PK 76000014

A paper to be submitted for publication in
Pakistan Journal of Medical Association.

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TRACE ELEMENT ANALYSIS : A DIAGNOSTIC TOOL

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SEPTEMBER,1976

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ABSTRACT

Human body continuously assimilates a variety of elements from the environment and the concentration of these elements in blood is regulated by means of various homeostatic mechanisms. Some of the elements, though present in very small amounts, perform highly specialized functions in initiating many biochemical reactions. These elements, known as essential trace elements, are closely related to human diseases as their deficiency or excess induces physiological changes. Many diseases such as hypertension, atherosclerosis, diabetes etc; are related to trace element imbalance. Therefore the measurement of trace elements in body fluids and tissues can be effectively employed for diagnostic tests.

TRACE ELEMENT ANALYSIS: A DIAGNOSTIC TOOL

Introduction

Analytical chemistry has played a prominent role in the study of environment and fundamental processes of life. The analysis of biological materials has provided vital information regarding the origin of life by showing that the complex organic compounds, which constitute the living organisms, are in fact formed from such primeval constituents as hydrogen, ammonia, water, methane and inorganic salts, In the field of clinical diagnosis analytical chemistry has made significant contributions by providing a sound base for quantitative chemical measurement. The elucidation and control of pathological processes are becoming evermore dependent on chemical analysis and the demand for diagnostic chemical tests has increased exponentially during the last two decades. Since diseases are simply manifestations of the malfunction of biochemical processes, if becomes necessary to study and understand the biochemical and physiological role of the elements and molecules that form biological systems. Such studies will unravel at least part of the mysteries of the operation of complex biological systems, will help to devise new diagnostic tests, and will give us an opportunity to contribute to the relief of suffering.

Living organisms are basically composed of macro-molecules such as proteins, lipids, carbohydrates and nucleic acids etc. These macro-molecules are formed mostly by the combination of only a few elements. However, studies of biological systems have shown that a considerable number of additional elements, though present in very small amounts, also influence many vital functions. Living organisms continuously assimilate a greater or smaller variety of molecular or elemental constituents from the environment for their proper functioning. This accumulation occurs through lungs, the skin gills, the epithelium of the gut or other organs, and the concentration of the elements in blood are regulated by means of various homeostatic mechanisms. The different body cells selectively collect elements from the blood according to their specific functions. For instance red blood corpuscles concentrate iron and zinc whereas the cells which produce haemocyanine concentrate copper.

Essential Trace Elements

In addition to 12 essential elements (Hydrogen, carbon, nitrogen, oxygen, sulphur, phosphorous, chlorine, potassium, sodium, magnesium, calcium and iron) which constitute the bulk of the living matter, the tissues have been found to contain a number of other elements, which occur only in small amounts and are therefore known as trace elements. Some of these trace elements have been accumulated due to environmental contamination whereas some belong to a special group of elements known as "Essential trace elements".

The essential trace elements are linked to specific organic compounds via coordination or covalent bonds and perform specialized functions in initiating biochemical reactions. Many of these reactions are catalyzed by enzymes which are highly selective in their action. The activity of these enzymes is in turn dependent upon the presence of certain trace elements in their chemical structure and in their absence they become inactive adversely affecting the functioning of biological systems. It is, therefore, important to know which trace elements are essential for biochemical processes.

The basic knowledge regarding the essential trace elements has mainly stemmed from the modern nutrition research and chemical analysis of the various body organs, fluids etc. These investigations have proved that thirteen trace elements namely copper, manganese, zinc, cobalt, iron, molybdenum, selenium, iodine, chromium, tin, vanadium, silicon and fluorine are essential for animal and man(1-4). In addition to these, eight other trace elements have been found to occur regularly in the body but no essential role has yet been established for them.

Trace Elements and Diseases:

Trace elements are closely related to human diseases as the deficiency or excess of these induces physiological changes and distrubs the normal functioning of the organs. For example, such common diseases as anaemia and goitre are well known to be related to iron and iodine deficiency respectively. The following examples illustrate the vital bearing of trace elements imbalance on various diseases.

The normal excretory pathway of copper, manganese and certain other metals in man is through the kidney, liver and biliary system. Therefore a major disturbance in the metabolism of trace elements would lead to an increase in the concentration of these metals in the liver. The Wilson's disease has been known to be related to genetically determined abnormal metabolism of copper (5). In this disease the amount of copper increases in the liver and urine whereas its concentration decreases in blood. A few cases of Wilson's disease were investigated by Fell (6) to determine the copper contents in the liver, urine and blood of the patients and the normal subjects. The liver, urine and blood of the patients were found to contain 145 μg-Cu/g, 521 μgCu/24 hours and 46.5μg Cu/100 ml as compared to

25.2 μgCu/g, 48 μgCu/24 hours and 113 μgCu/100 ml respectively found in normal persons. This investigation clearly indicates that the trace element analysis can be effectively employed for diagnostic tests.

The disturbance in the metabolism of zinc, iron and copper also affects the human growth. The retarded growth of dwarfs is believed to be closely related to zinc deficiency (7). This deficiency not only results in the failure of gonadal development and growth at the time of puberty but also produces anaemia and enlargement of liver and spleen. Retarded growth which could not be accounted for by the usual causes for impairment could be explained on the basis of zinc deficiency. The affected youths in Iran and Egypt were found to contain low concentration of zinc in the plasma, urine and hair as compared to the controlled subjects(7). The affected youths when fed supplementary zinc showed an accelerated uptake and decreased excretion of this element by tissues which clearly indicates the special role of zinc in the causation of the syndrome.

Zinc deficiency is related to the birth weight of newborn children. It has been observed that during pregnancy the blood and hair of women contain subnormal amount of zinc (8). A reasonable explanation of this deficiency is that the demand of zinc and calcium by foetus and placenta exceeds the ability to maintain normal levels of these elements.

Diabetes has been observed to be related to disturbance of chromium metabolism (4). Chromium which is a co-factor of insulin is required by the body to maintain normal glucose utilization. The deficiency of this element slows down the rate of removal of glucose from the blood stream and the impaired glucose tolerance is an early symptom of diabetes. Since chromium is transported by transferrin (an iron transporting specific protien) it competes with iron for transferrin in blood. Normally transferrin has enough binding capacity for transport of iron and chromium. However in certain diseases such as haemochromatosis iron begins to accumulate in large amounts and the free iron-binding capacity of transferrin in serum becomes almost zero. About 80% of such patients develop diabetes within a few years due to disturbance of chromium metabolism, This observation also indicates that a competition among essential trace elements and the disturbance of an equilibrium between two trace elements results in certain pathogenic conditions.

<u>Cardiovascular diseases:</u>

Cardiovascular diseases appear to be related to socio-economic and technological development in a country as is evident from the fact that the cardiovascular mortality rate in developed countries is much higher as compared to the under developed countries. However with the increasing technological progress these diseases may also become major health problem in developing countries. Although we possess good knowledge of medical and surgical treatment of cardiovascular diseases today, nevertheless it is necessary to adopt preventive measures to curb the alarming increase of heart diseases.

Systematic studies of trace elements in relation to cardiovascular disease would provide valuable information. These studies are important as the man made alterations in the environment through the use of fertilizers, food additives, food processing, treatment and softening of drinking water and industrial pollution of air and water tend to bring about changes in the mineral balance in man. Several clinical and experimental studies indicate that chromium, zinc, manganese, vanadium, fluorine, silicon and copper appear to be beneficial to cardio-circulatory functions whereas cadmium, lead and cobalt are harmful to cardiovascular health (9).

Cadmium plays a number of detrimental roles in heart diseases and produces hypertension and sclerosis of small arteries in kidney, heart and other organs (10-12). Although it decreases blood cholesterol but increases lipid deposition in the aorta walls and thus favours formation of atherosclerotic plaque. Some studies suggest that cadmium induces hypertension in man. This suggestion is supported by the clinical observations that hypertensive patients contain high concentration of this element in their kidneys and urine as compared to normal subjects (13-15).

Experimental studies show that the hypertension induced by cadmium in rat can be reversed by the administration of zinc (16). Since the two elements are chemically similar, they compete for the same binding sites and the removal of cadmium by zinc helps to reverse the hypertension as the blood pressure is directly related to the molar ratio of cadmium to zinc in the kidneys. The animal is likely to develop hypertension when the molar ratio is higher than 0.35. This observation seems to be true for man as the cadmium to zinc ratio in urine increases with age and reaches a maximum in the age group of 40 to 50 years. In this age group the people are becoming increasingly affected by heart diseases (17).

The elements manganese and chromium seem to exert beneficial effects against atherosclerosis and prevent the development of this disease in animals (18). Manganese facilitates lipid metabolism whereas chromium improves cholesterol catabolism and excretion (10). The heart and aorta of atherosclerotic patients contain subnormal amount of manganese and chromium as compared to healthy subjects. The chromium level in tissues of population with higher incidence of cardio-vascular diseases such as North Americans has been found to be about four times less than those (Africans) with lower incidence of this disease (14). Similarly the chromium level in kidneys and aortas is about seven times lower.

The elements zinc, manganese and nickel have been reported to be related to myocardial infarction (19). The injured heart tissue shows decrease in the concentration of zinc which may be related to the disappearance of lactic dehydrogenase from the infarcted heart tissue. The concentration of manganese and nickel also show a sharp decrease in injured myocardium which is followed by a marked increase of these elements in the plasma. The increase in concentration of these elements is so rapid and specific that it may be used as a diagnostic indicator of a recent myocardial infarction (20-22).

Conclusion

The above mentioned examples clearly show that trace elements play an important role in disease machanisms. Thus the determination of trace elements in body fluids and tissues can be effectively employed for clinical diagnostic tests.

However the information obtained from these analysis will be meaningful only if the test samples are properly collected and transported to the analytical laboratory without any contamination. This necessitates a close association between the analytical chemist and the physician. Therefore most physicians in advanced countries are arranging cooperative programs with analytical laboratories for trace element studies.

Since trace elements always occur in very small amounts in the body their determination renders it necessary to use highly sensitive and accurate analytical techniques such as neutron activation analysis, emission spectrography, atomic absorption spectrometry and mass spectrometry. In Pakistan all these and certain other analytical facilities are available at Pakistan Institute of Nuclear Science and Technology(PINSTECH), Rawalpindi. The analytical chemistry laboratory is collaborating with international organisations like IAEA and WHO on the measurement of trace elements in various types of biological materials. In our laboratory neutron activation analysis, a nuclear technique, has been extensively used for multitrace element analysis. The results of some of these analyses are shown in Table-I which indicates the sensitivity and precision of this technique.

It is high time that a serious planned effort be undertaken in the country to establish positive correlation between the incidence of major diseases and the deficiency or excess of trace elements. For this purpose, it is necessary first to launch a reliable statistical survey of the population in selected areas (such as the one performed some years ago in Gilgit for iodine deficiency) to determine the normal levels of trace elements in a healthy sample. Pathological cases can then be studied in relation to a control group. Since the facilities for trace element analysis are now available in Pakistan, we should try to utilise them to the full for the well-being of the populace.

TABLE - I

Results of the determination of trace elements in IAEA Samples(23).

Sample	*Amount of the element in µg/g		
	Copper	Manganese	Zinc
Powdered Milk (A/8)	1.4±0.1	4.4±0.2	32.8±1.6
Wheat Flour (V-2/1)	4.7±0.2	37.2± 1.9	31.8± 1.5
Mashed dried potatoes (V - 4)	4.2±0.2	2.7± 0.1	10.3± 0.5
Calcinated animal bones (A - 3/1)	5.1± 0.2	31.9± 1.8	171.0± 7.7
Fish Solubles(A-6)	4.6± 0.2	4.1± 0.2	17.3± 0.8
Oysters (MA-M-1)	336± 6.4	75.2± 5.7	2534± 190
Animal Muscle (H-4)	3.1± 0.1	2.1± 0.1	101.8±4.6

* Average of triplicate measurements.
The error quoted is the standard deviation of these runs.

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