

Elevated serum level of human alkaline phosphatase in obesity

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Abstract

Objective: To investigate a correlation between serum alkaline phosphatase level and body mass index in human subjects.

Methods: The comparative cross-sectional study was carried out at the National Institute for Biotechnology and Genetic Engineering, Faisalabad, Pakistan, from April 2012 to June 2013. Blood serum alkaline phosphatase levels were estimated and the subjects were divided into three sub-groups on the basis of their body mass index: normal weight ($\leq 25\text{kg/m}^2$), overweight ($25\text{-}27\text{kg/m}^2$) and obese ($>27\text{kg/m}^2$) subjects. The serum samples were used for the estimation of clinically important biochemical parameters, using commercial kits on clinical chemistry analyser.

Results: Of the 197 subjects, 97(49%) were obese and 100(51%) were non-obese. The serum alkaline phosphatase level increased in obese (214 ± 6.4 IU/L) compared to the non-obese subjects (184.5 ± 5 IU/L). Furthermore, a significant linear relationship ($r=0.3$; $p<0.0001$) was found between serum alkaline phosphatase and body mass index. Other biochemical variables were not correlated to the body mass index.

Conclusion: Over activity and higher amounts of alkaline phosphatase were linked to the development of obesity.

Keywords: Alkaline phosphatase, ALP, Obesity, Body mass index, BMI. Biomarker. (JPMA 65: 1182; 2015)

Introduction

Obesity is a complex metabolic disorder which is characterised by an excessive accumulation of fat in various body parts. The prevalence of obesity along with other metabolic disorders such as type 2 diabetes mellitus (T2DM or T2D), cardiovascular diseases (CVD), certain types of cancers and lung-related abnormalities is challenging for the global population.¹⁻⁴ Both genetic and environmental factors play a role in the development of obesity.⁵ It is well studied that in normal physiological state, a product of obese gene (Leptin or Ob), a leptin hormone regulates body weight within permissible limits by creating a balance between fat intake and energy expenditure.⁶ It is also reported that obese subjects have increased serum leptin levels and become resistant to leptin.⁷ Such leptin resistance reduces appetite, suppressing effect of leptin in obesity, resulting in excessive calories being stored in various body tissues primarily in the adipose tissue. Uncontrolled and abnormal fat overloading and distribution among different body tissues promotes severity of obesity, which is associated with insulin resistance, T2D, CVD and pulmonary problems.^{6,8}

Apart from several other biochemical, molecular and

genetic factors, an enzyme called alkaline phosphatase (ALP) has also been reported to increase in obesity.⁹ A study showed that serum level of ALP in obese was significantly higher than non-obese subjects.¹⁰ Interestingly, it has recently been studied that besides other body tissues (e.g. liver, bone, bile duct, kidney, intestinal mucosa and placenta), ALP is also expressed in adipose tissue which regulates intracellular fat deposition in preadipocytes during adipogenesis.¹¹ In obesity, the activity of ALP is enhanced and anticipated disproportionate intracellular fat depots. In return, ALP is released from adipose tissue into the blood circulation in excessive amount.⁹ Yet, no comprehensive clinical study has been conducted to find out an association of serum ALP level with obesity.

On the basis of such observations, the present study was planned to evaluate the relationship between obesity and blood serum level of ALP enzyme. Additionally, correlations of obesity with aspartate aminotransferase (AST), alanine aminotransferase (ALT), albumin and bilirubin were also investigated.

Subjects and Methods

The comparative cross-sectional study was carried out at the National Institute for Biotechnology and Genetic Engineering (NIBGE), Faisalabad, Pakistan, from April 2012 to June 2013. Study subjects comprised adult obese and non-obese individuals. For blood sample collection, camps were organised at several sites in and around the district of Faisalabad. All subjects were informed to attend the blood collection spots on a specific day in the

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morning after 12-14 hours overnight fast. Prior to blood sample collection, information regarding demographic and socio-economic status was also obtained through a pre-designed questionnaire. Measurements of height and weight were taken to calculate body mass index (BMI as kg/m²). In line with literature, BMI of 27 was used as cutoff point to distinguish obese and non-obese subjects.¹² Subjects based on BMI were categorised as: lean/normal weight (BMI ≤25), overweight (BMI = 25-27) and obese (BMI >27). None of the subjects suffered from bone disorders and hepatobiliary abnormalities.

After approval by the institutional ethics committee, and informed consent was obtained from each participant, a total of 10ml venous blood sample was taken in gel containing vacutainer without anticoagulant from each subject between 8am and 10am after an overnight fast. The blood sample of every subject was allowed to clot at room temperature for about 30 minutes and was then centrifuged and stored in Eppendorf tubes. Subsequently, serum was used for biochemical analyses, including fasting glucose, cholesterol, triglycerides, albumin, bilirubin, AST, ALT and ALP. These biochemical parameters were determined by colorimetric methods using commercial kits (DiaSysHolzheim, Germany), on a semi-automatic clinical chemistry analyser (Microlab-300, Merck, Germany).

Statistical analysis was carried out in MS Excel 2010 and GraphPadPrism 5. All results are presented as mean ± standard deviation (SD). Threshold for two-tailed p-value significance was set at p<0.05. Correlations between BMI and other variables such as glucose, cholesterol, triglycerides, albumin, ALT, AST, ALP and total bilirubin were carried out using Pearson correlation coefficient (r).

Results

Of the 197 subjects, 116(59%) were women and 81(41%) were men. Overall, 97(49%) subjects were obese and 100(51%) were non-obese. The mean age was 45.2±11.2 years for obese subjects and 48.1±8.8 years for non-obese. The obese had higher mean BMI value

Table-1: Baseline characteristics.

Subjects Parameters	Non-obese (n= 100)	Obese (n= 97)	p-value
Age (years)	48.1±8.8	45.2±11.2	-
BMI (Kg/m ²)	23.8±3.1	32.9±5.4	0.0001*
Glucose (mg/dl)	88.2±18	89.04±12.4	0.69
Cholesterol (mg/dl)	192.06±42.6	197.47±42.6	0.41
Triglycerides (mg/dl)	149±6.3	164.1±8.3	0.1514
Albumin (g/dl)	4.6±0.8	4.5±0.5	0.2
Bilirubin (mg/dl)	0.49±0.01	0.53±0.01	0.08
AST (IU/l)	26.69±1.2	28.3±1.5	0.39
ALT (IU/l)	26.14±1.49	28.38±1.6	0.3
ALP (IU/l)	184.5±5	214±6.4	0.0001*

*Shows significant p-value <0.05

BMI: Body mass index

AST: Aspartate aminotransferase

ALT: Alanine aminotransferase

ALP: Alkaline phosphatase.

32.9±5.4Kg/m² compared to 23.8±3.1 Kg/m² for non-obese. In case of biochemical variables only serum level of ALP was increased significantly (p=0.0001) in obese compared to the non-obese subjects (Table-1).

BMI was correlated significantly (r=0.3; p=0.0001) to the serum level of ALP, but the correlation for the remaining parameters with BMI were non-significant (Table-2).

The subjects were divided into three BMI sub-groups i.e. normal weight (BMI ≤25), overweight (BMI = 25-27) and obese (BMI >27) groups. The analysis revealed that serum ALP level was linearly increased with BMI and was significantly different in group-wise comparison, with maximum difference found between lean and obese at BMI cutoff point 27. Moreover, the increase of serum ALP levels in overweight and obese were 4.5% and 15% respectively relative to the lean subjects. The correlation between ALP and BMI was significant (r=0.3; p=0.0001); whereas ALT and AST were not correlated with BMI i.e. (r=0.035; p=0.62) and (r=0.038; p=0.59) respectively.

Table-2: Correlations of BMI with various biochemical variables.

Parameters	Glucose		Cholesterol		TG		Albumin		ALT		AST		ALP		T o t a l		
	r	p	r	p	r	p	r	p	r	p	r	p	r	p	r	p	
Bilirubin																	
BMI	0.03	0.66	0.03	0.62	0.01	0.87	0.054	0.2	0.035	0.62	0.038	0.43	0.3*	0.0001	0.12	0.06	

*correlation was considered significant at p-value<0.05 Pearson correlation (r), p (p-value)

TG: Triglycerides

ALT: Alanine transaminase

AST: Aspartate transaminase

ALP: Alkaline phosphatase

BMI: Body mass index.

Discussion

Obesity is a precursor for several metabolic diseases such as T2DM, hypertension, CVD, chronic obstructive pulmonary disorders (COPD) and asthma.^{3,8} The rate of adipogenesis is increased in obese subjects, resulting in excess amount of fat that is accumulated in the adipose tissue.⁵ Several genetic factors are reported that take part in obesity development.¹³ However, molecular mechanisms underlying development of obesity are not completely understood. It is well established that a defect in adipocyte-derived hormone leptin is significantly contributing to the obesity progress.⁸ A high serum leptin level reflects excessive accumulation of fat in the adipose tissue and other peripheral parts of the body.⁷ Recently, an ALP isozyme was reported to be expressed in adipocytes, which is implicated to raise fat depots during the course of enhanced adipogenesis in obesity. This high fat deposition might add surplus leptin into the blood circulation of the obese subjects.^{8,9,11}

The presence of an ALP isozyme in adipocytes is conceivable that adipose tissue might be a source of serum ALP. Furthermore, increased serum ALP concentration in obese versus non-obese subjects is a clue for additional release of ALP from adipose tissue in obesity. Hence, the present study was undertaken to establish a relationship between serum ALP level and BMIs of the study subjects. BMI is used ubiquitously to define whether a subject is normal weight or overweight/obese.^{12,14} Importantly, the present study with clinical samples from normal weight, overweight and obese Pakistani Punjabi subjects revealed that serum levels of ALP were significantly increased in obese subjects, which was 15 percent higher compared to the normal-weight subjects. Moreover, its level was linearly increased with BMI value. A study indicated a positive relationship for the activity of ALP enzyme and fat accumulation in adipose tissue.⁹ Thus, in obese subjects, both over-activity and increased production of ALP seem to be contributing in leptin overspill from adipose tissue into the blood stream. Result in elevation of serum leptin occurs, which leads to the development of leptin resistance by adducting with circulating serum leptin interacting proteins (SLIPs) in obesity.^{6,8,9,11,15} Recently a study manifested that serum leptin level increases with BMI and becomes maximum in severely obese subjects, using BMI >27 as cutoff point.¹⁴ This perception is also supported by other past studies that revealed ALP is a precursor of adipocytes and may participate in the regulation of adipogenesis.^{16,17} A study in animal models demonstrated that ALP activity has been raised markedly in obese versus non-obese subjects.¹⁸ A study predicted a

link between ALP activity and insulin secretion/action from the pancreatic beta cells.¹⁹ Furthermore, former studies have manifested that inhibitors of ALP, such as histidine and levamisole, impede adipogenesis with concomitant reduction of fat accumulation in human fat cells.^{11,20}

In addition to ALP, other biochemical parameters like albumin, bilirubin, ALT and AST were estimated in sera of these obese and non-obese human subjects. ALT is mainly produced from hepatocytes and in case of any infection or inflammation its serum level exceeds normal range. Thus, it is regarded as a well-known marker of liver infection.²¹ However, AST is not a better predictor of hepatocytes injuries, because it is produced from various body organs such as liver, kidney, lung, brain and placenta. Serum level of albumin and bilirubin characterise hepatocytes functionality and biliary obstruction respectively.²¹⁻²³ No increase in ALT, AST and other liver function related biomarkers somehow exclude the liver for the release of ALP and thus support its origin from adipose tissue in the obese subjects in the current study. Overall, our findings are in accordance with previously published results,^{9,10,24} but these warrant further investigations.

Conclusion

Our findings, together with previously published results, indicate that both increased expression and activity of ALP during adipogenesis may be an important contributing factor for the development of fat depots in adipose tissue and subsequent induction of leptin resistance in obese compared to non-obese subjects. These investigations might be helpful to provide a new surrogate biomarker and therapeutic options for the research struggle to tackle obesity. However, this correlation between serum ALP level and BMI needs to be validated in large population based studies before considering its translational potential for patients.

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