Proceeding S.Z.P.G.M.I. Vol: 29(2): pp. 101-105, 2015.

Determination of Alanine-Aminotransferase Levels in First-Degree Relatives of Diabetics

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ABSTRACT

Background: Hepatic transaminases test such as alanine aminotransferase and aspartate aminotransferase are part of the panel of Liver Function tests in out-patient departments. Their levels are two of the most reliable markers of hepato-cellular injury or necrosis. Out of the two, alanine aminotransferase is thought to be more specific for hepatic injury. **Aims and objectives:** The objective of the present study was to determine alanine aminotransferase levels in norm-glycemic individuals with family history of diabetes because they are two to three times at risk of developing type2 diabetes. As alanine aminotransferase is a valuable screening test to detect other-wise in apparent liver disease and is also cost-effective and sensitive to indicate the presence of liver disease, it will help us in detecting a group of people in which diet and life-style modifications can help to prevent future development of diabetes and cardio-vascular disease. **Subjects and methods:** A total of 100 males and females were investigated with and without family history of diabetes. Fasting levels of serum lipids, glucose and liver enzymes was performed by automated chemistry analyzer. Student t-test was applied to see the differences between the values of controls and subjects. **Results:** A significant difference of p-value <0.05 was present between age, blood pressure, triglycerides, HDL-C, alanine aminotransferase, gamma glutamyl transferase and bilirubin levels of controls and subjects. The male subjects were younger and showed higher values for alanine aminotransferase and other metabolic markers. **Conclusion:** In norm-glycemic first degree relatives of diabetics serum alanine aminotransferase levels were higher and can predict the future disease.

Key words: Alanine aminotransferase (ALT), type 2 diabetes mellitus (type 2 DM), non-alcoholic fatty liver disease (NAFLD), metabolic syndrome (Met S).

INTRODUCTION

Diabetes Mellitus (DM) is a group of metabolic disease characterized by hyperglycemia resulting from defects in insulin secretion, insulin action or both. The chronic hyperglycemia is associated with long term damage, dysfunction and failure of various organs. The basis of abnormalities in carbohydrate, fat and protein metabolism in diabetes is deficient action of insulin on target tissues. Impairment of insulin secretion and defects in insulin action frequently co-exist in the same patient[1].There are different type of diabetes, type 1, type2 and other specific types.

Type 2 DM comprises approximately 90% of all individuals with diabetes[2]. Over the last three decades the number of people with DM has doubled globally, making it one of the most important health challenges[3]. Type 2 is primarily a life-style disorder and highest prevalence rates are occurring in populations that are undergoing "modernization" or westernization. Asian population is one of these populations. The metabolically obese (*i.e.* normal body weight with increased abdominal adiposity) is common on Asian population[4]. Under these circumstances the genetic susceptibility is interacted with environmental change (*i.e.* adopting sedentary life-style and changing nutrition)[5]. As the risk of

having type 2 diabetes is also increased 2-6 fold if a parent or sibling has a disease[6], in these circumstances screening for type 2DM becomes important because only early detection and prompt treatment may reduce the burden of diabetes and its complications. The estimated prevalence of diabetes among adults is 8.7%.

Insulin resistance (IR) is defined as a state when a given concentration of insulin produces a less than expected biological effect and in turn causes the body to not utilize the insulin efficiently. Reduced insulin sensitivity is seen in liver and adipose tissue. Adipose tissue becomes resistant to the effects of insulin and causes peripheral lipolysis which causes increased delivery of free fatty acids (FFA) to the liver[7] and this has the potential to increase hepatic triglyceride accumulation[8]. Abnormalities of triglyceride storage and lipolysis in liver are an early manifestation of IR. Under normal physiological conditions fatty acids enter the cell and are either oxidized or stored. When the influx/out flux ratio is altered, fatty acids accumulate, leading to hepatic steatosis, infiltration, altered mitochondrial function and increased lipid intermediates[9].

Biochemical tests used for screening diabetes are urine glucose, random blood glucose, fasting plasma glucose, glycated hemoglobin, oral glucose challenge test and liver enzymes[10]. All the liver enzymes have been reported to be associated with increased risk of diabetes. Several studies have found association between ALT and GGT levels with increased risk of diabetes[11,12]. Hepatic transferases such as alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are part of Liver function test panel. Their levels are two of the most reliable markers of hepatocellular injury and necrosis. ALT is considered to be more specific for hepatic injury. ALT is an enzyme that catalyzes the transfer of amino group to form hepatic metabolite oxaloacetate. ALT is found in the cytosol and mitochondria of the liver. Its activity is about 3000 times that of serum. Its activity varies day-to-day by 10-30%. In acute hepato-cellular injury AST levels rise immediately, however within 24-48 hours ALT rises because of its longer half-life. In chronic hepato-cellular injury ALT is more commonly elevated than AST. Also ALT is a valuable screening test to detect otherwise in apparent liver disease such as asymptomatic viral hepatitis and non-alcoholic liver disease. Apart from liver disease ALT levels may be affected by gender, body mass index (BMI), triglyceride and cholesterol levels and alcohol. Several prospective studies have previously shown that increased ALT concentration within the reference interval predicts the future development of diabetes and cardio-vascular events independent of other known risk factors[13,14]. Chang reported that ALT concentration also predicted the development of non-alcoholic liver disease in healthy Koreans[15]. A study by Baffy also reported that people with un-diagnosed diabetes also have increased levels of ALT and AST. This maybe due to allostatic load or physiological response to environmental stress which is linked to accumulation of fat, inflammation and oxidative stress.[16]

This study is to determine the ALT levels in first-degree relatives of diabetics with normal blood glucose to observe whether these levels rise earlier than other risk factors.

SUBJECTS AND METHODS

Prior to the start of study informed and written consent was taken from all participants. A total of 100 subjects were selected including 50 males and 50 females to carry out this study. The control group consisted of 25 males and 25 females with normal blood glucose levels and without any family history of diabetes. Whereas in the subject group, there were 25 males and 25 females with normal blood glucose levels with family history of diabetes mellitus in a first-degree relative. Participants were selected from general population. The age of participating was ranging from 25-55 years. Various demographic parameters including height, weight, body mass index (BMI) and blood pressure were recorded. The height was measured on a standard height scale and weight was measured on CAMRY weight scale. BMI was calculated by the following formula; BMI=weight (kg)/height (m^2).

After an overnight fast, 6ml of blood was drawn aseptically and was centrifuged to separate the serum for further analysis. It was divided into 2 aliquots, one for glucose, and one for lipid profile

and liver function tests. All the biochemical parameters including lipid profile, glucose and liver function tests were analyzed by Auto- chemistry analyzer Dimension RXL by DADE Behring (USA) using standard protocols. Insulin resistance was calculated from the formula Tg/HDL-C ratio. The data was analyzed by using SPSS17. Mean±SEM is given for quantitative variables. Independent t-test was applied to observe group mean differences. Pearson Correlation was applied. Correlation was calculated with p value of <0.05 was considered as statistically significance.

RESULTS

In this cross-sectional study, total 50 males and 50 females were included. Controls and subjects were age and sex matched. Other demographic parameters including blood pressure and BMI are given in Table 1.

The male subjects were younger with higher blood pressure. No significant difference was present in female controls and subjects. There was no significant difference between the BMI of male controls and subjects. The BMI of 25.22±0.66 kg/m ^{2} is considered to be grade 1 in overweight category. No significant difference was seen in BMI of female controls and subjects.

Table 1: Demographic parameters.

Parameters	Control	Subject	P-value
Male			
Age (years)	45.88±1.04	41.28±1.73	0.038*
BMI (kg/m^2)	24.72±0.52	25.22±0.66	0.67
BP (systolic)	122±1.53	127±1.66	0.043*
BP (diastolic)	78.00±1.12	83.00±1.29	0.01**
Female			
Age (years)	43.24±1.43	46.12±1.12	0.79
BMI (kg/m^2)	26.48±0.77	28.11±0.80	0.53
BP (systolic)	126±2.57	126.80±2.14	0.64
BP(diastolic)	84.88±1.40	83.60±1.74	0.23

p<0.05significantly different from controls
p<0.01 highly significant as compared to controls

In Table 2 the male subjects showed significant difference in serum triglyceride and HDL-C whereas no significant difference was seen in female controls and subjects.

In Table 3 of the Liver Function test panel the normal values of ALT is 4-36U/L, GGT is 5-40U/L and Direct Bilirubin they are <0.03mg/dl. There was a significant difference between the values of ALT, GGT and Direct bilirubin in male subjects; however no significant difference is seen between female controls and subjects.

Table 2: Represents the comparison of serum glucose, lipid profile and liver function tests of controls and subjects.

Parameters	Control	Subject
Male		
Cholesterol(mg/dl)	178.88±5.10	184.24±7.88
Triglycerides(mg/dl)	105.72±5.48	134.48±9.97*
HDL(mg/dl)	40.96±1.16	36.24±1.33*
LDL(mg/dl)	116.68±4.42	114.64±5.77
Glucose	84.40+2.54	91.72+3.89
Female		
Cholesterol(mg/dl)	190.52±5.41	184.00±6.75
Triglycerides(mg/dl)	107.64±10.32	101.76±6.95
HDL(mg/dl)	46.20±1.45	45.04±1.55
LDL(mg/dl)	130.76±6.07	116.32±6.00
Glucose	81.24+3.09	81.24+2.68

*p<0.05 significantly different as compared to controls

Table 3: Liver function tests of both the groups:

Parameters	Control	Subject
Male		
Alkaline Phosphatase (U/L)	81.40±3.20	87.20±3.46
Alanine aminotransferase (U/L)	41.28±1.88	46.88±1.89*
Aspartate aminotransferase (U/L)	23.88±1.15	24.52±1.05
Total Bilirubin (mg/dl)	0.55±0.04	0.60±0.06
Direct Bilirubin (mg/dl)	0.09±0.01	0.13±0.02*
GGT(U/L)	24.84±1.77	35.64±3.30**
Female		
Alkaline Phosphatase (U/L)	77.56±4.50	76.28±3.75
Alanine aminotransferase(U/L)	42.28±2.12	38.36±2.19
Aspartate aminotransferase(U/L)	26.28±1.70	22.40±1.07
Total Bilirubin (mg/dl)	0.51±0.04	0.49±0.03
Direct Bilirubin (mg/dl)	0.09±0.02	0.07±0.01
GGT(U/L)	26.12±2.08	26.56±3.38

*p<0.05 shows significant difference as compared to controls
**p<0.01 shows significant as compared to controls

DISCUSSION

The Metabolic syndrome (met S) is a combination of multiple disorders including insulin resistance, abdominal obesity, increased blood

pressure, hypocholesteremia and pro-inflammatory state. Met S has become a leading cause of morbidity and mortality in industrial countries[17]. The relationship between Met S and NAFLD is bidirectional. Liver fat content is significantly increased in subjects with Met S as compared to those without it, independent of age, gender and BMI and in turn the presence of NAFLD is a strong predictor of Met S. Markers of NAFLD are associated with future risk of type 2 DM and cardio-vascular mortality[18].

The serum ALT activity has been regarded as a reliable and sensitive marker of liver disease. ALT levels may also be a good indicator of overall health, particularly in context of obesity, metabolic syndrome and presence of cardiovascular disease. Despite all these considerations abnormal ALT activity is often ignored or minimized as most patients are asymptomatic. Minor elevations are often considered clinically insignificant, in part because of lack of perspective about the impact of abnormal ALT on long-term outcomes such as end-stage liver disease or premature mortality. ALT measurements provide a readily available, low-cost blood test for detection of liver disease or a valuable screening test to detect otherwise in apparent liver disease. The average serum ALT level considered as the upper normal limit is 40IU/L ranging from 30-50IU/L among various populations.

The recognized markers for metabolic disease are high BMI, total cholesterol, triglycerides, uric acid, low HDL-C levels and ALT levels. These markers predict NAFLD. A limited number of studies evaluated the association of parental history of type 2DM with levels of some diabetes related metabolic bio-markers. These studies found no association of parental history with triglycerides and HDL-C. They however found a positive association with history of type2 DM with liver enzymes such as GGT and ALT in non-diabetic off-springs[19,20]. Abbassi and associates found that parental history of type 2DM was associated with higher levels of non-fasting levels of ALT, AST and GGT than in general population without history of type 2 DM[21]. Chang and associates reported that ALT concentrations also predicted the incidence of NAFLD[27]. Several other prospective studies have previously shown that increased ALT levels within the reference interval also predict the future development of diabetes and cardiovascular events[22]. In non-diabetic individuals ALT has direct relationship with triacylglycerol and low HDL-C concentrations[14].

Kim J and Jo I in a population based study found a strong relationship of BMI with ALT in non-diabetic Koreans population. The risk tended to increase according to the severity of obesity. Similarly NHANES III data has revealed that the risk of ALT is two to three fold higher in overweight (BMI>25 and <30)[23]. However Kotrenan and associates explained that although a relationship exists between ALT and liver fat, however serum ALT concentrations explained only 15-19% of this fat, implying that this enzyme is a poor marker of liver fat content in non-diabetic subjects[24]. Marchesini has also reported association between ALT activity and hyperlipidemia[25].

This biochemical, clinical and epidemiological information suggests that ALT can be used to screen chronic liver disease. Higher ALT levels represent not only a status of sub-clinical hepato-cellular inflammation but they also might predict a pre-diabetic status.

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