COLLECTIVE EXPERT APPRAISAL: SUMMARY AND CONCLUSIONS

Regarding the expert appraisal on setting occupational exposure limits for chemical agents

Evaluation of biomarkers of exposure and recommendations for biological limit values and biological reference values for hexavalent chromium and its compounds

This document summarises and presents the work of the Expert Committee on expert appraisal for recommending occupational exposure limits for chemical agents (OEL Committee) and the Working Group on biomarkers of exposure.

Presentation of the issue

AFSSET, which became ANSES in July 2010, received a formal request on 12 June 2007 from the French Directorate General for Labour to conduct the scientific expert appraisal work required for setting occupational exposure limit values (OELs) for chromium(VI) and its compounds.

France had a mean eight-hour exposure value for chromium(VI) and its compounds of 0.05 $mg.m^{-3}$. This value was set in the Circular of 13 May 1987[1] of the Ministry of Labour (not published in the OJ).

The Directorate General for Labour asked the Agency to reassess this value and, if necessary, to propose new occupational exposure limit values based on health considerations.

Examination of this request was entrusted to the ANSES OEL Committee, which issued a report in September 2009, indicating, in particular, that:

- individual excess risk for lung cancer was estimated at 10^{-3} and 10^{-4} for atmospheric concentrations of 0.1 and 0.01 µg Cr(VI).m^{-3} respectively (taking into account the inherent limitations to the interpretation of the results of the key study);
- the limits of quantification of the measurement methods made their application inappropriate for an OEL below 1 $µg.m^{-3}$;
- a “skin” notation should be assigned;
- the ALARA principle should be applied in the case of a non-threshold carcinogen;
- in the absence of available data, it recommended that exposure should not exceed five times the 8-hour OEL over a 15 minute period, in order to limit the magnitude of exposure levels for short exposure times.

[1] Supplementing and amending the Circular of 19 July 1982 on the acceptable values for concentrations of certain hazardous substances in workplace atmospheres.

On the basis of this expert appraisal, restrictive limit values applicable from 1 July 2014 were established by Decree No. 2012-746 of 9 May 2012, i.e. an 8-hour OEL of 1 µg.m^{-3} and a 15 minute short-term limit value (15-min STLV) of 5 µg.m^{-3}.

The OEL Committee decided to supplement its expert appraisal with an assessment of the biological monitoring data on hexavalent chromium in an occupational environment, in order to establish whether it was relevant to recommend the monitoring of one or more indicators in addition to the OEL and the establishment of biological limit values for the selected biomarker(s).

The following report is based on the exposure limit value recently established for calculating concentrations of biomarkers of exposure when it is necessary to link these to atmospheric concentrations.

Scientific background

Biological monitoring of exposure in the workplace has emerged as a complementary method to atmospheric metrology for assessing exposure to chemical agents. Biological monitoring assesses a worker's exposure by including all the routes by which a chemical penetrates the body (lung, skin, digestive tract). It is particularly effective when a substance has a systemic effect, and:

- when routes other than inhalation contribute significantly to absorption;
- and/or when the pollutant has a cumulative effect;
- and/or when the working conditions (personal protection equipment, inter-individual differences in respiratory ventilation, etc.) determine large differences in internal dose that are not taken into account by atmospheric metrology.

With regard to prevention of chemical risk in the workplace, the French Labour Code authorises the use of biological monitoring of exposure and biological limit values.

OEL Committee definitions

Biomarker of exposure: parent substance, or one of its metabolites, determined in a biological matrix, whose variation is associated with exposure to the agent targeted. Biomarkers of early and reversible effects are included in this definition when they can be specifically correlated to occupational exposure.

Biological limit value (BLV): This is the limit value for the relevant biomarkers.

Depending on the available data, the recommended biological limit values do not all have the same meaning:

- if the body of scientific evidence is sufficient to quantify a dose/response relationship with certainty, the biological limit values (BLVs) are established on the basis of health data (no effect for threshold substances or risk levels for non-threshold carcinogens);
- in the absence of such data for substances with threshold effects, BLVs are calculated on the basis of the expected concentration of the biomarker of exposure (BME) when the worker is exposed to the 8-hour OEL. For carcinogens, in the absence of sufficient quantitative data, the biological limit value is calculated on the basis of another effect (pragmatic BLV). These last values do not guarantee the absence of health effects, but aim to limit exposure to these substances in the workplace.

Whenever possible, the OEL Committee also recommends biological reference values (BRVs). These correspond to concentrations found in a general population whose characteristics are similar to those of the French population (preferentially for biomarkers of exposure) or in a

control population not occupationally exposed to the substance under study (preferentially for biomarkers of effect).

These BRVs cannot be considered to offer protection from the onset of health effects, but do allow a comparison with the BME levels measured in exposed workers. These values are particularly useful in cases where it is not possible to establish a BLV.

Organization of the expert appraisal

The Agency entrusted examination of this request to the Expert Committee on Expert appraisal for recommending occupational exposure limits for chemical agents (OEL Committee). The Agency also mandated the Working Group on biomarkers for this expert appraisal.

The methodological and scientific aspects of the Working Group's work were regularly submitted to the OEL Committee. The report produced by the Working Group takes account of observations and additional information provided by the Committee members.

This expert appraisal was therefore conducted by a group of experts with complementary skills. It was carried out in accordance with the French Standard NF X 50-110 "Quality in Expertise Activities".

Preventing risks of conflicts of interest

ANSES analyses interests declared by the experts before they are appointed and throughout their work in order to prevent potential conflicts of interest in relation to the points addressed in expert appraisals.

The experts' declarations of interests are made public on ANSES's website (www.anses.fr).

Description of the method

A rapporteur from this working group was mandated by the Agency to produce a summary report on biomarkers of exposure (BMEs) and the recommendation of biological limit values (BLVs) and biological reference values (BRVs) for the BME(s) considered relevant. An ANSES employee also contributed to this report.

The summary report on the BMEs for hexavalent chromium and its compounds was based on bibliographical information taking into account the scientific literature published on this substance until 2012.

The bibliographical research was conducted in the following databases: Medline, Toxline, HSDB, ToxNet (CCRIS, GENE-TOX, IRIS) and ScienceDirect. The rapporteur reassessed the source articles or reports cited as references whenever he considered it necessary, or whenever the Committee so requested.

The report, the summary and conclusions of the collective expert appraisal work were adopted by the Expert Committee on expert appraisal for recommending occupational exposure limits for chemical agents on 11 October 2013.

The collective expert appraisal work and the summary report were submitted to public consultation from 28/04/2014 to 30/06/2014. The people or organizations who contributed to the public consultation are listed in appendix of the report (only available in French). The comments received were reviewed by the OEL Committee (term of office 2014-2016) who adopted this version on 16 December 2014.

Result of the collective expert appraisal

Introduction

The scientific articles selected for evaluating biomonitoring data on hexavalent chromium and its compounds were identified using the following keywords: “chromium”, “biomarker”, “biomonitoring”, “urine”, “blood” and “occupational”, while limiting the search to human data.

Toxicokinetics data

Toxicokinetic properties are generally linked to the valence state of the chromium atom (VI in this document), and the nature of the compound, which primarily determines the solubility.

Although absorption by the pulmonary route is predominant, dermal absorption is non-negligible and hexavalent chromium and its compounds carry a “skin” notation.

To simplify, chromium(VI) is absorbed more efficiently by the pulmonary route than chromium in its other valences.

As a general rule, the quantity, the deposition and absorption of inhaled chromium are determined by the factors that influence the behaviour of the particles in the respiratory tract and the nature of the compound. The most water-soluble compounds have a lower pulmonary retention time than compounds with low water-solubility (ATSDR, 2012) and are therefore absorbed more rapidly. Several studies on volunteers have assessed oral absorption.

Examinations and autopsies carried out on workers exposed to chromium (mostly in valence(VI)) showed higher levels of chromium in different tissues and organs (liver, brain, lungs, heart, lymph nodes, bone marrow, muscle, adrenal glands, etc.) than the levels observed in subjects not exposed in the workplace (ATSDR, 2012).

Chromium(VI), or Cr(VI), is reduced in the body (e.g. stomach, lung) to Cr(III), after transition through Cr(V) and Cr(IV), which can then be eliminated. This reduction occurs inside cells. Once Cr(VI) enters the blood, it can be reduced into Cr(III) in the plasma or penetrate in the erythrocytes. There are divergent hypotheses explaining the potential passage of Cr(III) into cells.

Following exposure via inhalation, the compounds of Cr(VI) are mostly excreted via the urinary route and rather less via the faeces (OSHA, 2006). Whether in urine or faeces, excretion occurs via complex substances formed from Cr(III) and proteins with a low molecular weight. In cases of oral exposure, unabsorbed Cr VI is eliminated mostly in the faeces. As a general rule, excretion occurs in urine, with very little being excreted in exhaled air.

Choice of biomarkers of exposure and effect

Only red blood cell (RBC) chromium levels are specific to exposure to Cr(VI). There is insufficient data from the general population or from the workplace to establish reference values or limit values for this biomarker. No study in the workplace was identified that linked levels of this BME with potential health effects of Cr(VI), nor even with atmospheric concentrations. The Committee considers it important that research be continued, given the importance of this BME (specific to exposure to Cr(VI)).

Chromium, whether measured in total blood, plasma or urine, is not specific to occupational exposure to Cr(VI) and also includes exposure to Cr(III) (by inhalation and/or the dietary route). Regarding Cr in total blood or plasma, the lack of field data means that there is insufficient data to recommend exposure biomarkers (in addition to the fact that sampling would be invasive). However, several studies carried out in occupational environments were found that reported measurements of urinary Cr associated with the study of potential renal toxicity or oxidative mechanisms linked to exposure to chromium(VI).

Although urinary chromium is not specific, this BME can be recommended for biomonitoring of exposure to Cr(VI) by taking into account levels in the general adult population not exposed to Cr(VI) in the workplace.

For information purposes only, some studies report the formation of DNA-adducts of Cr(VI) and its reaction products, especially at the N^8 site of guanine (Singh et al., 1999; Wise et al., 2002; Wise et al., 2004). However, technical difficulties related to their detection and the absence of reference values for interpreting their frequency of occurrence mean that it is difficult to build upon these exposure biomarkers.

Impaired renal function in association with exposure to Cr(VI) was suggested in several field studies on Cr(VI) but the toxicity mechanisms are not clearly established.

Oxidative stress and possible lesions to the DNA related to exposure to Cr(VI) are currently under study. However, the lack of reference data makes it impossible to use such effect indicators. These will be incorporated into the scientific report once they have been studied in association with measurements of exposure biomarkers for Cr(VI), although it will not be possible to recommend any monitoring measures.

Information on biomarkers of exposure identified as relevant for the biological monitoring of exposed workers

Name	**URINARY CHROMIUM (Cru)**
Other substances producing this BME	Compounds of Cr(III)
Levels found in exposed workers or volunteers	- Field studies: (atmospheric samples taken throughout the duration of the shift and urine levels measured after spot samples) **Open arc welding** Edme et al. (1997): Cr(VI)atmo = 45 µg.m^{-3} (AM); Cru = 8 µg.L^{-1} (AM; EWES) **Chromium plating** Benova et al. (2002): Cr(VI)atmo = 18 µg.m^{-3} (AM); Cru = 73 µg.L^{-1} (AM; ES day NS) Chen et al. (2002): Cr(VI)atmo = 2 µg.m^{-3} (AM); Cru = 3 µg.g^{-1} cr (AM; EWES) Chen et al. (2002): Cr(VI)atmo = 25 µg.m^{-3} (AM); Cru = 46 µg.g^{-1} cr (AM; EWES) - Studies on volunteers: NS
Conversion factor	Molecular weight: 52 1 µg.L^{-1} = 0.02 µmol.L^{-1} 1 µmol.L^{-1} = 52 µg.L^{-1} 1 µg.g^{-1} creatinine = 2.17 µmol.mol^{-1} creatinine 1 µmol.mol^{-1} creatinine = 0.46 µg.g^{-1} creatinine
Levels in the general population	France-ENNS (1939 people from the general population) 95th percentile: 0.65 µg.L^{-1}; 0.54 µg.g^{-1} creatinine (Fréry et al., 2011)

	The MAK Commission in Germany gives a concentration of 0.6 µg.L^{-1} as the 95th percentile in the distribution of urine levels of total chromium in the general population who are of working age, non-smokers, and not exposed in the workplace (Biologische Arbeitsstoff-Referenzwerte "BAR" value) (Deutsche Forschungsgemeinschaft, 2012)	
Recommended limit values for exposed workers (INRS, 2012)	USA – ACGIH (for water-soluble aerosols)	EWES: 25 µg.L^{-1} Maximum difference between SS and ES: 10 µg.L^{-1} (ACGIH, 2004)
	Finland – FIOH (for exposure to chromium and its inorganic derivatives)	EWES: 0.01 µmol.L^{-1} (0.5 µg.L^{-1}) (FIOH, 2012)
	USA – OSHA	NS
	Quebec – IRSST (for water-soluble compounds)	EWES: 65 nmol.mmol^{-1} cr (28.5 µg.g^{-1} cr) Maximum difference between SS and ES: 22 nmol.mmol^{-1} cr (10 µg.g^{-1} cr) (IRSST, 2012)

AM: arithmetic mean; cr: creatinine; GM: geometric mean; EW: end of week; ES: end of shift; SW: start of week; SS: start of shift; NS: not specified

Study of relationships between urine levels of chromium and certain health effects

Renal toxicity

Several field studies showed increased concentrations of certain markers of renal toxicity (markers of tubular and/or glomerular toxicity) related to high urine concentrations of Cr (Foa et al., 1988; Franchini and Mutti, 1988; Liu et al., 1998; Mutti et al., 1979; Nagaya et al., 1994; Verschoor et al., 1988). However, the mechanism of renal toxicity is not yet clear and it does not seem possible to identify a dose-response relationship with any certainty. There is no indication of what the target is for the potential renal toxicity of Cr(VI). Verschoor et al. (1988), for example, demonstrated an increase in levels of blood beta-2-microglobuline (β2MG) as markers of glomerular damage in exposed workers (with mean urine levels of Cr equal to 5 µg.g^{-1} of creatinine) relative to non-exposed workers (with mean urine levels of Cr 10 times lower), but no increase in levels of markers of tubular damage. Liu et al. (1998), on the other hand, do not show any evidence of an increase in levels of markers of glomerular damage but an increase in levels of a very early marker of tubular toxicity (urinary N-acetylglucosaminidase, NAG) in workers exposed in hard chromium plating (mean urine levels of Cr equal to 2.4 µg.g^{-1} of creatinine).

The results of the study of the literature reporting measurements of renal toxicity in relation to levels of urinary Cr are summarised in the following table.

Table 1: Summary of urinary chromium concentrations measured simultaneously with parameters of renal toxicity

Reference group (least exposed workers, n = 39) Mean: 5.3 µg.g^{-1} cr	Highly exposed workers Mean (chromium plating, n = 24): 24.5 µg.g^{-1} cr Mean (welding armoured steel, n = 36): 33.3 µg.g^{-1} cr Significant increase in urinary beta-glucuronidase in both groups of highly exposed workers, relative to the least exposed workers Significant increase in total urinary proteins in the "chromium plating" group relative to the least exposed workers No significant increase in urine levels of total proteins in the "welding" group relative to the least exposed workers	Mutti et al. (1979)
Reference group (non-exposed workers + slightly	Highly exposed workers (n = 74)	Verschoor et al. (1988)

exposed boilermakers, n = 89) Mean: 0.5 µg.g^{-1} cr (0.1 to 2 µg.g^{-1} cr)	Mean: 5 µg.g^{-1} cr (1 to 34 µg.g^{-1} cr) No significant increase in markers of glomerular toxicity in highly exposed workers relative to the reference group (slightly or non-exposed workers) No significant increase in markers of tubular toxicity in highly exposed workers relative to the reference group (slightly or non-exposed workers)	
Reference group (least exposed workers, n = 71) Mean: 0.4 µg.g^{-1} cr (0.1 to 2 µg.g^{-1} cr)	Highly exposed workers (n = 74) Mean: 5 µg.g^{-1} cr (1 to 34 µg.g^{-1} cr) Significant increase in markers of glomerular toxicity (principally blood β2M) in the most exposed workers relative to non-exposed workers No increase in markers of tubular toxicity in the most exposed workers relative to non-exposed workers	
Reference group (least exposed workers, n = 18) Mean: 1 µg.g^{-1} cr (0.3 to 1.5 µg.g^{-1} cr)	Highly exposed workers (n = 74) Mean: 5 µg.g^{-1} cr (1 to 34 µg.g^{-1} cr) Significant increase in markers of glomerular toxicity (principally blood β2M) in the most exposed workers relative to the least exposed workers No increase in markers of tubular toxicity in the most exposed workers relative to the least exposed workers	
Reference group; the authors do not specify whether they are workers not exposed to Cr(VI) < 2 µg.g^{-1} cr (n = 39)	Exposed workers (n = 43) > 15 µg.g^{-1} cr Significant increase in urine levels of BB50 in exposed workers relative to the reference group No dose-response relationship	Franchini and Mutti (1988)
Reference group (non-exposed workers) Urine levels of Cr are not reported in this publication (but it is indicated that they are < 1 µg.g^{-1} cr)	Exposed workers (n = 166) 1st quartile: 0.6 to 2.9 µg.g^{-1} cr 2nd quartile: 0.7 to 1.9 µg.g^{-1} cr 3rd quartile: 2 to 3.9 µg.g^{-1} cr 4th quartile: 4 to 19.9 µg.g^{-1} cr No significant increase in levels of markers of nephrotoxicity in exposed workers relative to non-exposed workers Statistical association between increase in urine levels of Cr and urine levels of total proteins (analysis per quartile of urinary Cr levels) Urine levels of the other markers (ALB and RBP) are not statistically associated with urine levels of Cr	Nagaya et al. (1994)
Reference group (least exposed workers, n = 46) Mean: 0.09 µg.g^{-1} cr (0.01 – 5.44)	Slightly exposed workers (n = 98) Mean: 0.31 µg.g^{-1} cr (0.02 to 19.84 µg.g^{-1} cr) No significant increase in urine levels of µALB and total proteins (markers of glomerular toxicity) in slightly exposed workers relative to the least exposed workers No significant increase in urine levels of markers of tubular toxicity (RBPu, BB50u) in slightly exposed workers relative to the least exposed workers	Liu et al. (1998)

Reference group (least exposed workers, n = 46) Mean: 0.09 µg.g^{-1} cr (0.01 – 5.44)	Most exposed workers (n = 34) Mean: 2.4 µg.g^{-1} cr (0.13 to 20.98 µg.g^{-1} cr) No significant increase in urine levels of µALB and total proteins (markers of glomerular toxicity) in the most exposed workers relative to the least exposed workers Significant increase in NAG activity (a marker of tubular toxicity) in the urine of the most exposed workers relative to the least exposed workers No significant increase in urine levels of β2M (another marker of tubular toxicity) in the most exposed workers relative to the least exposed workers	

* SWES: Start of week and end of shift; EWES: end of week and end of shift

Mechanisms of genotoxicity

Some authors investigated the mechanisms of the genotoxicity of Cr by measuring, in humans, markers of lipid peroxidation or levels of DNA-protein cross-links. There are still only a few results from human studies.

Huang et al. (1999) and Kalahasthi et al. (2006) demonstrated a significant increase in urine and/or blood levels of malondialdehyde (MDA), an indicator of lipid peroxidation in exposed workers but no increase in other markers of peroxidation, such as erythrocyte superoxide dismutase (SOD), blood or erythrocyte glutathione peroxidase (GPX) and erythrocyte catalase (CAT). Huang et al. (1999) also report correlation equations between urine or blood levels of MDA and urine levels of Cr.

Caglieri et al. (2006) report statistically significant correlations between levels of Cr and different markers of lipid peroxidation measured in exhaled air condensates. However, they did not demonstrate any relationship between urine levels of Cr and levels of markers of lipid peroxidation in exhaled air.

In a study of several publications, Costa et al. (1993 and 1996) and Taioli et al. (1995) present the following results concerning lymphocyte levels of DNA-protein cross-links:

- welder exposed to fumes containing Cr(VI): 1.8% (non-smokers) and 1.9% (smokers)
- general population exposed to Cr(VI): 1.3%
- general population not exposed to Cr(VI): 0.8%

The authors did not find any correlation between levels of cross-linking in lymphocytes and urine levels of Cr.

It should be noted that potential immunotoxic effects studied in relationship with urine levels of chromium have been reported in only one publication (Kuo and Wu, 2002). Similarly, local effects such as irritation (studied by questionnaire or clinical examination) have also been reported in two publications that provide results from measurements of urine levels of Cr (Kuo et al., 1997b; Lumens et al., 1993).

<u>A study of correlations between urine levels of chromium and atmospheric levels of Cr(VI)</u>

Table 2: Summary of studies in the literature reporting measurements of atmospheric levels of Cr(VI) in relation to urine levels of Cr

n	Atmospheric concentrations of Cr(VI) ($\mu g.m^{-3}$)	Urine levels		Reference
	Median Mean [min – max]			
		Arc welding		
25	NS [1 to 510]	NS	[Cru] ($\mu g.g^{-1}$ cr) = 0.26 [Cr(VI)a] ($\mu g.m^{-3}$) + 24.56 r = 0.88 Urine and atmospheric samples taken every day of the working week Atmospheric Cr(VI): LOD = 1 to 2 $\mu g.m^{-3}$ Cru: LOD not specified	Tola et al. (1977)
20	NS	(AM) 33.3 $\mu g.g^{-1}$ cr ± 12.5	Urine samples at end of shift on several consecutive days	Mutti et al. (1979)
5	(AM) 150 [30 – 960]	(AM) 37.8 $\mu g.g^{-1}$ cr [19.3 – 67.2]	[Cru] ($\mu g.g^{-1}$ cr) = 0.10 [Cr(VI)a] ($\mu g.m^{-3}$) + 25.8 r = 0.91 Urine levels measured on 48 hours of urine Atmospheric samples over the whole duration of the shift LODs were not specified	Rahkonen et al. (1983)
103	NS	(AM) 51.2 $\mu g.g^{-1}$ cr [5.4 – 229.4]	Atmospheric samples measured over 2 to 5h Urine samples taken at end of shift over several consecutive days	Angerer et al. (1987)
116	(AM) 45.3 [1 – 649]	(AM) 8 $\mu g.L^{-1}$ [NS]	Atmospheric samples taken over the whole working day Urine samples taken at end of week and end of shift	Edme et al. (1997)
16	(GM) 0.2 [0.02 – 1.5]	(GM) 0.9 $\mu g.g^{-1}$ creat. [0.2 – 7.7]	Atmospheric samples taken over 4 hours Urine samples taken at end of week and end of shift	Gianello et al. (1998)
		Chromium plating		
57	NS [0.2 – 20]	NS	[Cru] ($nmol.L^{-1}$) = 77 [Cr(VI)a] ($\mu g.m^{-3}$) – 33 r = 0.71 Atmospheric samples taken over full duration of shift (day not specified) Urine samples taken at end of shift on 2nd working day Atmospheric Cr(VI): LOD = 0.2 $\mu g.m^{-3}$ Cru: LOD = 0.25 $\mu g.L^{-1}$	Lindberg et al. (1983)
15	(AM) 18 [4.2 – 47]	(AM) 73 $\mu g.L^{-1}$ [6.7 – 245.6]	Log[Cru] ($\mu g.L^{-1}$) = 0.96 Log[Cr(VI)a] ($\mu g.m^{-3}$) + 0.5 r = 0.35 Samples taken at end of shift (day NS) Atmospheric samples throughout working day (day NS) Total atmospheric Cr: LOD = 0.08 $\mu g.m^{-3}$ (No information concerning LD for Cr(VI)) Cru: LOD not specified	Benova et al. (2002)

27	(AM) 1.9 [0.3 – 14.0]	(AM) 3.4 µg.g^{-1} cr [0.6 – 29.2]	Decorative chromium plating: [Cru] (µg.g^{-1} cr) = 1.75 [Cra] (µg.m^{-3}) + 0.05 r = 0.86 Atmospheric samples taken over the whole working day Urine samples taken at end of week and end of shift Atmospheric Cr(VI): LOD = 2 ng.m^{-3} Cru: LOD = 0.2 ng.L^{-1} Wearing of personal protective clothing (as specified in the study)	Chen et al. (2002)
30	(AM) 25.2 [4.4 – 96]	(AM) 46.2 µg.g^{-1} cr [7.7 – 187]	Hard chromium plating: [Cru] (µg.g^{-1} cr) = 1.86 [Cra] (µg.m^{-3}) - 0.33 r² = 0.81 Atmospheric samples taken over the whole working day Urine samples taken at end of week and end of shift Atmospheric Cr(VI): LOD = 2 ng.m^{-3} Cru: LOD = 0.2 ng.L^{-1} Personal protective clothing worn	
57	Decorative + hard chromium plating		[Cru] (µg.g^{-1} cr) = 1.86 [Cra] (µg.m^{-3}) – 0.21 r = 0.86 Atmospheric samples taken over the whole working day Urine samples taken at end of week and end of shift Atmospheric Cr(VI): LOD = 2 ng.m^{-3} Cru: LOD = 0.2 ng.L^{-1} Personal protective clothing worn	
All sectors				
137	(AM) 19.1 [0 – 212]	(AM) 19.2 [NS]	[Cru] (µg.g^{-1} cr) = 0.384 [Cr(VI)a] (µg.m^{-3}) + 10.62 r = 0.88 Regression performed based on means calculated per sector Urine samples taken at end of shift (day NS) Atmospheric samples taken over 30 to 60 min LODs not specified	Mutti et al. (1984)

AM: arithmetic mean; GM: geometric mean; EW: end of week; ES: end of shift; SW: start of week; SS: start of shift; NS: not specified; LOD: limit of detection

Establishment of BLVs and choice of biological reference values

In the scientific literature no dose-effect relationship between urinary Cr concentrations and the critical effect chosen by the OEL Committee to calculate risk excess (lung cancer) has been identified.

DNA oxidations and the formation of interstrand DNA or DNA-protein cross-links are included in the continuum between exposure to Cr(VI) and its genotoxic potential in the lung, as emphasised by the ATSDR. How this damage is repaired and the types of mutations it generates in humans are still poorly understood. It is not possible to undertake a quantitative

analysis of the data in order to investigate a possible dose-response relationship on the basis of this type of molecular mechanism.

Some studies can be referred to in order to calculate urine levels of Cr depending on atmospheric concentrations of Cr(VI). This makes it possible to calculate, indirectly, individual excess risk linked with urine levels of Cr. This approach is therefore considered to be the most relevant.

The calculations of urine levels of Cr are based on the exposure limit value set at 1 µg.m^{-3} applicable from 1 July 2014 (Decree of 9 May 2012) as the reference atmospheric concentration. The results of the calculations of urine levels of Cr on the basis of atmospheric concentrations of Cr(VI) (regression equation) are given in Table 2. It is worth noting that the calculations by the OEL Committee indicate that an atmospheric concentration of 1 µg.m^{-3} corresponds to a risk of an additional case of lung cancer for 100 workers exposed 8 hours a day for 40 years.

Most of the field studies, especially those concerning exposure to welding fumes, were carried out for very high exposures in the past when analytical capabilities were unable to measure atmospheric concentrations at the level of the OEL established in 2012. The urine levels of Cr calculated for low atmospheric concentrations of Cr on the basis of these relationships give lower values than those observed in workers not exposed to compounds of hexavalent chromium or in the general population (Tola et al., 1977; Rahkonen et al., 1983; Mutti et al., 1984; Mutti et al., 1979).

Furthermore, in the study on volunteers by Gube et al. (2013) the urine level of Cr corresponding to atmospheric concentrations of Cr of 1 µg.m^{-3} (or less) and calculated on the basis of the regression equation is lower than the limit of detection indicated in the study. In this study, the exposed subjects were not workers in the chromium sector and presented urine levels that seem low bearing in mind the exposure to which they were subject. Indeed, it is worth noting that in this case there is only a single exposure event and not chronic exposure as occurs in the occupational environment.

It is difficult to interpret the results of the study by Caglieri et al. (2006) as only the concentrations of total Cr are reported[2].

Two studies were carried out with a higher level of analytical capability, making it possible to extrapolate urine levels on the basis of the OEL (1 µg.m^{-3}) while remaining within the scope of validity of the methods used (Lindberg et al., 1983; Chen et al. 2002). Levels of urinary Cr calculated on the basis of these two studies are between 2.3 and 2.5 µg.L^{-1} (1.6 and 1.8 µg.g^{-1} of creatinine), with a mean of 2.4 µg.L^{-1} for exposure to the OEL of 1 µg.m^{-3}.

It was not deemed relevant to include the study by Benova et al. (2002) as the atmospheric concentrations of Cr(VI) reported did not include the concentration of 1 µg.m^{-3} within their range.

However, the question of the nature of the exposures concerned by the relationship identified between atmospheric and urinary concentrations of Cr needs to be investigated. This relationship was of course identified for a specific industrial sector, chromium plating, where Cr(VI) was measured, and cannot be applied to all Cr sectors where workers are not exposed to the same compounds (joint exposure to Cr(VI) and Cr(III)).

[2] It has been calculated that urine levels of Cr equating to exposure to 1 µg.m^{-3} of Cr(VI) would be between 5.75 and 6.5 µg.g^{-1} of creatinine. The calculation was based on the assumption that:

- either all the urinary Cr was the result of exposure to Cr(VI): Log[Cru] = 0.34 Log(1) + 0.76 → [Cru] = 5.75 µg.g of creatinine
- or the urinary excretion from Cr(III) and Cr(VI) was of equal proportions and the proportion of Cr(VI) relative to total Cr in the work atmosphere was 70%.. Atmospheric concentration calculated for total Cr, considering atmospheric concentration of 1 µg.m-1 for Cr(III), is therefore equal to 1.43 µg.m^{-3}: Log[Cru] = 0.34 Log(1.43) + 0.76 → [Cru] = 6.5 µg.g^{-1} of creatinine

The Committee proposes 2.5 µg.L^{-1} (1.8 µg.g^{-1} of creatinine) as the BLV for urinary Cr at end of week and end of shift and recommends applying this value only to exposure to compounds of Cr(VI).

In cases of joint exposure (Cr(III) and Cr(VI)) and bearing in mind the contribution of exposure to Cr(III) to urinary Cr, urine measurements could be made but should be interpreted in the light of the respective atmospheric concentrations of the different compounds of Cr.

The French ENNS study (*Etude Nationale Nutrition Santé*) in the general population can be used to establish a biological reference value. The level of chromium in urine, which corresponds to the 95th percentile of the distribution in this study, is 0.65 µg.L^{-1} or 0.54 µg.g^{-1} of creatinine (Fréry et al., 2011).

The biological reference value chosen for urinary chromium is 0.65 µg.L^{-1} or 0.54 µg.g^{-1} of creatinine.

Conclusions of the collective expert appraisal

The biological values proposed for monitoring exposure to hexavalent chromium are:

Urinary chromium:

BLV based on exposure to the 8-hour OEL (1 µg.m^{-3}): **2.5 µg.L^{-1} (1.8 µg.g^{-1} of creatinine) (end of week)**

This value only applies to exposures to CrVI in the chrome-plating sector.

Biological reference value: **0.65 µg.L^{-1} or 0.54 µg.g^{-1} of creatinine.**

Sampling method and factors that may affect the interpretation of results

Considering the uses and the kinetics data, the Committee recommends sampling at end of week and end of shift. These measurements reflect exposure from the preceding days and also long-term exposure. Normal sampling equipment may be used as long as the necessary precautions are taken to avoid contaminating samples (sampling outside the workplace and, at the least, after the subject has taken a shower).

In addition to samples at end of week and end of shift, the ACGIH proposes taking samples at the beginning and end of shifts in order to assess the difference between these concentrations. The ACGIH considered that this excludes exposures that are not of occupational origin. However, the 8-hour OEL of 1 µg.m^{-3} means that the available field studies cannot be used to quantify this difference.

Samples may be stored for up to 15 days at 4°C but no preserving agent must be added to samples.

Biomonitoring

URINARY CHROMIUM					
Interlaboratory quality control		Institute and out-patient clinic for occupational, social and environmental medicine of the University Erlangen-Nuremberg (Germany): G-EQUAS National Public Health Institute of Quebec, Toxicology Centre: PCI			
Analytical technique	**Limit of detection Limit of quantification**	**Reliability**	**Precision**	**Benchmark**	**Bibliographic reference**
Electrothermal atomic absorption spectrometry (ETAAS)	LD: 0.5 µg.L^{-1} (standard tubes) LD: 0.1 µg.L^{-1} (graphite tubes)	Standard deviation = 0.5 µg.L^{-1} (between 5.9 and 50.7 µg.L^{-1})	NS	Standard solution of potassium dichromate at 0.1 g of chromium	Fleischer (2012)
Inductively Coupled Plasma Mass Spectrometry (ICP-MS)	LD: 5 nmol.L-1	Within-day: SD < 3% Between-day: SD < 7% at 380 nmol.L^{-1}	NS	NS	HSL (2013)

References

ACGIH. (2004). Chromium (VI), water soluble fume in 'Threshold limit values for chemical substances and physical agents and biological exposure indices'. 7th ed. (American Conference of Industrial Hygienists: Cincinnati, United States). 9 p.

ANSES. (2010). *Évaluation des effets sur la santé et des méthodes de mesure des niveaux d'exposition sur le lieu de travail pour les composés du chrome hexavalent* [Assessment of health effects and methods for measuring exposure levels in the workplace for compounds of hexavalent chrome]. (French Agency for Food, Environmental and Occupational Health & Safety: Maisons-Alfort, France). 95 p.

Angerer J, Amin W, Heinrich-Ramm R, et al. (1987). Occupational chronic exposure to metals: I. Chromium exposure of stainless steel welders biological monitoring. Int Arch Occup Environ Health. 59:503-512.

ATSDR. (2008). Toxicological Profile for chromium. (Agency for toxic substances and disease registry: Atlanta, United States). 610 p.

Benova D, Hadjidekova V, Hristova R, et al. (2002). Cytogenetic effects of hexavalent chromium in Bulgarian chromium platers. Mutat Res. 514:29-38.

Caglieri A, Goldoni M, Acampa O, et al. (2006). The effect of inhaled chromium on different exhaled breath condensate biomarkers among chrome-plating workers. Environ Health Perspec. 114(4):542-546.

Chen, JH, Guo YL, Tsai PJ, Su LF. (2002). Use of inhalable Cr+6 exposures to characterize urinary chromium concentrations in plating industry workers. J Occup Health. 44: 46-52.

Costa M, Zhitkovich A, Taioli E, Toniolo P. (1993). Preliminary report on a simple new assay for DNA-protein cross-links as a biomarker of exposures experienced by welders. J Toxicol Environ Health. 40(2-3):217-222.

Costa M, Zhitkovich A, Toniolo P, et al. (1996). Monitoring human lymphocytic DNA-protein cross-links as biomarkers of biologically active doses of chromate. Environ Health Perspect. 104 Suppl 5:917-919.

Deutsche Forschungsgemeinschaft. (2012). BAR, in List of MAK and BAT Values 2012: Maximum Concentrations and Biological Tolerance Values at the Workplace. Wiley-VCH: Weinheim, Ge.

Edme JL, Shirali P, Mereau M, et al. (1997). Assessment of biological chromium among stainless steel and mild steel workers in relation to welding processes. Int Arch Occup Environ Health. 70:237-242.

Foa V, Riboldi L, Patroni M, et al. (1988). Effects derived from long-term low-level chromium exposure in ferro-alloy metallurgy. Study of absorption and renal function in workers. Sci Total Environ. 71:389-400.

FIOH. (2012). Biomonitoring of exposure to chemicals – Guideline for specimen collection 2011-2012. (Finnish Institute of Occupational Health: Helsinki, Finland). 64 p.

Fleischer M. (2012). Chromium [Biomonitoring Methods, 1988]. The MAK Collection for Occupational Health and Safety. 97–115.

Franchini I, Mutti A. (1988). Selected toxicological aspects of chromium(VI) compounds. Sci Total Environ. 71(3):379-87.

Fréry N, Saoudi A, Garnier R, et al. (2011). Exposition de la population française aux substances chimiques de l'environnement – Tome 1. (Institut de veille sanitaire: Saint-Maurice, France). 151 p.

Gianello G, Masci O, Carelli G, et al. (1998). Occupational exposure to chromium—An assessment of environmental pollution levels and biological monitoring of exposed workers. Ind Health. 36:74-77.

Gube M, Brand P, Schettgen T, et al. (2013). Experimental exposure of healthy subjects with emissions from a gas metal arc welding process-Part II: biomonitoring of chromium and nickel. Int Arch Occup Environ Health. 86:31-37.

HSL. (2013). Guidance sheet for: Method for Chromium in Urine. Available on website http://www.hsl.gov.uk/online-ordering/analytical-services-and-assays/biological-monitoring/bm-guidance-values.aspx consulted 07-03-2013

Huang Y, Chen C, Sheu J, et al. (1999). Lipid peroxidation in workers exposed to hexavalent chromium. J Toxicol Environ Health, A. 56:235-247.

IRSST. (2012). Guide de surveillance biologique de l'exposition. 7th ed. (Institut de Recherche Robert-Sauvé en Santé: Montréal, Québec). 107 p.

Kalahasthi RB, Rao RH, Murthy RB, et al. (2006). Effect of chromium(VI) on the status of plasma lipid peroxidation and erythrocyte antioxidant enzymes in chromium plating workers. Chem Biol Interact. 164(3):192-199.

Kuo HW, Lai JS Lin TI. (1997b). Nasal septum lesions and lung function in workers exposed to chromic acid in electroplating factories. Int Arch Occup Environ Health 70: 272-276.

Kuo HW and Wu ML. (2002). Effects of chromic acid exposure on immunological parameters among electroplating workers. Int Arch Occup Environ Health. 2002 Mar;75(3):186-190.

Lindberg E, Vesterberg O. (1983). Monitoring exposure to chromic acid in chromeplating by measuring chromium in urine. Scand J Work Environ Health 9:333-340.

Lumens ME, Ulenbelt P, Géron HM, et al. (1993). Hygienic behaviour in chromium plating industries. Int Arch Occup Environ Health. 64(7):509-14.

Liu CS, Kuo HW, Lai JS, et al. (1998). Urinary N-acetyl-B-glucosaminidase as an indicator of renal dysfunction in electroplating workers. Int Arch Occup Environ Health. 71:348-352.

Mutti A, Cavatorta A, Borghi L, et al. (1979). Distribution and urinary excretion of chromium: Studies on rats after administration of single and repeated doses of potassium dichromate. Med Lav. 3:171-179.

Mutti A, Pedroni C, Arfini G, et al. (1984). Biological monitoring of occupational exposure to different chromium compounds at various valency states. Int J Environ Anal Chem. 17(1):35-41.

Nagaya T, Ishikawa N, Hata H, et al. (1994). Early renal effects of occupational exposure to low-level hexavalent chromium. Arch Toxicol. 68(5):322-324.

OSHA. (2006). Occupational exposure to hexavalent chromium. Final rule: Federal Register, v. 71, no. 39, p. 10099-10385.

Rahkonen E, Junttila ML, Kalliomäki PL, et al. (1983). Evaluation of biological monitoring among stainless steel welders. Int Arch Occup Environ Health. 52(3):243-255.

Singh J, Pritchard DE, Carlisle DL, et al. (1999). Internalization of carcinogenic lead chromate particles by cultured normal human lung epithelial cells: formation of intracellular lead-inclusion bodies and induction of apoptosis. Toxicol Appl Pharm. 161: 240–248.

Taioli E, Zhitkovich A, Kinney P, et al. (1995). Increased DNA-protein crosslinks in lymphocytes of residents living in chromium-contaminated areas. Biol Trace Elem Res. 50(3):175-180.

Tola S, Kilpio J, Virtamo M, et al. (1977). Urinary chromium as an indicator of the exposure of welders to chromium. Scand J Work Environ Health. 3:192-202.

Verschoor MA, Bragt PC, Herber RFM, et al. (1988). Renal function of chrome-plating workers and welders. Int Arch Environ Health 60: 67-70.

Wise SS, Elmore LW, Holt SE, et al. (2004). Telomerase-mediated lifespan extension of human bronchial cells does not affect hexavalent chromium-induced cytotoxicity or genotoxicity. Mol Cell Biochem. 255: 103–111.

Wise Sr. JP, Wise SS, Little JE. (2002). The cytotoxicity and genotoxicity of particulate and soluble hexavalent chromium in human lung cells. Mutat Res. 517:221–229.