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An analysis of micronutrient trace elements in children dependent on various modes of feedings

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ABSTRACT

Micronutrient trace elements Zinc, Iron & Copper among 100 children's, dependent on various feeding methods, randomly selected from various parts of District Bannu, Khyber Pakhtunkhwa, Pakistan were determined by Atomic Absorption Spectrometer. Out of 100 children, 64 were male and 36 were female. The serum Zinc, Iron and Copper contents among male children's were 65.38 ±1.7 µg/dl, 64.69 ±2.2 µg/dl and 113.35 ±3.2 µg/dl and among female children were 59.09 ±2.4 µg/dl, 54.23 ±2.4 µg/dl and 123.23 ±5.2 µg/dl respectively, showing significant differences ($p<0.05$) among serum Zinc & Iron levels, while non-significant differences among Serum Copper level. Among 100 subjects, seven (07) were on mixed pattern, twenty eight (28) were bottled fed and sixty five (65) were mother / breast fed. BMI were 16.38 ±0.8, 15.66 ±0.5 and 14.11 ±0.4 in mixed, bottle fed & mother fed groups, showing significant difference from each other ($p < 0.05$). The serum Zinc, Iron and Copper levels in children dependent on mixed food were 56.33 ±3.8 µg/dl, 55.60 ±5.0 µg/dl and 81.61 ±8.04 µg/dl, on bottle feeding were 60.57 ±2.03 µg/dl, 58.86 ±3.3 µg/dl and 112.99 ±4.6 µg/dl, while on mother feed were 64.94 ±3.8 µg/dl, 62.39 ±2.2 µg/dl and 121.54 ±3.8 µg/dl respectively, showing significant (<0.05) differences among groups. It is concluded that the higher concentration of Zinc, Iron and Copper were found among children who were fed on mother milk as compared to those who were fed either on mixed food or on bottles milk.

INTRODUCTION

Although micronutrient trace elements are required in a very small quantities, but still human beings need them to have strong bodies and mental vitality, resist various diseases, and produce normal and healthy children. In most cases, the deficiency of micronutrient trace elements is either caused by inadequate access to food or high burden of disease or parasitic infestations. In addition the dietary and feeding practices of particular community also play a part. The deficiency of micronutrient trace elements has negative effects on the heath and body functions of human beings [1].

Vitamins and minerals, being required in very minute amount of our body tissues, but no doubt they have a vital role in growth and development and rightly support the term "micronutrients" [2]. The deficiencies of most micronutrient trace elements rich foods or their poor absorption have devastating health consequences on our bodies and mind [3]. Minerals can be defined as inorganic compounds, which are not obtained from animals or plants, but rather obtained from the earth crust [4]. Their lack increases the overall risk of mortality and morbidity, including poor cognition and intellectual development, poor immune functions and poor capacity of work. In fact the devastating effects of micronutrient deficiencies more pronounced on children, pregnant and lactating women [5]. It is evident that around the world, at least two billion people live with vitamin and mineral deficiencies. Each year 3.5 million children under five years die because of malnutrition and it also impairs hundreds of thousands of growing minds [6]. Moreover it account for an estimated 11% of the global burden of disease [7]. It is believed that gradually and silently micronutrient trace elements deficiencies affect people and entire communities in a vicious cycle of poor health, poor productivity and ultimate poverty. In majority of cases the affected victims never know the actual cause for their sufferings [8]. In this connection the present study was planned to analyze the level of micronutrient trace elements Zinc, Iron & Copper among children, who were on in various feeding methods with an objective to explore comparison among these methods.

MATERIALS AND METHODS

After getting approval from ethical committee of the institute of this cross sectional analytical study, the following protocols were adopted, for the determination of micronutrient trace elements in blood samples of children's<5 years age, belonging to various parts of District Bannu, Khyber Pakhtunkhwa, Pakistan.

Apparatus and Glassware:

To prevent contamination with trace elements, all glassware and bottles were beforehand drenched in weak 10% Nitric Acid (HNO_3) overnight and rinsed carefully with deionized water and double distilled water.

Instruments:

Atomic Absorption Spectrophotometer (AAS) model Hitachi Polarized Zeeman Atomic Absorption Spectrometer, Z-8200, Japan was used for elemental (metals) analysis.

Selection of Subjects:

Total of one hundred (100) children (age < 5 years), from different parts of District Bannu , who were fed on mother milk , on mixed food or bottles milk were selected for analysis.

Inclusion & Exclusion Criteria:

Children of 1 - 5 years age group were selected, while children with any organic illnesses, malnourished or children with family history of genetic disorders were excluded. Similarly children with hemoglobinopathies or using medicines were also not included in the study.

Collection and Storage of blood Samples:

Blood samples of approximately 5mL were drawn from antecubital vein using disposable syringes of 5ml with butterfly needles of 22G with all aseptic measures. The blood was transferred to a sterilized and already labeled glass tubes and serial numbers were endorsed. The blood was centrifuged in centrifuged machine (IEC Model DORP 6000 Centrifuged Machine) at 1500 2000 rpm for 10 minutes [9]. The separated layer of serum was clipped out with micropipette into sterile and properly labeled disposable plastic capped (eppendorf) tubes. All the tubes were stored at 20 ^{0}C to be used for determination of the desired micronutrient trace elements.

Serum Samples Preparation for (AAS):

The collected serum samples were subjected to wet digestion before elemental (Cu, Fe, Zn) determinations by using Atomic Absorption Spectrophotometer (AAS) model Hitachi Polarized Zeeman Atomic Absorption Spectrometer, Z-8200, Japan. One ml of serum was taken into a digestion flask and 10 ml of concentrated HNO_3 was added. Mixture was heated at 60 to 70 ^{0}C for about 15 minutes. After cooling, 5mL of $HClO_4$ was added in the flask. The contents of the flask were heated vigorously till the volume reduces to 1-2 ml. The contents were diluted up to the desired volume by adding de-ionized water [10].

Statistical analysis

All statistical analysis including the calculation of mean SD, range and variance were calculated. Level of significance was calculated using *t* test. p values > 0.05 were considered non significant.

RESULTS

Results of the study are tabulated at tables 1-3.

Table 1 shows that overall mean serum Zinc, Iron and Copper contents in 64 male and 36 female children. The overall mean serum zinc, iron and copper contents among male children were 65.38 ±1.7 µg/dl, 64.69 ±2.2 µg/dl and 113.35 ±3.2 µg/dl and the overall mean serum Zinc, Iron and Copper contents among female children were 59.09 ±2.4 µg/dl, 54.23 ±2.4 µg/dl and 123.23 ±5.2 µg/dl respectively. A significant differences (p<0.05) were obtained among serum Zinc & Iron levels, while non-significant differences were obtained among male and female children. The results in table 2, shows the distribution of serum Zinc, Iron and Copper contents in urban and rural children. The overall mean serum Zinc, Iron and Copper contents amongst 48 urban children were 62.84 ±1.94 µg/dl, 61.12±2.36 µg/dl and 117.80±4.68 µg/dl respectively and the overall mean serum zinc, Iron and copper contents among rural children were 63.37 ±1.99 µg/dl, 60.75 ±2.52 µg/dl and 116.08 ±3.67 µg/dl respectively, showing no significant difference among the rural and urban children however, the serum zinc levels of rural children were slightly higher than the urban children. Same is the case of copper which is found low in urban when compared to rural. The BMI and age were also not significantly different in both urban and rural children (P > 0.05).

The results shown in table 3, represent the distribution of serum zinc, iron and copper levels with respect to different mode of feeding including mother feeding, bottles or mixed feedings in the study subjects. In total of 100 subjects, seven (07) were on

Table 1 : Gender wise distribution of Micronutrients level:

Gender	N	BMI (kg/cm^2)	Age (months)	Zinc (µg/dl)	Iron (µg/dl)	Copper (µg/dl)
Male	64	15.17 ±0.37 a	48.91 ±1.83 a	65.38 ± 1.65 a	64.69 ±2.19 a	113.35 ±3.51 a
Female	36	13.86 ±0.45 b	43.25 ±2.68 a	59.09 ±2.38 b	54.23 ±2.44 b	123.23 ±5.15 a

Means sharing a common letter do not differ significantly, others differ significantly ($p < 0.05$).

Table 2 : Trace elements in urban & rural populations of the study

Residence	n	BMI (kg/cm^2)	Age (months)	Zinc (µg/dl)	Iron (µg/dl)	Copper (µg/dl)
Urban	48	14.72 ± 0.39 a	47.19 ± 2.21 a	62.84 ± 1.94 a	61.12 ± 2.36 a	117.80 ± 4.68 a
Rural	52	14.68 ± 0.43 a	46.58 ± 2.15 a	63.37 ± 1.99 a	60.75 ± 2.52 a	116.08 ± 3.67 a

Means sharing a common letter do not differ significantly, others differ significantly ($p < 0.05$).

Table 3 : Trace elements level in various mode of feeding

Method	N	BMI (kg/cm^2)	Age (months)	Zinc (µg/dl)	Iron (µg/dl)	Copper (µg/dl)
Mixed	7	16.38 + 0.78 a	55.00 + 2.66 a	56.33 + 3.79 a	55.60 + 5.01 a	81.61 + 8.04 A
Bottle	28	15.66 ± 0.50 ab	53.21 ± 2.17 ab	60.57 ± 2.03 ab	58.86 ± 3.29 ab	112.99 ± 4.59 B
Mother	65	14.11 + 0.37 c	43.26 + 2.02 c	64.94 + 1.87 b	62.39 + 2.18 ab	121.54 + 3.78 b

Means sharing a common letter do not differ significantly, others differ significantly ($p < 0.05$).

mixed pattern, twenty eight (28) were bottled fed and sixty five (65) were mother / breast fed. BMI in mixed group was 16.38 ±0.8 as well in bottle fed & mother fed it is 15.66 ±0.5 and 14.11 ±0.4 respectively. The result showed significant difference from each other (P = 0.05). The serum Zinc, Iron and Copper levels in children dependent on mixed food were 56.33 ±3.8 µg/dl, 55.60 ±5.0 µg/dl and 81.61 ±8.04 µg/dl. The serum zinc, iron and copper levels in children dependent on bottle feeding were 60.57 ±2.03 µg/dl, 58.86 ±3.3 µg/dl and 112.99 ±4.6 µg/dl respectively. The serum Zinc, Iron and Copper levels in children dependent on mother milk were 64.94 ±3.8 µg/dl, 62.39 ±2.2 µg/dl and 121.54 ±3.8 µg/dl respectively, showing a higher concentration of Zinc, Iron and Copper among children who were fed on mother milk as compared to those children who were fed on mixed food or bottles milk.

DISCUSSION

Zinc, Iron & Copper trace elements deficiencies are important nutritional problems and are widespread in many developing countries [11, 12]. Trace element micronutrients have long been accepted as indispensable for optimal health. The medical implication of trace elements is still somewhat contentious. Among the trace elements, iron, copper and zinc are of scrupulous interest.

In the present study zinc, iron, and copper and their relationships were investigated in children less than 05 years of age, living in the region of district Bannu (Pakistan). The result of the study shows that girls are more likely to suffer from under nutrition than boys, as it is evident from our results that serum zinc, iron and copper contents among male children were 65.38 ±1.65 µg/dl, 64.69±2.19 µg/dl and 113.35±3.15 µg/dl and among female children were 59.09 ± 2.38µg/dl , 54.23±2.44 µg/dl and 123.23±5.15 µg/dl respectively.

The health and nutrition profile of the girl child through the transition from early childhood to early womanhood does not come through as clearly as the threatened chances of her survival, unequal distribution of food in the family, with a definite gender discrimination against the female has been observed even in rich families in our society, but the lines of discrimination in poorer societies seem to be etched deeper [13]. Even when conservative families can afford to feed a good diet to both male and female children only the boys are given the rich diet and girls a much poorer one. This is mainly because of the conviction that a girl given rich food comes of age and starts menstruating earlier [14]. Otherwise in poorer families the female children are generally neglected and sons get preference in terms of best as well as larger share of food.

When we decided to undertake this study we were expecting that copper deficiency if present, would most likely affect the nutritionally vulnerable sectors of the children population. Contrary to our expectations, female children had the highest levels of serum copper in our study, as the copper contents among male children in our study were 113.35±3.15 µg/dl and among female children were 123.23±5.15 µg/dl respectively, showing a bit high concentration in female children as compared to male children. (Table 1). The observation that females tend to have higher serum copper levels than male children is not clear to us, although zinc deficiency has been reported with copper excess [15]. We think that the copper status of male subjects needs more clarification/evaluation with larger sample size.

In Pakistan there is significant difference between urban and rural children's population in terms of economic status ,life style and nutritional status. Ample evidence shows that urban children generally have a better nutritional status than their rural counterparts. Our results showed that serum zinc, iron and copper contents among urban children were 62.84 ±1.94 µg/dl, 61.12±2.36 µg/dl and 117.80±4.68 µg/dl, while in rural children, it were 63.37 ±1.99 µg/dl, 60.75±2.52 µg/dl and 116.08±3.67 µg/dl respectively, showing deficiency of micronutrients in rural children in comparison with urban children of District Bannu, Khyber Pakhtunkhwa, but the difference was not significant, which is an agreement to previous studies[16].

The results by urban-rural classification showed that iron deficiency anemia was more prevalent in rural than in urban children, while no significant differences were observed in the prevalence of zinc between urban and rural children respectively (table 2).The same has been supported by other investigators, in both urban and rural regions, females were more attributed to anemia [17]. In children, severe anemia can impair growth and mental development. Children with severe iron-deficiency anemia may also have an increased risk for stroke [18]. The low Hb and MCV are associated with low serum ferritin concentration in anemic children[19]. In the population survey it has been reported that children and women of childbearing age are more affected. It was found that iron deficiency anemia may vary from 17% to 70% in pre-school children; while in adolescents it may vary between 14% to 42% and in child bearing age from 11% to 40% [20].

Breast milk is the basic right of child and is the most excellent first food for babies [21].The nutrients present in the breast milk are the species specific formula for excellent growth and development. In the present study the children who were fed by their mother showed no deficiency of micronutrients as compared to bottle fed children and children who were dependent on mixed food . The period from birth to two years of age is widely recognized as the critical period, during which growth retardation and micronutrient deficiencies can occur. Nutritional vulnerability during this period results from poor breastfeeding and addition of complementary food, coupled with high rates of infectious diseases[22].At present, the feeding bottle are s viewed as a" symbol of modernity"; the majority of infants appear to be fed on bottles which can be purchased in the smallest shops(kokas), even in remote villages [23] .Breast feeding directly promotes overall health of the child and results in decreased childhood morbidity and mortality [24].Although breast feeding is well experienced in our community but the ratio of exclusive breast feeding is low. Mothers have meager knowledge of benefits of breast feeding. The lack of exclusive breastfeeding, early introduction of harmonizing foods and wrong preparation of formula milk may also be the causes of micronutrient deficiencies in bottle fed and mixed food dependent children.

CONCLUSION

It is concluded in our study that low serum Zinc and Iron, while some what high copper level were found in female as compared to male children of <5 year age (pre-school) of District Bannu, Khyber Pakhtunkhwa. Similarly, by comparing results, it showed a higher concentration of serum Zinc, Iron and Copper among children who were fed on mother milk as compared to those who were fed either on mixed food or on bottles milk.

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