COLLECTIVE EXPERT APPRAISAL: SUMMARY AND CONCLUSIONS

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

Assessment of health effects and methods for the measurement of exposure levels in workplace atmospheres for

n-butylamine (CAS No. 109-73-9)

This document summarises the work of the Expert Committee on expert appraisal for recommending occupational exposure limits for chemical agents (OEL Committee) and the Working groups on health effects and on metrology.

Presentation of the issue

On 12 June 2007, AFSSET, which became ANSES in July 2010, was requested by the Directorate General for Labour to conduct the expert appraisal work required for establishing recommendations on measures to be taken in the event of specific exposure profiles such as those with peaks.

A first report[1] published in June 2009 issued recommendations on measures to be taken in the event of an 8h-OELV with no short-term exposure limit (STELV).

In 2010, ANSES published a second report that recommended studying the 36 substances in France with a short-term exposure limit but no time-weighted average (TWA) to recommend health values taken from the most recent scientific literature (ANSES, 2010).

In this context, an assessment was undertaken for n-butylamine, which has a short-term exposure limit in France set at 15 $mg.m^{-3}$ in a Circular[2] of 1982 but no TWA.

Scientific background

The French system for establishing OELVs has three clearly distinct phases:

- Independent scientific expertise (the only phase entrusted to ANSES);
- Proposal by the Ministry of Labour of a draft regulation for the establishment of limit values, which may be binding or indicative;
- Stakeholder consultation during the presentation of the draft regulation to the French Steering Committee on Working Conditions (COCT). The aim of this phase is to discuss the effectiveness of the limit values and if necessary to determine a possible implementation timetable, depending on any technical and economic feasibility problems.

[1] http://www.anses.fr/ET/DocumentsET/VLEP_Picsdexpo_Avis_0906.pdf
[2] Circular of 19 July 1982 on the acceptable values for concentrations of certain hazardous substances in workplace atmospheres.

The organisation of the scientific expertise phase required for the establishment of Occupational Exposure Limits (OELVs) was entrusted to AFSSET in the framework of the 2005-2009 Occupational Health Plan (PST) and then to ANSES after AFSSET and AFSSA merged in 2010.

The OELs, as proposed by the Committee on expert appraisal for recommending occupational exposure limits for chemical agents (OEL Committee), are concentration levels of pollutants in workplace atmospheres that should not be exceeded over a determined reference period and below which the risk of impaired health is negligible. Although reversible physiological changes are sometimes tolerated, no organic or functional damage of an irreversible or prolonged nature is accepted at this level of exposure for the large majority of workers. These concentration levels are determined by considering that the exposed population (the workers) is one that excludes both children and the elderly.

These concentration levels are determined by the OEL Committee experts based on information available from epidemiological, clinical, animal toxicology studies, etc. Identifying concentrations that are safe for human health generally requires adjustment factors to be applied to the values identified directly by the studies. These factors take into account a number of uncertainties inherent to the extrapolation process conducted as part of an assessment of the health effects of chemicals on humans.

The Committee recommends the use of three types of values:

- 8-hour occupational exposure limit (8h-OEL): this corresponds to the limit of the time-weighted average (TWA) of the concentration of a chemical in the worker's breathing zone over the course of an 8-hour work shift. In the current state of scientific knowledge (toxicology, medicine, epidemiology, etc.), the 8h-OEL is designed to protect workers exposed regularly and for the duration of their working life from the medium- and long-term health effects of the chemical in question;
- Short-term exposure limit (STEL): this corresponds to the limit of the time-weighted average (TWA) of the concentration of a chemical in the worker's breathing zone over a 15-minute reference period during the peak of exposure, irrespective of its duration. It aims to protect workers from adverse health effects (immediate or short-term toxic effects such as irritation phenomena) due to peaks of exposure;
- Ceiling value: this is the limit of the concentration of a chemical in the worker's breathing zone that should not be exceeded at any time during the working period. This value is recommended for substances known to be highly irritating or corrosive or likely to cause serious potentially irreversible effects after a very short period of exposure.

These three types of values are expressed:

- either in $mg.m^{-3}$, i.e. in milligrams of chemical per cubic metre of air and in ppm (parts per million), i.e. in cubic centimetres of chemical per cubic metre of air, for gases and vapours;
- or in $mg.m^{-3}$, only for liquid and solid aerosols;
- or in $f.cm^{-3}$, i.e. in fibres per cubic centimetre for fibrous materials.

The 8h-OELV may be exceeded for short periods during the working day provided that:

- the weighted average of values over the entire working day is not exceeded;
- the value of the short term limit value (STEL), when it exists, is not exceeded.

In addition to the OELs, the OEL Committee assesses the need to assign a "skin" notation, when significant penetration through the skin is possible (ANSES, 2014a). This notation

indicates the need to consider the dermal route of exposure in the exposure assessment and, where necessary, to implement appropriate preventive measures (such as wearing protective gloves). Skin penetration of substances is not taken into account when determining the atmospheric limit levels, yet can potentially cause health effects even when the atmospheric levels are respected.

The OEL Committee assesses the need to assign an “ototoxic” notation indicating a risk of hearing impairment in the event of co-exposure to noise and the substance below the recommended OELs, to enable preventionists to implement appropriate measures (collective, individual and/or medical) (ANSES, 2014a).

The OEL Committee also assesses the applicable reference methods for the measurement of exposure levels in the workplace. The quality of these methods and their applicability to the measurement of exposure levels for comparison with an OEL are assessed, particularly with regards to their compliance with the performance requirements in the NF-EN 482 Standard and their level of validation.

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 health effects to conduct the expert appraisal work on health effects;
- The working group on metrology to assess measurement methods in workplace atmospheres.

The methodological and scientific aspects of the work of this group were regularly submitted to the Expert 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

For the assessment of health effects:

A summary report on the health effects of n-butylamine was prepared by the working group on health effects and submitted to the OEL Committee, which commented on it and added to it.

The summary report was based on bibliographic information taking into account the scientific literature that had been published on this substance up to 2013. The literature search was carried out using the summary report written by ACGIH[3] (2001) and the Health Council of the Netherlands (2003) and articles found in the Medline, Toxline and HSDB (ToxNet) databases.

For assessment of methods for measuring exposure levels in workplace:

A summary report was prepared by the working group on metrology and submitted to the OEL Committee, which added its own comments.

The summary report presents the various protocols for measuring n-butylamine in workplace atmospheres grouped together based on the methods they use. These methods were then assessed and classified based on the performance requirements set out particularly in the French Standard NF EN 482: "Workplace atmospheres - General requirements for the performance of procedures for the measurement of chemical agents" and the decision-making criteria listed in the methodology report (ANSES, 2014a).

A list of the main sources consulted is detailed in the methodology report (ANSES, 2014a).

These methods were classified as follows:

- Category 1A: the method has been recognized and validated (all of the performance criteria in the NF-EN 482 Standard are met);
- Category 1B: the method has been partially validated (the essential performance criteria in the NF-EN 482 Standard are met);
- Category 2: the method is indicative (essential criteria for validation are not clear enough);
- Category 3: the method is not recommended (essential criteria for validation are lacking or inappropriate).

A detailed comparative study of the methods in Categories 1A, 1B and 2 was conducted with respect to their various validation data and technical feasibility, in order to recommend the most suitable method(s) for measuring concentrations for comparison with OELs.

The collective expert appraisal work and its conclusions and recommendations were adopted on 17 May 2014 by the OEL Committee.

The collective expert appraisal work and the summary report were submitted to public consultation from 03/06/2015 to 03/08/2015. 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 2014-2017) who adopted this version on 7 march 2016.

[3] American Conference of Governmental Industrial Hygienists

Results of the collective expert appraisal on the health effects

Kinetics

No data were found in the literature regarding the absorption, distribution, metabolisation or excretion of n-butylamine in humans or animals.

Its chemical structure (a short-chain, non-substituted, primary amine) suggests the oxidation of this substance by monoamine oxidase in tissues (formation of the corresponding aldehyde) followed by further oxidation to carboxylic acid and then ammonia, which is probably excreted as urea (Health Council of the Netherlands, 2003). Moreover, acetoacetic acid is one of the metabolites of n-butylamine identified *in vitro* from guinea pig liver slices (Benya and Harbison, cited in Health Council of the Netherlands, 2003).

General toxicity

Toxicity in humans

Acute and subacute toxicity

Exposure to n-butylamine vapours causes severe irritation of the skin, eyes and mucosa (upper airways). The symptoms described following acute exposure to n-butylamine are red, watery eyes, nasal discharge, throat irritation and headaches. Exposure to (unspecified) high concentrations causes cyanosis, pulmonary oedema, convulsions and coma. Confidential reports (industrial reports; Beard and Noe, 1981 and Benya and Harbison, 1994 cited in Health Council of the Netherlands, 2003) show the following effects:

- no complaints at concentrations below 5 ppm;
- 5 to 10 ppm: nose, eye and throat irritation and headaches;
- 10 to 25 ppm: concentrations deemed intolerable by some workers.

Toxicity in animals

Acute and subacute toxicity

The Health Council of the Netherlands reported two studies in which all of the rats (n=4) died within five minutes for exposure to 4,000 ppm (12,000 $mg.m^{-3}$), and half of the rats (n=2-4/6) died within four hours of exposure to 4,000 ppm.

In mice, a lethal concentration (for 50% of the animals, LC_{50}) of 800 $mg.m^{-3}$ (264 ppm) was reported for two hours of exposure.

Only the study by Gamer *et al.* (2002) reported irritation of the upper respiratory tract confirmed by histopathology. In this study, pregnant Wistar rats were exposed, using an inhalation chamber, to concentrations of 17, 50 or 152 ppm, six hours per day for 14 days (25 animals per dose + one group of 25 unexposed animals).

The results of the histopathological examinations are broken down in the following table.

Table 1: prevalence of the effects observed in the nasal mucosa

	0	17 ppm	50 ppm	152 ppm
	Percentage of animals			
Squamous metaplasia	NS	10	50	100
Cell infiltration	NS	30	90	100
Necrosis	NS	NS	NS	50

In the preliminary study described by the authors in the 'methodology' section of the publication, (pregnant) rats were exposed (with an inhalation chamber) to concentrations of 84, 165 or 337 ppm of n-butylamine for 14 days. The authors observed, in particular, severe histopathological lesions in the respiratory tract for the concentrations of 165 and 337 ppm.

An oral exposure study (single dose of 100, 200, 300, 400, 500 or 600 $mg.kg^{-1}$) reported sedation, ataxia, convulsions and nasal discharge in the animals (Cheever *et al.*, 1982). They were gasping and had hypersalivation.

In mice, the threshold of irritation (determined by the decrease in respiratory rate) was identified at 110 to 120 ppm for CF-1 mice, and around 250 ppm for NMRI mice exposed by inhalation, and around 300 ppm for CF-1 mice and 360 ppm for NMRI mice exposed by intra-tracheal administration (Gagnaire *et al.*, 1989; Nielsen and Vinggaard, 1988; Vinggaard and Nielsen, 1989).

Carcinogenic effects - genotoxicity

No data on the carcinogenicity of n-butylamine were identified in the literature.

Reproductive toxicity

Gamer *et al.* (2002) (study cited above) studied the effects of n-butylamine on development. All of the animals were sacrificed 20 days post-coitum. The authors report that at 17 ppm (the highest concentration), no effects were observed on embryonic or foetal development (particularly foetal morphology). The statistical analysis of results shows no significant difference between the exposed and unexposed female rats for the following measurements: average gravid uterine weight, average weight gain (six days post-coitum), average number of corpora lutea and implantation sites, proportion of pre- and post-implantation losses, proportion of foetal resorptions, average number of live and dead foetuses per litter, average placental weight, average foetal weight, proportion of litters with at least one foetal malformation and proportion of foetuses per litter with at least one malformation. The authors concluded that there were no effects on development for exposure levels less than or equal to 152 ppm.

Establishment of OELs

Exposure to n-butylamine causes severe irritation of the skin, eyes and mucosa.

No conclusions can be drawn from the few data available in the literature as to the medium- or long-term systemic toxicity of n-butylamine.

Thus, according to the methodological document on the establishment of limit values for irritants and corrosives, a 15min-STEL can be recommended to protect workers from the irritating

effects of n-butylamine but it does not appear relevant to recommend an 8h-OEL (ANSES, 2014b).

15min-STEL

The study by Gamer *et al.* (2002) described above was used to establish the 15min-STEL. Histopathological examination showed, in the nasal mucosa, epithelial hyperplasia, squamous metaplasia, inflammation (cell infiltration) correlated with exposure levels, and necrosis.

The study's authors indicate that the NOAEL[4] for nasal epithelial hyperplasia, metaplasia, inflammation and necrosis was less than 17 ppm. The OEL Committee's experts interpreted this concentration of 17 ppm as a LOAEL[5].

The experts considered this study to be sufficiently robust to establish a 15min-STEL. It was a recent study, there were a sufficient number of animals used and concentrations tested and these concentrations were sufficiently spaced out to highlight potential toxic effects in the study groups. Furthermore, this study clearly showed a dose-response relationship.

The OEL Committee proposes applying the following adjustment factors (AF) to the LOAEL of 17 ppm:

- AF=3 for LOAEL-to-NOAEL adjustment
- AF=3 for inter-individual variability

No adjustment factors were introduced to take into account inter-species variability; according to the methodology of the OEL Committee, in the case of a non-systemic effect, the mechanism of action of the chemical substance varies little, irrespective of the species in question (bioavailability, metabolism, excretion and detoxification mechanisms do not impact the occurrence of local effects).

Therefore: 17 ppm / 9=1.88 ppm or 5.74 mg.m^{-3} (20°C conversion factor and 101 kPa[6]).

This value is rounded to recommend a 15min-STEL of 6 mg.m^{-3}.

"Skin notation"

In the absence of a conclusion on systemic toxicity, the "skin" notation was not assigned for n-butylamine.

"Ototoxic notation"

In the absence of scientific data on the ototoxic effects of n-butylamine, the "ototoxic" notation was not assigned for this substance.

[4] No Observed Adverse Effect Level.
[5] Lowest Observed Adverse Effect Level.
[6] conversion factor: 1 mg.m^{-3}= 0.328 ppm

Results of the collective expert appraisal on measurement methods in workplace atmospheres

Assessment of methods for measuring n-butylamine in workplace atmospheres

Two methods for measuring concentrations of n-butylamine in workplace atmospheres were identified and assessed (see Table 2).

Table 2: Assessment of methods for measuring concentrations of n-butylamine in workplace atmospheres

No	Methods	Protocols[7]	Category[8]	
			For regulatory technical control of the 15min-STEL	For monitoring short-term exposure
1	Sampling in a tube containing silica gel, desorption with an acetonitrile/m-toluoyl chloride mixture and NaOH or KOH, high-performance liquid chromatography with ultra-violet detection (HPLC-UV)	INRS MétroPol 026:2004	**3**	
2	Sampling in a tube containing silica gel impregnated with sulphuric acid. Desorption in a methanol/water mixture (50/50). Neutralisation with KOH. The extract is analysed by gas chromatography with flame ionisation detection.	NIOSH 2012: 1994	**3**	**2**

The graph below presents the ranges for which the various methods have been tested and their limits of quantification.

[7] INRS: National Research and Safety Institute; NIOSH: National Institute for Occupational Safety and Health.

[8] Validation and performance criteria for methods for monitoring STELs are defined in the NF EN 482 Standard from 0.5 to 2 times the STEL. Under the French regulations, for the technical control of the exposure limit, the measurement method must be able to measure one-tenth of the 15min-STEL (Ministerial Order of 15 December 2009 on technical controls of occupational exposure limits in workplace atmospheres and conditions for accrediting the organisations in charge of controls, published in the OJ of 17 December 2009). As such, when a method cannot measure one-tenth of the 15min-STEL, it cannot be classified in category 1A or 1B for regulatory control of the 15min-STEL. However, it may be classified in category 1A or 1B solely for assessing occupational exposure.

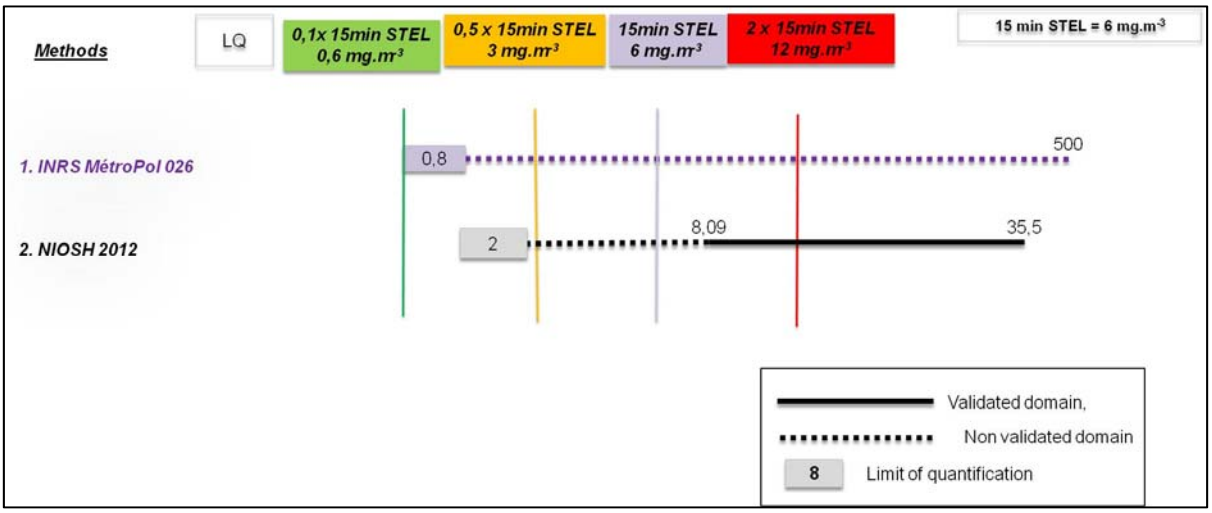

Figure 1: ranges of validity and limits of quantification for the various compared methods from 0.1 to 2 times the 15min-STEL recommended by the OEL Committee for n-butylamine

Method 1 has no validation data, except for a recovery rate determined by doping. It has therefore been classified in category 3 for technical control of the 15min-STEL recommended by the OEL Committee and the monitoring of short-term exposure.

Method 2 meets the main requirements of the NF EN 482 Standard. However, no information about interferences is mentioned in the NIOSH 2012 protocol. The method has therefore been classified in category 2 for the monitoring of short-term exposure.

The limit of quantification is higher than one-tenth of the 15min-STEL recommended by the OEL Committee. The method has therefore been classified in category 3 for regulatory technical control of the 15min-STEL. It should be noted that it should be possible to measure one-tenth of this value by using a more sensitive detector (NPD).

Conclusions of the collective expert appraisal

Based on the data currently available, the OEL Committee recommends establishing a 15min-STEL of 6 mg.m^{-3} and does not recommend an 8h-OEL considering the lack of expected medium- or long-term systemic effects.

The OEL Committee does not recommend a “skin” notation.

The OEL Committee does not recommend an “ototoxic” notation.

In light of the assessment of measurement methods, the OEL Committee emphasises that none of the identified methods are validated or indicative for regulatory technical control of the 15min-STEL.

For the monitoring of short-term exposure, the OEL Committee recommends the method described by the NIOSH 2012 protocol which involves sampling in a silica gel tube impregnated with sulphuric acid, desorption in a methanol/water mixture (50/50) followed by neutralisation with potassium hydroxide and then analysis by gas chromatography with flame ionisation detection. This method is indicative (category 2 classification) due to the lack of data on interferences.

Further details: General information on the substance

1) Identification of the substance:

Name	**n-butylamine**
CAS No	109-73-9
EINECS No	203-699-2
Molecular weight	73.1
Empirical formula	$C_4H_{11}N$
Physical form, appearance	Colourless/slightly yellow liquid with an ammonia odour

2) Physico-chemical properties:

Boiling point:	78°C
Solubility in water:	complete
Relative density of the mixture saturated with air (air=1)	1.1
Relative density (water=1)	0.7
Explosive limits (in % vol air)	1.7-10 (% v/v)
Conversion factor	1 $mg.m^{-3}$ = 0.328 ppm

3) Professional uses:

N-butylamine is used as an intermediate for the production of plasticisers, agrochemical products, pharmaceutical products, emulsifiers, dyes, tanning agents, and also as a rubber vulcanisation accelerator and as a curing agent for polymers.

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