Comparison of Transudates and Exudates Using Malondialdehyde and Lipid Profile

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Received: 22/03//2014 Revised: 10/04/2014 Accepted: 19/04/2014

ABSTRACT

Aim: The aim of present study was to evaluate the levels of oxidative stress and local inflammatory status in the pleural fluid and blood of patients with pleural effusion of various etiologies. We also examined whether the absolute values of all parameters or the relative ratios between pleural fluids to serum provide the better diagnostic accuracy.

Materials and methods: All the parameters are analyzed on Siemens Dimension Expand plus fully automated random access analyzer. MDA is done using TBARS method.

Results and discussion: MDA values in pleural fluid showed significant increased levels in exudates compared to corresponding values in transudates. However when values in blood of exudates show no increase compared to control group indicate no systemic involvement only local peroxidation in pleura takes place. Mean cholesterol levels show significant increase in exudates in pleural fluid compared to transudates. But when compared to controls, serum levels of cholesterol in exudates do not show significant increase. We have compared even the pleural fluid/ serum ratios of transudates and exudates for MDA and lipid profile.

Conclusion: The present study showed that oxidative stress was more in exudates compared to transudates, probably due to production of reactive oxygen species and it may serve as a marker for differentiation between transudates and exudates

KEY WORDS:- Pleural fluid, serum, transudates, exudates, MDA, lipid profile

INTRODUCTION

The imbalance between oxidant and antioxidant, referred as oxidative stress has been associated with various respiratory disorders. (1) Determining the cause of pleural effusion requires use of different diagnostic techniques, some of which are invasive and not completely free of morbidity. (2-5) The use of criteria established by Light et al for segregating transudates and exudates led to correct classification of pleural effusion tested in 99 % of cases. However recent reports have shown that the low specificity may lead to unwanted invasive interventions in upto 20 – 30 % of patients with transudates. (6-8) If an exudative effusion is present, further diagnostic procedures are imperative such as
cytopathology, pleural biopsy and sometimes even thoracotomy, so that a definitive diagnosis can be made and specific therapy may be instituted. On the other hand, if the fluid is clearly a transudate, one need not worry about therapeutic maneuvers directed at pleura and need treat only the congestive heart failure, nephrosis, cirrhosis or hypoproteinememia.

MDA is a lipid peroxidation product, produced by oxidative deterioration of unsaturated fatty acids. Two sources could be suggested for MDA in pleural fluid. (9) The first possible source is plasma proteins. When pleura are inflamed there is increased leakage of plasma proteins into pleural space. Plasma MDA is mostly bound to plasma proteins. Second source is local production by inflammatory cells. Lipid peroxides are important products of the eicosanoid synthetic pathways responsible for inflammatory mediator production. (10) The lipid peroxidation starts with the removal of hydrogen atom from the chain of a polyunsaturated fatty acid in the membrane structure due to the effect of a free radical formed in vivo. Consequently the structure of cell membrane changes and the protective part disappears. Therefore it is suggested that free oxygen radicals and lipid peroxidation have role in the formation of the exudative pleural effusion and lipid peroxidation products may be used in distinction between transudates and exudates. (11, 12)

The cause of the rise in cholesterol levels in pleural exudates is unknown and two possible explanations have been put forward. According to the first, the cholesterol is synthesized by pleural cells themselves for their own needs (13) (extra hepatic synthesis of cholesterol is now known to be much greater than was once thought, depends on the metabolic needs of cells and is in dynamic equilibrium with cholesterol supply by LDL and cholesterol removed by HDL) (14) and the concentration of cholesterol in the pleural cavity is increased by degeneration of leukocytes and erythrocytes, which contain large quantities. The second possible explanation is that pleural cholesterol is derived from plasma; some 70% of plasma cholesterol is bound to LDL and the rest to HDL or VLDL and the increased permeability of pleural capillaries in pleural exudates patients would allow plasma cholesterol to enter pleural cavity.

MATERIALS AND METHODS

The present study was conducted in the department of biochemistry, Rangaraya Medical College, Kakinada, Andhra Pradesh, India. The present study was undertaken to determine biochemical changes in patients with pleural effusion with different etiologies to compare transudates from exudates in both pleural fluid and venous blood consisting of 100 patients with 50 patients of exudative etiology (TB, Pneumonia, Malignancy with metastasis) and 50 patients of transudative etiology (CHF, Cirrhosis, Nephrotic syndrome). Pleural fluid and blood samples were collected simultaneously. These values are compared with 50 normal healthy persons. All of these samples were taken from TBCD, Government General Hospital, Kakinada. Demographic and clinical data was collected at TBCD ward. The following parameters were analyzed in pleural fluid and serum.

MDA was estimated by TBARS method, Mahalouz et al. Under acidic conditions, lipid peroxides break down to form MDA which complex with TBA. The resulting MDA – TBA chromogen is measured at 530nm against distilled water in spectrophotometer. Cholesterol by CHOD-PAP method HDL and LDL by direct estimation.
RESULTS AND DISCUSSION

The mean oxidative stress expressed by MDA values in pleural fluid (transudates -145 ± 11.02, exudates -264.58 ± 68.52) showed significantly increased levels in exudates compared to corresponding values in transudates (p<0.001). However when values in blood samples of exudates showed no significant elevation compared to control groups (254.06 ± 59.86) indicating no systemic involvement only local peroxidation in pleura takes place.

We have included MDA as one of the criteria because an imbalance between oxidants and antioxidants results in oxidative stress and has been associated with various respiratory disorders. Increased oxidative stress participates in pathogenesis of both airways and parenchymal lung diseases. Lung represents unique tissue in both its exposure to high oxygen tension and its high concentration of antioxidants. (Kinmula & Crapo, 2003). Inflammatory cells generate free radicals in patients with exudative etiology. Mycobacterium can induce reactive oxygen species production by activating phagocytes and although these are an important part of host defense against mycobacterium, increased ROS may promote tissue injury and inflammation (Beers & Sizer, 1979). This may contribute to immunosuppression (Grimble, 1994; Jack et al, 1994; Nathan et al, 1979), particularly in those with impaired antioxidant capacity, such as HIV infected patients (Aukrust and Muller, 1999; Favier et al, 1994; Muller et al, 2000). Moreover the malnutrition which is commonly present in patients with TB can add to impaired antioxidant capacity in these patients. During pulmonary inflammation increased amounts of reactive nitrogen intermediates are produce as a consequence of phagocytic respiratory burst (Kwiatkowska et al, 1999) which causes cell damage and may induce many pathological events.

Local production of oxidants in pleural cavity has not been exclusively studied. There is in vitro evidence in animal models that receive oxygen and nitrogen species may be implicated in the pathogenesis (Choe et al, 1998). In the diseases studied in our patients, there is little evidence in the literature regarding the local production of oxidative stress, oxidants have been shown to play an important role in carcinogenesis; serving not only as tumor initiators but also as tumor promoters and regulators of gene expression (Upaham and Wagner , 2001). TB has been associated with increased levels of several markers of oxidative stress and decreased antioxidant capacity (Mabeto et al, 2003). Mesothelial cells are responsible for the release of oxidants in pleural space infections (Antony and Mohammed, 1999).

Mean cholesterol levels show significant increase in exudates in pleural fluid (transudates – 28.6 ± 8.53, exudates – 95.54 ± 17.93) compared to transudates (p<0.001). But when compared to controls (176.04 ± 14.38), serum levels of cholesterol in exudates do not show significant increase. Mean triglyceride levels (pleural fluid transudates - 35.26 ± 7.30, exudates - 50.64 ± 9.59) and LDL-Cholesterol (pleural fluid transudates -55.26 ± 6.63, exudates – 67.04 ± 7.01) levels in pleural fluid show significant increase in exudates compared to transudates. But serum level of triglycerides in exudates do not show significant increase when compared to control group (105.60 ± 36.89) whereas HDL shows decreased levels in pleural fluid (transudates-28.32 ± 4.74, exudates- 20.58 ± 3.26) of exudates compared to transudates.

Pleural fluid/ serum ratio of MDA (transudates- 0.52 ± 0.06, exudates- 0.82 ± 0.17, p<0.001) and that of cholesterol (transudates- 0.18 ± 0.06, exudates- 0.55 ±
0.12, p<0.0001) of transudates are significantly low compared to exudates thus giving better distinction of transudates from exudates.

CONCLUSION

By this study, we came to the conclusion that the oxidative stress was higher in exudative pleural effusions compared to transudative effusions, probably due to the production of more reactive oxygen species in the pleura locally in exudates group. The systemic involvement is ruled out by comparing it in serum samples. Pleural fluid/ serum cholesterol ratio is giving better discrimination between the two groups as compared to corresponding ratio of MDA indicating increased capillary permeability to cholesterol involved in lipid peroxidation.

ACKNOWLEDGEMENTS

I acknowledge my thanks to Dr.S.Sanjeevi Rao, Professor and Head of department of Biochemistry, Rangaraya Medical College, Kakinada and my family members for their guidance and support.

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International Journal of Health Sciences & Research (www.ijhsr.org) 69
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How to cite this article: Usha KP, Komala P. Comparison of transudates and exudates using malondialdehyde and lipid profile. Int J Health Sci Res. 2014;4(5):66-70.

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