CHEMICAL ENGINEERING TRANSACTIONS
VOL. 77, 2019
A publication of AIDIC
The Italian Association of Chemical Engineering
Online at www.cetjournal.it
Guest Editors: Genserik Reniers, Bruno Fabiano

ISBN 978-88-95608-74-7; **ISSN** 2283-9216
DOI: 10.3303/CET1977005

The Use of Probit Functions for Toxic Substances in Quantitative Risk Analysis

Karola Imbrechts

Government of Flanders, Department of Environment & Spatial Development, Koning Albert II-laan 20 bus 8, 1000 Brussels, Belgium
Karola.imbrechts@vlaanderen.be

When calculating the risk of a Seveso-establishment with toxic substances, probit functions are used to have a relationship between the human lethality on one hand and the concentration of the toxic substance on the other hand. These probit functions have a substantial impact on the calculated effect of a toxic release and so the risk of the establishment. Probit functions are derived from animal experiments with a lot of assumptions, among others the classification of the acute inhalation studies and the extrapolation factors for animal to human extrapolation.
In this paper, the methodology to derive probit functions will be described for Flanders and The Netherlands with emphasis on the differences. The probit function for the toxic substance hydrogen chloride (HCl) will be studied more in detail, because, in this case, there is another important assumption, namely whether or not considering a specific study about baboons.

1. Introduction

The Flemish regulations require Seveso-establishments to draw up (1) a safety report to obtain an environmental permit and (2) a safety report in the context of the European Seveso-Directive. These safety reports contain the risk map of the establishment. The risk map includes the location specific risk as well as the societal risk and they both must meet the applicable risk criteria. In these calculations the guideline on probit functions for toxic substances is used.
A probit function for the acute inhalation toxicity of a substance describes the human lethality rate in an exposed population as a function of any combination of the exposure concentration and exposure duration. The toxicity probit functions are of the form $Pr = a + b.\ln(C^n.t)$ with concentration C in mg/m^3 and exposure time t in minutes. The parameters a, b and n are constants with a value depending on the substance and are derived from experiments on animals.

2. Method to derive probit functions for humans from experiments on animals

The method used in Flanders to derive a probit function for humans for toxic substances is based on the old method from The Netherlands that is described in the Green Book (VROM, 1990, 2005a). It has been used to derive all new probit functions since 2000 (Hooghe, Wouters, 2000) (Bloemen et al., 2005) (Cornelis et al., 2018), with one modification since 2005. In the meantime, in the Netherlands a new method has been developed to derive new probit functions (Ruijten et al., 2015). The general concept of both and the main differences will be described in the following paragraphs.

2.1 General concept

The method to derive a probit function for a toxic substance is described in detail in the above mentioned reports. The general concept consists of (1) the collection of relevant acute inhalation studies on animals and the selection of useful data, (2) the derivation of a probit function for the animals and (3) the conversion of the probit function for the animals to a probit function for humans. In the first step, the studies are classified as A, B or C studies based on their quality and the available data. In the second and third step, assumptions have to be made.

Paper Received: 9 February 2019; Revised: 20 May 2019; Accepted: 27 June 2019

Please cite this article as: Imbrechts K., 2019, The use of probit functions for toxic substances in Quantitative Risk Analysis, Chemical Engineering Transactions, 77, 25-30 DOI:10.3303/CET1977005

Both the Flemish and the Dutch methodology use this general concept. The main differences in its application can be summarized as (1) the subjective interpretation of the available data, (2) whether or not applying an extra factor of 2 when data on multiple animal species is selected and (3) the extrapolation factors for converting animal data to human data.

2.2 Flanders

The relevant aspects of the current methodology of Flanders will be described. The acute inhalation studies are classified as follows. (A) A-studies are studies with data generated for a combination of different concentrations and exposure times and in which the lethality is dose-related. (B) B-studies are studies in which either one concentration and different exposure times are used, or one exposure time and different test concentrations. Lethality is either concentration or time dependent and is between 1 and 100 % for the different test conditions allowing the calculation of the lethal concentration and exposure time with 50 % lethality (LC50 and LT50). (C) C-studies are studies with low quality but reporting LC50 or LT50 values or studies providing relevant data that do however not allow calculating LC50 or LT50.
The method for deriving the values for the constants a, b and n in the probit function is dependent on the available studies. (A) For A-studies, the values are calculated from the experimental data using the software DoseResp (Ten Berghe, 2015). Data from animals is extrapolated to humans by species specific extrapolation factors (see Table 1). The value for n is the same value as provided for the animal and b = 2/n. The value for a is calculated with the probit function and calculated LC50 value. (B) For B-studies, default values are used, namely n = 2 and b = 1. Animal data are extrapolated to humans by species specific extrapolation factors in the same way as for A-studies. (C) C-studies are only used when no A or B studies are available and only when LC50 or LT50 values are provided. The same procedure as for B studies is then followed. When the quality of the study is not sufficient, no probit function is calculated.
If data with multiple animal species are selected, a mean value for the LC50 is calculated for the animal data. In the original method, this mean value is multiplied with an extra factor of 2. This has been applied in 2000. In (Bloemen et al., 2005) this factor of 2 was questioned and probit functions with and without this extra factor were calculated. When publishing the probit functions, Flanders decided to use the probit functions without the extra factor of 2. Therefore, in (Cornelis et al., 2018) this factor was ignored.

Table 1: Extrapolation factors for animal species in Flanders

Animal species	Rat	Mouse	Cavia	Hamster	Others
Extrapolation factor	0.25	0.5	0.2	0.3	To be estimated per substance

After derivation, the quality and reliability of the probit function is assessed by comparison to available reference values and experimental data. The final score of the evaluation is based on the aspects given in Table 2.

Table 2: Score of studies and probit functions

Score	The quality of the base studies	Fitting of lethality data for human exposure	Fitting of reference values
++	Not applicable	Human lethality data are available and they fit within the probit data.	Not applicable.
+	Data are provided that allow to evaluate the experiment procedure, so that the study is reliable and well documented.	No human data. Experimental data on other species can be extrapolated to human and these fit within the probit data.	Reference values fit.
+/-	The study is rather well described to reconstruct the exposure, but details are missing and reliability is lower than in the previous case.	Some of the data do not fit, but most of them do. Or, there is only a limited number of values and they fit.	Some values do not fit, but most of them do. Or, there is only a limited number of values available and they fit.
-	The study is not well described, lot of information is lacking and reliability cannot be evaluated.	Most of the data do not fit or no data are available.	There is a systematic deviation or no data are available.

2.3 The Netherlands

The document of Ruijten et al. (2015) describes the new methodology used for the derivation of probit functions in The Netherlands. The methodology, that was formerly described in the Green Book (VROM, 2005a), has been updated and thoroughly revised. After introducing the basic philosophy of deriving a probit function, the methodology describes in detail the interpretation and use of animal data and the derivation of the probit function. The revised method makes use of various assessment factors to account for e.g. data quality, sensory irritation, and inter- and intra-species variability.
For this paper, an important aspect of the new methodology is the fact that the extrapolation factor for animal to human extrapolation is now equal for all animal species and set at a default value of 1/3. Before, the values of Table 1 were used. Also, the procedure to raise the LC_{50} value when data from multiple animal species is available has been dropped. This confirms the omitting of the extra factor of 2 in Flanders.

3. Derived probit functions for HCl

In this section, the resulting probit functions for Hydrogen Chloride (HCl) (CAS number: 7647-01-0) are discussed, as well for Flanders as for The Netherlands.

3.1 Flanders

The current probit function for HCl for Flanders is derived by (Hooghe, Wouters, 2000) based on the (old) Green Book (VROM, 1990). In this derivation, the extra factor of 2 for multiple animal species was still used.
In (Cornelis et al., 2018), a new probit function was derived, because of the availability of more and more reliable studies. The global quality assessment according to Table 2 is A+, because of the fact that (1) the study category is A, (2) the quality of the studies used is +, (3) the testing against limit values is + and (4) the assessment of human lethality is -. Comparison with the data for LC50 of the B and C studies shows that the probit is relatively stringent, but also yields values that correspond to those derived from the experiments on test animals. Therefore, Cornelis et al. (2018) states that the derived probit is considered as reliable.

3.2 The Netherlands

In The Netherlands, the current probit function for HCl is the one from (RIVM, 2015), which is adopted from the Purple Book (VROM, 2005b), where is mentioned that the probit functions for toxic substances are described in the (old) Green Book (VROM, 1990) and that the values for the toxic constants a, b and n for hydrogen chloride are taken from (VROM, 1989). In (Ruijten, 2017), a new probit function for HCl is derived with the new method.

3.3 Comparison

From the above, it can already be concluded that the currently used probit functions of Flanders and The Netherlands are based on the same method, namely the one of (VROM, 1990).
In Table 3, the currently used and newly derived probit functions for Flanders and The Netherlands are represented together with the value for $LC_{50,30'}$ (lethal concentration for 50 % lethality and 30' exposure time). If the extra factor of 2 for multiple species is not applied in the derivation of the current probit function, the probit function would be $-16{,}63 + 2 \cdot \ln(C \cdot t)$ with $LC_{50,\,30'}$ = 1,662 mg/m³.

Table 3: Currently used and newly derived probit functions for HCl in Flanders and The Netherlands

Country	Current probit function [mg/m³]	New probit function [mg/m³]
Flanders	$-18.02 + 2 \cdot \ln(C \cdot t)$	$-16.03 + 1.70 \cdot \ln(C^{1.18} \cdot t)$
	with $LC_{50,\,30'}$ = 3,324 mg/m³	with $LC_{50,\,30'}$ = 2,062 mg/m³
The Netherlands	$-37.7 + 3.69 \cdot \ln(C \cdot t)$	$-17.09 + 1.463 \cdot \ln(C^{1.367} \cdot t)$
	with $LC_{50,\,30'}$ = 3,536 mg/m³	with $LC_{50,\,30'}$ = 5,198 mg/m³

These 5 probit functions are shown in Figure 1. This demonstrates that there can be a very big difference in the results depending on the studies used, the method used and the assumptions made. For Flanders, the only difference between the probit functions "Flanders 2000 – current" and "Flanders 2000 – without factor 2" is the extra factor 2 for taking into account different kind of animal species. And for the probit functions "Flanders 2000 – without factor 2" and "Flanders 2018 – new" in principle the same method is used, namely the one of the Green Book, and the extra factor 2 is not applied. The difference is due to the availability of more, more recent and more reliable studies. For the Netherlands, the difference between the 2 probit functions is due to the application of the whole new and revised method and due to the use of more recent studies.

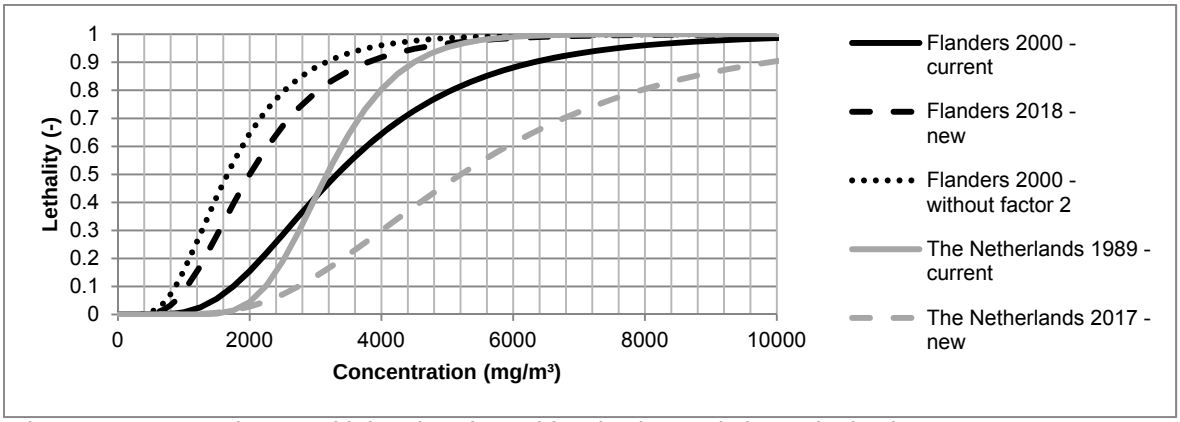

Figure 1: Current and new probit functions for HCl for Flanders and The Netherlands

In Table 4, an overview is given of the studies that have been used to derive the new probit functions. From this, it can be concluded that the newly derived probit functions are based on the same studies, although they were not always given the same classification and in (Cornelis et al., 2018) some of them, namely the studies classified as B-studies, were only used to verify the calculated probit function according to the methodology, while they were really taken into account in (Ruijten, 2017) to calculate the probit function.

Table 4: Studies used for the different probit functions for HCl in Flanders and The Netherlands, with indication of the classification (studies between brackets are not taken into account for the calculation)

Study reference	Animal species	Flanders 2018	The Netherlands 2017
Arts et al., 2000	Rat	A	A
Hartzell et al., 1985	Rat	A	A
Lapin, 1981	Rat	A	A
Darmer et al., 1974	Rat	A	B1
Darmer et al., 1974	Mouse	A	B1
Jean et al., 2006	Rat	[B]	B1
Wohlslagel et al., 1976	Rat	[B]	B1
Wohlslagel et al., 1976	Mouse	[B]	B1

4. The influence of baboons

There is still one more big difference between the 2 newly derived probit functions. And that is the processing of the studies of Kaplan et al. (1988 and 1993). In these studies, experiments are carried out on baboons. However, due to lack of information, these studies are classified as C-studies, as well in (Cornelis et al., 2018) as in (Ruijten, 2017). This means that the results itself from the studies can not be taken into account in the same manner as the other studies with rat and mice to derive the probit function. The only thing that can be done with the results is adapting the extrapolation factors for the animal species.

4.1 Flanders

(Cornelis et al., 2018) states (translated from Dutch): *"The probit function was calculated with the classic interspecies extrapolation factors. In the RIVM substance dossier (Ruijten, 2017), these corrective interspecies extrapolation factors are not applied because the Kaplan study shows that baboons are less susceptible to HCl than rodents. … On the basis of these data, RIVM concludes that primates (and therefore also humans) are less sensitive than rodents, so that extrapolation factors would unnecessarily deepen the probit function.*

Moreover, it concerns only 2 reported values for baboons. In rodent animals too, LC_{50} values are sometimes reported in individual studies, but in several other studies on the same species significantly lower values are found (…). This shows that one has to be careful with data for specific species when only a few studies on the relevant animal species are available. Moreover, the reported effect values for humans do not confirm that humans would have the same sensitivity as baboons. … Therefore, based on the available data, there is insufficient convincing evidence that HCl sensitivity in humans is comparable to that of baboons. The baboon studies alone therefore do not form an adequate basis for omitting or adapting the extrapolation factors, since no further studies are available that can support the comparison between baboon and human with regard to

the sensitivity to HCl. The original … studies are not available, but the data can not be ignored. The values are used by eg. NIOSH when deriving IDLH values. Other institutions that derived reference values also did not charge these studies on baboons.
In order to be able to estimate the impact of the application of the extrapolation factors, the probit function was also calculated without applying the factors. ... The derived probit function therefore underestimates the lethality and is experienced as insufficiently stringent. Based on this, the precautionary principle is applied in this dossier and the probit is derived on the basis of the available A studies on rats and mice, using the classic interspecies extrapolation factors."

4.2 The Netherlands

(Ruijten, 2017) states: *"In addition to the rodent data, the expert panel made the following observations:*

- *3/3 baboons survived a 15-minute exposure to approximately 15,000 mg/m³ HCl (Kaplan et al., 1988), while HCl exposure to 15,000 mg/m^3 (Hartzell et al., 1985) and 12,000 mg/m^3 (Lapin, 1981) for 15 minutes is fatal to 6/6 rats.*
- *The respiratory response of baboons (and therefore maybe in man) is clearly different from that in rats (Kaplan et al., 1988, 1993).*

Based on the primate data from Kaplan et al (1988, 1993), it was argued that the derivation of a probit function from rat and mouse data may overestimate the acute lethality of HCl in non-human primates and therefore most likely also in humans, assuming that baboons are a better model for lethality in humans than rats."
Therefore, the human equivalent LC50 was calculated by applying an extrapolation factor of 1 for animal to human extrapolation, while the default value is 1/3.

4.3 Comparison

To demonstrate the influence of the assumption of whether or not incorporating the studies of Kaplan about baboons, the newly derived probit functions are recalculated with adapted extrapolation factor for Flanders and adapted extrapolation factor for animal to human extrapolation for The Netherlands. The different probit functions are shown in Table 5 and Figure 2. This shows that the influence of this factor is very significant.

Table 5: New probit functions for HCl for Flanders and The Netherlands without and with incorporating the studies about baboons

Country	Probit function without baboons [mg/m³]	Probit function with baboons [mg/m³]
Flanders 2018	$-16.03 + 1.7 \cdot \ln(C^{1.18} \cdot t)$ with $LC_{50,\ 30'}$ = 2,062 mg/m³ Extrapolation factor: see Table 1	$-18.64 + 1.7 \cdot \ln(C^{1.18} \cdot t)$ with $LC_{50,\ 30'}$ = 7,347 mg/m³ Extrapolation factor = 1
The Netherlands 2017	$-14.89 + 1.463 \cdot \ln(C^{1.367} \cdot t)$ with $LC_{50,\ 30'}$ = 1,733 mg/m³ Extrapolation factor = 1/3 (default)	$-17.09 + 1.463 \cdot \ln(C^{1.367} \cdot t)$ with $LC_{50,\ 30'}$ = 5,198 mg/m³ Extrapolation factor = 1

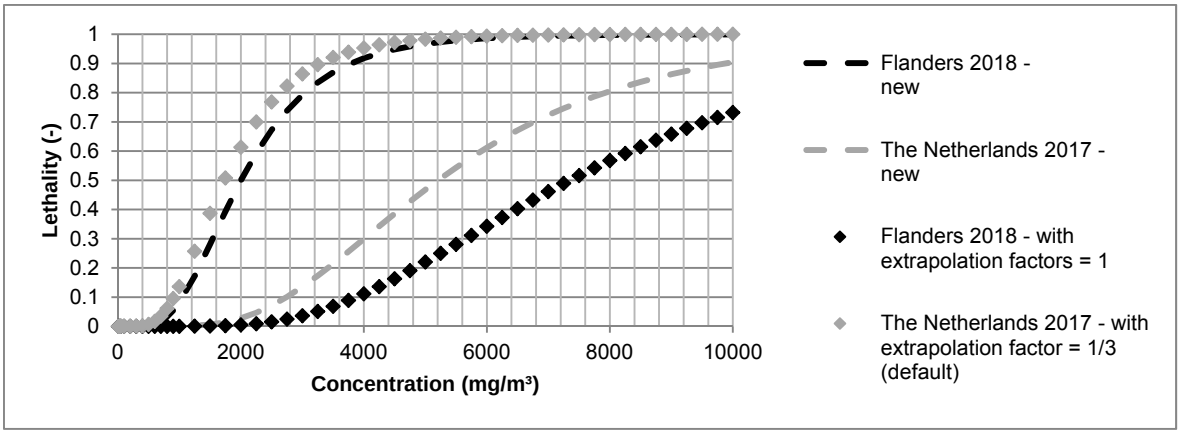

Figure 2: Probit functions for HCl for Flanders and The Netherlands with and without taking into account the studies about baboons

5. Conclusions

The probit functions for humans are derived from experiments on animals with a specific method in which a lot of assumptions have to be made. The result is that different organisations derive different probit functions for the same substance. Comparison of the Flemish and Dutch probit functions for HCl shows that there is a big difference in the resulting probit functions. Looking closer at the method, learns that the difference is due to (1) the classification process of the studies, (2) whether or not taking into account the extra factor of 2 when using studies with multiple animal species, (3) the extrapolation factors for converting animal data to human data and very specifically for HCl, (4) whether or not taking into account the Kaplan-studies about baboons. As these probit functions are used to calculate the risk for Seveso-establishments, which must meet the risk criteria to get an environmental permit, it is important to have a more unified approach.

References

Arts J.H.E., C. Mommers, H. Muijser, 2000, Toxic Effects from Accidental Releases of Hazardous Substances (TEARHS) – Lethal and non-lethal effects in rats upon exposure during short periods of time, TNO Nutrition and Food Research, report V99.11. Zeist.

Bloemen K., Cornelis C., Wouters G., Schoeters G., 2005, Opstellen van toxiciteitsprobitfuncties ten behoeve van veiligheidsrapporten, VITO-rapport 2005/IMS/R/039, Departement Leefmilieu en Infrastructuur, Brussel.

Cornelis C., Geerts L., Weltens R., 2018, Inhalation probit functions – 2017/2018, VITO-rapport 2016/MRG/R/0853 (revised version 2018), Departement Leefmilieu, Natuur en Energie, Brussel.

Darmer K.J., Kinkead E.R., DiPasquale L.C., 1974, Acute toxicity in rats and mice exposed to hydrogen chloride gas and aerosols, Am. Ind. Hyg. Assoc. J., 20 623-631.

Hartzell G.E., Grand A.F., Switzer W.G., 1985, Modeling of toxicological effects of 26 fire gases: VI. Further studies on the toxicity of smoke containing hydrogen chloride, J. Fire Sci., 5: 368-391.

Hooghe R., Wouters G., 2000, Herevaluatie van de toxiciteitsprobitfunctie voor waterstofchloride in het kader van de externe veiligheidsrapportering, VITO rapport 2000/TOX-IMS/R/211, Departement Leefmilieu en Infrastructuur, Brussel.

Jean, PA, RH Gallavan, GB Kolesar et al., 2006. Chlorosilane Acute Inhalation Toxicity and Development of an LC50 Prediction Model. Inhal. Tox., 18,515-522.

Kaplan H.L., Anzueto A., Switzer W.G., Hinderer R.K., 1988, Effects of Hydrogen Chloride on Respiratory Response and Pulmonary Function of the Baboon, J. Toxicol. Environ. Health 1988; 23: 473-493.

Kaplan H.L., Switzer W.G., Hinderer R.K., Anzueto A., 1993, A Study on the Acute and Long-Term Effects of Hydrogen Chloride on Respiratory Response and Pulmonary Function and Morphology in the Baboon, J. Fire Sci. 1993; 11: 459-484.

Lapin C.A., 1981, Inhalation toxicity of common combustion gases, E.I. Dupont de Nemours and Company, Haskell Laboratory for toxicology and Industrial Medicine, report 238-81, Newark.

RIVM, 2015, Handboek Risicoberekeningen BEVI 3.3, Bilthoven.

Ruijten M.M.W.M., Arts J.H.E., Boogaard P.J., Bos P.M.J., Muijser H., Wijbenga A., 2015, Method for derivation of probit functions for acute inhalation toxicity, RIVM Report 2015-0102, National Institute for Public Health and the Environment.

Ruijten M.M.W.M., 2017, Probit function technical support document – hydrogen chloride, RIVM report 20170606-hydrogen chloride-INTERIM.

Ten Berge W., 2015, Doseresp: Concentration-time response in acute inhalation toxicity. home.planet.nl/~wtberge/doseresp.html (as of March 2015).

VROM, 1989, Knelpuntenoverleg EVR, Gebruik toxiciteitsgegevens, KO 24-2, Den Haag, Ministerie van Volkshuisvesting, Ruimtelijke Ordening en Milieu.

VROM, 1990, Groene Boek - Methoden voor het bepalen van mogelijke schade aan mensen en goederen door het vrijkomen van gevaarlijke stoffen, CPR 14, Directoraat-Generaal van de Arbeid, 1990.

VROM, 2005a, Groene Boek - Methoden voor het bepalen van mogelijke schade aan mensen en goederen door het vrijkomen van gevaarlijke stoffen, Publicatiereeks Gevaarlijke Stoffen 1 (Deel 4), Den Haag, Ministerie van Volkshuisvesting, Ruimtelijke Ordening en Milieu.

VROM, 2005b, Paarse Boek - Guidelines for quantitative risk assessment, Publicatiereeks Gevaarlijke Stoffen 3, Den Haag, Ministerie van Volkshuisvesting, Ruimtelijke Ordening en Milieu.

Wohlslagel J., DiPasquale L.C., Vernot E.H., 1976, Toxicity of solid rocket motor 16 exhaust: Effects of HCl, HF, and alumina on rodents. J. Combust. Toxicol. 3: 17 61-70.