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# Detection of carcinogens as mutagens in the *Salmonella*/microsome test: Assay of 300 chemicals*

**(rapid *in vitro* screening/environmental carcinogens and mutagens)**

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**ABSTRACT** **About 300 carcinogens and non-carcinogens of a wide variety of chemical types have been tested for mutagenicity in the simple *Salmonella*/microsome test. The test uses bacteria as sensitive indicators for DNA damage, and mammalian liver extracts for metabolic conversion of carcinogens to their active mutagenic forms. Quantitative mutagenicity data from linear dose-response curves are presented: potency varies over a $10^6$-fold range. There is a high correlation between carcinogenicity and mutagenicity: 90% (156/174) of carcinogens are mutagenic in the test and despite the severe limitations inherent in defining non-carcinogenicity, few "non-carcinogens" show any degree of mutagenicity. The results also demonstrate the great utility, and define the limitations, of the test in detecting environmental carcinogens.**

There is considerable evidence that a large proportion of human cancer may be caused by exposure to toxic chemicals in the environment, very few of which have been tested for carcinogenicity or mutagenicity. A program of cancer prevention aimed at identifying and eliminating human exposure to hazardous chemicals requires the development of rapid, inexpensive, screening methods as complements to expensive, long-term animal tests, to pinpoint dangerous chemicals among the thousands to which humans are exposed. The *Salmonella*/microsome mutagenicity test (1–4) has been sufficiently developed and validated to be seriously considered for widespread use in this way. The considerable evidence (5), much of it obtained using this test (1–4, 6–13), that with few exceptions carcinogens are mutagens, supports the desirability of using this type of rapid and economical test system as a screening technique (1–3).

Chemicals are tested for mutagenicity on petri plates with several specially constructed mutants of *Salmonella typhimurium* (2–4). Homogenates of rat (or human) liver, (S-9 Mix), are added directly to the petri plates, thus incorporating an important aspect of mammalian metabolism into the *in vitro* test (2). In this way, a wide variety of carcinogens requiring metabolic activation can be detected easily as mutagens (2, 4, 5). The system has been recently reviewed (6) and the test method described in detail (7).

The present paper presents mutagenicity data on a large number of carcinogens and non-carcinogens of many different classes that have been examined in the system using a standard methodology (7) to determine the correlation between mutagenicity and carcinogenicity, and the utility of the test, at this stage of its development, for the detection of various types of carcinogens.

## DISCUSSION AND RESULTS

Results are in Table 1. For each chemical, quantitative data are presented as revertants per plate (histidine revertants on a petri plate/number of micrograms tested). For mutagenic compounds, data are from the linear region of dose-response curves, and for non-mutagenic chemicals, data are presented as less-than figures at the highest dose-level tested. The number of revertants per nmol is an indication of the mutagenic potency of the chemical in the test system.

All non-mutagens, and most mutagens, have been tested, using the recently improved standard methodology (7), on the new R factor tester strains (4) as well as the earlier standard tester strains (3). Non-mutagens have been tested over a wide dose range both with and without the liver microsome activating system.

In addition to our previously published studies and new work presented here, results have been contributed to this compilation by a number of laboratories using this test, including a large contribution from Japan. Some of the chemicals tested are also specified in a contract sponsored by the National Cancer Institute (V. Simmon and H. Rosenkranz, to be published). Some of the non-carcinogens we tested were specified in a contract sponsored by the Environmental Protection Agency to B. Commoner.

We have not reported on any metal carcinogens, though three or four that have been tested are negative in the standard test. The test system is not suitable for metals entering the bacteria because of the large amount of Mg salts, citrate, and phosphate in the minimal medium. A number of carcinogenic metals have been shown to be mutagens in bacteria by means of a different methodology, e.g. ref. 14.

In addition to the compounds presented we have tested 46 common biochemicals that are non-carcinogens, or presumed non-carcinogens, and have found that all were negative in the test (<0.01 revertants/nmol). These data will be presented in a companion paper that will appear in this journal. The companion paper will also discuss the results presented here, the utility and limitations of the test in detecting chemicals likely to be environmental mutagens and carcinogens for humans, quantitation of mutagenic potency, and somatic mutation as the most attractive hypothesis to explain chemical carcinogenesis.

B.N.A. would like to dedicate this paper to the memory of Gordon M. Tomkins (1926–1975), friend and colleague of 20 years. We thank S. Barnes Jolley, D. Streitwieser, G. Jen, V. Donahue, G. Stark, L. Haroun, D. Maron, and T. Keng, who were of invaluable assistance in this work. We also gratefully acknowledge V. Simmon, T. Sugimura, T. Matsushima, H. Bartsch, E. and J. Weisburger, J. and E. Miller, J. Arcos, L. Tomatis, H. Rosenkranz, L. Poirier, and numerous colleagues, who provided criticisms and unpublished information. This work has been supported by Energy Research and Development Administration Grant AT (04-3)34 P.A. 156 to B.N.A. J.McC. was supported by a postdoctoral fellowship from the California Division of the American Cancer Society.

* This is part I of a series.

Table 1. Carcinogenicity versus mutagenicity in the *Salmonella* test

| | Car. | Mut. | Revertants per nmol | Revertants per plate | strain | S9 | Ref. |
|---|---|---|---|---|---|---|---|
| **A) Aromatic amines etc.** | | | | | | | |
| 1) N-acetoxy-2-acetylaminofluorene[w] | + | + | 50 | 806/5 | 98 | - | 15/X,9 |
| 2) 2-aminofluorene[s] | + | + | 205 | 11300/10 | 87 | P | 16/2 |
| 3) 2-acetylaminofluorene[a] | + | + | 108 | 4823/10 | 8907 | P | 16/X,2 |
| 4) N-hydroxy-2-acetylaminofluorene[w] | + | + | 48 | 1004/5 | 98 | A- | 15/X,3,11 |
| 5) N-hydroxy-2-aminofluorene[w] | + | + | 583 | 1480/.5 | 8 | - | 15/11,9 |
| 6) 2-nitrosofluorene[w] | + | + | 1040 | 1327/.25 | 8 | - | 15/3,9 |
| 7) 7-hydroxy-2-acetylaminofluorene[w] | c0 | 0[#] | 0.42[#] | 436/250 | 890 | P | 15/X,k |
| 8) 1-hydroxy-2-acetylaminofluorene[w] | 0 | 0 | <0.02 | <70/1000 | 0957 | A | 15/X,9,k |
| 9) 3-hydroxy-2-acetylaminofluorene[w] | 0 | 0 | <0.02 | <70/1000 | 0957 | A | 15/X,9,k |
| 10) 5-hydroxy-2-acetylaminofluorene[w] | 0 | w+[#] | 0.04 | 84/500 | 809 | A | 15/X,9,k |
| 11) 4-acetylaminofluorene[v] | 0 | +[#] | 0.3 | 272/200 | 8 | P | 15/X,k |
| 12) 2,7-bis-acetylaminofluorene[s] | + | +[#] | 9.5 | 338/10 | 8 | P | 15/X |
| 13) 2,7-diaminofluorene[g] | + | + | 22 | 2243/20 | 98 | A- | 16/X,3 |
| 14) 6-aminochrysene[s] | + | + | 156[*] | 638/1 | 8907 | A | 18/2,X |
| 15) 1-aminopyrene[s] | ? | + | 23 | 5370/50 | 9087 | A | ?/X,2 |
| 16) 9-aminoacridine[g] | ? | + | 10[*] | 7900/150 | 7 | - | ?/3 |
| 17) acridine orange[e] | + | + | 66 | 4370/20 | 097 | A | 15/X,k |
| 18) proflavine[g] | ? | + | 38 | 708/5 | 8 | A | 15/X |
| 19) 2-aminoanthracene[s] | + | + | 510 | 5251/2 | 9085 | P | 15/X,4,2 |
| 20) 1-aminoanthracene[a] | w+ | +[#] | 22 | 2300/20 | 90 | P | 15/X,k |
| 21) β-naphthylamine[g] | + | + | 8.5 | 590/10 | 058 | A | 17/X,2 |
| 22) α-naphthylamine[g] | c0 | +[#] | 0.42 | 290/100 | 08 | A | 15/X |
| 23) β-naphthylhydroxylamine[v] | + | + | 21 | 671/5 | 0985 | - | 15/X,k |
| 24) α-naphthylhydroxylamine[v] | + | + | 229 | 1439/1 | 09 | - | 17/X |
| 25) 2-nitrosonaphthalene[w] | + | + | 0.16 | 102/100 | 8 | - | 15/X,9 |
| 26) 4-aminobiphenyl[s] | + | + | 31 | 9130/50 | 087 | A | 17/X,2 |
| 27) 2',3-dimethyl-4-aminobiphenyl[i] | + | + | 75 | 3240/10 | 098 | A | 15/X,k |
| 28) 2-aminobiphenyl[a] | ? | +[#] | 0.51 | 606/200 | 0 | A | 15,16/X |
| 29) benzidine[a] | + | + | 1.4 | 265/50 | 8 | P | 17/2 |
| 30) N,N,N',N'-tetramethylbenzidine[a] | ?0 | 0 | <0.02 | <70/1000 | 9075 | A | 15/X |
| 31) 3,3',5,5'-tetramethylbenzidine[a] | 0 | 0 | <0.03 | <70/500 | 0957 | A | 20/X |
| 32) 4-amino-trans-stilbene[s] | + | + | 19 | 800/10 | 80 | P | 15/2,X |
| 33) 4-dimethylamino-trans-stilbene[s] | + | + | 22 | 843/10 | 80 | P | 15/2,X |
| 34) 4,4'-methylene-bis-2-chloro aniline [MOCA][v] | + | + | 2.7 | 1050/100 | 0 | A | 17/X,k |
| 35) 4,4'-[(4-imino-2,5-cyclohexadien-1-ylidene)methylene] dianiline·HCl[para-rosaniline] | ?+ | 0 | <0.02 | <70/1000 | 0957 | A | 17/X |
| 36) p-aminodiphenylamine[g] | 0 | 0 | <0.01 | <70/1000 | 0957 | A | 15/X |
| 37) bis-dimethylaminodiphenylmethane[i] | ? | 0 | <0.036 | <70/500 | 0957 | A | 15/X,k |
| 38) p-aminophenol[a] | 0 | 0 | <0.01 | <70/500 | 0957 | A | 17/X |
| 39) aniline[a] | ? | 0 | <0.005 | <70/1000 | 0957 | A | 17,15/X,k |
| 40) sulfaguanidine[g] | ?0 | 0 | <0.003 | <70/5000 | 0957 | A | 16/X |
| 41) o-toluidine[e] | c0[T] | 0 | <0.0007 | <70/10[4] | 0957 | A | 15/X |
| 42) 2,4-diaminotoluene[e] | + | + | 0.43 | 270/100 | 80 | A | 15/13,X |
| 43) auramine[a] | + | 0 | <0.01 | <70/2000 | 0957 | A | 15/X,2 |
| 44) ethidium bromide[c] | ? | + | 80 | 1012/5 | 98 | A | ?/X,s |
| **B) Alkyl halides etc.** | | | | | | | |
| 1) methyl iodide | ?+ | w+ | | G | 0 | - | 16/k |
| 2) carbon tetrachloride[m] | + | 0 | <0.001 | <70/10[4] | 05 | A | 17/X |
| 3) ethylene dibromide[a] | + | w+ | 0.06 | 128/4344 | 05 | - | 35/X,1 |
| 4) vinyl chloride[d] | + | w+ | | G | 05 | P- | 17/36,22,8 |
| 5) vinylidene chloride | ?+[T] | w+ | | G | 05 | P | 34/34,k |
| 6) chloroacetic acid[a] | 0 | 0 | <0.007 | <70/1000 | 0957 | A | 37/X,8 |
| 7) 2-chlorobutadiene [chloroprene] | ?+[T] | +0 | | G | 0 | P- | 38/34,r |
| 8) bis-chloromethyl ether [BCME] | + | + | | | 4 | - | 17/39 |
| 9) bis-(2-chloroethyl)amine[a] | ? | + | 9.7 | 2710/50 | 50 | - | /4 |
| 10) methyl-bis-(2-chloroethyl) amine [nitrogen mustard][a] | + | + | 1.3 | 333/50 | 50 | - | 16/X,l |
| 11) uracil mustard[a] | + | + | 0.40 | 157/100 | 5 | - | 16/X,f,n |
| 12) cyclophosphamide[e] | + | + | 1.4 | 1079/200 | 50 | A | 16/8,m |
| 13) isophosphamide[i] | + | + | 0.26 | 1001/1000 | 50 | A | 25/f |
| 14) chlornaphazin[j] | + | + | 5.6 | 207/10 | 0 | A | 16,70/X |
| 15) melphalan[i] | + | + | 0.29 | 187/200 | 50 | A- | 40/f |
| 16) 2-methoxy-6-chloro-9-[4-bis(2-chloroethyl)amino-1-methylbutylamino]-acridine·2HCl [ICR-10] | + | + | 90 | 1660/10 | 7 | - | d/X |
| 17) 2-methoxy-6-chloro-9-(3-(ethyl-2-chloroethyl)aminopropylamino) acridine·2HCl [ICR-170][u] | + | + | 260 | 6410/10 | 7 | - | d/X |
| 18) 2-methoxy-6-chloro-9-(3-(2-chloro ethyl)aminopropylamino) acridine·2HCl [ICR-191][u] | ?0 | + | 511 | 566/.5 | 7890 | - | d/3 |
| 19) dimethylcarbamyl chloride[a] | + | w+ | 0.04 | 380/1060 | 50 | - | 16,37/4 |
| 20) benzyl chloride[a] | ?+ | w+ | 0.02 | 230/2000 | 0 | - | 16/4 |
| 21) captan[p] | +[T] | + | 25 | 820/10 | 095 | - | n/4,41 |
| 22) folpet[p] | 0 | + | 64 | 2170/10 | 05 | - | 16/X,42 |
| 23) DDE[k,l] | + | 0 | <0.004 | <70/5000 | 0957 | A | 17/X |
| 24) dieldrin[l] | + | 0 | <0.003 | <70/10[4] | 0957 | A | 17/X |
| **C) Polycyclic aromatics** | | | | | | | |
| 1) dibenzo(a,e)pyrene | + | | | | 8 | + | 17/56 |
| 2) dibenzo(a,i)pyrene[t] | + | + | 20 | 1290/20 | 098 | A | 17/X,56 |
| 3) benzo(a)pyrene[a] | + | + | 121 | 2398/5 | 0987 | A | 17/4,2 |
| 4) 6-hydroxymethylbenzo(a)pyrene[r] | + | + | 270 | 482/.5 | 08 | A | 17/X,q |
| 5) benzo(a)pyrene-4,5-oxide[q] | + | + | 295 | 5500/5 | 87 | - | q/7 |
| 6) 3-hydroxybenzo(a)pyrene[t] | ?0 | + | 111 | 416/1 | 09 | A- | 16/X |
| 7) 7,8-dihydrobenzo(a)pyrene[q] | + | + | 20 | 797/10 | 87 | A | b/X |
| 8) benzo(e)pyrene[t] | w+ | + | 0.60 | 143/60 | 0 | A | 17/X |
| 9) dibenz(a,j)acridine[t] | + | + | 18 | 318/5 | 07 | A | 17/X |
| 10) dibenz(a,c)anthracene[q] | + | + | 175 | 6280/10 | 0987 | A | 15/X |
| 11) dibenz(a,h)anthracene[q] | + | + | 11 | 401/10 | 097 | A | 17/X,56 |
| 12) dibenz(a,h)anthracene-5,6-oxide[q] | 0 | + | 5.3 | 90/5 | 7 | - | 57/10 |
| 13) 3-methylcholanthrene[e] | + | + | 58 | 2160/10 | 087 | A | 15/X,2 |
| 14) chrysene[t] | w+ | + | 38 | 1670/10 | 09 | A | 17/X |
| 15) chrysene-5,6-oxide[q] | ? | + | 2.2 | 450/50 | 07 | - | 57/X |
| 16) benz(a)anthracene[q] | + | + | 11 | 2640/57 | 09 | A | 17/X,56,k |
| 17) benz(a)anthracene-5,6-oxide[q] | w+ | + | | | | - | 57/10 |
| 18) 7-methylbenz(a)anthracene[q,r] | + | + | 22 | 897/10 | 0 | A | 57/X |
| 19) 7,12-dimethylbenz(a)anthracene[g] | + | + | 19 | 1458/20 | 0978 | A | 15/4,2 |
| 20) 7-hydroxymethyl-12-methylbenz(a) anthracene[r] | + | + | 30 | 547/5 | 0 | A | 16/X |
| 21) 7-chloromethyl-12-methylbenz(a) anthracene[u] | + | + | 80 | 274/1 | 07 | - | d/X |
| 22) 7-chloromethylbenz(a)anthracene[u] | + | + | 121 | 440/1 | 0 | - | d/X |
| 23) 7-bromomethyl-12-methylbenz(a) anthracene | + | + | 20 | 604/10 | 98 | - | 16/k |
| 24) 7,9-dimethylbenz(c)acridine[g] | + | + | 9.4 | 730/20 | 09 | A | 15/X |
| 25) 7,10-dimethylbenz(c)acridine[g] | + | + | 15 | 2830/50 | 0 | A | 15/X |
| 26) pyrene[a] | 0 | 0 | <0.02 | <100/1000 | 0957 | A | 15/X |
| 27) anthracene[s] | c0 | 0 | <0.01 | <70/1000 | 0957 | A | 15/X |
| 28) 1,8,9-trihydroxyanthracene[i] | π | ? | <16 | <70/1 | 0957 | A | 15/X |
| 29) 9,10-dichloromethylanthracene[u] | + | + | 88 | 32/.1 | 7 | - | d/X |
| 30) 10-chloromethyl-9-methylanthracene[u] | + | + | 4.6 | 96/5 | 7 | - | d/X |
| 31) 10-chloromethyl-9-chloroanthracene[u] | + | + | 55 | 211/1 | 0 | - | d/X |
| 32) 10-bromomethylanthracene[u] | + | + | 35 | 1298/10 | 0 | - | d/X |
| 33) phenanthrene[t] | 0 | 0 | <0.25 | <70/50 | 0957 | A | 15/X,k |
| 34) 15,16-dihydro-11-methylcyclopenta-(a)phenathrene-17-one[o] | + | + | 84 | 3420/10 | 098 | A | 59/X,e |
| 35) 15,16-dihydro-3-methylcyclopenta-(a)phenanthrene-17-one[o] | 0 | 0 | <0.03 | <70/500 | 0957 | A | 59/X,e |
| 36) fluorene[e] | 0 | 0 | <0.01 | <70/1000 | 0957 | A | 16/X |
| 37) naphthalene[t] | 0 | 0 | <0.09 | <70/100 | 0957 | A | 16/X |
| 38) 1-naphthol[t] | ?0 | 0 | <0.01 | <70/1000 | 0957 | A | 16/X |
| **D) Esters, epoxides, carbamates, etc.** | | | | | | | |
| 1) vinyl acetate[a] | 0 | 0 | <0.0006 | <70/10[4] | 0957 | A | 16/X,47 |
| 2) n-propyl p-hydroxybenzoate ester[a] | 0 | 0 | <0.007 | <70/2000 | 0957 | A | 68/X |
| 3) methyl methanesulfonate[a] | + | + | 0.63 | 3244/570 | 05 | - | 17/4 |
| 4) ethyl methanesulfonate[g] | + | + | 0.16[*] | 12934/10[4] | 05 | - | 17/X,4 |
| 5) ethyl p-toluenesulfonate[v] | w+ | w+ | 0.005 | 115/4700 | 5 | - | 16/n,k |
| 6) diethyl sulfate[f] | w+ | + | [*] | | 50 | - | 17/4,1 |
| 7) malathion[p] | 0 | | <0.05 | <70/500 | 0957 | A | 16/X,k |
| 8) succinic anhydride[g] | ?+ | ? | <0.035 | <70/200 | 0957 | A | 16/X,k |
| 9) β-propiolactone[g] | + | + | 4.1 | 6460/114 | 05 | - | 17/X,1 |
| 10) β-butyrolactone[e] | + | + | | | 4 | - | 16/1 |
| 11) ε-caprolactone[a] | ? | 0 | <0.0008 | <70/10[4] | 0957 | A | ?/X |
| 12) 1,3-propane sultone[a] | + | + | 6.6 | 3770/70 | 5 | - | 17/X,1 |
| 13) styrene oxide | c0[T] | + | 0.37 | 1292/500 | 05 | - | 16/58 |
| 14) 2,3-epoxy-1-propanol [glycidol][a] | ?0 | + | 0.58 | 1730/223 | 50 | - | 16/X,k |
| 15) 2,3-epoxypropionaldehyde [glycidaldehyde][i] | + | + | 19 | 5260/20 | 05 | - | 16/X,k |
| 16) 1,2-epoxybutane[a] | ?0 | w+ | 0.006 | 333/4200 | 05 | - | 16/X,n |
| 17) 1,2,3,4-diepoxybutane[t] | + | + | 0.12 | 71/50 | 5 | - | 16/X |
| 18) 1,2,7,8-diepoxyoctane[a] | + | + | 0.10 | 690/990 | 05 | - | 16/X |
| 19) thiourea[i] | w+ | 0 | <0.001 | <70/5000 | 0957 | A | 17/X |
| 20) methyl carbamate[k] | ?0 | 0 | <0.0005 | <70/10[4] | 0957 | A | 16/X |
| 21) ethyl carbamate [urethane][g] | + | 0 | <0.0006 | <70/10[4] | 0957 | A | 17/X |
| 22) 1-naphthyl-N-methyl carbamate [carbaryl/Sevin][p] | c0 | 0 | <0.008 | <70/2000 | 0957 | A | 16/X,24 |
| 23) 1-phenyl-1-(3,4-xylyl)-2-propynyl-N-cyclohexylcarbamate[w] | + | + | 7.5 | 2087/100 | 05 | - | o/4 |
| 24) 1,1-diphenyl-2-butynyl-N-cyclohexyl carbamate[w] | + | + | 2.5 | 714/100 | 0 | - | 69/X,4 |
| 25) 1,1-diphenyl-2-propynyl-N-cyclohexyl carbamate[w] | + | + | 2.8 | 844/100 | 0 | - | 16/X,4 |
| 26) diphenylethinyl carbinol[w] | 0 | 0 | <0.03 | <70/500 | 0957 | - | o/X |
| 27) thioacetamide[i] | w+ | 0 | <0.001 | <70/5000 | 0957 | A | 17/X |
| 28) acetamide[g] | w+ | 0 | <0.0008 | <70/5000 | 0957 | A | 17/X |

Table 1. (*Continued*)

| | Car. | Mut. | Revertants per nmol | Revertants per plate | strain | S9 | Ref. |
|---|---|---|---|---|---|---|---|
| **E) Nitro Aromatics & Heterocycles** | | | | | | | |
| 1) α,α,α-trifluoro-2,6-dinitro-N,N-dipropyl-p-toluidine[l] | ?[†] | 0 | <0.01 | <70/2500 | 0957 | A | /X |
| 2) 4-nitrobiphenyl[a] | + | + | 11 | 5490/100 | 09 | - | 17/X,a |
| 3) 2-nitronaphthalene[f] | + | + | 8.7 | 5250/100 | 05 | - | 16/X,n |
| 4) 5-nitroacenaphthene | + | + | 17 | 4344/50 | 09 | A- | 15,66/62 |
| 5) 2-nitrofluorene[a] | + | + | 18 | 4200/50 | 809 | - | 15/3,9,a |
| 6) 4-nitroquinoline-1-oxide[s] | + | + | 2906 | 7640/.5 | 0985 | - | 15/4,27 |
| 7) 4-hydroxyaminoquinoline-1-oxide[f] | + | + | 76 | 4300/10 | 08 | - | 15/X,3 |
| 8) p-nitrophenol[t] | ?0 | 0 | <0.02 | <70/500 | 0957 | A | 16/X |
| 9) chloramphenicol[g] | ? | ? | <4.5 | <70/5 | 0957 | A | 16/X |
| 10) 1-(2-hydroxyethyl)-2-methyl-5-nitroimidazole [metronidazole][c] | + | + | 1.1 | 1650/250 | 0 | - | 16/X,63,71 |
| 11) 1,2-dimethyl-5-nitroimidazole[m] | + | + | 3.5 | 1391/50 | 0 | - | 16/X,1,71 |
| 12) 1-(5-nitro-2-thiazolyl)-2-imidazolidinone [niridazole][c] | + | + | 1752 | 1636/.2 | 08 | - | 72/4,63 |
| 13) 1,2-dihydro-2-(5-nitro-2-thienyl)-quinazolin-4-(3H)-one | + | + | | | 09 | - | 64/1 |
| 14) 4-(2-hydroxyethylamino)-2-(5-nitro-2-thienyl)quinazoline | + | + | | | 09 | - | 64/1 |
| 15) 4-bis(2-hydroxyethyl)amino-2-(5-nitro-2-thienyl)quinazoline | + | + | | | 90 | - | 64/1 |
| 16) 2-(2-furyl)-3-(5-nitro-2-furyl)-acrylamide [AF-2][b] | + | + | 20800 | 1674/.02 | 09 | - | 61/4,60 |
| 17) N-[4-(5-nitro-2-furyl)-thiazolyl]-formamide [FANFT][i] | + | + | 16500 | 3260/.05 | 0 | - | 16/X,60 |
| 18) 5-nitro-2-furamidoxime[a] | 0 | + | 5.3 | 678/20 | 0 | - | 16/X,60 |
| 19) 1-[(5-nitrofurfurylidene)-amino]-hydantoin[g] | 0 | + | 230 | 4820/5 | 0 | - | 16/X,60 |
| 20) 5-nitro-2-furoic acid[a] | 0 | + | 0.26 | 420/250 | 0 | - | 1/X,60,1 |
| 21-34) 14 nitrofurans | + | + | | | 0 | - | 61/60 |
| **F) Misc. Aliphatics and Aromatics** | | | | | | | |
| 1) cyclohexane[e] | ?0 | 0 | <0.006 | <70/1000 | 0957 | A | 16/X |
| 2) acetone[m] | 0 | 0 | <0.0004 | <70/$10^4$ | 0957 | A | 16/X |
| 3) acetic acid[e] | 0 | 0 | <0.004 | <70/1000 | 0957 | A | 16/X |
| 4) ethyl alcohol[h] | 0 | 0 | <0.0003 | <70/$10^4$ | 09 | A | 16/X |
| 5) n-butyl alcohol[m] | ?0 | 0 | <0.0005 | <70/$10^4$ | 0957 | A | 16/X |
| 6) ethylene glycol[t] | 0 | 0 | <0.0004 | <70/$10^4$ | 0957 | A | 16/X |
| 7) ethionine[c] | + | 0 | <0.02 | <70/500 | 0957 | A | 16/X,k,23 |
| 8) ethylenediaminetetraacetic acid, disodium [EDTA][t] | ?0 | 0 | <0.002 | <70/$10^4$ | 0957 | A | 16/X |
| 9) dimethyl sulfoxide [DMSO][z] | 0 | 0 | <0.00001 | <70/5x$10^5$ | 0957 | A | 16/X |
| 10) benzoic acid[a] | ?0 | 0 | <0.009 | <70/1000 | 0957 | A | 16/X |
| 11) bromobenzene[a] | ? | 0 | <0.01 | <70/750 | 0957 | A | /X |
| 12) salicylic acid[a] | ?0 | 0 | <0.02 | <70/500 | 0957 | A | 16/X |
| 13) resorcinol[g] | ?0 | 0 | <0.008 | <70/1000 | 0957 | A | 16/X, 13 |
| 14) trans-stilbene[d] | ?0 | ? | <0.13 | <70/100 | 0957 | A | 16/X |
| 15) safrole[w] | + | 0 | <0.01 | <70/1000 | 0957 | A | 17/X |
| 16) 1'-hydroxysafrole[w] | + | 0 | <0.006 | <70/2000 | 0957 | A | 33/X |
| 17) 1'-acetoxysafrole[w] | + | + | 2.4 | 556/50 | 0 | - | 33/4 |
| 18) 1-naphthylisothiocyanate[a] | 0 | ? | <0.65 | <70/20 | 0957 | A | 16/X |
| 19) diethylstilbesterol[g] | + | ? | <0.38 | <70/50 | 0957 | A | 17/X |
| 20) 12-O-tetradecanoylphorbol-13-acetate[s] | + | 0 | <0.09 | <70/500 | 0957 | A | 16/X |
| **G) Nitrosamines, etc.** | | | | | | | |
| 1) dimethylnitrosamine | + | w+ | 0.02[†] | 1100/4440 | 03 | + | 17/21,43,44 |
| 2) diethylnitrosamine | + | w+ | 0.01[†] | 380/4080 | 03 | + | 17/21,45,44 |
| 3) di-n-propylnitrosamine | + | w+ | 0.08 | 395/650 | 3 | + | 46/47,21 |
| 4) di-n-butylnitrosamine | + | + | 0.15 | 384/395 | 3 | + | 46/47,21 |
| 5) di-n-pentylnitrosamine | + | w+ | 0.05 | 115/465 | 3 | + | 46/47,21 |
| 6) N-nitrosopyrrolidine | + | w+ | 0.02 | 180/1000 | 3 | + | 16/47,51 |
| 7) N-nitrosomorpholine | + | w+ | 0.06 | 300/580 | 3 | + | 16/47,51 |
| 8) N-nitrosopiperidine | + | w+ | 0.01 | 466/5000 | 053 | + | 16/X,47 |
| 9) dibenzylnitrosamine[x] | 0 | ? | <0.03 | <70/500 | 0957 | A | 16/X |
| 10) diphenylnitrosamine[e] | 0 | ? | <0.03 | <70/500 | 0957 | A | 16/X |
| 11) N-methyl-N'-nitro-N-nitroso-guanidine[a] | + | + | 1375* | 18700/2 | 05 | - | 17/4,3 |
| 12) N-ethyl- " | + | + | 350 | 35000/16 | 5 | - | 48,16/a |
| 13) N-propyl- " | + | + | 40 | 4000/18 | 5 | - | 48/a |
| 14) N-butyl- " | + | + | 49 | 5000/19 | 5 | - | a/a |
| 15) N-isobutyl- " | + | + | 77 | 7700/19 | 5 | - | a/a |
| 16) N-pentyl- " | + | + | 22 | 2200/20 | 5 | - | a/a |
| 17) N-hexyl- " | ? | + | 5.3 | 532/22 | 5 | - | /a |
| 18) N-nitrosomethylurea | + | + | 4.4 | 6000/140 | 50 | - | 17/j,h,23 |
| 19) N-nitrosoethylurea | + | + | 1.1 | 932/100 | 5 | - | 17/n,k |
| 20) N-nitroso-N-methylurethane | + | + | | | | | 17/1 |
| 21) cycasin[a] | + | 0 | <0.04 | <70/500 | 0957 | A | 17/X,49 |
| 22) methylazoxymethanol | + | + | | | 4 | - | 17/49 |
| 23) methylazoxymethanol acetate ester | + | + | | | 5 | - | 17/k,n |
| 24) streptozotocin | + | + | 1949 | 4265/1 | 05 | - | 17/X,50 |
| **H) Fungal Toxins & Antibiotics** | | | | | | | |
| 1) aflatoxin $B_1$[c] | + | + | 7057 | 2260/0.1 | 098 | A | 17/4,2 |
| 2) aflatoxin $B_2$[c] | w+ | + | 2.1 | 664/100 | 09 | A | 17/X,p |
| 3) aflatoxicol | + | + | 2200 | 346/.05 | 9 | P | 19/p |
| 4) aflatoxin $M_1$ | w+ | + | 112 | 274/.8 | 9 | P | 17/p |
| 5) aflatoxin $G_1$ | + | + | 116 | 142/.4 | 9 | P | 17/p |
| 6) aflatoxin $P_1$ | 0 | | <0.38[p] | <20/16 | 9 | P | p/p |
| 7) aflatoxin $G_2$ | ?0 | | <0.43[p] | <20/16 | 9 | P | 16/p |
| 8) sterigmatocystin[a] | + | + | 915 | 282/.1 | 098 | A | 17/4,2 |
| 9) gibberellic acid[a] | 0 | 0 | <0.005 | <70/5000 | 0957 | A | 16/X |
| 10) penicillin G, potassium[g] | ?0 | 0 | <0.01 | <70/3000 | 0957 | A | 16/X |
| 11) adriamycin-HCl[g] | + | + | 108 | 3740/20 | 908 | - | 16/X,f,m |
| 12) daunorubicin-HCl[g] | + | + | 356 | 3790/6 | 908 | - | 16/X |
| 13) tetracycline[g] | ?0 | ? | <31 | <70/1 | 0957 | A | 16/X |
| 14) mitomycin C[g] | + | ? | <23 | <70/1 | 0957 | - | 16/X,4 |
| **I) Mixtures** | | | | | | | |
| 1) cigarette smoke condensate[h] | + | + | 18200/cigarette | | 8 | A | 16/12 |
| **J) Miscellaneous Heterocycles** | | | | | | | |
| 1) phenobarbital, sodium[m] | w+ | 0 | <0.004 | <70/5000 | 0957 | A | 16/X |
| 2) 3,3-dimethyl-1-phenyl-3-pyrazolin-5-one [antipyrene][a] | ?0 | 0 | <0.01 | <70/3000 | 0957 | A | 16/X,n |
| 3) 4-amino-2,3-dimethyl-1-phenyl-3-pyrazolin-5-one [4-aminoantipyrene][g] | ?+ | 0 | <0.001 | <70/$10^4$ | 0957 | A | 16/X,n |
| 4) 4-butyl-1,2-diphenyl-3,5-pyrazolidine-dione [phenylbutazone][g] | ?0 | 0 | <0.01 | <70/2000 | 0957 | A | 16/X,m |
| 5) 3-amino-1H-1,2,4-triazole [amitrole][a] | + | 0 | <0.001 | <70/5000 | 0957 | A | 17/X |
| 6) 5-iododeoxyuridine[a] | c0 | 0 | <0.01 | <70/2000 | 0957 | A | 16,65/X |
| 7) 4,4'-propylenedi-2,6-piperazine-dione [ICRF 159][o] | ?0[T] | 0 | <0.04 | <70/500 | 5097 | A | 25/X,e |
| 8) nicotine[g] | 0 | 0 | <0.001 | <70/$10^4$ | 0957 | A | 16/X |
| 9) caffeine[c] | ?0[T] | 0 | <0.002 | <70/6000 | 0957 | A | 16/X |
| 10) atropine sulfate[a] | ?0 | 0 | <0.01 | <70/5000 | 0957 | A | 16/X |
| 11) hycanthone methanesulfonate[f] | + | + | 2.5 | 346/50 | 8 | - | 26/X,27 |
| **K) Miscellaneous Nitrogen compounds** | | | | | | | |
| 1) propyleneimine[f] | + | + | 2.0 | 5230/150 | 5 | - | 28/X,k,n |
| 2) ethyleneimine[y] | + | + | 2.0 | 469/10 | 50 | - | 16/X |
| 3) tris(1-aziridinyl)phosphine sulfide [thio-TEPA][f] | + | + | | | 50 | - | 16/f |
| 4) hydrazine sulfate[f] | + | w+ | | | 3 | | 16/1 |
| 5) 1,2-dimethylhydrazine[a] | + | 0 | <0.0008 | <70/5000 | 0957 | A | 17,32/X |
| 6) N-(2-hydroxyethyl)hydrazine[a] | + | w+ | 0.01 | 134/1100 | 50 | - | 16/X,30 |
| 7) natulan [procarbazine][f] | + | 0 | <0.0015 | <70/$10^4$ | 0957 | A | 16/X |
| 8) maleic hydrazide[a] | c0[T] | 0 | <0.0008 | <70/$10^4$ | 0957 | A | 16/X,24 |
| 9) 1-phenyl-3,3-dimethyltriazene | + | + | † | | 4 | P- | 16/g |
| 10) 1-(4-chlorophenyl)-3,3-dimethyltriazene | + | + | † | | 4 | P | 67/g |
| 11) sodium azide[t] | 0[T] | + | 150 | 2240/1 | 05 | - | 16/X,31 |
| 12) hydroxylamine hydrochloride[e] | 0 | 0 | <0.001 | <70/5000 | 0957 | A | 29/X |
| 13) sodium nitrite[b] | 0 | w+ | 0.01 | 975/9000 | 50 | - | 16/X,1 |
| **L) Azo dyes and diazo compounds** | | | | | | | |
| 1) azobenzene[a] | ?[T] | + | 1.4 | 379/50 | 0 | A | 17/X |
| 2) 4-aminoazobenzene[s] | w+ | + | 0.29 | 147/100 | 098 | A | 15/X,2 |
| 3) o-aminoazotoluene[s] | + | + | 15 | 1340/20 | 098 | A | 15/X,2 |
| 4) 3-methoxy-4-aminoazobenzene | + | + | 747* | 16454/5 | 89 | A | 15/X,k,n |
| 5) N-methyl-4-aminoazobenzene | + | + | 0.14[†] | 140/200 | 09 | A | 15/52 |
| 6) N,N-dimethyl-4-aminoazobenzene | + | + | 0.12[†] | 120/210 | 90 | A | 17/52 |
| 7) 3'-methyl-4-dimethylamino-azobenzene[s] | + | + | 0.34 | 71/50 | 8 | P | 15/2 |
| 8) N,N-diethyl-4-aminoazobenzene[v] | 0 | 0 | <0.03 | <70/500 | 09 | A | 15/X |
| 9) N-hydroxy-4-aminoazobenzene | ?0 | + | 0.35[†] | 350/210 | 90 | A | 17/52 |
| 10) 2-methyl-4-dimethylamino-azobenzene | w+ | + | 0.60[†] | 300/120 | 90 | A | 15/52 |
| 11) N-benzoyloxy-4-methylamino-azobenzene | + | + | † | | 90 | - | 16/52 |
| 12) methyl orange[a] | 0 | 0 | <0.01 | <70/2500 | 0957 | A | 15/X |
| 13) p-dimethylaminobenzenediazo sodium sulfonate [dexon][p] | ? | + | 1.8 | 719/100 | 0957 | - | 15/X,53 |
| 14) azaserine[f] | + | + | 12000 | 14000/.2 | 0 | - | 55/X,55 |
| 15) diazoacetylglycine amide | + | + | 28 | 2000/10 | 5 | - | 16/54 |
| 16) diazoacetylglycine hydrazide | + | + | 7.9 | 500/10 | 5 | - | 16/54 |
| 17) diazoacetylglycine ethyl ester | ? | + | 17 | 1000/10 | 5 | - | /54 |

Legend to Table 1 (*on preceding pages*).

Explanations and footnotes are given below for each column in the Table.

*Chemicals.* The division of chemicals into groups was, of necessity, somewhat arbitrary. Commercial chemicals were the purest grades available. Sources of chemicals are indicated by superscripts, and are as follows: [a]Aldrich, [b]Baker, [c]Calbiochem, [d]BDH, [e]Eastman, [f]Fisher, [g]Sigma, [h]Commercial Solvents, [i]National Cancer Institute, [j]Allied Chemical, [k]K & K, [l]Eli Lilly, [m]Mallinckrodt, [n]NBC, [o]gift of M. Coombs, [p]Analabs, [q]gift of P. Sims and P. Grover, [r]gift of J. Flesher, [s]Schuchardt, [t]MC/B, [u]gift of R. Peck and H. Creech, [v]gift of V. Simmon, [w]gift of J. A. and E. Miller, [x]gift of W. Lijinsky, [y]Dow, [z]Schwarz/Mann, [aa]gift of G. L. Laqueur, [ab]gift of T. Sugimura, [ac]gift of E. Bueding, [ad]Union Carbide, [ae]Mead Johnson, [af]gift of P. Hartman, [ag]gift of N. Bachur, [ah]gift of R. Kouri, [ai]gift of W. Benedict, [aj]gift of D. E. V. Wilman, [ak]gift of H. Bartsch, [al]gift of J. Casida, [am]gift of Salsbury Laboratories.

*Car.* Carcinogenicity. Classification, especially non-carcinogenicity, is usually difficult because of the varying completeness and modes of treatment in many studies and the statistical limitations inherent in animal tests. + = carcinogen; 0 = non-carcinogen; w+ = weak carcinogen (occasionally used for comparison to potent relative; in general, we have not evaluated carcinogenic potency, an inherently complex area); ?+ or ?0 = carcinogen, or non-carcinogen, in limited studies, further confirmatory work required; c0 = non-carcinogen in most studies, with some reports of weak or marginal activity; ? = inadequate data available for classification as a carcinogen or non-carcinogen. [T] = under test [chemicals **B5** and **B7**, personal communication B. McKusick (du Pont)]; $\pi$ = promoter.

*Mut.* Mutagenicity. The standard *Salmonella* plate test (7) was used except where indicated. + = mutagenic; 0 = non-mutagenic; w+ = weakly mutagenic (<0.10 revertants/nmol); ? = chemical toxicity prevented adequate mutagenicity test. Data is presented when a complete dose-response curve (almost always linear; * in *Revertants per nmol* column = non-linear) was obtained. In cases where a spot (7) or well test (61) was used, or a complete dose-response curve was not available, and the mutagenicity was clear, the compound is reported mutagenic but quantitative data are not given. † in *Revertants* column = liquid test (7, 21, 44), a modification where the bacteria are preincubated with the S-9 Mix before pouring plates: chemicals **K10, L5, L9, L11,** not tested in standard plate test; chemicals **L6, L10, K9, G1** (47), **G2** (47) negative in standard plate test; chemicals **G3, G4, G5** less active in liquid test (21, 47); chemical **L6,** NADH and ATP added to S-9 Mix. = = analyzed for impurities by high pressure liquid chromatography: weak activity of chemical **A7** all due to 0.25% **A3** impurity; of chemical **A22** not all due to trace of **A21;** of chemical **A28** not all due to trace of **A26;** of chemical **A20** not due to several impurities; chemical **A10,** insufficient material available for chromatography (if 0.1% **A3** impurity present, it could account for very weak activity: see chemical **A7**); of chemical **A11** not due to several impurities; chemical **A12** data given is for purified material [direct mutagenic activity previously reported (3) due to impurities] (V. Donahue, J. McCann, and B. N. Ames, in preparation).

*Revertants per plate.* Numbers given are revertant colonies on a petri plate per micrograms of chemical incorporated in the plate, e.g., 1630/10 = 1630 revertant colonies per 10 $\mu$g tested. For mutagenic compounds the numbers are single points from linear dose-response curves. Spontaneous revertant colonies have been subtracted: usually 20 (tester strain TA1535), 7 (TA1537), 25 (TA1538), 140 (TA100), or 40 (TA98). For non-mutagenic compounds, a less-than figure is used to indicate the result: the denominator is the maximum dose tested—each non-mutagenic compound was tested over a wide dose range, usually 10, 100, and 500 or 1000 $\mu$g (often higher for TA100) for non-toxic compounds, and up to the maximum allowable concentration if the compound was inhibitory; the numerator is 1/2 the spontaneous reversion rate of the strain for which results are reported (usually <70, for TA100). Each non-mutagenic compound was tested both with and without S-9 (usually Aroclor-induced microsomes were used) on at least strains TA1535, TA1537, TA98, and TA100. A compound was called non-mutagenic (0) if there was no dose-response. G = the chemical is a gas or very volatile liquid, the test was done in a dessicator, and quantitation is difficult.

*Revertants per nmol.* We have calculated the number of revertants per nmol from the revertants per plate and the molecular weight. The value represents the mutagenic potency of the particular chemical in reverting the strain indicated in the standard assay. The *Salmonella*/microsome test is a back mutation test using tester strains containing different types of mutations. The relationship of the mutagenic potency of a mutagen on a particular strain to the overall mutagenic potential of the compound for DNA in general, and to carcinogenic potency, remains to be determined. In addition the standard assay represents a compromise between various factors such as amount of S-9, induction procedure, etc. Thus, comparisons between potency of different chemicals must be undertaken with caution. This important and complex subject will be discussed further. * = non-linear dose-response. † = liquid test (see *Mut.*).

*Strain.* The standard tester strains (3, 4, 7) are abbreviated by a single digit as follows: 0(TA100), 5(TA1535), 7(TA1537), and 9(TA98); 8(TA1538) contains the same histidine mutation as TA98, which has replaced it as a more sensitive indicator for some mutagens; 3(TA1530) and 4(*hisG46*) are old tester strains and are the same as TA1535 except they lack the *uvrB* or deep rough mutations (3). The first digit of the series means that the data come from the test with that strain: each other digit, in the case of a mutagen, means that the chemical is also mutagenic on that strain or, in the case of a non-mutagen, that it has been tested on that strain. Chemicals have been designated non-mutagenic only if they have been tested on at least strains 0, 9, 5, and 7, the complete set of strains recommended for general mutagenesis testing (7).

*S-9.* The S-9 fraction (9000 × *g* supernatant of rat liver) was added (A or P or +) or not (−) as indicated. Liver was from rats induced with Aroclor 1254 (A) (12, 7) or phenobarbital (P) (2, 7). (+) = S-9 added, induction procedure not A or P, or unspecified (see individual reference). P- or A- indicates that the data were from an experiment with S-9, but that the compound was somewhat active without S-9.

*Ref.* References are given for carcinogenicity/mutagenicity data, respectively. We have mainly cited three compendia for carcinogenicity (15–17), which contain the primary source references, rather than citing the large number of primary sources for carcinogenicity or non-carcinogenicity. Where individual papers have been cited no attempt has been made to determine priority. For mutagenicity the first reference in the series refers to the source of the data quoted (X = this study) and others to previous reports, or independent observations. All non-mutagens reported here have been tested both with and without S-9 on the new tester strains TA100 and TA98 and over a wide dose-range on all strains: as a general rule we have not cited negative mutagenicity studies unless they fulfill these criteria. We have also not cited mutagenicity studies in other organisms, some of which predate results in the *Salmonella* test. Personal communication citations are as follows: a = T. Matsushima, M. Nagao, and T. Sugimura; b = P. Sims; d = R. Peck; e = M. Coombs; f = W. Benedict; g = H. Bartsch and C. Malaveille; h = A. E. Auletta; i = C. Y. Wang, K. Muraoka, and G. T. Bryan; j = K. Lum and J. Richards; k = V. Simmon; m = M. Legator; n = H. Rosenkranz; o = P. Harris; p = J. Wong and D. Hsieh (chemicals **H6** and **H7** not tested at higher doses); q = J. W. Flesher and E. Swim; r = B. McKusick (du Pont) negative (see ref. 34 for +); s = I. Mattern.


1. Ames, B. N. (1971) in *Chemical Mutagens: Principles and Methods for their Detection,* ed. Hollaender, A. (Plenum Press, New York), Vol. 1, pp. 267–282.
2. Ames, B. N., Durston, W. E., Yamasaki, E. & Lee, F. D. (1973) *Proc. Nat. Acad. Sci. USA* **70,** 2281–2285.
3. Ames, B. N., Lee, F. D. & Durston, W. E. (1973) *Proc. Nat. Acad. Sci. USA* **70,** 782–786.
4. McCann, J., Spingarn, N., Kobori, J. & Ames, B. N. (1975) *Proc. Nat. Acad. Sci. USA* **72,** 979–983.
5. Miller, E. C. & Miller, J. A. (1971) in *Chemical Mutagens: Principles and Methods for their Detection,* ed. Hollaender, A. (Plenum Press, New York), Vol. 1, pp. 83–119.
6. McCann, J. & Ames, B. N. (1975) *Ann. N.Y. Acad. Sci.,* in press.
7. Ames, B. N., McCann, J. & Yamasaki, E. (1975) *Mutat. Res.,* in press.
8. McCann, J., Simmon, V., Streitwieser, D. & Ames, B. N. (1975) *Proc. Nat. Acad. Sci. USA* **72,** 3190–3193.

9. Ames, B. N., Gurney, E. G., Miller, J. A. & Bartsch, H. (1972) *Proc. Nat. Acad. Sci. USA* **69**, 3128–3132.
10. Ames, B. N., Sims, P. & Grover, P. L. (1972) *Science* **176**, 47–49.
11. Durston, W. E. & Ames, B. N. (1974) *Proc. Nat. Acad. Sci. USA* **71**, 737–741.
12. Kier, L. D., Yamasaki, E. & Ames, B. N. (1974) *Proc. Nat. Acad. Sci. USA* **71**, 4159–4163.
13. Ames, B. N., Kammen, H. O. & Yamasaki, E. (1975) *Proc. Nat. Acad. Sci. USA* **72**, 2423–2427.
14. Nishioka, H. (1975) *Mutat. Res.* **31**, 185–189.
15. Arcos, J. C. & Argus, M. F. (1974) *Chemical Induction of Cancer* (Academic Press, New York), Vol. II (A&B).
16. *Survey of Compounds which have been Tested for Carcinogenic Activity*, USPHS Publication no. 149; Washington, D.C. (through 1972–73 volume).
17. *IARC Monograph on the evaluation of carcinogenic risk of chemicals to man* (1972–1975) (IARC, Lyon), Vols. 1–9.
18. Lambelin, G., Roba, J., Roncucci, R. & Parmentier, R. (1975) *Eur. J. Cancer* **11**, 327–334.
19. Schoenhard, G. L. (1974) Ph.D. Dissertation, Oregon State University.
20. Holland, V. R., Saunders, B. C., Rose, F. L. & Walpole, A. L. (1974) *Tetrahedron* **30**, 3299–3302.
21. Sugimura, T., Yahagi, T., Nagao, M., Takeuchi, M., Kawachi, T., Hara, K., Yamasaki, E., Matsushima, T., Hashimoto, Y. & Okada, M. (1975) in *Screening Tests in Chemical Carcinogenesis*, eds. Montesano, R., Bartsch, H. & Tomatis, L. (IARC, Lyon), in press.
22. Bartsch, H., Malaveille, C. & Montesano, R. (1975) *Int. J. Cancer* **15**, 429–437.
23. Odashima, S. (1975) in *Screening Tests in Chemical Carcinogenesis*, eds. Montesano, R., Bartsch, H. & Tomatis, L. (IARC, Lyon), in press.
24. Rashid, K. A. (1974) Masters Thesis, Pennsylvania State University.
25. Stoner, G. D., Shimkin, M. B., Kniazeff, A. J., Weisburger, J. H., Weisburger, E. K. & Gori, G. B. (1973) *Cancer Res.* **33**, 3069–3085.
26. Haese, W. H., Smith, D. L. & Bueding, E. (1973) *J. Pharmacol. Exp. Ther.* **186**, 430–440.
27. Hartman, P. E., Levine, K., Hartman, Z. & Berger, H., (1971) *Science* **172**, 1058–1060.
28. Ulland, B., Finkelstein, M., Weisburger, E. K., Rice, J. M. & Weisburger, J. H. (1971) *Nature* **230**, 460–461.
29. Yamamoto, R. S., Weisburger, E. K. & Korzis, J. (1967) *Proc. Soc. Exp. Biol. Med.* **124**, 1217–1220.
30. Shirasu, Y., Moriya, M., Kato, K. & Kada, T. (1975) *Mutat. Res.* **31**, 268–269.
31. Nilan, R. A., Sideris, E. G., Kleinhofs, A., Sander, C. & Konzak, C. F. (1973) *Mutat. Res.* **17**, 142–144.
32. Reddy, B. S., Narisawa, T., Wright, P., Vukusich, D., Weisburger, J. H. & Wynder, E. L. (1975) *Cancer Res.* **35**, 287–290.
33. Borchert, P., Miller, J. A., Miller, E. C. & Shires, T. K. (1973) *Cancer Res.* **33**, 590–600.
34. Bartsch, H., Malaveille, C., Montesano, R. & Tomatis, L. (1975) *Nature* **255**, 641–643.
35. Powers, M. B., Voelker, R. W., Page, N. P., Weisburger, E. K. & Kraybill, H. F. (1975) *14th Ann. Mtg. Soc. of Toxicol.*, paper no. 123, p. 99.
36. Rannug, U., Johansson, A., Ramel, C. & Wachmeister, C. A. (1974) *Ambio* **3**, 194–197.
37. Van Duuren, B. L., Goldschmidt, B. M., Katz, C., Seidman, I. & Paul, J. S. (1974) *J. Nat. Cancer Inst.* **53**, 695–700.
38. Khachatryan, E. A. (1972) *Probl. Oncol.* **18**, 85–86.
39. Mukai, F. H. & Hawryluk, I. (1973) *Mutat. Res.* **21**, 228.
40. Weisburger, J. H., Griswold, D., Jr., Prejean, J. D., Casey, A., Wood, H., Jr. & Weisburger, E. K. (1975) *Recent Results in Cancer Res.* **52**, 1–17.
41. Ficsor, G. & Nii, G. M. (1970) *Environ. Mutagen Soc. Newslett.* **3**, 38.
42. Shirasu, Y., Moriya, M., Kato, K., Furuhashi, A. & Kada, T. (1974) *Mutat. Res.* **26**, 438.
43. Malling, H. V. (1971) *Mutat. Res.* **13**, 425–429.
44. Bartsch, H., Malaveille, C. & Montesano, R. (1975) *Cancer Res.* **35**, 644–651.
45. Malling, H. V. & Frantz, C. N. (1973) *Environ. Health Perspect.* **6**, 71–82.
46. Magee, P. N. & Barnes, J. M. (1967) *Adv. Cancer Res.* **10**, 163–246.
47. Bartsch, H., Malaveille, C. & Montesano, R. (1975) in *Screening Tests in Chemical Carcinogenesis*, eds. Montesano, R., Bartsch, H. & Tomatis, L. (IARC, Lyon), in press.
48. Kogure, K., Kawachi, T., Tokunaga, A., Kazama, S., Shimosato, Y., Sugimura, T., Takayama, S., Kuwabara, N., Murakami, T. & Nakadate, M. (1974) *Abs. XIth Int. Cancer Congr.* pp. 671–672.
49. Smith, D. W. E. (1966) *Science* **152**, 1273–1274.
50. Gabridge, M. G., Denunzio, A. & Legator, M. S. (1969) *Nature* **221**, 68–70.
51. Gomez, R. F., Johnston, M. & Sinskey, A. J. (1974) *Mutat. Res.* **24**, 5–7.
52. Yahagi, T., Degawa, M., Seino, Y., Matsushima, T., Nagao, M., Sugimura, T. & Hashimoto, Y. (1975) *Cancer Lett.*, in press.
53. Kada, T., Moriya, M. & Shirasu, Y. (1974) *Mutat. Res.* **26**, 243–248.
54. Monti-Bragadin, C., Tamaro, M. & Banfi, E. (1974) *Antimicrob. Agents Chemother.* **6**, 655–656.
55. Longnecker, D. S. & Curphey, T. J. (1975) *Cancer Res.* **35**, 2249–2258.
56. Teranishi, K., Hamada, K. & Watanabe, H. (1975) *Mutat. Res.* **31**, 97–102.
57. Grover, P. L., Sims, P., Mitchley, B. C. V. & Roe, F. J. C. (1975) *Br. J. Cancer* **31**, 182–188.
58. Milvy, P. & Garro, A. J. (1975) *Mutation Res.*, in press.
59. Coombs, M. M., Bhatt, T. S. & Croft, C. J. (1973) *Cancer Res.* **33**, 832–837.
60. Yahagi, T., Matsushima, T., Nagao, M., Seino, T., Sugimura, T. & Bryan, G. T. (1975) *Mutat. Res.*, in press.
61. Yahagi, T., Nagao, M., Hara, K., Matsushima, T., Sugimura, T. & Bryan, G. T. (1974) *Cancer Res.* **34**, 2266–2273.
62. Yahagi, T., Shimizu, H., Nagao, M., Takemura, N. & Sugimura, T. (1975) *Gann*, in press.
63. Legator, M. S., Connor, T. H. & Stoeckel, M. (1975) *Science* **188**, 1118–1119.
64. Cohen, S. M. & Bryan, G. T. (1973) *Fed. Proc.* **32**, 825.
65. Veselý, J. & Čihák, A. (1973) *Experientia* **29**, 1132–1133.
66. Takemura, N., Hashida, C. & Terasawa, M. (1974) *Br. J. Cancer* **30**, 481–483.
67. Preussman, R., Ivankovic, S., Landschütz, G. J., Flohr, E. & Griesbach, U. (1974) *Z. Krebsforsch.* **81**, 285–310.
68. Matthews, C., Davidson, J., Bauer, E., Morrison, J. & Richardson, A. (1956) *J. Am. Pharm. Assoc.* **45**, 260–267.
69. Harris, P., Gibson, W. & Dillard, R. (1971) *Proc. Am. Assoc. Cancer Res.* **12**, 26.
70. Thiede, T. & Christensen, B. Chr. (1969) *Acta Med. Scand.* **185**, 133–137.
71. Voogd, C. E., Van der Stel, J. J., & Jacobs, J.J.J.A.A. (1974) *Mutation Res.* **26**, 483–490.
72. Urman, J. K., Bulay, O., Clayson, D. P., & Shubik, P. (1975) *Cancer Letters*, in press.