COLLECTIVE EXPERT APRAISAL: SUMMARY AND CONCLUSIONS

Regarding the "expert appraisal on recommending occupational exposure limits for chemical agents"

On the evaluation of biomarkers of exposure and recommendation for biological limit values and biological reference values for perchloroethylene

[CAS n°:127-18-4]

This document summarises the work of the Expert Committees on "expert appraisal for recommending occupational exposure limits for chemical agents" (OEL Committee), on "health reference values" and the Working Group on biomarkers (Biomarkers WG).

Presentation of the issue

On 12 June 2007, AFSSET, which became ANSES in July 2010, was requested by the Directorate General for Labour to carry out the necessary assessment for setting occupational exposure limits for perchloroethylene.

The European expert committee in charge of expert appraisals on occupational exposure limits for chemical agents (SCOEL) issued an opinion on the health effects of perchloroethylene. This report, submitted for public consultation by the European Commission until 15 January 2009, recommended, on the basis of an analysis of health effects, an 8 hour-TWA of 20 ppm (138 $mg.m^{-3}$) and a 15 minute short term exposure limit (STEL) of 40 ppm (275 $mg.m^{-3}$). The European committee proposed assigning a "skin" notation and a biological limit value of 0.4 mg perchloroethylene per litre in blood.

The Directorate General for Labour asked the Agency to undertake a critical review of the SCOEL report on perchloroethylene (dating from 2008) and, if necessary, to propose new occupational exposure values based on health considerations.

This request was entrusted to OEL Committee which, in June 2009, issued a report recommending for perchloroethylene:

- establishing an 8h-OEL of 20 ppm (138 $mg.m^{-3}$)
- establishing a short-term limit value (STEL) of 40 ppm (275 $mg.m^{-3}$)
- not assigning a "skin" notation

ANSES decided to supplement its appraisal by assessing the data on biological monitoring in the workplace for perchloroethylene in order to assess the suitability of recommending monitoring one or more biomarkers in addition to the atmospheric OEL and elaboration of biological limit values for the selected biomarker(s).

It should be noted that further to the public consultation phase, the SCOEL published in June 2009 a final report recommending an 8h-OEL of 20 ppm (138 $mg.m^{-3}$) and a STEL (15 min) of 40 ppm (275 $mg.m^{-3}$). In addition to assigning a "skin" mention, it recommends two biological limit values, namely: 0.4 $mg.L^{-1}$ for perchloroethylene in blood and 3 ppm for perchloroethylene in exhaled air.

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 worthwhile 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 provides for the use of biological monitoring of exposure and biological limit values.

Committee definitions

Biomarker of exposure (BME): parent substance, or one of its metabolites, determined in a biological matrix, whose variation is associated with exposure to the targeted agent. 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 BLVs will be 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 latter values do not guarantee the absence of health effects, but aim to limit exposure to these substances in the workplace.

Whenever possible, the 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 BMEs) or in a control population not occupationally exposed to the substance under study (preferentially for biomarkers of effects).

These BRVs cannot be considered to offer protection from the onset of health effects, but do allow a comparison with the concentrations of biomarkers assayed in exposed workers. These values are particularly useful in cases where it is not possible to establish a BLV (Anses, 2017).

Organisation of the expert appraisal

ANSES 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 (Biomarkers WG) for this expert appraisal.

The methodological and scientific aspects of the work of this group 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

An ANSES employee and a rapporteur of the Biomarkers WG produced a summary report on biomarkers of exposure and the recommendation of biological limit values (BLVs) and biological reference values for the BME(s) considered relevant.

The summary report on the BMEs for perchloroethylene was based on bibliographical information taking into account the scientific literature published on this substance until 2016. The bibliographical research was conducted in the following databases: Medline, Scopus and the Public Health Database.

The scientific articles selected for evaluating biomonitoring data on perchloroethylene were identified using the following keywords: “perchloroethylene”, “tetrachloroethylene” “biomarker”, “biomonitoring”, “biological monitoring”, “urine”, “blood”, “occupational”, “analysis method”, and the search was limited to human data.

The rapporteur reassessed the original articles or reports cited as references whenever he considered it necessary, or whenever the Committee requested it.

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 (term of office 2014-2017) on 16 May 2017.

This collective expert appraisal work and the summary report were submitted to public consultation from 13/12/2017 to 13/02/2018. The people or organizations that contributed to the public consultation are listed in appendix 1 of the report (only available in French). The comments received were reviewed by the Committee on Health Reference Values (term of office 2017-2020) who finally adopted this version on the 3rd May 2018.

Result of the collective expert appraisal

Toxicokinetics data

Inhalation is the main route of absorption of perchloroethylene (PERC). Pulmonary absorption is rapid (blood/gas partition coefficient is between 15 and 18) and high: at equilibrium, i.e. when the concentration of PERC in exhaled air per unit of time is constant, pulmonary absorption is estimated at 78-93% in humans (Benoit 1985, Monster & Zielhuis 1983, Chiu *et al.* 2007, US EPA 2012). Pulmonary absorption gradually decreases with PERC saturation in blood and body tissues (Monster *et al.* 1979). Absorption is influenced by physical activity, body fat, and exposure duration and intensity (Opdam 1989).

Percutaneous absorption of PERC occurs in humans when the skin is exposed to the substance in a liquid state, but dermal absorption of PERC vapours is negligible compared with pulmonary absorption.

Available data are scarce on the absorption of PERC via the oral route. Results from studies conducted on animals indicate rapid oral absorption, with rates of nearly 100% (Dallas *et al.* 1994a).

PERC is distributed throughout the body after absorption, regardless of the exposure route. Due to its lipophilicity, PERC accumulates the most in human and animal organs and tissues with high fat content (e.g. liver, kidneys, brain, lungs), with a high retention in adipose tissue. It can also be found in breast milk, as reported in a human case study (Bagnell & Ellenberger 1977) and in a study on rats (Byczkowski & Fisher 1994). It can also cross the placenta and distribute to the foetus and the amniotic fluid of mice exposed via inhalation (Ghantous *et al.* 1986).

In humans, most absorbed PERC is not metabolised, but is excreted unchanged in exhaled air (80-100%) for all routes of absorption (Chiu *et al.* 2007, Monster *et al.* 1979). An estimated 1 to 3% of inhaled PERC is transformed into chlorinated metabolites. The metabolised fraction can follow two metabolic pathways. The main pathway is cytochrome P450 (CYP2E1)-mediated oxidation, which takes place primarily in the liver. In humans, trichloroacetic acid (TCA) is the major urinary metabolite of this oxidative pathway. Trichloroethanol (TCOH) has been detected in the urine of people exposed to PERC in some studies (Birner *et al.* 1996), but not in others. The secondary pathway is the glutathione conjugation pathway, involving glutathione S-transferases (GSTs). This pathway predominates when the enzymes associated with oxidative metabolism are saturated; it transforms quantitatively less PERC at low exposure levels. The importance of this pathway lies in the production of reactive metabolites that are particularly associated with the nephrotoxicity of PERC (involving N-acetyl-S-(1,2,2-trichlorovinyl)-L-cysteine (NAcTCVC)) and its carcinogenicity in rats. In humans, the metabolism of PERC appears to saturate after exposure via inhalation at high concentrations (i.e. > 50 or 100 ppm, depending on the study).

Humans show polymorphism for enzymes involved in PERC metabolism (CYP and GST in particular). Thus, there is measurable variation in the percentage or rate of PERC metabolisation according to ethnic group, sex, alcohol consumption, pre-existing diseases or under the influence of chemical substances (US EPA 2012).

Regardless of the exposure route in humans or laboratory animals, the main route of excretion is exhalation of the unchanged compound (95% in humans), with small quantities of metabolites present in the urine. PBPK modelling considers that pulmonary excretion of PERC occurs in three phases: rapid elimination from highly vascularised tissues, slower elimination from muscle, skin, conjunctive and pulmonary tissues, and very slow elimination from adipose tissues (Guberan & Fernandez 1974). Urinary elimination of unchanged PERC is minimal, but detectable at an estimated quantity of 0.03% of total absorbed PERC (Furuki *et al.* 2000). The

mechanism of urinary excretion of non-metabolised PERC is governed by simple diffusion according to Fick's law with first-order kinetics. Therefore, the renal excretion kinetics of urinary PERC generally follows the kinetics of blood elimination with an (apparent) very rapid elimination half-life of several minutes.
In humans, excretion of PERC metabolites makes up only a small percentage of the absorbed dose after exposure via inhalation: urinary excretion of TCA is estimated at 1-2% of the total absorbed dose (Chiu *et al.* 2007).

The main toxicokinetic parameters for PERC are summarised in Table 1.

Table 1- Summary of toxicokinetic parameters for PERC and its metabolites

	Estimated percentage of the absorbed dose	Time of peak concentration	Half-life
PERC in blood		**5.75 h** after the beginning of 6 h exposure period (Chiu *et al.* 2007)	Triphasic elimination with half-lives similar to those for PERC in exhaled air: **12-16 h** then **30-40 h** then **55-65 h** (Monster 1979)
PERC in urine	**0.03%** (Furuki *et al.* 2000)		Follows triphasic elimination pattern observed for PERC in blood
PERC in alveolar air	**80 - 100%** (Monster *et al.* 1979) **90 - 99%** (Chiu & Ginsberg 2011)		*__5 - 20 min__ (Chien 1997. IARC 2014) *__12 - 16 h__ (highly vascularised tissues +++) then **30 - 40 h** (muscles, skin, vascularised tissues +) then **55 - 65 h** (adipose tissues) (Monster *et al.* 1979) *__71.5 h__ (adipose tissues, body burden) (Guberan & Fernandez 1974) *__79 h__ (Benoit *et al.* 1985)
TCA in blood		*__20 - 50 h__ after the end of exposure (Monster *et al.* 1979) *__47 h__ after the beginning of exposure at 1 ppm (Chiu *et al.* 2007)	*__75 - 80 h__ (Monster *et al.* 1979) > 60h after exposure *__90 h__ (Monster *et al.* 1983)
TCA in urine	**1.8%** 64 h after exposure for 3 h at 87 ppm (Ogata *et al.* 1971) **2%** (Monster *et al.* 1979) **1.28%** (Furuki *et al.* 2000)	*__24 48 h__ after the end of exposure (Fernandez *et al.* 1976)	*__65 h__ (Monster *et al.* 1983) *__45.6 h__ (Volkel *et al.* 1998)
NAcTCVC in urine			**14.1 h** (Volkel *et al.* 1998)

Selection of biomarkers of exposure and effect

Biomarkers of exposure (BME)

The analysis of data from the literature identified six BMEs: PERC in blood, PERC in urine, PERC in exhaled air, TCA in blood, TCA in urine and NAcTCVC in urine.

A clear advantage of the three BMEs that involve measuring the unchanged PERC in blood, urine and exhaled air is their specificity and their good correlation with atmospheric exposure. The slow elimination kinetics of PERC from adipose tissue, associated with the long time to equilibrium in these tissues, allow PERC concentrations in blood and exhaled air to be used as indicators of cumulative exposure over several days, in particular if the sample is taken after the first rapid phase of pulmonary PERC elimination.

TCA and NAcTCVC are not specific to PERC exposure because they are also metabolites of other solvents (e.g. trichloroethylene, TCE). The degree of metabolisation of TCE into TCA is higher than that of PERC, because CYP seem to have higher affinity for TCA than PERC. Moreover, the concentrations of the metabolites (TCA and NAcTCVC) are affected by inter-individual variation in metabolic capacity or kinetics. Finally, correlations with exposure have not been frequently reported for TCA in blood and NAcTCVC in urine, or are weak for TCA in urine.

Therefore, **PERC in blood, urine and exhaled air were used as the relevant BMEs for the biomonitoring of occupational exposure to PERC.**

Biomarkers of effect

After acute or chronic exposure, PERC is a neurotoxin for humans. Although the neurotoxic effects can be assessed (symptomatic or neurological examination, neuro-behavioural tests), they are not specific to PERC exposure. Markers of kidney damage (increase in urine protein levels, e.g. albuminuria) or liver damage (e.g. increase in bilirubin or alkaline phosphatase levels in the blood) have been described following exposure to PERC (ATSDR 1997).

These effects described in the literature do not lend themselves to propose biomarkers of effect for the biomonitoring of occupational exposure.

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

Name	**Perchloroethylene in blood**
Other substances giving rise to this biomarker	None
Concentrations found in exposed workers or volunteers (with exposure levels and sampling times)	• **<u>Field studies</u>:** **- Lauwerys R. *et al.* (1983)** N = 26 workers from 6 dry-cleaning facilities [PERC-TWA]atmo = 20.8 ppm (8.9 - 37.5) (individual active sampling; NIOSH protocol; gas chromatography (GC)) ***[PERC in blood] Wed start of shift (SS) mean = 0.4 mg.L^{-1} (0.1 - 0.8); [PERC in blood] Wed 30 min after the end of shift (ES) = 1.2 mg.L^{-1} (0.4 - 3.1)** (analysis of PERC in blood using HS-GC) **- Skender *et al.* (1991)** N = 18 dry-cleaning workers [PERC-8h TWA] = 33-53 ppm (individual active sampling; GC)

***[PERC in blood] start of week and start of shift (SWSS)= 0.62 mg.L^{-1} (0.29 - 5.27)** (GC)

***[PERC in blood] Wed ES = 1.48 mg.L^{-1} (0.79 - 13.27)** (GC)

- Jang *et al.* (1993)

N = 13 Korean metal degreasers, 20-29 years old; 8 h workday

[PERC]atmo mean = 22.4 ppm (individual active sampling; NIOSH protocol; GC)

***[PERC in blood] EWSS mean = 0.85 (± 0.72) mg.L^{-1} (0.2-2.5)** (HS-GC-ECD)

- Gobba *et al.* (2003)

N = 26 workers from 7 dry-cleaning facilities; mean age: 40 years (± 14); average length of exposure: 11 years (± 12)

[PERC- 8h TWA]atmo mean = 6.4 ppm; median = 2.81 ppm (standard deviation (SD) 6.30) (individual passive sampling; GC)

***[PERC in blood] Wed ES mean = 0.7256 mg.L^{-1}; median = 0.3355 mg.L^{-1} (SD 0.9371)** (GC-ECD; LOD = 0.1 ng; N = 26)

- McKernan *et al.* (2008)

N = 18 female workers in dry-cleaning facilities with limited exposure to other solvents; mean age: 41 years

[PERC-TWA]atmo mean = 3.15 ppm (SD 4.51); geo. mean = 1.64 ppm (geo. SD 3.26) (individual active sampling over 2 consecutive days; NIOSH 1003 protocol; GC-FID; LOD = 0.0008 - 0.002 mg/sample)

***[PERC in blood] Thurs SS mean = 0.705 mg.L^{-1} (SD 0.1064); median = 0.0367 mg.L^{-1} (geo. SD 0.0334)** (purge-and-trap GC-MS; LOD = 0.02 ppb; n = 15)

- Furuki *et al.* (2000)

N = 44 workers in the textile degreasing sector; 8 h workday

[PERC-TWA]atmo geo. mean = 13 ppm; max value = 46 ppm (exclusion if co-exposure with TCE; individual passive sampling; GC-FID; LOD = 1 ppm)

***[PERC in blood] Wed, Thurs or Fri ES mean = 0.0011 mg.L^{-1}; max value = 0.0033 mg.L^{-1}** (HS-GC-ECD; LOD = 1 µg.L^{-1}; N = 54)

- Trevisan *et al.* (2000)

N = 40 dry-cleaning workers

[PERC]atmo mean = 8.65 ppm (individual passive sampling; HS-GC-ECD)

***[PERC in blood] Thurs ES mean = 0. 69 (± 0. 54) mg.L^{-1}; [PERC in blood] Friday SS mean = 0. 35 (± 0.28) mg.L^{-1}** (HS-GC-ECD)

- Toraason *et al.* (2003)

N = 18 female workers from 7 dry-cleaning facilities, < 70 years

[PERC-TWA]atmo Wed = 2.4 (± 3.4) ppm; Thurs = 3.8 (± 5.3) ppm (NIOSH 1003 protocol; GC-FID)

***[PERC in blood] Thurs SS mean = 0.075 (± 0.104) mg.L^{-1}** (GC-MS)

- Emara *et al.* (2010)

N = 40 male dry-cleaning workers (Egypt). 8 h workday

[PERCatmo] ≤ 140 ppm (colorimetric tubes; mean of 5 measurements)

***Non-smokers: [PERC in blood] mean = 1.681 (± 0.372) mg.L^{-1}; Smokers: [PERC in blood] mean = 1.695 (±0.454) mg.L^{-1}** (GC-ECD. LOD = 0.5 µg.L^{-1})

- Maccà *et al.* (2012)

N = 71 workers (42 females + 29 males) from 40 dry-cleaning facilities

[PERC]atmo mean = 7.58 ppm; median = 5.06 ppm (individual passive sampling; GC-ECD; LOD = 2 µg.L^{-1})

***[PERC in blood] Thurs ES mean = 0.617 mg.L^{-1}; median = 0.453 mg.L^{-1} (SD 0.519); [PERC in blood] Fri SS mean = 0.304 mg.L^{-1}; median = 0.266 mg.L^{-1} (SD 0.258)** (HS-GC-ECD; LOD = 2 µg.L^{-1}; N = 71)

- Lucas *et al.* (2015)

	N = 50 workers from 22 dry-cleaning facilities. Length of time badge worn/day: 3.25 h - 8 h [PERC]atmo mean = 7 ppm (0.22 - 33); median = 3.8 ppm (individual passive sampling; GC according to DFG) ***[PERC in blood] EWSS = 0.1259 mg.L^{-1} (0.0118 - 0.5440); median = 0.0736 mg.L^{-1}** (HS-GC-ECD according to DFG; LOD = 2 µg.L^{-1}; N = 49) • **<u>Studies on volunteers</u>:** **- Jang *et al.* (1997)** N = 12 male volunteers (6 Caucasians and 6 Asians) [PERC]atmo = 50 ppm for 6 h in an exposure chamber ***[PERC in blood] 30 min after the end of exposure Caucasian = 1.69 (± 0.35) mg.L^{-1}; Asian = 1.60 (± 0.47) mg.L^{-1}** (HS-GC)
Conversion factor	1 µmol.L^{-1} = 166 µg.L^{-1} 1 mg.L^{-1} = 6.0 µmol.L^{-1}
Concentrations in the general population[1]	N = 1458 subjects of 20-59 years old, non-occupationally exposed **[PERC in blood] 95th percentile = 0.13 µg.L^{-1}** [PERC in blood] mean not determined (high % of samples < LOD = 0.048 µg.L^{-1}) (NHANES IV (2007-2008); (CDC 2017) N = 590 subjects non-occupationally exposed **[PERC in blood] mean = 0.190 µg.L^{-1}** (Brugnone *et al.* (1993) cited in Ashley *et al.* (1996)) N = 248 subjects from the general population in Italy (107 rural subjects, 106 urban subjects and 35 urban workers potentially occasionally exposed to solvents) Analysis of PERCs using HS-GC-MS and cryogenic trapping techniques: **[PERC in blood] 95th percentile = 0.36 µg.L^{-1}** [PERCs] mean = 0.149 µg.L$^{-1;}$ median = 0.039 µg.L^{-1} (60% of samples > LOD) Rural subjects vs urban subjects vs urban subjects potentially exposed to solvents: [PERC in blood] = 0.062 µg.L^{-1} vs 0.263 µg.L^{-1} vs 0.231 µg.L^{-1} (Brugnone *et al.* 1994) N = 2453 subjects (1225 females + 1228 males). 12 - 79 years old **[PERC in blood] 95th percentile = 0.17 µg.L^{-1}** (0.10 - 0.23) (with 60.8% of samples < LOD = 0.02 µg.L^{-1}) (Canadian Health Measures Survey, Cycle 3 (2012-2013), Health Canada (2015a)) N =543, 20-39 years old **PERC in blood] 95th percentile = 0.15 µg.L^{-1}** (0.080 - 0.23) (with 60.04% of samples < LOD = 0.02 µg.L^{-1}) (Canadian Health Measures Survey, Cycle 3 (2012-2013), Health Canada (2015a)) N=587, 40 to 59 years **PERC in blood] 95th percentile = 0.13 µg.L^{-1}** (0.089 - 0.17) (with 65.08% of samples < LOD = 0.02 µg.L^{-1}) (Canadian Health Measures Survey, Cycle 3 (2012-2013), Health Canada (2015a))

[1]Or failing that, in a non-occupationally exposed control population; 95th percentile, or failing that the median or the mean (number of people in the study if this information is available)

	N = 218 subjects of a German cross-sectional study **[PERC in blood] 95th percentile = 0.38 µg.L-1** (with 42.7% of samples < LOD = 0.5 µg.L-1) (Angerer 2002)	
Recommended limit values for exposed workers	USA - ACGIH (BEI)	**0.5 mg.L-1** SS after at least 2 days of exposure (2015, last update 2008) (value based on a TLV-TWA of 25 ppm)
	Germany - DFG (EKA)	**At 10 ppm: 0.2 mg.L-1; 20 ppm: 0.4 mg.L-1; 30 ppm: 0.6 mg.L-1; 50 ppm: 1 mg.L-1**, 16 h after ES (2015, last update 2005) (BAT value no longer recommended since 1982: 1 mg.L-1 16 h ES)
	Finland - FIOH (BAL)	**0.2 mg.L-1** (1.2 µmol.L-1) Morning specimen before the work shift at the end of the working week or exposure period. In occasional exposure the specimen should be collected in the morning after the day of exposure (2015, last update)
	Switzerland - SUVA (VBT)	**1 mg.L-1** (6 µmol.L-1) SS (date of publication: 2016)
	Quebec - IRSST (IBE)	**0.5 mg.L-1** (3 µmol.L-1) before the last shift of a WW (2012) (value corresponds to the mean weighted exposure value of 25 ppm)
	EU - SCOEL (BLV)	**0.4 mg.L-1** before the last shift of a WW (2009, last update) (mean value corresponds to an 8h TWA exposure of 20 ppm)

*BAT= Biological Tolerance Value at the workplace; ES = end of shift; EWSS = end of week and start of shift; Fri = Friday; geo. Mean = geometric mean; LOD = limit of detection; SD = standard deviation; SS = start of shift; SWSS = start of week and start of shift; Thurs = Thursday; Wed = Wednesday; WW = work week.

Name	**Perchloroethylene in urine**
Other substances giving rise to these biomarkers	None
Concentrations found in exposed workers or volunteers (with exposure levels and sampling times)	• **<u>Field studies</u>:** **- Furuki *et al.* (2000)** N = 44 workers in the textile degreasing sector; 8 h workday [PERC-TWA]atmo geo mean = 13 ppm; max value = 46 ppm (exclusion if co-exposure with TCE; individual passive sampling; GC-FID; LOD = 1 ppm) ***[PERC in urine] ES geo. mean = 167 µg.L-1; max value = 422 µg.L-1** (HS-GC-ECD, LOD = 1 µg.L-1, N = 54) **- Trevisan *et al.* (2000)** N = 40 dry-cleaning workers [PERC]atmo mean = 8.65 ppm (individual passive sampling; HS-GC-ECD) ***[PERC in urine] Thurs ES = 32.4 (± 51.2) µg.g-1 creatinine; [PERC in urine] Fri SS = 9.1 (± 8.4) µg.g-1 creatinine** (HS-GC-ECD) **- Gobba *et al.* (2003)** N = 26 workers from 7 dry-cleaning facilities; mean age: 40 years (± 14) [PERC-8h TWA]atmo mean = 6.4 ppm; median = 2.81 ppm (SD 6.30) (individual passive sampling; GC) ***[PERC in urine] Wed ES mean = 29.8 µg.L-1; median = 20.2 µg.L-1 (SD 25.8)**

	(bladder emptied at midday; GC-ECD; LOD = 0.1 ng; N = 25) **- Poli *et al.* (2005)** N = 39 dry-cleaning workers, mean age: 37.2 years; [PERCatmo] not given ***[PERC in urine] median = 0.58 µg.L^{-1} (0.27-1.85)** (HS-SPME-GC-MS; LOD = 5 ng.L^{-1}) **- Rastkari *et al.* (2011)** N = 30 male dry-cleaning workers, 3 groups of 10 subjects according to washing machine capacity, non-smokers, mean age: 41 years (27-57 years) Analysis of [PERC]atmo using individual passive sampling and SPME-HS-GC-quadrupole MS (LOD = 20 ng.L^{-1}) Analysis of PERC in urine using HS-SPME-GC-quadrupole MS (LOD = 20 ng.L^{-1}): - 8 L washing machine: [PERC]atmo mean = 4.50 ppm (SD 2.00): ***[PERC in urine] SS = 6.58 µg.L^{-1} (SD 2.49); [PERC in urine] ES = 18.04 µg.L^{-1} (SD 7.28)** - 12 L washing machine: [PERC]atmo mean = 7.37 (SD 2.43): ***[PERC in urine] SS = 14.17 µg.L^{-1} (SD 4.40); [PERC in urine] ES = 36.77 µg.L^{-1} (SD 12.45)** - 18 L washing machine: [PERC]atmo mean = 17.53 (SD 2.94): ***[PERC in urine] SS = 21.95 µg.L^{-1} (SD 6.85); [PERC in urine] ES = 63.55 µg.L^{-1} (SD 13.80)** **- Maccà *et al.* (2012)** N = 71 workers (42 females + 29 males) from 40 dry-cleaning facilities [PERC]atmo mean = 7.58 ppm; median = 5.06 ppm (individual passive sampling; GC-ECD; LOD = 2 µg.L^{-1}) ***[PERC in urine] Thursday ES mean = 24 µg.L^{-1}; median = 15 µg.L^{-1} (SD 25); [PERC in urine] Friday SS mean = 12 µg.L^{-1}; median = 10 µg.L^{-1} (SD 8)** (HS-GC-ECD; LOD = 1 µg.L^{-1}; N = 71)
Conversion factor	1 µmol.L^{-1} = 166 µg.L^{-1} 1 mg.L^{-1} = 6.0 µmol.L^{-1}
Concentrations in the general population[2]	N = 120 subjects from the general population, mean age: 38.6 (± 6.6) years **[PERC in urine] mean = 0.08 µg.L^{-1}**; median = 0.05 µg.L^{-1} (0.01 - 0.70) (SD 0.11) (SPME-GC-MS; 68% of samples > LOD = 0.005 µg.L^{-1}) (Poli *et al.* 2005) N = 136 subjects from the general population in Italy (94 rural subjects, 42 urban subjects) Analysis of PERC in urine using HS-GC-MS and cryogenic trap techniques: **[PERC in urine] 95th percentile = 0.407 µg.L^{-1}** **[PERC in urine] mean = 0.110 µg.L^{-1}**; median = 0.016 µg.L^{-1} (76% of samples > LOD) Rural vs urban subjects: [PERC in urine] = 0.094 µg.L^{-1} vs 0.042 µg.L^{-1} (Brugnone *et al.* 1994)
Recommended limit values for exposed workers	ND

*ES = end of shift; Fri = Friday; ND = not determined; SS = start of shift; Thurs = Thursday; Wed = Wednesday

[2] Or failing that, in a non-occupationally exposed control population; 95th percentile, or failing that the median or the mean (number of people in the study if this information is available

Name	Perchloroethylene in exhaled air
Other substances giving rise to these biomarkers	None
Concentrations found in exposed workers or volunteers (with exposure levels and sampling times)	• **<u>Field studies</u>:** **- Lauwerys *et al.* (1983)** N = 26 workers from 6 dry-cleaning facilities [PERC-TWA]atmo = 20.8 ppm (8.9 - 37.5) (individual active sampling; NIOSH protocol; HS-GC; N = 26) ***[PERC in exhaled air] Wed SS = 1.9 ppm (0.1 - 5.5); [PERC in exhaled air] Wed ES = 5.1 ppm (0.2 - 10)** (HS-GC; N = 26) **- Solet *et al.* (1990)** N = 195 workers from 13 dry-cleaning facilities (12 shops + 1 industrial facility) [PERCatmo]: passive sampling according to the NIOSH S335 protocol with direct-injection GC [PERC in exhaled air]: analysis of mixed-exhaled air in 12 L Saran bags, after at least 3 h of activity, not on Mon or Fri if possible - Non-operators: [PERC-8 h TWA]atmo = 12.85 (± 12.8) ppm (n = 27): ***[PERC in exhaled air] mean = 4.94 (± 4.7 ppm) (n = 95)** - Operators: [PERC-8 h TWA]atmo = 46.50 (± 34.1) ppm (n = 12): ***[PERC in exhaled air] mean = 12.45 (± 9.7 ppm) (n = 11)** **- Aggazzotti *et al.* (1994)** N = 60 workers from 26 dry-cleaning facilities; 2 exposure measurement methods: 1) sampling every hour in glass tubes (6 to 8 samples/d) then GC-ECD (LOD = 1 $mg.m^{-3}$): [PERC]atmo median = 0.087 - 10.87 ppm according to dry-cleaning facility 2) Individual badges changed in the middle of the day; NIOSH protocol; GC-ECD (LOD = 3.5 $mg.m^{-3}$): [PERC-8 h TWA]atmo mean= 0.38 - 32.10 ppm according to dry-cleaning facility (N = 52 full-time workers) ***[PERC in exhaled air] mean = 15.42 $mg.m^{-3}$; median = 16.45 $mg.m^{-3}$ (0.49 - 353)** (glass tubes, GC-ECD, LOD = 1 $mg.m^{-3}$) **- Gobba *et al.* (2003)** N = 26 dry-cleaning workers [PERC-8 h TWA]atmo mean = 6.4 ppm; median = 2.81 ppm (SD 6.30) (individual badges; GC) ***[PERC in exhaled air] mean = 7.7 ppm (44 $mg.m^{-3}$); median = 2.3 ppm (19 $mg.m^{-3}$) (SD: 11.9 ppm)** (glass tubes; GC; LOD = 0.1 ng; N = 26) **- McKernan *et al.* (2008)** N = 18 dry-cleaning workers [PERC-TWA]atmo mean = 3.15 ppm (SD 4.51); geo. mean = 1.64 ppm (geo. SD 3.26) (individual active sampling taken over 2 consecutive days; NIOSH 1003 protocol; GC-FID; LOD = 0.0008 - 0.002 mg/sample) ***[PERC in exhaled air] tot SS mean = 0.51 ppm (SD 0.37); geo. mean = 0.51 ppm (geo. SD 0.37)** (N = 51) ***[PERC in exhaled air] tot ES mean = 1.21 ppm (SD 0.87); geo. mean = 0.87 ppm (geo. SD 2.51)** (N = 45) **- Azimi Pirsaraei *et al.* (2009)** N = 179 workers from 69 dry-cleaning facilities [PERC-8 h TWA]atmo: individual active sampling; NIOSH protocol; GC-FID; [PERC in exhaled air]: air exhaled normally in a 1 L Tedlar bag; GC-FID

	- Machine operators (N = 71): [PERC-TWA]atmo = 11.5 (± 16.9) ppm ***[PERC in exhaled air] SWSS = 1.7 (± 2.5) ppm; [PERC in exhaled air] EWES = 2.4 (± 3.4) ppm** - Ironers (N = 63): [PERC-TWA]atmo = 9.6 (± 20.4) ppm ***[PERC in exhaled air] SWSS = 1.5 (± 3.0) ppm; [PERC in exhaled air] EWES = 2.0 (± 4.1) ppm** - Reception workers (N = 45): [PERC-TWA]atmo = 7.2 (± 11.9) ppm ***[PERC in exhaled air] SWSS = 1.1 (± 1.7) ppm; [PERC in exhaled air] EWES = 1.5 (± 2.5) ppm** • **<u>Studies in volunteers</u>:** **- Jang *et al.* (1997)** N = 12 male volunteers (6 Caucasians + 6 Asians); [PERCatmo] = 50 ppm for 6 h in an exposure chamber ***[PERC in exhaled air] ES Asians = 8.3 (± 0.9) ppm; Caucasians = 9.5 (± 1.7) ppm** ***[PERC in exhaled air] SS following workday Asians = 1.27 (± 0.15) ppm; Caucasians = 1.33 (± 0.23) ppm**	
Conversion factor	1 µmol.L^{-1} = 166 µg.L^{-1} 1 mg.L^{-1} = 6.0 µmol.L^{-1}	
Concentrations in the general population[3]	N = 54 healthy volunteers, from an urban population in Chicago **[PERC in exhaled air] mean = 0.0026 µg.m^{-3} = 0.37 ppb** (traces of PERC in exhaled air for 30.2% of subjects) (Krotoszynski *et al.* 1979) N = 300 New Jersey residents **[PERC in exhaled air] mean = 13.3 µg.m^{-3} = 1.92 ppb** (detection in 93% of samples of exhaled air) (Wallace (1986) cited in ATSDR (1997)) N = 10 adults living near a factory or a waste disposal site **[PERC in exhaled air] mean = 7.8 µg.m^{-3} = 1.13 ppb**; [PERC in exhaled air] max = 4 ppb (Monster & Smolders (1984) cited in ACGIH (2009))	
Recommended limit values for exposed workers	USA - ACGIH (BEI)	**3 ppm** (0.435 mg.m^{-3}) in end-exhaled air, pre-shift after at least 2 days of exposure (2015, last update 2008)
	Germany - DFG (BAT, EKA)	Value dating from 1982: 9.5 ppm (64 mg.m^{-3}), no longer recommended since 1983
	EU - SCOEL (BLV)	**3 ppm** (0.435 mg.m^{-3}) in end-exhaled air, prior to the last shift of a work-week (2009. last update)

ES = end of shift EWSS = end of week and start of shift; Fri = Friday; geo. = geometric standard deviation ; GC = gas chromatography; geo. SD = geometric ; HS = headspace; LOD: limit of detection; Mon = Monday; N = sample size; SD = standard deviation; SS = start of shift; SWSS = start of week and start of shift; Thurs = Thursday; TWA = time weighted average; Wed = Wednesday; WW = workweek

<u>Study of the relationship between concentrations of biomarkers of exposure and health effects</u>

[3] Or failing that, in a non-occupationally exposed control population; 95th percentile, or failing that the median or the mean (number of people in the study if this information is available

According to the analysis of the available scientific literature, no correlations between the biological concentrations of the three selected BMEs (PERC in blood, in urine or exhaled air) and health effects were identified.

Study of the relationship between concentrations of biomarkers of exposure and atmospheric concentration

Several studies have examined the relationship between concentrations of BMEs of PERC and atmospheric concentrations of PERC. The main data on BMEs considered relevant/valuable are shown in the tables below.

PERC in blood

Equation linking exposure to PERC concentration in blood (with sampling time)	Calculation of **[PERC in blood]** for exposure at the 8h-OEL (20 ppm; 138 $mg.m^{-3}$)	Reference
Field studies		
Blood sampled end of shift		
[PERC in blood] end of shift Wed ($\mu g.L^{-1}$) = -93.918 + 27.322 [PERC]atmo ($mg.m^{-3}$) (1) ([PERC]atmo mean = 6.4 ppm; r = 0.938; p < 0.001; N = 20)	**3676 µg.L^{-1}**	Gobba *et al.* (2003)
[PERC in blood] end of shift after at least 2 days of exposure ($\mu g.L^{-1}$) = 331 + 51.5 [PERC]atmo (ppm) (1) ([PERC]atmo mean =13 ppm; r = 0.770; p < 0.01; N = 54)	**1361 µg.L^{-1}**	Furuki *et al.* (2000)
[PERC in blood] end of shift Thurs ($\mu g.L^{-1}$) = 295.28 + 5.96 [PERC]atmo ($mg.m^{-3}$) (1) ([PERC]atmo mean = 7.7 ppm; r = 0.68; p < 0.001; N = 71)	**1118 µg.L^{-1}**	Maccà *et al.* (2012)
ln [PERC in blood] 15-30 min after the end of shift and the end of week (µmol.L-1) = -7.03 + 1.26 ln [PERC-WW TWA] (µmol.m^{-3}) (2) ([PERC]atmo = 1.6 - 159.4 ppm; r^2 = 0.953; r = 0.976; p = 0.05; N = 21)	**701 µg.L-1**	Monster *et al.* (1983)
ln [PERC in blood] end of shift and end of week (µmol.L-1) = (-5.66 + ln [PERC- WW TWA] (µmol.m^{-3})) / 0.756 (2) ([PERC]atmo = 1.6 - 159.4 ppm; r^2 = 0.953; r = 0.976; p = 0.05; N = 21)	**673.7 µg.L-1**	Monster *et al.* (1983)
Blood sampled start of shift		
[PERC in blood] start of shift Fri ($\mu g.L^{-1}$) = 145.17 + 3.03 [PERC]atmo ($mg.m^{-3}$) (1) ([PERC]atmo mean = 7.7 ppm; r = 0.70; p < 0.001; N = 71) *Average workday: 8-9 h (5-6 d/week)*	**563.3 µg.L^{-1}**	Maccà *et al.* (2012)
[PERC in blood] start of shift and end of week ($\mu g.L^{-1}$) = 44.44 + 1.6904 [PERC]atmo ($mg.m^{-3}$) (3) (r = 0.64; p < 0.01; N = 49) and temporal adjustment for median daily exposure of 5.75 h and not 8h *Median daily workday length on the day badges were worn: 5.75 h*	**386.4 µg.L^{-1}**	Lucas *et al.* (2015)
ln [PERC in blood] start of shift and start of week ($\mu mol.L^{-1}$) = (-6.15 + ln [PERC-WW TWA] ($\mu mol.m^{-3}$)) / 0.941 (2) ([PERC]atmo = 1.6 - 159.4 ppm; r^2 = 0.786; r = 0.887; p = 0.05; N = 19) *Daily workday length: 8 h* *Note: study conducted on metal degreasing workers*	**294 µg.L^{-1}**	Monster *et al.* (1983)
[PERCs] start of shift and end of week ($mg.L^{-1}$) = 0.277 + 0.0258 [PERC]atmo (ppm) (4) ([PERC]atmo mean = 7 ppm; r = 0.825; p < 0.001; N = 13)	**793 µg.L^{-1}**	Jang *et al.* (1993)

Daily workday length: 8 h		
[PERCs] start of shift and end of week (after 3 days of exposure) for an individual atmospheric exposure (active charcoal tubes) of 20.8 ppm (N = 26)	**400 µg.L^{-1}**	Lauwerys *et al.* (1983)

(1) Passive measurement of PERCatmo over 8 h; (2) Active measurement of PERCatmo over 5 days, for 4 - 6 h/d; (3) Passive measurement of PERCatmo over 3.25 - 8 h; (4) Active measurement of PERCatmo over 8 h

PERC in urine

Equation linking exposure to **PERC concentration in urine** (with sampling time)	Calculation of **[PERC in urine]** for exposure at the 8h-OEL (20 ppm; 138 mg.m^{-3})	Reference
Field studies		
[PERC in urine] end of shift Wed (8 h of exposure; 4 h urine collection) (µg.L^{-1}) = 16.411 + 0.303 [PERC]atmo (mg.m^{-3}) (1) ([PERC]atmo mean = 6.4 ppm; r = 0.667; p < 0.001; N = 26)	**58.2 µg.L^{-1}**	Gobba *et al.* (2003)
[PERC in urine] noon (4 h of exposure; 4 h urine collection) (µg.L^{-1}) = 12 + 0.33 [PERC]atmo (mg.m^{-3}) (2) ([PERC]atmo median = 9.5 ppm; r = 0.88; p = ND; N = 55)	**57.5 µg.L^{-1}**	Imbriani *et al.* (1988)
[PERC in urine] noon (4 h of exposure; 4 h urine collection) (µg.L^{-1}) = 11.9 + 0.33 [PERC]atmo (mg.m^{-3}) (1) ([PERC]atmo = 0-70 ppm; r = 0.87; p = ND; N = 40)	**57.4 µg.L^{-1}**	Ghittori *et al.* (1987)
[PERC in urine] end of shift (8 h of exposure) (µg.L^{-1}) = 67.5 + 7.75 [PERC]atmo (ppm) (1) ([PERC]atmo mean = 13 ppm; r = 0.722. p < 0.01. N = 54)	**222.5 µg.L^{-1}**	Furuki *et al.* (2000)
[PERC in urine] end of shift Thurs (8 h of exposure) (µg.L^{-1}) = 8.74 + 0.29 [PERC]atmo (mg.m^{-3}) (1) ([PERC]atmo mean = 7.6 ppm; r = 0.68; p < 0.001; N = 71)	**48.8 µg.L^{-1}**	Maccà *et al.* (2012)

(1) Passive measurement of PERCatmo over 8 h; (2) Passive measurement of PERCatmo over 4 h

PERC in exhaled air

Equation linking exposure to **PERC concentration in exhaled air** (with sampling time)	Calculation of **[PERC in exhaled air]** for exposure at the 8h-OEL (20 ppm; 138 mg.m^{-3})	Reference
Field studies		
Exhaled air sampled end of shift		
[PERC in exhaled air] end of shift Wed (mg.m^{-3}) = 1.681 + 1.172 [PERC]atmo (mg.m^{-3}) (1) * ([PERC]atmo mean = 6.4 ppm; r = 0.808. p < 0.001. N = 26)	**23.7 ppm (163.42 mg.m^{-3})**	Gobba *et al.* (2003)
Log [PERC in exhaled air] end of shift 2nd or 3rd day (mg.m^{-3}) = 0.31 + 0.783 log [PERC]atmo (mg.m^{-3}) (2) *	**14.0 ppm (96.6 mg.m^{-3})**	Aggazzotti *et al.* (1994)

([PERC]atmo median = 2.2 ppm; r = 0.758; p < 0.001; N = 49)		
[PERC in exhaled air] end of shift and end of week (ppm) = 0.072 + 0.201 [PERC]atmo (ppm) (3) ** ([PERC]atmo mean = 7.2 - 11.5 ppm; r = 0.98; p < 0.001; N = 179)	**4.1 ppm (28.2 mg.m^{-3})**	Azimi Pirsaraei *et al.* (2009)
ln [PERC in exhaled air] end ofshift Wed/Fri (15-30 min after the end of the shift) (µmol.m^{-3}) = -2.79 + 1.345 ln [PERC-TWA-4h]atmo (µmol.m^{-3}) (4) *** ([PERC]atmo mean = 7.2 - 11.5 ppm; r^2 = 0.927; r = 0.963; N = 63	**1.2 ppm (50.9 µmol.m^{-3}; 8.5 mg.m^{-3})**	Monster *et al.* (1983)
ln [PERC in exhaled air] end of shift and end of week (µmol.m^{-3}) = (-2.78 + ln [PERC-WW TWA] (µmol.m^{-3})) / 0.708 (4) *** ([PERC]atmo mean =1.6 to 159.4 ppm; r^2 = 0.931; r = 0.965; p < 0.05; N = 21)	**6.3 ppm (260.7 µmol.m^{-3}; 43.3 mg.m^{-3})**	Monster *et al.* (1983)
ln [PERC in exhaled air] end of shift = ß0 + ß2 (BMI) + ß3 ln [PERC]atmo **ln [PERC in exhaled air] end of shift = -0.46 + 0.042*28 + 0.52 ln [PERC]atmo** (r^2 = 0.61)	**9.7 ppm (66.9 mg.m^{-3})**	McKernan *et al.*, (2008)
[PERC in exhaled air] end of shift and end of week (15-30 min after the end of shift on day 3) for an individual atmospheric exposure (active charcoal tubes) of 20.8 ppm (N = 26)	**5.1 ppm (35.0 mg.m^{-3})**	Lauwerys *et al.*, (1983)
Exhaled air sampled start of shift		
[PERC in exhaled air] start of shift and start of week (ppm) = 0.031 + 0.147 [PERC]atmo (ppm)(3) ** (r = 0.99; p < 0.001; N = 179)	**3.0 ppm (20.5 mg.m^{-3})**	Azimi Pirsaraei *et al.* (2009)
ln [PERC in exhaled air] start of shift and start of week (µmol.m^{-3}) = (-2.30 + ln [PERC-WW TWA] (µmol.m^{-3})) / r²=0.927 (4) *** (r^2 = 0.85; r = 0.921; p < 0.05; N = 20)	**2.0 ppm (83 µmol.m^{-3}; 13.8 mg.m^{-3})**	Monster *et al.* (1983)
ln [PERC in exhaled air] start of shift= ß0 + ß1 (Day) + ß2 (BMI) + ß3 ln [PERC]atmo **ln [PERC in exhaled air] start of shift = -0.42 + 0.15*Day + 0.028*28 + 0.15 ln [PERC]atmo** (r^2 = 0.47)	**0.96 ppm (6.6 mg.m^{-3})**	McKernan *et al.* (2008)
[PERC in exhaled air] start of shift (15 h after the end of shift) (ppm adjusted to 5.5% of CO_2) = 0.817 + 0.022 [PERC]atmo (ppm.h) (5)**** (r = 0.96; N = 38) Adjusted for exposure at 20 ppm for 8 h	**4.3 ppm (29.7 mg.m^{-3})**	Droz & Guillemin (1986)

(1) Passive measurement of PERCatmo over 8 h; (2) Passive measurement of PERCatmo over 4 h; (3) Active measurement of PERCatmo over 8 h; (4) Active measurement of PERCatmo over 5 days for 4 - 6 h/d; (5) Individual measurement method of PERCatmo not detailed

* End-exhaled air, sampled in glass tubes; ** End-exhaled air, sampled in Tedlar™ bags; *** End-exhaled air after holding breath for 5 s, sampled in glass tubes; ****Forced exhaled air, sampled in Tedlar™ bags

BMI: Body Mass Index

Establishment of BLVs and choice of biological reference values

In the absence of carcinogenic mechanisms of action elucidated in humans and sufficiently robust quantitative data to establish a dose-response curve, the 8h-OEL recommended by the OEL Committee (20 ppm or 138 mg.m^{-3}) is based on another type of effect for PERC. Neurotoxicity, the most sensitive effect caused by PERC exposure, was chosen to set the 8h-OEL values.

Field studies have failed to establish a dose-response relationship between the concentrations of PERC in blood, PERC in urine or PERC in exhaled air and neurotoxic effects. Therefore, establishment of the BLVs based on exposure at the 8h-OEL drew on studies linking atmospheric concentrations of PERC to biological concentrations of the selected BMEs.

PERC in blood

PERC in blood is a specific indicator, and correlates well with atmospheric concentrations, even for levels clearly lower than the 8h-OEL of 20 ppm.

The PERC in blood measured at the end of shift and end of the workweek was not chosen despite its very good correlation with atmospheric concentrations because of the rapid decline in blood concentrations after the end of the shift; this would require sampling immediately at the end of the shift, which is not easily practicable in the field. Furthermore, samples taken a half-hour or 1 h after the end of the shift would run the risk of manifestly underestimating concentrations at the end of the shift.

PERC in blood measured at the end of the week and start of the shift (i.e. at the beginning of the last workshift of the workweek or 16 h after the end of the last shift) has also shown a very good correlation with atmospheric levels of PERC and lower variability in concentrations if sampling is not done exactly at the ideal time. The field studies of Maccà *et al*. (2012), Lucas *et al*. (2015), Jang *et al*. (1993), and Lauwerys *et al*. (1983) make it possible to estimate a concentration of 535 µg.L^{-1} of PERCs resulting from an exposure to the OEL-8h (i.e. 20-ppm), by averaging the concentrations obtained from each study. The Monster *et al.* (1983) study was excluded due to the sampling time considered (start of week and start of shift).

The BLV based on the 8h-OEL of 20 ppm recommended by the Committee for PERC in blood based on sampling at the end of week and start of shift is thus rounded to 500 µg.L^{-1}.

<u>Proposition of biological reference values</u>

There are no French data reporting PERC blood levels on large sample sizes in the general population.

The blood samples collected in 2007-2008 for the American NHANES study (n=1482) give a value for the 95th percentile of the distribution of PERC blood concentrations in men and women aged from 20 to 59 years equal to 0.102 µg.L^{-1} (NHANES IV (2007-2008), CDC (Centers for Disease Control and Prevention (2017)).

Other studies in the general population (Brugnone *et al.* (1993 &1994), Health Canada (2015), Angerer (2002)) provide 95th percentiles for PERC concentrations between 0.17 and 0.38 µg.L^{-1}. Based on the Health Canada study data, the 95th percentile for PERC concentrations of 0.14 µg.L^{-1} was determined for the 20-59 year-old age class (n=1130).

Considering the mean of the 95th percentiles of the two general population studies with the largest sample sizes (CDC and Health Canada), the blood concentration of **0.12 µg.L^{-1} for PERC in blood is recommended as the BRV.**

PERC in urine

There is a correlation between atmospheric and urinary PERC concentrations even at exposure levels lower than the 8h-OEL.

The urinary PERC concentration in a sample taken at the end of week and end of shift is proposed as a BME. The Furuki *et al.* (2010) study was excluded due to the high value for the y-intercept of the regression line equation, leading to a BLV very different from the other selected studies. The field studies of Gobba *et al.* (2003), Imbriani *et al.* (1988), Ghittori *et al.* (1987) and Maccà *et al.* (2012) give similar results and lead to an estimate of the PERC concentration in urine after exposure at the 8h-OEL (i.e. 20 ppm) of **55 µg.L^{-1}** by calculating the average of the concentrations from each study**.**

The BLV based on the 8h-OEL of 20 ppm recommended by the Committee for PERC in urine from a sample taken at the end of the week and end of shift is thus rounded to 50 µg.L^{-1}.

Proposition of biological reference values

There are no French data reporting PERC urine levels on large sample sizes in the general population[4]. The Brugnone *et al.* (1994) study, carried out on 136 Italian subjects (including 94 rural and 42 urban subjects), was thus selected to set the BRV.

The 95th percentile of the distribution of urinary PERC concentrations in the subjects of this study was **0.41 µg.L^{-1}**.

The concentration of 0.40 µg.L^{-1} for PERC in urine is thus the recommended BRV.

PERC in exhaled air

The PERC in exhaled air measured at the end of the week and end of the shift was not chosen to recommend a BLV or BRV despite its very good correlation with atmospheric levels because of the rapid decline in exhaled air concentrations after the end of shift; this would require sampling immediately at the end of the shift, which is not easy to implement in the field. Furthermore, samples taken a half-hour or 1 h after the last shift would run the risk of manifestly underestimating the end of shift concentrations.

The exhaled PERC concentrations in a sample of exhaled air collected at the end of the week and the start of the shift is a good candidate as BME. Nevertheless, this indicator was not selected to recommend a BLV or BRV due to:

- the low number of studies available and the discrepancies among the results obtained in these studies (exhaled PERC concentrations differ by a factor of 1 to 4.5 for an exposure at 20 ppm);
- lack of a standard sampling protocol and the practical difficulties of carrying it out (specific material, training workers in the exhalation technique used to sample alveolar air) and storing the samples prior to analysis.

Therefore, no BLV or BRV can be recommended for exhaled PERC concentrations.

[4] No results for BME assays of perchloroethylene in French national surveys (ENNS and Esteban).

Conclusions of the collective expert appraisal

The biological values proposed for monitoring occupational exposure to PERC are:

Blood PERC at the end of week and start of shift:

BLV based on a health effect	None
BLV based on an 8h-OEL exposure (20 ppm or 138 mg.m^{-3})	500 µg.L^{-1}
Biological reference value	0.12 µg.L^{-1}

Urinary PERC at the end of week and end of shift:

BLV based on a health effect	None
BLV based on an 8h-OEL exposure (20 ppm or 138 mg.m^{-3})	50 µg.L^{-1}
Biological reference value	0.40 µg.L^{-1}

Sampling method and factors that may affect the interpretation of results

Blood perchloroethylene

Samples must be taken prior to the workshift, outside of the workplace. The samples must be taken in vacuum blood collection tubes containing heparin or EDTA, filled to their maximum capacity. The samples are to be immediately transferred to a glass tube with a Teflon® stopper (for solvent analysis) and sealed and stored between +2 and +8°C until analysis (ACGIH 2009). The samples can be stored for 5 days at 4°C (UCL – LTAP[5]).

Urinary perchloroethylene

Samples must be taken outside of the workplace. To prevent the risk of external biological contamination of the sample, it is recommended that the subject wash his/her hands, take a shower and change his/her clothes before sample collection. The samples are to be collected in polypropylene containers. The minimum volume is 10 mL. The samples must be maintained between °2 and +8°C and ideally frozen (-20°C) during storage and transport.

Absorption of PERC is influenced by physical activity (by a factor of 3 compared with the resting state) as well as the body mass index.

Biological samples must be taken after several weeks of exposure, to allow equilibrium to be reached between absorption, storage in fatty tissues and elimination.

[5] Louvain Centre for Toxicology and Applied Pharmacology at the Université catholique de Louvain avalaible at http://www.toxi.ucl.ac.be/biological_monitoring/biomarqueur/1083

Biometrology

Some analytical methods described in the literature have been listed and are shown in the table below for the relevant BMEs. The objective of this section is not to recommend a measurement method, but to provide information on certain characteristics of the analysis methods.

PERC IN BLOOD			
Analytical methods			
	Method 1	**Method 2**	**Method 3**
Analytical technique	**Headspace gas chromatography coupled with electron capture detection (HS-GC-ECD)**	**Gas chromatography coupled with electron capture detection (GC-ECD)**	**Purge and trap gas chromatography with mass spectrometry detection (Purge & trap-HR-GC-MS)**
References	Jang *et al.* (1993); Monster *et al.* (1983); Furuki *et al.* (2000); Maccà *et al.* (2012); Trevisan *et al.* (2000); Emara *et al.* (2010); Lucas *et al.* (2015)	Gobba *et al.* (2003)	McKernan *et al.* (2008)
Limit of detection	2 $\mu g.L^{-1}$ (Jang *et al.* (1993), Maccà *et al.* (2012), Lucas *et al.* (2015)) 0,5 $\mu g.L^{-1}$ (Emara *et al.* (2010))	0.1 $ng.mL^{-1}$ or 0.1 $\mu g.L^{-1}$	0.02 ppb or 0.02 $\mu g.L^{-1}$
Fidelity		CV: 1.5-4.5%	

PERC IN URINE			
Analytical methods			
	Method 1	**Method 2**	**Method 3**
Analytical technique	**Headspace gas chromatography coupled with mass spectrometric detection (HS-GC-MS)**	**Gas chromatography coupled with electron capture detection (GC-ECD)**	**Headspace gas chromatography coupled with electron capture detection (HS-GC-ECD)**
References	Imbriani *et al.* (1988); Ghittori *et al.* (1987)	Gobba *et al.* (2003)	Furuki *et al.* (2000), Trevisan *et al.* (2000); Maccà *et al.* (2012)
Limit of detection	0.5 $\mu g.L^{-1}$ (Imbriani *et al.* (1988)) 0.02 $\mu g.L^{-1}$ (Rastkari *et al.* (2011))	0.1 $\mu g.L^{-1}$	1 $\mu g.L^{-1}$
Fidelity			
Precision			
Reference standard			

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