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Is Body Mass Index an Important Determinant in Screening Serum Amylase Level?

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ABSTRACT

Amylase enzyme is an enzyme involved with digestion of carbohydrates. Unusual levels of serum amylase accompany with medical complications. Determination of serum amylase level is not done routinely in the medical laboratories and it is recommended for the patients diagnosed with acute and chronic pancreatitis at its severe stage. Thus, screening of serum amylase levels in early stages has a great importance. Hence, the objective of this study was to investigate whether there is an association between serum amylase level and Body Mass Index (BMI). Healthy individuals of 120 belonged to 22-35 years were recruited to the study. BMI calculation was done according to the standard protocol. The serum amylase level was determined by MISPA VIVA semi-automated Clinical Chemistry Analyzer. The minimum and maximum serum amylase levels of the study population were 15.6 and 132.19 U/L, respectively. The mean serum amylase level of females was 72.35 U/L and in males, 63.28 U/L. The mean serum amylase enzyme level in underweight, normal, overweight and obese groups were 69.32, 69.30, 70.25 and 70.28 U/L, respectively. In males, the mean serum amylase enzyme levels in same BMI categories were 63.30, 48.82, 51.32 and 80.06 U/L, respectively and in females, 70.24, 74.42, 81.20 and 62.79 U/L, respectively. Results showed that serum amylase enzyme level was significantly higher in females compared to males ($p=0.042$). There was no significant linear correlation between serum amylase enzyme level and BMI in both males ($r=0.204$, $p=0.248$) and females ($r=-0.046$, $p=0.671$) of the study population.

1. Introduction

Amylases are group of enzymes that degraded complex carbohydrates into small fragments by hydrolysis reactions. There are three types of amylases; (i) α -amylase (ii) β -amylase and (iii) γ -amylase, which are categorized according to their ability to hydrolyse polysaccharide bonds. In humans, amylase is produced by pancreatic cells, salivary glands, small intestinal mucosa, ovaries, placenta, liver and fallopian tubes. Pancreatic and salivary amylases are isoenzymes [1]. In total amylase, a higher percentage is produced by the pancreas and smaller percentage is by the salivary glands. Salivary amylase is a glucose-polymer cleavage enzyme, produced by salivary glands [2]. It is a hydrolase enzyme. Salivary amylase cleaves α -1,4 glycosidic bond of complex carbohydrate into simple sugars such as glucose and maltose [3].

Pancreatic amylase plays the main role in luminal digestion of carbohydrate in small intestine [4]. Acinar cells of the exocrine pancreas synthesize the amylase enzyme and secrete it into the gastrointestinal tract via pancreatic duct [1].

High serum levels of amylase enzyme activity could be due to a variety of factors such as acute and chronic pancreatitis, pancreatic pseudocysts formation, cancers in pancreas, colon, ovary, breast or lung, ascites, macroamylasemia, peptic ulcer, intestinal infarction, appendicitis, intestinal obstructions, swelling of salivary glands, acute cholecystitis, burns, peritonitis, diabetic ketoacidosis, renal failure, Morphine drug usage, alcohol intake, prostatic tumours, anorexia nervosa and hypertriglyceridemia. On the other hand, low

levels of amylase enzyme are due to liver failure and cystic fibrosis [5]. In addition to that, cardio-metabolic conditions such as obesity, metabolic syndrome and type 2 diabetes lead a major role in low serum amylase concentration in humans. Smoking, exercise, stress and eating disorders also can affect serum amylase enzyme activity [6].

Usually, serum amylase test is recommended for the patients who are diagnosed with clinical complications such as acute and chronic pancreatitis, at its severe stage. Generally, abnormal serum levels of amylase enzymes can be mostly prevailed as an asymptomatic condition and in an undiagnosed state. So, it has a great importance in screening serum levels of amylase enzyme in early stages, thereby people can obtain proper treatments before falling into risk.

Although it has been reported that obesity has an association with serum amylase level, contradictory findings have been shown by previous studies on salivary, pancreatic and serum amylases. According to Aldossari *et al*, Bonnefond *et al*, and Nakajima *et al*, amylase enzyme level is inversely correlated with BMI [7-9]. Contrast to this observation, Mennella *et al* mentioned that amylase activities are higher in overweight than in normal weight subjects [10].

However, if it is possible to build a correlation between serum amylase enzyme level and BMI, people can go for screening tests by concerning their BMI. Individuals can assume their degree of risk for the elevated or decreased amylase levels with the aid of BMI, and thereby they can manage their food habits or life styles. Hence, it has a great importance in analyzing the correlation between BMI and serum amylase enzyme levels based on Asia-Pacific BMI categorization as applicable to Sri Lanka. Therefore, the objective of this study was to investigate the association between the level of serum amylase enzyme and BMI in healthy individuals according to the Asia-Pacific cut-off point categorization of BMI.

2. Material and Methods

2.1 Study design

This was a descriptive cross sectional study with laboratory investigations.

2.2 Study population

Subjects were selected according to the inclusion and exclusion criteria by examining the answers provided by each individual to the self-administered questionnaire.

2.3 Inclusion criteria

Study population included healthy males and females within 22-35 year age group.

2.4 Exclusion criteria

Exclusion criteria included individuals clinically diagnosed with acute and chronic pancreatitis, patients suffering from renal problems, type 1 & 2 diabetes, intestinal infarction, intestinal blockages, appendicitis, ascites, peptic ulcer and acute cholecystitis, liver failure, cancers, diabetic ketoacidosis and hypertriglyceridemia. In addition to that, alcoholics, people undergone medications and surgeries, individuals with burns and pregnant & lactating women were excluded from the study.

2.5 Sample size calculation

Sample size was calculated according to the following equation ensuring higher precise sample size.

$$n = Z^2 \sigma^2 / e^2$$

Where;

Z = Confidence level

σ^2 = Variance

e = Chance of sampling error

n = Sample size

According to the above equation, sample size was obtained as 30 subjects per one BMI category. As there are four BMI categories, 120 subjects were recruited in the study.

2.6 Data collection

The study was conducted after obtaining ethical approval from the Ethical Review Committee, Faculty of Medical Sciences, University of Sri Jayewardenepura, Sri Lanka. Subjects were selected by convenience sampling method. Data collection was done by using a self-administered questionnaire. Before commencing the study, an information sheet was given to the participants. All the subjects were informed about the study including implications, outcomes and method of sample collection in their own language. The voluntary written consent was obtained from each participant.

2.7 Calculation of Body Mass Index (BMI)

Each individual was weighted using a pre-calibrated electrical scale to the nearest decimal place in kilogram. Standing height was measured using a stadiometer fixed on a wall with a flat floor and the reading on the scale was taken in meters as the standing height by keeping the investigator's eye and the level of the head-piece at same level. BMI was calculated using following formula,

$$\text{BMI} = \text{Weight (kg)} / \text{Height}^2 (\text{m}^2)$$

After that, BMI value of each individual was included in the appropriate BMI category according to the Asia-Pacific cut-off values of BMI [11] as follows.

1. BMI <18.5 Kg/m² (underweight)
2. BMI between 18.5 to 22.9 Kg/m² (normal weight)
3. BMI between 23.0 to 24.9 Kg/m² (overweight)
4. BMI ≥ 25.0 Kg/m² (obese)

2.8 Blood samples collection and serum separation

Venipuncture was performed and 3 mL of whole blood was collected from each individual. The blood was transferred into the pre-labeled plain tube with red cap which was free from anticoagulants. All the collected blood samples were left undisturbed at room temperature for 30 minutes by keeping the plain tubes with blood in an upright position in a tube rack. After 30 minutes, the samples were checked for clot formation. All the samples had been clotted after 30 minutes. Then, the tubes were gently tapped in order to detach the blood clots from the bottom of the tubes. Next, the clotted samples in plain tubes were centrifuged at 3000 rpm for 5 minutes. After centrifugation, tubes were observed for haemolysis before serum was separated. Finally, the supernatant was separated carefully as the serum. Then, the serum samples were stored at -20°C until the analysis was carried out.

2.9 Determination of serum amylase enzyme level

The level of total serum amylase enzyme was measured using the MISPA VIVA semi-automated Biochemistry Analyzer. Quality control (QC) samples were run before processing the serum samples. Serum amylase enzyme level was measured by kinetic method with 2-chloro-p-nitrophenyl-α-D-maltotrioxide (CNPG3) as the substrate.

2.10 Data analysis

Data analysis was done using IBM Statistical Package for Social Sciences (SPSS) software version 25.

3. Results and Discussion

3.1 Distribution of BMI in the study population

The distribution of BMI in the study population was symmetrical. Similarly, the distribution of BMI in males and females separately was also symmetrical. On account of the symmetrical distributions, we considered the mean value for each BMI category in the study population.

3.2 Distribution of serum amylase enzyme level in the study population

The distribution of serum amylase level in the study population was also symmetrical. Similarly, the distribution of serum amylase levels in males and females separately was symmetrical. Since the symmetrical distributions, we considered the mean values of serum amylase levels in the study population.

3.3 Distribution of serum amylase enzyme levels according to the gender

Females showed a higher mean level of serum amylase compared to males. The mean serum amylase level in the females was 72.35 U/L whereas in males it was 63.28 U/L. The mean serum amylase level of females was higher by 9.07 U/L than males. According to the t-test for equality of means, there was a significant difference in mean serum amylase levels between males and females (p=0.042). Accordingly, the mean serum amylase level was significantly higher in females compared to males.

3.4 Distribution of serum amylase enzyme levels according to the BMI

The mean serum amylase enzyme levels in the study population within four BMI groups are indicated in the Table 1.

Table 1: Mean serum amylase enzyme levels within four BMI groups

BMI category	Mean serum amylase enzyme level (U/L)
Underweight	69.32
Normal	69.30
Overweight	70.25
Obese	70.28

3.5 Distribution of serum amylase enzyme levels according to the gender

The distribution of mean serum amylase enzyme levels within four BMI groups according to the gender is indicated in the Table 2.

3.6 Comparison of mean values of serum amylase enzyme level across categories of BMI in male and female

One-way ANOVA test was used to compare the mean values of serum amylase enzyme level across categories of BMI in female and male study populations. In females, though there was a reduction in serum amylase level in obese group compared to underweight and normal groups it was not statistically significant (p>0.05). However, in

males there were significant differences ($p < 0.05$) in mean serum amylase levels across some of the categories of BMI.

Table 2: Distribution of mean serum amylase enzyme levels within four BMI groups according to the gender

BMI group	Mean serum amylase enzyme (U/L)	
	Male	Female
Underweight	63.30	70.24
Normal	48.82	74.42
Overweight	51.32	81.20
Obese	80.06	62.79

3.7 Identification of significantly different BMI groups with respect to serum amylase enzyme levels in female and male study populations

According to One-way ANOVA test, results showed that serum amylase enzyme level of each BMI category was significantly different in male study population and not in female study population. In order to check individual differences of mean serum amylase level between BMI groups in males, Post-Hoc comparisons using the Tukey test was selected. There was a significant difference between mean serum amylase enzyme levels of normal weight and obese males ($p = 0.006$). The mean serum amylase level of obese males was 31.2 U/L higher than that of normal weight males. Also, there was a significant difference between mean serum amylase level of overweight and obese males ($p = 0.002$). Mean serum amylase level of obese males was 28.7 U/L higher than that of overweight males. The difference between mean serum amylase levels of underweight & normal weight males, underweight & overweight males, underweight & obese males and normal weight & overweight males were not statistically significant ($p > 0.05$).

3.8 Linear correlation between serum amylase enzyme level and BMI

Pearson's correlation coefficient (r) was used to evaluate the correlation between serum amylase enzyme level and BMI. Pearson's correlation coefficient (r) and p value obtained from statistical analysis are given in the Table 3.

Since $p > 0.05$, there was no significant linear correlation between serum amylase enzyme level and BMI in both females and males.

Table 3: Correlation between serum amylase enzyme level and BMI in the study population

Gender	Pearson's correlation coefficient (r)	p Value
Females	-0.046	0.671
Males	0.204	0.248

It has been reported that serum amylase level is influenced by several conditions/factors and BMI is one of them. Aldossari *et al* had conducted a study on association between salivary amylase enzyme activity and obesity whereas Bonnefond *et al* investigated on relationship between salivary/pancreatic amylase and BMI. Nakajima *et al* revisited cardiometabolic relevance of serum amylase. According to the above mentioned past studies increased body weight tends to decrease serum amylase activity [7-9]. However, according to the study conducted by Mennella *et al* on salivary α -amylase activities and body weight, amylase activities are higher in overweight than in normal weight subjects [10].

The minimum serum amylase level reported from our study population was 15.6 U/L, while the maximum level was 132.19 U/L. Data were presented as mean values because, symmetrical distributions were shown by both BMI and serum amylase level within the study population and it was fitted to the standard normal distribution. The minimum serum amylase level in the females was 30.95 U/L, while the maximum level was 132.19 U/L. The mean serum amylase level of the females was 72.35 U/L. The mean serum amylase level of males was 63.28 U/L. The minimum and maximum levels were 15.6 and 104.85 U/L, respectively. According to that, the serum level of amylase enzyme was significantly higher in females than males. This finding was supported by the previous studies. Uede *et al* stated that total serum amylase, salivary and pancreatic type isoamylase levels were significantly higher in females than males [12]. Segawa *et al* reported that mean serum amylase level was higher in females than in males [13]. However, we obtained a statistically significant association between serum amylase enzyme levels and gender. This finding is supported by the previous studies [12,13].

The mean serum amylase levels in females of underweight, normal weight, overweight and obese were 70.24, 74.42, 81.20 and 62.79 U/L, respectively while the mean serum amylase levels in males of underweight, normal weight, overweight and obese were 63.30, 48.82, 51.32 and 80.06 U/L, respectively. Therefore, according to the results of the present study, in males, normal weight group

had the minimum mean value and obese group had the maximum mean value for serum amylase level. But in females, obese group had the minimum mean value and overweight group had the maximum mean value for serum amylase levels. Also, underweight, normal weight and overweight group females had higher mean serum amylase enzyme level than males. But, obese group males had higher mean serum amylase enzyme level than females. Further, there was a statistically significant difference of mean serum amylase levels between normal weight and obese groups and between overweight and obese groups in males. In the females, though there was a reduction in serum amylase level in obese group compared to underweight and normal groups it was not statistically significant ($p>0.05$). According to these findings we could not get significant linear correlation between serum amylase level and BMI in both males and females.

In comparison of serum amylase levels of present study with the previous studies the main observation made was in the past studies authors had expressed the results as Mean \pm SD in relation to serum amylase. But, we could not be able to calculate standard deviations (SD) for any of the BMI group of both genders due to wide variations in serum amylase levels. This is obvious because the reference range of serum amylase shows a wide variation ie. 26-102 U/L [1]. So, it is clear that why we could not get a linear relationship between serum amylase and BMI as far as the mean serum value is considered. In addition to that, vast variations in serum amylase levels may be due to food intake of the subjects prior to blood sample collection. This is because increased carbohydrate utilization is associated with lower plasma amylase level [7].

4. Conclusion

Females showed a statistically significant higher mean level of serum amylase level compared to males. In the females there was a reduction in serum amylase level in obese group compared to underweight and normal groups but it was not statistically significant. There was a significant difference between mean serum amylase enzyme levels in the males of normal weight & obese and overweight & obese males. There was no linear correlation between serum amylase levels and BMI in both genders.

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