Hypoadiponectinemia in Obesity and Type 2 Diabetes: Close Association with Insulin Resistance and Hyperinsulinemia. *The Journal of Clinical Endocrinology & Metabolism*. 2001;86(5):1930-1935.