POLICY PAPER – HAIR ANALYSIS

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1. Background

Occupational Medicine

Occupational medicine involves dealing with hazards at work - both prevention and dealing with and adverse health effects. Hazards may be physical, chemical, psychological, ergonomic or biological. Assessing exposure to chemical hazards includes monitoring of workplace air, history and physical examination,

as well as biological monitoring in certain situations. Biological monitoring includes sampling and analysis of blood and urine, analysis of exhaled breath, and in some cases, hair analysis.

Occupational physicians are involved in giving advice to individuals and employers regarding appropriate monitoring techniques at work, and in interpreting results of tests ordered by themselves or others. They may be required to give expert opinions to environmental workers compensation authorities in a legal context, or appear in court as an expert witness.

Some chemicals such as heavy metals have been used in manufacturing and other industry for hundreds of years. For example cases of lead poisoning have been documented throughout most of recorded history. As many heavy metals have been used widely for industrial purposes and are persistent in the environment, over-exposure in the occupational and general societal settings has been the subject of extensive study for many years.

Biological monitoring and measurements

Occupational and general environmental exposure to chemicals may be assessed using measurements of levels

- present in the environment (air and soil) and
- present in the human body (known as biological monitoring).

Measurement in air or soil gives an indication of environmental levels, but biological monitoring gives a more precise measure of the total amount of substance which has been absorbed via different routes such as inhalation, ingestion, or skin absorption. It also takes account of excretion through exhalation, urine and faeces.

Although biological monitoring is often more accurate in assessing body burden of chemical substances, there may be advantages and disadvantages associated with the specific matrices that are collected and analysed:

- Urine collection is non-invasive but usually indicates the concentrations of water-soluble metabolites and represents recent exposure.
- Blood samples are invasive, requiring clinical expertise for sampling and are not well tolerated, particularly by children.
- Exhaled air may be useful for volatile materials, but requires specialised sampling, storage and transport procedures.

- Hair, nails and deciduous teeth are tissues in which chemicals may be deposited over the medium to longer-term, so could represent a more integrated measure of exposure over such periods.
- Children's deciduous teeth may be collected and analysed as they naturally fall out in childhood to be replaced by permanent adult teeth, although sampling is opportunistic and there may be disadvantages in the time delay while waiting for the teeth to shed.

Individuals who believe they have suffered over-exposure and adverse health effects from chemical substances sometimes feel the tests carried out by their medical practitioners may not be sufficiently comprehensive. Despite extensive blood tests and other investigations, they may not accept a conclusion that their symptoms do not arise from chemical exposure. Some people look for further information, different analytical methods and different methods of biological monitoring.

In response to this, environmental toxicological analysis and diagnosis services have been established to provide alternative analytical approaches. In these cases, hair and nails may be viewed as preferred tissues for analysis, since they are robust and convenient to collect, usually without particular technical expertise or other considerations. They may be dispatched by mail or other routes without need for specialist handling or storage.

Aims and objectives of this paper

Hair analysis can be used in research and in limited clinical applications. It is a useful tool in limited circumstances.

However it is also being used by a limited number of practitioners, diagnosing individuals with chemical related diseases, often for people with non specific symptoms. Rigour in the application of the test results is often missing, and the negative impact on people's lives when an inappropriate diagnosis is given can be substantial.

The aim of this paper is to provide useful and practical advice, based on the science of the topic. The objectives are

- To assist practitioners' understandings of the potential pitfalls of hair analysis
- To assist practitioners' understandings of the issues that need to be addressed if hair analysis is going to be used
- To highlight the fallacies in using the procedure in many clinical situations.

2. Applications

Biological monitoring for chemical exposure using hair samples has been reported in both occupational and environmental settings for decades (Clarkson et al 1988). It has been applied to a wide range of putative chemical exposures ranging from drugs, inorganic compounds (metals, fluoride) to organic pollutants (pesticides, PCBs).

Metals

Occupational exposure to metals is the greatest reported application of hair analysis. Examples have included overtly toxic metals such as lead, mercury, chromium, cadmium and others as well as essential metals including selenium, copper, zinc, and iron. These investigations are predominantly hygiene-based, using the levels of metals detected in hair as an index of relative exposure rather than an index of toxicity and may demonstrate correlations between hair concentrations of selected metals and blood concentrations (e.g. Lee and White 1980).

Environmental applications of hair analysis to metals exposure have been well reported for lead, mercury (and

methylmercury) and arsenic from seafood, groundwater and other sources (Barbosa et al 2005; Weihe et al 2005; Hughes 2006).

Pesticides

Pesticide levels in hair, particularly the organochlorines, organophosphates and pyrethroids, have been reviewed by Tsatsakis and Tudaki (2004). This review also examined the use of hair for other persistent pollutants such as dioxins (PCDD), dibenzofurans (PCDF) and biphenyls (PCBs). Maternal hair has also been examined during pregnancy to determine exposure to a wide range of pesticides (Ostrea et al 2006). In each case the authors were generally supportive of the use of these indicators as exposure markers, and demonstrated correlations between hair and blood pesticide concentrations, but did not establish any population norms or reference ranges.

Pharmaceuticals

Measurement of therapeutic drugs in hair may be a useful adjunct to more traditional blood and urine analysis techniques (Laloup et al 2005; Kaddoumi et al 2004) but has also been used to test patient treatment compliance with antipsychotic drugs (Shen et al 2002) and in forensic assessments (Maurer 2000).

Nicotine

Recent papers have examined the use of hair as a matrix for environmental exposure to tobacco smoke, especially in children and in service industry workers (Chetiyanukornkul et al 2004; Groner et al 2005; Dimich-Ward et al 2005).

Illicit drugs

Environmental exposure to illicit drugs, either through their manufacture, use or as incidentally-exposed bystanders (especially children) (Wolff et al 1999; De Giorgio et al 2004; Pichini et al 2006) have been explored. These studies have generally not used reference ranges as sample comparators, but have established ‘cut-off’ values to indicate a ‘positive’ or ‘negative’ test outcome.

See also the following papers for applications of hair testing to determine exposure to drugs and problems associated with such testing (Kintz 2007 Bioanalytical procedures for detection of chemical agents in hair in the case of drug-

facilitated crimes, Anal Bioanal Chem 2007 March 6 e-pub and Pragst and Balikova State of the art in hair analysis for detection of drug and alcohol abuse Clin Chim Acta. 2006; 370:17-49).

3. Principles of Use

There has been a significant growth in the number of laboratories offering commercial tests of hair for metals, trace elements and toxins. Seidel et al [i] reported over 225,000 such tests by nine US laboratories in a single year, at an estimated cost of $9.6 million. The US Agency for Toxic Substances and Disease Registry established an expert panel to discuss the methodological and analytical issues surrounding the use for hair analysis (ATSDR 2001[ii]).

Appropriate versus current use of hair analysis

Analytical approaches to assist diagnosis of disease are best used from an a priori basis. That is, medical screening based upon a report of symptoms, leading to a hypothesized condition, which may have a suspected cause. Analytical chemistry may permit samples of tissues to be examined for the presence or concentration of chemicals of interest. The confirmed presence or the measured concentration of a toxic quantity of suspect chemicals would enable practitioners to determine causal relationships and initiate appropriate interventions.

In the case of commercial hair analysis, the main source of clients appears to be individuals with vague symptoms of unwellness, or with concerns about environmental chemical exposure. These then have a broad spectrum of potential contaminants determined in hair samples. These are compared post hoc with in-house norms, reference ranges or standards to deduce ‘toxic’ overexposure to selected contaminants. This ‘expeditionary’ approach is not well based in the scientific method, nor is it toxicologically sound.

Figure 1 demonstrates an example of a hair analysis report. This de-identified report is from a provider that takes referrals directly from the public, and forwards the report to the person or their medical practitioner.

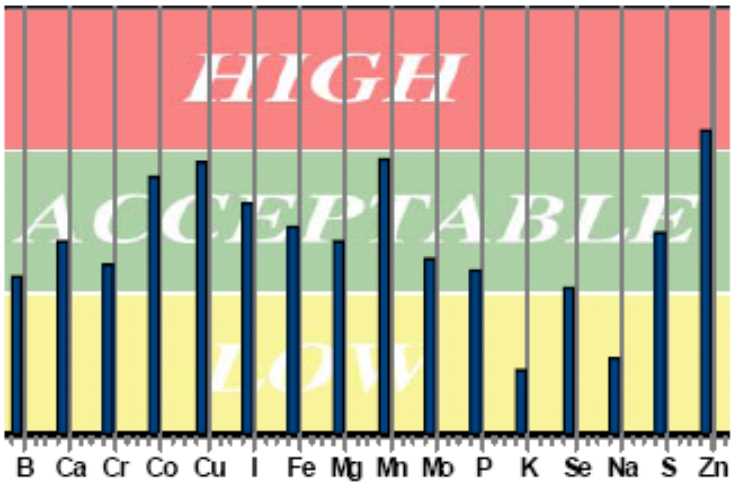
Figure 1 – Example of hair analysis report available directly to the public

First Name: Jane Last Name: Doe Address: City: State:
Post Code: Country: Gender: Female DOB:
Home Phone: Fax: E-mail:

Test:
Type:
Date:

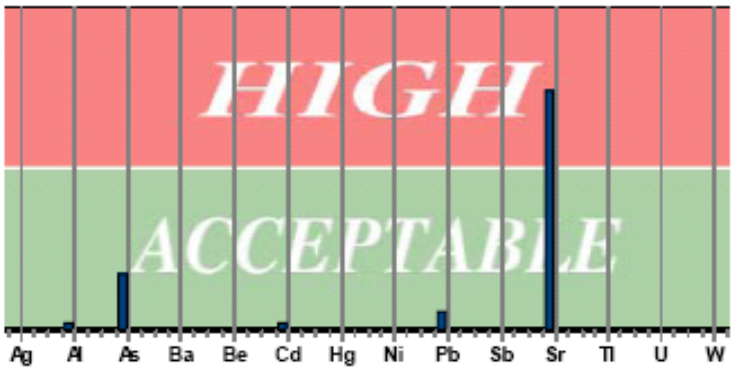
Sample Weight: Sample Location:
Hair Colour: Occupation: Pregnant:
Colour Agent: Shampoo: Conditioner:
Health:
Medication:

Essential Elements



Essential Element	Result	Acceptable Range
Boron (B)	0.842	0.5 - 3.5
Calcium (Ca)	346.719	200 - 600
Chromium (Cr)	0.359	0.23 - 0.9
Cobalt (Co)	0.089	0.02 - 0.105
Copper (Cu)	28.27	9 - 30
Iodine (I)	0.918	0.25 - 1.3
Iron (Fe)	14.941	6 - 25
Magnesium (Mg)	51.629	30 - 90
Manganese (Mn)	0.573	0.18 - 0.6
Molybdenum (Mo)	0.048	0.035 - 0.09
Phosphorus (P)	106.234	80 - 240
Potassium (K)	22.728	50 - 150
Selenium (Se)	0.434	0.4 - 1.2
Sodium (Na)	66.621	125 - 375
Sulphur (S)	38704.7	30000 - 50000
Zinc (Zn)	218.777	100 - 200

Toxic Elements



Element	Result	Acceptable Range
Aluminium (Al)	0.606	< 15
Antimony (Sb)	n.d.	< 0.08
Arsenic (As)	0.027	< 0.08
Barium (Ba)	n.d.	< 1.6
Beryllium (Be)	n.d.	< 0.02
Cadmium (Cd)	0.006	< 0.15
Lead (Pb)	0.113	< 1
Mercury (Hg)	n.d.	< 0.4
Nickel (Ni)	n.d.	< 0.4
Silver (Ag)	n.d.	< 0.13
Strontium (Sr)	9.949	< 5
Thallium (Tl)	n.d.	<0.05
Tungsten (W)	n.d.	<0.5
Uranium (U)	n.d.	< 0.012

This report would leave many people with a high level of concern, without good reason.

Limitations to interpretation

Limitations in interpreting the hair analysis findings include:

- Those based on the interpretation of data collected
- The method of analysis used.

Most commercial hair analysis providers examined by the US review of hair analysis provided no toxicological basis for their analytical conclusions. In particular,

- the establishment of dose-response relationships was missing
- there was no basis to form the view about expected adverse outcomes at 'high' analyte concentrations.

Seidel et al [i] described the results of a round-robin submission of split hair samples, collected from the scalp of a single donor, to six commercial hair analysis laboratories in the USA. They found the following significant problems in their comparison of laboratory analyses:

1. Significant variations, **for some analytes exceeding 10-fold**, were observed in the reported concentrations of metals between different laboratories
2. Five of the laboratories reported at least one element exceeding the reference range, but different laboratories did not report the same element in each case
3. Reference ranges, sample handling and methodological conditions were different amongst laboratories
4. Diagnostic interpretations between laboratories were in poor agreement, in some cases suggesting overexposure even in cases where elements were determined to be within their normal range
5. Two thirds of laboratories recommended proprietary interventions in the form of supplements or diet modification.

The IARC (2001)**Error! Bookmark not defined.** also concluded that;

> *For most substances, insufficient data currently exist that would allow the prediction of a health effect from the concentration of the substance in hair. The presence of a substance in hair may indicate exposure (both internal and external) but does not necessarily indicate the source of exposure."*

Professional advice and assistance

A variety of professional bodies are able to provide expert advice on the use and practical application of analytical methods for detecting and quantitating chemical exposure.

1. **For assistance with the medical aspects of the case,**

   Australasian Faculty of Occupational and Environmental Medicine.
   http://afom.racp.edu.au/

   AFOEM members can provide specific expertise in medical aspects of occupational and environmental chemical exposure

2. **For assistance with the technical aspects of the case, such as use of accredited laboratories, standards of testing.**

   Australian Institute of Occupational Hygienists
   http://www.aioh.org.au/

   Chemical exposures in occupational and environmental settings may be determined by occupational hygienists and environmental health professionals respectively. These groups have expertise in evaluating sources of chemical contaminants and may be able to provide technical methods for measuring chemicals in specific environments.

   Australasian College of Toxicology and Risk Assessment. (ACTRA) No website available

   Toxicologists (who may be registered or certified as professional practitioners, for example in Europe and the UK, USA and elsewhere) can provide specific advice on the chemical, physiological and toxicological basis for determining whether an adverse effect may be associated with particular chemical exposures. Toxicological risk assessment is a specialized branch of toxicology practice, and the Australian College of Toxicology and Risk Assessment may provide technical support in this area.

3. **For interpretation of results**

All of the above groups can assist with interpretation of the results. Making sense of the results is a difficult area, with a number of laboratories reporting significantly different reference ranges.

Government departments, such as:

- Australian Government Department Health and Aging
- The state workplace based safety authorities may also be able to assist.

4. Analytical Issues

Hair may be useful to establish pre-exposure to substances including heavy metals from occupational settings many weeks to months prior to collection.

Entry of substance into hair

Substance entry into hair is complicated and involves a number of processes. Incorporation by entrapment from the blood bathing the growing follicle is a major mechanism, although incorporation through direct contact of mature hair with sweat and/or sebaceous secretions may also be a significant source of entry.

Because of the ability of hair to directly absorb foreign substances, contamination of hair by direct environmental exposure should also be reasonably excluded, if hair results are to be used.

- There is a wide range of uptake into hair even for the same substance and for the same exposure. Large variations occur between substances.

Retention of the substance into hair

There are a number of factors that affect retention of substances into hair. These include

- Hair colour
- Hair structure
- The chemical properties of foreign substance
- Bleaching and excessive use of shampoo and conditioners can reduce the concentration of substance in hair

Technical issues to be addressed

External contamination can occur, making the interpretation of hair data difficult. Laboratories can reduce this artefact by incorporating washing techniques.

Metal concentrations by themselves in hair will rarely provide sufficient proof of excessive exposure unless compared to and correlated with the history and the medical details of any exposure. Segmental analysis of 1-2 cm sections can provide some picture of changing exposure over a longer period of time. This may assist in determining if exposure has occurred and a possible time frame of exposure. Whilst the majority of hair is in the growing phase, a substantial portion of hair may no longer be growing. Consequently, segmental analysis rarely provides a sharp history of exposure if more than one hair is taken for analysis.

Collection of hair should follow standard procedures to avoid contamination of hair by the collector and to ensure collection from the nape of the head which shows the most consistent growth of hair. Laboratories conducting testing will usually have collection kits and protocols available.

Analysis methods vary widely but generally involve spectroscopic methods. These are very specific techniques and can detect several elements in one test. However, given the large number of elements targeted search for presumed exposed substances is warranted rather than examining hair for any substance. For some metals, e.g. arsenic and mercury organic forms derived from food are fat less toxic than inorganic forms. Consequently, the hair test will need to differentiate the chemical form of substance.

5. Recommendations

Because this area is complex, and has the potential to be misunderstood with significant negative consequences if not dealt with appropriately, it is strongly recommended that expert advice and input is sought.

There are early reports that other testing methods, such as toenail analysis, may be useful in some circumstances. The Seidal[i] review indicates confusion with setting of normal ranges, and the interpretation of results needs to be undertaken with caution.

We recommend advice from accredited providers, accredited medical practitioners, and analytical services accredited by National Association of Testing Authorities (NATA). http://www.nata.asn.au/

6. Case Studies

Case Study 1 – productive use of hair analysis

A young woman was attending a nightclub with friends. Some alcohol was consumed. Hours later the woman woke up and found herself on the floor in a room adjacent to the club. She felt dizzy and had difficulty walking. Her clothing was in disarray suggesting a possible sexual assault. Several hours later she reported the event to the police which resulted in an examination by a forensic medical officer.

Subsequent tests revealed a likely sexual assault with suspicion of surreptitious administration of a drug, possibly from a drink given to her by a man at the club. Analysis of her urine confirmed the presence of the benzodiazepine temazepam. Hair analysis was activated to not only confirm the presence of this benzodiazepine in another tissue but also establish the possible time frame of exposure and eliminate personal use of this drug.

Hair was taken 2 weeks later to ensure externalization of the embryonic hair exposed to drug and her 10-cm long hair was cut into 1-cm segments and analysed for benzodiazepines. Temazepam was only found in the first segment of hair (closest to scalp).

These results supported the claim of surreptitious administration of temazepam at about the time of the assault and show no prior recent use of the drug. While these results do not of themselves prove a drug facilitates sexual assault hair analysis they provide useful information to support a prosecution.

Case Study 2 - productive use of hair analysis

Hair analysis can provide a history of drug exposure by the analysis of segments of hair. For example, in a case of a drug –dependent person seeking treatment it is hoped that the treatment (pharmacotherapy, counselling etc) at least reduces (illicit) drug use.

In the case of 30-year male who had been drug-dependent (heroin, amphetamines and benzodiazepines) for 12 years the entry into a drug treatment program provides an opportunity to monitor drug use and hopefully to successfully treat his addiction. In addition to clinical and psychological signs of improvement the use of a hair test at the 6-month point can provide an objective measure of declining drug use.

In this case, segmental analysis of his 8-cm long hair will provide a history of about 8-months of drug use (hair grows at about 1-cm per month). Results from the laboratory showed that the concentrations of the heroin metabolites (6-acetylmorphine & morphine) and methamphetamine were steadily decreasing from the time before treatment to the most recent growth and indeed at this point the concentrations of drugs were quite small suggesting little use of these drugs. The benzodiazepine (alprazolam) was present at similar concentrations throughout treatment, but this was expected since this drug was being prescribed to treat his anxiety and insomnia. A further hair test at 12-months would be useful to further monitor his drug use.

Case study 3 – counterproductive use of hair analysis

A 58 year old woman complaining of headaches and neurological disturbances had sent a hair sample to a commercial laboratory to determine if she had been exposed to environmental chemicals.

The analysis confirmed very high hair concentrations of mercury, and indicated that she should begin chelation therapy to be detoxified. She was also advised to cease consumption of potential mercury sources in diet, especially fish (tuna and shark).

Following on from this advice the individual approached the local health department to help her identify the source of her exposure. During the course of the discussion, she revealed that “some years ago” she had been provided with a lotion to treat a scalp condition. She had discovered a bottle of lotion in her cabinet in the previous months and recommenced treatment. The lotion was a mixture of salicylic acid and mercurous chloride.

Although her symptoms were consistent with mercury exposure, the assessment of hair in this case yielded a very high mercury concentration as a result of the direct application of lotion to the hair. Hence the analysis revealed mercury deposited during the hair growth as well as from direct surface application, overestimating the extent of her exposure. Without a detailed history, the analytical report also gave poor advice regarding the source of her exposure.

Case study 4 – unproductive use of hair analysis

A 49 year old woman complaining of non-specific symptoms (headache, malaise) sought specific advice from her local GP suspecting some form of environmental chemical exposure may have been responsible.

Although no specific agent was identified, she sent a hair sample to a commercial laboratory. Results were reported to her that she had elevated mercury and needed to detoxify and exclude mercury-contaminated foods, especially fish. The subject suffered from food intolerance and could not consume meat, but could only eat fish protein. She was concerned that she would not be able to include fish in her diet in future.

In spite of blood analysis and urinalysis for mercury yielding unremarkable results, and extensive analysis of fish fillets (including tuna) from her suppliers demonstrating no significant mercury contamination, her symptoms have persisted. The cause is still unknown.

7. References

[i] Seidel S et al, Assessment of commercial laboratories performing hair mineral analysis. JAMA 2001; 285:67-72.

[ii] ATSDR 2001. Hair analysis panel discussion: exploring the state of the science. Atlanta. GA: Agency for Toxic Substances and Disease Registry. Available from http://www.atsdr.cdc.gov/hac/hair_analysis/. Accessed 20 March 2007.

Drasch G, Roider G, Assessment of hair mineral analysis commercially offered in Germany. J Trace Elem Med Biol. 2002; 16(1):27-31

Barrett S, Commercial hair analysis. Science or scam? JAMA. 1985; 254:1041-5.

8. Website details for relevant Agencies

Australasian Faculty of Occupational and Environmental Medicine

http://afom.racp.edu.au/

National Association of Testing Authorities
http://www.nata.asn.au

Forensic & Medical Sexual Assault Clinicians Australia Inc
http://www.famsacaustralia.org.au/

Department of Health and Aging
http://www.health.gov.au/

The International Association of Forensic Toxicologists (TIAFT)
http://www.tiaft.org

Society for Hair Testing (SOHT)
http://www.soht.org