

Lung Dosimetry and Risk Assessment of Nanoparticles: Evaluating and Extending Current Models in Rats and Humans

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Risk assessment of occupational exposure to nanomaterials is needed. Human data are limited, but quantitative data are available from rodent studies. To use these data in risk assessment, a scientifically reasonable approach for extrapolating the rodent data to humans is required. One approach is allometric adjustment for species differences in the relationship between airborne exposure and internal dose. Another approach is lung dosimetry modeling, which provides a biologically-based, mechanistic method to extrapolate doses from animals to humans. However, current mass-based lung dosimetry models may not fully account for differences in the clearance and translocation of nanoparticles. In this article, key steps in quantitative risk assessment are illustrated, using dose-response data in rats chronically exposed to either fine or ultrafine titanium dioxide (TiO₂), carbon black (CB), or diesel exhaust particulate (DEP). The rat-based estimates of the working lifetime airborne concentrations associated with 0.1% excess risk of lung cancer are approximately 0.07 to 0.3 mg/m³ for ultrafine TiO₂, CB, or DEP, and 0.7 to 1.3 mg/m³ for fine TiO₂. Comparison of observed versus model-predicted lung burdens in rats shows that the dosimetry models predict reasonably well the retained mass lung burdens of fine or ultrafine poorly soluble particles in rats exposed by chronic inhalation. Additional model validation is needed for nanoparticles of varying characteristics, as well as extension of these models to include particle translocation to organs beyond the lungs. Such analyses would provide improved prediction of nanoparticle dose for risk assessment.

The overall objective of the research pertaining to this article is to develop a scientifically reasonable strategy to estimate the risk of exposure to nanoparticles (also known as ultrafine

particles)* in the workplace. Few studies are available to assess the risk of engineered nanoparticles, including limited toxicological studies and no epidemiological studies. Thus, it is useful to analyze the dose-response data of inhaled fine and ultrafine particles in rodents for use in quantitative risk assessment (QRA)

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*The terms *ultrafine particle* and *nanoparticle* are often used interchangeably, although nanoparticle generally refers to engineered particles. Both ultrafine particles and nanoparticles have primary particle diameters of less than 0.1 μm, and are respirable (i.e., capable of depositing in the gas-exchange region of the lungs), as are fine particles (0.1–2.5 μm) and coarse particles (2.5–10 μm) in humans.

in humans. Established QRA methods can be applied to the available animal dose-response data to provide risk estimates of exposure to those particles, and to provide a basis for comparison and evaluation of possible adverse health risks from exposure to engineered nanoparticles.

The existing risk assessment paradigm (National Research Council [NRC], 1983) provides a scientifically based framework for the risk assessment of substances that may be encountered in the workplace or other environments. The risk assessment steps in the NRC paradigm include: (1) hazard identification, (2) exposure assessment, (3) dose-response assessment, and (4) risk characterization. Quantitative risk assessment is defined as “the estimation of the severity and likelihood of adverse responses associated with exposure to a hazardous agent” (Piegorsch & Bailer, 2005). A fundamental step in QRA is the dose-response assessment, which is performed using statistical and/or biologically-based models. These models provide estimates of the risk associated with a specified dose, or the dose associated with a specified risk. The benchmark dose (Crump, 1984) is an example of the latter approach.

The extrapolation from the rat lung doses to the human-equivalent lung doses and airborne exposures involves adjustment for species differences in the relationship between external exposure and internal dose (Brown et al., 2005; Jarabek et al., 2005). Extrapolation based on allometric relationships between species is often used in risk assessment in the absence of internal dose data. Lung dosimetry models provide a biologically-based, mechanistic approach to predict the fate of inhaled particles, by describing the physical and physiological factors that influence the deposition, clearance, and retention of inhaled particles.

In this article, we illustrate the quantitative risk assessment steps for estimating human-equivalent exposures using rat dose-response data. Both allometric and lung dosimetry model-based methods are illustrated. We also compare observed and predicted particle lung burdens in rats, and discuss issues and research needs for rat and human lung dosimetry modeling of nanoparticles. Since the methods illustrated here are based on established risk assessment practices, this approach would be useful for estimating disease risks from other inhaled particles for which rodent dose-response data are available, and for evaluating steps in the risk assessment process for which improved models may be required.

METHODS

Excess Risk and Benchmark Dose

The excess risk of lung cancer associated with either the airborne mean particle mass concentration or the retained lung dose (as particle surface area or mass) was estimated using chronic inhalation data in rats exposed to poorly soluble particles (fine or ultrafine TiO₂, ultrafine carbon black [CB], or ultrafine diesel exhaust particulate [DEP]). Excess (or added) risk at dose X , $ER(X)$, is defined here as the difference in the tumor propor-

tion in the exposed, $P(X)$, and the unexposed or control, $P(0)$, rats at the end of a 2-yr study, that is, $ER(X) = P(X) - P(0)$.

A critical lung dose (i.e., benchmark dose) was identified as the estimated retained dose associated with a specified excess risk. The benchmark dose (BMD) is defined as “a statistical lower confidence limit on the dose corresponding to a small increase in effect over the background level” (Crump, 1984). More recently, the 95% lower confidence limit of the benchmark dose is referred to as the BMDL, while the point estimate, such as the maximum likelihood estimate (MLE), is called the BMD (U.S. EPA, 2003).

QRA Steps

The steps in a QRA using animal dose-response data, as illustrated in this article, include the following:

1. Select the animal model, dose metric, and disease response. The analyses in this article are based on chronic inhalation data in rats exposed to fine or ultrafine TiO₂, ultrafine CB, or ultrafine DEP (Lee et al., 1985; Mauderly et al., 1987; Muhle et al., 1991; Heinrich et al., 1995; Nikula et al., 1995). The dose metric used is either (a) the retained particle surface area dose in rat lungs (which is estimated from the measured mass dose in the lungs and the specific surface area [m²/g] of the bulk particle material); (b) the retained mass lung dose; or (c) the average airborne exposure concentration in the chronic inhalation study. The response evaluated is lung cancer.
2. Analyze the dose-response relationships to estimate a critical dose, that is, the dose associated with a specified risk level of an adverse response. The statistical dose-response model used here is the multistage model to estimate the BMD and BMDL at 10% excess risk of lung cancer, with linear extrapolation to 0.1% excess risk (U.S. EPA, 2003).*
3. Extrapolate the critical dose (BMD, BMDL) in animals to humans by adjusting for species differences in lung mass or lung surface area. In the absence of other data, an equal response is assumed at an equivalent (or normalized) dose in both species (Brown et al., 2005; Jarabek et al., 2005).
4. Determine the human external exposure (e.g., working lifetime average exposure concentration) that is equivalent to the animal-based critical dose. For the BMD and BMDL based on mean airborne exposure concentrations, the species differences in inhalation rate, lung mass, and exposure conditions are taken into account. For the BMD and BMDL based on retained lung dose, human lung dosimetry models are used to estimate the working lifetime exposure concentrations that would yield those retained doses.

*This approach was used because the BMD and BMDL estimates at 10% excess risk are essentially model independent—i.e., the various models in the BMD software (U.S. EPA, 2003), including the multistage model used here, provide similar estimates. Also, model-based extrapolation beyond the range of the data is not required; since those estimates would vary depending on the model used, linear dose response is assumed below 10% excess risk.

Extrapolation of Rat BMDL to Humans: Lung Dose

In QRA steps 2 and 3, the dose metrics of particle surface area or mass dose retained in the lungs were normalized across species (from different rat strains to humans) by adjusting for differences in lung weights. In the BMD model for TiO_2 , the retained particle surface area dose (per gram lung) was used because it was required to adequately describe the dose-response data of both the fine and ultrafine TiO_2 (Figure 1a). After obtaining the BMD and BMDL for TiO_2 , it was necessary to convert the surface area doses to mass doses of fine or ultrafine TiO_2

(using the specific surface area of each) in order to later estimate the human-equivalent airborne mass concentrations. The ultrafine CB or ultrafine DEP data were adequately described using particle mass dose in the lungs.

In QRA step 4, two different human lung dosimetry models (CIIT & RIVM, 2002; Kuempel et al., 2001a) were used to estimate the human-equivalent airborne mass concentrations associated with the human-equivalent BMDL, as retained lung mass, based on the rat data. The multiple-path particle deposition (MPPD) model (CIIT & RIVM, 2002) includes the International

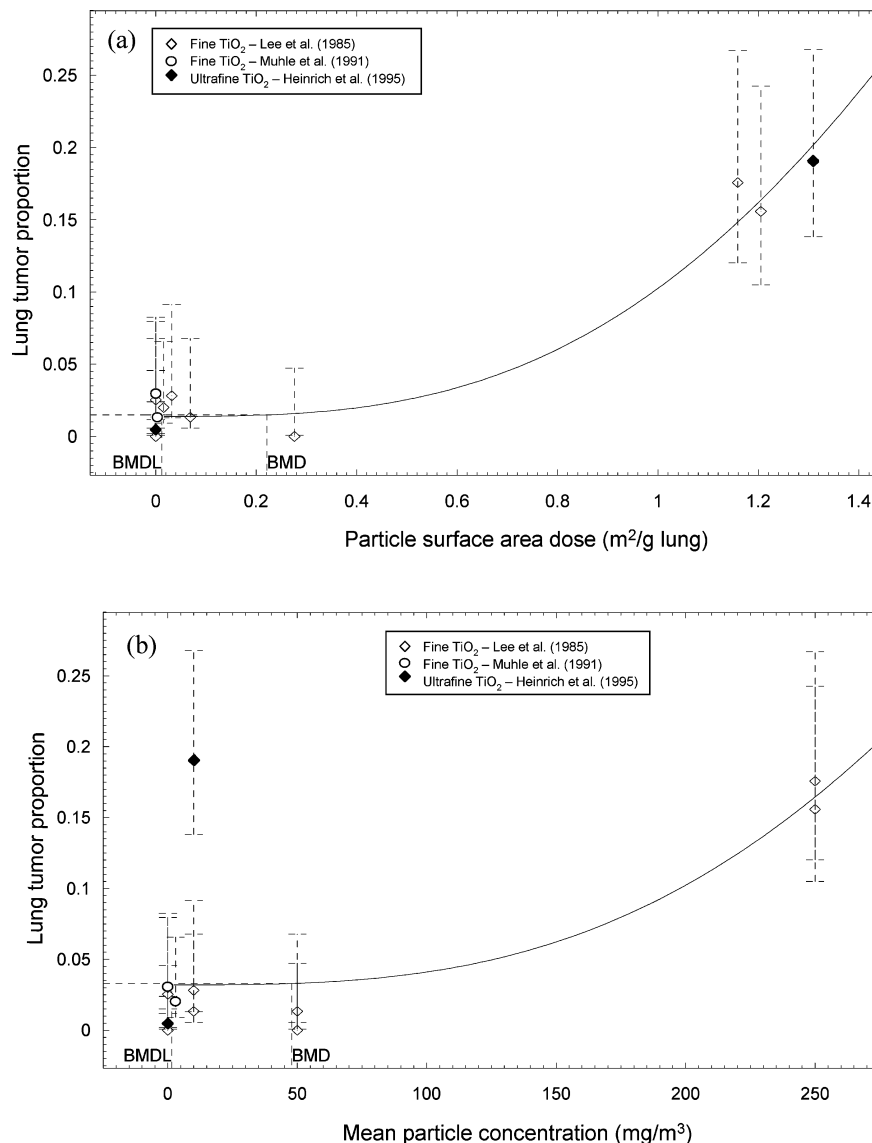


FIG. 1. Relationship between lung tumor proportion and (a) particle surface area dose retained in lungs or (b) mean airborne particle concentration—in rats exposed to fine or ultrafine titanium dioxide by inhalation for 2 yr. (Tumor proportion is the number of rats with tumors in a dose group divided by the total number of rats in that dose group.) Benchmark dose estimates shown here are associated with 0.1% excess risk (estimated directly from a multistage model, third-degree polynomial), and are provided for illustration only. Adequate model fit was not achieved in (b); thus, BMD and BMDL estimates are unreliable. See Methods for approach used. BMD, benchmark dose point estimate; BMDL, lower 95% confidence limit of BMD.

Commission on Radiological Protection (ICRP, 1994) clearance and retention model, and the Yeh and Schum (1980) lung geometry was selected for the deposition estimates. The interstitial/sequestration model (Kuempel, 2000; Kuempel et al., 2001a, 2001b; Tran & Buchanan, 2000) is a three-compartment model of the long-term clearance and retention of respirable particles, which was developed using data of two independent coal miner cohorts; the deposition fractions were also from the Yeh and Schum (1980) lung geometry in the MPPD model. Particle clearance in each of these lung dosimetry models is described as the mass transfer of particles from a given lung region over time.

The parameter values used in the lung dosimetry models are from previously published sources. The mass median aerodynamic diameter (MMAD), geometric standard deviation (GSD), and density of particles were reported in the rat studies (Lee et al., 1985; Mauderly et al., 1987; Muhle et al., 1991; Heinrich et al., 1995; Nikula et al., 1995). (MMAD was used since the aerodynamic particle size, not the primary particle size, determines the particle deposition probability.) Reference worker breathing parameters were used (e.g., 9.6 m³ air inhaled per 8-h day), as reported by ICRP (1994). A working lifetime exposure duration of 45 yr was assumed (8 h/day, 5 days/wk, 50 wk/yr). The clearance rate parameters used in the MPPD model were the default values in that model, and the clearance rate parameters used in the interstitial/sequestration model (described in Kuempel et al., 2001a) were the arithmetic mean estimates from the Tran and Buchanan (2000) coal dust model because those estimates were based on the most complete exposure data.

Extrapolation of Rat BMDL to Humans: Airborne Exposure

When the rat mean airborne mass concentration is used as the dose metric in QRA steps 2 and 3, then the extrapolation of the BMDL to humans is performed using allometric relationships as follows:

$$\begin{aligned} \text{BMDL human} &= [\text{BMDL (rat) (mg/m}^3\text{)} \times \text{air inhaled (rat) (m}^3\text{/d)} \\ &\quad \times \text{hours exposed (rat) (6/24)} \\ &\quad \times \text{days exposed (rat:human)(260/240)} \\ &\quad \times \text{(allometric factor)}] / \\ &\quad \text{air inhaled in 8-h workday (human) (m}^3\text{/day)} \end{aligned}$$

Where: the “allometric factor” is based on either the total lung mass or lung alveolar epithelial surface area (SA):

$$\begin{aligned} \text{Lung mass} &= \text{lung mass (human)/lung mass (rat)} \\ \text{Lung SA} &= \text{lung SA (human)/lung SA (rat)} \end{aligned}$$

The BMDL values were derived from the rat studies using the BMD software (U.S. EPA, 2003). The remaining values used in these equations include: air inhaled (rat), 0.36 m³/day (U.S. EPA, 1987); air inhaled in 8-h workday (human), 9.6 m³ (ICRP, 1994); lung mass (human), 1000 g (ICRP, 1994); lung mass

TABLE 1
Benchmark doses associated with 0.1% excess risk of lung cancer in rats

Substance ^a	BMD (BMDL) as average airborne mass concentration over 2 yr (mg/m ³) ^b	BMD (BMDL) as retained lung burden after 2 yr (mg/g lung) ^b
TiO ₂ (ultrafine)	—	0.22 (0.18)
CB (ultrafine)	0.051 (0.039)	0.21 (0.19)
DEP (ultrafine)	0.066 (0.058)	0.43 (0.37)
TiO ₂ (fine)	2.2 (1.8)	2.1 (1.7)

Note. Dose-response data from Lee et al. (1985), Mauderly et al. (1987), Muhle et al. (1991), Heinrich et al. (1995), and Nikula et al. (1995). BMD: Maximum likelihood estimate of benchmark dose; BMDL: 95% lower confidence limit of benchmark dose.

^aDEP: diesel exhaust particulate; CB: carbon black.

^bBased on multistage model (third degree polynomial; coefficients restricted to be positive).

(rat), 2 g (average control lung weight across studies); alveolar epithelial surface area (human), 143 m²; and alveolar epithelial surface area (rat), 0.48 m² (Parent, 1992).

Comparison of Observed and Predicted Rat Lung Burdens

An additional analysis was performed to compare the observed and predicted lung burdens in rats. Three different rat lung dosimetry models (CIIT and RIVM, 2002; Tran et al., 2000; and Tran et al., 2001, 2002) were used to predict the rat retained lung mass dose of fine or ultrafine TiO₂, ultrafine CB, or ultrafine DEP. These predicted rat lung burdens were not used in the human risk estimates, but were obtained to provide a preliminary evaluation of how well the current models may predict nanoparticle lung doses. The fractional deposition predicted by the MPPD model (CIIT & RIVM, 2002) was used in the other two models, which describe particle clearance and retention in the lungs. An inhalability adjustment* was used in each MPPD model.

RESULTS

Table 1 provides the benchmark dose estimates (BMD and BMDL) from the rat dose-response models using airborne exposure or retained lung burden data for ultrafine or fine TiO₂, ultrafine CB, or ultrafine DEP (QRA step 2). These values were used to estimate the working-lifetime exposure concentrations in Table 2. The BMD and BMDL for ultrafine TiO₂ as airborne concentrations could not be estimated because the ultrafine TiO₂ data alone are insufficient (a single exposure group) (Heinrich et al., 1995)—hence the missing values in Tables 1 and 2—and

*The inhalability adjustment multiplies the inhaled concentration by a factor that adjusts for attenuation of the probability that particles are inhaled as their size increases (CIIT & RIVM, 2002).

TABLE 2
Airborne particle concentrations (8-h TWA) over a 45-year working lifetime associated with 0.1% excess risk of lung cancer in humans

Substance ^b	Human-equivalent exposure concentration (mg/m ³)			
	Allometric adjustment—BMDL as air mass concentration ^a		Lung dosimetry model—BMDL as mass lung dose ^a	
	Lung mass extrapolation factor	Lung surface extrapolation factor	MPPD/ICRP models (CIIT & RIVM, 2002)	Interstitial/sequestration model ^c
TiO ₂ (ultrafine)	—	—	0.14	0.073
CB (ultrafine)	0.19	0.11	0.24	0.12
DEP (ultrafine)	0.28	0.17	0.28	0.18
TiO ₂ (fine)	8.7	5.1	1.3	0.68

^aBMDL: Lower 95% confidence limit of benchmark dose (from multistage model, deg. 3). See Methods for details.

^bDEP: diesel exhaust particulate; CB: carbon black.

^cKuempel et al. (2001a); Tran and Buchanan (2000).

the divergent exposure-response relationships for fine and ultrafine TiO₂ (Figure 1b) precluded combining the data to fit a single multistage model for both.

Table 2 shows the human-equivalent exposure concentration (mg/m³) (over a 45-yr working lifetime) associated with a 0.1% excess risk of lung cancer (QRA step 4). Among the ultrafine particles, the estimates are within a factor of 2, whether from allometric extrapolation or lung dosimetry modeling. For fine TiO₂, the human-equivalent exposure estimates are several times higher than those for any of the ultrafines, using either the allometric extrapolation or the lung dosimetry modeling approach. The differences between fine and ultrafine estimates are greater using the allometric approach.

Table 3 shows results for the evaluation of the observed vs. predicted retained mass lung burdens for the three rat lung dosimetry models. These results show that each of these models predicts reasonably well (within a factor of approximately 2) the fine or ultrafine particle mass doses retained in the lungs following chronic inhalation at the highest exposure in each study. The

retained mass burden of fine TiO₂ was underpredicted by each model, while the lung burden of ultrafine particles tended to be overpredicted.

DISCUSSION

In this article, we have illustrated a quantitative risk assessment approach using dose-response data from rats exposed by chronic inhalation to poorly soluble fine or ultrafine particles. We compared both allometric adjustment and lung dosimetry modeling approaches to extrapolate animal airborne exposures or lung doses to humans. We also compared the observed versus predicted lung burdens of fine or ultrafine particles following chronic inhalation exposure in rats.

Human-Equivalent Exposure Estimates

Both of the human lung dosimetry models predict lower human-equivalent exposure concentrations for fine TiO₂ compared to the allometric approaches (Table 2), suggesting that the

TABLE 3
Ratio of predicted vs. observed mass particle burden in rat lungs

Substance ^a	Two-cpt model: MPPD with inhal adj (CIIT & RIVM, 2002)	Nine-cpt model for low toxicity dust (Tran et al., 2000)	Four-cpt model for high toxicity dust (Tran et al., 2001, 2002)
TiO ₂ (ultrafine)	1.5	0.92	1.3
CB (ultrafine)	2.0	1.6	2.0
DEP (ultrafine)	1.4	1.1	1.4
TiO ₂ (fine)	0.46	0.36	0.43

Note. Abbreviations: cpt, compartment; inhal adj, inhalation adjustment.

^aHighest dose in each study: TiO₂ (ultrafine), CB (carbon black), DEP (diesel exhaust particulate) (Heinrich et al., 1995) (female rats); TiO₂ (fine) (Lee et al., 1985) (average lung burdens of male and female rats).

human lung dosimetry models are more sensitive to the factors influencing the relationship between respirable particle exposure and internal lung dose in humans. The similar human-equivalent exposure estimates for the ultrafine particles from the allometric extrapolation and the lung dosimetry models suggest that external exposure is a good estimate of the internal dose—which would be expected if there were high lung retention of inhaled ultrafines. These estimates may also reflect some of the complexity of the processes influencing particle size-specific deposition and retention in the lungs, and the capability of the current lung models to predict these processes for ultrafines. For example, if the deposited dose of ultrafine particles is greater than predicted by the MMAD and GSD (e.g., if the airborne fraction includes unmeasured primary particles), or if the clearance of ultrafines is less than that of an equal mass of fine particles, then the dosimetry models would overestimate the working-lifetime exposure concentration associated with a given critical lung dose. That is, a higher exposure concentration would be predicted to be associated with a given excess risk based on lung dose.

Comparison of Observed Versus Predicted Rat Lung Burdens

The three rat lung dosimetry models evaluated provide reasonable estimates of the retained mass burden of the fine or ultrafine poorly-soluble particles in the lungs following chronic inhalation (Table 3). The data that had been used in developing these rat lung dosimetry models were from subchronic inhalation exposure to fine-sized respirable particles including TiO₂ (CIIT & RIVM 2002; Tran et al., 2000) and SiO₂ (Tran et al., 2001, 2002). The predicted lung burdens following chronic inhalation exposure were similar to observed values, although fine TiO₂ was underpredicted by each model. A possible reason is that the rat strain used in the fine TiO₂ study (Lee et al., 1985), the Sprague-Dawley rat, is larger in size than the rat strains used to calibrate these models (Wistar, Fischer 344, and Long Evans) (Tran et al., 2000; Tran et al., 2001, 2002; and CIIT & RIVM, 2002, respectively). For the ultrafine particles evaluated, the models tended to overpredict the lung burdens following chronic exposure. This is consistent with greater clearance and/or translocation of ultrafine particles from the lungs. However, the uncertainty and variability in factors such as particle size distributions and model assumptions could also account for these differences in predicted and observed lung burdens. Further evaluation of these models with additional data is needed, including exposure to nanoparticles of varying characteristics (e.g., size, shape, chemical composition, degree of agglomeration).

Experimental Evidence of Modeling Results

Experimental studies have shown that ultrafine particles are cleared less effectively and may be retained in the lungs to a greater extent than larger respirable particles (Ferin et al., 1992; Oberdörster et al., 1994), and may also translocate to other

organs (Geiser et al., 2005). The reduced lung clearance may be due to less effective phagocytosis by alveolar macrophages (Renwick et al., 2001, 2004). Chemical composition can also influence particle clearance and translocation. In rats, relatively rapid translocation of carbon nanoparticles from the lungs to other organs was observed (Oberdörster et al., 2002), compared to iridium nanoparticles (Kreyling et al., 2002) (1 to 7 days post-exposure). However, long-term (up to 6 mo) lung retention of iridium nanoparticles was similar to that for other poorly soluble, micrometer-sized particles (Semmler et al., 2004). In humans, quartz particles were estimated to translocate to the lung-associated lymph nodes at a faster rate than coal particles (Tran & Buchanan, 2000). These studies illustrate that the clearance and translocation of inhaled respirable particle may depend on several factors, including particle size, chemical composition, and surface properties.

Thus, it is not certain to what extent the current mass-based lung dosimetry models may predict the clearance and translocation of respirable particles of various sizes and compositions. These models describe the mass transfer of *all* particles that deposit in a given region of the respiratory tract. To a certain extent, the deposition models intrinsically account for particle size-specific clearance by predicting where particles (given certain characteristics and conditions) will deposit within the respiratory tract. Once deposited, the particles encounter region-specific clearance mechanisms, such as mucociliary clearance in airways or alveolar macrophage-mediated clearance in the gas-exchange region. Particles in the alveolar region that are not cleared may translocate to the lung interstitium, where they may enter the lymph or blood circulation. Thus, for a given mass of deposited particles, the extent to which the clearance and translocation processes depend on particle properties will influence how well the current lung dosimetry models predict the disposition of nanoparticles.

In addition, there is a need to better describe the fate of inhaled nanoparticles beyond the lungs including by pathways that may have been previously unrecognized. For example, nanoparticles depositing in the nasal region have been shown to translocate via the olfactory bulb into the brain in rats (Oberdörster et al., 2004). Better understanding of the biology associated with the disposition of inhaled nanoparticle is expected to lead to better dose estimation in any QRA.

Lung Dosimetry Model Structure

Differences in structure and parameter values of the various lung dosimetry models can result in large variation in predicted doses, and therefore in predicted risks (Kuempel & Tran, 2002). The human lung models used here were relatively similar, with the MPPD model predicting approximately twice the exposure concentrations as that predicted from the interstitial/sequestration model (Table 2). The differences are likely due to the clearance and retention features of the models, since the deposition estimates were similar. The MPPD human model uses the ICRP (1994) clearance model, which consists of three

first-order compartments describing three clearance phases of particles that deposit in the gas-exchange region of the lungs. In contrast, the interstitial/sequestration model (Kuempel et al., 2001a) is a higher-order model in which particles depositing in the gas exchange region of the lungs may be cleared to the tracheobronchial region or may be translocated to an interstitial/sequestration compartment, where only very slow clearance to the lymph nodes may occur. That model structure was found to be necessary to adequately predict the retained lung burdens in two independent cohorts of coal miners (Kuempel, 2000; Kuempel et al., 2001a, 2001b; Tran & Buchanan, 2000).

The rat models also differ in structure. The two-compartment MPPD rat clearance model has one alveolar compartment and one lymph node compartment, with a dose-dependent clearance parameter based on the particle volume dose (to describe overloading of lung clearance). In contrast, the nine-compartment model (Tran et al., 2000) includes alveolar, interstitial, and lymph node compartments, as well as subcompartments for free and sequestered particles, and mobile and decayed macrophages. Dose-dependent clearance is based on the particle surface area dose in that model. The four-compartment model (Tran et al., 2001, 2002) includes alveolar, interstitial, and lymph node compartments, with an additional compartment to distinguish the free and phagocytized particles in the alveolar region.

It should be noted that these lung dosimetry models use average parameter values. Interindividual variability in the rat and human lung model parameter values has been explored to some extent (Kuempel et al., 2001b; Tran et al., 2000), but more work is needed to better characterize the distribution of parameter values in a population.

Future Studies and Research Needs

The examples of QRA in this article use data from chronic inhalation studies. Because of the high cost of these studies and the long duration before results are obtained, it would be beneficial if reliable, shorter term *in vivo* or *in vitro* toxicity tests could be developed. To validate the shorter term *in vivo* or *in vitro* tests, these assays would need to be shown to be predictive of the chronic studies, for example, by detecting an early response that has been causally linked on the pathway to a chronic disease endpoint. For many inhaled particles, including the poorly-soluble, low-toxicity particles, pulmonary inflammation has been shown to be an early response that is also involved in the mechanism of lung tumor development in rats (ILSI, 2000). Using pulmonary inflammation as the response of interest, Faux et al. (2003) showed equivalent dose-response *in vivo* and *in vitro* when the dose metric was surface area dose of particles per surface area of epithelial cells (m^2/m^2) in the centriacinar region of the lungs or in the petri dish, respectively. *In vivo*, the response was the elevation in polymorphonuclear leukocytes (PMNs) in bronchoalveolar lavage (BAL) fluid from rats, while *in vitro* the response was elevation of the inflammatory cytokine interleukin-8 (IL-8) mRNA in a human alveolar

epithelial cell line (A-549). These studies are currently being extended to evaluate other particles and to determine whether the doses associated with increased production of inflammatory cytokines *in vitro* are predictive of the initiation of inflammation *in vivo*. If so, data from these assays could be useful in risk assessment by providing alternative estimates of the critical dose to the lung epithelial cell surface and the relative potency of particles including nanoparticles.

Research is needed in several areas to address gaps identified in lung dosimetry modeling and to better predict the risk of occupational exposure to nanoparticles. The specific aims of ongoing research to develop lung dosimetry and risk assessment approaches for nanoparticles include: (1) estimate disease risk as a function of particle composition and size, using existing data and models; (2) revise and extend the current rat lung dosimetry models to include particle size- and composition-specific particle clearance from the lungs and translocation beyond the lungs; (3) quantitatively evaluate the model structures and the population variability in parameter values, including deposition and clearance; (4) validate the revised and extended rat lung dosimetry models using data from ongoing studies; (5) extrapolate the validated rat model to extend the human lung dosimetry model to include key processes for nanoparticle clearance and translocation; and (6) estimate the internal dose and disease risk associated with occupational exposures to nanoparticles.

CONCLUSIONS

Established QRA methods, such as those described in this article, can be used to estimate the risk of occupational exposures to fine and ultrafine particles, and to provide a scientific basis for evaluating the possible risk of exposure to engineered nanoparticles. The method used to extrapolate the rat data to humans influences the estimate of dose, and therefore risk. Validation of lung dosimetry models is needed for nanoparticles of varying physicochemical properties, and extension of these models is needed to describe the translocation of nanoparticles beyond the lungs as observed in rodent studies.

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