ISSN 0975-6299

Vol 2/Issue 2/Apr-Jun 2011

International Journal of Pharma and Bio Sciences

RESEARCH ARTICLE

BIO CHEMISTRY

ASSOCIATION OF TNF-A WITH OBESITY IN TYPE2- DIABETES MELLITUS

D.RAJARAJESWARI*[1], K.RAMALINGAM[2], M.KRISHNAMMA[1] AND T.SHARMILA KRISHNA[1].

[1]Department of Biochemistry, Narayana Medical College and Hospital, Nellore, A.P., India.
[2]Department of central laboratory, Narayana Medical College and Hospital, Nellore, AP, India.

D.RAJARAJESWARI

Department of Biochemistry, Narayana Medical College and Hospital, Nellore, A.P., India

*Corresponding author

ABSTRACT

Recent work in the area of Obesity has confirmed that Obesity is a state of low grade chronic inflammation and is associated with increased levels of TumorNecrosisFactor-α, a pro-inflammatory adipocytokine, which plays a key role in the pathogenesis of Diabetes. In this study we analysed the interrelations between TumorNecrosisFactor-α with anthropometric and clinical variables. The study was carried out on a total of 100 subjects attending outpatient department of General Medicine, Narayana Medical College, Nellore. Serum TumorNecrosisFactor-α was measured by sandwich ELISA method and insulin by chemiluminescence method. TumorNecrosisFactor-α was significantly elevated in diabetic cases[215.18+ 119.58] compared to controls[p= 0.0001]. There was significant positive correlation between TumorNecrosisFactor-α and Body Mass Index. TNF alpha levels were significantly elevated in obese than in non-obese diabetic subjects. Our results suggest a possible role of TNF-alpha in the pathophysiology of Type2 Diabetes mellitus.

ISSN 0975-6299 Vol 2/Issue 2/Apr-Jun 2011

KEY WORDS

Obesity, TNF-alpha, Type2-DiabetesMellitus, Insulin resistance.

INTRODUCTION

Obesity with an increasing prevalence has become the most common metabolic disorder in the world[1]. Obesity is now considered as a state of low grade chronic inflammation[2]. Subclinical inflammation increases the risk of cardiovascular diseases in both metabolic syndrome, the pre diabetic state and in overt Diabetes Mellitus[3,4]. The production of TNF alpha, a pro-inflammatory adipocytokine is noticeably enhanced in Obesity[5]. TNF-alpha is a pleiotropic cytokine with diverse functions and occurs in many pathological diseases like cancer, cardiovascular disease, Type2 Diabetes Mellitus (T2DM) etc[6] . It is produced by macrophages in response to inflammation, endotoxemia and cancer[7] and plays a key role in the pathogenesis of peripheral insulin resistance in Obesity. TNF α inhibits Tyrosine kinase activity at the insulin receptor level and cause obesity induced insulin resistance[8]. Previous studies have reported a significant association between serum TNF-α and insulin resistance when a combined analysis of Diabetic and non–Diabetic subjects were employed, but when examined in obese T2DM alone failed to find a significant relation between TNF alpha and Insulin resistance[9]. Many studies have shown increased serum levels of TNF alpha in obese patients in comparison with lean subjects [9,10,17] . Emerging clinical data suggests that inflammation precedes the development of clinically overt Diabetes and also predicts the subsequent cardiovascular events[11-13]. TNF alpha may serve as an inflammatory biomarker and as an important risk indicator for the future development of T2DM and may prove a novel target for therapeutic intervention[14]. This study was undertaken to estimate the TNF- alpha levels in T2DM and to analyse its association with the anthropometric (Body Mass Index and Waist Hip Ratio) and clinical variables (Fasting glucose and insulin) related to insulin resistance, in obese and non-obese T2DM and control subjects.

MATERIALS & METHODS

The study was carried out on 100 subjects attending outpatient department of General Medicine, NMCH, Nellore. 50 subjects(M=28 F=22) were newly diagnosed Type 2 diabetic patients and 50 subjects (M=26 F=24) were normal healthy controls who attended for their periodic health checkups. All individuals were subjected to a complete medical evaluation by a physician including a full medical history and physical examination. Both males and females between 25-65 years of age were included in the study. Patients with evidence of acute or chronic inflammatory or infectious diseases, cancer, persons on insulin, or other medications that could affect glucose metabolism and pregnant or lactating women were excluded from the study.

Anthropometric measurements: Height (cm), waist and Hip circumference (cm) were noted using a measuring tape to the nearest 0.1cm. waist circumference was measured at the mid point between the lower border of rib cage and the iliac crest. Hip circumference was measured at the level of trochanter, the widest part of the hip region. Weight(kg) was measured to the nearest 0.1 kg using a weighing machine simultaneously . Waist hip ratio (WHR) was calculated as waist circumference divided by hip circumference. BMI was calculated as weight(kg) divided by

height (m^2).Obesity is defined as BMI>30kg/m^2 . ***Biochemical assays:*** Plasma glucose was analysed by glucose oxidase method using Human kits(GmbH) by automated chemistry analyser, Humaster-300(GmbH, Germany). Serum TNF-α was measured by using sandwich Elisa kit method [e-Bioscience, Bender med systems] which has an inter assay co-efficient of variations of 7.5-10.4% and a lower limit of detection 0.5pg/ml. Serum Insulin by using chemiluminescence immunoassay method (Beckman coulter, Virginia) using kits by Bayer diagnostics. All samples were processed and examined according to principles of good laboratory practice at central laboratory, Narayana Medical college and Hospital , Nellore. Homeostasis Model of Assessment (HOMA-IR) method was used for the calculation of Insulin resistance . This method has been validated as a reliable measure of insulin resistance in vivo in humans. High HOMA-IR scores denote lower insulin sensitivity and greater insulin resistance.

$$\text{HOMAIR} = \frac{\text{Fasting serum Insulin (µU/ML)} \times \text{Fasting plasma Glucose (Mm/l)}}{22.5}$$

The study was conducted after obtaining informed consent from all the subjects . the study was approved by the local ethical committee. Statistical analysis was performed using SPSS-13 soft ware version. P<0.05 was considered as statistically significant.

RESULTS

Table-1 summarizes the Mean ± SD values of clinical variants in obese and non- obese Diabetic and control subjects. The mean ± SD value for TNF-α in obese diabetic cases was 215.18 ± 119.58 and was statistically significant between cases and control (p<0.0001). Our results demonstrated that, compared to Age/gender/BMI/ matched obese controls, obese Diabetics had significantly higher circulating TNF-α (p<0.0001), fasting blood glucose(p<0.0001) and fasting Insulin concentrations (p<0.0001). fig-1 shows the elevated levels of TNF-α in obese diabetics compared to non-obese diabetics and obese and non-obese controls. Fig-2 shows a significant positive correlation between TNF-α and BMI within cases (r=0.383 P=0.06). Positive correlation was found between TNF-α & Insulin resistance(r=0.172). All the coefficients were stronger for obese T2DM subjects than for non- obese T2DM & obese controls.

TABLE-1

Mean± SD values of variants in obese and non obese diabetic and control subjects

Type 2 diabetics				Controls		
Variable	Obese	Non obese	P	Obese	Non Obese	P
N	22	28	-	21	29	-
M/F	13/9	15/13	-	14/10	18/11	-
Age(years)	41.3 ± 5.1	51.4 ± 11.3		52.4 ± 2.1	48.1 ± 3.6	
BMI kg/m^2	28.13 ± 10.2	22.3 ± 1.6	0.0001	25.12 ± 4.4	21.3 ± 2.8	0.0001
HOMAIR	7.46 ± 10.8	6.68 ± 6.8	NS	4.58 ± 0.94	3.58 ± 0.5	0.0001
TNF-α	215.8 ± 119.5	168.5 ± 32.5	0.007	42.3 ± 18.5	22.9 ± 15.1	0.0001

FIG- 1

TNF α in obese and non-obese Diabetics and controls

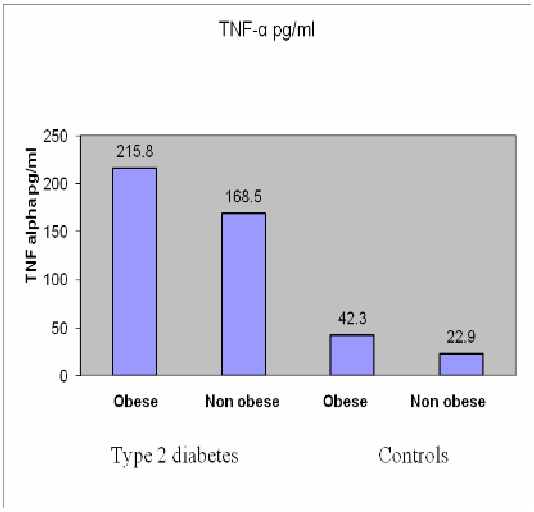
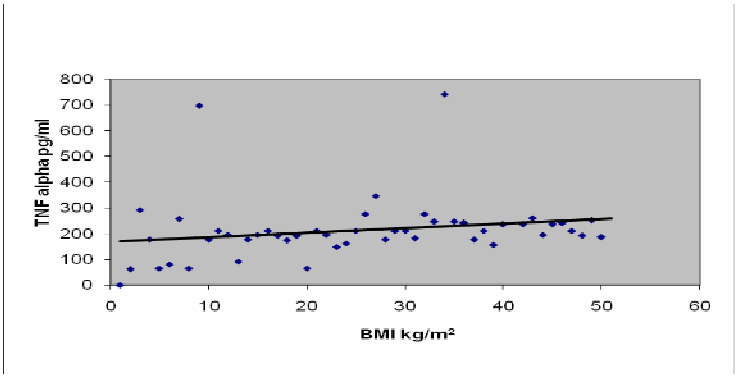


FIG-2

Relation between TNF α and BMI in obese Diabetic subjects



DISCUSSION

TNF-α is hypothesized to link obesity to insulin resistance. studies in human and animal models have indicated that TNF-α expression in the adipose tissue is significantly elevated in obesity [15,18]. In our study TNF-α concentration was significantly high in obese T2DM than in non obese subjects. Our results demonstrated that increased levels of TNF- α were associated with increased levels of glucose in T2DM and was related to the degree of obesity. Nilsson et al[16] reported that the plasma TNF- α levels were increased by 23% in lean T2DM compared to 51% in obese T2DM subjects with more severe insulin resistance. Katsuki et al [17] reported that TNF-α is elevated in obese T2DM but not in lean T2DM. According to Hotamisligil et al body weight reduction in obese individuals is also associated with a reduction in TNF-α levels and in improved insulin sensitivity[18].our present results clearly demonstrated that circulating TNF-α levels were significantly elevated in T2DM compared to normal healthy subjects

particularly in obese subjects , and is strongly correlated with BMI . Our observation is consistent with numerous previous studies which have documented a strong correlation between TNF-α and BMI[19-21]. Elevated levels of TNF-α were also found to predict Cardiovascular events with Diabetes from the nurses health study[22] . All these data provide a strong associative evidence supporting subclinical inflammation as a unifying factor accelerating the progression of Insulin resistance and Type 2 Diabetes mellitus. our data suggest a possible role of TNF-α in the pathophysiology of Insulin Resistance particularly in obese individuals.

REFERENCES

1. Y.Miyazaki . TNFalpha and Insulin Resistance in Type 2Diabetes Mellitus. International Journal of Obesity. 27:88-94(2003).
2. Tzanavari T, Giannogonosa P., Karalis KP. TNF-alpha and obesity. Current directions in Auto immunity. 11: 145-156 (2010)
3. Aruna Pradhan. Obesity, Metabolic Syndrome and Type2 Diabetes: inflammatory basis of Glucose Metabolic Disorders. Nutrition Reviews. 65 : s152-156 (2007).
4. Bronwen Dalziel, Alison K. Gosby, Rose Mary M. Richman, Janet M. Bryson and Ian D.Caterson. Association of the TNF-alpha-308G/Apromoter polymorphism with Insulin Resistance. Obesity Research. 10 (5) 401-407 (2002).
5. Hotamisligil GS, Bdavari A, Murray D, Spiegelman BM. Reduced tyrosine kinase activity of the insulin receptor in Obesity-diabetes. Central role of TNF-alpha. Journal of Clinical Investigation. 94: 1543-1549 (1994).
6. Simona Gwozdziewiczova, Radka Lichnovska, Rabha Ben Yahia, Rudolf Chulp, Jiri Hrebicek. TNF-alpha in the development of Insulin Resistance and other disorders in Metabolic Syndrome. Biomed. 149(1):109-117 (2005).
7. B.Zahorska Markiewicz, J. Janowska, M.Olszanecka Glinianowicz and A.Zurakowski. serum concentration of TNF alpha and soluble TNF alpha receptors in Obesity.24 : 1392-1395(2000)
8. D.Dixon, R.Goldberg, N. Schneiderman and Delamater. Gender differences in TNF-alpha levels among obese vs non obese Latino children. European journal of clinical nutrition. 58, 696-699 (2004).
9. Mohammed Hassan Saiem Al-Dhar and Essan H- Jiffri. Adipose tissue expression of TNF-alpha and insulin resistance in obese subjects with type 2 Diabetes mellitus. World journal of Medical sciences 5(2) : 30-35 (2010).
10. B.Zahorska Markiewicz, J.Janowska, M.Olszanecka Glinianowicz andMajewski T. Serum concentration of TNFalpha in obese women. Journal of endocrinology investigations. 22: 66-72 (1999).
11. Schmidt MI, Duncan BB, Sharrett AR et al. Markers of inflammation and prediction of Diabetes Mellitus in adults(Atherosclerosis risk in community study) a cohort study. Lancet. 353:1649-1652(1999).
12. Barzilay JI, Abraham L, Heckbert SR et al. The relation of markers of inflammation to the development of glucose disorders in the elderly : The Cardiovascular health study. Diabetes. 50: 2384-2389(2001).
13. Spranjer J, Korke A, Mohlig M,et al. Inflammatory cytokines and the risk to develop t2 Diabetes Mellitus. Results of the prospective population based European prospective investigation into

cancer and nutrition (EPIC)-Postsdam study. Diabetes. 52:812-817(2003).
14. Shoelson SE, Lee J, Gold fine AB. Inflammation and Insulin Resistance. J Clin Invest. 116: 1793-1801 . (2006).
15. Hotamisligil G S, Arner P, Atkinson R L, Spiegelman B M. Differential regulation of the P80 TNFfactor receptor in human obesity and insulin resistance. Diabetes. 46: 451-455 (1997).
16. Nilsson J, Jowinge S, Niemann A, Rene lend R, Lithell H. Relationship between plasma TNF alpha and Insulin sensitivity in elderly men with type2 Diabetes Mellitus. Arterio Sclero. Throm Vas Bio. 18:1199-1202 (1998)
17. Katsui A, Sumida Y, Murashimha S, Murata K, Jakarada Y, Ito K et al. Ser. Levels of TNF-alpha are increased in obese patients with NIDM. Jou.Clin. End. Met. 83: 859-862 (1998).
18. Hotamisligil GS, Shargil NS, Spiegelman BM. Adipose expression of TNF alpha : A direct role in obesity induced insulin resistance. Science. 25: 87-91(1993)
19. Mishima Y, Kuyama A, Tada A, Takahaslin K, Ishioka T, Kibata M. Relationship between TNF-alpha and Insulin Resistance in obese men with Type2 Diabetes Mellitus. Diabetes Res clin Pract.52: 119-123 (2001).
20. Dandona P, Weinstock R, Thusuk K, Abdel Rahman A, Aljada A, Wadden T. TNFalpha in serum of obese patients; fall with weight loss. J.Clin.End Met. 83:2907-2910 (1998).
21. Tsigos C, Kyrou I, Chala E, Tsapogas P, Sturidis JC, Raptis SA, Katsilambros N. Circulating TNF-alpha concentrations are hiegher in abdominal verses peripheral obesity. Metabolism. 48:1332-1335 (1999).
22. Shai I, Schulze MD, Manson JE,et al. A prospective study of soluble TNF alpha receptor 2 and risk of coronory heart disease among women with type2 Diabetes Mellitus. Diabetes care. 28: 1376-1382 (2005).