

Ear Wax: A New Biological Monitoring Medium for Metals?

U. Krishnan¹ and S. S. Que Hee²

¹Department of Environmental Health, University of Cincinnati Medical Center, 3223 Eden Avenue, Cincinnati, Ohio 45267-0056, USA and ²Department of Environmental Health Sciences, UCLA School of Public Health, 10833 Le Conte Avenue, Los Angeles, California 90024-1772, USA

Well known media for biological monitoring include urine, blood, breath, nails, hair, teeth, saliva, feces, sweat, tears, milk and sebum (Waritz 1979). Ideally sampling should be non-invasive to humans: that is, urine, breath, nails, hair, saliva, feces, sweat, tears, milk and sebum are favored for biological monitoring. Of these only urine and breath have been adjudged adequate for quantitative dose-effect relationships between the concentration of the biological marker and the exposure amount as shown by their use for the Biological Exposure Indices (ACGIH 1990). A medium not much investigated for xenobiotics exposure is ear wax or cerumen. The pesticides lindane and DDT have been detected in ear wax (Wang et al. 1988).

Ear wax consists of the secretions of the sebaceous and ceruminous glands as well as shed skin cells (Ballantyne 1965). Histologically and functionally, the ceruminous glands are identical to the apocrine sweat glands. Since xenobiotics have been detected in sweat (Waritz 1979), they should be also present in ear wax. Metals in ear wax will probably be present at lower concentrations than in sweat because of dilution with the sebaceous gland secretions. Lipid-soluble compounds may be at higher concentrations than found in sweat.

We report the first finding of metals in ear wax.

MATERIALS AND METHODS.

Ear wax from two volunteers (one Eurasian male in his middle forties and one female from the Indian subcontinent in her thirties) were separately collected from

Send reprint requests to SS Que Hee, Department Environmental Health Sciences, UCLA School of Public Health, 10833 Le Conte Avenue, Los Angeles CA 90024-1772

the ear by a clean Q tip and transferred by a Teflon-coated spatula to separate preweighed 25 mL Teflon containers (previously acid-washed in 10% nitric acid) comprising the digestion cup of a Parr 4745 digestion bomb (Fisher Scientific 01-023-2). The samples were stored in a vacuum desiccator until constant daily weight (10.5 and 43.7 mg respectively). Dry weights of 10.5 and 17.9 mg for the two individuals were analysed respectively. A volume of 2.75 mL of concentrated nitric acid (Ultrex distilled from Vycor; GF Smith) was added to each sample and to each of 2 empty Teflon containers used as controls. The containers were heated on a hot plate to 50 C for 15 min with Teflon covers on, and then allowed to stand overnight for 16 hours. Each Teflon cup was placed in its Parr Bomb which was then heated in an oven at 140 C for 2 hr, cooled, and 0.1 mL of concentrated perchloric acid (Ultrex distilled from Vycor; GF Smith) added. The solutions were evaporated at 80 C to about 0.050 mL. A 2-mL aliquot of 11.6% hydrochloric acid/2.8% nitric acid (v/v) was added. The hydrochloric acid originated from Fisher Scientific A-144C. Each container was then heated for 5 min at 90 C with Teflon cover on, and each solution transferred using an acid-washed Pasteur pipet to a calibrated measuring cylinder. This washing procedure was repeated two more times and the volumes of the combined transferred solutions were adjusted to 10 mL with 11.6% hydrochloric acid/2.8% nitric acid. This procedure has been shown by Que Hee and Boyle (1988) to be the reference digestion method for organic biological specimens like blood and organs. The samples were then analysed for 38 elements by inductively coupled plasma atomic emission spectroscopy (Que Hee and Boyle 1988) using a vacuum simultaneous Jarrell-Ash/Allied Chemical ICAP-9000 equipped with 40 fixed channels, variable wavelength N+1 scanner, spectrum shifter, peristaltic pump, and argon purification train.

The results in Table 1 were expressed on a μg element/g dry weight basis after correction for elemental content of controls and accounting for digest total volume.

RESULTS AND DISCUSSION.

Table 1 shows that the elements analysed in the ear wax of two people represented between 1.6-4.8% of the total dry weight of ear wax. The major elements in mg/g were: K, 5.7-19; S, 3.8-13; Na, 3.6-8.8; Ca, 0.69-2.2; and Mg, 0.54-1.1. The most surprising result was the moderately high concentration of As (0.15-0.22 mg/g). The non-detection of Ag, Be, Co, Hg, Mn, Ni, Se, and V may be suggestive of a potential usefulness of ear wax as a biological monitoring medium for these toxic elements in people exposed to high concentrations in the

Table 1. Concentrations of some elements in the ear wax of two individuals

Element	Concentration in $\mu\text{g/g}$ dry weight	
	Person 1	Person 2
Al	49	83
As	150	220
Au	7.2	6.0
Ba	nd	61
Ca	2200	690
Cd	1.6	0.67
Cr	99	110
Cu	4.8	23
Fe	150	170
I	2400	nd
In	65	40
K	19000	5700
Li	0.50	0.29
Mg	1100	540
Mo	1.7	nd
Na	8800	3600
Pb	13	14
Pt	100	82
S	13000	3800
Sb	13	72
Si	36	nd
Sn	400	350
Sr	17	6.4
Ti	6.7	4.0
Tl	100	60
Zn	88	103

nd, not detected; relative errors are <10%.

Table 1 Note 1: Ag, B, Be, Co, Hg, Mn, Ni, P, Se, and V were not detected.

Table 1 Note 2: The lowest quantifiable levels were in $\mu\text{g/mL}$: 1-5, C; 0.1-1, I, In, Sn; 0.05-0.1, As, B, K, Tl; 0.01-0.05, Ag, Hg, Na, P, Pb, S, Se, Zn; 0.005-0.01, Au, Ba, Cr, Fe, Ni, Sb, Si; 0.001-0.005, Al, Ca, Cd, Co, Cu, Mg, Mn, Mo, Pt; <0.001, Be, Li, Sr, Ti, V.

Table 1 Note 3: The dry weights for the ear wax samples analysed for Persons 1 (Indian subcontinent female) and 2 (Eurasian male) were 17.9 and 10.5 mg, respectively. The elements listed above comprised 4.8 and 1.9 % of the respective ear waxes.

environment or in the workplace since no baseline correction is required unlike for the other elements. Non-detection of the latter, however, could be caused by analytical artifacts even though volatilization should not occur since a Parr Bomb is closed. Nevertheless to

check this point, the remaining 25.8 mg of the sample for Person 1 was digested in a Microwave Parr Bomb by the microwave digestion procedure of Que Hee and Boyle (1988). None of these elements were detected also after this treatment. The elements showing less than three-fold variation in the ear wax samples from the two volunteers were: Al, As, Cd, Cr, Fe, In, Li, Mg, Na, Pb, Pt, Sn, Sr, Ti, Tl and Zn. Less than twofold variation was shown for Al, As, Au, Cr, Fe, In, Li, Mg, Pb, Pt, Sn, Ti, Tl, and Zn. The concentrations of the latter group of elements may reflect homeostatic control. In contrast, the following elements were present in concentrations differing more than threefold from one another: Ba, Ca, Cu, I, K, Mo, S, Sb, and Si. More earwaxes from different people need to be analysed to determine reference ranges.

The ultimate origin of the ceruminous secretion is blood plasma. Thus, it is interesting to compare in Table 2 the elemental concentrations of human blood plasma, sweat and skin (Iyengar et al. 1978), sweat and skin being the known proximal contributors to ear wax (Ballantyne 1965). None of the three media explain the ear wax K/S ratio (1.49-1.50). The K/Ca ratio varies between 8.3-8.6 for the two ear wax samples consistent with the ratios expected for sweat and skin. Sweat is the source explaining the ratios of K/Mg (11-17), Ca/Zn (6.7-25) and Mg/Zn (5.2-13). Skin is the origin consistent with ear wax Na/S ratios of 0.68-0.95 and K/Na ratios of 1.6-2.2. The high concentration of K relative to Na might be expected if the apocrine sweat glands discharge their intracellular contents. There are very few excreted fluids with K/Na ratios >1 , one being human milk (Iyengar et al. 1978). The source of As is unknown. Arsenic compounds may contribute to the bacteriostatic properties of cerumen which hitherto have been attributed to its acid pH and enzyme activity (Ballantyne 1965). The fact that lead and cadmium were detectable in both samples signifies possible use of ear wax to assess their external exposure, baseline sampling before exposure being necessary to interpret results.

Clearly more measurements need to be performed to define representative reference ranges, and to assess the effects of gender, ethnicity, food habits, race, life style, and the other factors known to influence the composition of biological monitoring media (Waritz 1979). The fat composition of ear wax is affected by gender and season (Cipriani et al. 1990). The sampling time required to obtain samples with detectable concentrations must also be defined. A technique for measuring the rate of cerumen production is available (Cipriani et al. 1988).

Table 2. Ranges of some elements in human blood plasma, sweat and skin of people unexposed occupationally to these elements (Iyengar et al. 1978)

Element	Concentration in		
	Plasma mg/L	Sweat µg/g	Skin µg/g
Al	0.28-0.41		5.1-1000
As	<0.01(2)		0.06-0.23
Ba	0.050-0.059		14-17 (2)
Be	<0.004(1)		0.02(1)
Ca	89-106	4-206	0.034-20
Cd	<0.003(1)		0.096-36
Co	0.0007-0.012		0.05-300
Cr	0.026-0.164		0.44-41
Cu	0.61-1.41	0.058-1.48	6.9-143
Fe	0.71-1.27	0.46-1.5	27-1900
Hg	0.002-0.011		0.003-3.34
I	0.058-0.085	0.01(1)	0.35(1)
K	137-208	176-350	0.846-118
Li	0.03(1)		0.29(1)
Mg	13.0-27.4	0.002-217	0.009-0.87
Mn	0.00059-0.068	0.06(2)	0.003-1.08
Mo	0.013(1)		0.06-1.7
Na	2990-3330	1017-3370	69-4968
Ni	0.010-0.066	0.052-0.163	0.05(1)
P	112-130(2)	0.24-1.55(2)	0.30-40
Pb	0.012-0.18	0.07-2.74	0.1-796
S	330(1)		1380-1850
Sb	0.0032-0.054(2)		0.03-0.22
Se	<0.03-0.59		0.061-1.2
Si	0.43(1)		128-1390
Sn	<0.004-0.105(3)		21(1)
Sr	0.028-0.044		0.023-13
Ti	<0.04-0.11		1.91-72(2)
V	<0.01-0.56(2)		<1(2)
Zn	0.79-1.7	0.50-1.58	15.6-1000

No value indicates not reported

Reference range and reported ranges are indicated

The bracketed numbers indicate 1 or 2 studies only

This is the first report of the use of ear wax as a biological monitoring medium for metals. Some advantages include non-invasive sampling, and the fact that the ear canal is much more protected from the external environment than the skin so that the concentrations of xenobiotics may reflect absorbed dose better than skin sampling of sweat and sebum, and than nails. Sampling could be performed simply in the workplace by the insertion of ear plugs, especially if workers are already wearing them in any hearing conservation program. It

would be interesting to assess whether hircismus, a condition in which a person sweats excessively in the armpits, will affect the metal content since it is known that this changes the wax composition of ear wax (Inaba et al. 1987). Ear wax like nails, hair and teeth, may indicate chronic exposure since the last three media facilitate cumulative deposition of xenobiotics. Bioaccumulatory xenobiotics like lindane and DDT have been detected in cerumen (Wang et al. 1988). The present study shows metals are also detectable.

Acknowledgments. Funding for this work was from the consulting funds of Dr. Que Hee.

REFERENCES.

- ACGIH (1990) Documentation of the biological exposure indices. American Conference of Governmental Industrial Hygienists, Cincinnati, Ohio
- Ballantyne J (1965) Anatomy of the ear. In: Diseases of the ear, nose and throat, Volume 2, Scott-Brown WG, Ballantyne J, Groves J (eds), Butterworths, London, pp 231-282
- Cipriani C, Taborelli C, Cardo PP, Rabora A (1988) A technique for measuring the rate of cerumen production. *Laryngoscope* 98:204-205
- Cipriani C, Taborelli C, Caddia C, Malagrana A, Rabora A (1990) Production rate and composition of cerumen: influence of sex and season. *Laryngoscope* 100:275-278
- Inaba M, Chung TH, Kim JC, Choi YC, Kim JH (1987) Lipid composition of ear wax in hircismus. *Yonsei Med J* 28:49-51
- Iyengar GV, Kollmer WE, Bowen HJM (1978) The elemental composition of human tissues and body fluids. Verlag Chemie, New York
- Que Hee SS, Boyle JR (1988) Simultaneous multielemental analysis of some environmental and biological samples by inductively coupled plasma atomic spectrometry. *Anal Chem* 60:1033-1042
- Wang XQ, Gao PY, Lin YZ, Chen CM (1988) Studies on hexachlorocyclohexane and DDT contents in human cerumen and their relationships to cancer mortality. *Biomed Environ Sci* 1:130-151
- Waritz RS (1979) Biological indicators of chemical dosage and burden. In: Patty's Industrial Hygiene and Toxicology, vol III, Cralley LJ and Cralley LV (eds), John Wiley and Sons, New York, pp 257-318

Received July 29, 1991; accepted November 30, 1991.