Some Antioxidants Level In Seropositive Toxoplasmosis Woman In Mosul

Elham M. AL-Khshab

Department of Physiology , College of Veterinary Medicine , Mosul , Iraq (Received 1/9/2008 , Accepted 17/5/2009)

Abstract

This Study was conducted to estimate the level of some antioxidant , including (Vitamin c, Glutathion (GSH), albumin, and Superoxide dismutase (SOD) activity). Also to determine Peroxynitrite radical (one of the reactive oxygen species ROS) and total protein in (88) infected women with Toxoplasmosis (Whose – anti – Toxoplasma gondii antibodies were Positive), in addition to (31) women, (whose anti- Toxoplasma gondii antibodies were negative as control group). The results showed that , serum vitaminC and glutathione levels were significantly lower (P<0.05) in toxoplasmosis women than control group, however there was a significant increase (P<0.05) in serum total protein and peroxynitrite radical in toxoplasmosis women, compared with control group . The study showed a significant reduction (P<0.05) in albumin / globulin ratio in toxoplasmosis woman, while there were no changes in serum albumin and superoxide dismutase (SOD) activity in these infected women when compared with control group .

This study concluded that , infection with Toxoplasma play a role in oxidative stress induction and reduced the antioxidant – defens systems .

Introduction

Toxoplasma gondii is widespread prodozoan Parasite, that infects most types of warm-blooded mammals and causes opportunistic disease in humans(1).

Maternal infection with *Toxoplasma gondii* acquired during Pregnancy causes a serious health risk to the fetus. Human infection usually occurs by ingestion of materials contaminated with cat feces or by the eating poorly cooked meat, which contains Toxoplasma cysts(2).

In Europe, congenital toxoplasmosis occur in1–10/1000 new born babies, of whom1-2%develop learning difficulties and 4-27% develop retinochoroidal lesions leading to Permanent unilateral impairment of vision (3,4). Usually asymptomatic in immunocompetent individuals. Toxoplasmosis may cause sever disorders in immunocompromised patients and in Pregnant women, because of high risk of transplacental transmission and the occurrence of multiple congenital lesions in the fetus (5). During infection, the immune effector cells are able to kill or inhibit its intracellular growth. This antiprotozoan activity produces a number of toxic products such reactive oxygen intermediates. While, within the host cell, T.gondii itself produces oxidants as by products of normal metabolism. Reactive oxygen species(ROS) are potentially destructive, capable of oxidizing proteins or lipids and causing chemical modifications to nucleic acids(6). It is well documented that, under normal physiological conditions, an estimated 1-3% of inspired oxygen is converted to superoxide radicals and H₂O₂. The existence and development of cells in an oxygen containing environment would not be possible with out the presence of complicated defense system that include enzymatic and nonenzymatic antioxidant components(7). The aim of the study is to estimate some antioxidante level in serum of positve toxoplasmosis women.

Materials and Methods

Blood samples were collected from (88) seropositive—Toxoplasmosis women and from (31) sero negative—Toxoplasmosis women (Control group). All these women attended the central laboratory in Mosul city from (Febriuary—July 2005). Non of toxoplasmosis women and control group took any medication or supplemention.

Serological test was done using Latex Agglutination with Toxo-Cell-Latex kit (Bio Kit – Span).

Serum vitamin C was determined according to Stanley and others(8) which depends on the oxidation of ascorbic acid by copper and by measuring the Product at 520 nm. Glutathione (GSH) was detected by using the modified method of Sedlak and Lindsay (9) this method based on the reaction of glutathione with Elman's reagent DTNB [(5,5 –dithiobis (2-nitrobenzoic acid)], the product was measured at 412 nm.

Modified photochemical Nitroblue Tetrazolum –method was used for estimation the SOD activity(10)which depends on the measurement of absorbance, where the increase in absorption , reflects the decrease in activity .

Method by Vanuffelen and others(11)was used for determination the peroxynitrite radical level which based on the nitration of phenol to produce nitrophenol, that was measured at 410 nm.

Biuret method was used for detection the total Protein concentration(12). Albumin was measured using kit supplied from syrbio company / syria.

Statistical analysis was done by using student T test & the P< 0.05 was regarded as significant & all data were presented as mean \pm S-D .

Results & Discussion

The results in table(1) and Fig.(1) revealed a significant decrease(P< 0.05) in serum vitamin C in women affected with T.gondii Compared with control group, this decrease in vitamin C level in the affected women may be due to the production oxidant as free radicals by the parasite T-gondii therefore vitamin C as antioxidant, prevents the body from these radicals by oxidized itself, in addition to the chemical properties of vitamin C, that allow the direct interaction with O_2 and OH in the aqueous phase of plasma, thus preventing damage to cells. Vitamin C plays a role in reproduction of-Tocopherol from Tocopherol radical E, which is produced from the inhibition of lipid Peroxidation by vitamin E, therefore both vitamin C and E conserve the cell wall from damage. (13, 14).

As shown in table(1) and Fig.(2)glutathione decreased significantly (P<0.05)in serum of Toxoplasmosis woman, compared with control group.(-glutamyl cysteinyl glycine)GSH is the most abundant non protein thiol

source in the cell which serves multiple functions in protecting tissues from oxidative damage and keeping the intracellular environment in the reduced state. GSH reduces hydrogen and organic - peroxides via a reaction catalyzed by GSH Peroxidase, it serves as a scavenger of (OH)and siglet oxygen(${}^{1}O_{2}$)(15).GSH reduces tocopherol radicals, either directly or in directly by reducing semidihydroascorbate therby preventing Free radical chain reaction and lipid Peroxidation (16). In addition, GSH depletion may occur during oxidative stress, caused by cellular demand and lead to impaired cell function. Glutathione is the most significant component, which directly quenches ROS such as lipid peroxides and plays major role in xenobiotic metabolism, when an individual is exposed to high levels of xenobiotics, more glutathione is utilized for conjugation making it less available to serve as antioxidant. Furthermore, it maintains vitamin C and E in their reduced form, which exerts an antioxidant effect by quenching free radicals (17).

Peroxynitrite radical level elevated significantly(P<0.05)in serum of Toxoplasmosis women, compared with control group as shown in table(1)and Fig.(3). The peroxynitrite is mainly formed closer to O₂ generation sites, under condition such as, inflammatory states, in which both formation rates of No and O₂ increase (18) *Toxoplasma gondii* could damage the cells, this may lead to release of nitric oxide, which is considered as vasodilator agent that contributes to the increase in the ability of the parasite to attack the arterial walls(19).

Table(1) and Fig.(4) show that, there was no significant differences in serum SOD activity between Toxoplasmosis women and control group, although others pointed to the presence of endogenous SOD, GPX and catalase, that related to the protection from ROS attacks (20).

Serum total protein increased significantly(P<0.05)in Toxoplasmosis women compared with control group as shown in table(1)and Fig(5). This result agreed with others(21)they noted that serum protein increased as increasing in gama globulin fraction in infected dogs with T. gondii. Also this result agree with AL-Zuhairy (22) who noted a significant increase in total protein concentration in sheep infected with Toxoplasma and in experimental infected rats with Toxoplasmosis and it has been explained that this elevation may be due to the increase in globulin levels that caused by the infection. In addition, Abernga and Anosa showed an increase in total protein concentration in serum of experimental Gambian trypanosomosis and attributed this increase to the higher levels of globulins with out any change in albumin concentration (23).

In the present study albumin/globulin ratio decreased significantly in sero positive Toxoplasmosis woman in compared to the control group, this may be because of the increase in the globulin concentrations, where there was no changes in albumin concentration in the infected women. table(1)Fig. (6). Fig.(7).

Table (1) Antioxidants levels in Toxoplasmosis women

Parameter	Control group $n = 31$	Toxoplasmosis women $n = 88$	P-Value
	mean \pm SD	mean \pm SD	
Vitamin C µg/ml	6.08 ± 0.92	3.78 ± 0.77	< 0.05
Glutathione µmol/L	1.54 ± 0.102	0.948 ± 0.15	< 0.05
Albumin g/dL	3.5 ± 0.18	3.6 ± 0.27	> 0.05
T.Protein g/dL	6.9 ± 0.13	8.8 ± 0.39	< 0.05
SOD	$0.0156 \pm 6.88 \times 10^{-3}$	$0.0151 \pm 5.39 \times 10^{-3}$	> 0.05
(oxidant) Peroxynitrite µmol/L	76.2 ± 7.4	123.43 ± 24.26	< 0.05
Albumin /globulin	1.029 ± 0.07	0.69 ± 0.029	< 0.05
Thousann / Stocum	1.02/ _ 0.0/	0.07 = 0.027	\ 0.05

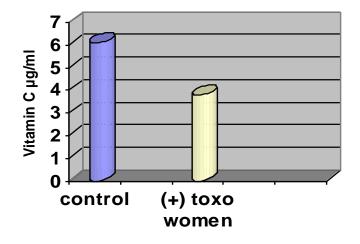
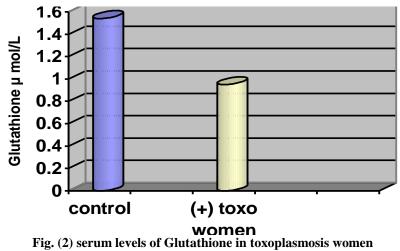


Fig. (1) serum levels of vitamin C in toxoplasmosis women



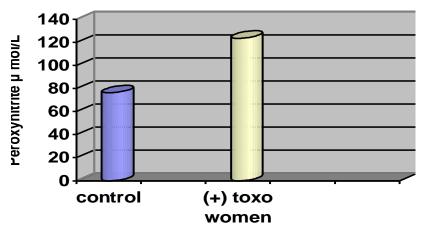


Fig. (3) serum levels of peroxynitrite in toxoplasmosis women

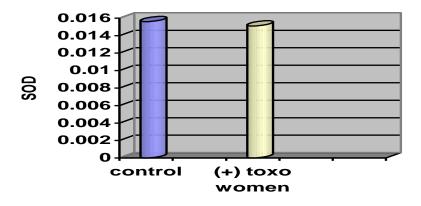


Fig. (4) serum levels of SOD in toxoplasmosis women

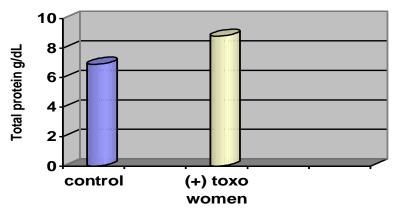


Fig. (5) serum levels of total protein in toxoplasmosis women

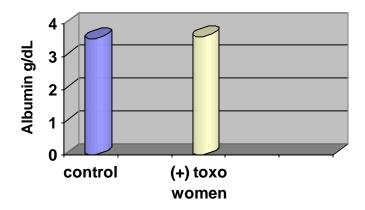


Fig. (6) serum levels of albumin in toxoplasmosis women

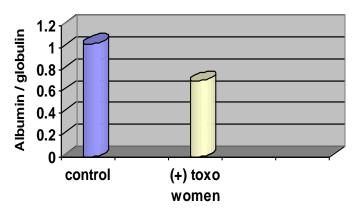


Fig. (7) Albumin / globulin ratio in toxoplasmosis women

Refences

- Dubey J., Chunlei su., Daniel K., Howe K. and David S. Identification of quantitative trait Ioci controlling acute virulence in *Toxoplasma* gondii. Am ,Vet, Med, Assoc.205:1593-1598. (1994).
- 2- Bowie W. R., King A. S., Werker D. H., et al., Outbreak of toxoplasmosis associated with municipal drinking water . *Lancet* 350:173-177. (1997).
- 3- Gilbert R. E., Buffolano W., Zufferey J., Petersen E., Jenum P. A., Foulon W. and

- Semprini E. European Research Network on Congenital Toxoplasmosis. *BMJ*, 321:142-7.(2000).
- 4- Lebech M., Anderson O., Christensen N. C. and Hertel J. Feasibility of neonatal screening to toxoplasma infection in the absence of prenatal treatment . *Lancet.*, 353:1834-7.(1999).
- 5- Foulon W., Villena I. and Stray-petersen B. Treatment of toxoplasmosis during pregnancy a multicenter study of impact on fetal transmission and children's seqwelae at age 1 year . *Am. J. Obstet. Gyneco* . 180:410-415. (1999).
- 6- Halliwell, B. and Gutteridge, J. M. Antioxidant defenses in Free radicals in biology and medicine. PP, 105 245. Oxford University Press, New York. (1999).
- 7- Guohua, C. and Ronald, L. Postrandial increase in serum antioxidant capacity in older women. *J. Appl Physiol*. 89: 877 883.(2000).
- 8- Stanley T., David T.and Howerds S. "Selected method for determination of ascorbic acid in animal cells, tissues and fluids" Method in Enzymology, Vol. 62. Vitamins and Coenzymes Part D.(1979).
- 9- Sedlak J. and Lindsay R. H. "Analytical biochemistry" p.192 (1968). cited by Al-Zamely(2001). Al-Zamely O. M., Al-Nimer M. S., and Muslih R. K. "Detection the level of peroxynitrite and related with antioxidants status in the serum of patient with acute myocardial infraction" National Journal of Chemistry, 4:625-637.(2001).
- 10- Brown M. S. and Goldstein Ann. Rev. Biochem. 52,223.(1983). cited by Al- Zamely et al., 2001.
- 11- Vanuffelen B. E., Van Derzec J. Dekoster B. M. *Bio Chem. J.*, 330-19.(1998).
- 12- Plummer D.T. "An Introduction to Practical Biochemistry". 2nded., Mc Gaw Hill Book company, UK. P. 142. (1987).
- 13- Lee S. H., Oe T.and Blair I. A. Vitamin C-induced decomposition of lipid hydro peroxides to endogenous genotoxins. Science., 292:2083-2086 (2001).
- 14- Grodner M., Long S. and De Young S. "Foundations and Clinical Applications of Nutrition" 3rded. (2004).15-Hill M. F. and Singal P. K. Right and left myocardial responses during heart failure subsequent to myocardial infarction. Circulation. 96:2414-2420. (1997).
- 16- Yvonne V., Yuan and David D. Dietary (n⁻³)fat and cholesterol Alter tissue antioxidant enzymes and susceptibility to oxidation in SHR and WKY Rats. The American Society of Nutritional sciences J. Nutr., 133:679-688. (2003).
- 17- Singh, R. P., Sharad, S. and Kapur, S. Free radicals and oxidative stress in Neurodegenrative Diseases: Relevance of Dietary Antioxidants. *JIACM*, 5(3):218-25 .(2004).18- Beckman, J. S., Beckman, T. W., Chen, J., Marshall, P. A. and Freeman, B. A. Apparent hydroxyl radical production by peroxynitrite: Implications for endothelial injury from nitric oxide and superoxide *Pro* . *Natl. Aead. Sci.* USA., 87: 1620 1624 (1990).
- 19- Denkers E. Y., Butcher B.A., DelRio L. and Bennouna S. Neutrophiles, dendritic cells, and Toxoplasma. Int J. Parasitol., 34:411-421. (2004).
- 20- Kaasch, A. j. and Joiner, K. A. Targeting and subcellular localization of *toxoplasma gondii* catalase.

Identification of peroxisomes in an opicomplexan parasite *J. Biol Chem*, 275:1112 – 1118.(2000).

- 21- Nisbet, C, Yarim, G., Cenesiz, D., Oncel, T. and Cittci, G. Serum protein alterations in dogs naturally infected with *Toxoplasma gondii Parasitol*. Res. (2007). 22- AL Zuhairy, A. S. Some Biochemical Parameters in sheep infected with Toxoplasmosis M. Sc A thesis college of Medicine, University of Mosul, Iraq.
- 23- Abenga, J. N.and Anosa, V. O. serum total protein and creatine levels in experimental Gambian trypanosomosis of Vervet monkeys, African Journal of Biotechnology; 4(2): 187 190.(2005).

مستوى بعض مضادات الأكسدة في النساء المصابات بداء القطط في الموصل الهام محمد الخشاب

فرع الفسلجة ، كلية الطب البيطري ، جامعة الموصل ، الموصل ، العراق (تاريخ الاستلام: ١/ ٩ / ٢٠٠٩)

الملخص

تستقصي هذه الدراسة مستويات بعض مضادات الأكسدة والتي نتضمن (فيتامين C، الكلوتا الألبومين، وفعالية إنزيم السوبراوكسايد، دسميوتيز) ومستوى جذر البيروكسي نيتريت (احد الأصناف الفعالة للأوكسجين ROS) ومستوى البروتين الكلي في أمصال (88) امراءة مصابة بداء القطط بالإضافة إلى (31) امراءة غير مصابة (اعتبرت مجموعة سيطرة).

بينت النتائج وجود انخفاض معنوي (P < 0.05) في مستوى فيتامينP = P والكلوناثايون في أمصال النساء المصابات بداء القطط مقارنة مع مجموعة السيطرة في حين كان هناك ارتفاعا" معنويا" (P < 0.05) في مستوى البروتين الكلي وجذر البيروكسي نيتريت في النساء المصابات مقارنة بمجموعة السيطرة .

كما بينت الدراسة أيضا وجود انخفاض معنوي (P<0.05) في نسبة الألبومين/ الكلوبيولين في النساء المصابات مقارنة بالسيطرة في حين لم يكن هناك تغيير في مستوى الألبومين وفعالية إنزيم السويراوكسايد دسميوتيز في النساء المصابات بداء القطط مقارنة بمجموعة السيطرة .

تستنتج هذه الدراسة، أن الإصابة بداء القطط يلعب دورا في إحداث الكرب التاكسدي وبالتالي إضعاف الأنظمة الدفاعية المضادة للأكسدة .