EXPEDITED CANCER POTENCY VALUES AND NO SIGNIFICANT RISK LEVELS (NSRLs) FOR SIX PROPOSITION 65 CARCINOGENS: CARBAZOLE, MeIQ, MeIQx, METHYL CARBAMATE, 4-N-NITROSOMETHYLAMINO)-1-(3-PYRIDYL)-1-BUTANONE, TRIMETHYL PHOSPHATE

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SUMMARY OF FINDINGS

Cancer potencies were estimated for six Proposition 65 carcinogens with dose-response data summarized by Gold and colleagues (Gold and Zeiger, 1997; Gold *et al.*, 1999) in the Carcinogenic Potency Database (CPDB) (http://potency.berkeley.edu/database.html), using an expedited methodology. The expedited approach represents the first level of a three-tiered risk assessment procedure currently in place for timely and efficient development of cancer potencies and Proposition 65 "no significant risk levels" (NSRLs) and has been shown previously to produce reliable potency values (OEHHA, 1992; Hoover *et al.*, 1995). Values generated using the expedited approach may be reevaluated if scientific considerations indicate that more detailed analysis associated with a conventional risk assessment is warranted. The chemicals assessed here are carbazole, MeIQ (2-amino-3,4-dimethylimidazol[4,5-*f*]quinoline), MeIQx (2-amino-3,8-dimethylimidazol[4,5-*f*] quinoxaline), methyl carbamate, 4-(N-nitrosomethylamino)-1-(3-pyridyl)-1-butanone (NNK), and trimethyl phosphate (TMP). This report describes the methodology used and the basis for cancer potency estimation for each of these compounds. The upper 95 percent confidence bound on the linear term of the multistage model fit to cancer dose response data is taken as the estimate of cancer potency. The derivation takes into account species differences and length of the bioassay. The Proposition 65 NSRL is defined in regulation as the daily level posing a 10^{-5} lifetime risk of cancer. Cancer potency estimates and the corresponding NSRLs are given in Table 1 below.

Table 1. Cancer Potencies and NSRLs

Chemical	CAS #	Cancer Potency $(mg/kg\text{-}day)^{-1}$	NSRL (µg/day)
Carbazole	86-74-8	0.17	4.1
MeIQ (2-Amino-3,4-dimethylimidazo[4,5-*f*] quinoline)	77094-11-2	1.5	0.46
MeIQx (2-Amino-3,8 -dimethylimidazo[4,5-*f*] quinoxaline)	77500-04-0	1.7	0.41
Methyl carbamate	598-55-0	0.0044	160
4-(N-nitrosomethylamino)-1-(3-pyridyl)-1-butanone	64091-91-4	49	0.014
Trimethyl phosphate	512-56-1	0.029	24

INTRODUCTION

This report describes the derivation of cancer potency values and "no significant risk levels" (NSRLs) for the six Proposition 65 carcinogens (California Health and Safety Code 25249.5 *et seq*.) listed in Table 1. An expedited procedure was applied in the derivation (OEHHA, 1992; Hoover *et al.*, 1995); the methodology is summarized below. The studies used as the basis of the potency derivation for each chemical, and the relevant data are described. The bases for selecting the cancer potency estimates are discussed. The final cancer potency estimates and NSRLs are presented for each chemical.

METHODOLOGY

In a typical, non-expedited assessment, a full literature search is undertaken to locate all data on the carcinogenicity and dose response characteristics of the compound. This is followed by a review of the pharmacokinetic and mechanistic (*e.g.*, genotoxicity) data, and a dose response review of all adequate bioassays. Occasionally the data support a pharmacokinetic analysis in the derivation of target dose estimates, or a dose response model different from the default. The expedited procedure differs from this usual practice in two ways. First, it relies on cancer dose response data evaluated and extracted from the original literature by Gold and colleagues (Gold and Zeiger, 1997; Gold *et al.*, 1999) and contained in the Carcinogenic Potency Database (CPDB) currently

electronically available at http://potency.berkeley.edu/database.html. Second, under the expedited procedure the choice of the multistage model is automatic and pharmacokinetic adjustments are not employed. The default procedures used to derive expedited cancer potency values are specified in the administrative regulations for Proposition 65 (Title 22 California Code of Regulations [CCR] 12703). The expedited approach, which represents the first level of a three-tiered risk assessment procedure currently in place for development of cancer potencies and Proposition 65 NSRLs, has been shown to be a reliable means for generating potency values and NSRLs in a timely and efficient manner. As described in Title 22 CCR 12703, an NSRL generated using the expedited approach may be reevaluated if scientific considerations indicate that more detailed analysis associated with a conventional risk assessment is warranted.

The methods for expediting potency estimation incorporate the following assumptions:

- The dose-response relationship for carcinogenic effects in the most sensitive species tested is representative of that in humans.
- Observed experimental results can be extrapolated across species by use of the interspecies factor based on "surface area scaling."
- The dose to the tissue giving rise to a tumor is assumed to be proportional to the administered dose.
- The multistage polynomial can be used to extrapolate potency outside the range of experimental observations to yield estimates of "low" dose potency.
- Cancer hazard increases with the third power of age.

Data Set Selection: The following criteria are used for data selection:

- Data sets with statistically significant increases in cancer incidence with dose ($p \leq 0.025$, two-tailed as reported in the CPDB) are used.
- When several studies are available, and one study stands out as being of higher quality due to numbers of dose groups, magnitude of the dose applied, duration of study, or other factors, the higher quality study is chosen as the basis for potency calculation.
- When there are multiple studies of similar quality in the sensitive species, the geometric mean of potencies derived from these studies is taken. If the same investigators tested both sexes of the same species/strain under the same laboratory conditions, and no other adequate studies are available for that species, the data set for the more sensitive sex is selected.
- Potency is derived from data sets that tabulate malignant tumors, combined malignant and benign tumors, or tumors that would have likely progressed to malignancy.

Mathematical Model: Cancer potency is defined as the slope of the dose response curve at low doses. Following the default approach, the Crump linearized multistage polynomial (Crump *et al.*, 1977) describes the dose response relationship:

$$\text{Probability of cancer} = 1 - \exp[-(q_0 + q_1 d + q_2 d^2 + ...)] \quad (1)$$

The slope (q_1) is estimated by fitting the polynomial to dose response data collected at high doses using one of the statistical curve-fitting packages developed for this purpose [*e.g.*, "Tox_Risk" (Crump *et al.*, 1993); "MSTAGE" (Crouch, 1992)]. For bioassays with exposures throughout the study period, dose (d) is the average daily dose over the experimental period. Cancer potency is estimated from the upper 95 percent confidence bound on the linear coefficient q_1, which will be termed q_1^*.

For a given chemical, the model is fit to one or more data sets. As discussed in the section above, the default is to select the data for the most sensitive species and sex. When there are several bioassays of equivalent quality, a geometric mean is taken. For multi-site carcinogens, a distribution of estimates corresponding to the 0.1 through 99.9 percentiles of the linear term (q_1) of the multistage model was generated for each treatment-related tumor site in the most sensitive species/sex with the MSTAGE 2.01 computer program (created by Edmund Crouch), which had been modified to tabulate percentile values. In general, the distribution of q_1 for a given site was discretized into 100 segments. The distribution of the sum of q_1s for each site affected by the chemical was obtained using a Monte Carlo simulation (250,000 trials) run in Crystal Ball (Crystal Ball 2000 software, Decisioneering, Inc., Denver, Colorado). The upper 95 percent confidence bound on the summed q_1s was taken as the basis of the cancer potency estimate for the combined tumor sites.

Standard bioassays on mice and rats last approximately two years. In standard risk assessments, this is the assumed lifespan for these species. Animals in experiments of shorter duration are at a lower risk of developing tumors than those in the standard bioassay; thus potency is underestimated unless an adjustment for experimental duration is made. In estimating potency, short duration of an experiment is taken into account by multiplying q_1^* by a correction factor equal to the cube of the ratio of the assumed standard lifespan of the animal to the duration of the experiment (T_e). This assumes that the cancer hazard would have increased with the third power of the age of the animals had they lived longer:

$$q_{animal} = q_1^* \bullet (104 \text{ weeks}/T_e)^3 \quad (2)$$

To estimate human cancer potency, q_{animal} values derived from bioassay data are multiplied by an interspecies scaling factor (K; the ratio of human body weight (bw_h) to test animal body weight (bw_a), taken to the 1/3 power; see Anderson *et al.* (1983) for details):

$$K = (bw_h/bw_a)^{1/3} \quad (3)$$

Thus,

$$\text{Cancer potency} = q_{human} = K \bullet q_{animal} \quad (4)$$

To calculate K, unless otherwise specified, default body weights of 0.5 and 0.35 kg were used for male and female rats, and 0.03 and 0.025 for male and female mice, respectively (Gold and Zeiger, 1997). Values for interspecies scaling and correction for study duration, as well as the recommended cancer potency value are reported in tabular form for each of the chemicals addressed in this report. From these human cancer potencies, exposures associated with a given level of cancer risk can be derived. For example, the NSRL for Proposition 65 is the intake associated with a lifetime cancer risk of 10^{-5} or lower. For a 70-kg adult, this level is calculated according to the following equation:

$$I = \frac{10^{-5} \times 70\ \text{kg} \times 1000 mg/\text{mg}}{q_{human}} \quad (5)$$

where q_{human} is given in units of $(\text{mg/kg-day})^{-1}$ and I in units of µg/day.

DERIVATION OF HUMAN CANCER POTENCY VALUES AND NSRLS

Cancer potency estimates and NSRLs were derived for six Proposition 65 carcinogens, as described below for each chemical.

CARBAZOLE (CAS No. 86-74-8)

Results from the studies by Tsuda *et al.* (1982) were listed in the CPDB. Tsuda *et al.* exposed male and female $B6C3F_1$ mice to carbazole via diet for 22 months. The animals were sacrificed at 24 months. Female mice were exposed to 180, 360, or 739 mg/kg-day. Males were exposed to 166, 332 or 665 mg/kg-day. The effective tumor incidence was reported by the authors and tabulated in the CPDB. Statistically significant increases in the incidence of hepatocellular tumors and forestomach tumors were observed in male and female mice. Cancer potency estimates based on these dose-response data are provided in Table 2.

As indicated in Table 2, the most sensitive sex/species/site is female mouse liver. The final potency estimate was derived based on data for both tumor sites, liver and forestomach, in female mouse. The corresponding dose-response data are shown in Table 3. The dose-response data for the liver tumors in female mice were highly supralinear (*i.e.*, the trend in tumor incidence is less than linear with increasing doses). Following the U.S. Environmental Protection Agency (U.S. EPA) procedures described in Anderson *et al.* (1983), whenever the multistage model does not fit the data adequately, data at the highest dose are deleted and the model fitted to the remaining data. This is repeated until an acceptable fit is obtained, as measured by the chi-square goodness-of-fit test. For the analysis of the female mouse liver data, the mid- and high-dose groups were dropped. The multistage model fit adequately to the forestomach

tumor data in female mice, and all dose groups were retained. The probability distributions for q_1 were generated for both tumor sites, using likelihood methods and the modified MSTAGE computer program (see Methodology). The distribution of the sum of the q_1s for each site was obtained, as described under Methodology, using a Monte Carlo simulation. The upper 95 percent confidence bound on q_1 was determined based on the distribution of the sum of potencies from the carbazole-affected sites. The human cancer potency is estimated to be 0.17 $(mg/kg\text{-}day)^{-1}$ and the associated NSRL 4.1 µg/day.

Table 2: Values Used in Calculating Human Cancer Potency Values for Carbazole based on Tsuda *et al.* (1982)

Sex/species/site	q_1* $(mg/kg\text{-}day)^{-1}$	Interspecies Scaling Factor (kg/kg)	Correction for Experiment Duration (wk/wk)	q_{human} $(mg/kg\text{-}day)^{-1}$	Goodness-of-Fit Test[1]
Female mouse hepatocellular carcinoma	0.009030	$(70/0.025)^{1/3}$	$(104/95.333)^3$	0.17	NA[2]
Female mouse forestomach (combined benign and malignant)	0.0004976	$(70/0.025)^{1/3}$	$(104/95.333)^3$	0.0091	p = 0.1146
Female mouse liver and forestomach, combined[3]	0.009357	$(70/0.025)^{1/3}$	$(104/95.333)^3$	0.17	--
Male mouse liver hepatocellular carcinoma	0.001823	$(70/0.03)^{1/3}$	$(104/95.333)^3$	0.031	p = 0.7625
Male mouse forestomach (combined benign and malignant)	0.0001209	$(70/0.03)^{1/3}$	$(104/95.333)^3$	0.0021	p = 0.9696

1 A p-value of greater than 0.05 for the chi-square goodness of fit test indicates an adequate fit.
2 Not applicable; see text.
3 Potency estimate is based on the combined distribution for q_1 for these sites.

Table 3: Incidence of Hepatocellular Carcinoma and Forestomach Tumors in Female B6C3F$_1$ Mice Treated with Carbazole Via Diet (Tsuda *et al.*, 1982)

Average Dose[1] (mg/kg-day)	Hepatocellular Carcinoma	Statistical Significance[2]	Forestomach Tumors	Statistical Significance[2]
0	2/45	$p < 0.0005$	0/45	$p < 0.009$
180	35/49	$p < 0.001$	5/49	$p < 0.05$
360	24/43[3]	$p < 0.001$	8/43	$p < 0.01$
749	30/46[3]	$p < 0.001$	6/46	$p < 0.05$

1 As reported by CPDB.

2 P-value listed next to control incidence is the statistical significance associated with testing whether the dose-response curve is different from zero (two-tailed), as reported in the CPDB. P-value listed next to dose groups is the result of pairwise comparison with the control group using the Fisher exact test.

3 Dose group dropped due to nonlinearity, as determined by Mstage. If the p-value for the goodness-of-fit test is less than 0.05, nonlinearity is indicated. Following the U.S. EPA (Anderson *et al.*, 1983), the mid- and high-dose groups were dropped in this analysis.

MeIQ (2-Amino-3,4-dimethylimidazo[4,5-*f*]quinoline) (CAS No. 77094-11-2)

Results from the studies of Ohgaki *et al.* (1986) are listed in the CPDB. Ohgaki *et al.* (1986) exposed male and female CDF$_1$ mice to MeIQ (2-amino-3,4-dimethylimidazo[4,5-f]quinoline) via diet for 91 weeks. Females were exposed to average doses of 13 and 52 mg/kg-day, while males were exposed to 12 and 48 mg/kg-day. The effective tumor incidence was reported by the authors and tabulated in the CPDB. Statistically significant increases in forestomach tumors were observed in males and females. Liver tumors in female mice also showed statistically significant increases with increasing dose. Cancer potency values associated with sites having significant tumor increases are shown in Table 4.

As evident from Table 4, the most sensitive sex/species is the female mouse and the most sensitive site is the forestomach. The potency is calculated based on the incidence of forestomach tumors and liver tumors, shown in Table 5. The probability distributions for q_1 were generated for both tumor sites, using likelihood methods and the modified MSTAGE computer program (see Methodology). The distribution of the sum of the q_1s for each site was obtained using a Monte Carlo simulation. The upper 95 percent confidence bound on q_1 was determined based on the distribution of the sum of potencies from the MeIQ-affected sites. The human cancer potency for MeIQ is estimated to be 1.5 $(mg/kg\text{-}day)^{-1}$ and the associated NSRL is 0.46 µg/day.

Table 4: Values Used in Calculating Human Cancer Potency for MeIQ based on Ohgaki *et al.* (1986)

Sex/species/site	q_1* (mg/kg-day)$^{-1}$	Interspecies Scaling Factor (kg/kg)	Correction for Experiment Duration (wk/wk)	q_{human} (mg/kg-day)$^{-1}$	Goodness-of-Fit Test[1]
Female mouse forestomach	0.06337	$(70/0.025)^{1/3}$	$(104/91)^3$	1.3	p = 0.6549
Female mouse liver	0.01719	$(70/0.025)^{1/3}$	$(104/91)^3$	0.36	p = 1
Female mouse forestomach and liver, combined[2]	0.07253	$(70/0.025)^{1/3}$	$(104/91)^3$	1.5	NA
Male mouse forestomach	0.02304	$(70/0.0.03)^{1/3}$	$(104/91)^3$	0.46	p = 1

1 A p-value of greater than 0.05 for the chi-square goodness of fit test indicates an adequate fit.
2 Potency estimate is based on the combined distribution for q_1 for these sites.

Table 5: Incidence of Forestomach and Liver Tumors in Female CDF_1 Mice Treated with MeIQ Via Diet (Ohgaki *et al.*, 1986)

Average Dose[1] (mg/kg-day)	Forestomach Tumors	Statistical Significance[2]	Liver Tumors	Statistical Significance[2]
0	0/40	p < 0.0005	0/40	p < 0.0005
13	19/36	p < 0.001	4/36	p < 0.05
52	34/38	p < 0.001	27/38	p < 0.001

1 As reported in the CPDB.
2 P-value listed next to control incidence is the statistical significance associated with testing whether the dose-response curve is different from zero (two-tailed), as reported in the CPDB. P-value listed next to dose groups is the result of pairwise comparison with the control group using the Fisher exact test.

MeIQx (2-Amino-3,8-dimethylimidazo[4,5-*f*]quinoxaline) (CAS No. 77500-04-0)

The CPDB lists several groups of studies. Results from the first set of studies were reported in two publications (Ohgaki *et al*., 1987; Wakabayashi, 1992). Kato *et al.* (1988) reported a second set of studies and Kushida *et al.* (1994) a third set. In the studies by Ohgaki *et al.* and Wakabayashi, male and female CDF_1 mice were exposed to MeIQx (2-amino-3,4-dimethylimidazo[4,5-f]quinoxaline) via diet for 84 weeks. Average doses were 78 and 72 mg/kg-day for female and male mice respectively. Kato *et al.* exposed male and female F344/DuCrj rats to MeIQx via diet for 61 weeks. Average doses were 20 and 16 mg/kg-day for female rats, and male rats respectively. Kushida *et*

al. exposed male F344/DuCrj rats to MeIQx via diet for 56 weeks. The effective tumor incidence was reported by the authors for all three sets of studies and tabulated in the CPDB. Statistically significant increases in liver tumors were observed in male and female mice. Lung tumors in female mice also showed statistically significant increases with increasing dose. Statistically significant increases in clitoral gland and Zymbal's gland squamous cell carcinomas were observed in female rats. In both the Kato *et al.* and Kushida *et al.* studies of males rats, Zymbal's gland, skin and liver tumors were significantly increased. Rats were determined to be the most sensitive species and males the most sensitive sex. The Kushida *et al.* study was superior for dose-response analysis, because of the multiple low dose groups. This study was selected as the basis for the cancer potency analysis. Cancer potency values associated with sites having significant tumor increases in the Kushida *et al.* study are shown in Table 6. The dose-response data from Kushida *et al.* are shown in Table 7. The probability distributions for q_1 were generated for Zymbal's gland, liver and skin tumors from the Kushida *et al.* incidence data, using likelihood methods and the modified MSTAGE computer program (see Methodology). The distribution of the sum of the q_1s for each site was obtained using a Monte Carlo simulation. The upper 95 percent confidence bound on q_1 was determined based on the distribution of the sum of potencies from these MeIQx-affected sites. The human cancer potency for MeIQx is estimated to be 1.7 $(mg/kg\text{-}day)^{-1}$, with an associated NSRL of 0.41 µg/day.

Table 6: Values Used in Calculating Human Cancer Potency for MeIQx from Data in Male Rats based on Kushida *et al.* (1994)

Site	q_1* $(mg/kg\text{-}day)^{-1}$	Interspecies Scaling Factor (kg/kg)	Correction for Experiment Duration (wk/wk)	q_{human} $(mg/kg\text{-}day)^{-1}$	Goodness-of-Fit Test[1]
Liver	0.04164	$(70/0.5)^{1/3}$	$(104/56)^3$	1.4	p = 0.8574
Zymbal's gland	0.01886	$(70/0.5)^{1/3}$	$(104/56)^3$	0.63	p = 0.9608
Skin	0.01426	$(70/0.5)^{1/3}$	$(104/56)^3$	0.47	p = 0.6752
Liver, Zymbal's gland, and skin, combined[2]	0.05107	$(70/0.5)^{1/3}$	$(104/56)^3$	1.7	NA

1. A p-value of greater than 0.05 for the chi-square goodness of fit test indicates an adequate fit.
2 Potency estimate is based on the combined distribution for q_1 for these sites.

Table 7: Incidence of Liver, Zymbal's Gland, and Skin Tumors in Male Rats Treated with MeIQx Via Diet (Kushida *et al.*, 1994)

Average Dose[1] (mg/kg-day)	Liver Tumors	Statistical Significance[2]	Zymbal's Gland Tumors	Statistical Significance[2]	Skin Tumors	Statistical Significance[2]
0	0/15	P < 0.0005	0/15	p < 0.0005	0/15	p < 0.002
4	5/30	P = 0.1166	1/30	p = 0.6667	0/30	p = 1.000
8	26/29	p < 0.001	5/29	p = 0.1094	3/29	p = 0.2759
16	16/16	p < 0.001	13/16	p < 0.001	6/16	p < 0.05

1 As reported in the CPDB.

2 P-value listed next to control incidence is the statistical significance associated with testing whether the dose-response curve is different from zero (two-tailed), as reported by Gold and Zeiger (1997). P-value listed next to dose groups is the result of pairwise comparison with controls using the Fisher exact test.

METHYL CARBAMATE (CAS No. 598-55-0)

Results from the studies of NTP (1987) in male and female $B6C3F_1$ mice and male and female F344/N rats are listed in the CPDB. NTP (1987) exposed the animals to methyl carbamate via gavage for 24 months. Groups of 50 mice were exposed to 354 mg/kg-day or 707 mg/kg-day. Groups of 50 rats were exposed to 70.7 mg/kg-day or 142 mg/kg-day. Groups of 50 mice and rats were used as concurrent controls. Animals were sacrificed at 24 months. A statistically significant increase in liver tumors was observed in high dose female rats. The CPDB listed no other statistically significant result but did note that NTP (1987) interpreted the finding of liver tumors in male rats as providing additional evidence of carcinogenicity. The most sensitive sex/species/site is the female rat liver. The values used in the potency calculation are shown in Table 8. The potency is calculated based on the dose-response data for liver tumors in female rats, shown in Table 9. The human cancer potency for methyl carbamate is estimated to be 0.0044 $(mg/kg\text{-}day)^{-1}$, and the associated NSRL is 160 $\mu g/day$.

Table 8: Values Used in Calculating Human Cancer Potency for Methyl Carbamate based on NTP (1987)

Sex/species/site	q_1* $(mg/kg\text{-}day)^{-1}$	Interspecies Scaling Factor (kg/kg)	Correction for Experiment Duration (wk/wk)	q_{human} $(mg/kg\text{-}day)^{-1}$	Goodness-of-Fit Test[1]
Female rat liver	0.0007560	$(70/0.35)^{1/3}$	$(104/104)^3$	0.0044	p = 0.4503

1 A p-value of greater than 0.05 for the chi-square goodness of fit test indicates an adequate fit.

Table 9: Incidence of Liver Tumors in Female F344/N Rats Treated with Methyl Carbamate Via Gavage (NTP, 1987)

Average Dose[1] (mg/kg-day)	Tumor Incidence	Statistical Significance[2]
0	0/50	$p < 0.006$
70.7	0/50	$p = 1$
142	6/50	$p < 0.05$

1 As reported in the CPDB.

2 P-value listed next to control incidence is the statistical significance associated with testing whether the dose-response curve is different from zero (two-tailed), as reported in the CPDB. P-value listed next to dose groups is the result of pairwise comparison with the control group using the Fisher exact test.

4-(N-NITROSOMETHYLAMINO)-1-(3-PYRIDYL)-1-BUTANONE (CAS No. 64091-91-4)

Results from the studies of Rivenson *et al.* (1988), Furukawa *et al.* (1994) and Hecht *et al.* (1996) are listed. Rivenson *et al.* (1988) exposed male F344/N rats to 4-(N-nitrosomethylamino)-1-(3-pyridyl)-1-butanone (NNK) via drinking water for 27 months. Dose levels were 0, 0.025, 0.050 and 0.250 mg/kg-day on average. Eighty animals were in each group with the exception of the highest dose level, which had 30 animals in the group. Animals were sacrificed at 30 months. Statistically significant increases in lung tumors, pancreas exocrine tumors, liver tumors and nasal tumors were observed. Hecht *et al.* (1996) exposed male F344/N rats to NNK via drinking water for 26 months. Dose levels were 0 and 0.100 mg/kg-day on average. The control group had approximately 20 animals and the exposed group approximately 60 animals (the CPDB reported effective number, so the precise group size cannot be determined). Statistically significant increases in lung tumors were observed. Furukawa *et al.* (1994) exposed Syrian golden hamsters to the chemical via drinking water for 87 weeks. No significant increases in tumor incidences were observed. The study by Rivenson *et al.* was selected as the highest quality study for dose-response analysis, because it had the greatest number of animals per group, the largest number of dose groups, and the lowest doses.

Potency estimates based on the Rivenson *et al.* study are shown in Table 10. The dose-response data used in the analysis are shown in Table 11. The multistage model did not fit adequately to dose-response data for pancreatic tumors. Following U.S. EPA methodology described in Anderson *et al.* (1983), whenever the multistage model does not fit the data adequately, data at the highest dose are deleted and the model fitted to the remaining data. This is repeated until an acceptable fit is obtained, as measured by the chi-square goodness-of-fit test. For the analysis of the pancreas data, the high-dose group was dropped. The probability distributions for q_1 were generated for lung, pancreas, liver and nasal cavity tumors from the Rivenson *et al.* incidence data, using likelihood

methods and the modified MSTAGE computer program (see Methodology). The distribution of the sum of the q_1s for each site was obtained using a Monte Carlo simulation. The upper 95 percent confidence bound on q_1 was determined based on the distribution of the sum of potencies from the NNK-affected sites. The human cancer potency for this chemical is estimated to be 49 $(mg/kg\text{-}day)^{-1}$, with an associated NSRL of 0.014 µg/day.

Table 10: Values Used in Calculating Human Cancer Potency for NNK from Data in Male Rats based on Rivenson *et al.* (1988)

Site	q_1* $(mg/kg\text{-}day)^{-1}$	Interspecies Scaling Factor (kg/kg)	Correction for Experiment Duration (wk/wk)	q_{human} $(mg/kg\text{-}day)^{-1}$	Goodness-of-Fit Test[1]
Lung	5.412	$(70/0.5)^{1/3}$	$(104/104)^3$	28	0.4005
Pancreas	3.434	$(70/0.5)^{1/3}$	$(104/104)^3$	18	NA
Liver	2.309	$(70/0.5)^{1/3}$	$(104/104)^3$	12	0.074
Nasal cavity	1.039	$(70/0.5)^{1/3}$	$(104/104)^3$	5.4	1
Lung, pancreas, liver, and nasal, combined[2]	9.373	$(70/0.5)^{1/3}$	$(104/104)^3$	49	NA

1 A p-value of greater than 0.05 for the chi-square goodness of fit test indicates an adequate fit.
2 Potency estimate is based on the combined distribution for q_1 for these sites.

Table 11: Incidence of Lung, Pancreas, Liver and Nasal Tumors in Male F344/N Rats Treated with NNK Via Drinking Water (Rivenson *et al.*, 1988)

Average Dose[1] (mg/kg-day)	Lung Tumors	Statistical Significance[2]	Pancreas Exocrine Tumors	Statistical Significance[2]	Liver Tumors	Statistical Significance[2]	Nasal Tumors	Statistical Significance[2]
0	6/80	p < 0.0005	1/80	p < 0.006	6/80	p < 0.0005	0/80	p < 0.0005
0.025	9/80	p = 0.2945	5/80	p <= 0.1049	3/80	p < 0.001	1/80	p < 0.001
0.050	20/80	p < 0.01	9/80	p < 0.01	11/80	p < 0.001	2/80	p < 0.001
0.250	27/30	p < 0.001	2/30[3]	p = 0.1801	12/30	p < 0.001	5/30	p < 0.001

1 As reported in the CPDB.
2 P-value listed next to control incidence is the statistical significance associated with testing whether the dose-response curve is different from zero (two-tailed), as reported by Gold and Zeiger (1997). P-value listed next to dose groups is the result of pairwise comparison with the control group using the Fisher exact test.
3 Dose group dropped due to nonlinearity, as determined by Mstage. If the p-value for the goodness-of-fit test is less than 0.05, nonlinearity is indicated. Following the U.S. EPA (Anderson *et al.*, 1983), the high-dose group was dropped in this analysis.

TRIMETHYL PHOSPHATE (CAS No. 512-56-1)

Results from the studies of NCI (1978) in male and female $B6C3F_1$ mice and male and female F344/N rats are listed in the CPDB. NCI (1978) exposed male and female mice and rats to trimethyl phosphate (TMP) via gavage for 24 months. Groups of 50 mice were exposed to 107 mg/kg-day or 214 mg/kg-day. Groups of 50 rats were exposed to 21.2 mg/kg-day or 42.5 mg/kg-day. Groups of 20 mice and rats were used as concurrent controls. Animals were sacrificed at 24 months. Statistically significant increases in uterus endometrium adenocarcinoma were observed in female mice and subcutaneous fibromas in male rats. Potency estimates associated with these data are shown in Table 12. The most sensitive sex/species is the female mouse. The potency is calculated based on the incidence of uterus endometrium adenocarcinoma, shown in Table 13. The human cancer potency for TMP is estimated to be 0.029 $(mg/kg\text{-}day)^{-1}$, with an associated NSRL of 24 µg/day.

Table 12: Values Used in Calculating Human Cancer Potency for TMP based on NCI (1978)

Sex/species/site	q_1* $(mg/kg\text{-}day)^{-1}$	Interspecies Scaling Factor (kg/kg)	Correction for Experiment Duration (wk/wk)	q_{human} $(mg/kg\text{-}day)^{-1}$	Goodness-of-Fit Test[1]
Female mouse uterine endometrium adenocarcinoma	0.002024	$(70/0.025)^{1/3}$	$(104/104)^3$	0.029	p = 1
Male rat subcutaneous fibroma	0.005129	$(70/0.5)^{1/3}$	$(104/104)^3$	0.027	P = 0.9635

1 A p-value of greater than 0.05 for the chi-square goodness of fit test indicates an adequate fit.

Table 13: Incidence of Uterus Endometrium Adenocarcinoma in Female $B6C3F_1$ Mice Treated with TMP Via Gavage (NCI, 1978)

Average Dose[1] (mg/kg-day)	Uterus Endometrium Tumors	Statistical Significance[2]
0	0/20	p < 0.002
107	7/50	p = 0.08
214	13/49	p < 0.01

1 As reported in the CPDB.

2 P-value listed next to control incidence is the statistical significance associated with testing whether the dose-response curve is different from zero (two-tailed), as reported in the CPDB. P-value listed next to dose groups is the result of pairwise comparison with the control group using the Fisher exact test.

REFERENCES

Anderson EL and the U.S. Environmental Protection Agency Carcinogen Assessment Group (1983). Quantitative approaches in use to assess cancer risk. *Risk Anal* **3**:277-295.

Crouch E (1992). MSTAGE (Version 1.1). E.A.C. Crouch, Cambridge Environmental Inc., 58 Buena Vista Road, Arlington, Massachusetts 02141.

Crump KS, Guess HA, Deal LL (1977). Confidence intervals and test of hypotheses concerning dose-response relations inferred from animal carcinogenicity data. *Biometrics* **33**:437-451.

Crump KS, Howe RB, Van Landingham C, Fuller WG (1993). TOX_RISK Version 3. TOXicology RISK Assessment Program. KS Crump Division, Clement International Corporation, 1201 Gaines Street, Ruston, Louisiana 71270.

Furukawa F, Nishikawa A, Yoshimura H, Mitsui M, Imazawa T, Ikezaki S, Takahashi M (1994). Effects of 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) on N-nitrosobis(2-oxopropyl)amine (BOP)-initiated carcinogenesis in hamsters. *Cancer Lett* **86**:75-82.

Gold LS and Zeiger E (1997). *Handbook of Carcinogenic Potency and Genotoxicity Databases*. CRC Press, Inc. Boca Raton.

Gold LS, Manley NB, Slone TH, Rohrbach L (1999). Supplement to the Carcinogenic Potency Database (CPDB): Results of animal bioassays published in the general literature in 1993 to 1994 and by the National Toxicology Program in 1995 to 1996. *Environ Hlth Perspec* **107** (Suppl. 4):527-600.

Hecht SS, Trushin N, Rigotty J, Carmella SG, Borukhova A, Akerkar S, Rivenson A (1996). Complete inhibition of 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone-induced rat lung tumorigenesis and favorable modification of biomarkers by phenethyl isothiocyanate. *Cancer Epidemiol Biomark Prev* **5**:645-652.

Hoover SM, Zeise L, Pease WS, Lee LE, Hennig MP, Weiss LB, Cranor C (1995). Improving the regulation of carcinogens by expediting cancer potency estimation. *Risk Anal* **15**(2):267-80.

Kato T, Ohgaki H, Hasegawa H, Sato S, Takayama S, Sugimura T (1988). Carcinogenicity in rats of a mutagenic compound, 2-amino-3,8-dimethylimidazo[4,5-f] quinoxaline. *Carcinogenesis* **9**:71-73.

Kushida H., Wakabayashi K, Sato H, Katami M, Kurosaka R, Nagao M (1994). Dose-response study of MeIQx carcinogenicity in F344 male rats. *Cancer Lett* **83**:31-35.

National Cancer Institute (NCI, 1978). *Bioassay of Trimethylphosphate for Possible Carcinogenicity (CAS No. 512-56-1).* NIH Technical Reports Series No. 81. NIH

Publication No. 78-1331. U.S. Department of Health, Education and Welfare, Public Health Service, National Institutes of Health.

National Toxicology Program (NTP, 1987). *Toxicology and Carcinogenesis Studies of Methyl Carbamate (CAS No. 598-55-0) in F344/N Rats and B6C3F$_1$ Mice (Gavage Studies).* NTP Technical Report Series No. 328. NIH Publication No. 88-2584. U.S. Department of Health and Human Services, NTP, Research Triangle Park, NC.

Office of Environmental Health Hazard Assessment (OEHHA, 1992). *Expedited Cancer Potency Values and Levels for Certain Proposition 65 Carcinogens.* California Environmental Protection Agency, OEHHA, Reproductive and Cancer Hazard Assessment Section, April 1992.

Ohgaki H, Hasegawa H, Suenaga M, Kato T, Sato S, Takayama S, Sugimura T (1986). Induction of hepatocellular carcinoma and highly metastatic squamous cell carcinomas in the forestomach of mice by feeding 2-amino-3,4-dimethylimidazo[4,5-f]quinoline. *Carcinogenesis* **7**:1889-1893.

Ohgaki H, Hasegawa H, Suenaga M, Sato S, Yakayama S, Sugimura T (1987). Carcinogenicity in mice of a mutagenic compound, 2-amino-3,8-dimethylimidazo[4,5-f]quinoxaline (MeIQx) from cooked foods. *Carcinogenesis* **8**:665-668.

Rivenson A, Hoffmann D, Prokopczyk B, Amin S, Hecht S (1988). Induction of lung and exocrine pancreas tumors in F344 rats by tobacco-specific and areca-derived N-nitrosamines. *Cancer Res* **48**:6912-6917.

Tsuda H, Hagiwara A, Shibata M, Ohshima M, Ito N (1982). Carcinogenic effect of carbazole in the liver of (C57BL/6N x C3H/HeN)F_1 mice. *J Natl Cancer Inst* **69**:1383-1387.

Wakabayashi K, Nagao M, Esumi H, Sugimura T (1992). Food-derived mutagens and carcinogens. *Cancer Res* **52**(Suppl):2092s-2098s.