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Proposal of new uncertainty factor application to derive tolerable daily intake

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ABSTRACT

We propose new uncertainty factors (UFs) and a new subdivision of default factors in chemical risk assessment using a probabilistic approach based on the latest applicable information. Rounded values of 150 for mice, 100 for hamsters and rats, and 40 for rabbits, monkeys and dogs for inter- and intra-species differences (UF_{AH}) were derived from the probabilistic combination of two log-normal distributions. Further calculation of additional UFs when chronic data (UF_S) or NOAEL (UF_L) are lacking was conducted using available log-normal distribution information. The alternative UF_S and UF_L values of 4 are considered to be appropriate for both cases where data are lacking. The default contributions of inter-species difference (UF_A) and intra-species difference (UF_H) to the UF_{AH} of 100 for hamsters and rats as an example are considered to be 25 and 4, respectively. The UF_A of 25 was subdivided into $25^{0.6}$ (i.e., 7.0) for pharmacokinetics (PK) ($UF_{A,PK}$) and $25^{0.4}$ (i.e., 3.6) for pharmacodynamics (PD) ($UF_{A,PD}$), and the UF_H of 4 was evenly subdivided into $4^{0.5}$ (i.e., 2) ($UF_{H,PK}$ and $UF_{H,PD}$), to account for chemical-specific difference data between humans and laboratory animals for PK and/or PD. These default UFs, which come from actual experimental data, may be more appropriate than previous default UFs to derive tolerable daily intake values.

1. Introduction

Principle uncertainty factors (UFs) consisting of inter-species differences (or extrapolation from laboratory animals to humans, referred to as "UF_A") and intra-species differences (human variability, referred to as "UF_H") have commonly been used when extrapolating from animal experimental data to human risk values in chemical risk assessment. The current combined default UF of 100 ($10_A \times 10_H$) for extrapolation from animal data was introduced in the US in 1954 (Lehman and Fitzhugh, 1954) for food contaminants with a rationale for its suitability for environmental contaminants provided by Dourson and Stara (1983) years later. The physical size of laboratory animals is variable, with animals as small as mice to larger animals like dogs. In some cases the size difference results in more than a 500-fold difference in body weight indicating that some type of variable adjustment might be needed, rather than just a 10-fold factor.

Body surface area correction, (human body weight/animal body weight)$^{1/3}$ was the first data supported size adjustment (Freireich et al., 1966). It has been applied to cancer endpoints in US Environmental Protection Agency (US EPA) assessments and was also used in the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) residual solvent guidelines (Connelly et al., 1997). Recently, allometric scaling according to caloric demand or metabolic size, (human body weight/animal body weight)$^{1/4}$ was introduced as a more appropriate adjustment (Schneider et al., 2004) and is currently used by US EPA in cancer risk assessment (US EPA, 1992, 2005a). Size adjustment might be more appropriately based on allometric scaling as discussed by Falk-Filipsson et al. (2007). However, the use of allometric scaling in non-cancer endpoints remains untested by US EPA and other organizational assessments. The caloric demand adjustment factor for a mouse (0.030 kg) or a dog (16 kg) compared to a human (70 kg) based on body weight is 7 or 1.4, respectively, which is significantly lower than the default of 10. However, Schneider et al. (2004) demonstrated that caloric demand scaling was effective for predicting median differences between humans and animals on the basis of body weight in maximal tolerated dose (MTD) ratios of anti-cancer drugs, and also calculated the combined geometric standard deviation (GSD) of the empirical distribution.

Useful experimental data are quite limited for human intra-species differences, specifically variability between different ages (Dourson and Stara, 1983; Dourson et al., 1996, 2002). However, some insights can be gained from experimental animal work. For

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example, a recent comparative investigation of no observed adverse effect levels (NOAELs) in repeat-dose studies of newborn and young rats for 18 chemicals was conducted (Hasegawa et al., 2007). The data provided the median and distribution of NOAEL ratios.

The default UF of 100 for inter- and intra-species differences is typically applied by multiplication of 10_A and 10_H. However, the default UF of 100 is not always appropriate to use. For example, multiplication of two log-normal distributions for inter- and intra-species differences also produces a log-normal distribution, and simple multiplication such as 10×10 causes overestimation if both individual values are in the 95th percentile. Kodell and Gaylor (1999) recommended standard statistical techniques that could be used to estimate the upper tolerance limits on the distribution of sums which can also be used for other UFs (e.g., the ratio of subchronic to chronic NOAELs). Swartout et al. (1998) also addressed this problem and gave hypothetical examples of UF combinations.

Another method for division of the default UF of 100 ($10_A \times 10_H$) for inter- and intra-species differences was proposed by Renwick (1993). He proposed a subdivision of these UFs into two parts, pharmacokinetics (PK) and pharmacodynamics (PD). Based on his analysis of experimental data and physiological parameters between animals and humans, the contribution ratios of PK and PD are 60:40 for inter-species differences and 50:50 for intra-species differences, leading to $10^{0.6}$ (4.0) $\times$ $10^{0.4}$ (2.5) and $10^{0.5}$ ($\sim$3.2) $\times$ $10^{0.5}$ ($\sim$3.2), respectively (IPCS, 1994). When chemical-specific data for the differences between animals and humans for PK and/or PD are available, the data should be used to develop chemical-specific adjustment factors instead of the default PK/PD factors (WHO, 2005). However, the default subdivision factors should be re-estimated if animal size-specific UFs are adopted as inter-species differences.

In this article, we propose new default UFs by a probabilistic approach using appropriate log-normal distribution data, taking animal size into consideration. We also propose development of new default values according to animal size for the subdivision of inter- and intra-species differences.

2. Data for each uncertainty

2.1. Inter-species difference data

Eight publications featuring chemical toxicity comparisons between humans and laboratory animals for anti-cancer drug toxicity were located. The first study by Freireich et al. (1966) showed MTD differences between humans and five animal species (mice, hamsters, rats, monkeys and dogs) in the analysis of 18 drugs. Recently, Schneider et al. (2004) extracted correlated human and animal data sets for 63 anti-cancer drugs from six additional publications (Goldsmith et al., 1975; Schein et al., 1979; Travis and White, 1988; Rozencweig et al., 1981; Grieshaber and Marsoni, 1986; Paxton et al., 1990) to demonstrate that caloric demand scaling was a suitable adjustment factor for the differences of inter-species median MTDs. Schneider et al. (2004) also derived a GSD of 3.23 from the combined distribution of all MTD ratios for humans versus the five animal species stated above.

Alternatively, inter-species differences in susceptibility could be derived based on the differences in NOAELs rather than MTDs. Schneider et al. (2004) also analyzed inter-species differences for pesticide NOAELs between mice/rats, rats/dogs and mice/dogs, providing further support to the caloric demand adjustment. Therefore, the median and GSD derived from MTD ratios of anti-cancer drugs might be equivalent to those based on NOAEL ratios between humans and animals. No other publications featuring an estimated direct comparison of chemical toxicity between humans and animals were identified.

2.2. Intra-species difference data

The NOAEL ratio of a sensitive subpopulation compared to that of the general population is a source of uncertainty for intra-species differences in risk assessment (Dourson et al., 2002). Occasionally the sensitive subpopulations are directly addressed in the risk assessment. For example, the Reference Dose (RfD) for nitrate on the Integrated Risk Information System (IRIS) used methemoglobinemia in children as the critical effect, therefore an intra-species UF may not be needed (US EPA, 2009). However, in most cases, the sensitive subpopulations are only considered protected with the use of an intra-species UF. Generally, infants, pregnant women, the elderly and other specified groups are considered high-susceptibility groups, although exceptions are not uncommon. For example, Tylenol overdose is more of a problem in adults than in children because the toxic metabolite is more readily formed in adults. Effects during pregnancy and gestation are considered to be adequately evaluated in the reproductive/developmental toxicity studies, while lifetime toxicity studies cover the potential for effects to the elderly. For a well-tested chemical, the only remaining sensitive subpopulation to be protected by an intra-species UF are infants. Currently, there are no experimental animal test guidelines intended for direct exposure of neonatal animals to chemicals. Other specified groups may include patients exhibiting hepatic or renal dysfunction and persons with a specific genetic background. These subpopulations need specific risk management and should not be the target population for a chemical risk assessment for public health because it is possible that their susceptibility to specific chemicals may be unexpectedly high owing to significantly reduced metabolism or excretion of toxic substances.

The comparative data between human adults and children/infants was assessed by many scientists. Glaubiger et al. (1981) compared MTDs in patients for 17 anti-cancer drugs demonstrating that children's MTDs were 50% higher than those of adults, indicating that children were less sensitive. Calabrese (1985) investigated the variation in physiological response to exogenous stress in humans, and judged that 80–95% of the variation in a human group for a given agent was less than 10-fold. Hattis et al. (1987) analyzed 101 PK parameter data sets for 49 substances (mostly medications) and showed that 96% of the human variation was also less than 10-fold. Ginsberg et al. (2002) compared PK in adults and children using a database of approximately 45 medications, and showed that the half-lives of medications for 1-week to 2-month old infants were twice as long as the half-lives in adults. Hattis et al. (2003) also showed significantly longer half-lives of medications in infants and children compared to adults.

Animal data has also been reviewed. Dourson and Stara (1983) analyzed acute rat toxicity data for 490 substances reported by Weil (1972). They concluded that the LD_{50}/non-lethal dose ratio for 92% of the chemical substances would be less than 10. In a meeting abstract, Sheehan and Gaylor (1990) stated that the LD_{50} of 238 substances in adult rats was about 2.6 times higher than the LD_{50} in newborn rat pups, and the LD_{50}/LD_{50} ratio for 86% of substances was less than 10. Calabrese (2001) showed that the LD_{50} in younger animals was within a 10-fold range of older animals for 86.3% of 313 substances. Charnley and Putzrath (2001) examined the influence of age on carcinogenesis caused by chemicals, but were unable to reach a clear conclusion. Similarly, the US EPA considered the effect of age in their most recent guidelines for carcinogen risk assessment. They estimated the geometric mean ratio of early-life to adult cancer potencies was 10.4 based on repeated and lifetime exposure data in the available scientific literature for six chemicals acting through a mutagenic mode of action

(US EPA, 2005b; Barton et al., 2005). As for chemicals causing cancer through other modes of action, the ratio was 3.4 for lifetime exposure (5 chemicals) and 2.2 for repeated exposure (6 chemicals).

The quantitative human and experimental animal data for severe endpoints and kinetic parameters are useful. However, a study design similar to the repeat-dose exposure studies used in risk analysis would be ideal to derive an intra-species UF. The UF is applied to the NOAEL derived from the results of repeated dose toxicity studies, therefore a comparative analysis of NOAELs from repeat-dose toxicity studies of newborn and young rats for 18 chemicals was considered more appropriate (Hasegawa et al., 2007). In this study, Hasegawa et al. (2007) strictly compared the NOAEL ratios for newborn and young rats in a repeat-dose study. The NOAEL ratios were log-normally distributed. The ratio median was 3, and 5 was equivalent to 94.4% of the whole data set, from which the GSD can be calculated (see below).

2.3. Data for supplemental uncertainty factors[1]

The appropriate adjustment from short-term NOAEL to lifetime NOAEL for risk assessment was evaluated using 33 data sets of subchronic (3 months)/chronic (2 years) NOAELs in rats and mice reported by Weil and McCollister (1963) and 68 additional data sets from analyses of published reports or papers that we previously summarized (Hasegawa, 1991). Comparison of NOAELs from published 3-month and 2-year repeated dose toxicity studies, unpublished data). The combined data sets yielded a median of 1.7 with a GSD of 3.30. If only a LOAEL was identified, the median LOAEL/NOAEL ratio of 3.5 with a GSD of 1.82 from Abdel-Rahman and Kadry (1995) from other chemicals can be adapted as an UF for this area, with the usual upper bound value of 10. However, it is recognized that the application of the benchmark dose approach is usually more appropriate in cases where only a LOAEL is available, and as such this UF is not used as frequently.

3. Calculation of new uncertainty factors based on experimental data by probabilistic approach, an example of rats

The distribution of both inter- and intra-species differences is log-normal because each component consists of the NOAEL ratios for two groups. If the default values of 10 are used, simple multiplication of 10 by 10, resulting in 100, leads to overestimation for the 95th percentile of the combined distribution, more appropriately it should be 51, as shown by Monte Carlo simulation (Swartout et al., 1998). Generically, the *N*th percentile of a log-normal distribution can be expressed as *N*th percentile = Exp [LN (median) + $\alpha_n \times$ LN (GSD)]. For the 95th percentile, $\alpha_n = 1.645$. The equation for the combination of two log-normal A and B distributions can be shown as follows: 95th percentile of (A × B) = Exp [LN (median_A) + LN (median_B) + $1.645 \times ((\text{LN}(\text{GSD}_A))^2 + (\text{LN}(\text{GSD}_B))^2)^{0.5}$] (Kodell and Gaylor, 1999).

Inter-species differences were calculated using an analytical method presented by Schneider et al. (2004). A median of 4 was reported for the caloric demand adjustment, rounded from $3.76 = (70/0.35)^{1/4}$ (70 kg human body weight and 0.35 kg that of rats). A GSD of 3.23 was adopted from a combined distribution of MTD ratio for humans versus the 5 animal species previously described. For the 95th percentile, $\alpha_n = 1.645$.

LN (95th percentile) = LN (4) + 1.645 × LN (3.23)

95th percentile = UF (95%) = Exp [1.39 + 1.645 × 1.17] = 27.5.

Intra-species differences were calculated using rat young/newborn NOAEL ratios in repeat-dose toxicity studies (Hasegawa et al., 2007). The median was 3 for 18 data sets and 5 was equivalent to 94.4% of all the data sets. For the 94.4th percentile, $\alpha_n = 1.590$.

LN (5 as 94.4th percentile) = LN (3) + 1.590 × LN (GSD)
Rearranging,
LN (GSD) = (1.61 − 1.10)/1.590 = 0.321
Therefore,
GSD = Exp [0.321] = 1.38
95th percentile = UF (95%) = Exp [1.10 + 1.645 × 0.321] = 5.09.

From the above data for inter- and intra-species differences, the combined UF_{AH} was calculated as follows:

$\text{LN}\ (4) + \text{LN}\ (3) + 1.645 \times ((\text{LN}\ (3.23))^2 + (\text{LN}\ (1.38))^2)^{0.5} = 1.39 + 1.10 + 1.645 \times (1.17^2 + 0.321^2)^{0.5} = 4.48$
Exp[4.48] = 88.7.

For adjustment of short-term NOAEL to lifetime NOAEL, all 101 data sets of subchronic NOAEL/chronic NOAEL were used. The median was 1.7 with 10 equivalent to 93.1% of all the data sets. For the 93.1th percentile, $\alpha_n = 1.483$.

LN (10 as 93.1th percentile) = LN (1.7) + 1.483 × LN (GSD)
Rearranging,
LN (GSD) = (2.30 − 0.531)/1.483 = 1.20
Therefore,
GSD = Exp [1.20] = 3.30
95th percentile = UF (95%) = Exp [0.531 + 1.645 × 1.20] = 12.1.

From the above UF calculations, the combined UF_{AHS} was calculated as follows:

$1.39 + 1.10 + 0.531 + 1.645 \times (1.17^2 + 0.321^2 + 1.20^2)^{0.5} = 5.82$
Exp[5.82] = 337.

If a benchmark dose approach cannot be applied, an additional UF should be applied when using LOAEL data. The LOAEL/NOAEL ratio for 24 chemicals was reported by Abdel-Rahman and Kadry (1995). The median was 3.5 and 10 was equivalent to 96% of the whole data. For the 96.0th percentile, $\alpha_n = 1.751$.

LN (10 as 96.0th percentile) = LN (3.5) + 1.751 × LN (GSD)
Rearranging,
LN (GSD) = (2.30 − 1.25)/1.751 = 0.600
Therefore,
GSD = Exp [0.600] = 1.82
95th percentile = UF (95%) = Exp [1.25 + 1.645 × 0.600] = 9.39.

From the above UF calculations, the combined UF_{AHSL} was calculated as follows:

$1.39 + 1.10 + 0.531 + 1.25 + 1.645 \times (1.17^2 + 0.321^2 + 1.20^2 + 0.6^2)^{0.5} = 7.24$
Exp [7.24] = 1400.

4. Summary of combined uncertainty factors for six animal species by probabilistic approach

All fundamental values for the median, GSD and UF (95%) are shown in Table 1. The median for inter-species differences was derived using caloric demand adjustment from the standard human and animal body weights and rounded to a simple value. The

[1] The uncertainty factor used by several organizations for missing certain studies in the database (e.g., Dourson et al., 1992, 2002) was not considered here at this time, as it is being studied for applicability in Japan.

Table 1
Median, GSD and UF (95%) of inter-species differences for 6 animal species and other uncertainties.

	Median	GSD	UF (95%)
Inter-species differences (caloric demand)[a]			
Mice to humans	6.95 → 7		48.2
Hamsters to humans	4.86 → 5		34.4
Rats to humans	3.76 → 4	3.23	27.5
Rabbits to humans	2.04 → 2		13.8
Monkeys to humans	1.77 → 1.8		12.4
Dogs to humans	1.44 → 1.4		9.63
Intra-species differences[b]	3.0	1.38	5.09
Subchronic to chronic[b]	1.7	3.30	12.1
LOAEL to NOAEL[b]	3.5	1.82	9.39

[a] Use of caloric demand and distribution from MTD ratios of 63 anti-cancer drugs between humans and 5 animals given by Schneider et al. (2004). Medians were calculated as caloric demand adjustment ((human body weight/animal body weight)$^{1/4}$) on the bases of body weight: humans = 70 kg, mice = 0.03 kg, hamsters = 0.125 kg, rats = 0.35 kg, rabbits = 4 kg, monkeys = 7 kg and dogs = 16 kg.
[b] Calculation details are shown in the previous section.

GSD for inter-species differences was obtained by combining the distribution of all the MTD data sets. This distribution may contain some additional, but unquantifiable, conservatism since humans are more heterogeneous than laboratory animals; thereby inflating the upper limits. The 95th percentile of UFs for six laboratory animal species ranged from approximately 10–50, a 5-fold difference.

All possible cases of UFs for six laboratory animal species were calculated by a probabilistic approach (Table 2) using the values from Table 1. The UF_{AH} for each animal is calculated by combining inter- and intra-species differences. We propose a rounded UF_{AH} of 150 for mice, 100 for hamsters and rats, and 40 for rabbits, monkeys and dogs. Additional single UFs for either subchronic to chronic (UF_S) or LOAEL to NOAEL (UF_L) extrapolation, resulted in a 3.8-fold increase for the UF_{AHS} from the UF_{AH} and a 4.4-fold increase for the UF_{AHL} from the UF_{AH}, giving UFs approximately 4-fold higher than the UF_{AH} in either case. Finally, the four combined UFs, UF_{AHSL}, when chronic data and NOAEL are lacking, resulted in a 16-fold increase from the UF_{AH}. All the UFs obtained by Monte Carlo simulation, based on the default UF of 10, are slightly lower than our proposed UFs for rats. Simple multiplication of the default value of 10, resulted in much larger values than all three or four combined UFs (UF_{AHS}, UF_{AHL}, UF_{AHSL}) for all animals.

5. Application of subdivision and replacement of uncertainty factors for inter- and intra-species differences (chemical-specific adjustment factors)

In the present article, we propose animal size-specific inter-species UFs and new combined UFs (UF_{AH}) by using probabilistic approaches. For the cases of hamsters and rats, UF_{AH} is set at 100 but the contributions of inter- and intra-species differences are not equal. The application of the same default subdivision factor shown by Renwick (1993) is not appropriate, if the UF_A values of Table 2 are used as the basis of the assessment. However, the concept established by Renwick (1993) is appropriate because we also recommend that actual and reliable experimental data for PK or PD differences should be incorporated into the risk assessment processes wherever possible. Therefore, we subdivided the new UF_A to determine the contribution ratio of inter- and intra-species differences. In the case of hamsters and rats, the average UF_A is approximately 30 (hamsters = 34.4 and rats = 27.5) and the intra-species difference is 5.09, (calculated above from the Hasegawa et al. (2007) data), resulting in a ratio contribution of ~6:1. The UF_{AH} for hamsters and rats is set at 100, which can be divided into factors of 25 and 4, according to the above ratio of 6:1. Considering the contribution ratios of PK and PD as 60:40 for inter-species differences and 50:50 for intra-species differences, 25 will be subdivided into $25^{0.6}$ (7.0) for PK and $25^{0.4}$ (3.6) for PD, and 4 will be evenly subdivided into $4^{0.5}$ (2) (Table 3).

Similar approaches can be used elsewhere. For example, the mice UF_{AH} of 150 can be divided into 38 and 4, then 38 will be subdivided into $38^{0.6}$ (9.0) for PK and $38^{0.4}$ (4.3) for PD. For rabbits, monkeys and dogs, the UF_{AH} of 40 can be divided into 10 and 4, then 10 will be subdivided into $10^{0.6}$ (4.0) for PK and $10^{0.4}$ (2.5) for PD.

If actual data for the difference between humans and animals for PK and/or PD are available, those data can be used as chemical-specific adjustment factors instead of respective default subdivision factors.

Table 2
Combined UFs for six animal species by probabilistic approach (95th percentile), Monte Carlo simulation and simple multiplication of UF 10.

Species	UF_A	UF_{AH}	UF_{AHS}	UF_{AHL}	UF_{AHSL}
Mice	48.2	155	589	684	2440
Hamsters	34.4	111	421	488	1740
Rats	27.5	88.7	337	391	1400
Rabbits	13.8	44.3	168	195	698
Monkeys	12.4	39.9	152	176	628
Dogs	9.63	31.0	118	137	488
All animals					
Monte Carlo[a]	10	51	234	234	1040
Default[b]	10	100	1000	1000	3000

A, inter-species differences; H, intra-species differences; S, subchronic to chronic; L, LOAEL to NOAEL.
[a] Data from Swartout et al. (1998).
[b] Note that US Environmental Protection Agency (USEPA) combines the default values of 4 UFs into 3000, because of the generally conservative nature of combining 10-fold factors that are each somewhat conservative (Dourson, 1994).

6. Discussion

The proposed written document to address chemical safety assessment methodology is needed because officially agreed upon guidelines do not exist in Japan. For this purpose, the latest scientific information has been collected to reduce the uncertainty in the risk assessment process. It would be more reliable for UFs to be estimated on the basis of actual experimental data rather than use conventional default UFs. Furthermore, the values are more representative of the data if they are developed using statistical components such as the median with distribution of differences rather than point estimates. A tolerable daily intake can be derived by probabilistic approaches, using the median or geometric mean (GM) and GSD to combine two or more distributions.

Recently, Falk-Filipsson et al. (2007) reviewed a wide variety of assessment factors in various historical and scientific ranges, including guidelines from national and international bodies. They reported that "over-conservatism" should be avoided by using a probability distribution for the various assessment factors. However, such an approach was only applied to the UF for inter-species

Table 3
Subdivision of uncertainty factors for inter- and intra-species differences.

Species	UF_{AH}	UF_A UF_H	Subdivision PK × PD
Mice	150	38 4	9.0 × 4.3 2 × 2
Hamsters Rats	100	25 4	7.0 × 3.6 2 × 2
Rabbits Monkeys Dogs	40	10 4	4.0 × 2.5 2 × 2

Table 4
Median or GM with GSD for each uncertainty in four different methodologies.

	Inter-species differences		Intra-species differences		Subchronic to chronic		LOAEL to NOAEL	
	Median/GM	GSD	Median/GM	GSD	Median/GM	GSD	Median/GM	GSD
Baird et al. (1996) (GM)	AF[a]	4.9	2.7	2.3	2.0	2.1	3.4	1.70
Swartout et al. (1998)	10[b]		10[b]		10[b]		10[b]	
Kodell and Gaylor (1999) (median)	1	5.27	1	5.15	2	3.67	3.5	1.82
Present experiment (median)	AF[c]	3.23	3.0	1.38	1.7	3.30	3.5	1.82

[a] Adjustment factor for each animal on the basis of body surface correction.
[b] Use of 10 for every traditional default factor.
[c] Adjustment factor for each animal on the basis of caloric demand.

differences because appropriate distribution data for intra-species differences could not be located.

This study is the fourth trial following those of Baird et al. (1996), Swartout et al. (1998) and Kodell and Gaylor (1999) to use a probabilistic approach to estimate UFs for chemical risk assessment. Table 4 shows the median/GM and GSD for the four methodologies and Table 5 shows combined UFs for inter- and intra-species differences, and two other uncertainties. Swartout et al. (1998) estimated four UFs by Monte Carlo simulation using a traditional default UF of 10 for each uncertainty. Baird et al. (1996) also performed Monte Carlo simulation with specific software, but used actual data instead of default values. On the other hand, Kodell and Gaylor (1999) used standard statistical techniques, as we do here. Key differences in the three methodologies result from the original data used for inter- and intra-species extrapolation. For inter-species differences, the data used in this assessment are considered appropriate because the data are a direct comparison between humans and animals (Schneider et al., 2004). However, Baird et al. (1996) used comparative data within laboratory animals from pesticide safety studies (Dourson et al., 1992) and Kodell and Gaylor (1999) used toxicity comparisons of marine-life LD_{50} (Calabrese and Baldwin, 1995).

A similar analysis can be done for intra-species differences. This assessment used comparative NOAEL data from newborn and young rat repeat-dose studies as a sensitive subpopulation compared to the general population (Hasegawa et al., 2007). However, the other groups (Baird et al., 1996; Kodell and Gaylor, 1999) used lethality distribution data from acute toxicity studies (Dourson and Stara, 1983).

The different methodologies resulted in similar UF_{AH} values for Kodell and Gaylor (1999) and Baird et al. (1996), but were different from Swartout et al. (1998), as shown in Table 5. However, the Baird et al. (1996) UF_{AH} does not include a scaling adjustment factor, thus the median of inter-species differences of Baird's data was calculated as 1. As presented in this assessment, the body surface area correction factor, such as 13.3 for mice, 5.8 for rats, and 1.6 for dogs, should be used to reduce the uncertainty. This assessment calculated the expected UFs for rats using Baird et al. (1996) data (found in Table 4) and using the standard statistical techniques described in the previous sections of this paper. The results of these calculations are shown as "Baird et al., 1996 Our Calc" in Table 5.

Table 5
Combined UFs at 95% confidence limit by four methodologies.

UFs	Baird et al. (1996)[a]	Baird et al. (1996)[b]Our Calc	Swartout et al. (1998)[c]	Kodell and Gaylor (1999)[c]	Present study[b]	Default[c]
U_{AH}	50	300	51	46	89	100
U_{AHS}	126	764	234	161	337	1000
U_{AHL}	192	1156	234	184	400	1000
U_{AHSL}	484	2920	1040	629	1400	3000

[a] Not including inter-species scaling.
[b] Specific to rats.
[c] For all laboratory animals.

The calculated values were almost six times larger for each UF than those without the scaling adjustment factor (Baird et al., 1996 in Table 5). The calculated UFs in this assessment are relatively similar to Swartout et al. (1998) and much smaller than the default UF values.

The actual data used for our probabilistic estimation of the four UFs are considered suitable at this moment, and the combined UF_{AH} values for several commonly used laboratory animal species were given by standard statistical techniques (Table 2). However, as a rounded value is preferred for risk assessment, we propose size-specific UFs of 150 for mice, 100 for hamsters and rats, and 40 for rabbits, monkeys and dogs. As for other UFs such as UF_{AHS}, UF_{AHL} and UF_{AHLS}, the average uncertainty values for each (UF_{AHS}/UF_{AH}, UF_{AHL}/UF_{AH} and UF_{AHLS}/UF_{AH}) were 3.8, 4.4 and 15.7, respectively. Therefore, we propose to uniformly use a factor of 4 when a NOAEL (UF_L) and/or chronic data (UF_S) is lacking.

The application of an alternative subdivision of UFs should be considered in order to address the new concept of including animal size-specific UFs in the contribution of inter- and intra-species differences. The values of the new subdivision described in this study may be too precise, but this is inevitable, because the contribution of inter- and intra-species differences is definitively different. When further data on human and animal PK/PD differences are available, a more practical risk assessment can be implemented.

7. Conclusions

We propose an animal size-specific UF for UF_{AH} of 150 for mice, 100 for hamsters and rats, and 40 for rabbits, monkeys and dogs, for inter- and intra-species differences using a probabilistic approach. An additional default factor of 4 could be applied for either lack of chronic data or lack of a NOAEL. In addition to the proposed animal size-specific UFs, new subdivided PK/PD default factors for each animal are also proposed according to the different contribution of inter- and intra-species differences.

Conflict of interest statement

The authors declare that there are no conflicts of interest.

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