# BIOCHEMICAL ANALYSIS OF SERUM ENZYMES AND ELECTROLYTES OF TOXOPLASMA GONDII INFECTED WOMEN

# SYED ANWAAR HUSSAIN, RAZIA SULTANA, MUHAMMAD NAVEED AKHTAR, AZHAR MAQBOOL,

# Department of Livestock and Dairy Development, Govt. of the Punjab, Lahore (SAH, RS, MNA); Pakistan, University of Veterinary and Animal Sciences, Lahore (AM), Pakistan

Abstract: Biochemical analysis of serum enzymes (SGPT, SGOT, LDH) and serum electrolytes (Na, K, Ca and Mg) were recorded. A total of 50 serum samples from cases of pregnant and aborted women were collected at random. On the basis of serological examination of serum samples, Toxoplasma gondii positive at 1/16 dilution (Group A), T. gondii positive at 1/256 dilution (Group B) and T. gondii negative (Group N) were subjected to biochemical analysis. There was significant difference (P < 0.01) for serum enzymes among all the three treatment groups of women. The level of serum enzymes was much higher in the cases positive for Toxoplasmosis at 1/256 dilution (Group B) then in the cases positive for Toxoplasmosis at 1/16 dilution (group A) and in normal cases (Group N) of women. However, there was no significant difference (P > 0.01) for serum electrolytes among all the three treatment groups of women. Thus the results showed that the serum enzymes were affected by T. gondii but serum electrolytes remained unaffected during Toxoplasma gondii infection in women.

**Keywords:** Protozoan parasite, toxoplasmosis, zoonotic disease, disease biochemistry, transaminases, SGOT, SGPT, LDH

# INTRODUCTION

T oxoplasma gondii is a complex protozoan parasite with world wide in distribution. It is a zoonotic parasite. It is estimated that approximately one third of the world population has been exposed to *T. gondii* (Rizwan *et al.* 1999). Cats serve as the only definitive host of *T. gondii* and are the only host in which the organism undergoes sexual reproduction within the intestines.

0079-8045/08/0073-0080 \$ 03.00/0

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#### S. A. HUSSAIN ET AL.

Toxoplasmosis is of serious concern in pregnant women. The fetus is in danger if mother is exposed first time during pregnancy. It is estimated that in U.S.A. at least 3000 babies are born each year with congenitally acquired Toxoplasmosis showing lymphadenopathy (Markell *et al.*, 1992)

Very little work has been done on the biochemical analysis of serum enzymes and electrolytes in the cases of toxoplasmosis in women. Keeping in view the importance of this zoonotic disease the current project was designed to study the biochemical aspects of serum of women for enzymes and electrolytes and to compare the recorded values with the standard values.

# **MATERIALS AND METHODS**

#### **Collection of Samples**

A total of 50 serum samples from cases of pregnant and aborted women were collected at random from Gynecology Ward, Divisional Head Quarter Hospital, Faisalabad. All the relevant information were recorded on Performa regularly. Under aseptic measures, 6-8ml of blood was withdrawn by vein-puncture with the help of disposable syringes and was transferred to a screw capped sterile clean test tube slowly to avoid haemolysis (Benjamin, 1985).

#### Separation of Serum

All the blood samples were labeled with the species, outdoor registered number and the date of collection. The samples were left for about an hour for blood clotting to occur and were further processed for analysis (Samaha *et al.*, 1995). Reported freezing and thawing was avoided.

#### Analysis of the Serum Samples

All the serum samples were analyzed for specific IgG and IgM antitoxoplasma antibodies using Latex Agglutination (LA) test. For this purpose, the commercial test kit used was "Toxoplasmosis Latex" manufactured by Quimica Clinica Aplicada, S.A. Amposta Spain.

On the basis of serological examination of serum samples, *Toxoplasma gondii* positive at 1/16 dilution (Group A), at 1/256 dilution

#### BIOCHEMICAL ANALYSIS OF SERUM ENZYMES AND ELECTROLYTES 75 OF *T. GONDII* INFECTED WOMEN

(Group B) and *T.gondii* negative (Group N) samples were subjected to biochemical analysis.

# Serum Enzymes Assay

All the collected serum samples were analyzed on UVIDEC 430B double beam spectrophotometer, by JASCO company of Japan, in the biochemical laboratory Punjab Medical College, Faisalabad. Diagnostic kits were used in the above mentioned automated analyzer for the quantitative estimation of following enzymes.

- a. Alanine Aminotransferase-ALT (SGPT)
- b. Aspartate Aminotransferase-AST (SGOT)
- c. Lactate Dehydrogenase-LDH

Following diagnostic kits were used during this enzyme kinetic study.

- 1. Medela Diagnostic (cat. No. 330008) ALT (SGPT) SCE Method
- 2. Medela Diagnostic (cat. No. 330006) AST (SGOT) SCE Method
- 3. Menagent LDH/LD (Code B-8130) LDH

# **Biochemical Analysis**

# a. Estimation of Alanine Amino transferase-ALT (SGPT)

Elevated ALT (SGPT) levels in blood, which was determined through following procedure.

L-Alanine + $\alpha$ -Ketoglutara	ate <u>ALT</u>	Glutamate + Pyruvate
Pyruvate + NADH	LDH	Lactate + $NAD^+$

This reaction was monitored by the measurement of the decrease in absorbance at 340nm as NADH is converted to NAD in serum samples.

# b. Estimation of Aspartate Amino Transferase

Elevated AST levels in blood were estimated through following procedure.

#### S. A. HUSSAIN ET AL.

 $\begin{array}{c|c} Aspartate + \alpha - Ketoglutarate & AST \\ Oxaloacetate + NADH & MDH \end{array} \\ \begin{array}{c} Oxaloacetate + Clutamate \\ Malate + NAD^+ \end{array} \end{array}$ 

This reaction was monitored by measurement of decrease in absorbance at 340nm as NADH was converted to NAD in serum samples.

#### c. Estimation of Lactate Dehydrogenase

Lactate dehydrogenase catalyze the following reaction.

 $Pyruvate + NADH + \underline{H^{+} LDH} Lactate + NAD^{+}$ 

This absorbance diminution of NADH + H<sup>+</sup> in time when measured at 340nm is directly proportional to the LDH value.

#### Serum Electrolytes Assay

# Determination of serum Na and K

The serum Na and K concentrations were determined by means of atomic spectrophotometer (Flame Photometry) using lithium or cesium as an internal standard. The procedure was described by Frenkel *et al.* (1970).

#### Determination of Serum Ca and Mg

The concentrations of Ca and Mg in serum determined by Atomic absorption spectrophotometer using, Perkin Elmer Analyst 100.

# **RESULTS AND DISCUSSION**

The results of the different serum enzyme levels *i.e.*, Aspartate Amino-Transferase (SGOT), Alanine Amino Transferase (SGPT) and Lactate Dehydrogenase (LDH) and serum electrolytes *i.e.*, Sodium (Na), Potassium (K) Calcium (Ca) and Magnesium (Mg) in normal and *T. gondii* infected cases of women, at different screening dilution of 1:16 and 1:256 were given in the present study.

#### A.SERUM ENZYMES

# 1. Aspartate Amino Transferase – AST (SGOT)

Aspartate aminotransferase catalyzes the transamination of Laspartate and 2-oxoglutarate to oxaloacetate and glutamate. The presence

#### BIOCHEMICAL ANALYSIS OF SERUM ENZYMES AND ELECTROLYTES 77 OF *T. GONDII* INFECTED WOMEN

of AST in so many tissues makes their serum level a good marker of soft tissue damage but precludes its use an organ specific enzyme (Boyd, 1983).

Table I showed the enzyme activity in units/1, for women (N = 11.55 to 15.50, A = 18.70 to 24.60 and B = 28.45 to 32.75). The results were found to be highly significant when analysis of variance was applied (P<0.01). LSD test shows that the three treatment groups had significant difference among each other which revealed that *T. gondii* affected enzyme activity in women, significantly.

Moss and Buttorworth (1974) also stated that the use of enzyme estimation in serum in the detection of acute or chronic damage to cells which cause the leakage of enzymes into extracellular fluid and then in the blood provides an extremely sensitive index of deterioration of plasma membrane. Since enzymes can be detected by their catalytic activity as in case of AST level in serum of affected women.

#### 2. Alanine Aminotransferase-ALT (SGPT)

ALT in the serum is a sensitive live-specific indicator of damage so that it is used as an indicator of hepatopathy in toxicological studies (Boyd, 1983) Table I showed the enzyme activity in units/1, for women (N = 13.45 to 16.67, A = 20.25 to 24.70 as B = 29.35 to 33.40).

The statistical results were found to be highly significant when analysis of variance was applied (P < 0.01). L.S.D. value indicated that all the three treatment groups had significant difference among each other, which also revealed that *T. gondii* affected significantly enzyme activity in women.

# 3. Lactate Dehydrogenase-LDH

Tissue contains various amounts of the LDH-isoenzymes and serum isoenzymes profiles have been used to identify specific tissue damage by electrophoretic separation (Prasse, 1969).

Table I shows the enzyme activity in units/1, for women (N = 120.40 to 190.70, A = 180.90 to 250.55 and B = 210.70 to 320.45).

The results were found to be highly significant (P < 0.01). LSD value indicated that all the three treatment groups had significant difference among each other, which also revealed that *T. gondii* affected significantly enzyme activity in women, as was also noted by Benjamin (1978).

According to this study LDH may be elevated in many disease processes in which there is cell necrosis.

# **B. SERUM ELECTROLYTES**

# 1. Sodium (Na), Potassium (K), Calcium (Ca) and Magnesium (Mg)

Serum Na, K, Ca and Mg concentration (m Eq/1) in Normal (N), seropositive for Toxoplasmosis at 1/16 dilution (A) and seropositive for Toxoplasmosis at 1/256 dilution (B) cases of women, had been presented in Table II.

Serum concentrations of Na, K, Ca and Mg were statistically found similar in Normal cases and *T. gondii* affected cases of women. The results were found to be non-significant (P > 0.01) which also revealed that *T. gondii* did not affect the Na, K, Ca and Mg concentrations in women.

Table-I: Serum Alanine Aminotransferase (SGOT) serum Aspartate Aminotransferase (SGPT), serum Lactate Dehydrogenase (LDH) activity (unit/1) in Normal (N), seropositive for Toxoplasmosis at 1/16 dilution (A) and seropositive for Toxoplasmosis at 1/256 dilution (B) cases of women.

	Treatment groups	Mean value	Range	S.E
				•
SGOT	Ν	12.75	11.55-15.50	0.27
	А	21.44	18.70-24.70	0.48
	В	30.31	28.45-32.75	0.40
SGPT	Ν	14.07	13.45-16.67	0.20
	А	21.68	20.25-24.70	0.35
	В	30.62	29.35-33.40	0.29
LDH	Ν	153.57	120.40-190.70	1.87
	А	241.11	180.90-250.55	3.29
	В	283.85	210.70-320.45	2.74

#### BIOCHEMICAL ANALYSIS OF SERUM ENZYMES AND ELECTROLYTES 79 OF *T. GONDII* INFECTED WOMEN

Table II: Serum Concentrations of Sodium (Na), Potassium (K), Calcium (Ca)Magnesium (Mg) (mEq/1) in Normal (N), seropositive forToxoplasmosis at 1/16 dilution (A) and seropositive forToxoplasmosis at 1/256 dilution (B) cases of women.

Element	Treatment groups	Mean value	Range	S.E.
Na	Ν	146.79	140.50-149.70	1.23
	А	149.44	147.40-154.60	2.17
	В	146.31	141.70-149.80	1.86
K	Ν	4.49	3.75-5.70	0.14
	А	4.59	3.90-5.85	0.25
	В	4.30	3.660-5.40	0.21
Са	Ν	7.84	6.90-8.00	0.04
	А	7.82	7.10-8.00	0.06
	В	7.85	7.05-8.10	0.05
Mg	Ν	2.48	2.10-2.70	0.06
	А	2.49	2.5-2.65	0.11
	В	2.56	2.20-2.90	0.09

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(Received: April 11, 2008; Revised: May 22, 2008)