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A New Method for Determining Allowable Daily Intakes[1]

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A New Method for Determining Allowable Daily Intakes. CRUMP, K. S. (1984). *Fundam. Appl. Toxicol.* 4, 854–871. The usual method for establishing allowable daily intake (ADI) for a chemical involves determining a no-observed-effect level (NOEL) and applying a safety factor. Even though this method has been used for many years, there appear to be no general guidelines or rules for defining a NOEL. The determination of a NOEL is particularly uncertain for lesions which occur naturally in untreated animals. NOELs also have shortcomings in that smaller experiments tend to give larger values (this should be reversed because larger experiments can provide greater evidence of safety) and that the steepness of the dose response in the dose range where effects occur plays little or no role in the determination of a NOEL. This paper proposes and illustrates the use of a "benchmark dose" (BD) as an alternative to a NOEL. A BD is a statistical lower confidence limit to a dose producing some predetermined increase in response rate such as 0.01 or 0.1. The BD is calculated using a mathematical dose–response model. This approach makes appropriate use of sample size and the shape of the dose–response curve. The BD normally will not depend strongly upon the mathematical model used because the method does not involve extrapolation far below the experimental range. Thus the method sidesteps much of the model dependency often associated with extrapolation of carcinogenicity data to low doses. The method can be applied to either "quantal" data in which only the presence or absence of an effect is recorded, or "continuous" data in which the severity of the effect is also noted.

I. INTRODUCTION

A common approach to quantifying permissible human exposure to a toxic agent is to establish a no-effect level using experimental animal data and then to apply a safety factor—or uncertainty factor, as it is sometimes called—to arrive at a permissible exposure level for humans. Allowable daily intakes for chemicals (ADIs), such as were employed by EPA in calculating water quality criteria (EPA, 1980), furnish one example of such calculations. Threshold limit values (TLVs), which are provided by the American Conference of Government and Industrial Hygienists (ACGIH) for many chemicals to which workers are exposed, are calculated in a similar fashion (ACGIH, 1976). The term "daily intake (DIL) has also been employed. In this we will refer to ADIs as a matter of nience, although the discussion will a all such estimates. The calculation of a by applying a safety factor to a no-effec will be referred to as a NOEL–SF app

In recent years, ADIs for carcinogen sometimes been calculated by fitting m matical models to experimental dose–re data. These models are used to estima dose corresponding to some specified amount of additional risk. EPA (1980) this approach to set water quality crite carcinogens, and used a NOEL–SF m for noncarcinogens. This dichotomy wa upon the supposition that carcinogens a likely to have a threshold; consequen NOEL–SF approach would be inappro because it assumes the existence of a thre

The object of this paper is to present

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matical and statistical approaches to ting ADIs for effects other than cancer. next section some potential shortcom- the NOEL–SF approach are discussed. llowing section describes some math- al models and related statistical meth- hese methods have two features that are hat novel. First, some of the models e the possibility of thresholds below no effect will occur. Second, methods gested for application to "severity" or nuous" data rather than just on inci- data. In the next section, a recommen- is made for replacing the NOEL in the -SF approach with a "benchmark This benchmark represents a statistical confidence limit on the dose corre- g to a small increase in effect over the ound level. The amount of increase in sed to define the benchmark is small so that the estimate of the benchmark ll reflect the shape of the dose–response nd it is large enough so that the lower nce limit will not depend critically e mathematical model used in its cal- . A number of examples are presented ing the calculation of benchmark doses nparing them with NOELS.

FICULTIES WITH THE NOEL–AFETY FACTOR METHOD

on of a NOEL

rst problem one faces with the NOEL is one of definition: Just what con- a NOEL? For effects which are un- us because they do not occur in un- nimals, such as acute toxicity or the nce of rare tumors, determination of can be reasonably straight-forward; ct is seen in any animal a NOEL is ed—otherwise a NOEL is not deter- For less well-defined effects, such as or cloudy swelling of the liver, de- ion of a NOEL requires the use of t. This problem is compounded when considering effects which have nonzero background levels. Consider, for example, liver weight; all animals have nonzero background levels of this "effect." Liver weights constitute a continuous measure (as opposed to "incidence" or "quantal" data) which can be obtained for each animal. It might happen that the average liver weight in some, or even all, of the treated groups is above that of the control group. Since this could happen by chance, usually the NOEL is taken to be the largest dose for which the increase in liver weight is not statistically significant. However, such a decision can seem rather arbitrary when there is a smooth dose–response trend which overlaps the region where the increase is not statistically significant.

A NOEL must be one of the experimental doses.[2] This constraint can appear unnecessarily restrictive in some cases. Consider, for example, an experiment to detect liver effects which involves three dose levels. Suppose at the highest dose level there are very severe effects, at the middle there are barely discernible effects, and at the low dose no effects at all are seen. Then the low dose likely will be designated the no-effect level even if the dose–response from the middle to high dose indicates that a much higher dose (one slightly less than the middle dose) would have had no discernible effect. Furthermore, if the data at the lowest dose had not been available, this experiment could not be used at all to define a no-effect level.

Effect of Sample Size

It would be appropriate for larger studies to tend to produce larger ADIs because they

[2] A NOEL is not an inherent property of the animal system but depends upon the experimental design and outcome. Thus it represents, in statistical terms, a statistic or an estimate of a "true no-effect level." This latter term refers to the highest dose which is absolutely safe and thus is an inherent property of the animal system, or, in statistical terms, a parameter. For an effect for which no threshold exists, the "true no-effect level" is zero.

involve less random variation. However, the NOEL approach has the opposite tendency. A larger study has a better chance of showing a statistically significant result and thus will, on average, produce a smaller ADI. As an illustration, suppose at the control and one treated dose in a study involving 100 rats per dose, the resulting mean liver fat per animal was 15.1 g in the control group and 18.4 g in the treatment group with a standard deviation of 10.0 g in each group. Then the t statistic for a difference between the two groups is 3.3, which is significant at the 1% level. However, if the identical results came from a study involving only 25 rats per group, the t statistic is 1.14, which is not significant at the 10% level. Thus, a NOEL would possibly be estimated for the smaller study but not the larger.

Therefore, rather than encouraging larger studies to demonstrate greater evidence of safety, the NOEL–SF instead penalized proponents of chemicals for conducting large studies. This topsy-turvy state of affairs has made it necessary for regulatory agencies to set minimally acceptable sample sizes. Quite naturally, many studies use these minimal values.

Utilization of Dose Response

A NOEL is determined solely by information relating to whether or not an effect was observed; the magnitude of positive effects and relationships among the effects at the various doses (i.e., the dose–response trend) is largely ignored. Consider for example the hypothetical data in Fig. 1. Experiment A shows a sharply increasing dose response. Experiment B shows a much flatter dose response, which is, in fact, consistent with a linear response through the origin. It appears, because of the sharp decrease in response with decreasing dose in Experiment A, as opposed to Experiment B, that the NOEL for A should be larger than the NOEL for B. However, because the response at dose d_{2A} was barely significant, the NOEL for Experiment A is d_{1A}

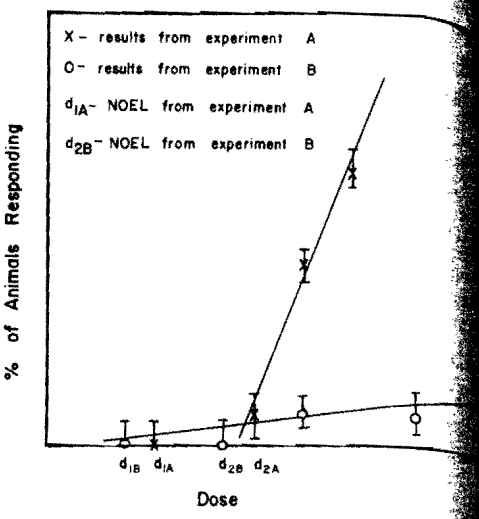
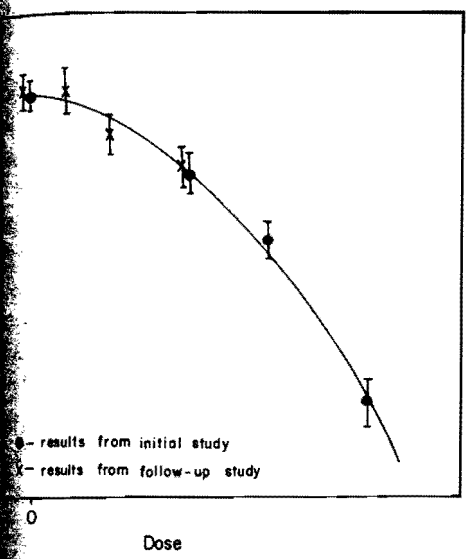


FIG. 1. Hypothetical responses with 95% con limits for two experiments.

which is less than the NOEL d_{2B} for Ex ment B. The dose–response methods discussed in later sections are capable of utilization of dose–response trends.

The NOEL–SF Approach Can Entail U essary Restrictions and Expense

Consider the following scenario: A com wishing to market a new product imple a thorough toxicological testing progra required by the regulatory agency inv Included in this program is a 2-year ch toxicity and carcinogenesis bioassay, a two-generation reproduction and terat study. Each study involves three treatmen one control group with doses and sample approved by the agency. The reproductio teratology study is negative. In fact, the t animals reproduce better than the contr imals. This is apparently related to th that the control animals are obese. The 2 study likewise shows no effects of treat except for a dose-related weight redu which is apparently due to the fact tha animals were fed the chemical in such concentrations that their food was distas As illustrated in Fig. 2, this weight loss fo



2. Hypothetical average weights with 95% confi-mits from two studies (see text for description).

dose–response trend and the effect is cally significant at the lowest dose tested. ency rules that a no-effect level has not etermined and thus there is no basis culating a ADI. The company is then d to conduct another 2-year study. This also uses three treatment levels, the of which coincides with the lowest used previous study. A dose–response trend obtained in the follow-up study as il-d in Fig. 2. The data at the highest produce almost exactly the results in ier study at that dose level. The weight he middle dose in the follow-up study nally significant and the average weight ow dose is comparable to that of the group. The agency rules that the low the follow-up study is a NOEL; an then calculated by applying a safety and the company is finally allowed to its product.

re 2 shows that the follow-up study was ssary and only verified the dose–re-trend of the initial study. If dose–re-methods had been used for determining he extra expense of the follow-up study 2-year delay could have been avoided.

Arbitrariness of Safety Factor

The NAS Safe Drinking Water Committee made the following recommendations for uncertainty factors (safety factors):

> 1. Valid experimental results from studies on prolonged ingestion by man with no indication of carcinogenicity.
>
> Uncertainty Factor = 0.
>
> 2. Experimental results of studies of human ingestion not available or scanty (e.g., acute exposure only). Valid results of long-term feeding studies on experimental animals or in the absence of human studies, valid animal studies on one or more species. No indication of carcinogenicity.
>
> Uncertainty Factor = 100.
>
> 3. No long-term or acute human data. Scanty results on experimental animals. No indication of carcinogenicity.
>
> Uncertainty Factor = 1000.
>
> These uncertainty factors are used in every case as a divisor of the highest reported long-term dose which is observed not to produce any adverse effect. (NAS, 1977)

The application of a 100-fold safety factor to results from long-term animal studies is a long-standing practice. It has been interpreted as resulting from the product of two 10-fold safety factors: one factor to account for animal-to-animal variation, and another to translate results from animal to man (Weil, 1972). However, the use of the 100-fold safety factor probably developed simply because some operational basis for setting allowable exposures was needed and a factor of 100 seemed "reasonable" or "prudent." The fact that humans have 10 fingers undoubtedly played a role in the specific factor selected.

When a safety factor is applied it is implicitly assumed that a threshold exists and the resulting ADI is below the threshold and hence safe. However, whether a threshold exists for a specific effect and, if so, whether the ADI is below that threshold are open to question. Also, as economic costs of regulations become more critical, there is increasing need for balancing the level of safety provided with

the costs involved. The NOEL–SF approach does not lend itself to cost–benefit analyses.

III. FITTING DOSE RESPONSE MODELS TO TOXICOLOGICAL DATA

Quantitative toxicological data are basically of two types: quantal and continuous. Quantal or incidence data specify the number of animals affected, but not the degree of harm. The numbers of animals with tumors or some genetic anomaly are examples of quantal data. On the other hand, with continuous data the level of harm is specified for each animal. Organ weights, triglyceride levels in liver, and serum measurements are examples of effects that are usually recorded as continuous data.

Dose Response Methods for Quantal Data

Quantal data from a toxicological experiment can be represented as a collection of triplets (N_i, X_i, d_i)—one triplet for each treatment or control group—where N_i is the number of animals in the ith group, X_i is the number of affected animals, and d_i is the dose. Let $P(d)$ represent a dose–response model applicable to quantal data, where $P(d)$ is the probability that an effect will occur in an animal subject to a dose d. The parameters of the model can be estimated by fitting the model to quantal dose–response data using maximum likelihood procedures. A number of dose–response models have been suggested for use with cancer data. Some of these, such as the one-hit, multistage, multihit, and Weibull models can be derived from detailed assumptions about carcinogenic mechanisms. Other models, such as the probit or logit models, can be thought of as representing the distribution of individual tolerances in a large population (Krewski and Van Ryzin, 1981).

In the next section some ways are suggested for applying mathematical models to noncancer data. The toxic endpoints to which these methods could be applied are ca
diverse mechanisms, most of which are p
understood. Therefore, it seems that it
not be fruitful to attempt to develop
response models from detailed assum
regarding these mechanisms. Instead, we
pose the use of relatively simple generic
els. For illustrative purposes we shall co
the following models: the quantal
regression (QLR) model

$$P(d) = c + (1 - c)\{1 - \exp[-q_1(d - d_0)$$

for d

$$= c \quad \text{for } d < d_0$$

where $0 \leq c \leq 1$, $d_0 \geq 0$, $q_1 \geq 0$; the qu
polynomial regression (QPR) model

$$P(d) = c + (1 - c)\{1 - \exp[-q_1(d - d_0)$$

$$- \cdots - q_k(d - d_0)^k]\} \quad \text{for } d$$

$$= c \quad \text{for } d < d_0$$

where $0 \leq c \leq 1$, $d_0 \geq 0$, $q_i \geq 0$ for
$\ldots, k$; the quantal Weibull (QW) mo

$$P(d) = c + (1 - c)[1 - \exp(-ad^k)]$$

where $0 \leq c \leq 1$, $a \geq 0$, and $k \geq 1$; a
log-normal (LN) model

$$P(d) = c + (1 - c)N(a + b \log d)$$

where $0 \leq c \leq 1$, $b \geq 1$ and N is the sta
normal distribution function.

Readers familiar with the carcino
dose–response literature will recognize
(2) as slightly modified versions of,
tively, the one-hit and multistage mo
applied to carcinogenesis data (Krews
Van Ryzin, 1981). Each has been m
here to include a threshold dose d_0; do
low this threshold produce no effect.
these models allow for the possibility
thresholds could exist for some effects;
ever, the models could be applied wi
threshold fixed to 0. Although we hav
done so, thresholds could also be inclu
the Weibull and log-normal models.

te that the restrictions $k \geq 1$ and $b \geq 1$ sumed for the Weibull and log-normal ls, respectively. Some restrictions of this seem necessary with these models; oth- these models can exhibit very extreme logically implausible behavior. The re- on $k \geq 1$ was selected for the Weibull because $k < 1$ corresponds to a su- ar curve shape which is implausible for logical effect (Crump, 1984). Although triction $b \geq 1$ for the slope parameter log-normal model does not have a strong tical basis, it does have a precedent, as recommended by Mantel *et al.* (1975) ncer data.

Response Methods for Continuous Data

tinuous dose–response data consist of e level and the response level for each With most continuous effects there variation about a nonzero value in the group. There has been little experience lying dose–response models to such is possible to convert continuous data tal data by considering all animals with es beyond a particular value as "af- and all others as "unaffected." How- us procedure entails a considerable loss rmation as well as requiring the arbi- oice of a cut-off value. The following makes more complete use of the data. method will be based upon the sup- that the responses in an animal group to a dose d_i are normally distributed an $m(d_i)$ and variance σ_i^2. There is heoretical reason and a pragmatic rea- assuming the normal distribution. the Central Limit Theorem of prob- heory (Loeve, 1963) the sample means pproximately normally distributed for mples regardless of the form of the ng distribution. Second, with the nor- ribution, maximum likelihood meth- be applied knowing only the doses d_g, the numbers of animals at each $\ldots, n_g$, and the corresponding sample means and standard errors $(\bar{x}_1, s_1)$, $(\bar{x}_2, s_2)$, $\ldots$, $(\bar{x}_g, s_g)$. If a non-normal distribution were assumed, maximum likelihood methods would require knowledge of the individual animal responses, which usually are not readily available. The choice of the normal distribution is not a critical decision as this distribution only determines the error structure, and not the dose response. The mean function $m(d)$, which represents the average response at a dose d, determines the dose response. We do not require any assumptions regarding the variances (other than that they are finite); it is not necessary to assume, for example, that the variances in the different dose groups are all equal.

For illustrative purposes, we will consider the following forms for $m(d)$: the continuous linear regression (LCR) model

$$m(d) = c + q_1(d - d_0) \quad \text{for } d \geq d_0$$
$$= c \quad \text{for } d < d_0 \qquad (5)$$

where $d_0 \geq 0$, but c and q_1 are unrestricted; the continuous polynomial regression (CPR) model

$$m(d) = c + q_1(d - d_0) + \cdots + q_k(d - d_0)^k \quad \text{for } d \geq d_0$$
$$= c \quad \text{for } d < d_0 \qquad (6)$$

where $d_0 \geq 0$ and the q_i's are restricted to be either all positive (increasing dose response) or all negative (decreasing dose response); and the continuous power (CP) model

$$m(d) = c + q_1(d - d_0)^k. \qquad (7)$$

These models are analogous to (1), (2), and (3), which were suggested for use with quantal data.

With both quantal and continuous data, in addition to the selection of a dose–response model, the proper use of statistical confidence limits is also of critical importance. Often, different confidence limit procedures yield different results; this makes it important to use the same procedures when comparing dose–

response models. A standard method for computing confidence limits is to base them upon the asymptotic normal distribution of maximum likelihood estimates. However, these confidence limits have been shown to behave poorly in a low dose extrapolation setting (Crump and Masterman, 1979; Krewski and Van Ryzin, 1981; Crump and Howe, 1983); the upper and lower limits are often too close together to be believable. Further, these limits are not invariant under parameter transformations, and different transformations applied to the same model at low doses can yield vastly different confidence bounds. Cox and Lindley (1974) noted these difficulties in a more general context, and argued that confidence limits based upon the asymptotic distribution of the likelihood ratio statistic "can be expected to behave much more sensibly" than those based upon the asymptotic normality of maximum likelihood estimates. Crump and Howe (1983) reviewed confidence limit procedures for use in dose–response evaluations and recommended limits based upon the distribution of the likelihood ratio statistic as the method of choice. This method for constructing confidence limits is outlined in the Appendix and will be used exclusively throughout this paper.

IV. THE BENCHMARK–SAFETY FACTOR METHOD FOR COMPUTING ADIs

In this section we examine the implications of modifying the NOEL–SF method for calculating ADIs by replacing the NOEL by a "benchmark dose" (BD) calculated using the methods described in the last section. A BD is defined as a lower statistical confidence limit for the dose corresponding to a specified increase in level of health effect over the background level. The increased level of effect upon which the BD is based would be near the lower limit of the experimental range; i.e., near the lower limit of increases in health effects which can be measured with reasonable accuracy in toxicological studies. This value is estimated to be something on the order of a 10% c from background at typical sample Benchmark doses calculated in this f will have several advantages over NO They will reflect the dose–response patt a much greater degree than NOELs. The also make more reasonable use of sampl (larger experiments will tend to produce BDs, which is not true of NOELs). It wi be necessary to define a NOEL in or determine an ADI. Because these BDs respond to risks in the experimental r their value will not depend strongly upo particular dose–response model used in calculation.

For quantal data we define the BD to dose d which corresponds to a specified for the extra risk

$$[P(d) - P(0)]/[1 - P(0)].$$

Extra risk can be interpreted as the probab of an effect at dose d given that no effect have occurred in the absence of the dose interpretation is valid irrespective of wh there is independent action between the ground and the stimulus. Extra risk greater weight upon the same increase for a common lesion than for a rare l For example, it takes an increase of 9% the background level for a lesion with a background rate to attain a 10% extra compared to only a 5% increase for a with a 50% background rate. Because property, some may prefer using the addi risk $P(d) - P(0)$ to extra risk.

For continuous data we define the B be a dose d which corresponds to a spe amount of absolute change in the mean relative to the mean value in the absen the dose—i.e., the dose d correspondin specific value for the "extra response"

$$\frac{m(d) - m(0)}{m(0)}.$$

Other terms, such as the standard error responses in control group, could be us the denominator in place of $m(0)$ to norm this expression.

TABLE 1

QUANTAL DATA USED TO ILLUSTRATE QUANTITATIVE DOSE–RESPONSE METHODOLOGY

ethiourea (ETU) (Khera, 1973)

Fetal anomalies in rats

s (mg/kg)	0	5	10	20	40	80
ffected/total No.	0/167	0/132	1/138	14/81	142/178	24/24

Tetrachlorodibenzo-p-dioxin (TCDD) (Khera and Ruddick, 1973)

Intestinal anomalies in rat fetuses

(μg/kg)	0	0.125	0.25	0.5	1.0
ected/total No.	0/24	0/38	1/33	3/31	3/10

Tetrachlorodibenzo-p-dioxin (TCDD) (Murray et al., 1979)

Rats dead at birth

(μg/kg/day)	0	0.001	0.01
cted/total No.	22/318	16/224	17/100

robenzene (HCB) (Khera, 1974)

14th rib anomaly in rat fetuses

(mg/kg)	0	10	20	40	60
cted/total No.	0/80	4/79	8/91	15/87	25/96

toxin—Type A (Food Research Inst., Univ. of Wisconsin) (FSC, 1978)

Death due to Botulinum

g)	.01	.015	.020	.024	.027	.030
d/total No.	0/30	0/30	0/30	0/30	0/30	4/30
g)	.034	.037	.040	.045	.050	
d/total No.	11/30	10/30	16/30	26/30	26/30	

es of Benchmark Doses Calculated Quantal Data

strate application of the benchmark to quantal data, we have applied it s of quantal dose–response data, in- posures to ethylenethiourea (ETU); trachlorodibenzo-*p*-dioxin (TCDD) sets); hexachlorobenzene (HCB); inum toxin—Type A (BT–A). The isted in Table 1. A summary of the four models to these data is given Graphs of the data, along with the QPR model (which was the only t fit all five data sets adequately) are Figs. 3–7. Doses corresponding to vels of extra risk are furnished in 7.

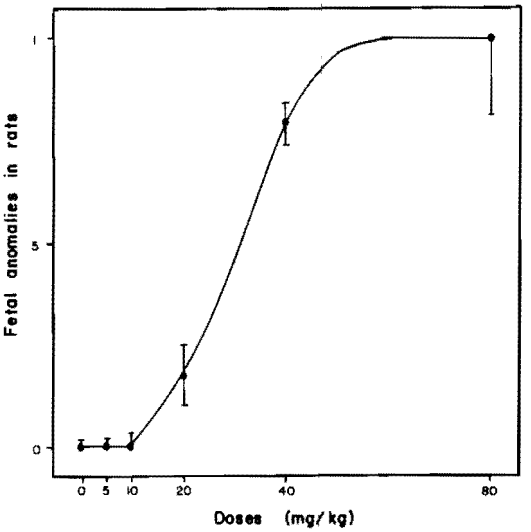


FIG. 3. Probability of fetal anomaly in rats (Khera *et al.*, 1977) from exposure to ETU with 90% confidence bars and best-fitting polynomial regression model.

TABLE 2

SUMMARY OF FITS OF MODELS TO QUANTAL DATA IN TABLE 1

Data	Model[a]	χ^2	*df*	*p* value
ETU	QLR	17	3	0.0007
	QPR	0.0	2	1.0
	QW	1.3	2	0.73
	LN	0.46	2	0.93
TCDD (Khera and Ruddick, 1973)	QLR	0.17	2	0.92
	QPR	0.014	1	0.91
	QW	0.32	3	0.85
	LN	0.23	3	0.89
TCDD (Murray *et al.*, 1979)	QLR	0	0	
	QPR	0	0	
	QW	0	0	
	LN	0	0	
HCB	QLR	0.11	2	0.95
	QPR	0.09	1	0.76
	QW	0.11	2	0.95
	LN	0.31	2	0.86
Botulinum toxin	QLR	7.0	8	0.54
	QPR	4.4	7	0.73
	QW	162	8	0.00001
	LN	159	8	0.00001

[a] Code: QLR = quantal linear regression, QPR = quantal polynomial regression, QW = quantal Weibull, LN = log-normal.

The ETU data involve a sizable number of animals and are characterized by a NOEL at 5 mg/kg followed by a steeply rising dose response that reaches 100% response at 80 ppm. Each of the models except the QLR model fits these data quite adequately. Also, except for the QLR model, MLEs and lower confidence limits for doses corresponding to various levels of increased risk computed using the various models agree rather closely for extra risks of 0.1, 0.05, and 0.01. However, doses corresponding to extra risks of 10^{-6} differ by larger amounts.

The Khera and Ruddick (1973) TCDD data on intestinal anomalies involve a dose-related increase in response for doses larger than a NOEL of 0.125 μg/kg. However, the confidence intervals on responses at the experimental doses are wider than those for the ETU

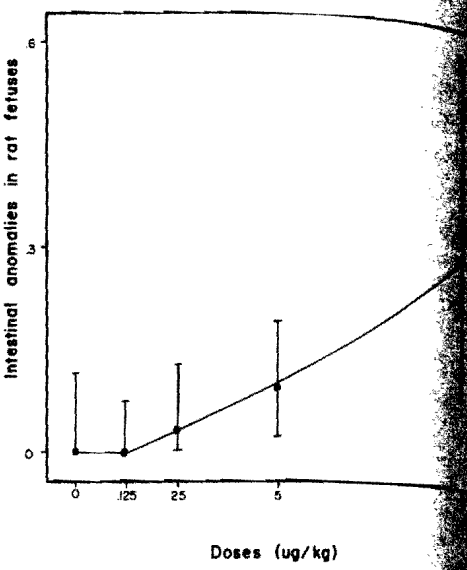


FIG. 4. Probability of intestinal anomaly in r... from exposure to TCDD (Khera *et al.*, 1973), w... confidence bars and best-fitting polynomial r... model.

data (Figs. 3 and 4). As a result, all ... the models fit these data quite well (T... Predictions of the four models also ... closely down to extra risks of 0.01 bu... considerably at extra risks of 10^{-6}.

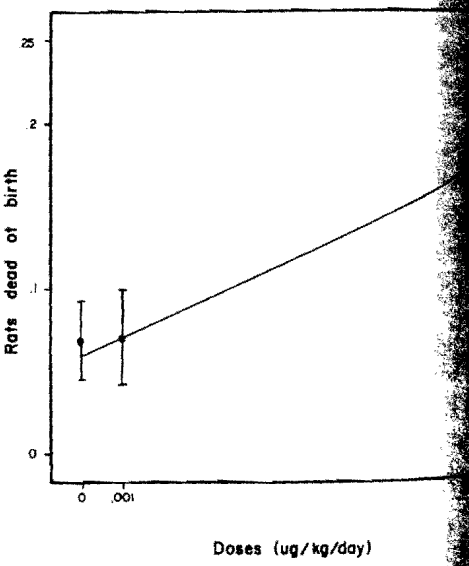
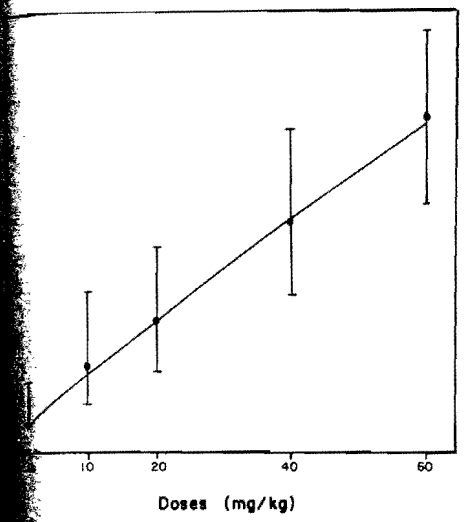


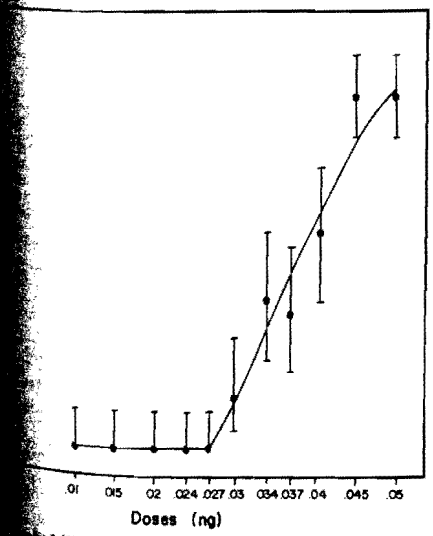
FIG. 5. Probability of fetal death in rats from ... to TCDD (Murray *et al.*, 1979), with 90% confide... and best-fitting polynomial regression model.



robability of 14th rib anomaly from exposure Khera, 1974), with 90% confidence bars and polynomial regression model.

he Murray *et al.* (1979) TCDD data atal survival and the Khera (1974) a on rib anomalies are nearly linear escribed well by all four of the mod- calculated from the models corre- to extra risks of 0.1, 0.05, and 0.01 n close agreement.

ta on probability of death after ex- Botulinum Toxin (FSC, 1978) ex-



ability of death from exposure to botulinum A (Food Research Inst., Univ. of Wisconsin,), with 90% confidence bars and best-fitting gression model.

TABLE 3
DOSES CORRESPONDING TO GIVEN LEVELS OF EXTRA RISK FOR QUANTAL ETU DATA[a]

		Dose (mg/kg)		
Model	Extra risk	MLE	95% lower	99% lower
QLR	0.1	12.2	11.8	11.6
QPR		16.4	13.9	13.2
QW		17.9	15.7	14.7
LN		17.1	15.3	14.6
QLR	0.05	11.0	10.6	10.4
QPR		13.2	11.6	11.0
QW		14.5	12.2	11.3
LN		14.8	13.0	12.2
QLR	0.01	10.1	9.7	9.5
QPR		10.2	7.2	6.0
QW		8.9	7.0	6.2
LN		11.2	9.5	8.8
QLR	1×10^{-6}	9.9	9.5	9.2
QPR		9.4	4.0-1	1.5-3[b]
QW		5.9-1	2.9-1	2.1-1
LN		4.2	3.1	4.7

[a] *Source.* Khera, 1977.
[b] 1.5-3 means 1.5×10^{-3}.

TABLE 4
DOSES CORRESPONDING TO GIVEN LEVELS OF EXTRA RISK FOR QUANTAL TCDD DATA[a]

		Dose (mg/kg)		
Model	Extra risk	MLE	95% lower	99% lower
QLR	0.1	4.6-1[b]	3.2-1	2.8-1
QPR		5.0-1	3.2-1	2.8-1
QW		5.2-1	3.6-1	3.0-1
LN		4.9-1	3.5-1	2.9-1
QLR	0.05	3.1-1	2.1-1	1.5-1
QPR		3.3-1	2.2-1	1.5-1
QW		3.6-1	2.0-1	1.5-1
LN		3.5-1	2.1-1	1.4-1
QLR	0.01	2.0-1	5.7-2	3.0-2
QPR		1.7-1	4.9-2	3.0-2
QW		1.6-1	4.3-2	2.9-2
LN		1.8-1	6.4-2	3.0-2
QLR	1×10^{-6}	1.7-1	7.8-3	3.0-6
QPR		1.3-1	4.9-6	3.0-6
QW		1.6-3	4.3-6	2.9-6
LN		2.0-2	5.4-4	1.1-4

[a] *Source.* Khera and Ruddick, 1973.
[b] 4.6-1 means 4.6×10^{-1}.

TABLE 5

DOSES CORRESPONDING TO GIVEN LEVELS OF EXTRA RISK FOR QUANTAL MURRAY POSTNATAL SURVIVAL DATA[a]

		Dose (μg/kg/day)		
Model	Extra risk	MLE	95% lower	99% lower
QLR	0.1	9.3-3[b]	5.3-3	4.4-3
QPR			Same as QLR	
QW		9.6-3	5.3-3	4.4-3
LN		9.3-3	4.6-3	3.5-3
QLR	0.05	4.9-3	2.6-3	2.1-3
QPR			Same as QLR	
QW		6.2-3	2.6-3	2.1-3
LN		5.5-3		1.5-3
QLR	0.01	1.6-3	6.1-4	4.2-4
QPR			Same as QLR	
QW		2.3-3	5.1-4	4.2-4
LN		2.0-3	4.1-4	3.2-4
QLR	1×10^{-6}	8.0-4	5.1-8	4.2-8
QPR			Same as QLR	
QW		9.6-6	5.0-8	4.2-8
LN		6.0-5	1.5-6	1.2-6

[a] *Source.* Murray *et al.*, 1979.
[b] 9.3-3 means 9.3×10^{-3}.

TABLE 6

DOSES CORRESPONDING TO GIVEN LEVELS OF [illegible] RISK FOR QUANTAL HCB DATA[a]

		Dose (mg/kg)	
Model	Extra risk	MLE	95% lower
QLR	0.1	21.7	17.4
QPR		22.3	17.4
QW		22.0	17.4
LN		21.0	14.2
QLR	0.05	10.6	8.5
QPR		11.0	8.5
QW		10.8	8.5
LN		11.1	6.1
QLR	0.01	2.1	1.7
QPR		2.2	1.7
QW		2.2	1.7
LN		3.4	1.3
QLR	1×10^{-6}	2.1-4[b]	1.7-4
QPR		2.2-4	1.7-4
QW		2.6-4	1.7-4
LN		5.0-2	4.8-3

[a] *Source.* Khera, 1974.
[b] 2.1-4 means 2.1×10^{-4}.

TABLE 7

DOSES CORRESPONDING TO GIVEN LEVELS OF [illegible] RISK FOR QUANTAL BOTULINUM TOXIN D[illegible]

		Dose (ng)	
Model	Extra risk	MLE	95% lower
QLR	0.1	3.0-2[b]	2.8-2
QPR		3.0-2	2.9-2
QW		3.3-2	2.7-2
LN		3.3-2	3.1-2
QLR	0.05	2.9-2	2.8-2
QPR		2.8-2	2.9-2
QW		3.0-2	2.2-2
LN		3.1-2	2.9-2
QLR	0.01	2.9-2	2.7-2
QPR		2.7-2	2.5-2
QW		2.4-2	1.6-2
LN		2.9-2	2.5-2
QLR	1×10^{-6}	2.9-2	2.7-2
QPR		2.7-2	2.3-2
QW		7.3-3	1.8-3
LN		2.0-2	1.6-2

[a] *Source.* FSC, 1978.
[b] 3.0-2 means 3.0×10^{-2}.

hibit a very rapid rise in response for doses larger than the NOEL of 27 ng. Neither of the nonthreshold models—QW or LN—fit these data. However, both the QLR and QPR models fit quite adequately (Table 2). All four of the models give comparable doses corresponding to given extra risk levels, even down to levels of extra risk of 10^{-6}.

Table 8 compares NOELs with BDs corresponding to three levels of extra risk. With the exception of the HCB data, the NOELs generally correspond to the BDs for extra risks between 0.01 and 0.05. However, these were all reasonably large studies and involved effects not seen in control animals; for smaller studies or for effects which can occur spontaneously, NOELs are liable to be larger relative to the BDs.

The data for HCB illustrate a particular advantage the benchmark approach has over the

TABLE 8

COMPARISON OF BENCHMARK DOSES WITH NOELS FOR QUANTAL DATA

Data set	Dose units	NOEL	Benchmark doses[a] corresponding to % extra risk		
			10%	5%	1%
(Khera, 1973)	mg/kg	5	13.9	11.6	7.2
(Khera and Ruddick,	μg/kg	0.125	0.32	0.22	0.049
(Murray *et al.*, 1979)	μg/kg/day	1.0-3	5.3-3	2.6-3	5.1-4
(Khera, 1974)	mg/kg	ND[b]	17.4	8.5	1.7
m toxin (Food Research te, Univ. of Wisconsin)	ng	0.027	0.029	0.027	0.025

hmark doses = 95% lower limits derived from QPR model.
= not determined.

approach. Since a NOEL was not de- d, the NOEL–SF method can not be ith these data to determine an ADI. r, these data would present no diffi- determining an ADI from a BD.

les of Benchmark Doses Calculated Continuous Data

9 contains dose–response data on in rats after exposure to carbon tetrachloride (Alumot *et al.*, 1976), mean body weights in rats after exposure to hexachlorobutadiene (HCBD) (Kociba *et al.*, 1977), and thymus weights in rats after exposure to TCDD (Murray *et al.*, 1979). Figures 8–10 contain graphs of the responses and 90% confidence intervals, along with the dose–response curve obtained by fitting the continuous polynomial regression (CPR) model to the data. In the Kociba *et al.* data numbers of animals were not provided and the total number on

TABLE 9

CONTINUOUS DATA USED TO ILLUSTRATE QUANTITATIVE DOSE–RESPONSE METHODOLOGY

on tetrachloride (Alumot et al., 1976)

	Average liver fat in male rats			
e (ppm in diet)	0	150	275	520
. ± SE (mg/g)	61.0 ± 6.6	71.0 ± 6.0	136 ± 21	229 ± 49
of animals	6	6	6	6

hlorobutadiene (HCBD) (Kociba et al., 1977)

	Mean body weight of male rats			
e (mg/kg/day)	0	0.2	2.0	20.0
. ± SE (gm)	586 ± 43	568 ± 53	557 ± 52	494 ± 15
of animals	90	40	40	40

8-Tetrachlorodibenzo-p-dioxin (TCDD) (Murray et al., 1979)

	Thymus weights of male offspring, f_3 generation		
e (μg/kg/day)	0	0.001	0.01
. ± SE (g)	0.19 ± 0.01	0.19 ± 0.06	0.08 ± 0.02
of animals	5	5	4

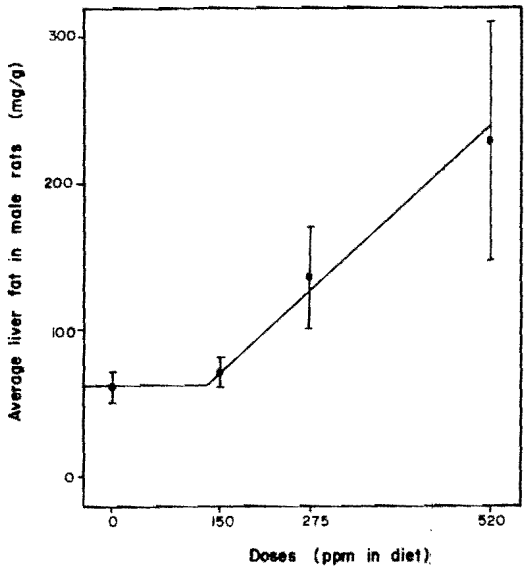


FIG. 8. Mean liver fat in rats exposed to carbon tetrachloride (Alumot *et al.*, 1976), with 90% confidence bars and best-fitting continuous linear regression model.

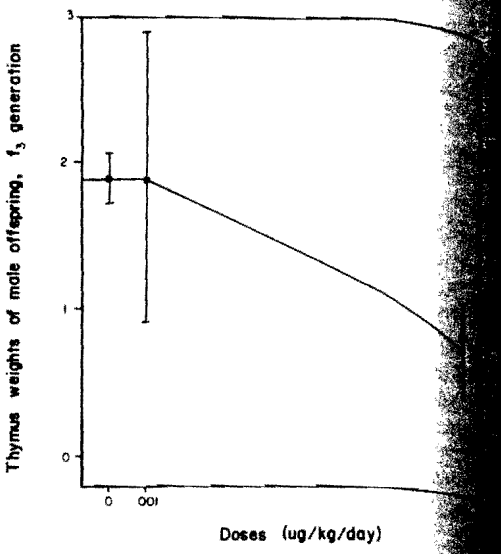


FIG. 10. Mean thymus weights of male offsp... generation (Murray *et al.*, 1979), with 90% co... bars and best-fitting continuous linear regression ...

test was assumed. Also, values reported by Kociba *et al.* as "s.d." were assumed to mean "s.e." As Table 10 shows, all of the models fit each of these data sets adequately.

Tables 11–13 show that the estimates of doses corresponding to given levels of extra response calculated using the four models are quite similar. In fact, corresponding ... confidence limits are almost identical in ... 12–14. In Table 11 the 95% lower limit ... by as much as a factor of 2 for an ex... sponse of 0.01 and by larger amou... smaller values of extra response.

Table 14 compares BDs with NOE... the continuous data. Question marks ... cluded beside the NOELs because it ... clear when a NOEL has been determin... example, although for the data for carb... rachloride the average liver fat in 15... animals is not statistically different fro... of control animals, there is an increase ... ppm that appears to be part of a dose–re... trend (Fig. 8). For these three data se... BD corresponding to an extra response ... are roughly comparable to the NOELs.

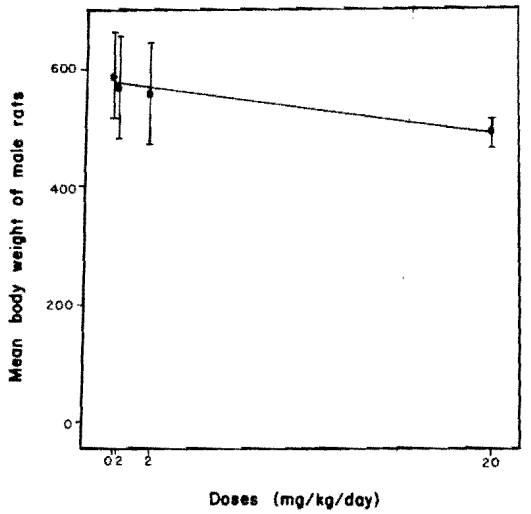


FIG. 9. Mean body weights in rats exposed to HCBD (Kociba *et al.*, 1977), with 90% confidence bars and best-fitting continuous linear regression model.

V. DISCUSSION

In this paper we have examined an... native to the NOEL–SF approach whi... volves fitting a mathematical model to ... cological dose–response data. The mo... used to define a BD, which represents ... tistical lower limit on the dose corresp...

TABLE 10

SUMMARY OF FITS TO MODELS TO CONTINUOUS DATA IN TABLE 9

Data	Model[a]	*F* statistic	*df*	*p* value
tetrachloride (Alumot 1976)	CLR	0.29	(1, 20)	NS[b]
	CPR	0.29	(1, 20)	NS
	CP	0.29	(1, 20)	NS
	CP (no threshold)	1.25	(2, 20)	NS
(Kociba *et al.*, 1977), body weights	CLR	0.14	(2, 206)	NS
	CPR	0.14	(2, 206)	NS
	CP	0.14	(2, 206)	NS
	CP (no threshold)	0.14	(2, 206)	NS
(Murray *et al.*, 1979)	CLR	0		NS
	CPR	0		NS
	CP (no threshold)	0		NS

[a] CLR = continuous linear regression, CPR = continuous polynomial regression, CP = continuous power.
[b] not significant (*p* value greater than 0.1).

cific increase in risk between 1 and is suggested that such a BD replace itional NOEL. We believe this approach mitigates several of the problems raised in Section II concerning the NOEL–SF method.

TABLE 11

ORRESPONDING TO GIVEN LEVELS OF EXTRA FOR CONTINUOUS CARBON TETRACHLORIDE

el	Extra response	Dose (ppm) MLE	95% lower
	0.1	141	102
		141	63
		141	67.2
shold)		95.6	47.1
	0.05	134	94.1
		134	37.6
		134	44.7
shold)		68.0	29.2
	0.01	129	87.9
		129	9.48
		129	17.3
hold)		30.8	9.53
	0.001	127	86.5
		127	1.03
		127	4.4
hold)		9.90	1.89

Alumot *et al.*, 1976.

TABLE 12

DOSES CORRESPONDING TO GIVEN LEVELS OF EXTRA RESPONSE FOR CONTINUOUS HCBD DATA ON MEAN BODY WEIGHTS[a]

Model	Extra response	Doses (mg/kg/day) MLE	95% lower
CLR	0.1	14.1	9.14
CPR		14.1	9.14
CP		14.1	9.14
CP (no threshold)		14.1	9.14
CLR	0.05	7.03	4.57
CPR		7.03	4.57
CP		7.03	4.57
CP (no threshold)		7.03	4.57
CLR	0.01	1.41	9.14-1[b]
CPR		1.40	9.14-1
CP		1.40	9.14-1
CP (no threshold)		1.41	9.14-1
CLR	0.001	1.40-1	9.14-2
CPR		1.41-1	9.14-2
CP		1.41-1	9.14-2
CP (no threshold)		1.41-1	9.14-2

[a] *Source.* Kociba *et al.*, 1977.
[b] 9.14-1 means $9.14 \times 10^{-1} = 0.914$.

TABLE 13

DOSES CORRESPONDING TO GIVEN LEVELS OF EXTRA RESPONSE FOR CONTINUOUS TCDD DATA[a]

Model	Extra response	Dose (μg/kg/day) MLE	95% lower
CLR	0.1	2.55-3[b]	1.32-3
CPR		2.55-3	1.32-3
CP (no threshold)		6.37-3	1.32-3
CLR	0.05	1.78-3	6.61-4
CPR		1.78-3	6.61-4
CP (no threshold)		5.53-3	6.61-4
CLR	0.01	1.16-3	1.32-4
CPR		1.16-3	1.32-4
CP (no threshold)		1.21-3	1.32-4
CLR	0.001	1.02-3	1.32-5
CPR		1.02-3	1.32-5
CP (no threshold)		1.96-3	1.32-5

[a] *Source.* Murray *et al.*, 1979.
[b] 2.55-3 means $2.55 \times 10^{-3} = .00255$.

A BD is calculated using a mathematical dose–response curve estimated from all of the dose–response data. Thus the benchmark should better reflect the shape of the dose response than the NOEL. Because a benchmark represents a statistical lower limit, larger experiments will tend on average to give larger benchmarks, thus rewarding good experimentation. As we pointed out, NOELs have the opposite tendency. With the NOEL approach, ADIs cannot be determined until a NOEL has been established. An otherwise well-conducted experiment may therefore be considered inappropriate for calculating an ADI if no NOEL is established. In such a case, determining an ADI could require an additional experiment resulting in considerable additional cost and delays. On the other hand, the original experiment might be quite acceptable for calculating a BD. This situation is illustrated by the quantal data for HCB (Fig. 6).

A BM–SF approach to setting ADIs would allow proponents of chemicals more latitude in the design of experiments than is possible under the NOEL–SF approach. With the BM method minimum sample sizes must be specified by the regulatory agency in order to ensure that NOELs are established to the agency's satisfaction. With a BM–SF approach the agency would still in some cases need to specify methods for choosing the maximum dose and the sample size to be used at this dose, because otherwise important effects might not be detected at all. Beyond this requirement, however, proponents of a chemical could be given wide latitude in selecting dose levels and sample sizes. Of course, the larger a study is, the better designed it is to estimate the BD, and the higher the benchmark is liable to be. The more accurate benchmark is considered critical, experimentors may wish to conduct a pilot study and consider carefully the placement of the experimental doses; otherwise, a study

TABLE 14

COMPARISON OF BENCHMARK DOSES WITH NOELS FOR CONTINUOUS DATA

Data set	Dose units	NOEL	Benchmark doses[a] corresponding to % extra risk 10%	5%	
Carbon tetrachloride (Alumot *et al.*, 1976)	ppm	150?[b]	141	134	12
HCBD (Kociba *et al.*, 1977) mean body weights	mg/kg/day	2.0?	14.1	7.0	
TCDD (Murray *et al.*, 1979)	μg/kg/day	0.001?	.0026	.0012	

[a] Benchmark doses = 95% lower limits derived from QPR model.
[b] ? indicates that it is doubtful whether a NOEL has been established.

may be considered adequate. Any prior nation on the shape of the dose–response could be used in optimally designing an ment. Such prior information might from pilot studies or studies of similar cals. Given such choices, proponents of als should be able to design studies n keeping with their needs and bud- constraints without compromising

n example of how experimental design erations could be put to effective use, e a company knows the smallest ADI ould permit the marketing of their t. It would be simple to calculate the ark that would produce this ADI. uld then design an experiment that e optimal under the assumption that ded benchmark is in fact the true ark. If the true benchmark were lower at they were hoping for, the statistical used in calculating the benchmark sure that human safety would not be mised. On the other hand, if the ark were near that for which the ex- t was designed, the extra care that the design might allow the marketing duct that could not have been mar- a less optimal design had been used. afety factors are largely arbitrary, one for choosing safety factors to use with rks would be to make the resulting mparable, on average, to those cal- previously using the NOEL–SF This could be accomplished by cal- benchmarks for a number of sub- or which ADIs have been developed NOEL–SF method, and then deter- he safety factor that, when applied to marks, would on average yield the I. Of course, ADIs calculated using L–SF and BM–SF methods could dif- ciably in specific cases.

gh we have not discussed the use of tical models for extrapolation of iogenesis data to low dose and thus g safety factors, this is another pos- ication of these methods. Our reluctance to recommend this application stems from the uncertainty as to the shape of the dose–response curves at low doses for toxic effects in general. Dose–response curves which are linear at low doses have been used to set upper bounds for low dose cancer risks (EPA, 1980). This approach has been justified on the grounds that cancer mechanisms that would produce linear dose responses at low doses appear quite plausible and those that would produce supralinear responses seem highly implausible. The low dose linearity concept could be used to determine upper limits of risks of noncarcinogenic effects as well. However, many of these effects appear threshold-like. The assumption of a linear response could greatly overestimate risk in cases where a threshold exists. The threshold models discussed in this paper might be used to determine risks at low doses for effects which appear to be threshold-like. However, we have not recommended this in this paper because of both the uncertainty as to the existence of a threshold and because these threshold estimates are apt to differ widely depending upon the specific model used.

The model-fitting techniques proposed here have fairly minimal data requirements. When quantal data are used, the basic needs are the doses, number of animals in each group, and the number of these animals which are affected. With continuous data one needs the doses, number of animals in each group, the average response in each group, and the standard errors of these responses. Some effects, such as cloudy swelling of the liver, are inherently difficult to quantify and are normally classified qualitatively, such as by present/absent or mild/severe. Even for effects which are quantifiable, the data needed to apply dose–response methods are frequently not reported in the literature. Thus, it will not be possible to apply these methods universally. However, the introduction of these methods would encourage more complete presentation of data, as well as generally encouraging the use of quantitative methods in toxicology.

It should be kept in mind that determining

ADIs does not involve purely statistical methods. Toxicological evaluation of data on numerous species and biological endpoints may be required. Included in the many considerations should be differences in species sensitivities to various chemicals and the need for affording different levels of protection for different toxicological effects. The statistical methods proposed in this paper should be useful in this process but they should not supplant a careful toxicological evaluation of all the data.

APPENDIX

Description of Maximum Likelihood Procedures

Likelihood for Quantal Data

Consider an experiment with g dose levels $d_1, \ldots, d_g$, and let N_i and X_i be, respectively, the number of animals tested and the number of animals affected at the ith dose level. Let $P(d)$ be the probability of a response at a dose d. Assuming that X_i has a binomial distribution with parameter N_i and $P(d_i)$, the likelihood of the data can be written as

$$L = \prod_{i=1}^{g} X_i^{N_i} P(d_i)^{X_i} [1 - P(d_i]^{N_i - X_i}.$$

Likelihood for Continuous Data

Consider an experiment with g dose levels $d_1, \ldots, d_g$; let N_i be the number of animals in the ith dose group, and let x_{ij}, $j = 1, \ldots, N_i$, $i = 1, \ldots, g$ represent the response of the jth animal in the ith dose group. It is assumed that x_{ij} has a normal distribution with mean $m(d_i)$ and variance σ_i^2. The parameters in the model consist of those involved in the definition of $m(d)$, plus $\sigma_1, \ldots, \sigma_g$. Let $\bar{x}_i$ be the sample mean in the ith dose group, i.e.,

$$\bar{x}_i = \sum_{j=1}^{N_i} x_{ij}/N_i.$$

and s_i^2 the sample variance, i.e.,

$$s_i^2 = \sum_{j=1}^{N_i} (x_{ij} - \bar{x}_i)^2/(N_i - 1).$$

Then the likelihood of the data can be [illegible] as

$$L = (2\pi)^{-g/2} \prod_{i=1}^{g} \sigma_i^{-1} \exp[-(N_i - 1)s^2$$

$$- N_i(\bar{x}_i - m(d_i)$$

Estimation and Confidence Intervals

The parameters are estimated as the [illegible] which maximize the appropriate like[illegible] The "likelihood method" (Cox and [illegible] 1974; Crump and Howe, 1983) is used [illegible] culate confidence limits. For example, [illegible] using quantal data the lower 95% limit [illegible] dose d corresponding to an extra risk [illegible]

$$\frac{P(d) - P(0)}{1 - P(0)} = 0.1$$

is calculated as the smallest d which [illegible]

$$\frac{P(d) - P(0)}{1 - P(0)} = 0.1$$

and

$$2 \log(L_{\max}/L) = (1.645)^2$$

where $L_{\max}$ is the maximum value of th[illegible] lihood L. When using continuous da[illegible] same approach is followed except the [illegible] for extra response replaces the one for [illegible] risk.

Computer Programs

These methods require iterative nu[illegible] calculations. We have developed co[illegible] programs to perform these calculatio[illegible] intend to have them available for the [illegible] public in the near future.

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necessarily reflect the views of the Agency, and
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