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Levels of Serum Total Proteins and Fractions along with Serum Cu, Fe and Zn in Diseased Malnourished Children of 0-4 Years of Age

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The research work was conducted to find out the differences in the serum of Cu, Fe, Zn and serum proteins in children of different degrees of malnutrition as a result of some disease. For this purpose a total of 100 children of pre-schooling age (0-4 years) having 85 malnourished and 15 apparently healthy were selected. Results revealed that diarrhoea, fever and malnutrition were the major diseases/ailments. Malnutrition was more prevalent in children of 12 and < 12 months of age, in males and in children of low socioeconomic status. All types of milk were more frequently offered to males and breast feeding was more frequently offered by mothers of uneducated class and undermatric. Levels of serum copper (Cu) and iron (Fe) were significantly ($P < 0.05$) higher in malnourished children. Serum total proteins and globulins were significantly ($P < 0.05$) lower in malnourished children of first degree in low socioeconomic group.

Key words: Serum, Cu, Zn, total protein, albumin, socioeconomic status, malnutrition, body mass index

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Introduction

In the modern age, malnutrition has still remained a devastating problem in certain parts of the world, although the proportion and absolute number of chronically undernourished people have declined. The term malnutrition was coined to describe over or more importantly under consumption of food nutrients. Undernutrition is the most serious form of malnutrition which is universal among the poor families and nations resulting from consumption of poor diet over a long period of time (Awan, 1997). Protein energy malnutrition has been a common health problem of third world. Nevertheless, it is of much serious concern among children of school going age that are deprived of good and ample nutrition on account of their poor socioeconomic status, ignorance and lack of health promotional facilities (Khan *et al.*, 1990). According to Rehman *et al.* (1993) about 13 million infants and children less than five years of age have been reported to die each year in developing countries and most of these deaths were attributed to undernutrition. El-Bushra and Ash (1993) found a significant relationship among protein energy malnutrition and diarrhoea in boys of two years age.

It is a well known that trace elements play important role in human nutrition which are widely distributed in nature (Williams, 1994). Copper (Cu) and zinc (Zn) are induced in many biochemical processes supporting life. The most important of these processes are cellular respiration, cellular utilization of oxygen, DNA and RNA reduction, maintenance of cell membrane integrity and sequestration of free radicals. Trace elements deficient patients usually present with common symptoms such as malaise, loss of appetite, anaemia, infection, skin lesions and low grade neuropathy (Chan *et al.*, 1998). Zinc deficiency is especially likely to be associated with longer episodes of diarrhoea such as persistent diarrhoea (Chaudhary, 1996). Long term exposure to high Zn intakes (substantially in excess of requirements) has been shown to result in interference with the metabolism of other trace elements, e.g., Cu utilization is especially sensitive to an excess of zinc. This Cu/Zn interaction has been responsible for the inadvertent induction between serum Zn, Cu and Fe, age and sex of the children (Alarcon *et al.*, 1997). Lower levels of trace elements increases the susceptibility to recurrent infection (Onerci *et al.*, 1997). The levels of Zn and Cu were reported to be higher in obese children (Yakinci *et al.*, 1997) while infection leads to decrease in their concentration in blood particularly with resultant malnutrition as in leukemia (Sgarbieri *et al.*, 1999). Serum Cu or ceruloplasmin levels could be used as an indicator of malnutrition as the level decreases with degree of malnutrition (Squali *et al.*, 1997). Therefore, the research work was conducted to determine the serum Fe, Cu, Zn, total proteins and fractions in children of pre-schooling (0-4 years) age suffering from different degrees of diseased-malnutrition with special reference to socioeconomic status, sex, age and area of living.

Materials and Methods

The research work was conducted on children admitted in pediatric wards of Allied and National Hospitals of Faisalabad. Children included in the study were those suffering from some common diseases and had developed signs of malnutrition. Children from Outdoor Patient Department, Allied Hospital were also included.

A total of 100 children of 0-4 years of age were randomly selected. Of these, 85 were suffering from some common diseases including diarrhoea, pneumonia, jaundice, gastroenteritis, while 15 apparently healthy children were also selected. These subjects were grouped according to

socioeconomic status, age and sex. Children were divided into two socioeconomic status, i.e., low (maximum earning of 5,000 per month) or middle class (monthly earning between Rs. 5000 to 15000).

Children were also divided into four age groups, i.e., 12 and < 12, 13-23, 24-35 and 36-48 months. History from the close family members of these children was obtained including age, sex, disease suffering, duration of illness, family income, etc. Anthropometric measurements were also made which included height, body weight and mid-arm circumference. Percent weight of each child was also calculated by dividing the actual weight with the target weight at that age.

Children were classified into three degrees of malnutrition according to their weight by using the Gomez classification (Hamill *et al.*, 1979).

Ist degree malnutrition: If weight is 75-90% of the target or expected weight.

IInd degree malnutrition: If weight is 60-75% of the target or expected weight.

IIIRD degree malnutrition: If weight is below 60% of the target or expected weight.

$$\text{Body mass Index} = \frac{\text{Weight}}{(\text{Height})^2}$$

Blood samples of about 3ml were collected from these children with the help of disposable syringes. Blood samples were allowed to clot, then were transferred into test tubes and centrifuged to obtain the serum. The serum thus collected was stored at -20°C for further studies. Serum total protein was determined by Biuret method as described by Oser (1976). Serum albumin was determined following the method of Gowenlock *et al.* (1988). The globulin was estimated by subtracting albumin from total protein and results were expressed in gm globulin 100 ml⁻¹ of samples. Serum Zn, Cu and Fe were determined by atomic absorption spectrophotometer (Z-8200 polarized Zeeman) at 329, 324.8 and 24.3nm wavelength, respectively (Gowenlock *et al.*, 1988).

Data obtained were analyzed by an analysis of variance technique (GLM procedure) and means were compared by LSD and DMR test by using SAS 6.1.2 statistical programme.

Results and Discussion

Data showed that malnutrition was more prevalent in children of 12 and < 12 months of age including 1st, 2nd and 3rd degree of malnutrition, while the prevalence decreased with the increase in age. This suggested that very young children become sick more frequently and weight loss was also rapid than in elderly patients as has been reported by Khan *et al.* (1990). It might be because the requirement of the protein is greater in very young children (Khan *et al.*, 1990). Results of this study suggested that malnutrition occurs more frequently in the male than female which might be due to metabolic and hormonal differences between male and female. The other reason could be that there is more likeliness for the male children, were hospitalized earlier and given more care, while female children mostly receive treatment at home, or much longer period and are hospitalized rarely and at later stages of illness. However, higher prevalence of all degrees of malnutrition in children of low socioeconomic status was in concordance with the results reported by Underwood *et al.* (1967).

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Table 1: Means \pm SD of mid-arm circumference, percent weight and body mass index of children of different ages in different degrees of malnutrition

Age Groups	Degree of Malnutrition			
	1st	2nd	3rd	Control
Mid-arm Circumference				
12 and < 12 months	13.00 \pm ABa	10.25 \pm Bb	10.50 \pm b	13.16 \pm Ba
13-23 months	12.33 \pm Bab	12.83 \pm Aab	11.20 \pm b	14.25 \pm Aa
24-35 months	13.50 \pm ABab	12.00 \pm Aabc	10.61 \pm c	14.67 \pm Aa
36-48 months	14.50 \pm Aa	12.70 \pm Ab	10.94 \pm c	14.50 \pm Aa
Overall mean	13.14 \pm b	11.69 \pm c	10.49 \pm d	14.14 \pm a
Percent Weight				
12 & < 12 months	77.71 \pm b	60.00 \pm Bc	52.60 \pm d	101.68 \pm a
13-23 months	77.00 \pm b	65.83 \pm Ac	56.60 \pm d	104.50 \pm a
24-35 months	81.00 \pm b	63.81 \pm ABc	53.72 \pm d	99.93 \pm a
36-48 months	79.00 \pm b	66.40 \pm Ac	58.88 \pm d	96.56 \pm a
Overall mean	78.21 \pm b	63.36 \pm c	53.57 \pm d	100.32 \pm a
Body Mass Index				
12 & < 12 months	15.62 \pm a	13.39 \pm b	11.77 \pm b	17.33 \pm a
13-23 months	19.07 \pm a	13.47 \pm b	11.87 \pm b	13.69 \pm b
24-35 months	14.49 \pm 0.53	17.87 \pm 10.60	12.12 \pm 1.88	15.06 \pm 0.95
36-48 months	15.89 \pm 8.49	17.11 \pm 6.76	12.72 \pm 1.58	16.66 \pm 2.66
Overall mean	15.87 \pm a	15.28 \pm a	12.05 \pm b	15.87 \pm a
	3.55	6.11	1.63	2.10

Values with different small letters in row and capital letters in column are significantly different at (P < 0.05)

Table 2: Means \pm SD of Cu, Zn and Fe in children of different ages in different degrees of malnutrition

Age Groups	Degree of Malnutrition			
	1st	2nd	3rd	Control
Cu				
12 & < 12 months	63.43 \pm a	51.00 \pm a	46.00 \pm a	10.33 \pm b
13 - 23 months	61.33 \pm a	55.33 \pm ab	58.00 \pm ab	16.00 \pm b
24 - 35 months	62.00 \pm a	52.50 \pm a	47.33 \pm a	11.67 \pm b
36 - 48 months	59.00 \pm a	47.20 \pm a	50.44 \pm a	8.00 \pm b
Overall Mean	62.14 \pm a	51.65 \pm ab	48.61 \pm b	10.00 \pm c
Zn				
12 & < 12 months	58.00 \pm	53.00 \pm	47.80 \pm	47.33 \pm
13 - 23 months	42.66 \pm	55.00 \pm	44.00 \pm	52.00 \pm
24 - 35 months	51.00 \pm	46.00 \pm	49.55 \pm	50.33 \pm
36 - 48 months	48.00 \pm	52.80 \pm	47.55 \pm	49.33 \pm
Overall Mean	52.29 \pm	51.45 \pm	47.67 \pm	50.00 \pm
Fe				
12 & < 12 months	418.28 \pm a	422.20 \pm a	315.90 \pm a	98.67 \pm b
13 - 23 months	286.00 \pm	353.00 \pm	289.60 \pm	18.00 \pm
24 - 35 months	306.00 \pm a	306.00 \pm a	271.55 \pm ab	106.33 \pm b
36 - 48 months	286.00 \pm ab	356.00 \pm a	256.66 \pm ab	80.00 \pm b
Overall Mean	354.86 \pm a	364.41 \pm a	291.16 \pm a	64.50 \pm b

They reported that crowded environment, poor nutrition and unhygienic conditions were more likely factors of sickness in

Table 3: Means \pm SD of serum total protein, albumin and globulins in children of different ages in different degrees of malnutrition

Age Groups	Degree of Malnutrition			
	1st	2nd	3rd	Control
Total protein				
12 & < 12 months	8.01 \pm	9.30 \pm	9.16 \pm	9.22 \pm
13 - 23 months	8.35 \pm	8.56 \pm	7.59 \pm	9.79 \pm
24 - 35 months	7.55 \pm	11.09 \pm	9.49 \pm	9.94 \pm
36 - 48 months	8.89 \pm	10.31 \pm	9.53 \pm	9.59 \pm
Overall Mean	8.14 \pm	9.82 \pm	9.13 \pm	9.62 \pm
Albumin				
12 & < 12 months	3.19 \pm	3.13 \pm	3.18 \pm AB	3.44 \pm
13 - 23 months	3.12 \pm	3.29 \pm	2.61 \pm B	3.30 \pm
24 - 35 months	2.90 \pm	3.13 \pm	3.28 \pm A	3.41 \pm
36 - 48 months	3.30 \pm	2.96 \pm	3.19 \pm AB	3.44 \pm
Overall Mean	3.15 \pm	3.13 \pm	3.13 \pm	3.41 \pm
Globulin				
12 & < 12 months	4.82 \pm	6.17 \pm	5.98 \pm	5.78 \pm
13 - 23 months	5.23 \pm	5.27 \pm	4.99 \pm	6.49 \pm
24 - 35 months	4.66 \pm	7.96 \pm	6.21 \pm	6.53 \pm
36 - 48 months	5.60 \pm	7.35 \pm	6.34 \pm	6.14 \pm
Overall Mean	4.99 \pm	6.68 \pm	5.99 \pm	6.21 \pm

Values with different capital letters in a column are significantly different at (P < 0.05)

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Table 4: Means \pm SD of serum total protein, albumin and globulin of children of different socioeconomic status in different degrees of malnutrition

Socioeconomic Status	Degree of Malnutrition			
	1st	2nd	3rd	Control
Total Protein				
Low	7.89 \pm b	9.83 \pm ab	9.10 \pm b	11.94 \pm a
	1.27	2.66	2.98	2.69
Middle	8.79 \pm	10.84 \pm	9.23 \pm	8.69 \pm
	2.90	4.87	3.75	3.51
Albumin				
Low	2.94 \pm	3.11 \pm	3.08 \pm	3.53 \pm
	0.54	0.49	0.59	0.46
Middle	3.66 \pm	3.22 \pm	3.36 \pm	3.44 \pm
	0.72	0.50	0.73	0.24
Globulin				
Low	4.95 \pm b	6.71 \pm ab	6.02 \pm ab	12.85 \pm a
	1.06	2.66	2.80	2.65
Middle	5.13 \pm	7.62 \pm	5.88 \pm	5.25 \pm
	2.36	4.41	3.60	3.27

Values with different small letters in row are significantly different at (P<0.05)

Table 5: Means \pm SD of mid-arm circumference, percent weight and body mass index of children of different sex in different degrees of malnutrition

Sex	Degree of Malnutrition			
	1st	2nd	3rd	Control
Mid-arm Circumference				
Male	13.18 \pm a	11.92 \pm b	10.24 \pm c	13.90 \pm a
	0.98	1.57	1.72	0.74
Female	13.00 \pm a	11.32 \pm b	10.83 \pm b	14.50 \pm a
	1.73	1.68	1.20	0.41
Percent Weight				
Male	78.18 \pm a	63.86 \pm b	52.26 \pm c	100.20 \pm a
	2.36	5.91	5.88	3.19
Female	78.33 \pm b	62.55 \pm c	55.39 \pm d	100.41 \pm a
	2.08	4.39	5.40	6.79
Body Mass Index				
Male	16.35 \pm a	16.24 \pm a	12.09 \pm b	16.31 \pm a
	3.85	7.59	1.62	1.68
Female	14.08 \pm ab	13.71 \pm ab	11.99 \pm b	15.50 \pm a
	1.32	1.45	1.68	3.19

Values with different small letters in row are significantly different at (P<0.05)

Table 6: Means \pm SD of Cu, Zn and Fe in children of different sexes in different degrees of malnutrition

Sex	Degree of Malnutrition			
	1st	2nd	3rd	Control
Cu				
Male	62.91 \pm	52.44 \pm	51.68 \pm	10.00 \pm
	18.07	15.93	22.51	4.92
Female	59.33 \pm a	50.36 \pm a	44.33 \pm a	10.83 \pm b
	18.90	17.20	19.32	4.90
Zn				
Male	52.36 \pm	55.44 \pm A	47.36 \pm	50.20 \pm
	13.47	13.80	12.22	3.70
Female	52.00 \pm	44.91 \pm B	48.11 \pm	48.50 \pm
	8.72	6.35	6.84	2.89
Fe				
Male	352.00 \pm	402.88 \pm	292.96 \pm	104.20 \pm
	173.81	166.26	157.22	12.62
Female	365.33 \pm a	301.45 \pm a	288.66 \pm a	64.50 \pm
	174.13	107.16	146.09	57.79

Values with different small letters in row and capital letters in a column are significantly different at (P<0.05)

Table 7: Means of total protein, albumin and globulin in children of different sexes in different degrees of malnutrition

Sex	Malnutrition degree			
	1st	2nd	3rd	Control
Total protein				
Male	7.98 \pm	9.24 \pm	8.90 \pm	9.49 \pm
	2.00	2.92	3.33	1.59
Female	8.76 \pm	10.76 \pm	9.44 \pm	9.72 \pm
	0.55	2.89	2.79	4.74
Albumin				
Male	2.99 \pm	3.07 \pm	3.26 \pm	3.30 \pm
	0.62	0.51	0.67	0.37
Female	3.72 \pm a	3.24 \pm ab	2.96 \pm b	3.49 \pm ab
	0.54	0.44	0.50	0.21
Globulin				
Male	4.99 \pm	6.17 \pm	5.64 \pm	6.19 \pm
	1.65	2.79	3.05	1.58
Female	5.04 \pm	7.53 \pm	6.48 \pm	6.22 \pm
	0.06	3.14	2.74	3.79

Values with different small letters in row are significantly different at (P<0.05)

Table 8: Means \pm SD of mid-arm circumference, percent weight and body mass index of children of different ages in different degrees of malnutrition

Socioeconomic Status	Degree of Malnutrition			
	1st	2nd	3rd	Control
Mid-arm Circumference				
Low	13.40 \pm a	11.88 \pm b	10.53 \pm b	14.27 \pm a
	1.171.65	1.40	1.65	
Middle	12.50 \pm ab	11.00 \pm bc	10.31 \pm c	14.25 \pm a
	0.581.41	1.79	0.35	
Percent Weight				
Low	78.70 \pm b	63.94 \pm c	53.24 \pm d	97.70 \pm a
	2.31	3.94	6.19	3.25
Middle	77.00 \pm b	59.25 \pm c	55.00 \pm c	102.70 \pm a
	1.63	10.94	3.85	8.92
Body Mass Index				
Low	16.25 \pm	15.58 \pm	14.00 \pm	14.00 \pm
	4.17	6.69	1.57	2.64
Middle	14.91 \pm a	13.89 \pm ab	12.01 \pm b	14.34 \pm a
	0.72	1.29	1.99	3.30

Values with different small letters in row are significantly different at (P<0.05)

Table 9: Means \pm SD of Cu, Zn and Fe in children of different ages in different degrees of malnutrition

Socioeconomic status	Degree of Malnutrition			
	1st	2nd	3rd	Control
Cu				
Low	56.20 \pm a	50.08 \pm a	49.09 \pm a	13.00 \pm b
	16.72	15.51	23.02	9.65
Middle	77.00 \pm a	61.00 \pm ab	46.50 \pm b	11.00 \pm c
	9.31	21.32	12.08	7.07
Zn				
Low	52.60 \pm	51.33 \pm AB	48.23 \pm	49.37 \pm
	12.04	12.88	10.11	11.25
Middle	51.50 \pm	47.00 \pm B	45.25 \pm	51.00 \pm
	14.82	5.29	11.00	14.14
Fe				
Low	329.40 \pm a	358.30 \pm a	296.10 \pm a	107.67 \pm b
	151.38	163.31	154.51	96.52
Middle	418.50 \pm a	403.00 \pm a	269.80 \pm ab	61.00 \pm b
	211.08	107.51	141.46	60.81

Values with different small letters in row and capital letters in column are significantly different at (P<0.05)

the children of low socioeconomic status. Higher prevalence of malnutrition in children living in towns, as observed, might be suggesting the same above mentioned factors.

This investigation revealed that all types of milk-offering including breast, formula or cow/buffalo were more frequent to males than females, while solid food offering was more frequently offered to females. This has confirmed the earlier statement that male children were given more care than females. It was also observed that breast feeding was less frequently offered by educated mothers, while more frequently by uneducated mothers. This might be due to that educated mothers probably have less time for their children than for themselves and give less attention to the children.

Age: Data revealed that significantly (P<0.05) lower values of mid-arm circumference and percent weight in malnourished children than apparently healthy children (Table 1) which was in concordance with the results reported by Rao (1980). Body mass index showed significant difference in children below 23 months of age with lower values in 3rd degree of malnutrition which indicated that loss of weight was more rapid and that children below 23 months of age suffered more frequently from serious illness.

Results of the micro-elements showed higher levels of copper in malnourished children of all ages (Table 2). These malnourished children were actually suffering from various diseases. It has already been reported that the level increases in children suffering from leukaemia (Chan *et al.*, 1998), tonsillitis (Koltas *et al.*, 1997), Fe deficiency anaemia (Ece *et al.*, 1997) and common variable immune disease (Litzman *et al.*, 1995). However, it has been observed that copper decreases with severity of malnutrition (Guler *et al.*, 1994). The possibility of high Cu levels as observed might be related with release of copper containing enzymes thus resulting in

high serum copper levels or that the children were suffering from Wilson's disease, a condition of defective copper metabolism. However, the levels of serum total proteins including albumin were significantly or otherwise lower in malnourished children (Tables 4 and 3). Thus it might be possible that proteins were less available for the Cu binding. Hence free Cu levels were higher that ultimately resulted in overall higher levels of it in malnourished children. It is however, emphasized that further research must be undertaken to investigate the real reasons for the higher Cu levels in malnourished children. Zinc level, however, have showed non-significant difference between malnourished and apparently healthy children (Table 2). As previously stated that Zn concentration in serum decreases in children suffering from diarrhoea (Bhutta *et al.*, 1999). Present results have, however, not supported the earlier findings and the reason may be investigated in further studies. However, it might be possible that Zn levels are not affected by the such diseases. However, present results were in line with the findings of Khaldi *et al.* (1995) as higher Cu levels were observed during this study also.

Similar to Cu, significantly ($P < 0.05$) higher iron concentration was observed in malnourished than apparently healthy children (Table 2). It might also be possible that high levels of micro-elements in the diseased malnourished children could be due to medication containing these micro elements. It has previously been reported that recurrent infections and other diseases occurred more frequently in children with low levels of Zn (Hambidge, 1972) and Cu (William, 1982). However, the infection established in these children was probably not cured even though the micro elements level was raised. Rather it may be possible that high level particularly of copper (Hambidge, 1972) and iron (Garrow and James, 1993) might have become toxic and further resulted in toxic damage in the body tissues. However, results of serum proteins showed non-significant difference (Table 3).

Sex: Mid-arm circumference, percent weight and body mass index was significantly ($P < 0.05$) lower in severely malnourished children of both sexes (Table 5). Levels of Cu and Fe were significantly ($P < 0.05$) higher in female malnourished children than apparently healthy children of same sex (Table 6). Flodin (1953) in a study reported close correlation between plasma Cu and Fe with age and sex of children.

Similar to the age groups, present data regarding total proteins, albumin and globulins showed non-significant difference between malnourished and apparently healthy children of both sexes except for albumin which was lower ($P < 0.05$) in severe degree of malnutrition (Table 7) which might be related with the protein malnutrition in female.

Socioeconomic Status: The values for mid-arm circumference and percent weight were lower in malnourished children irrespective of socioeconomic status and these were further lowered in severe degree of malnutrition and was highly related with degree of malnutrition (Table 8) which was in concordance with the findings of Flodin (1953). It may be mentioned that in families of middle socioeconomic status, more attention is given towards other house and personnel related affairs except nutrition and hence they face protein malnutrition. The levels of Cu and Fe were also significantly ($P < 0.05$) higher in malnourished children of each socioeconomic class (Table 9). Serum proteins revealed close to normal levels (Table 4). However, lower levels in first degree malnutrition in children of low socioeconomic status might be related with the protein intake in the children of

this group.

Therefore, from this research work concluded that Diarrhoea, fever and malnutrition were the major diseases observed at pre-schooling age. Mid-arm circumference, percent weight and body mass index were lower in malnourished children and among malnourished children in 3rd degree of malnutrition. Malnutrition was more prevalent in children of 12 and <12 months of age and in males. In each sex, age and socioeconomic status groups, serum Fe, and Cu were higher in diseased malnourished children.

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