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RESEARCH ARTICLE

INCREASED OXIDATIVE STRESS REDUCES THE LEVEL OF SERUM PARAOXONASE-1(PON-1) AND OTHER BIOCHEMICAL MARKERS IN PATIENTS WITH PULMONARY TUBERCULOSIS (PTB) IN NEPALESE POPULATION.

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Abstract

Objectives: The aim of this study was to assess the total peroxides level and paraoxonase (PON1) activities in patients with Pulmonary tuberculosis (PTB) in Nepalese population.

Material and methods: A total of 70 (40 newly diagnosed active PTB and 30 healthy control) subjects were recruited for this study. Anthropometric variables, total peroxide and PON1 activities were determined in control and PTB subjects.Serum glucose, urea, total protein, albumin, globulin and uric acid levels were also determined in the participants.

Results: Significant difference in BMI, SBP and DBP was observed between PTB and control subjects ($p<0.001$, $p<0.01$ and $p<0.01$ respectively). Total protein, albumin, TC and HDL were significantly lower in PTB subjects ($p<0.05$, $p<0.001$, $p<0.01$ and $p<0.01$ respectively). TG and LDL levels were also decreased though not statistically significant. However, the level of uric acid and globulin were significantly increased ($p<0.01$, $p<0.001$ respectively).The levels of Total peroxides (µmol H_2O_2/litre) activity in patients and control were 16.17±0.29 and 12.69±0.32, respectively ($p=<0.001$). Similarly, the levels of PON1 activity (µmol/min/ml) in patients and control were 109.72±6.43 and 161.55±15.49, respectively ($p=0.004$).

Conclusions: According to this study, we conclude that patients with active PTB are exposed to potent oxidative stress and they have decreased PON1 activity. These predisposal factors play a role in the pathogenesis of pulmonary tuberculosis in a group of Nepalese population which needs more clarification.

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Introduction:-

Tuberculosis is primarily a disease of the lung and infects one in three worldwide and kills more people each year than any other bacterial pathogen and occurring predominantly in socio-economically deprived populations [**1**].Pulmonary tuberculosis (TB) is a chronic disease caused by *Mycobacterium tuberculosis* and is still a major health problem in the world.

The problem occupies in the ability of M. tuberculosis to persevere in a presumably dormant state and bestows to a latent tuberculosis infection which serves as a tremendous reservoir of infection [**2,3**]. Worldwide, there are 8 million new cases of symptomatic tuberculosis and 3 million deaths from the disease every year. It is believed that one third of all the people in the world have a dormant (latent) tuberculosis infection, although only about 5 to 10% progress to active tuberculosis disease (URL-1). The South Asian Association for Regional Cooperation (SAARC) region accounts more than 32% of global burden with 2.5 million new cases and 0.6 million deaths annually [**4**]. More than 75% of morbidity and mortality due to disease occurred in the most economically productive age groups of 15-49 years [**5**]. TB is one of the Nepal's top health challenges, where 15 million people about 45% of the total population is infected with TB, out of which 60% are in productive age groups and an estimated 20,000 new infectious cases of TB are reported each year [**6**].

Recent research suggests that in pulmonary Tuberclosis can induce reactive oxygen species (ROS), indicating ongoing oxidative stress and decrease in the antioxidant activity which may contribute to development of lung function abnormalities [**7**]. Under normal conditions, oxygen and nitrogen derived radicals are generated continuously in small amounts and are kept in a state of balance by antioxidants, also known as oxidation-reduction balance [**8**]. When free radical overcomes antioxidant system in the body is called oxidative stress and is one of the key pathogenetic mechanisms in the development of the disease [**9**]. Thus, the oxidant and antioxidant valance system needs to treatment for its homeostasis. Recent research suggests that oxygen and its relative species (oxidants) may contribute to the pathogenesis of a number of important lung diseases.

Though ROS and reactive nitrogen intermediates (RNI) are important parts of the host defense against mycobacterium, their increased amount may in turn contribute towards pulmonary inflammation [**10**]. The inflammatory response is an important endogenous cause of chronic oxidative stress [**11**]. Therefore, excess amount of these free radicals cause protein oxidation, DNA damage and lipid peroxidation [**12**]. Lipid peroxidation is a chain process that affects polyunsaturated fatty acids mainly localized in cell membranes, in which such products as conjugated dienes (CD) and malondialdehyde (MDA) are generated. These products diffuse from the site of inflammation and can be measured in the blood, and the products of lipid peroxidation increase in active pulmonary TB [**13**]. The increase in circulating free radical activity by decreasing antioxidant activity will lead to oxidative stress and contribute to the pathogenesis of lung disease [**14**]. Oxidant-antioxidant balance is essential for the normal lung function. In tuberculosis patients, there are also some reports of poor antioxidants defence that may expose to oxidative host tissue damage [**15,16**]

Paraoxonase1 (PON1) is a calcium dependent enzyme that contributes to the hydrolysis of lipid peroxides and homocysteine-thiolactone and may play a role in the organism's antioxidant system. It is a protein of 354 amino acids with a molecular mass of 43 kDa and widely distributed among tissues such as liver, kidney, intestine, and also in serum [**17**]. There is a wide range of serum concentrations and activities of PON1 in humans. These have been traced, in part, to a number of polymorphisms within promoter and coding regions of PON1 gene affecting both enzyme activity and peptide concentration.Polymorphisms in the noncoding region of the PON1 gene can, on the other hand, have a major impact on gene expression and, consequently, serum levels of the enzyme. PON1 is also sensitive to inflammatory conditions. Thus, it can consider a negative acute phase protein whose serum level and hepatic synthesis are reduced during infection [**18**].

Oxidative stress downwards serum PON1 expression due to the changes in the "oxidation-reduction" status [1**9**]. There is no existing report on PON1 activity in pulmonary TB in Nepalese population.The objective of the present study was to investigate the levels of Total peroxides, Paraoxonase1 (PON1) and other biochemical parameters status in Pulmonary Tuberculosis (PTB) patients and normal healthy subjects.

Materials and Methods:-

This study analyzed 70 individuals participant where 40 were diagnosed with active pulmonary tuberculosis selected randomly from the Regional Tuberculosis Center, Pokhara, Nepal and 30 healthy subjects enrolled in this study during the period of 3rd February 2017 to 26th November 2017. An informed written consent was obtained from all the study subjects who were enrolled in the study. Each participant were given a validated questionnaire form to obtain their basic information which included name, age, sex, history of hypertension, diabetes mellitus, anti-tuberculosis therapy, HIV positive. The age range of participants was 20-80 years. Individuals with at least one sputum smear positive were selected for the study. Anthropometric measurement, taken by standard instruments and techniques, included blood pressure (SBP and DBP) measurement by aneroid and mercury sphygmomanometer, pulse, weight, height of participants wearing light clothing and without shoes. BMI (in kg/m^2) was used to measure general or weight loss due to tuberculosis.The estimation of biochemical parameters such as glucose, uric acid, total protein, albumin and lipid profile were done by colorimetric method. Total peroxide concentrations of the plasma samples were determined using the FOX2 method with minor modificationswas measured by spectrophotometer. In addition, Phenyl acetate assay was used to measure Paraoxonase1 (PON1) activity where one unit of arylesterase activity equal to 1 mmol of phenyl acetate hydrolyzed per minute was used to measure enzyme activity.

Inclusion criteria:-

Those individuals of age between 20-80 years, with normal lipid profile, controlled hypertension, absence of tuberculosis and any chronic disease/s were included in the study for control. For cases, the individuals with confirmed pulmonary tuberculosis on the basis of clinical symptoms, radiologic findings, and bacteriologic data (acid fast microscopy) with normal and abnormal lipid profile were included in the study.

Exclusion criteria:-

Those individuals under anti-tuberculosis therapy, HIV positive, previous myocardial infarction, coronary heart disease or any other cardiovascular disorder, diabetes mellitus, malignant disease were excluded from our study.

Statistical analysis:-

All statistical analyses were performed using Statistical Package for Social Science (SPSS) version 12.0 (SPSS Inc., Chicago, IL, USA) and Microsoft excel 2003. Data were presented as mean ± Standard error mean (SEM).Correlations between measured parameters were assessed using the analytical method of Pearson Coefficient. Comparison of parameters between healthy controls and pulmonary tuberculosis subjects was performed by Mann-Whitney test. Two-tailed probability values were calculated throughout, and $p<0.05$ was considered statistically significant.

Results:-

A total of 70 subjects from Pokhara Valley and its surroundings were enrolled in our study. Out of 70 subjects, 30 (16 female and 14 male) were healthy controls and 40 (10 female and 30 male) newly diagnosed active pulmonary tuberculosis subjects.

Table 1:- Gender Distribution in Control and Pulmonary tuberculosis (PTB) Subjects

Gender	Control	PTB Subjects	Total
Male	14	30	44
Female	16	10	26
Total	30	40	70

Demographic Characteristics:-

The demographic characteristics of the healthy controls and pulmonary tuberculosis subjects are presented in the **Table 2**. The mean age of study group was 37.31±1.72 years. The study group population had a greater proportion of male than female in pulmonary tuberculosis subjects in contrast to the greater proportion of female than male in healthy controls. The percentage of male and female subject was 62.85% and 37.15% respectively as shown in Figure .There were 30 males in pulmonary tuberculosis and 14 in control subjects out of 44 males in whole study group. There were 10 females in pulmonary tuberculosis and 16 in control subjects out of 26 females in whole study group. The mean and SEM of age (yrs) were 39.95±2.77, 33.80±1.40 for pulmonary tuberculosis and control subjects, respectively. Similarly, mean BMI (kg/m2), SBP (mmHg) and DBP (mmHg) were 18.61±0.37, 107±2.05

and 68.62±1.22; 22.93±0.35, 114.83±1.87 and 75.83±1.66 for pulmonary tuberculosis and control subjects, respectively (as shown in Table 3).

Table 2:- Demographic Details of Study Group

Parameters	**Control(n**=30)	**Pulmonary Tuberculosis(n** =40)	***P* -value**
Age (yrs)	33.80±1.40	39.95±2.77^{NS}	0.078
BMI (kg/m^2)	22.93±0.35	18.61±0.37***	<0.001
SBP (mmHg)	114.83±1.87	107±2.05**	0.009
DBP (mmHg)	75.83±1.66	68.62±1.22**	0.001

Values quoted as mean ±SEM ** p<0.01; ***p<0.001; NS: non significant when compare with control subjects

Biochemical Characteristics:-

The biochemical characteristics of control and pulmonary tuberculosis subjects are shown in Table. The mean (in mg/dl) and SEM of RBS and Urea are 79.15±2.58, 21.93±0.94 and 87.81±4.16, 21.37±0.73 for control and pulmonary tuberculosis subjects, respectively. The mean (in g/dl) and SEM of total protein, albumin and globulin are 7.00±0.16, 4.57±0.06, 2.42±0.17 and 7.77±0.19, 3.73±0.09, 3.73±0.22 for control and pulmonary tuberculosis subjects, respectively. The mean and SEM of A/G ratio for control and pulmonary tuberculosis are 2.27±0.21 and 1.15±0.08 respectively. Similarly, the mean (in mg/dl) and SEM of TC, TG, HDL and LDL are 155.80±5.71, 150.10±12.87, 42.94±1.93, 85.39±4.53 and 132.51±5.34, 134.78±5.05, 34.40±2.02, 72.28±5.04 for control and pulmonary tuberculosis subjects, respectively. The mean and SEM of uric acid (mg/dl) for control and pulmonary tuberculosis subjects are 5.08±0.30 and 6.19±0.23, respectively.

Table 3:- Biochemical Characteristics in Control and Pulmonary Tuberculosis Subjects

Parameters	**Control(n**=30)	**Pulmonary Tuberculosis (n** =40)	***P* -value**
RBS(mg/dl)	79.15±2.58	87.81±4.16^{NS}	0.107
Urea(mg/dl)	21.93±0.94	21.37±0.73^{NS}	0.636
TP(g/dl)	7.00±0.16	6.47±0.16*	0.029
Albumin(g/dl)	4.57±0.06	3.73±0.09***	0.000
Globulin(g/dl)	2.42±0.17	2.74±0.22***	0.000
TC(mg/dl)	155.80±5.71	132.51±5.34**	0.004
TG(mg/dl)	150.10±12.87	134.78±5.05^{NS}	0.275
HDL(mg/dl)	42.94±1.93	34.40±2.02**	0.003
LDL(mg/dl)	85.39±4.53	72.28±5.04^{NS}	0.057
Uric Acid(mg/dl)	5.08±0.30	6.19±0.23**	0.005

Values quoted as mean ±SEM * p<0.05; ** p<0.01; *** p <0.001; NS: non significant when compare with control subjects.

When all the biochemical parameters were grouped into control and pulmonary tuberculosis subjects, there were significant difference in TP, Albumin, Globulin, TC, HDL and Uric Acid value between these groups (TP, p <0.05; Albumin, p<0.05; Globulin, p<0.05; TC, p<0.01, HDL, p<0.05; and Uric acid, p<0.01). There was no significant difference in sugar, urea, TG and LDL value.

When serum paraoxonase 1(arylesterase)activity and total peroxide level were grouped into control and pulmonary tuberculosis subjects, PON1 arylesterase activity was significantly lower in Pulmonary Tuberculosis subjects than Controls(p<0.01), while Total peroxide level was significantly higher(p<0.001).

Table 4:-Total Peroxide level and PON1 arylesterase activity in Control and Pulmonary Tuberculosis Subjects

Parameter	**Control** (**n**=30)	**Pulmonary Tuberculosis** (**n** =40)	***p* value**
Total peroxide (µmol H_2O_2/litre)	12.69±0.32	16.17±0.29***	<0.001
PON1(ARE) (µmol/min/ml)	161.55±15.49	109.72±6.43**	0.004

Values quoted as mean ±SEM ** p<0.01; *** p <0.001

Parameter	TC	TG	HDL	LDL	Total peroxide	PON1
BMI	r=0.360** p=0.002	r=0.221 p=0.066	r=0.237* p=0.049	r=0.276* p=0.021	r=-0.391** p=0.001	r= 0.267* p= 0.025

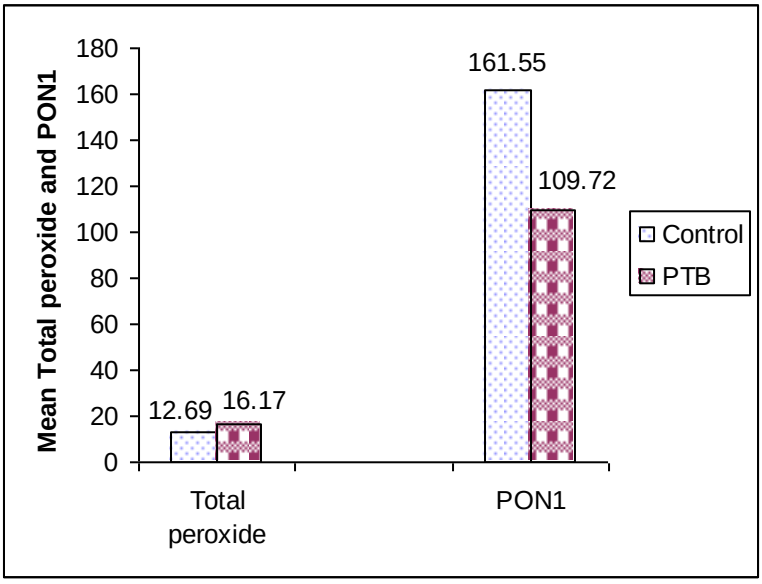

Figure 1:-Total peroxide and PON1 activity in Control and Pulmonary Tuberculosis Subjects

Pearson correlation analysis in the Control and Pulmonary Tuberculosis Subjects showed positive and statistically significant correlation of BMI with TC, HDL, LDL and PON1, positive though not statistically significant with TG but negative and statistically significant correlation with total peroxide. Similarly, correlation of Total peroxide with TC, TG, HDL and LDL was negative but statistically significant only with TC and TG where as correlation of PON1 with TC, TG and HDL was positive but negative with LDL even though it was not statistically significant. Moreover, correlation of PON1 with total peroxide was negative and statistically significant.

Table 5:-Correlation of BMI with Lipid Profile, Total peroxide and PON1 in Pulmonary Tuberculosis subjects.

Table 6:-Correlation of Total peroxide and PON1 with Lipid Profile in Pulmonary Tuberculosis subjects.

Parameter	TC	TG	HDL	LDL
Total peroxide	r= -0.290* p= 0.015	r= -0.248* p= 0.036	r= -0.204 p= 0.091	r= -0.184 p= 0.127
PON1	r= 0.005 p= 0.961	r= 0.186 p= 0.122	r= 0.008 p= 0.951	r= -0.071 p= 0.557

* p<0.05; ** p<0.01; *** p <0.001

Table 7:-Correlation of PON1 with Total peroxide in Pulmonary Tuberculosis subjects.

Parameter	Total peroxide
PON1	r= -0.390** p= 0.001

* p<0.05; ** p<0.01; *** p <0.001

Discussion:-

Understanding the biology of Mycobacterium tuberculosis and its pathogenesis is one of the primary challenges in current tuberculosis research.Our study was done to find the oxidative stress and other biochemical changes among the subjects suffering from pulmonary tuberculosis (PTB), apart from this the PON1 level in these subjects was also

estimated. The comparison was done with normal age sex matched controls. The experimental group consisted of 40 and the control group comprised of 30 subjects. Majority of the subjects were middle aged adults and is the first epidemiological study in western development region and probably the first in Nepal. The present study reveals that higher total peroxide products and significantly lowers antioxidants potential in TB patients. Additionally, this study supports a role for oxidative stress in the pathogenesis of TB and indicates lower antioxidant capacity and higher ROS levels in the TB patients than in control subjects. The finding in this study is also consistent with previous reports [**20].**

The present study shows significantly lower levels of total protein and albumin in subjects with pulmonary tuberculosis. This agrees with some previous studies reported by Sasaki et al. [**21**], and Akiibinu et al. **[22**] reported that the total protein, albumin, were significantly lower in pulmonary tuberculosis patients when compared with healthy control group. Low levels of total protein and albumin in this study might have been caused by anorexia, malnutrition and mal-absorption commonly observed in tuberculosis. However, the level of globulin observed in pulmonary tuberculosis subjects was significantly high which might have arisen from combination of elevation of different globulin fractions. In addition, BMI were significantly lower in pulmonary tuberculosis patients than in controls. BMI is an estimator of body fat and the total mass of all the cellular elements in the body represents the metabolically active component of the body. However, lower BMI in PTB patients was still a sensitive indicator of malnutrition in this study. In a study from Nigeria, patients with PTB and more severe lung disease had similar finding reported [**23**].

Similarly, we have found that the level of uric acid in PTB subjects was noticed significantly higher than the healthy controls. This data is in agreement with the study done by Akiibinu et al. 2007 [**22**]. This high level of uric acid could be due to continuous tissue break down or a compensatory mechanism of reducing the free radical load in the tuberculosis patients. In addition, lower levels of TC, TG, LDL and HDL were observed in PTB patients. We have noticed that total cholesterol and HDL were significantly different between the test and control group. However, we observed no significant variation in serum triglyceride and LDL level among PTB patients and healthy subjects. The low level of TC, TG, HDL and LDL could be the result of impaired rate of lipid production and enhanced lipid catabolic rate associated with tuberculosis. This is also consistent with previous reports from Mexico conducted to evaluate the hypothesis that low serum levels of cholesterol might be a risk factor in development of pulmonary tuberculosis, levels of TC, HDL, LDL and TG were found to be lower in tuberculosis patients [**24**]. Furthermore, similar to other studies, in these studies also levels of HDL and LDL were found to decrease due to inflammation in tuberculosis patients [**25**].

In this study we noticed that serum PON1 activity was decreased in pulmonary tuberculosis (PTB) patients relative to control subjects due to the high levels of oxidative stress in TB patients. In tuberculosis, oxidative stress is a result of tissue inflammation, poor dietary intake of micronutrients due to illness, free radical burst from activated macrophages and anti-tuberculosis drugs. In our study, we have demonstrated that total peroxide is elevated in patients with PTB which provides evidence for enhanced free radical mediated processes.In addition, we found the positive correlation of BMI with lipid profile. Kwiatkowska et al. and Reddy et al. [**26,10**] reported high level of lipid peroxidation in all categories of pulmonary tuberculosis patients, irrespective of treatment status and this might have caused reduction in the concentration of serum lipids as observed in our study. This proves that though the BMI and lipid profile is decreased because of malnutrition and mal-absorption, free radical load is increased as a result of respiratory burst leading to elevated oxidative stress in PTB patients.

On the other hand, we noticed that PON1 activity was negatively correlated with total oxidants levels in the PTB patients. The high oxidative stress markers and low antioxidant levels could be related to low PON1 activity which led to increased susceptibility to lipid peroxidation and is associated with cardiovascular diseases as reported by Serdar et al. **[27]** and affects PON1 expression and activities reported by Aviram et al. **[19]**. Decrease in PON1 activity under oxidative stress was mostly attributed to changes in the redox status of the protein's free sulfhydryl groups since sulfhydryl compounds prevented the inhibition of PON1 activity caused by ROS [**28**]. Previous studies have indeed showed that PON1 is inactivated on the oxidative conditions [**29**] and its activities is reduced in pathologies associated with atherosclerosis, e.g. CVD, diabetes and chronic renal failure [**30**]. In the present study, we showed that PON1 arylesterase activity was significantly lower in patients with PTB than control while total peroxide level was significantly higher.

Furthermore, we found the positive correlation of PON1 with BMI, TC, TG and HDL but negative with LDL and total peroxide. H_2O_2 is a major ROS produced by arterial wall cells during atherogenesis, and it is converted under oxidative stress into a more potent ROS leading to LDL oxidation. The ability of HDL-associated PON1 to hydrolyze H2O2 (in addition to peroxides) may play an important role in eliminating potent oxidants that are involved in atherosclerosis [**31**]. Thus we can suggest that increased total peroxides and decreased PON1 activity may, in part prone patients with PTB to the development of atherosclerosis.

Conclusion:-

Pulmonary tuberculosis is a major cause of death around the world despite the fact that the causative organism discovered more than hundred years ago, and highly effective drugs available making tuberculosis a preventable and curable disease. However, our knowledge of the oxidant and antioxidant profile in tuberculosis patients is very limited. Although, substantial effort has been directed towards enlightening biochemical events governing the unveiling and progression of PTB, much remains indecipherable. As such, a better understanding of the mechanisms leading to PTB and their clinical outcomes is urgently needed and one of the most serious challenges in medicine. Finally, this study revealed that total peroxide is significantly increased in PTB patients, which is a marker of oxidative stress whereas serum PON1 activity is significantly decreased in PTB patients when compared with control. However, these results should be considered preliminary because the number of participants enrolled was relatively low and need further study to clarify the possible mechanism.

References:-

1. WHO Tuberculosis Factsheet; March 2010
2. Nuermberger, E, Bishai, WR, Grosset, JH. Latent tuberculosis infection. Semin. Respir. Crit. Care Med. 2004; 25: 317-36.
3. Gomez, JE, McKinney, JD. M. tuberculosis persistence, latency, and drug tolerance. Tuberculosis (Edinb). 2004; 84:29-44.
4. WHO (2004) Health Development in the South -East Asia Region: An Overview. World Health Organization, Geneva.
5. Park K. Epidemiology of Communicable Disease. In: Park's Textbook of Preventive and Social Medicine, Bhanol Publishers, India pp. 2002;137-150.
6. Sreeramareddy CT, Panduru KV, Verma SC, Joshi HS, Bates MN. Comparison of pulmonary and extrapulmonary tuberculosis in Nepal- a hospital-based retrospective study. BMC Infect Dis 2008; 8:8
7. Ragunath RR, and Madhavi SP. Plasma oxidant antioxidant status in different respiratory disorder. Indian J. of Clinical Biochemistry.2006;21(2):161-4.
8. Droge W. Free radicals in the physiological control of cell function. Physiol Rev 2002;82:47-95.
9. Repine J. E, Bast A, Lankhorst I. Oxidative stress in chronic obstructive pulmonary disease. Oxidative Stress Study Group. Am J Respir Crit Care Med. 1997;156:341-57.
10. Reddy YN, Murthy SV, Krishna DR and Prabhakar MC. Role of free radicals and antioxidants in tuberculosis patients. Indian Journal of Tuberculosis. 2004; 51: 213-8.
11. Cerutti PA and Trump BF. Inflammation and oxidative stress in carcinogenesis. Cancer Cells. 1991; 3:1-7.
12. Kocyigit A, Keles H, Selek S, Guzel S, Celik H and Erel O. Increased DNA damage and oxidative stress in patients with cutaneous leishmaniasis. Mutation Research. 2005; 585:71-8.
13. Devici F and Ilhan N. Plasma malondialdehyde and serum trace element concentrations in patients with active pulmonary tuberculosis. Biological Trace Element Research. 2003; 95: 29-38.
14. Rai, RR and Phadke, MS. Plasma Oxidant Antioxidant Status in Different Respiratory Disorder. Indian Journal of Clinical Biochemistry. 2006; 21:161-4.
15. Jack, CIA, Jackson, M.J. and Hind, CRK. Circulating Markers of Free Radical Activity in Patients with Pulmonary Tuberculosis. Tubercle and Lung Disease. 1994; 75: 132-7.
16. Plit, ML, Theron, A.J., Fickl, H., Van Rensburg, C.E., Pendel, S. and Anderson, R. Influence of Antimicrobial Chemotherapy and Smoking Status on the Plasma Concentrations of Vitamin C, Vitamin E, Beta Carotene, Acute Phase Reactants, Iron and Lipid Peroxides in Patients with Pulmonary Tuberculosis. The International Journal of Tuberculosis and Lung Disease. 1998; 2: 590-6.
17. Li HL, Liu DP and Liang CC. Paraoxonase gene polymorphisms, oxidative stress and diseases. Journal of Molecular Medicine. 2003;81:766-79.
18. James RW and Deakin SP. The importance of high density lipoproteins for paraoxonase 1 secretion, stability and activity. Free Radical Biology and Medicine. 2004; 37(12):1986-94.

19. Aviram M, Rosenblat M: Paraoxonases 1, 2 and 3, oxidative stress, and macrophage foam cell formation during atherosclerosis development. Free Radic Biol Med 2004;37:1304–1316
20. Ramesh, Sudha K , Amareshwara M, Sameer R.M. Study of protein oxidation and antioxidants status in pulmonary tuberculosis patients. Intl J of Pharma and Bio Sciences. 2011;2:3,104-109
21. Sasaki Y. Yamagishi F, Yasi T, Mizutani F. A case of pulmonary tuberculosis case with pancytopenia accompanied to bone marrow gelatinous transformation. Kekaku 1999;74 (4):361-4
22. Akiibinu MO, Arinola OG, Ogunlewe JO and Onih EA. Non-enzymatic antioxidants and nutritional profiles in newly diagnosed pulmonary tuberculosis patients in Nigeria. African Journal of Biomedical Research. 2007; 10: 223-228.
23. Harries AD, Thomas J, Chugh KS. Malnutrition in Africa patients with pulmonary tuberculosis. Hum Nutr Clin Nutr 1985; 39: 361-363.
24. Pérez-Guzmán C, Vargas MH, Salas-Mártir C, Trejo-Santacruz T, Gallegos-Discua C, Flores-López F. Lipidprofile in household contacts of patients with pulmonary tuberculosis. Rev Med Inst Mex Seguro Soc. 2008; 46:247-52.
25. Volpato S, Palmieri E, Fellin R, Zuliani G. Acute phase markers are associated with reduced plasma lipid concentrations in a population of hospitalized elderly patients. Gerontology. 2000; 46: 22-7.
26. Kwiatkowska S, Piasecka G, Zieba M and Piotrowski D. Increased serum concentrations of conjugated diens and malondialdehyde in patients with pulmonary tuberculosis. Respiratory Medicine. 1999;93(4):272-6.
27. Serdar Z, Aslan K, Dirican M, Sarandol E, Yesilbursa D and Serdar A. Lipid and protein oxidation and antioxidant status in patients with angiographically proven coronary artery disease.Clinical Biochemistry. 2006;39:794-803.
28. Rozenberg O, Aviram M. S-Glutathionylation regulates HDL-associated paraoxonase 1 (PON1) activity. Biochem Biophys Res Commun 2006;351:492–8.
29. Rosenblat M, Hayek T, Aviram M. Anti-oxidative effects of pomegranate juice (PJ) consumption by diabetic patients on serum and on macrophages. Atherosclerosis. 2006; 187:363-71.
30. Seres I, Paragh G, Deschene E, Fulop Jr T and Khalol A. Study of factors influencing the decreased HDL associated PON1 activity with aging. Experimental Gerontology. 2004; 39:59-66.
31. Aviram M, Rosenblat, M Bisgaier CL, Newton RS, Primo-Parm SL and La Du BN. Paraoxonase inhibits high-density lipoprotein oxidation and preserves its functions: A possible peroxidative role for paraoxonase. Journal of Clinical Investigation. 1998;101: 1581-90.