

Biological monitoring of nickel in humans

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SUNDERMAN FW Jr. Biological monitoring of nickel in humans. *Scand J Work Environ Health* 1993;19:34—8. The literature since 1985 on the biological monitoring of occupational, environmental, or iatrogenic exposures of humans to nickel is surveyed from the author's perspective. Urine and serum are the body fluids commonly analyzed for nickel. Nickel concentrations in urine specimens from unexposed persons usually exceed the current analytical detection limits, while nickel concentrations in their serum specimens are close to the detection limits. For this reason, and because urine collection is painless, noninvasive, and convenient, urine is more practical than serum for the biological monitoring of nickel-exposed workers. Electrothermal atomic absorption spectrometry with Zeeman background correction is currently the method of choice for the analysis of nickel in biological materials. Procedures for quality assurance and strategies for biological monitoring are summarized, as performed in the author's laboratory, and current reference values are listed for nickel concentrations in urine and serum specimens from healthy unexposed persons.

Key terms: biological monitoring, reference values, serum nickel, urinary nickel.

In a report to the Steering Committee for Nickel Research (Sir Richard Doll, Chairman), my co-workers and I (1) reviewed the scientific literature to 1985 on nickel measurements in body fluids, excreta, and tissues from humans with occupational, environmental, or iatrogenic exposures to nickel compounds and critically evaluated the reported correlations between levels of human exposures to various classes of nickel compounds via inhalation, oral, or parenteral routes and the corresponding concentrations of nickel in biological samples. The conclusions of this report included four major points, as follows.

First, measurements of nickel concentrations in body fluids, especially urine and serum, provide meaningful insights into the extent of nickel exposures, provided these data are interpreted with knowledge of (i) the exposure routes, sources, and durations, (ii) the chemical identities and physical-chemical properties of the nickel compounds, and (iii) relevant clinical and physiological information, such as renal function.

Second, nickel concentrations in body fluids should not be viewed as indicators of specific health risks, except in persons exposed to nickel carbonyl, for whom urine nickel concentrations provide prognostic guidance on the severity of the poisoning.

Third, for persons exposed to soluble nickel compounds (eg, nickel chloride, nickel sulfate), nickel concentrations in body fluids are generally proportional to exposure levels; the absence of increased values usually indicates nonsignificant exposure; and the presence of increased values should be a signal to reduce the exposure.

Fourth, for persons exposed to less soluble nickel compounds (eg, nickel subsulfide, nickel oxide), increased concentrations of nickel in body fluids are indicative of significant nickel absorption and should be a signal to reduce the exposures to the lowest levels attainable with available technology; the absence of increased values does not necessarily indicate freedom from the health risks (eg, cancers of the lung and nasal cavities) associated with exposures to certain relatively insoluble nickel compounds.

These four major conclusions have been corroborated and strengthened by the numerous investigations of nickel metabolism and toxicology that have been reported during the past eight years. The important advances during this period have included (i) a mathematical model for the kinetics of nickel absorption, distribution, and elimination in humans (2), (ii) a reference method for nickel measurements in biological materials, the method being based on stable isotope dilution and gas chromatography-mass spectrometry of volatile nickel chelates (3), (iii) lower detection limits and higher precision of nickel determinations in body fluids, attributable to improved analytical instruments and methods, as well as reduced nickel contamination during specimen collection, sampling, processing, and analysis (4—6), and (iv) reliable reference materials for the quality control of nickel determinations in urine and serum (7—10).

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Absorption, distribution and elimination of nickel in humans

Humans absorb 15–50% of the nickel that is ingested in drinking water after an overnight fast, compared to <15% of the nickel that is ingested in foods (2). Approximately 30% of inhaled nickel reaches the lungs and 20% of inhaled nickel is absorbed from the respiratory tract into blood circulation. The remainder is mostly trapped by mucus, mobilized by ciliary activity, and expectorated or swallowed. A small proportion of inhaled nickel accumulates in lung tissue and regional lymph nodes (11). Cutaneous absorption of nickel is quantitatively minor, but it may be important in the pathogenesis of contact dermatitis caused by nickel hypersensitivity (12). In adult humans, the body burden of nickel averages $\sim 0.5 \text{ mg} \cdot 70 \text{ kg}^{-1}$ (ie, $\sim 7.3 \mu\text{g} \cdot \text{kg body weight}^{-1}$) (13). Nickel levels are highest in lung, thyroid, adrenal, and bone (14). In human serum, nickel is bound to albumin, alpha-2 macroglobulin, and ultrafiltrable ligands (eg, histidine) (15, 16). In tissue cytosol, nickel is bound to several proteins and peptides (16, 17). Urinary excretion is the major route for the elimination of absorbed nickel (2). Fecal excretion primarily reflects the nickel that is unabsorbed from the diet and passes through the gut (2, 18). Small amounts of absorbed nickel are excreted in bile, sweat, hair, saliva, and mother's milk (19, 20).

Media for the biological monitoring of nickel exposures

Urine is generally the most practical specimen for the biological monitoring of nickel exposure, since (i) a metal-free plastic container is the only collection equipment that is needed, (ii) the collection procedure is painless and noninvasive, and (iii) nickel concentrations in urine specimens from healthy, unexposed persons are above the current detection limits of electrothermal atomic absorption spectrometry (EAAS) with Zeeman background correction. The disadvantages of urine as a specimen for the biological monitoring of nickel exposures include high matrix variability, fluctuating specific gravity, difficulty of obtaining timed urine collections, and contamination from nickel-containing dust on clothing (1, 19–22).

For the biological monitoring of nickel exposure, serum has the advantages of controlled sample collection, small matrix variation, and little diurnal fluctuation (1, 19–22). In patients with renal insufficiency who are treated by extracorporeal hemodialysis, urine is usually unavailable, so serum is the specimen of choice (5, 23). Serum has the disadvantage of requiring venepuncture with a plastic canula to avoid nickel contamination from stainless steel needles. The reference values for serum nickel concentrations are barely above the detection limit of current analytical methods. Plasma can be used in lieu

of serum, but tests are needed to verify that the heparin is free of nickel contamination or that the ethylenediaminetetraacetic acid does not cause analytical interference. Analyses of nickel concentrations in whole blood have no practical advantages for biological monitoring and pose problems of matrix interference (eg, from iron in hemoglobin). Nickel concentrations in the whole blood of persons without occupational exposures are often below the analytical detection limit (1, 19–22).

Feces is the most reliable specimen for the biological monitoring of dietary nickel intake (1, 2, 18, 24). The disadvantages of fecal nickel analyses include the problems of collecting the feces for 3–5 d, pooling and homogenizing the specimens, digesting the samples with concentrated acids (eg, a mixture of nitric acid, sulfuric acid, and perchloric acid), and extracting the nickel as a chelate into an organic solvent prior to analysis by atomic absorption spectrometry. Nickel concentrations in saliva, hair, nails, sweat, milk, and blood lymphocytes have been analyzed in clinical investigations, but these specimens are seldom advantageous or practical for the biological monitoring of occupational, environmental, or iatrogenic exposures to nickel compounds (1, 19–22, 25).

Analytical methods for nickel in biological fluids

EAAS with Zeeman background correction is currently the most common technique for determining nickel in biological materials (5, 26–33). Minimal sample processing is required, and automated, computer-controlled instruments are available from several manufacturers. In my laboratory, the nickel detection limit by EAAS is $0.4 \mu\text{g} \cdot \text{l}^{-1}$ for urine and $0.05 \mu\text{g} \cdot \text{l}^{-1}$ for serum (26, 27). Lower detection limits for nickel ($\sim 1 \text{ ng} \cdot \text{l}^{-1}$ for all body fluids) can reportedly be achieved by differential pulse absorption spectrometry (DPAV) with a dimethylglyoxime-sensitized mercury electrode (34), but the procedure is time-consuming, requires sample digestion, and is susceptible to nickel contamination. The usual techniques of neutron activation analysis (NAA) are insufficiently sensitive for measurements of nickel in body fluids, but nickel detection limits of $\sim 10 \mu\text{g} \cdot \text{kg}^{-1}$ in solid samples have been obtained by a fast-neutron induced reaction (35, 36). Inductively coupled plasma-mass spectrometry (ICP-MS) gives a detection limit of $\sim 1 \mu\text{g} \cdot \text{l}^{-1}$ for nickel in urine; the use of ICP-MS for the analysis of nickel in body fluids is hampered by matrix interferences from calcium, sodium, and potassium (37). Isotope dilution gas chromatography-mass spectrometry is a reference procedure that can be used to validate nickel determinations by more convenient techniques (3, 38). The approach is expensive, difficult, and laborious. Sample cross-contamination is a problem that

must be circumvented, and the detection limit for nickel in urine is $\sim 1 \mu\text{g} \cdot \text{l}^{-1}$.

laboratory programs for harmonization and quality control of nickel determinations.

Quality assurance of nickel analyses

To obtain reliable measurements of nickel concentrations in urine or serum for the biological monitoring of occupational, environmental, or iatrogenic exposures to nickel compounds, the following ingredients are necessary (2, 21, 22). First, a clean laboratory room is needed that is solely used for trace-metal analyses, designed with precautions against metal contamination. The room should be equipped with a laminar-flow work station, a high-purity water system, and a dedicated centrifuge with hermetically sealed trunion cups. Second, the instrumentation for nickel analysis (eg, EAAS, DPAV, ICP-MS) must be in perfect adjustment and tested daily to ensure that the nickel detection limit is maintained. Third, the analyst should be skilled in trace-metal determinations and have ample time, patience, and motivation to achieve accuracy, to control contamination, and to perform the requisite preventive maintenance. Fourth, the support staff should be expert in acid washing the plasticware, collecting, transporting and processing the specimens, keeping the records, and reporting the analytical results. Fifth, the laboratory should be supervised by a clinical pathologist, clinical chemist, or toxicologist, who is knowledgeable about the analytical and clinical pitfalls of nickel determinations. Finally, the laboratory should perform the nickel analyses in duplicate or triplicate, include reference materials in every analytical run, regularly monitor the within-run and run-to-run precision, frequently determine the recovery of nickel added to serum and urine, and participate in inter-

Strategies for the biological monitoring of nickel exposures

The timing of specimen collection is critically important for the biological monitoring of persons exposed to soluble nickel compounds (eg, nickel chloride, nickel sulfate), since bivalent nickel has a short elimination half-time (1, 39, 40). The timing is not so important for the biological monitoring of persons exposed to inhaled particles of nickel-containing alloys or parenteral release of nickel from orthopedic prostheses (1, 41–43). In my laboratory, specimens of urine or serum are usually obtained from industrial workers on Wednesday or Thursday afternoons, at the end of the workshift, after the subjects have taken showers and changed into street clothes. Body fluids are collected in an uncontaminated dispensary by nurses or medical technologists, using plastic utensils and containers that have been checked for nickel contamination. Blood is withdrawn through plastic intravenous canulas, instead of through conventional needles. The blood tubes are sealed with plastic caps instead of rubber stoppers. Specimens are obtained from unexposed workers (eg, nurses, administrative staff) during each collection session so that nickel contamination can be monitored. The specimen containers are encased in polyethylene bags and transported to the trace-metal laboratory in a polystyrene ice chest. Immediately upon arrival in the laboratory, the blood specimens are centrifuged in sealed trunion cups, and the serum is removed with acid-washed Pasteur pipets. Aliquots of each urine specimen are removed for measurements of specific

Table 1. Reference values for nickel concentrations in serum and urine specimens from healthy persons without occupational exposure to nickel compounds. (NS = not stated)

Authors	Country	Number of specimens	Mean	SD	Range	Units
<i>Serum</i>						
Leach et al 1985 (44)	United States	30	0.28	0.24	<0.05–1.1	$\mu\text{g} \cdot \text{l}^{-1}$
Wills et al 1985 (25)	United States	18	0.44	0.18	NS	$\mu\text{g} \cdot \text{l}^{-1}$
Andersen et al 1986 (45)	Denmark	6	0.65	0.35 ^a	0.3–1.3	$\mu\text{g} \cdot \text{l}^{-1}$
Sunderman et al 1989 (42)	United States	42	0.2	0.1	<0.05–1.0	$\mu\text{g} \cdot \text{l}^{-1}$
Nixon et al 1989 (5)	United States	38	0.14	0.09	NS	$\mu\text{g} \cdot \text{l}^{-1}$
<i>Urine</i>						
Sunderman et al 1986 (27)	United States	34	2.0	1.5	0.5–6.0	$\mu\text{g} \cdot \text{l}^{-1}$
Kiilunen et al 1987 (4)	Finland	299	4.1 ^b	-	10.0 ^c	$\mu\text{g} \cdot \text{l}^{-1}$
Ghezzi et al 1989 (46)	Italy	97	1.6	2.1	NS	$\mu\text{g} \cdot \text{l}^{-1}$
Lin 1990 (33)	Taiwan	30	3.2	1.7	1.2–7.8	$\mu\text{g} \cdot \text{l}^{-1}$
Ong et al 1990 (32)	Singapore	20	3.1	2.3	0.9–8.3	$\mu\text{g} \cdot \text{l}^{-1}$
Angerer & Lehnert 1990 (47)	Germany	123	0.9	1.4	<0.1–13.3	$\mu\text{g} \cdot \text{l}^{-1}$
Ulrich et al 1991 (48)	Czechoslovakia	31	1.7	1.0	NS	$\mu\text{g} \cdot \text{l}^{-1}$

^a Plasma.

^b Geometric mean.

^c 95th percentile.

gravity and creatinine so that urine nickel concentrations can be normalized to a specific gravity of 1.024, or expressed as micrograms of nickel per gram of creatinine (27). Analyses performed on dilute urine specimens (specific gravity <1.010, or creatinine concentration <0.3 g · l⁻¹) are, whenever possible, repeated on a more concentrated specimen. If the subjects are hospital patients, timed urine collections are obtained for nickel analyses, instead of random urine samples; unfortunately, it is seldom feasible to obtain reliably timed urine collections from industrial workers.

Reference values for nickel in urine and serum

Current reference values for nickel concentrations in urine and serum specimens from healthy adult humans without occupational exposures to nickel compounds are listed in table 1. These values are substantially lower than those reported prior to 1980 and therefore reflect the improved detection limits that are achieved with recent EAAS instruments and the greater precautions that are now employed against nickel contamination. The current reference values for urine nickel are probably reliable, but those for serum nickel may still be higher than the true concentrations because of limited analytical sensitivity and persistent problems with nickel contamination during venepuncture and serum separation.

Acknowledgments

This work was supported by a grant from Northeast Utilities, Inc.

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