

Original Research Article

EVALUATION OF THE SERUM LIVER ENZYMES MARKERS, LIPID PROFILE AND KIDNEY

FUNCTION PARAMETERS IN TYPHOID PATIENTS

ABSTRACT

Aim

Typhoid fever is a life-threatening infection caused by the bacterium *Salmonella enterica serotype Typhi* occurring frequently in underdeveloped regions of the world due to overcrowding and poor sanitation. However, the biochemical changes induced by typhoid fever have not been fully understood as a guide to finding a possible counter measure to combating the disease. The study was designed to determine the biochemical changes associated with typhoid disease.

Place and duration of study

The study was carried out at the Department of Biochemistry, College of Natural Sciences, Michael Okpara University of Agriculture, Umudike, Nigeria, in the year 2014.

Method

The study evaluated kidney electrolyte, lipid profile, liver marker enzymes in the serum of typhoid patients. 140 subjects were recruited for this study, 70 of which were normal individual control group and 70 individual with typhoid test group with age range of 18 to 28years. Blood samples were analyzed to determine the lipid profile, kidney function and liver enzyme concentration spectrophotometrically.

Result

Urea and creatinine concentration in typhoid patients significantly increased ($P < 0.05$) when compared with the control group. Sodium ion, potassium ion, chloride ion and bicarbonate ion significantly decreased ($P = 0.05$) when compared to the control group. Result for liver function test revealed a significant increase in the concentration of AST, ALT and ALP in test group. HDL, LDL, Cholesterol, triacylglycerols and VLDL concentration significantly increased ($P = 0.05$) in typhoid patients when compared with normal individuals. Concentration of total protein, albumin, globulin significant decrease ($P = 0.05$) in test group when compared with the control group.

Conclusion

The study gave a clear indication of the changes induced by typhoid fever in humans. This study also revealed some physiological and biochemical conditions in typhoid patients that varies from normal individual, which explains the influence of typhoid morbidity.

Keywords: Typhoid Fever, Electrolytes, Urea, Creatinine, Cholesterol, Blood

INTRODUCTION

Typhoid fever is a systemic disease caused by *Salmonella typhi*, affecting only humans, [1]. In humans and animals, *Salmonella* infections are a significant cause of morbidity and mortality [2]. Typhoid fever is communicated by polluted food and water by feces and urine of patients and carriers [1].

Typhoid fever is endemic in developing countries and may cause very different clinical findings. Although hepatic involvement and abnormal liver function tests may be seen in 50% of the patients [3].

Typhoid fever, also known as typhoid is a common worldwide bacterial disease transmitted by the ingestion of food or water contaminated with the feces of an infected person, which contain the bacterium *Salmonella entericaserovarTyphi* [2].

The bacterium that causes typhoid fever may be spread through poor hygiene habits and public sanitation conditions, and sometimes also by flying insects feeding on faeces. Diagnosis is made by any blood, bone marrow or stoolcultures and with the Widal test (demonstration of salmonella antibodies against antigensO-somatic and H- flagellar).

The term *enteric fever* is a collective term that refers to typhoid and paratyphoid [2]. With an estimated 16–33 million cases of typhoid fever annually resulting in 216,000 deaths in endemic areas, the World Health Organization identifies typhoid as a serious public health problem.

With the high mortality rate associated with typhoid patients, the study was designed to determine the various biochemical parameters such as liver enzymes and kidney function in typhoid patient's serum to possibly recommend management measures for the disease condition.

MATERIALS AND METHOD

Blood collection and serum preparation

Blood sample of typhoid patients were obtained from Medical Center of the Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. For the test group, prior to blood collection, our team had visited the medical centre and informed the director about the research. We visited the centre daily speaking with typhoid patients and informed them about the research work. Those who get interested were given consent form to seek their approval to release blood samples to our team. Those that approved and signed the consent form duly, we drew blood samples from them and kept refrigerated till time for biochemical analysis. A total of 70 typhoid patients who were receiving treatment from the medical centre were surveyed for the purpose of this work. The study was approved by the ethical committee of the Department of Biochemistry, College of Natural Sciences, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria

Control group

For the control group, which comprises individuals of Michael Okpara University of Agriculture Umudike, within the age 25-40yrs, the study announcement was made open to the subjects nominated from those who replied to the announcement. A one day session was planned for the subjects giving them thorough details of the purpose of the research work. Afterward the participants were given informed consent forms and those who returned correctly filled form and signed were screened for enrolment. Prior to biochemical analysis, typhoid test (Widal test) was carried out on the control group to ensure they do not carry the typhoid vector. The control group was also screened to ensure that two weeks before the experimental day, they were not on any anti malaria therapy, typhoid treatment, not diabetic, obese and for female absence of pregnancy and were nonsmokers and alcoholics. We ensured that the control and test group were within the same age limit and were made up of both male and female.

Determination of serum urea concentration

This was done using the method of Chaney and Marbach [4]

Determination of serum creatinine concentration

This was done using the method of Taussky [5]

Determination of sodium concentration

This method was done using the method of AOAC [6] as contained in Teco test kit (U.S.A)

Determination of potassium concentration

This was done using the method of AOAC [6] as contained in Teco test kit (U.S.A)

87 **Determination of bicarbonate concentration**

88 The determination of bicarbonate ion was by the method described by Tietzel *et al.*, [7] as contained in Teco
89 bioassay test kit (U.S.A).

90 **Determination of chloride concentration**

91 The colometric determination of chloride in human was done by the method described by Tietzel *et al.*, [7] as
92 contained in Teco bioassay test kit (U.S.A)

93 **Determination of Alkaline Phosphatase Activity**

94 The activity of alkaline phosphatase was determined using the method described by King and King, [8].

95 **Determination of Aspartate and Alanine Aminotransferase (AST and ALT)**

96 The activities of AST and ALT were determined using Reitman and Frankel [9]

97 **Determination of Conjugated Bilirubin (CB) and Total Bilirubin (TB)**

98 The conjugated and total bilirubin were determined using the Max Discovery TM total bilirubin Assay Kit
99 (U.S.A.) and Diazyme's Direct Bilirubin Vanadate Oxidation assay by the colorimetric method described by
100 Jendrassik and Grof [10].

101 **Determination of Total Protein Concentration**

102 This was done using the method of Wiechselbum [11] as contained in the QuimicaClinicaApplicada (QCA)
103 Test kit (Spain).

104 **Determination of Albumin Concentration**

105 The determination was by the method of Grant [12] as contained in the Randox test kit (U.K).

106 **Total Cholesterol Estimation**

107 The serum total cholesterol was determined using the enzymatic method of Allain *et al.*, [13].

108 **High Density Lipoprotein (HDL) Cholesterol Estimation**

109 The High Density Lipoprotein (HDL) cholesterol was estimated using the method of Grove, [14]

110 **Triacylglycerol Estimation**

111 The Serum Triacylglycerol (TAG) was estimated via colorimetric method of Tietz, [15].

112 **Low Density Lipoprotein (LDL) Estimation**

113 The low density lipoprotein (LDR) cholesterol was estimated using the method described by Assman, [16].

114 **Very Low Density Lipoprotein (VLDL) Estimation**

115 Very low density lipoprotein (VLDL) cholesterol was estimated using the formula, VLDL = total cholesterol –
116 (HDL + LDL).

117 Statistical Analysis

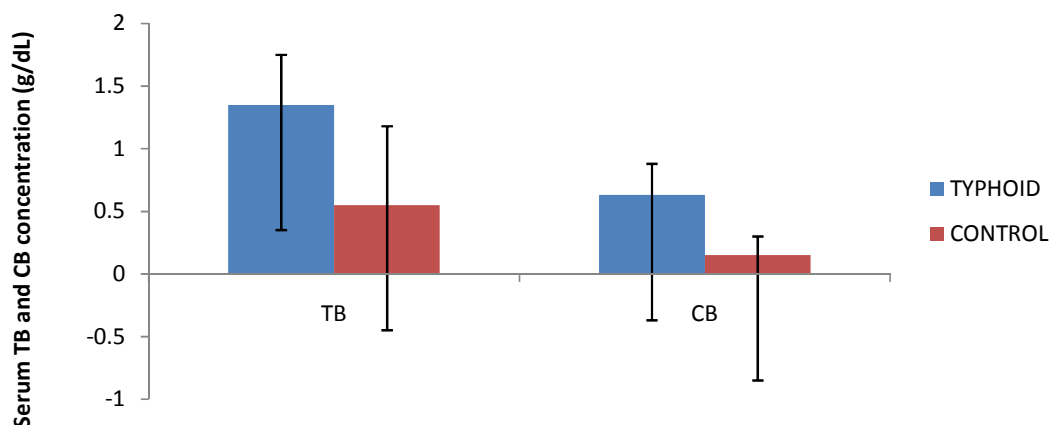
118 Statistical analysis was performed using Student T- test at 95% confidence level (SPSS VERSION 17)

119 Windows 7 Operating System

120 RESULT

121

TB AND CB CONCENTRATION IN TEST AND CONTROL GROUP



122

123 **Fig 1:** The mean serum total bilirubin and conjugated bilirubin concentration of typhoid and normal
124 individuals.

125 There was a significant ($P=.05$) difference between the mean total serum Bilirubin concentrations of the
126 typhoid infected individuals when compared with normal individuals. There was no significant ($P=.05$)
127 difference between the mean of serum Conjugated Bilirubin concentrations in typhoid infected individuals
128 when compared with normal individuals.

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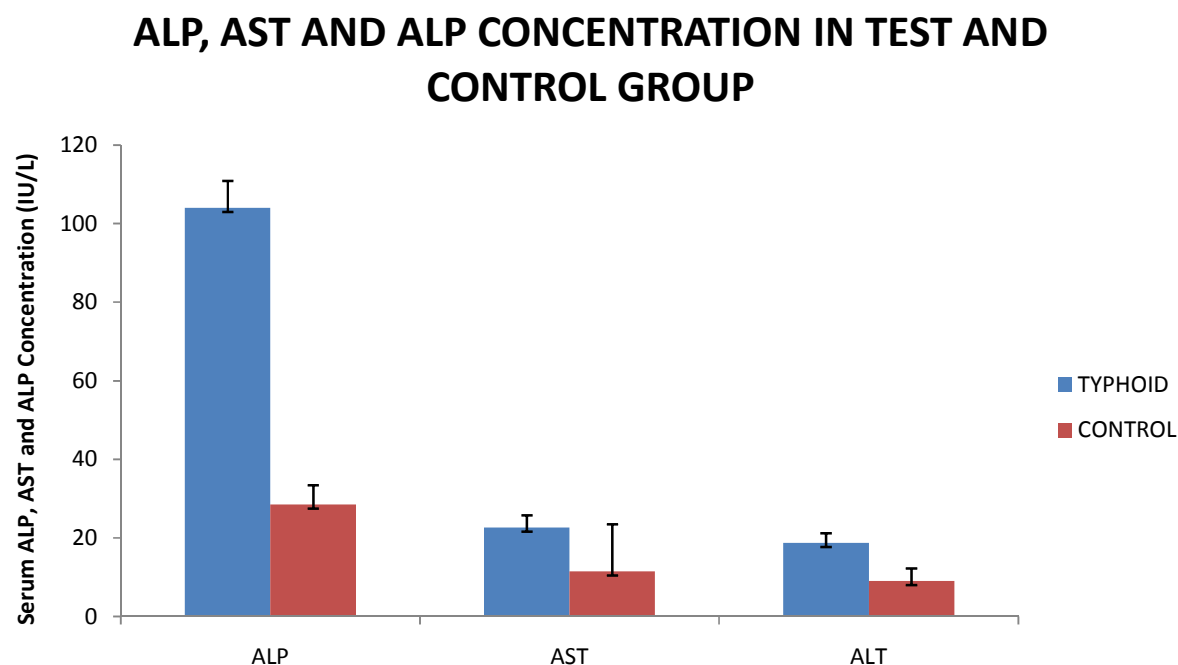


Fig. 2: The mean serum ALP, AST and ALP concentration of typhoid and normal individuals.

There was a significant ($P=.05$) difference between the mean of serum ALP concentrations in typhoid infected individuals when compared with normal individuals. Mean serum AST concentration was significantly ($P=.05$) higher in typhoid infected individuals when compared with normal individuals. Mean serum ALT concentration was significantly ($P=.05$) higher in typhoid infected individuals when compared with normal individuals.

TP, ALB AND GLOB CONCENTRATION IN TEST AND CONTROL GROUP

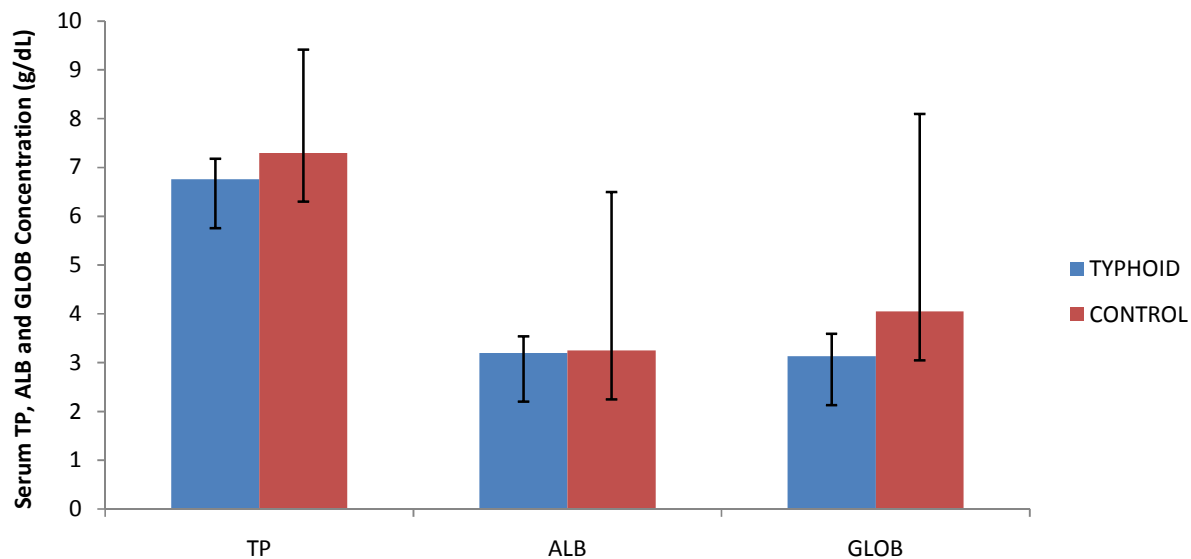


Fig 3: The mean serum TP, ALB and GLOB concentration of typhoid and normal individuals.

Mean serum TP concentration was significantly ($P=.05$) lower in typhoid infected when compared with normal individuals. Mean serum Albumin concentration was significantly ($P=.05$) lower in typhoid infected individuals when compared with normal individuals. Mean serum Globulin concentration was significantly ($P=.05$) lower in typhoid infected individuals when compared with normal individuals.

CHOL, TAGS, HDL, LDL AND VLDL CONCENTRATION IN TEST AND CONTROL GROUP

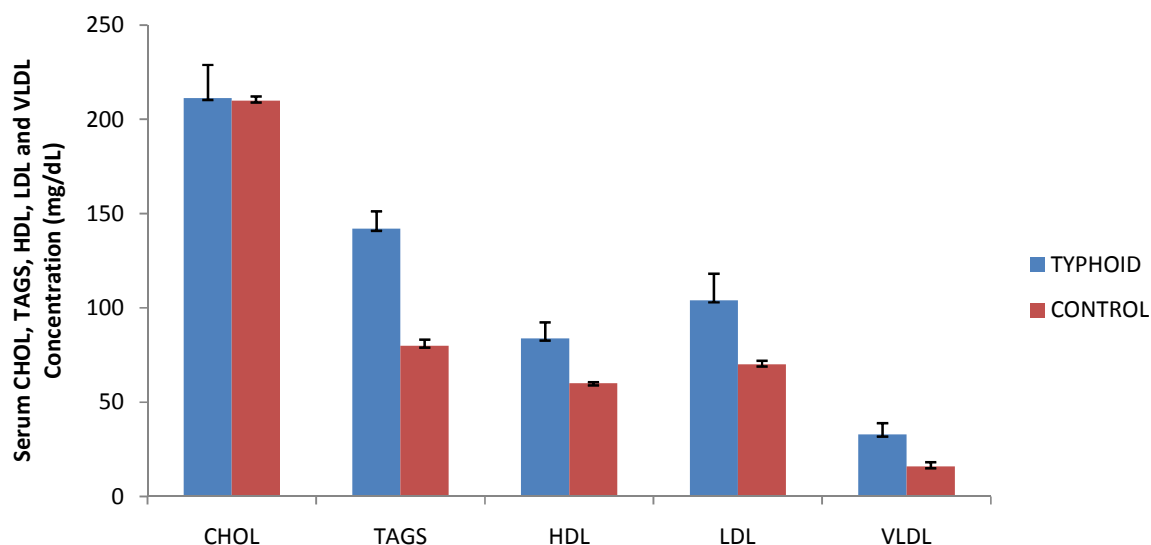


Fig. 4: The mean serum CHOL, TAGS, HDL, LDL and VLDL concentration of typhoid and normal individuals.

Mean serum cholesterol concentration was significantly ($P=.05$) higher in typhoid infected individuals when compared with normal individuals. Mean serum triacylglycerol concentration was significantly ($P=.05$) higher in typhoid infected individuals when compared with normal individuals. Mean serum HDL concentration was significantly ($P=.05$) higher in typhoid infected individuals when compared with normal individuals. Mean serum LDL concentration was significantly ($P=.05$) higher in typhoid infected individuals when compared with normal individuals. Mean serum LDL concentration was significantly ($P=.05$) higher in typhoid infected individuals when compared with normal individuals.

CREATININE AND POTASSIUM ION CONCENTRATION IN TEST AND CONTROL GROUP

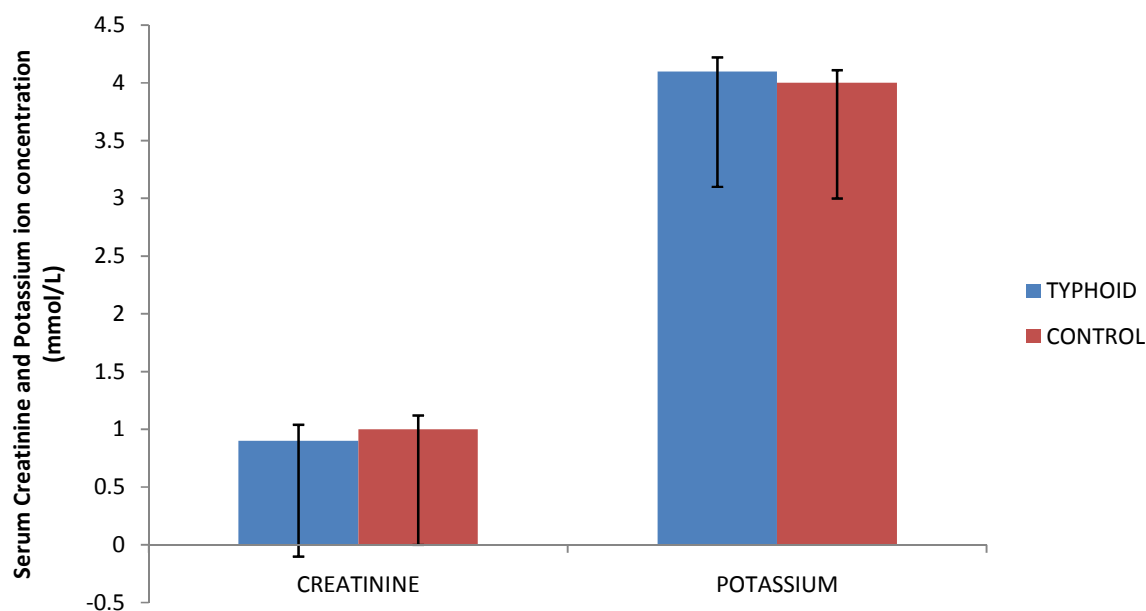


Fig. 5: The mean serum creatinine and potassium concentration of typhoid and normal individuals.

Creatinine and potassium ion concentration in typhoid and normal individual revealed no significant difference ($P=.05$)

UREA, SODIUM, BICARBONATE AND CHLORIDE CONCENTRATION IN TEST AND CONTROL GROUP

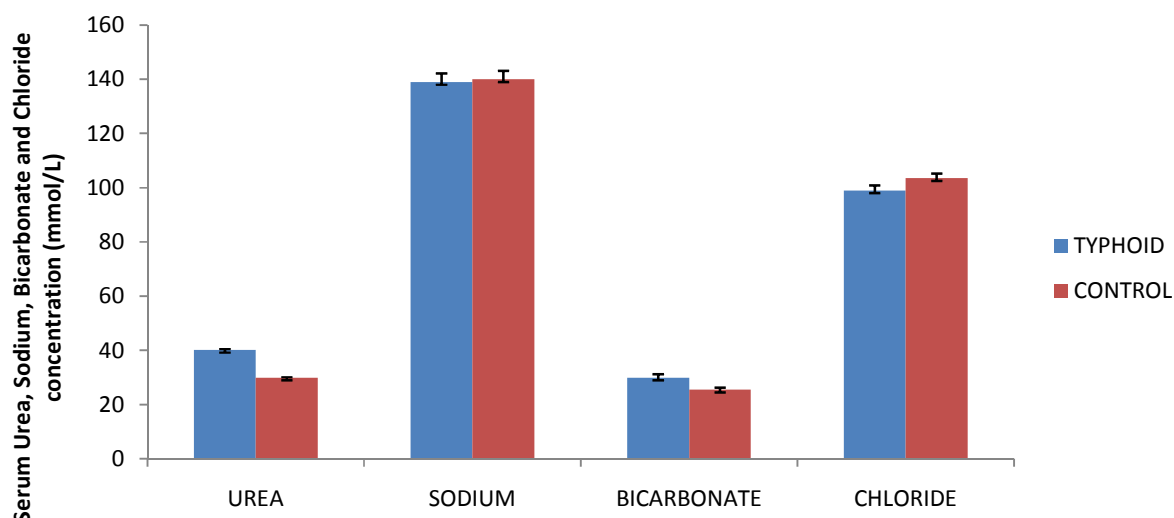


Fig. 6: The mean serum creatinine and potassium concentration of typhoid and normal individuals.

Mean value of urea concentration was significantly ($P=.05$) higher in the typhoid patient when compared with the normal individuals. Mean values for Chloride were not significantly ($P=.05$) different between the test and control. Mean values for Sodium were not significantly ($P=.05$) different between the test and control. Mean values for bicarbonate were not significantly ($P=.05$) different between the typhoid individual and normal individual control.

DISCUSSION

Typhoid fever presents with continuous fever, abdominal pain, constipation as symptoms and rose spot and relative bradycardia as signs usually. But sometimes it may present with variation in clinical finding such as diarrhea [17]. Several stages are seen in the pathophysiology of typhoid fever which is a complex process [18]. As the symptoms of typhoid fever are often nonspecific, they result in frequent diagnostic confusion with malaria, dengue fever, influenza or other febrile illness [1]. In the present study, evaluation of kidney markers (urea, creatinine, electrolytes and liver enzymes) were assayed for the purpose of selecting a marker with high sensitivity and specificity. These parameters were also assayed in healthy subjects as a control study.

Creatinine a breakdown product of creatine phosphate in muscles and is fairly constant rate by body, it only varies depending on the body muscle mass. From the result of this study, there was a significant difference between the creatinine concentrations of typhoid patient when compared to normal individual. The result revealed a significant increase of serum urea and creatinine of the typhoid patients ($P=.05$) when compared to the control. This is in contrast to the work of Sameera *et al.*, [19]. Elevated serum urea and creatinine level is often as a result of renal dysfunction due to the death of nephrons. There is a unique relationship between *S. typhi* and macrophages in the liver, spleen, intestinal lymphoid follicles, and mesenteric lymph nodes which could be responsible for the pathogenesis, [18]. Functionally active cytokines (TNF alpha, IL-1, interferon alpha and beta) are synthesized by macrophage cells and are an important source of arachidonate metabolites and reactive oxygen intermediates. These products can lead to cellular necrosis, other inflammatory cells recruitment, immune system stimulation, vascular instability, initiation of the clotting mechanism, and other abnormalities associated with typhoid fever [20].

Sodium is useful in controlling blood pressure and blood volume; it is also needed for proper functioning of the nerves and muscle, [21]. From this study, there was no significant change in serum sodium concentration of the typhoid patient when compared with the normal individual. This result was in disagreed with the findings of [22]. This bacteria *Salmonella typhi* did not affect serum sodium level in the test patient.

Result from our study also indicated that there was no significant difference in serum potassium concentration of typhoid patients when compared with normal individual. This result agrees with the findings of Sameera *et al.*, [19], and gave a clear indication that typhoid fever does not affect serum potassium level. This also implies that the typhoid does not affect the serum potassium.

Bicarbonate is a major element in our body secreted by the stomach, it is necessary for digestion, when ingested for example with mineral water, it helps buffers lactic acid generated during exercise and also reduce the acidity of dietary compound. Bicarbonate is present in all body fluid and organs; it plays a major role in acid base balance in the human body, [23]. Our findings also showed that there is no significant difference between the typhoid patient and normal individual bicarbonate ion concentration.

Chlorine is a major anion that is important in the maintenance of the cation/anion balance between intra and extra cellular fluids. This electrolyte is therefore essential to the control of proper hydration, osmotic pressure and acid- base balance [24]. In this study, there was no significant difference between the chloride ion concentration of the test and control group.

Serum total bilirubin, conjugated bilirubin also revealed no significant difference between the test and control group. Alkaline phosphatase revealed significant difference in typhoid patient when compared with normal individual. The concentration of Aspartate amino transferase (AST) and Alanine amino transferase (ALT) is for detecting hepato cellular injury and may help in monitoring the status of liver. Both enzymes increased in many hepatic diseases and have limited value in differential diagnosis. However, amino-transferases are considered useful in differentiating hepatocellular from cholestatic forms of liver injury. AST activity is related to damage of cell in kidney, pancrease and erythrocytes. ALT and AST were significantly higher in typhoid patients than normal individual. This was in contrast to the result obtained from Shaminet *al.*, [25] finding. In general, mechanism relating to association between liver marker and in typhoid fever may reflect elevations in ALT and AST. Serum total protein was significantly lower in typhoid patients as compared to normal counterparts.

Albumin and globulin were also significantly lower in typhoid patients as compared to normal individuals from the result of this study. This also disagrees with the finding of Shaminet *al.*, [25]. If the globulin levels fall below the normal ranges, it can be assign of several serious health conditions which are tonal diseases, hepatic dysfunctions; celiac disease etc. gammaglobulenemia and hypogammaglobulinemia can cause the globulin levels to drop. This is also a sign that proteins taken in by the digestive system are not being broken down or absorbed properly. High levels of globulin can be caused by an overproduction of globulin, an underproduction of albumin or a loss of albumin. Albumin can be lost due to kidney disorders which lead to an excessive amount of protein being shed from the body.

For lipid profile, our study revealed distinction in the lipid profile of the test group when compared with the control group as indicated by the result. The typhoid patients had increased cholesterol concentration when compared with the control group. This finding opposes the result of Amal [26], who reported a decrease in cholesterol concentration in typhoid patients.

243

244 HDL, LDL, VLDL and triacylglycerols also increased in typhoid patient when compared with normal
245 individuals. Unbalances in lipoproteins fractions can be attributed to lipid peroxidation attack which may
246 affect their regular metabolism and the successive delivery of lipid to peripheral organs in the body, [27].

247

248 The result revealed a significant increase in the level of total cholesterol in typhoid patients. Total
249 cholesterol (TC), triacylglycerol (TAG), and the lipoprotein system (HDL, LDL) concentrations can function
250 as marker or indicators of cardiovascular status.

251

252 In the studied typhoid group, there was an abnormal cholesterol level (hypercholesterolemia) that is, higher
253 concentrations of LDL when compared with the control group, which is strongly associated with
254 cardiovascular disease because these promote atheroma development in arteries (atherosclerosis). This
255 disease process leads to myocardial infarction (heart attack), stroke, and peripheral vascular disease [28].
256 LDL particles are often termed "bad cholesterol" because they have been linked to atheroma formation [29]

257

258 In the typhoid group, Increase in TAGs concentrations indicated an increase in total cholesterol level. High
259 levels of TAGs in the bloodstream have been linked to atherosclerosis and, by extension, the risk of heart
260 disease and stroke.

261

262 Typhoid fever is associated with inflammation and ulceration of the gut and liver of which oxidative lipid
263 modification plays an important role in exacerbating the disease. Increase in HDL levels and increase in
264 LDL-cholesterol is observed in patients with typhoid at the peak of fever.

265 The association between increased LDL cholesterol levels and increased risk of heart disease has been
266 well established and documented, independently of TG levels and other risk factors [30]. Intravascular
267 ultrasound studies demonstrate that patients with low HDL cholesterol and high TG levels have more
268 extensive coronary atheromas than those with an isolated elevation of LDL cholesterol [31]. Any substance
269 or condition that increases the LDLs concentration and lowers the HDLs tends to increase the probability of
270 plaque formation and a heart attack [32].

271

272 **CONCLUSION**

This study appears to be ample evidence based on the physiological and biochemical parameters in typhoid patients to explain influence of typhoid morbidity. Typhoid fever causes high incidence of biochemical enzymatic changes but these changes are covered up by antibacterial therapy. For proper management and possibly treatment of typhoid, more emphasis should be targeted towards increased excretion of the blood nitrogen based products, initiation of early treatment through early diagnosis can considerably decrease the morbidity and mortality caused by typhoid and drugs which have the ability to increase the rate at which urea and creatinine are eliminated from the body should be administered. Patients who suffer from typhoid have persistent fever, fatigue, loss of appetite, severe headache, swollen spleen, constipation or diarrhea situation. Such as lack of treatment, mortality may reach one-tenth, but if early treatment is administrated then the mortality rate can be dropped to one per cent or less.

ETHICAL APPROVAL

This study was approved by the ethical committee of the Department of Biochemistry, College of Natural Sciences, Michael Okpara University of Agriculture, Umudike. It was given approval number MOUAU/COLNAS/BCM/2014/005, specific for human study

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