Approaches to the Formulation of Standards for Carcinogenic Substances in the Environment

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After having agreed that standards are necessary for carcinogens that cannot be completely eliminated from the environment, two exchange groups in the U.S.S.R.-U.S. Cooperative Program present their different approaches to the problem. The Russian group has recommended a benzpyrene standard of 0.1 μg/100m³ of atmospheric air over populated regions and gives its experimental basis and theoretical rationale in the first part of this joint paper. Lifetime experiments in adult rats over a wide range of dose levels permit the determination of a largest ineffective dose level with respect to theoretical time of first tumor as well as incidence. The standard is set by extrapolation based on body weight and uses a safety factor of 10 to account for the additional susceptibility in embryogenesis and childhood. The U. S. group presents a mathematical model of time-to-tumor occurrence which permits the prediction of population incidence and life span shortening from time-to-tumor data in animals or man. It assumes the distribution of mortality-corrected time to tumor is lognormal with the nth power of time inversely proportional to dose and with dose independence of the variability of the logarithm of time to tumor. The prediction is made by combining this distribution, fitted to the data, with population mortality tables. Both groups emphasize that substantial research efforts are necessary to improve the scientific basis for setting standards.

Introduction

This paper represents a joint contribution under the auspices of the U.S.S.R.-U.S.A. Cooperative Program in Environmental Health Research. They summarize a series of discussions held at the Institute of General and Communal Hygiene in Kiev during the summer of 1973 concerning the general problem of formulating exposure standards for chemical carcinogens. In the U.S.S.R., there is official approval for an atmospheric exposure standard for benzpyrene in the occupational and general environment. The standard was developed in the U.S.S.R. after extended discussions and special research. It was recognized the carcinogen standards should be accepted as the legal basis for hygiene practices despite the fact that many aspects of the problem remain unsolved. In the U.S.A., the situation is complex. Exposure standards have evolved over the past 30 years in the atomic energy field and do have official sanction. Air quality criteria for

particulates developed by the Environmental Protection Agency represent an indirect standard for carcinogens, inasmuch as the particulates include the polycyclic aromatic hydrocarbons. The National Institute for Occupational Safety and Health has been developing criteria documents for specific substances which recommend exposure standards for carcinogens such as asbestos and arsenic. The Occupational Safety and Health Administration has developed standards for the safe handling of carcinogens without reference to permissible limits of exposure. The Delaney Clause of the Food, Drug and Cosmetic Act prohibits the addition of any carcinogens to food. Other regulations administered by the Food and Drug Administration do imply concentration limits for certain carcinogenic contaminants such as aflatoxin and the residues of carcinogenic feed additives in cattle and poultry. Thus, in the U.S.A., where there is considerable resistance to the idea of formalizing exposure standards for carcinogens, exposure standards are being used in various ways. However, there is no clearly defined or generally accepted methodology for developing such standards.

In view of the importance and diversity of approaches to this difficult problem, one aspect of the

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U.S.S.R.-U.S.A. Cooperative Program in Environmental Health Research is concerned with setting standards for carcinogens. The initial result of this cooperative program is this joint publication. The contribution from the U.S.S.R. is the work of scientists who developed the experimental data on benzpyrene carcinogenicity as well as the theoretical approach for applying this data to the formulation of benzpyrene and other exposure standards in air and water in the general and occupational environments. The contribution from the U.S.A. is a theoretical approach to the assessment of cancer risks, which uses animal and epidemiological data and characterizes risks both in terms of increased cancer incidence and life span shortening.

The participating groups from both countries agree that (1) standards are necessary for carcinogens that cannot be completely eliminated from the environment and (2) that a substantial research effort is required to further the development of the scientific basis for such standards.

Experimental Data for Assessment of Hazards from Small Doses of Carcinogens

At the present time, when no one has any doubts about the necessity of setting permissible standards for carcinogenic substances, acute importance attaches to the problems of a scientific basis and development of methodological approaches for establishing admissible limits for these compounds in the environment.

In view of the fact that up to now it is not clear whether a threshold exists for the action of carcinogenic substances, a genuine problem for hygiene is to determine the admissible doses of carcinogens, the specific action of which might show up only beyond the human life span. This can be attained by means of modeling, under the conditions of an oncological experiment, of the quantita-

tive dependence of the action of the carcinogenic agent, with subsequent extrapolation of the data to the human organism. The basic stages of such a process are the following: (1) the testing of various carcinogen doses, ranging from the maximally effective to the minimally effective and maximally noneffective doses, with animal observations throughout life span; (2) mathematical modeling of the relationships of carcinogen dose to tumorigenic effect and of carcinogen dose to time of tumor occurrence; (3) the quantitative prediction of the carcinogen doses whose theoretical times of first tumor formation extend beyond the limits of the life span in test animals; (4) extrapolation of the permissible carcinogen dose from animal to man and the calculation of the maximal permissible concentration in the environment.

The above will be illustrated using an example associated with the establishment of the maximal permissible concentration for benz(a)pyrene in the atmosphere which has been adopted in the U.S.S.R. Experimental study of the carcinogenic activity of various benz(a)pyrene doses by intratracheal administration served as the basis for this standard. In addition to the controls, six benz(a)pyrene treatment groups were tested with total doses ranging from 0.005 to 25 mg. The carcinogen was administered in 10 monthly doses by the method of Shabad (1). These tests made it possible to show the presence of a direct dose-effect relationship. Lung tumors were observed in 80% of the animals with the 25 mg carcinogen dose. Of the affected animals, 42.5% had malignant tumors. As the dose was reduced, the carcinogenic effect was reduced correspondingly, as can be seen from Table 1. The minimal effective benz(a)pyrene dose was 0.1 mg. This dose induced only benign tumors in 14.4% of the animals. Doses of 0.02 and 0.005 mg were inactive.

In addition to the dose-effect relationship, a pronounced dose-time relationship was observed. For example, in the case of the 25 mg benz(a)pyrene

Table 1. Occurrence of lung tumors in rats after ten intratracheal administrations of benz(a)pyrene at various total doses.

Benz(a)pyrene dose, mg	Animals with tumors, %[a]	Animals with malignant tumors, %[a]	Time to appearance of first tumor, months	Maximum life span, months
25.0	80.0	42.5	12	28
2.5	42.8	28.5	17	31
0.5	28.2	15.7	19	34
0.1	14.4	0	27	37
0.02	0	0	—	35
0.005	0	0	—	35
Controls	0	0	—	34

[a] Based on number of animals surviving 10 months.

dose, the first lung neoplasm was noted in the twelfth month after the initial exposure whereas in the case of the animals that received the minimally effective carcinogen dose (0.1 mg), the first tumor was observed only after 808 days, i.e., when most of the animals in the 25 mg dose group had died from neoplasms.

The third relationship noted by us was the dose dependence for the morphological structure of lung neoplasms, namely, the predominance of squamous carcinomas with large benz(a)pyrene doses and of adenocarcinomas with small doses. The importance of this observation lies in its agreement with the data on the different ratios of the histological types of human lung cancer in regions with different levels of pollution (2, 3).

The maximal noneffective benz(a)pyrene dose was determined on the basis of studies conducted under experimental conditions, whereas under natural conditions, man is exposed to a variety of carcinogenic substances. This fact required additional experiments involving the simultaneous introduction of benz(a)pyrene together with other substances into the lung (Table 2).

These combined exposure studies again showed that 0.1 mg is the minimal effective benz(a)pyrene dose, while 0.02 mg was the maximal noneffective dose. However, it is necessary to note the limitations of the experimental results, since the neoplasms were obtained on small groups of animals and the benz(a)pyrene was administered intermittently during only one-third of the total life span of the animals (tenfold, once a month), while the human organism is subjected to benz(a)pyrene ex-

posure during the entire life span.

The experimental results provided the basis for the mathematical modeling of the dose-response relationships in terms of the following logarithmic equation:

$$Y = 10 \ln [(X_n/X) + 1]$$

where Y is the percentage of animals with tumors, X_n is the carcinogen dose (mg), and X is the maximally noneffective dose (mg).

Based on the above model, the possible risk of tumor occurrence is predicted at the 6.9% level for a benz(a)pyrene dose of 0.02 mg as can be observed in Table 3.

Similar results were obtained in modeling the dose-effect relationship with the aid of the exponential function $Y = 100 (1 - e^{-ax})$. Under these conditions, it is assumed that the carcinogenic effect cannot be greater than 100% or less than 0. This model made it possible to estimate small benz(a)pyrene risks in terms of an upper limit obtained with the exponent ($a = 1.555$) determined by the 0.1 mg point and a lower limit with the exponent ($a = 0.064$) determined by the 25 mg point. As a result, the risk of tumor occurrence was determined for the 0.02 mg dose to have an upper limit of 3% and a lower limit of 0.023%.

Since the above considerations imply that benz(a)pyrene at doses which are noneffective in an experiment could induce neoplasms under certain conditions, we also used the dose-time relationship as a more realistic basis for extrapolation.

The calculation for the time t of first tumor occurrence with a small benz(a)pyrene dose d was done by using the function $t = (a/d) + b$, where $a = 1.021$ and $b = 16.79$. As shown in Table 4, the car-

Table 2. Occurrence of lung tumors in rats after intratracheal administration of benz(a)pyrene with various solvents and polycyclic aromatic hydrocarbons.

Mixture instilled	Benz(a)pyrene dose, mg	Tumor incidence, %
Benz(a)pyrene + india ink	0.1	14.4
	0.02	0
	Control	0
Benz(a)pyrene + dust	0.1	18.2
	0.02	0
	Control	0
Benz(a)pyrene + Tween 60	0.1	26.3
	0.02	0
	Control	0
Benz(a)pyrene + dibenz(a,h)-anthracene	0.1	14.2
	0.02	0
	Control	0
Benz(a)pyrene + benz(g,h,i)perylene + pyrene + anthracene + chrysene	0.1	0
	0.02	0
	Control	0

Table 3. Calculated risk for lung tumor occurrence in rats after the intratracheal introduction of various doses of benz(a)pyrene.

Benz(a)pyrene dose, mg	Animals with tumors, %
0.1	17.9
0.05	2.23
0.02	6.9
0.002	0.95
0.0005	0.24

Table 4. Calculated time t of the occurrence of the first lung tumor following ten intratracheal administrations of various total doses of benz(a)pyrene.

Benz(a)pyrene dose, mg	Time t, months
0.1	27.0
0.05	37.2
0.02	67.8
0.01	118.9
0.005	221.0
0.002	527.3

cinogenic effect from a 0.05 mg benz(a)pyrene dose will manifest itself towards the 38th month, which corresponds to the natural life span of the animals, while the effects from a 0.02 mg dose (68th month) and from a 0.01 mg dose (119th month) fall outside the limits of the life span.

Consequently, for calculation of the maximum permissible concentration of benz(a)pyrene in air, we can take the 0.02 mg benz(a)pyrene dose and use it in the formula for converting dose to concentration in air:

$$C_{benz(a)pyrene} /100 \text{ m}^3 = 100 \text{ } XP/BKV$$

where $C_{benz(a)pyrene}$ /100 m³ is the maximal permissible benz(a)pyrene concentration in 100 m³ of air, X is the maximal benz(a)pyrene dose for which the calculated time of the first tumor extends beyond the limits of maximal life span (0.02 mg). B is the average weight of the experimental animal (0.3 kg); P is the weight of the standard human (70 kg), K is a safety factor (10), and V is the volume of air inhaled by a human during his lifetime (383, 250 m³).

As can be seen from the formula presented, we make use of a safety factor because all of the calculations were based on results obtained from adult animals. The calculated concentration which is recommended as the benz(a)pyrene standard for the atmospheric air of populated areas will affect children who can be assumed to be much more sensitive to the action of carcinogenic substances than adults. In addition we must consider the possible effect of carcinogens during embryogenesis.

Thus, considering the safety factor, the relationship between human and experimental animal weights and the volume of air used by a human being during his lifetime, the average daily maximum benz(a)pyrene concentration should not exceed 0.1 μg/100 m³ of air.

In conclusion, it should be noted that the approach described above for deriving the maximal permissible concentration for benz(a)pyrene in air is the first attempt to set a carcinogen standard in the environment. There is no doubt that further investigations are necessary to establish more accurately the basis for extrapolation of animal data to man.

Use of the Temporal Aspects of Tumor Formation for the Estimation of Risks from Exposure to Environmental Carcinogens

The formulation of exposure limits to carcinogens in the environment is an important aspect of the control of hazards from these toxic agents. Some carcinogens can be eliminated from the environment without serious sacrifice. Other carcinogens are associated with essential technological activities where the complete elimination of environmental contamination is impossible; here, it is necessary to develop the methodology for estimating the levels of risk according to level of exposure in order to achieve a rational basis for selecting permissible exposure standards. We have advocated the position that in order to completely define the nature of the hazards from a carcinogen, it is necessary not only to define the increased risk of developing cancer at any given level of exposure, but also the age at which the extra cancers would occur and thus the amount of life-span which would be lost by the individuals who would not otherwise have developed cancer in the absence of such extra carcinogen exposure (4).

In order to define the age of occurrence of cancer, it is necessary to characterize carcinogenic response on a temporal basis. This requires a mathematical formulation which provides an accurate and systematic description of the time of cancer occurrence independent of the complications of extraneous mortality. Such a mathematical formulation can then be combined with population survival statistics to obtain the desired response in terms of cancer incidence and age of occurrence. Since human population survival has not been accurately described by a mathematical function, especially at the very advanced ages, the interaction of survival with the mathematical description of mortality-corrected tumor response is necessarily done by numerical computation.

A central issue is whether there is a mathematical formulation that satisfactorily describes the time pattern of cancer occurrence. A promising development in this regard is a formulation first proposed by Blum on the basis of the skin tumor response of mice to ultraviolet radiation (5) and later extended by Druckrey to a variety of chemical carcinogens and target organs on rodents (6). We have also shown that it is applicable to radiation cancer in terms of the osteogenic sarcoma response in mice to ²²⁶Ra and in humans to cigarette smoking (4). This formulation has two elements. The first is that at any given dose level of a carcinogen, administered for the lifetime of the exposed animals, the cumulative mortality-corrected tumor incidence will have a lognormal shape. In other words, the temporal pattern of response can be described completely by two parameters: the median or 50% time of tumor occurrence t and the geometric standard deviation σ_g. The second element of this formulation is that when a population of animals is subdivided into

different treatment groups and each treatment group is exposed to a different daily dose level d of a carcinogen, the tumor responses of each treatment group will differ only in the median time of tumor occurrence t, with the value of σ_g remaining unchanged. This means that the higher dose not only reduces the median time of tumor formation but also causes the same proportionate decrease in the time of tumor occurrence for all of the exposed animals. Furthermore, the relationship of t and d is expressed by the equation

$$dt^n = \text{constant}$$

where d is daily dose, t is time to 50% cancer incidence, and n is a constant exponent. When $n = 1$, the median appearance time is proportional to the reciprocal of the daily dose, e.g., doubling the dose rate halves the median tumor appearance time. When n has a high value, the median appearance time of tumors is affected relatively little by changes in dose rate.

Model calculations have been done to examine the interacting effects of σ_g and n on the various tumor response parameters, including tumor incidence (the proportion of responders) p, age at tumor occurrence τ, and life-span loss of the individuals developing cancer δ, and the life-span loss in the population as a whole $\triangle$. These calculations which assumed dt^n is constant and σ_g is constant and use U. S. vital statistics tables with dose normalized so that when dose is unity, median tumor time equals the average lifetime, have been presented in detail elsewhere and are useful to illustrate the general patterns of response (4).

The value of n determines the shape of the dose-response curve for incidence. The curves for incidence p and population life shortening $\triangle$ are S-shaped and asymmetrical. For a higher value of n, the early steplike increase, which is a quasi-threshold, occurs at a lower dose rate, and the later part if flatter at the higher dose rate. The higher the value of n, the greater the impact of low dose rates. This result reflects the greater insensitivity of median time t to changes in dose rate with higher n. These considerations suggest that a high value of n is unfavorable for a downward extrapolation of dose rate to achieve negligible effect levels.

At low incidence levels, where t is much greater than the maximum life-span, the incidence p and population life shortening, $\triangle$ are strongly dependent on the magnitude of the geometric standard deviation σ_g. The value of σ_g also influences the values of δ and τ; the larger the σ_g, the younger the average age of cancer occurrence τ and the greater the individual loss of life-span δ.

It is to be noted that, regardless of the value of n or σ_g, the dose-incidence curve is complex and does not fit any of the conventionally assumed forms, as for example, a linear or log-normal shape. This follows from the fact that the incidence curve includes two components: a simple systematic behavior of the mortality-independent temporal features of tumor response, and the complex and empirically determined survival pattern of the responding populations. The latter is determined by a large number of factors and the resultant incidence curve is necessarily complicated.

It also needs to be emphasized that at the present time, there are comparatively few experiments that have determined the temporal features of tumor formation. Hence, the analysis presented above represents no more than a suggested direction for further investigation of the general problem of determining the dose-dependence of tumor response. It is quite possible that no simple formulation will be applicable to all the various types of tumorigenic responses. Furthermore, the credibility of any formulation will depend on developing an understanding of the underlying mechanisms that determine the time pattern of tumor occurrence.

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