



“ASSESSMENT OF THE OXIDATIVE STRESS IN MODERATE & SEVERE UNDERNOURISHED CHILDREN (1-5 YEARS) BY ESTIMATING SERUM MDA LEVEL” IN ROHILKHAND MEDICAL COLLEGE AND HOSPITAL

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ABSTRACT

INTRODUCTION: Protein energy malnutrition (PEM) is most important public health problems in India¹. In malnutrition, there is excess production of free radicals within the erythrocytes which lead to oxidative stress⁽²⁻⁴⁾ and contribute to pathophysiology in malnutrition.^{5,6}

AIM and OBJECTIVES: the aim of study is to assess the oxidative stress in moderate and severe undernourished children (1–5 years).

MATERIAL AND METHODS: Moderate and severe undernourished children (1–5 years) according to WHO Classification included in the study. In oxidative stress there is lipid peroxidation, which is assessed by measuring intermediate like Malondialdehyde (MDA) by Thiobarbituric acid (TBA) method.

RESULT: An institutional based case control study was conducted in the Biochemistry department among undernourished children attending the outpatient department of Pediatric RMCH Bareilly U.P. A total of 202 children of age 1–5 yrs., out of this 103 were undernourished and 99 were healthy as control. The serum MDA level of control was 0.71 ± 0.18 mmol/mL and in moderate and severe undernourished children were 1.84 ± 0.38 and 3.44 ± 0.55 mmol/mL respectively. Compare to control, undernourished Children have high serum MDA level which is statistically significant (p value < 0.001).

CONCLUSION: In this study there was enhanced oxidative stress in the form of elevated serum MDA in moderate and severe undernourished children.

KEYWORDS : undernourished children, oxidative stress, MDA level etc.

INTRODUCTION

Protein energy malnutrition (PEM) is one of the most important public health problems in many developing countries including India, South East Asia and Africa. It is a wide-spread deficiency disease among children of low socio-economic groups. According to UNICEF in India, around 46 percent of all children below the age of three are malnourished and underweight. 1 in 3 of the world's malnourished children lives in India⁽¹⁾. In malnutrition, there is excess production of reactive oxygen intermediates such as superoxide anion, hydroxyl radical, singlet oxygen and hydrogen peroxide within the erythrocytes. All these events lead to oxidative stress⁽²⁻⁴⁾. Malondialdehyde (MDA) a product of lipid peroxidation, is generated in excess amounts⁽⁵⁻⁶⁾. This oxidative stress and a possible consequential accelerated apoptosis may contribute to pathophysiology in malnutrition.

MATERIAL AND METHOD

The proposed study was conducted in Biochemistry and Pediatric department, Rohilkhand Medical College and Hospital (RMCH), Bareilly, Uttar Pradesh, India, after getting approval from Institutional Ethical Committee. After explaining aim and objectives, informed consent was taken from parents of children for participation in this study.

SOURCE AND DATA:

The source of data for the study is from the patients attending Pediatrics OPD in Rohilkhand medical college and Hospital, Bareilly, Uttar Pradesh, India

STUDY PERIOD:

The study was completed in a period of one year i.e. from January 2014 to December 2014.

Study design: Institutional based Case control study.

Study population: Moderate and Severe undernourished children (age- 1-5 years) according to WHO Classification attending out patients department of Pediatrics, RMCH, Bareilly.

SAMPLING FRAME

Children under 5 years of age coming to pediatric out patient department of Rohilkhand Medical College & Hospital, Bareilly, whose parents/guardians gave voluntary consent for interview and examination of their child.

SAMPLE SIZE: Sample size was calculated according to the aim and objective of the study by using an appropriate statistical formula for determination of sample size for valid inferences a minimum sample

size was required, the procedure as given below.

Sample size is calculated on the basis by formula $4PQ/L^2$, where P is 46%⁽¹⁾, Q=100-P=100-46=52, L= (1-20%) of P which comes to be 117, which are rounded up 200.

INCLUSION CRITERIA:

Moderate and Severe undernourished children (age- 1-5 years) according to WHO classification.

EXCLUSION CRITERIA:

1. Child with sepsis or any acute illness.
2. Any systemic illness/ liver disease, nephrotic syndrome, Thalassemia etc.
3. Children on micronutrient supplementation (Zn, Se, Cu, Mg etc.)
4. Children on Vitamin C or Vitamin E supplementation.
5. Those not giving consent/ child refusal.

In the pediatric OPD of RMCH 200 below five year age children were examine (100 cases and 100 controls). Parents/Guardian of the children were interviewed and physical examinations were carried out on study subjects. Data collected was recorded on the proforma.

ASSESSMENT OF NUTRITIONAL STATUS

The nutritional status of the children was assessed by plotting the curve of weight and height of the children on WHO 2006 Growth Standards growth charts using z - scores. Weight and height for age assessment was done by plotting the study subject's weight and height on different growth charts for boys and girls. Weight for height assessment was done by plotting on different graphs, as per the study subject's age, separately for boys and girls. Nutritional status as per BMI for age criteria was also assessed by plotting the graph of study subject's BMI for their respective age.

Nutritional status of the children was classified as per the WHO classification which is mentioned below:

	Normal	Moderate Malnourishment	Severe malnourishment
Weight for age	± 2 SD	SD score < -2	SD score < -3 (severely underweight)
Height for age	± 2 SD	SD score < -2	SD score < -3(severe stunting)
Weight for height	± 2 SD	SD score < -2	SD score < -3(severe wasting)
BMI for Age	± 2 SD	SD score < -2	SD score < -3 (severe undernutrition)

Specimen collection & Processing

Five ml of venous blood was taken from each subject in a plain vacutainer with proper antiseptic precaution. After allowing 30 minutes for spontaneous blood clotting, the serum was separated from the blood cells by centrifugation at 2000 rpm for 10 minutes at 37 degree Celsius.

The investigations were performed in laboratory of Biochemistry department, RMCH on daily basis.

Biochemical analysis included

Estimation of serum MDA: The concentration of serum MDA is estimated by Kei Satoh Method. It is based on the principle of auto-oxidation of unsaturated fatty acids, involves the formation of semi stable peroxides, which then undergo a series of reactions to form malondialdehyde (MDA). MDA reacts with thiobarbituric acid (TBA) to form pink colored chromogen. The resulting chromogen is extracted with 4.0ml of n-butyl alcohol and the absorbance of which is measured at 530 nm.

RESULT:

An institutional based case control study was conducted in the Biochemistry department among undernourished children attending the out patient department of Pediatric RMCH Bareilly U.P. A total of 202 children of age 1 – 5 yrs, out of this 103 were undernourished and 99 were healthy as control. Table 1 depicts the distribution of cases and control subjects. Total of study subjects were divided into two groups of cases that is undernourished children and one group of control subjects, overall into three groups which are as follows:

- Group 1 – Severe undernourished children (n= 46)
- Group 2 – Moderate undernourished children (n= 57)
- Group 3 – Healthy children (n = 99)

Table 1 Distribution of Malnourished (cases) and control subjects

	Frequency	Percentage (%)
Severe Malnourished children	46	22.8
Moderate Malnourished children	57	28.2
Control	99	49
Total	202	100

Table -2 Depicts the comparison of serum MDA level in malnourished children and healthy children.

Parameter	Control	Moderate Malnourished children	Severe Malnourished children	F	P value
MDA (mmol/mL)	0.71 ± 0.18	1.84 ± 0.38	3.44 ± 0.55	881.09	<0.001

The serum MDA level of control were 0.71 ± 0.18 mmol/mL and moderate and severe undernourished children were 1.84 ± 0.38 and 3.44 ± 0.55 mmol/mL respectively. When compared to control, malnourished Children have high serum MDA level which is highly significant (p value < 0.001)

DISCUSSION:

Malnutrition among children under five years age are major public health problem in India⁽⁷⁾. Under nutrition impairs physical, mental, and behavioral development of millions of children and is a major cause of child death⁽⁸⁾. Oxidative stress is harmful because oxygen free radical attach biological with molecules like lipid, proteins and DNA. The scientist Golden and Ramdth postulated that there was involvement of oxidative stress in the severe oedematous malnourished syndrome⁽⁹⁾. In present study serum MDA level in moderate and severe undernourished children were extremely higher (p value < 0.001) as compared to control.

According to Mehmet Bosnak et al⁽¹⁰⁾ study several mechanism which could contribute to enhanced oxidative stress in severe acute malnutrition. The most important one was subnormal intake of nutrients such as carbohydrates, protein, vitamins which could lead to accumulation of ROS. Depleted concentrations of enzymatic and non enzymatic antioxidants, along with trace elements, have been reported in malnutrition. The second mechanism for increased oxidative stress in malnutrition may be a non-specific chronic activation of the immune system due to chronic inflammation. The elevated activity of MDA in malnourished children may be due to depletion in overall antioxidant enzymes as a compensatory mechanism from effects of free radicals. So malnourished cells, (the lipids and proteins) are more susceptible to auto oxidation⁽¹⁰⁾. The study conducted by Ghone R et al⁽¹¹⁾ showed

harsh deficiency of various nutrients in severe acute malnutrition leads to generation of heavy oxidative stress. These effect minimized with supplementation of antioxidants. Similar result was seen by Jain A et al⁽¹²⁾ which showed MDA concentration in malnourished children was significantly higher (p < 0.001) as compared to the control. A study conducted by B.Sharda et al⁽¹³⁾, show low antioxidant level which could be multifactorial viz. low zinc, selenium, vitamin A and C deficiency, recurrent infections elevated free iron and chronic starvation stage. According to Khare M et al⁽¹⁴⁾ study, stress is created as a result of PEM which is responsible for the overproduction of reactive oxygen species (ROS). The ROS will lead to membrane oxidation and thus an increase in lipid peroxidation by products such as MDA and product of protein oxidation. A study was conducted by Yoshikawa Y et al⁽¹⁵⁾ suggest an increased oxidative damage and lipid peroxidation in malnourished children.

CONCLUSION:

An institutional based case control study was conducted in the department of biochemistry and OPD of Pediatric department in RMCH Bareilly U.P. a tertiary care hospital.

The essence of current study lies in the fact that there was enhanced oxidative stress in the form of elevated serum MDA. Changes in oxidant and antioxidant levels may be responsible for grading in PEM. One way to increase survival and rapid growth rates patient s are inhibiting the effect of oxidative stress and improvement in the nutritional status. Regular antioxidant supplementation to malnourished children improves the nutritional as well as antioxidant status by neutralizing the free radicals formation, followed by protection of RBCs from anemia and boosting of the immune system.

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