Nutritional Values in Aspects of Essential and Non Essential Elements in Variety of Milk Samples by AAS and FES

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Summary: Milk makes a significant contribution to the human diet through provision of macro nutrient, vitamins and minerals. The exact composition of milk varies by species to naturally or contamination. It is recognized that imbalance quantity of minerals and trace element being a serious health hazards especially for infants. Therefore, some essentials elements like K, Fe, Co and Pb (as a non essential element) have been determined in locally available milk powder of infant formulas, milk powder of growing children, processed milk or tetra pack milk of different brands and fresh milk samples (cow and buffalo) by sophisticated analytical techniques flame emissions spectroscopy (FES) and atomic absorption spectroscopy (AAS). The range of mean concentration of elements (K, Fe and Co) in milk samples was found to be 650.00-1500.00mg/l, 2.76-8.93mg/l and 0.05mg/l respectively. The levels of these elements in milk powder of infant formulas (1&2) were compared with the standards of FAO/WHO, recommended values of the Committee on Nutrition of the American Academy of Pediatrics, human milk and cow’s milk.

Introduction
Milk and dairy products have been recognized all over the world for a long time as good for their sensory properties, as well as their beneficial influence on human health. On the other hand milk is an expensive raw material for different food products. As we know milk is a complete diet for infants, growing children and old peoples. For changing the taste various value added products are usually derived from the milk [1-3]. Furthermore, the market survey shows that to enhance the nutritional value of milk some additives are necessary for better results.

The composition of human milk is the gold standard by which infant nutrient needs are determined and the larger society benefits through reduced infant illness and health care cost. Babies who are not breastfed are given infant formula. In USA most infant formulas have a base of modified cow’s milk or soy protein. By the age of 6 months, a breastfed infant needs an additional iron source. Iron fortified infant cereals can meet this need. For formula-fed babies, iron supplementation is needed from birth. The AAP therefore recommends iron-fortified formula for all formula-fed babies [4].

The production of sterile milk of long keeping quality by continuous flow process at ultra high temperature (UHT) for short time, followed by aseptic packaging has been actively studied during the last two decades and accepted for liquid milk processing [5].

Milk is considered as complete diet rather than a drink, due to its contents of proteins, fats, carbohydrates, minerals and vitamins. Milk contains 30-36g/L of total proteins and its rate very high in nutrients quality [6]. Many methods are available in literature to estimate sugar, fats and proteins in milk [7]. Minerals contribute to the buffering capacity of milk, the maintenance of milk pH, the ionic strength of milk and milk’s osmotic pressure. Milk is known as an excellent source of calcium [8]. Calcium plays an important role in the pathogenesis of gallstones. On the other hand, maintenance of the appropriate blood Ca level involves bones, kidneys, intestine and parathyroid glands. Other minerals present in milk include sodium, potassium, magnesium and inorganic phosphates. Controlled quantities of sodium and fixed potassium/sodium ratio are recommended in case of some heart diseases. As magnesium is an essential factor in the glucose transport mechanism through cell membranes, it is important to determine its level in plasma and erythrocyte [9]. Trace elements in milk which are important in human nutrients include chromium (Cr), cobalt (Co) copper (Cu), iron (Fe), manganese (Mn) etc. Chromium (Cr) control glucose tolerance, cobalt (Co) present in vitamins B-12, copper (Cu) present in enzyme cofactor, iron (Fe) in hemoglobin and manganese (Mn) concern with hypocholestrol [10-13].

Iron is low in milk of many species relative to the needs of the neonate. Iron in milk is bound to lactoferrin, transferring, xanthine oxidase and some to caseins. Iron deficiency causes microcytic and hypochromic anemia. Potassium deficiency causes muscles weakness, vomiting and irregular heartbeat [14].

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Trace elements may enter the milk during milk synthesis or by contamination after the milk is removed from the animal; for example from metal containers, etc. Animal raised near industrial plants and near highways can have higher milk Cd and Pb.

It is well established that Pb and Cd are toxic and children are more sensitive to these metals than the adults. The symptoms of lead toxicity are low IQ, slow growth, iron deficiency, nervous system disorders, etc. in children, whereas in adults reproductive complications, hypertension and kidney failure observed [15].

ISO14000 has opened the field for enhancing the awareness towards healthy atmosphere and increment of every type of pollution degrading the quality of food.

In Pakistan various sources of contamination of milk sample exist, mainly due to poor environmental conditions of cattle farms, residential cattle colonies and food material, the animals take. WHO and others gave the concept of ISO 9000 and ISO 14000 to develop hygienic condition for industries (main sources of pollution) particularly mentioned here related to food. So it is very necessary to develop the education related to above shortcomings and for this purpose regular monitoring of milk samples should be under taken for establishing effective quality control of milk supplies available for human consumption [16].

In the metal elemental analysis of biological materials and food stuffs the digestion is very basic step, which brings the desired elements into solution. A variety of decomposition methods are available but wet digestion methods are generally preferred over dry ones. Atomic absorption spectrometry requires complete dissolution of elements [17-20].

Milk because of its extraordinary calorific value, nutrition importance, worldwide demand and being a very important commodity in different areas of food research [21-24].The theme of this study is to determine the concentration of some elements namely K, Fe, Co and Pb in variety of milk available in market for human consumption.

Results and Discussion

Table-1 shows that the average values of potassium, iron, cobalt and lead along with standard deviation in milk samples. In all types of samples, the mean concentration of potassium is the highest followed by iron, cobalt and the last is lead, as the potassium is micronutrient. It is clearly evident from Tables-1 and 2 that the concentration of potassium in infant formula-2 is the highest. The iron content in full cream milk powder sample is lower than the infant formula-1 and 2. Cobalt was found only in full cream milk powder.

Table-1: Concentration of metals in powdered milk samples.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Sample Brand</th>
<th>Potassium (mg/L ± SD)</th>
<th>Iron (mg/L ± SD)</th>
<th>Cobalt (mg/L ± SD)</th>
<th>Lead (mg/L ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Infant formula-1</td>
<td>850.00 ± 0.00</td>
<td>8.15 ± 0.34</td>
<td>ND*</td>
<td>ND</td>
</tr>
<tr>
<td>2</td>
<td>Infant formula-2</td>
<td>1500.00 ± 0.00</td>
<td>8.93 ± 0.89</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>3</td>
<td>Full cream milk powder</td>
<td>1216.70 ± 28.80</td>
<td>4.33 ± 1.53</td>
<td>0.05 ± 0.00</td>
<td>ND</td>
</tr>
</tbody>
</table>

* ND = Not Detected

Table-2 indicates that the mean concentration of potassium and iron in tetra pack 1, tetra pack 2, cow and buffalo milk samples. The level of potassium is found least in buffalo milk sample among all varieties of samples. While iron in tetra packs is lower than cow and buffalo milk samples. However, the iron level in buffalo milk sample almost identical to the cow milk sample. In South Asian countries, tetra packs, cow and buffalo milk are used alike in urban and rural areas, as a part of meal. The level of trace elements is greatly influenced by environmental conditions, food stuff they ingested and metabolic activities.

The concentration of cobalt was found in full cream milk powder, while in other cases cobalt was not detected. The level of lead in all samples was not detected according to Tables-1 and 2.

Table-2: Concentration of metals in tetra pack and fresh milk samples.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Sample Name</th>
<th>Potassium (mg/L ± SD)</th>
<th>Iron (mg/L ± SD)</th>
<th>Cobalt (mg/L ± SD)</th>
<th>Lead (mg/L ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Tetra Pack-1</td>
<td>1116.66 ± 20.86</td>
<td>2.76 ± 1.75</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>2</td>
<td>Tetra Pack-2</td>
<td>1066.66 ± 28.86</td>
<td>2.86 ± 1.01</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>3</td>
<td>Cow</td>
<td>1216.66 ± 28.86</td>
<td>3.30 ± 0.20</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>4</td>
<td>Buffalo</td>
<td>650.00 ± 0.00</td>
<td>3.31 ± 0.47</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

Table-3 shows the values of potassium and iron mentioned on the packets which meet the standards of FAO / WHO and recommended values of the committee on nutrition of the American Academy of Pediatrics and human milk and cow's milk [25].

Table-3: Concentration of potassium and iron in standard reference milk.

<table>
<thead>
<tr>
<th>Type of milk</th>
<th>Recommended Age</th>
<th>Concentration in mg/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mature Human Milk [25]</td>
<td>From 5days - 2 years</td>
<td>508b 0.3 – 0.9</td>
</tr>
<tr>
<td>Infant Formula-1</td>
<td>From birth – 6 months</td>
<td>662 8.1 (12.0)b</td>
</tr>
<tr>
<td>Infant Formula-2</td>
<td>Over 6 months - 2 years</td>
<td>810 7.8 (12.0)</td>
</tr>
<tr>
<td>Full Cream Milk Powder</td>
<td>Above 2 years – 11 years</td>
<td>NM 10</td>
</tr>
<tr>
<td>Cow [25]</td>
<td>NM</td>
<td>1440 Traces</td>
</tr>
</tbody>
</table>

b. Iron fortified formula
c. Not Mentioned
As compared with the data reported in Table-3, the values obtained for the concentration of potassium in infant formula (1 and 2) is found to be higher, but not exceeds the estimated safe and adequate daily dietary intake [26]. Although the concentration of iron is almost identical to the given standard in Table-3, but concentration of iron is lower than the iron-fortified formula.

The concentration of potassium in tetra pack (1 and 2), cow and buffalo is found to be lower than the literature values. This study, as mentioned in Table-2, did not show marked deviation from the literature with respect to iron content. National Academy of Sciences suggests a daily allowance of 11 mg/day iron for infant above 6 months to 12 months [14]. Human milk contains less iron than the others, but the amount and types of proteins in breast milk are ideally suited to the human infant. Certain proteins in human milk improve the absorption of iron, an important trait because breast milk is low in iron. Cow’s milk contains far too much protein for infant consumption, to and high casein content makes it much harder for the infant to digest and absorb [25]. Over-reliance on breast milk or formula can limit the infant intake of iron-rich food, resulting in a condition known as milk anemia. Anemia can affect a child’s energy level, attention span and mood. Between 6 months and the time of weaning, solid foods should gradually wake up an increasing proportion of the infant’s diet. Cobalt is a key mineral in the large vitamin B12 molecule, but it is not an essential nutrient and no recommendation has been established [15].

The selection of samples emphasize the fundamental needs of infant as milk provides the primary source of nutrition for young mammals before they are able to digest other meal. On the other hand neonatal period is one of the most critical with respect to nutrition, there is need to known the actual intakes of trace elements by fully breast feeding infants and fully top feeding infants during the first 6 months to 2 years.

**Experimental**

**Collection of Milk Samples**

The samples of milk powder (infant formula-1, infant formula-2 and powder milk for growing children or full cream milk powder), processed milk or tetra packs of different brands and fresh milk (cow and buffalo) were purchased from local market randomly. Before analysis, milk powder samples were oven dried at 60°C to constant weights. The fresh milk samples were stored in refrigerators airtight plastic bags till required.

**Preparation of Sample Solution for AAS and FES**

A two gram of powder milk samples, an aliquot of 50 mL of each sample of processed milk and fresh milk were taken separately in triplicate in 250 mL beakers. Powder milk samples, processed milk samples and fresh milk samples were treated with 50 mL mixture of concentrated HNO₃ and concentrated HClO₄ (1:1) directly, heated the samples on sand bath for four hours at 105°C, H₂O₂ was added and heated again until solution became colorless, The cold clear sample solution, filtered in 250 mL volumetric flask and made the volume up to the mark with deionized water. These digested samples were labeled as stock solution and diluted whenever required with deionized water. A blank was also prepared similarly under identical conditions.

**Reagents, Glasswares and Equipments**

All chemicals used were analytical AR grade (Merck) throughout the work; concentrated HNO₃, HClO₄ and H₂O₂ were used in highest purity range. Deionized water was used in the whole process. For filtration purpose Whatman no. 40 filter paper were used. Stock solutions of Fe, Co and Pb of 1000ppm from Merck were used, while standard stock solution of K was prepared by dissolving appropriate amount of KCl (Merck) of high purity solid.

All glassware which were used in this work were soaked out for 2 days in 20% nitric acid to ensure the surfaces were free from metals and then rinsed with deionized water at three time and dried in oven at 120°C.

Electrolyte K was analyzed using flame emission spectrophotometer (FES). For this task, a corning – 400 flame emission spectrophotometer (FES) was equipped. A Perkin Elmer – 3001 atomic absorption spectrophotometer equipped with standard burner and air-acetylene flame was used. Standard hollow cathode lamps were used as a radiation source for analyzing Fe, Co and Pb.

**Calibration Curve**

The calibration graph obtained for potassium is shown in Fig. 1. For the estimation of potassium concentration in the samples, a calibration
curve was drawn of the standard solution of potassium versus emission intensity values. Intensity of each standard solution was measured at least three times and the mean was plotted.

\[ R^2 = 0.9984 \]

Fig. 1: Concentration versus Intensity calibration curve for potassium.

Reference