Abstract

In forensic sciences, evidences play a vital role to establish relationship between the victim, suspect and the scene of occurrence. Evidences are recovered in various forms i.e. physical, biological and chemical which may reveal the actual story behind the crime. In case of a suspicious death, different organs of body are analyzed to determine the cause of death. A few of the substances which are used in form of drug may react like a poison. Some are consumed along with the food or already have been present in our body, but due to some infectious disease may give side effect and causes death. Now a day, the quantification of such elements can be done from the biological fluids by using advanced instrumentation and the cause of death can be determined easily. Zinc is an omnipresent trace element that occurs from the earth’s crust with an approximately concentration at about 70mg/kg. It is a blue white metal and soluble in water at a melting point 419.5 and boiling point 908 [1]. For the production of zinc, electrolytic method is accounted in which zinc-oxide is leached out by the calcined with sulfuric acid to form zinc sulfide solution. Later on, it is used to deposit on cathode from electrolyzed in cells. A few of minerals which contains zinc exist and are prepared for commercial use [2]. The list of such commercials is given below in (Table 1).

Table 1: Name of the few of minerals containing zinc and its percentage.

| Name of the mineral | Composition     | Percentage of zinc |
|---------------------|-----------------|--------------------|
| Sphalerite          | ZnS             | 67.00%             |
| Hemimorphite        | Zn₂SiO₇(OH)₂H₂O | 54.20%             |
| Smithsonite         | ZnCO₃           | 52.00%             |
| Hydrozinate         | ZnO             | 56.00%             |
| Zincite             | ZnO             | 80.30%             |
| Willemite           | Zn₃SiO₄        | 58.50%             |
| Franklinite         |                 | 15-20%             |

Zinc plays a vital role as a catalytic regularity ion with structural roles in enzymes molecule and for the growth of cell, cell proliferation, energy metabolism. Even, it is used for the DNA metabolism its repair and gene expression [3]. Zinc also serves as a component of hundreds of proteins and metalloenzymes including DNA polymerase, RNA polymerase, alkaline phosphatase, carbonic anhydrase, lactate dehydrogenase etc. There are zinc finger motifs in transcription proteins and these fingers links to their corresponding sites directly to DNA for initiate transcription and gene expression. It plays a vital role in vitamin A metabolism [4]. It helps in sequestration of free radicals and prevention against rapid peroxidation, which can be stated as one of an anti-cancerous effect, anti-aging, apoptosis and homeostasis [5]. Concentration of zinc in various age groups as per the requirement, estimated and lower are given below in (Table 2).
Age | Lower Nutrient reference intake (LNRI) | Estimated average requirement (EAR) | Recommended nutrient intake (RNI) | USA Recommended daily allowance (RDA) | World health Organization (WHO) | European Dietary reference intake
--- | --- | --- | --- | --- | --- | ---
0-3 months | 2.6 | 3.3 | 4 | 2 | | |
4-6 months | 2.6 | 3.3 | 4 | 3 | | |
7-12 months | 3 | 3.8 | 5 | 3 | 5.6 | 4 |
1-3 years | 3 | 3.8 | 5 | 3 | 5.5 | 4 |
4-6 years | 4 | 5 | 6.5 | 5 | 6.5 | 6 |
7-10 years | 4 | 5.4 | 7 | 8 | 7.5 | 7 |

| Males | | | | | | |
11-14 years | 5.3 | 7 | 9 | 8 | 12.1 | 9 |
15-18 years | 5.5 | 7.3 | 9.5 | 11 | 13.1 | 9.5 |
19-50+years | 5.5 | 7.3 | 9.5 | 11 | 9.4 | 9.5 |

| Females | | | | | | |
11-14 years | 5.3 | 7 | 9 | 8 | 10.3 | 9 |
15-18 years | 4 | 5.5 | 7 | 9 | 10.2 | 7 |
19-50+years | 4 | 5.5 | 7 | 8 | 6.5 | 7.1 |

Table 2: Concentration of zinc as per the age group.

**Toxicity of Zinc**

Zinc toxicity is a medical condition involving an overdose or toxic over exposure to zinc. Homeostatic ability of the biological systems aids in tight regulation of bodily zinc levels through the complicated framework for transport and distribution, import and export and sensing of zinc status to ensure that zinc do not participate in toxic reactions [6]. However, recent studies show that free ionic zinc is a potent killer of neurons, glia and other cell types. Serum zinc levels are important biomarker for zinc toxicity. Plus, fecal and urine zinc concentrations are also effective indications. (ATSDR, 2005). Acute toxicity results from ingestion of more than 20mg/day of zinc. Zinc concentration in brain is maintained within a narrow range of 600-800ng/L [7].

**Acute Toxic Effects of Zinc**

Although the zinc is a necessary component that is found in our body along with other metals. Still in some conditions, it works like a toxic material or substance in our human body. The conditions in which zinc will work like a toxicological substance includes are dermal contact, dermal irritants, eye irritants, ingestion etc. [8]. Dermal when brought in the contact with zinc powder or concentrated zinc solution, it can produce severe corrosive effects including blisters or permanent scarring. Zinc salt act as a dermal-irritants which can be absorbed by wounds [9].

Ocular acts as an eye-irritants causing pain, erythema, edema and cataract formation. Dilute solution of zinc chloride (<1%) are non-irritants and also used as eye drops. Ingestion is relatively non-toxic. However, when brought in contact with zinc chloride solution, microcytic anemia and decreased in blood platelets are reported. Hepatic Toxicity is when zinc exposure lead to an end stage of biliary cirrhosis. Nephrotoxicity is a high dose of zinc which leads to microscopic hematuria which is later accompanied by renal failure and mild albuminuria. Neurotoxicity is state as high doses ingestion leads to lethargy, staggering, difficult writing, anxiety, depression, somnolence, comatose. Immediate effects of zinc toxicity are vomiting, nausea, loss appetite, diarrhea, headache, gastric irritation, lethargy and anemia.

**Chronic Toxicity of Zinc**

Chronic toxicity results in 50-150mg of zinc consumption per day in people using zinc containing supplements and oral medicines. The effects caused by these supplements are mentioned i.e. weakened immune system with chills, fatigue and fever, copper metabolism disturbance, reduced levels of HDLP and iron functioning, impaired functioning of pancreatic enzymes, amylase and lipase, neutropenia, hypocupremia, hypoferrremia. Through available studies of oral zinc toxicity, a number of health effects in humans are identified which are named as: Decreased copper metalloenzyme activity, Hematological effects, Immunotoxicity. Reduced
levels in HDL cholesterol, Gastrointestinal effects, A zinc altered copper status, when zinc is induced it results in decreased in copper absorption [10].

Over exposure to zinc oxide results in fumes during the welding process and cutting or brazing on galvanized steel can result in metal fume. Often, it causes fever and flu like symptoms i.e., coughs, sore throat, chest pain, gastroenteritis which can be recordable within 3-10 hours of exposure and resolved within 1-2 days. According to OSHA, "The permissible exposure limit (PEL) is 5mg of zinc oxide fumes per meter cube of air."

Analysis of Zinc

Zinc has ubiquitous nature. Therefore, to avoid the contamination of biological sample, proper precautions should be taken. It can be done by avoiding the contamination during sample collection, inadequate reagent purity, atmospheric deposition. The trace evidence can be carried out on bodily elements like blood, plasma, serum, urine and on body tissue. Analysis of fluids requires proper knowledge and dynamics of its accumulation, metabolism and excretion [11]. It determines the level of exposure of that element and also measures the biological exposure indices. Biological monitoring of trace elements is the measurement of substance to assess the potential health risk which the substance may induce. The measurable unit is called as biological marker which helps to know individual's total exposure for that xenobiotic from all source including occupational exposure, diet, hobbies, medications and smoking etc. it also includes the host body to accumulate the trace elements and is also affected by inborn error in metabolism. Many factors affect excretion of metals over short periods, like State of hydration, Renal blood flow, Renal function. With the passage of time, the concentration variation of blood and urine is collected and equilibrium is set, where the average concentration during that period is directly proportional to the urine concentration.

Collection and Preservation of Urine Sample

To analyze the zinc from the urine sample, appropriate collection and preservation is a necessity. The collection and preservation process have three distinct categories i.e. ambient, refrigerated and frozen. If the analysis is to be performed with 24 hours or 48 hours, the sample can be preserved at 21-250 C temperature. Second type of preservation is prefect for less than 28 days in which, urine samples are preserved at 40 C temperature. And the third type of preservation is known as frozen in which samples are preserved at -200 C temperature for more than 28 days' latency period.

Although now a days, numerous preservatives are present although these three types of preservatives (50% acetic acid, 6N Hcl and toluene) are considered best in case of zinc for urine preservation. 50% acetic acid is oftenly firstly used preservative in which 25 ml of it is sufficient for 24 hours. Secondly toluene is used for preservation, in which 30 ml is mixed with urine solution for preservatives. In the last as an alternate preservative, 6N HCL is used for preservation of zinc suspected urine sample. In this preservative, 1 ml 6N HCL is mixed with 5 ml of urine for 24 hours preservation.

General Instructions for Specimen Collection

During the collection of urine samples, few instructions should be followed by the researchers. In this process, Specimens for trace elements should not be collected from patients who have received gadolinium, iodine or barium containing contrast material in the past 96 hours. Because the presence of these metal contrast agents is known for it interfere nature with lab techniques for trace elements analysis. Secondly, the collection of specimens in a dusty environment should be avoided to prevent contaminations [12]. The urine samples should never be done in colored containers, metallic containers i.e. pan or metals urinals. It should be always collected in a clean plastic container without a glued cap insert. Urine should be mixed well before aliquoting and a specimen identification label should be attached to the urine aliquot tube and capped tightly.

Interaction of Zinc with Other Essential Trace Elements

Copper and Zinc

The levels of copper containing enzymes (like superoxide dismutase, serum ceruloplasmin and plasma LDL cholesterol) are altered. In 1984, Fischer and his colleagues reported that copper metalloenzymes activity is more sensitive indicator of copper status than plasma copper levels. When zinc is induced, reduction in absorption of copper takes place. Hence, result in copper deficieny with unknown mechanism. Excessive intake of dietary zinc results in induction of intestinal metallothionein, which has higher affinity for copper than zinc. Therefore, copper get absorbed into intestinal mucosal cells. It suggests that increased intake of copper can result in reduction of zinc toxicity [13].

Calcium and Zinc

Calcium levels do not affect the absorption of zinc directly. In 2000, Lonnerda, Heth and Hoekstra (1965) had reported a decreased absorption of zinc during calcium administration in diet and increased dietary calcium enhance the rate of zinc loss because of short clearance half time.

Zinc and Iron

Women administrated with prenatal supplements containing (60mg/day) showed low percentage of zinc absorption as compared to ladies who were not taking such supplements. In 1999, Bougle and his colleagues conducted study on the significant correlation between iron content in diet and zinc absorption i.e., increased dietary iron results in reduction of zinc absorption but at lower iron intake levels, iron has no effect on the absorption of zinc Lonnerdal (2000).
Quantification of Zinc

Quantification of zinc can be done by: Atomic Absorption Spectroscopy which is an analytical technique that measures the concentration of elements by wavelength of light which is specifically absorbed by them [14]. Mass spectrometry is an analytical technique by which chemical substances are identified by sorting the gaseous ions in electric and magnetic fields according to their mass to change ratios. ICP-MS technique is used when metal ions are heated to such a high temperature that they are stripped off their electrons and are reduced to atomic nuclei, which are further analysed by atomic weight. This technique has a very high sensitivity with an error of ±5 of true concentration [15].

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