Hair Mineral Analysis
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Hair element analysis remains an important tool in the nutritional and environmental assessment of patients. In spite of various objections in the literature that the analyses may be poorly controlled, that different laboratories produce may different results from the same sample and also that the clinical interpretation of the results may be obscure [1], a measurement of the elemental concentration in recently-grown hair provides an integrated view of the element status in the follicular cells and their blood supply, unaffected by short term fluctuations in the nutrient intake of the subject.

Hair analysis at Biolab has recently been re-standardised and, with the introduction of the state-of-the-art trace element analysis, ICP-MS (Inductively Coupled Plasma – Mass Spectrometry) technology, we now report hair mineral analysis results with an adjusted reference range relevant to this technology.

Hair growth

The structural proteins of hair are formed as a filament arising from the matrix of follicular cells in the epidermal epithelium. Human hair is approximately 80% protein and 15% water, with smaller amounts of lipid and inorganic substances [2]. Hair growing from the human scalp normally follows a definite growth cycle: anagen (the growth phase), catagen (a period of controlled regression of the hair follicle, when the cells become inactive and the hair fibre stops growing) and telogen (when the follicle is in a resting state and the hair may fall out) [3]. Normally more than 90% of human head hair is in the growth phase and it is at this stage that elements from the follicular cells and their blood supply are incorporated into the structure of hair.

These may be nutrient elements, such as zinc and magnesium, present in the follicular cells as enzyme co-factors, or toxic elements, poorly handled by renal excretion, which are dumped into the hair as part of a detoxification mechanism. Hence mercury, lead and arsenic show considerable enrichment in hair as compared to blood levels. Other elements, such as zinc and magnesium, show some enrichment over blood levels (reflecting their high intracellular concentrations), while others, such as iron, show no enrichment over blood levels (which for iron are, of course, much higher than serum levels).

So elements are normally incorporated into the filamentous structure of hair for plausible, predictable biological reasons. As the hair follicle emerges through the skin surface, the process of keratinisation (incorportation of cysteine residues) seals the formed elements within the protein structure, making the hair a resilient and long-lasting tissue [4] that can be used for a variety of analyses. The presence of sulphydryl groups from cysteine means that hair will selectively chelate and indefinetly retain heavy metals.

Limitations to the use of hair analysis for assessment of minerals status

While the cycle of hair production normally continues for the whole lifespan, other factors, such as adverse reactions to drugs and tumors, radiation damage and other toxic reactions, may lead to the physical destruction of the hair follicle. This will affect the overall rate of hair growth and the incorporation of elements into the hair shaft. Human hair normally grows at about 1.0 cm per month, but, for example, prolonged zinc deficiency, or protein-energy malnutrition [5], slows down hair growth and may thus elevate the concentration of heavy metals in the hair (since the same mass of mercury, lead and cadmium is being incorporated into a smaller volume of hair tissue).

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Many cosmetic treatments and colourants contain metals, especially magnesium and copper. If hair that has had such treatments in incorporated into the analytical sample, very high values will be recorded for these elements, which may mask deficiencies. Shampoos may contain sodium, potassium, zinc and selenium in substantial amounts and it is not always possible to wash these elements from the hair, either with normal rinsing in the bathroom or *ex-vivo* in the laboratory [6].

Procedure for hair element analysis

Hair samples are analysed by ICP-MS for a number of nutritional elements (calcium, chromium, cobalt, copper, iron, magnesium, manganese, phosphorus, potassium, selenium, sodium and zinc) and also for toxic elements (aluminium, arsenic, cadmium, lead, mercury and nickel). Careful sample preparation is used to remove adhering particles and fluids which may also contain these elements. Since hair treatments, such as colourants and highlights, are mineral-based, patients are advised to wait for twelve weeks before submitting hair clippings for analysis if one of these treatments has been used.

To provide an index of normality, each set of hair element results is compared with reference intervals which have been derived from Biolab's extensive database. We have recently improved our sample preparation technique (the hairs are now dissolved by microwave in a closed container) and this has resulted in better recovery of lead and mercury, in particular. It is intended to introduce several more elements into the profile in due course, at which time the reference intervals will be reviewed again.

Quality control of the analyses is carried out by repeat re-analysis of pooled hair extracts, as well as by analysis of serum and blood samples with known elemental concentrations. There is no external quality assessment scheme for hair, but we have validated our results and reference ranges by comparison with certified reference hair samples from the National Institute for Environment Studies, Japan [7] and from the Community Bureau of Reference of the Commission of the European Communities [8]. The analytical methods (as opposed to the sample preparation techniques) used for hair analysis are the same as those used for plasma, blood and urine elements, which are subject to external quality assessment in recognized schemes.

Interpretation of results: are hair element concentrations of clinical significance?

A variety of scholarly and US government-funded studies have concluded that hair element analysis is a valid means to screen for mineral deficiencies and for toxic element exposure [2, 9, 10, 11]. A comprehensive review conducted by the U.S. Environmental Protection Agency (1979), which examined over 400 studies, concluded that …"if hair samples are properly collected and cleaned, and analyzed by the best analytic methods, using standards and blanks as required, in a clean and reliable laboratory by experienced personnel, the data are reliable" [9].

Systemic deficiencies of magnesium, zinc, selenium, chromium, manganese or copper will be reflected in low hair concentrations of these elements. Toxic levels of nutritionally important minerals will also be reflected in their hair concentration [6].

Enhanced exposure to, or ingestion of, aluminium, arsenic, cadmium, lead, mercury and nickel will result in higher than normal levels of these elements in hair tissue. This may not correlate directly with blood levels or with clinical symptoms, since it is part of a de-toxification mechanism, in which toxic metals are sequestered into hair to prevent the expression of their adverse biological effect. Hair toxic metal concentrations are, however, a sensitive measure of exposure to these elements [9, 10].

Much work, too extensive to detail here, has also been published by outstanding researchers on the relationship between measured hair elements and various clinical conditions, for example cardiovascular disease and myocardial infarction. A Finnish-Austrian study [12] reported in 2001 that men with the highest hair mercury content had double the risk of suffering a myocardial infarction and three times the risk of dying of cardiovascular disease, as compared to men with the lowest hair mercury content (this is a reflection of the transition metal activity of mercury and its ability to catalyse the oxidation of low-density lipoprotein). Another study [13] reported on low hair magnesium levels (along with other elements) which were recorded in subjects who had already suffered a myocardial infarction.

Our conclusion is that hair element concentrations, based on appropriate samples and analysed using the best methods, can provide important and valid information over a wide variety of clinical conditions.

The reference intervals currently used for hair elements are shown below in Table 1.

TABLE 1
Reference intervals for hair elements in micrograms per gram of hair [5]

ELEMENT	Reference Interval	COMMENTS
CALCIUM (Ca)	**200 - 2800**	Hair calcium levels are affected by PTH secretion and bone turnover, including periodontal disease, as well as by ingestion of hard water. High hair calcium and magnesium can be an indicator that hair treatments (eg bleaching and perming) have been used by the patients within three months prior to hair sampling.
CHROMIUM (Cr)	**0.10 – 1.50**	Results can be used to assess chromium status [14]
COBALT (Co)	**0.01 - 0.20**	Reflects B12 status as well as cobalt exposure
COPPER (Cu)	**10 - 100**	Low levels reflect copper deficiency; hair copper is increased in high oestrogen states. Zinc/copper ratios change in malignancy
IRON (Fe)	**5.0 - 30.0**	Reflects whole blood concentrations, but is not a good test of iron deficiency (which should be investigated by serum analysis)
MANGANESE (Mn)	**0.20 – 2.00**	High levels reflect manganese intoxication; low levels are often found in epilepsy, hyperactivity and neurological disorders
MAGNESIUM (Mg)	**60 - 160**	Low levels reflect tissue de-saturation of magnesium; high levels are caused by increased ingestion or exposure
PHOSPHORUS (P)	**100 - 200**	Reflects bone turnover and PTH secretion
POTASSIUM (K)	**50 - 300**	High hair potassium has been reported in coeliac disease; low levels may result from alcohol abuse. Plasma potassium should be used to assess current status and treatment
SELENIUM (Se)	**0.40 – 2.00**	Low hair selenium is an indicator of poor prognosis in a variety of conditions; high levels reflect selenium intake as well as selenium-containing anti-dandruff shampoos (eg Selsun) [14].
SODIUM (Na)	**50 - 1000**	Sodium levels above this range usually reflect exposure to certain shampoos
ZINC (Zn)	**160 - 240**	Low hair zinc reflects poor intake and zinc unavailability as an enzyme co-factor. High hair zinc can paradoxically be caused by slow hair growth as a result of inadequate zinc status, thereby allowing more time for low levels of zinc to be incorporated in the hair shaft. High hair zinc can also be a sign of use of zinc pyrithione-containing anti-dandruff shampoos (eg "Head & Shoulders").
ALUMINIUM (Al)	**< 50.0**	Much investigated for a possible correlation with CNS symptoms, especially in ageing subjects. May come from acidic food cooked in aluminium pots.
ARSENIC (As)	**< 1.00**	Elevated levels reflect increased arsenic exposure from drinking water, alcoholic beverages, shellfish or contact with treated wood and pesticides
CADMIUM (Cd)	**< 0.10**	From exposure to smoke and tobacco, but also Ni/Cd batteries, PVC plastics (yellow colour), motor exhausts, sludge and fertilisers
LEAD (Pb)	**< 2.00**	A useful measure of lead exposure; hair represents a detoxification pathway for lead, keeping the body burden at lower levels
MERCURY (Hg)	**< 1.00**	High hair mercury results from leaking dental amalgams, consumption of contaminated fish and other sources. Hair mercury reflects the body burden of organic mercury and is much studied as a medium for assessing exposure
NICKEL (Ni)	**< 1.40**	From jewellery, stainless steel, dental crowns and a variety of industrial sources.

Patient preparation

If the hair has been bleached or permed, none of the treated hair should be included in the sample for analysis. If the treatment has been carried out recently, a period of 12 weeks should be allowed to elapse before sampling. The subject may continue to take nutritional supplements, which will not factitiously distort the hair results.

Specimen requirements

Hair should be cut from the back of the head, or nape of the neck, as close to the scalp as possible. At least 0.5gm of hair is required, which is about one heaped tablespoon full. Only hair up to 1½" (4cm) from scalp can be used. Please allow for this when the hair is long by sending in a larger total sample, for example 2 tablespoons-full of hair.

Turn-around time

Hair analysis is now carried out weekly in the laboratory. The report will be sent directly to the referring doctor.

References:

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