The Director General

Maisons-Alfort, 30 October 2013

OPINION
of the French Agency for Food, Environmental and Occupational Health & Safety

on the development of TRVs for naphthalene by inhalation

ANSES undertakes independent and pluralistic scientific expert assessments.
ANSES primarily ensures environmental, occupational and food safety as well as assessing the potential health risks they may entail.
It also contributes to the protection of the health and welfare of animals, the protection of plant health and the evaluation of the nutritional characteristics of food.

It provides the competent authorities with all necessary information concerning these risks as well as the requisite expertise and scientific and technical support for drafting legislative and statutory provisions and implementing risk management strategies (Article L.1313-1 of the French Public Health Code).

Its Opinions are made public.

On 30 March 2009, ANSES issued an internal request with a view to developing toxicity reference values (TRVs) for naphthalene (CAS No. 91-20-3) by inhalation.

1. BACKGROUND AND PURPOSE OF THE REQUEST

Since 2004, ANSES has been pursuing work on the development of toxicity reference values (TRVs) along with methodological developments. This expertise work initially prioritised reprotoxic chemicals, then from 2007 it focused on carcinogenic chemicals. In this regard, a method for establishing TRVs based on carcinogenic effects was adopted as part of a pilot phase organised in 2008. Benzene, cadmium, ethanol, naphthalene and vinyl chloride were selected as the substances to be studied during a pilot phase. When this phase ended, the proposed carcinogenic TRVs for these substances were submitted for validation by the Expert Committee (CES) on Assessment of the risks related to chemical substances. This Opinion concerns the TRVs for naphthalene.

A TRV is a toxicity indicator. Comparing exposure levels with this indicator enables a risk to human health to be qualified or quantified. TRVs are specific to a duration (acute, subchronic or chronic) and route (oral or respiratory) of exposure, and to a type of effect (reprotoxic, carcinogenic, etc.). The way TRVs are established differs depending on the knowledge or assumptions made about a substance's mechanisms of action.

"Threshold dose" TRVs are established for substances that cause, above a certain dose, damage whose severity is proportional to the absorbed dose, while "non-threshold dose" TRVs are established for substances for which there is a probability, however small, that even a single molecule entering the body will cause harmful effects for the organism.

In practice, establishing a TRV involves the following steps:

- choice of the critical effect;
- identification of the assumption used to establish the TRV, whether it concerns a threshold or non-threshold dose, depending on the substance's mode of action;
- choice of a good quality scientific study enabling establishment of a dose-response relationship;
- choice or development of a critical dose from experimental doses and/or epidemiological data and, where necessary (for a critical dose obtained in animals), adjustment of this dose to humans;
- application of uncertainty factors to the critical dose to take uncertainties into account for threshold TRVs, or a linear extrapolation to the origin derived from the critical dose for non-threshold TRVs.

TRVs are established according to a highly structured and rigorous approach involving collective assessments by groups of specialists (AFSSET, 2010).

2. ORGANISATION OF THE EXPERT APPRAISAL

The expert appraisal was carried out in accordance with French standard NF X 50-110 "Quality in Expert Appraisals – General Requirements of Competence for Expert Appraisals (May 2003)".

This expert appraisal fell within the field of competence of the CES on "Assessment of the risks related to chemical substances". ANSES entrusted the appraisal to the "Toxicity reference values" working group. The methodological and scientific aspects of the work were submitted to the CES on 12 April and 31 May 2012, and 10 January, 28 March and 16 May 2013. The work was adopted by the CES on "Assessment of the risks related to chemical substances" at its meeting on 16 May 2013.

3. ANALYSIS AND CONCLUSIONS OF THE CES

General information

Naphthalene is a polycyclic aromatic hydrocarbon occurring naturally in the environment. Because of its volatility, exposure is predominantly via inhalation compared with other routes. Heaters are responsible for naphthalene emission in housing, while industry and traffic are the emission sources in outdoor air.

There are very few data in the literature on the toxicity of naphthalene in humans. All the information used to characterise the toxicological profile of the substance comes from studies conducted in animals.

Toxicokinetics

In animals, naphthalene is absorbed via the respiratory route and follows a process of metabolism via cytochromes P450 followed by conjugation with glutathione. Several reactive metabolites are then formed that may partly explain the toxicity of naphthalene (in particular 1,2-naphthoquinone and 1,4-naphthoquinone). The cytochrome P450 isoenzymes involved in naphthalene metabolism are 2F2 in mice and 2F4 in rats. Mouse lung microsomes metabolise naphthalene at a rate eight times higher than rat lung microsomes, and are found at levels six to 30 times higher than those in rats. It seems that

mice are more sensitive to the metabolic activation of naphthalene than rats, and also more than hamsters, monkeys and humans.

Chronic inhalation toxicity

In humans, several cases of hemolytic anemia following inhalation and dermal penetration have been described in newborns whose clothes and bedding were stored with mothballs. This same anaemia has also been reported in newborns exposed by inhalation to medicines containing naphthalene. The cases occurring in these newborns were sometimes associated with neurological disorders such as drowsiness.

In animals, one of the main studies concerns groups of male and female B6C3F1 mice (aged 6 to 7 weeks) exposed by inhalation to naphthalene vapours (> 99% purity) for 6 hours/day, 5 days/week for 2 years (NTP, 1992; Abdo *et al.*, 1992). The groups of mice were exposed to concentrations of 0 (75 animals/sex), 52 (75 animals/sex) and 157 $mg.m^{-3}$ (150 animals/sex).
The number of animals exposed to the highest concentration was doubled due to the lack of data on naphthalene toxicity during chronic exposure. Mortality and morbidity were noted twice a day, every day of the week.
A significant increase in the incidence of non-cancerous lesions was observed in the lungs and nasal cavity of the exposed male and female mice.

An NTP study (2000) (Abdo *et al.*, 2001; Long *et al.*, 2003) provides additional information on the lesions observed in the nasal epithelium of rats. This study was conducted in F344 rats (49 males and 49 females) exposed by inhalation to concentrations of 0 - 52 - 157 - 314 $mg.m^{-3}$ for 6 h/d, 5 d/w for 2 years (naphthalene purity > 99%). The concentration of 314 $mg.m^{-3}$ was introduced in the test because rats are a less sensitive species than mice and this corresponds to the maximum concentration that can be achieved without causing condensation of naphthalene. The animals were monitored regularly and a full anatomopathological study was conducted.
Mean weights of the exposed male rats were slightly lower than those of the control rats throughout the study, while those of female rats were unchanged. The survival time of exposed rats and controls was similar.
In animals of both sexes, naphthalene induced numerous non-cancerous nasal lesions: atypical basal cell hyperplasia, atrophy, chronic inflammation and hyaline degeneration. Hyperplasia, squamous metaplasia, hyaline degeneration and hyperplasia of goblet cells were observed in the respiratory epithelium. Naphthalene also caused glandular hyperplasia and squamous metaplasia of the glandular epithelium. In general, the severity of the effects increased with a rise in concentration. These non-cancerous lesions are usually observed during exposure to other chemicals that are irritant by inhalation (NTP, 2000).

Carcinogenic effects of naphthalene by inhalation in animals

The NTP study (1992) exposed 70 male and 70 female B6C3F1 mice for 6 h/d, 5 d/w for 104 weeks to doses of 0 and 52 $mg.m^{-3}$, as well as 135 male and 135 female mice to 157 $mg.m^{-3}$ (naphthalene purity > 99%).
This study reports the development of numerous cancerous lesions. In the group exposed to 157 $mg.m^{-3}$, an increase in the incidence of alveolar and bronchiolar adenomas was seen in female mice (28/134) compared to the control group. An increase in the incidence of adenomas was also observed in the males but this remains marginal due to the bias associated with differences in survival between control animals and exposed animals. A histopathological analysis showed that 5/135 of carcinomas were haemangiosarcomas, but due to a frequency of development similar to that of the controls, and their non-specific localisation, these carcinomas are not regarded as being specific to naphthalene exposure.

This study showed that naphthalene is carcinogenic in female mice (significant increase in the incidence of bronchoalveolar adenomas, non-significant increase in the incidence of carcinomas). In the males, these results are not statistically significant because of the high mortality rate in the control groups. A Lowest Observed Adverse Effect Concentration (LOAEC) of 157 $mg.m^{-3}$ can be established for the development of bronchoalveolar adenomas in female mice (NTP, 1992).

The NTP study (2000) was conducted in F344 rats (49 males and 49 females) exposed by inhalation to 0, 52, 157 and 314 $mg.m^{-3}$ for 6 h/d, 5 d/w for 105 weeks (naphthalene purity $>$ 99%). This study showed a significant increase in the incidence of neuroblastomas of the olfactory epithelium, respectively in 0/49, 2/49, 3/49 and 12/49 female rats. In addition, adenomas of the nasal respiratory epithelium were observed at a significant level in respectively 0/49, 6/49, 8/48 and 15/48 male rats.
Neuroblastomas occurred in male rats for exposures to 157 and 314 $mg.m^{-3}$. One male rat for each of the two concentrations of 157 and 314 $mg.m^{-3}$ also presented lung metastases.
The dose-response relationship was significant for all exposed groups among the females. The incidence in females was statistically different compared to the control at the 314 $mg.m^{-3}$ concentration.
A dose-response relationship in the incidence of adenomas was observed in both males and females, but only the incidences in males increased in a statistically significant manner for all exposed groups.

A second publication based on the 2000 NTP study (Long *et al.,* 2003) details the histological characteristics of cancerous (neuroblastomas and adenomas of the respiratory epithelium) and non-cancerous lesions (inflammatory lesions of the olfactory and respiratory epithelium, and of the *lamina propria* of the olfactory region).
Neuroblastomas of the olfactory epithelium are highly invasive masses of variable shape that develop in the ethmoidal region of the nasal cavity and may extend to the respiratory epithelia.
The neuroblastomas and adenomas reported above are regarded as carcinogenic effects of naphthalene due to their high incidence in exposed animals and the absence of these tumours both in the control animals and in historical laboratory controls.
Undifferentiated basal cells may be responsible for olfactory neuroblastomas, as these are able to differentiate into support cells or sensory cells (Long *et al.*, 2003).

Genotoxicity
In vitro, naphthalene did not induce mutation in bacteria, with or without metabolic activation (NTP, 2011; IARC, 2002), nor mutation on metabolically competent human lymphoblastoid cells at the two loci tested. Clastogenic potential (micronuclei, chromosomal aberrations and sister chromatid exchanges) has only been shown *in vitro* for naphthalene and two of its metabolites (1,2- and 1,4-naphthoquinone). *In vivo,* the few studies available show that naphthalene has a toxic effect by oxidative stress.
An overall analysis of these data was thus unable to rule out the genotoxic potential of naphthalene. According to the method of establishing carcinogenic TRVs (AFSSET, 2010), naphthalene is regarded as a genotoxic carcinogen, with a non-threshold mode of action.

Development of a chronic threshold TRV for non-cancerous effects
Choice of the critical effect
The olfactory and respiratory epithelia of the nasal cavity are the critical target organs for the toxic effects of naphthalene. From the lowest concentration tested, inflammatory lesions have been observed as well as hyperplasia and aplasia phenomena, indicating local cytotoxicity.

It is also considered that these inflammatory olfactory and respiratory irritant effects are observed at lower naphthalene concentrations than those associated with the development of hemolytic anemia, whose effects are the most widely documented in humans.
The critical effect selected is chronic inflammation of the respiratory and olfactory epithelia observed in rats.

Choice of the key study
Among the animal studies, two inhalation carcinogenicity studies have been conducted in rodents (rats and mice) (NTP, 1992 and 2000). Although no toxicity studies were conducted prior to the one from 1992, which would have enabled more relevant test concentrations to be determined (inflammatory and cytotoxic phenomena are described from 52 $mg.m^{-3}$, the lowest concentration tested), these studies are of good quality (valid without restriction according to the Klimisch rating).
The NTP study (2000) was therefore selected as the key study.

Choice of the critical concentration
The NTP study (2000) showed that non-cancerous lesions of the olfactory and respiratory epithelia (chronic inflammation, hyperplasia, etc.) were observed in almost all the rats exposed to the concentration of 52 $mg.m^{-3}$ (Table I:).

Table I: Incidence of non-cancerous lesions in rats of both sexes

Lesions	Sex	0 $mg.m^{-3}$	52 $mg.m^{-3}$	157 $mg.m^{-3}$	314 $mg.m^{-3}$
Atypical hyperplasia of the olfactory epithelium	M	0/49	48/49	45/48	46/48
	F	0/49	48/49	48/49	43/49
Atrophy of the olfactory epithelium	M	3/49	49/49	48/48	47/48
	F	0/49	49/49	49/49	47/49
Chronic inflammation of the olfactory epithelium	M	0/49	49/49	48/48	48/49
	F	0/49	47/49	47/49	45/49
Hyaline degeneration of the olfactory epithelium	M	3/49	45/49	40/48	38/48
	F	13/49	46/49	49/49	45/49
Hyperplasia of the respiratory epithelium	M	3/49	21/49	29/48	29/48
	F	0/49	18/49	22/49	23/49
Squamous cell metaplasia of the respiratory epithelium	M	0/49	15/49	23/48	18/48
	F	0/49	21/49	17/49	15/49
Hyaline degeneration of the respiratory epithelium	M	0/49	20/49	19/48	19/48
	F	8/49	33/49	34/49	28/49
Goblet cell hyperplasia of the epithelium	M	0/49	25/49	29/48	26/48
	F	0/49	16/49	29/49	20/49
Bowman's gland hyperplasia	M	1/49	49/49	48/48	48/48
	F	0/49	48/49	48/49	42/49
Squamous cell metaplasia of the Bowman's glands	M	0/49	3/49	14/48	26/48
	F	0/49	2/49	20/49	20/49

Due to the high proportion of animals developing lesions from the first concentration tested, the dose-response relationship cannot be used to determine a benchmark concentration (BMC). Therefore, the concentration of 52 $mg.m^{-3}$ is considered a LOAEC and can be used as a starting point for calculating the TRV.

Allometric adjustment
The aim is to reduce the value of the uncertainty related to inter-species variability in order to determine a human equivalent concentration (HEC). For the respiratory tract, the US EPA has developed various dosimetric adjustments that are applied based on the physico-chemical properties of the inhaled substance (particles or gases, highly soluble or

slightly soluble in water) and the site where the critical effects are observed (respiratory or extrarespiratory), leading to different equations (US EPA, 1994).
Naphthalene is only slightly soluble in water. Its metabolism involves the formation of reactive metabolites that are responsible for the toxic effects observed in animals, and which may also be formed in the liver (US EPA, 1998). In addition, studies conducted in rodents involving intraperitoneal administration of naphthalene have shown the appearance of cell lesions (swelling, vacuolation, exfoliation, necrosis) in the tracheobronchial epithelial cells (Clara cells) of mice and the olfactory epithelial cells of rats, mice and hamsters.
Based on the recommendations of the US EPA (1994), the toxicity of naphthalene in olfactory and respiratory epithelia in rodents is considered to be extrarespiratory effects of a category 3 gas (systemic toxicity). The allometric adjustment applied by default for a category 3 gas is as follows:

$$LOAEC_{HEC} = LOAEC_{rat} \times (Hb/g)_{rat} / (Hb/g)_{human}$$

Where (Hb/g): blood/air partition coefficient of naphthalene.

As the blood/air partition coefficients of naphthalene for humans and rats are not known, the US EPA proposes selecting the default value of 1. Indeed, data from the literature show that the blood/air partition coefficient in animals is higher than in humans (US EPA, 1994).

$$\mathbf{LOAEC_{HEC} = 52\ mg.m^{-3}}$$

Temporal adjustment
The animals were exposed for 6 hours/day, 5 days/week for two years. To take account of the discontinuity of exposure, a temporal adjustment was applied:

$$LOAEC_{\mathbf{HEC\ ADJ}} = LOAEC_{HEC} \times (5/7) \times (6/24) = \mathbf{9.29\ mg.m^{-3}}$$

Choice of uncertainty factors
The TRV was calculated from the $LOAEC_{\mathbf{HEC\ ADJ}}$ using the following uncertainty factors (AFSSET, 2007):

- **Interspecies variability ($UF_{A\text{-}TD}$) = 2.5**: an allometric adjustment was applied to take into account the variability between species and enable calculation of a human equivalent concentration, using the above-mentioned equation. To take account of the toxicodynamic variability and residual uncertainty, an additional uncertainty factor was set at 2.5 according to ANSES practice.
- **Interindividual variability (UF_H) = 10**: a factor of 10 was applied by default to take account of variability within the human species and the existence of vulnerable populations, especially G6PD-deficient individuals.
- **Use of a LOAEC (UF_L) = 10**: the critical concentration selected is a LOAEC, i.e. a concentration associated with an effect percentage. This concentration does not therefore ensure the absence of occurrence of toxic effects of naphthalene in the same way as a NOAEC (No Observed Adverse Effect Level). Moreover, the LOAEC selected according to the NTP study (2000) corresponds to the lowest concentration tested, which is known to be higher than the maximum tolerated doses (MTDs). Lastly, a very high proportion of animals developed lesions of the respiratory and olfactory epithelium from this first concentration. For these reasons, an additional uncertainty factor of 10 was applied.

Critical effect	Critical concentration	UF	TRV
Lesions of the respiratory and olfactory epithelium in F344 rats NTP, 2000	LOAEC = 52 mg.m^{-3} <u>Allometric adjustment</u> $LOAEC_{HEC}$ = 52 mg.m^{-3} <u>Temporal adjustment</u> $LOAEC_{HEC\ ADJ}$ = 9.29 mg.m^{-3}	250 UF_{A-TD} = 2.5 UF_H = 10 UF_L = 10	**TRV = 37 µg.m^{-3}** **Confidence level** MODERATE

Confidence level: MODERATE

- Choice of the critical effect: high confidence level (NTP study (2000) of good scientific quality; effects consistent with the results of other toxicology studies);
- Quality of the key study: moderate confidence level (questionable choice of test concentrations as they are higher than the MTDs, no prior chronic toxicity study, high proportion of animals developed lesions from the first concentration tested, inability to define a NOAEC);
- Establishment of the TRV: moderate confidence level (no modelling of a benchmark concentration (BMC), application of a high overall uncertainty factor).

Development of a non-threshold TRV for carcinogenic effects

Choice of assumption made to establish the TRV

After inhalation, naphthalene is distributed throughout the body to the site of its metabolism into genotoxic reagents. Although metabolism of naphthalene varies according to the species and anatomical sites, the isoenzymes involved predominate in mice compared to other species and are mainly present in the olfactory epithelium.

The genotoxic effects of naphthalene implicated in the occurrence of tumours of the olfactory epithelium involve the production of reactive and mutagenic metabolites. Although naphthalene does not induce mutations in either bacteria or human cells, formation of adducts and clastogenic potential have been shown *in vitro.* Some studies on oxidative stress (Bagchi *et al*., 1998, 2000, 2001 and 2002) have concluded that there is a cytotoxic mode of action responsible for the carcinogenicity of naphthalene. However, the hypothesis of a genotoxic potential of naphthalene cannot be ruled out and scientific views diverge on this matter.

In accordance with the method of establishing carcinogenic TRVs (AFSSET, 2010), the assumption chosen was a mode of carcinogenic action with no threshold dose.

Choice of the key study

There is no epidemiological study in the scientific literature that can be used to develop a carcinogenic TRV for naphthalene by inhalation.

Among the animal studies, two inhalation carcinogenicity studies have been conducted in rodents (rats and mice) (NTP, 1992 and 2000). Although no toxicity studies were conducted prior to the one from 1992, which would have enabled more relevant test concentrations to be determined (inflammatory and cytotoxic phenomena are described from the lowest concentration tested), these studies are of good quality (valid without restriction according to the Klimisch rating) and were selected as the key studies.

Choice of the critical effect

In both NTP studies, the described effects were alveolar and bronchiolar adenomas and carcinomas in mice, and adenomas of the nasal epithelium and neuroblastomas of the

olfactory epithelium in rats. These differences in anatomical tumour sites between mice and rats can be explained by inter-species metabolic variability. In rats, the olfactory and respiratory epithelia of the nasal cavity are the targets for the toxic and carcinogenic effects of naphthalene, which induces an increased incidence of adenomas of the respiratory epithelium in males and neuroblastomas of the olfactory epithelium in females.
In the current state of knowledge, it is not possible to state that the effects observed in mice (alveolar or bronchiolar adenomas or carcinomas) are more representative of possible effects in humans than those reported in rats (adenomas of the respiratory epithelium and neuroblastomas of the olfactory epithelium). The observations in mice have also been criticised for their relevance and causality with regard to exposure to naphthalene (US EPA, 1998; OEHHA, 2000). In addition, it does not seem appropriate to transpose the effects observed in Clara cells in mice, due to this species' greater susceptibility and a higher metabolic capacity in these cells compared to that in rats, primates and humans (WHO, 2010).
The adenomas of the respiratory epithelium observed in male rats are benign tumours. In the current state of knowledge, it is not possible to determine whether these adenomas have the potential to transform into malignant tumours. Considering their benign nature and in accordance with the method of establishing carcinogenic TRVs (AFSSET, 2010), the adenomas of the respiratory epithelium observed in male rats were not selected as the critical effect.
Neuroblastomas are tumours of the neuroblasts or nearby cells, arising from neural crest cells. These solid tumours are rare in humans and are mainly observed in children (8-10% of paediatric cancers and 1 case per 70,000 cases of solid tumours in children under 15 years). In the NTP study (2000), neuroblastomas were observed in adult animals. It is therefore difficult to transpose this observation to a similar situation in humans (neuroblastomas occurring in adulthood). However, considering the metabolism involving the CYP450 isoenzymes found in rodents and in humans, and the severity and poor survival prognosis associated with this type of tumour, the carcinogenic potential of naphthalene is regarded as being transferable to humans (WHO, 2010).
The critical effect selected is the increased incidence of neuroblastomas of the olfactory epithelium in female rats.

Choice of the starting point
The results provided by the NTP study (1992) in mice are insufficient for developing a BMC (only two concentrations were tested besides the controls). The results from the most recent study (NTP, 2000) conducted in rats were therefore used to calculate the BMC.
The data in the table below were modelled with version 18.2 of the RIVM's PROAST software (Netherlands) to determine a BMC. A significant dose-response relationship in female rats compared to male rats, between the increased incidence of neuroblastomas and the concentrations of naphthalene exposure, was confirmed by a trend test.

Table II: Incidence of neuroblastomas of the olfactory epithelium in rats

Lesions	Sex	0 mg.m^{-3}	52 mg.m^{-3}	157 mg.m^{-3}	314 mg.m^{-3}
Neuroblastomas of the olfactory epithelium	M	0/49 (0%)	0/49 (0%)	4/48 (0%)	3/48 (6%)
	F	**0/49 (0%)**	**2/49 (4%)**	**3/49 (6%)**	**12/48 (24%)**

The aim of the approach is to estimate the dose corresponding to a defined level of response or a defined percentage of additional response compared to a control. This level or percentage is called the Benchmark Response (BMR). This is predominantly the BMCL, i.e. the lower limit of the BMC confidence interval, which is considered to be a critical concentration for developing the TRV.

The model that offered the best fit with the experimental data was selected, using the maximum likelihood method (log likelihood). The log-logistic model was selected for estimating the lower limit of the 90% confidence interval[1] of a concentration corresponding to a 10% increased response compared to the non-exposed group (the 10% threshold is generally used in carcinogenicity studies). The lower limit of the 90% confidence interval of the BMC was selected, after being established:

- for the critical effect selected: neuroblastomas of the olfactory epithelium in female rats,
- for an effect level corresponding to 10% of the effect level observed in the control group,
- calculated from the Weibull model using RIVM's PROAST software (Netherlands).

So **$BMC_{10\%}$ = 155 mg.m^{-3}** and **$BMC_{10\%}L_{90\%}$ = 99.6 mg.m^{-3}**

Allometric adjustment
As indicated above, naphthalene is considered a category 3 gas, i.e. whose observed toxic effects are associated with systemic toxicity (according to the method of the US EPA, 1994).
Based on the recommendations of the US EPA regarding the use of dosimetry by default, the formula for a category 3 gas can be used:

$$BMC_{10\%}L_{90\%\ HEC} = BMC_{10}L_{90\%\ rat} \times (Hb/g)_{rat} / (Hb/g)_{human}$$

Where (Hb/g): blood/air partition coefficient of naphthalene.

As the blood/air partition coefficients of naphthalene for humans and rats are not known, the US EPA proposes selecting the value of 1.

So **$BMC_{10\%}L_{90\%\ HEC}$ = $BMC_{10\%}L_{90\%}$ = 99.6 mg.m^{-3}**

Temporal adjustment
The animals were exposed for 6 hours/day, 5 days/week for two years. To take account of the discontinuity of exposure, a temporal adjustment was applied:

$BMC_{10\%}L_{90\%\ HEC\ ADJ}$ = 99.6 x (5/7) x (6/24) = **17.8 mg.m^{-3}**

Calculation of the TRV
The carcinogenic TRV for naphthalene corresponds to the excess risk per unit (ERU) obtained after linear extrapolation from the previously determined $BMC_{10\%}L_{90\%\ HEC\ ADJ}$, namely:

$ERU = (0.1 / BMC_{10\%}\ L_{90\%\ HEC\ ADJ}) = (0.1 / 17.8) = 0.0056\ (mg.m^{-3})^{-1} = 5.6.10^{-3}\ (mg.m^{-3})^{-1}$

[1] The lower limit of the 95% confidence interval ("one-sided"), used by the US EPA is equivalent to the lower limit of the 90% confidence interval ("two-sided") used by the RIVM.

Critical effect	Critical concentration	TRV
Neuroblastomas of the olfactory epithelium in female F344 rats NTP, 2000	$BMC_{10\%}\ L_{90\%}$ = 99.6 $mg.m^{-3}$ Allometric adjustment $BMC_{10\%}\ L_{90\%\ HEC}$ = 99.6 $mg.m^{-3}$ Temporal adjustment $BMC_{10\%}\ L_{90\%\ HEC\ ADJ}$ = 17.8 $mg.m^{-3}$	After linear extrapolation to the origin: **ERU = 5.6.10^{-3} (mg.m^{-3})$^{-1}$** 0.18 µg.m^{-3} for a risk of 10^{-6} 1.8 µg.m^{-3} for a risk of 10^{-5} 18 µg.m^{-3} for a risk of 10^{-4}
		Confidence level MODERATE

Confidence level: MODERATE

- Choice of the critical effect: low confidence level (malignant tumours occurring in older animals that cannot be transposed to a situation in humans, rare tumours found in young children harbouring mutations),
- Quality of the key study: moderate confidence level (dose-response relationship not very robust),
- Establishment of the TRV: high confidence level.

4. AGENCY CONCLUSIONS AND RECOMMENDATIONS

The French Agency for Food, Environmental and Occupational Health & Safety endorses the conclusions and recommendations of the CES on Assessment of the risks related to chemical substances, concerning the development of toxicity reference values for naphthalene by inhalation.

Critical effect and source study	Method of establishment	TRV
Lesions of the respiratory and olfactory epithelium in F344 rats NTP, 2000	LOAEC = 52 $mg.m^{-3}$ Allometric adjustment $LOAEC_{HEC}$ = 52 $mg.m^{-3}$ Temporal adjustment $LOAEC_{HEC\ ADJ}$ = 9.29 $mg.m^{-3}$ Uncertainty factors UF = 250 (UF_{A-TD} = 2.5; UF_H = 10; UF_L = 10)	**37 µg.m^{-3}**
Neuroblastomas of the olfactory epithelium in female F344 rats NTP, 2000	$BMC_{10\%}\ L_{90\%}$ = 99.6 $mg.m^{-3}$ Allometric adjustment $BMC_{10\%}\ L_{90\%\ HEC}$ = 99.6 $mg.m^{-3}$ Temporal adjustment $BMC_{10\%}\ L_{90\%\ HEC\ ADJ}$ = 17.8 $mg.m^{-3}$ Extrapolation to low doses Linear extrapolation to the origin	**5.6.10^{-3} (mg.m^{-3})$^{-1}$** Concentrations associated with several risk levels: **10^{-4}: 18 µg.m^{-3}** **10^{-5}: 1.8 µg.m^{-3}** **10^{-6}: 0.18 µg.m^{-3}**

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KEY WORDS

Naphthalene, toxicity reference value, inhalation

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