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Review

Nonlinearity and thresholds in dose–response relationships for carcinogenicity due to sampling variation, logarithmic dose scaling, or small differences in individual susceptibility

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Abstract

Nonlinear and threshold-like shapes of dose–response curves are often observed in tests for carcinogenicity. Here, we present three examples where an apparent threshold is spurious and can be misleading for low dose extrapolation and human cancer risk assessment. Case #1: For experiments that are not replicated, such as rodent bioassays for carcinogenicity, random variation can lead to misinterpretation of the result. This situation was simulated by 20 random binomial samplings of 50 animals per group, assuming a true linear dose response from 5% to 25% tumor incidence at arbitrary dose levels 0, 0.5, 1, 2, and 4. Linearity was suggested only by 8 of the 20 simulations. Four simulations did not reveal the carcinogenicity at all. Three exhibited thresholds, two showed a nonmonotonic behavior with a decrease at low dose, followed by a significant increase at high dose ("hormesis"). Case #2: Logarithmic representation of the dose axis transforms a straight line into a sublinear (up-bent) curve, which can be misinterpreted to indicate a threshold. This is most pronounced if the dose scale includes a wide low dose range. Linear regression of net tumor incidences and intersection with the dose axis results in an apparent threshold, even with an underlying true linear dose–incidence relationship. Case #3: Nonlinear shapes of dose-cancer incidence curves are rarely seen with epidemiological data in humans. The discrepancy to data in rodents may in part be explained by a wider span of individual susceptibilities for tumor induction in humans due to more diverse genetic background and modulation by co-carcinogenic lifestyle factors. Linear extrapolation of a human cancer risk could therefore be appropriate even if animal bioassays show nonlinearity.

Keywords: Dose–response relationship; Nonlinearity; Threshold; Hormesis; Bioassay for carcinogenicity; Cancer risk assessment; Pharmacodynamic modeling

Contents

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Introduction

Deviation from linearity in dose–response relationships for tumor induction in rodent bioassays for carcinogenicity is important in the context of extrapolation to low dose. Strongly sublinear (up-bent) curves and apparent thresholds may allow for a rejection of the linear-no threshold (LNT) default assumption and for a discussion of threshold doses and safety factors to derive tolerable exposure levels. This appears to be appropriate if mechanistic considerations can explain the threshold-like shape of the dose–response curve (Lutz, 1998). Here, we draw your attention to situations where an apparent deviation from linearity has no mechanistic support. The first two examples are trivial mathematically but are often overlooked. The third refers to extrapolations when epidemiological data in humans are available.

Case #1: Random variation in bioassays for carcinogenicity

Sampling variability can produce nonlinear shapes even if the true dose response is linear. This is particularly important for bioassays for carcinogenicity, because this test is usually not repeated. We simulated 20 bioassays by random sampling from a binomial distribution, with 50 animals per group and a true linear dose–response relationship with 5–7.5–10–15–25% tumor incidence at arbitrary units of dose 0–0.5–1–2–4. Fig. 1 shows the results. Among the 20 simulations, 4 did not reveal significant carcinogenicity of the chemical (nos. 1, 7, 10, 19), and only 8 showed a more or less linear dose response (nos. 3, 5, 9, 12, 14, 15, 16, 20). Three could be interpreted as threshold-like (nos. 4, 11, 13), 2 showed even a nonmonotonic dose response, with a tumor incidence below control at the lowest dose, followed by a significant increase at high dose (nos. 2, 4).

This simulation shows that the hypothesis of a threshold should be tested appropriately. A statistical procedure for this purpose has been suggested recently (Lutz et al., 2002). It is based on the simplest case with the analysis of three data points: (i) the control group, (ii) the highest dose that did not give rise to a significant increase in tumor incidence (NOEL), and (iii) the lowest dose that resulted in a statistically significant increase (LOEL). Based on a minimum chi-square procedure, the probability P was estimated that the observed tumor incidences could be a chance result of an underlying true linear dose response. If this null hypothesis was rejected on a 5% level, the lower limit of a 95% confidence interval for the threshold dose, i.e., the lower bound breakpoint of a hockey stick, was estimated. The procedure runs on the free statistics software package 'R' (http://www.r-project.org) and the source file is available at http://www.stat.math.ethz.ch/~lutz/publ/RTPlintest.php. When this test was applied to the 3 simulations that showed the most prominent threshold appearance in Fig. 1 (nos. 4, 11, 13), linearity could

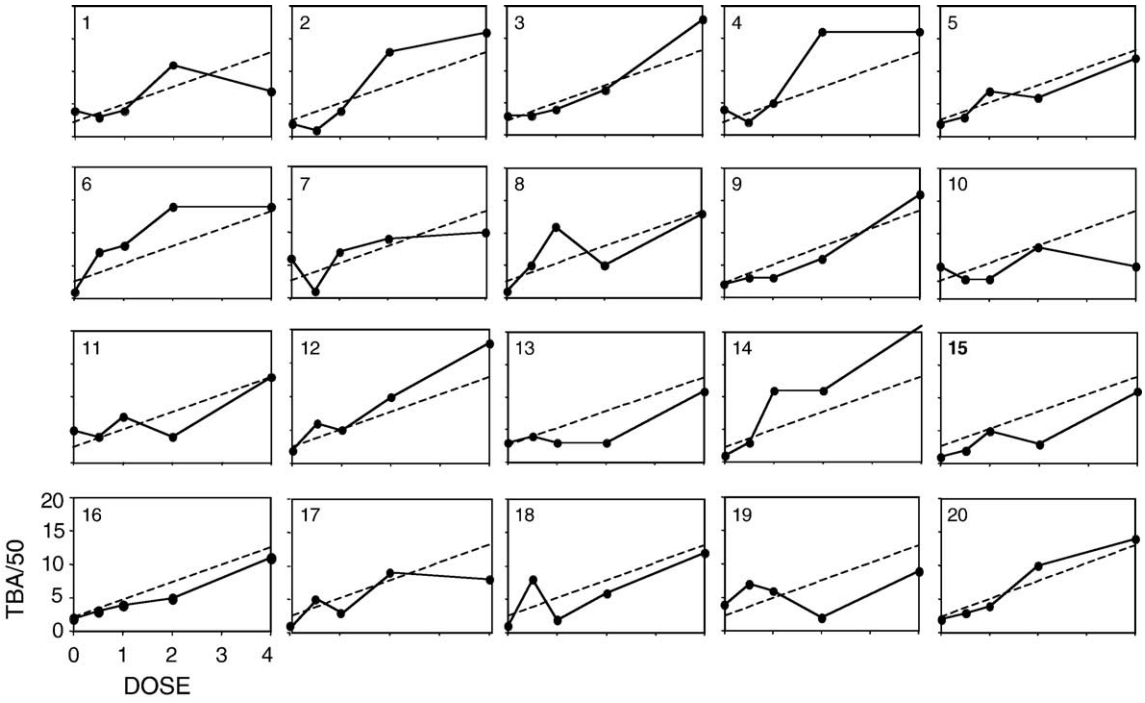

Fig. 1. Simulation of dose–response relationships for a standard rodent bioassay for carcinogenicity. Groups of 50 animals were sampled randomly from a binomial distribution, assuming a true linear dose response with a tumor incidence of 5% in the control group (dose 0) and tumor incidences of 7.5, 10, 15, and 25% at doses 0.5, 1, 2, and 4, respectively. TBA/50, number of tumor-bearing animals in groups of 50 animals. The dashed line indicates the true dose response.