

Spectroscopic Investigation of the Hair of Diabetic Patients

Alhakim Ali¹, Vishwa Nath Verma^{2,*}

¹Quality Assurance Chemist, E.B. Beharry and Company, Mandela Avenue, Georgetown, Guyana, South America

²Department of Chemistry, Faculty of Natural Sciences, University of Guyana, Turkeyen Campus, Georgetown, Guyana, South America

*Email address: professorverma@ymail.com

ABSTRACT

Diabetes is a major disease condition that is accountable for 5% of annual cause of death globally and it continues to escalate. In this research the levels of minerals in persons with type 2 diabetes were investigated using hair as the biopsy specimen. The philosophy behind is that minerals are involved in countless metabolic functions in all phases of the life process, an imbalance of which can have widespread implication, while hair due to its physiology serves an invaluable record of the body's exposure to minerals. Hair metal testing is a fascinating new diagnostic tool and often gives unexpected clues to mineral imbalances in the body. In executing the research samples of head hair collected from 6 type 2 diabetic patients were washed to remove exogenous contaminants, then subjected to open vessel acid digestion and subsequent analysis for the elements Ca, Cd, Cr, Cu Fe, K, Mg, Na and Zn using Flame Atomic Absorption Spectroscopy (FLAAS). After comparing the concentration of the elements to a range that is considered to be normal it was found that Ca, Zn, Fe, Mg, K, Cu, Cd and Cr were off range while Na was normal. Upon investigating the function and effects of the elements that were off range it was found that a deficiency of chromium and an excess of iron could be a possible cause of diabetes.

Keywords: Diabetes, Metals, Hair, AAS

1. INTRODUCTION

Diabetes is a set of related diseases in which the body cannot regulate the amount of sugar (glucose) in the blood. Blood glucose is metabolized by the liver from the food which you eat. In a healthy person, the blood glucose level is regulated by several hormones, one of which is insulin. Insulin allows glucose to move from the blood into liver, muscle, and fat cells, where it is used for fuel. People with diabetes cannot produce enough insulin (type 1 diabetes) or don't use insulin properly (type 2 diabetes), or both.

In 1975 the Global Environmental Monitoring System (GEMS) recommended that human hair be utilized as one of the important materials for biological monitoring of trace metal levels in human beings.

The hair is a fine structure that is found on the surface of the skin except on the eyelids, the palm of the hand and the sole of the feet. Human hair is 80% protein, 15% water and small amounts of lipids and inorganic materials. The inorganic content of the hair is 0.25% to 0.95% on a dry ash basis. Each strand of hair consists of three layers.

CUTICLE or outer layer of colourless cells, which forms a protective surface to the hair. It regulates the chemicals entering and damaging the hair, and protects the hair from excessive heat and drying. The cells overlap, like roof tiles.

CORTEX or middle layer which forms the bulk, consisting of long spiral chains of cells like springs. Each cell is made of bundles of fibres.

These are composed of small bundles of macro fibrils which in turn are formed from even smaller bundles of proto-fibrils - all long, spiralling, ladder like chains. The way these fibres and cells are held together determines the strength of hair, its thickness, curl and elasticity.

MEDULLA The central space of the hair. It serves no useful purpose, and is not always present.

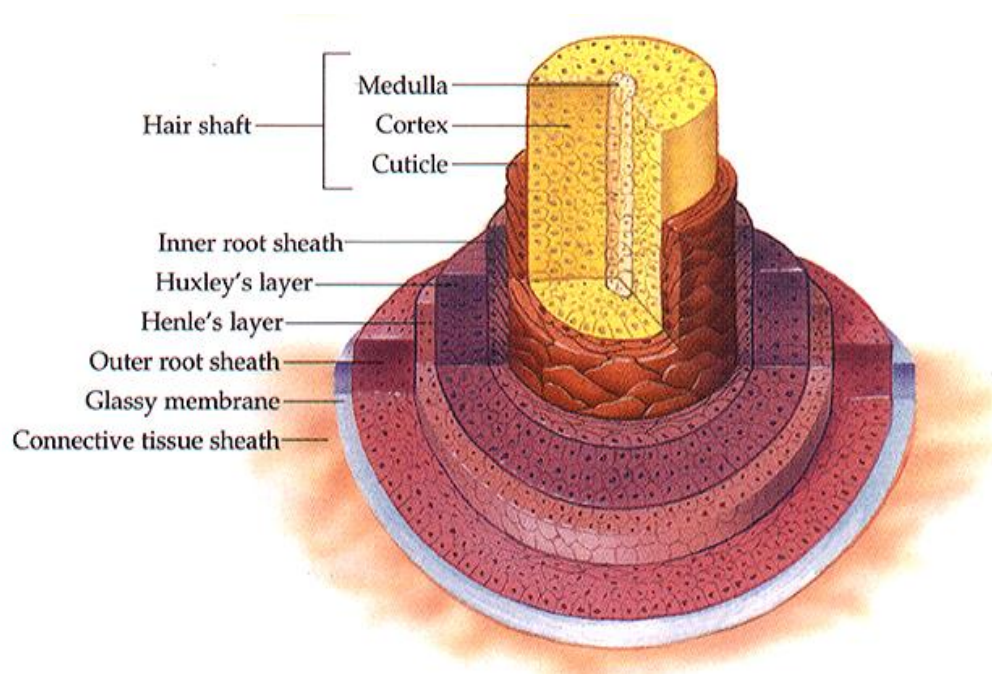


Fig.1. Structure of the hair shaft

Below the surface of the skin is the hair root, which is enclosed within a hair follicle. At the base of the hair follicle is the dermal papilla.

The dermal papilla is fed by the bloodstream which carries nourishment to produce new hair. The dermal papilla is a structure very important to hair growth because it contains receptors for male hormones and androgens.

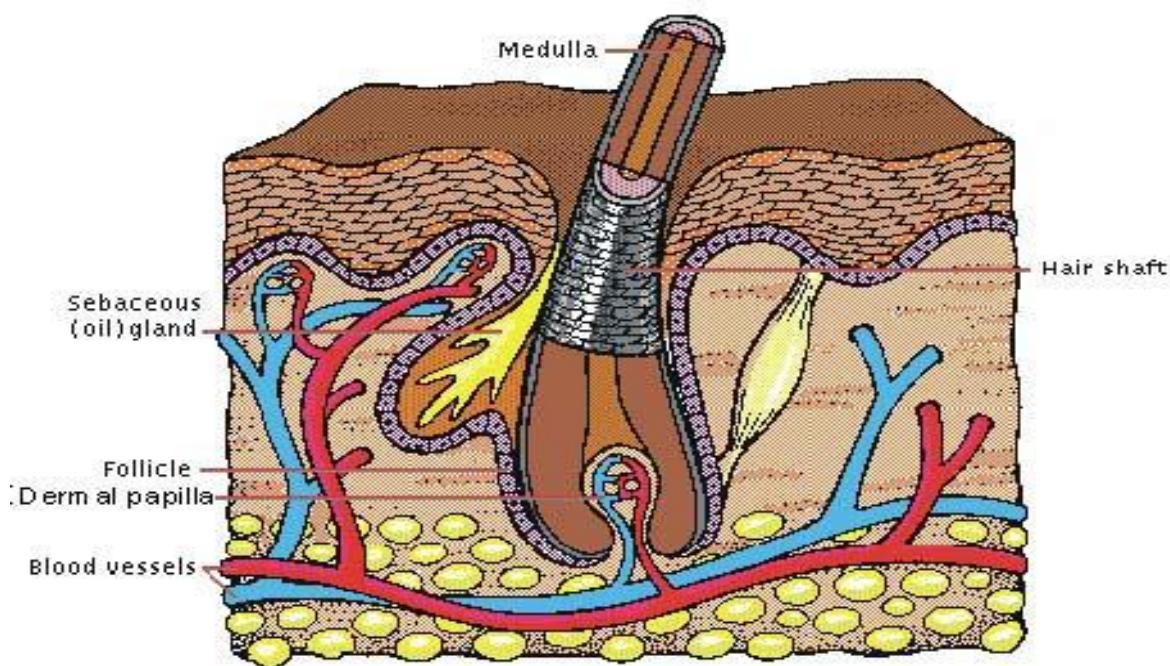


Fig. 2. Structure of the hair follicle

Trace mineral analysis or hair tissue mineral analysis (HTMA) is a soft tissue mineral biopsy. A biopsy is an analysis of a body tissue, in this case to detect mineral levels. Hair is classified as a soft tissue of the body. Hair analysis provides a reading of the mineral deposition in the cells and interstitial spaces of the hair over a 2-3 month period.

There are thousands of biochemical reactions that ultimately control our metabolism, digestion and the regeneration of body tissues. The vast majority of these reactions depend on certain trace minerals for their activity.

The most important nutrient metals include zinc, copper, calcium, magnesium, iron, selenium, lithium, cobalt, manganese, and phosphorus and the electrolytes sodium and potassium. If minute amounts of these essential minerals are not there to fuel the processes then your ability to regenerate, metabolize or breakdown noxious substances is compromised.

The growing hair follicle is richly supplied with blood vessels, and the blood that bathes the follicle is the transport medium for both essential and toxic elements. Because of the exposure of hair follicles to the blood supply during growth, elemental concentrations of the hair reflect concentrations in other body tissues.

Reinhold, Kfoury, Ghalambor, and Jean, C have studied zinc and copper concentration in hair of men (1). Analysis of zinc levels in hair of man was also reported by Strain, Steadman, Lankau, Berliner and Pories (2).

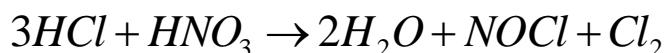
A brief information about this research was presented in the 61st. Southeast Regional Meeting & Conference, American Chemical Society, SERMACS, Puerto Rico, USA.(2009), October 21-25 (3).

2. EXPERIMENTAL

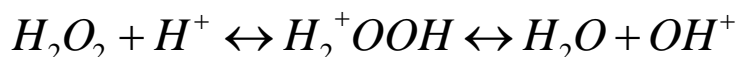
Hair samples were collected from the back of the head since hair from this area show least variability in element content¹. The back of the head include areas such as the nape of the neck, posterior vertex and posterior temporal regions. A representative sample of not more than 0.5g was weighed to the nearest 0.001g.

- 18 ± 0.1 ml of concentrated hydrochloric acid and 6 ± 0.1 ml of nitric acid was added to the sample in a fume hood.
- Addition of 5 ± 0.1 ml 35% hydrogen peroxide to aid in complete oxidation of organic matter.
- Addition of 5 ml HPLC grade water² to improve solubility of minerals and prevent temperature spikes due to exothermic reactions.
- The sample mixture was then placed on a hot plate, operating at the low setting, for about 45 minutes, and then allowed to boil down to about 13 ml.
- The digest was allowed to cool to room temperature.
- A reagent blank was also prepared using the same reagents and quantities used in sample preparation, placed in vessels of the same type, and processed with the samples.

The aim of the digestion is to solubalise the metals of interest in the hair sample. Digestion process is a combination of oxidation/destruction of organic matter and dissolution of the mineral phase to a certain extent to bring the elements of interest into solution. Aqua regia digestion consists of treating samples with a 3:1 mixture of concentrated Hydrochloric acid and concentrated Nitric acid respectively. This forms Nitrohydrochloric acid (NOCl), according to equation 1, which has a strong oxidising effect and gives almost complete dissolution of organic samples. The NOCl destroys organic matter to give carbon dioxide and water.



Also as part of the digestion hydrogen peroxide will be used. Under acidic conditions hydrogen peroxide undergoes an equilibrium reaction to give reactive hydroxyl radical (equation 2) which aids in oxidation of organic matter.



2. 1. Identification of metals present in the sample by Flame Atomic Absorption Spectrophotometer

In this technique a liquid sample is aspirated i.e. aerosolised and mixed with combustible gases such as acetylene and air or acetylene and nitrous oxide. The mixture is ignited in a flame whose temperature reaches from 2100 to 2800 °C. In the combustion process the atoms of elements of interest in the sample are reduced to free unexcited ground state atoms which absorbs light at a characteristic wavelength as illustrated in fig. 14 below.

The characteristic wavelength of the elements is specific and accurate to 0.01 to 0.1 nm.

To provide the element with specific wavelengths, a light beam from a lamp whose cathode is made of the element being determined is passed through the flame.

Then a device such as photonmultiplier can detect the reduction in the intensity of light due to absorption by the analyte, and this can be directly related to the concentration of the element in the sample.

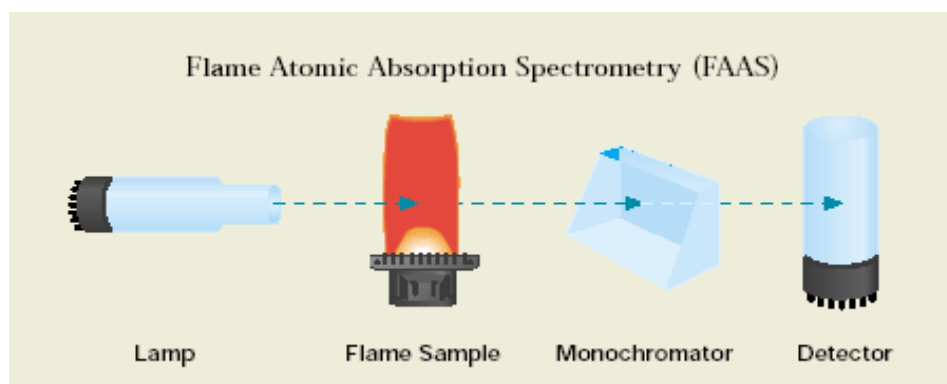


Fig. 3.Operational principle of an atomic absorption spectrometer

Table 1. Concentration of elements found in the six patients/samples

Metals tested for	Concentration of metals in parts per million (ppm)					
	Sample 1 ⁺	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6
Ca (ppm)	3327.16	594.63	1196.84	2054.65	466.09	2665.38
Zn (ppm)	327.23	830.48	1005.22	3066.31	1346.18	3959.02
Fe (ppm)	1380.10	725.11	784.70	2121.96	1295.04	995.36
Cu (ppm)	2.81	nd [‡]	2.07	0.29	1.35	31.63
Cd (ppm)	26.14	35.63	21.43	15.67	3.75	11.60
Mg (ppm)	302.20	33.75	280.47	347.57	147.90	393.80
K (ppm)	431.49	691.25	754.51	642.64	297.60	431.08
Cr (ppm)	nd	nd	nd	nd	Nd	nd
Na (ppm)	362.10	442.40	327.56	426.60	225.51	166.29

+ Sample or patients , nd = Not detected

Bar chart showing the distribution of the elements in the six Diabetic patients/samples

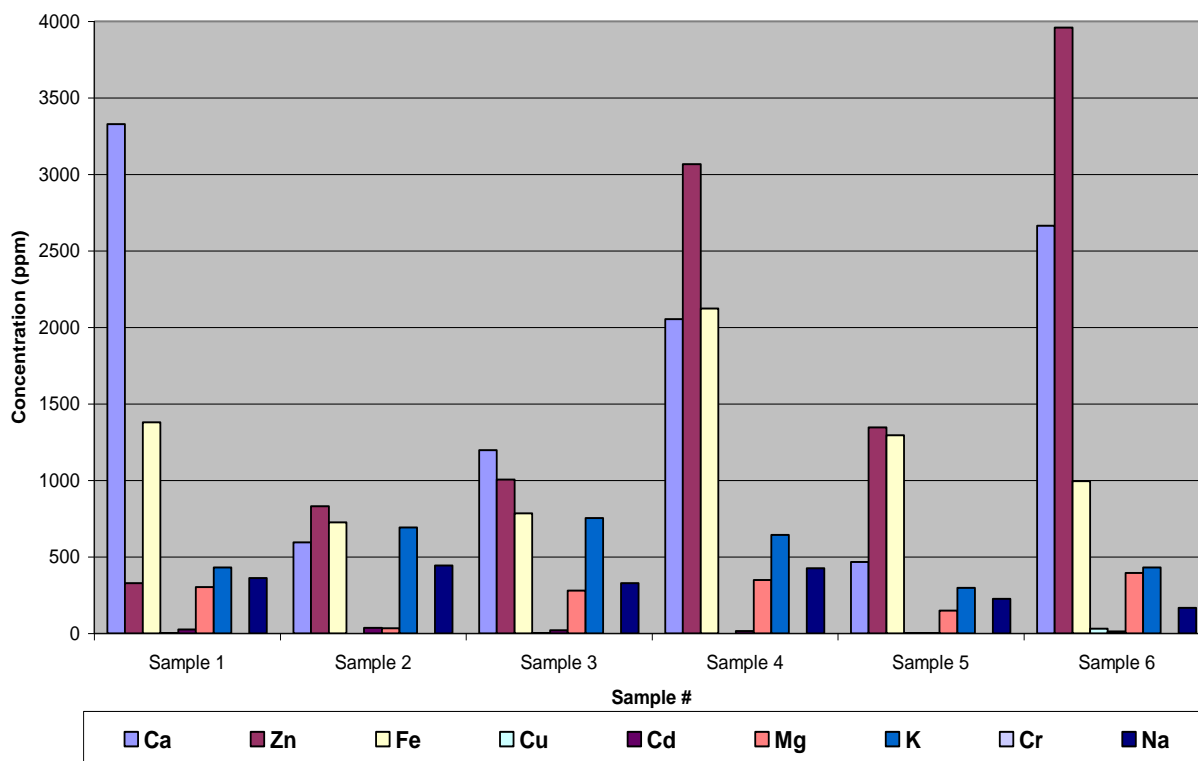


Fig. 4. Bar Chart showing the distribution of the elements in the six diabetic patients/samples

3. RESULTS AND DISCUSSION

3. 1. Statistical Analysis for Calcium

Ca (ppm)	3327.16	594.63	1196.84	2054.65	466.09	2665.38
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$$\text{Mean} = \frac{3327.16 + 594.63 + 1196.84 + 2054.65 + 466.09 + 2665.38}{6}$$

$$= \frac{10304.75}{6} = 1717.458333$$

3. 2. Standard deviation

x_i	$x_i - \bar{x}$	$(x_i - \bar{x})^2$
3327.16	1609.701667	2591139.457
594.63	-1122.828333	1260743.465
1196.84	-520.618333	271043.4487
2054.65	337.191667	113698.2203
466.09	-1251.368333	1565922.705
2665.38	947.921667	898555.4868
		$\Sigma = 6701102.774$

$$s = \pm \sqrt{\frac{\sum_{i=1}^N (x_i - \bar{x})^2}{N-1}} = \pm \sqrt{\frac{6701102.774}{6-1}} = \pm 1157.68$$

Relative Standard Deviation (S_r)

$$S_r = \frac{s}{\bar{x}} \times 100\% = \frac{1157.68}{1717.45833333} \times 100\% = 67.41\%$$

Calculations for the other elements were done similarly the results. Now what could have contributed to some of the elements being exceedingly high in concentration while some were very low ? Research indicates that there are antagonistic and synergistic interactions between different minerals(4). Antagonistic minerals are inversely related when the level of one goes up, the other goes down.

Examples of antagonistic minerals are sodium and zinc, calcium and magnesium, copper and potassium, iron and chromium. On the other hand synergistic minerals are directly related, when the level of one goes up, the level of the other goes up and examples are iron and aluminium, copper and calcium, manganese and sodium.

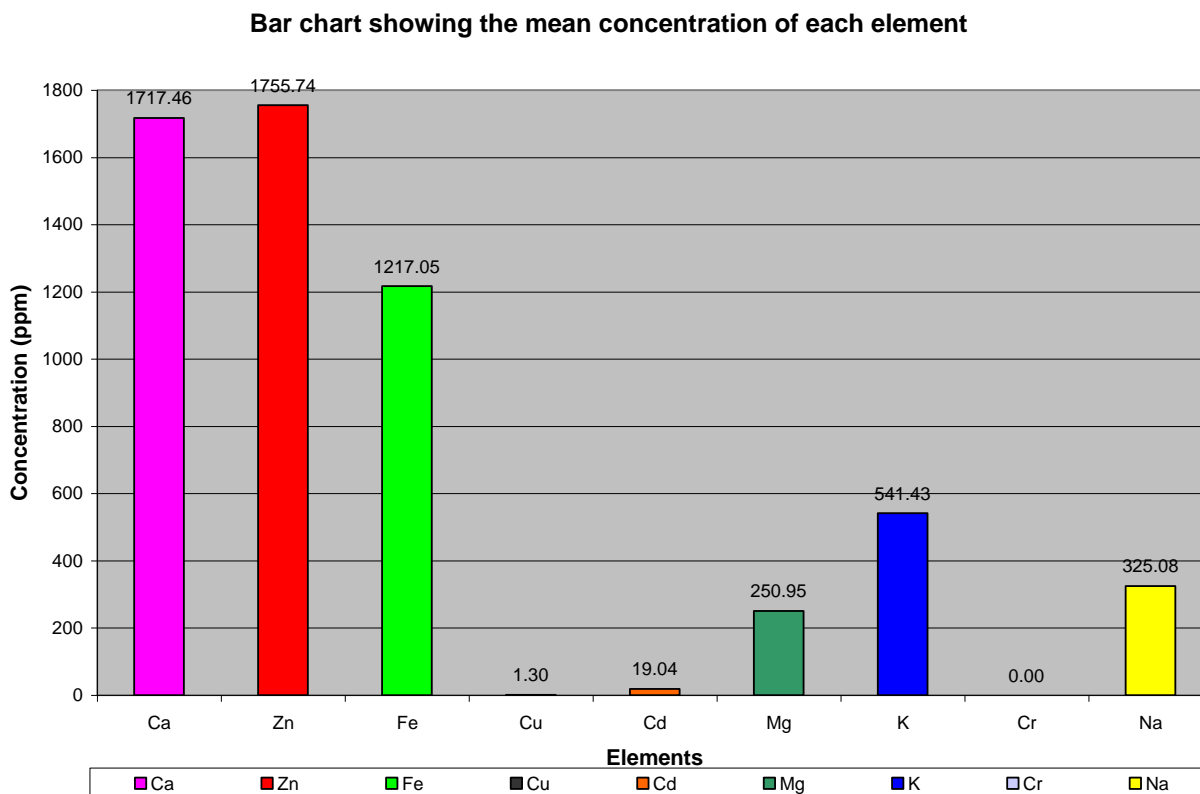


Fig. 5. Chart showing the mean concentration of each element.

4. CONCLUSION

From analysing the function, the possible effect of the elements that fell outside of their respective reference range and the different interactions between the minerals it can be recognised that a deficiency of chromium could be a contributing factor to diabetes since it is synergistic with insulin. Also an excess of iron can be indirectly linked to diabetes since it antagonises chromium. Apart from this scientists have found that eating foods high in simple sugars stimulate chromium loss through the urine. In addition, refined carbohydrates are devoid and heavy exercise deplete the body of chromium. In addition, chromium binds with transferrin, a transporter of iron in the body, so high levels can deplete chromium of chromium and other Trace mineral analysis of hair is very useful in determining a person's level of essential minerals as well as toxic burden. study trace mineral analysis was done on hair samples from six Type 2 Diabetic patients analysing the elements; Ca, Zn, Fe, Cu, Cd, Mg, K, Cr, and Na. Then analysing the function and the possible effect of the elements that fell outside of their respective reference range it was found that chromium which was deficient could be a contributing factor to diabetes since it increases insulin sensitivity and also excess iron indirectly since it antagonises chromium.

Further the study made on the general medicines given for the treatment of diabetic patients such as Glinil, Diamicon, Meftormine, Glucotrol, Chlorpropamide, Glucobayand, Gluburide contain good amount of chromium (5) which directly support to our findings reported here.

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