CARCINOGENICITY OF N-NITROSAMINES RELATED TO N-BUTYL-N-(4-
HYDROXYBUTYL)NITROSAMINE AND N,N-DIBUTYLNITROSAMINE IN ACI/N
RATS*1

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Carcinogenic effect of 14 N-nitrosamines related to N-butyl-N-(4-hydroxy-
butyl)nitrosamine (BBN) and N,N-dibutylnitrosamine (DBN) was studied in
ACI/N male rats by administration in the drinking water. BBN homologs having
methyl, ethyl, or pentyl group selectively induced urinary bladder tumors, but a
homolog with tert-butyl group did not have any carcinogenic effect. N-Ethyl-N-
(3-carboxypropyl)nitrosamine, the principal urinary metabolite of the ethyl
homolog of BBN, did also induce bladder tumors selectively, thus providing an
additional evidence that N-alkyl-N-(3-carboxypropyl)nitrosamines are responsi-
ble for the selective induction of bladder tumors by BBN homologs. N-Butyl-
N-(carboxymethyl)nitrosamine and BBN analogs having 3-hydroxypropyl chain
together with ethyl or butyl group were found to be noncarcinogenic. N-Propyl-
N-butylnitrosamine and DBN induced hepatomas, but simultaneous develop-
ment of esophageal tumors was observed only with the former. N-Butyl-N-(3-
hydroxybutyl)nitrosamine, one of the principal metabolites of DBN, did not
induce any tumors, but its further transformation product, N-butyl-N-(3-
oxobutyl)nitrosamine as well as N-butyl-N-(2-oxobutyl)nitrosamine, another
metabolic intermediate of DBN, induced hepatomas. Possible correlation of
structure and metabolism with organotropic carcinogenesis by N,N-dialkylnitro-
samines is discussed, with special reference to selective induction of urinary
bladder tumors.

In the course of our studies9~11,14~16,18) on
a possible correlation of structure and metab-
ilism with organotropic carcinogenesis by
N,N-dialkylnitrosamines with special refer-
ce to induction of urinary bladder tumors
in rats, carcinogenic effect of a number of
N-nitrosamines structurally related to N-
butyl-N-(4-hydroxybutyl)nitrosamine (BBN)
and N,N-dibutylnitrosamine (DBN) was inves-
tigated in ACI/N rats after oral admin-
istration. The present paper describes the
results of the carcinogenicity test carried out
with 14 kinds of N-nitrosamines listed in
Table I except N-butyl-N-(3-carboxypropyl)-
nitrosamine (BCPN), the carcinogenicity of
which was reported earlier.10) They are divid-
ed into the following 4 groups.
(A) BBN Homologs: N-Methyl-N-(4-hy-
droxybutyl)nitrosamine (MHBN), N-ethyl-
N-(4-hydroxybutyl)nitrosamine (EHBN), BB-
N, N-pentyl-N-(4-hydroxybutyl)nitrosamine
(AHBN) (pentyl = amyl), N-tert-butyl-N-(4-
hydroxybutyl)nitrosamine (t-BBN)
(B) BCPN Homolog and Analog: N-

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Table I. Structure and Abbreviation of N-Nitroso Compounds Tested

<table>
<thead>
<tr>
<th>Group</th>
<th>Compound</th>
<th>R</th>
<th>Abbreviation used</th>
</tr>
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<tbody>
<tr>
<td>A</td>
<td>ON-N</td>
<td>CH₂CH₂CH₂CH₂OH</td>
<td>R</td>
</tr>
<tr>
<td></td>
<td>CH₂CH₃</td>
<td>CH₃CH₂CH₂CH₂CH₃</td>
<td></td>
</tr>
<tr>
<td></td>
<td>CH₂CH₃CH₂CH₂CH₃</td>
<td></td>
<td>BBN</td>
</tr>
<tr>
<td></td>
<td>CH₂CH₃CH₂CH₂CH₃</td>
<td></td>
<td>AHBN</td>
</tr>
<tr>
<td></td>
<td>C(CH₂)₃</td>
<td>i-BBN</td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>ON-N</td>
<td>CH₂CH₂CH₂COOH</td>
<td>R</td>
</tr>
<tr>
<td></td>
<td>CH₂CH₂CH₂CH₃</td>
<td></td>
<td>BCPN</td>
</tr>
<tr>
<td>C</td>
<td>ON-N</td>
<td>CH₂COOH</td>
<td>R</td>
</tr>
<tr>
<td></td>
<td>CH₂CH₂CH₂CH₃</td>
<td></td>
<td></td>
</tr>
<tr>
<td>D</td>
<td>ON-N</td>
<td>CH₂CH₂CH₃CH₃</td>
<td>R</td>
</tr>
<tr>
<td></td>
<td>CH₂CH₂CH₃CH₃</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Ethyl-N-(3-carboxypropyl)nitrosamine (EC-PN), N-butyl-N-(carboxymethyl)nitrosamine (BCMN)

(C) BBN Analogs with 3-Hydroxypropyl
Group: N-Ethyl-N-(3-hydroxypropyl)nitrosamine (EHPN), N-butyl-N-(3-hydroxypropyl)nitrosamine (BHPN)

(D) DBN Homologs and Its Metabolites:
N-Propyl-N-butylnitrosamine (PBN), DBN, N-butyl-N-(3-hydroxybutyl)nitrosamine (BHBN-3), N-butyl-N-(3-oxobutyl)nitrosamine (BOBN-3), N-butyl-N-(2-oxobutyl)nitrosamine (BOBN-2)

Materials and Methods

Chemicals All the N-nitrosamines used in the present experiment were prepared in our laboratory or by Izumi Chemical Laboratory, Yokohama, according to our direction*4; MHBN, bp 143~144°/4 Torr; EHBN, bp 139~140°/4 Torr; BBN, bp 165~167°/5 Torr; AHBN, bp 164~167°/5 Torr; t-BBN, bp 130~131°/3 Torr; ECPN, mp <25°; BCMN, mp 61°; EHPN, bp 129~130°/4 Torr; BHPN, bp 144~146°/3 Torr; PBN, bp 74~76°/4 Torr; DBN, bp 114~115°/11 Torr; BHBN-3, bp 143~146°/5 Torr; BOBN-3, bp 123°/3 Torr; BOBN-2, bp 102~103°/0.15 Torr. The purity of these compounds was checked by thin-layer chromatography, and from infrared, nuclear magnetic resonance, and mass spectra.

Animals and Treatment Eight-week-old inbred ACI/N male rats obtained from Fuji Animal Farm, Tokyo, were kept until they were 10 weeks old. Five rats in each group were housed in one cage and they were maintained on the cube diet CE-2 (CLEA Japan Inc., Tokyo) throughout the experiment. The compounds dissolved in distilled water in a concentration of 2.87mM corresponding to the 0.05% solution of BBN as the control.10) Animals received the solution daily in the dose of 20 ml/rat as drinking water for 20 weeks and subsequently tap water except one group receiving BHPN. Usually some of the rats were sacrificed at 20 weeks and all the remaining animals at 30 weeks. When a group of rats developed large tumors during or at 20 weeks, however, all the rats in that group were killed at that time.

Histology Histological specimens were prepared in the same way as reported earlier.11)

Assay for Liver Function Activities of glutamic oxaloacetic transaminase (GOT) (EC 2.6.1.1), glutamic pyruvic transaminase (GPT) (EC 2.6.1.2.), and alkaline phosphatase (EC 3.1.3.1) in serum of rats receiving the compounds were assayed periodically as reported previously.13)

*4 The synthesis of these compounds will be reported elsewhere together with their metabolic studies.
RESULTS

Body Weight Changes in body weight of rats treated with the test compounds are shown in Fig. 1. Rats receiving compounds other than PBN gained body weight to the normal level.

Liver Function Activities of serum enzymes associated with the liver function were assayed with 5 compounds; EHBN, BBN, AHBN, BCMN, and PBN. As illustrated in Fig. 2, the compounds except PBN did not affect the enzyme activities throughout the exposure period. Significantly higher enzyme levels were observed only with PBN.

Carcinogenicity (Table II) (A) BBN Homologs: MHBN, EHBN, BBN (as a positive control), and AHBN induced urinary bladder tumors but no tumors in other sites. Especially EHBN produced large tumors (1~2.5 cm in diameter) in all the treated rats at 20 weeks, whereas MHBN and BBN induced only small tumors at 20 weeks (Photo 1a), although development of relatively large tumors (0.5~1.0 cm in diameter) was observed with these compounds at 30 weeks. Tumors induced by these 3 compounds were histologically diagnosed as transitional cell carcinoma with or without squamous metaplasia, the degree of which was found to be more marked
with tumors induced by EHBN than with those induced by the other compounds (Photo 2). Keratinizing pattern was observed not only in cancers but in papillomas (Photo 1b). Basal cell carcinoma was observed in the urinary bladder of one rat treated with EHBN (Photos 3 and 4). In contrast, AHBN did not induce cancer at 30 weeks, though 3 out of 7 rats developed papillomas which were microscopically detectable. t-BBN did not induce any tumors.

(B) BCPN Homolog and Analog: As reported earlier,9) ECPN selectively induced bladder cancer in all the treated rats at 20 weeks as effectively as the original compound, EHBN. BCMN did not induce any tumors.

(C) BBN Analogs with 3-Hydroxypropyl Group: Any histological changes were not observed in the rats treated with EHPN and BHPN, either in the urinary bladder or in other organs. All the rats receiving BHPN for 20 weeks as well as for 52 weeks did not have any tumors.

(D) DBN Homologs and Its Metabolites: PBN induced liver and esophageal tumors in all the rats and 6 out of 10 rats respectively at 16–19 weeks. Liver tumors were diagnosed as poorly or well-differentiated hepatomas (Photo 8), and esophageal tumors as papillomas. Hyperplasia of the urinary bladder was observed in 2 out of 10 treated rats but no cancer of the bladder. DBN also induced hepatomas but no esophageal tumors. One and 5 out of 9 rats respectively developed papillomas and focal hyperplasia in the bladder, but no bladder cancer was induced by DBN in the present experiment. BHBN-3 did not induce any tumors under the present experimental conditions, although degenerative changes in the liver were observed histo-

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Table II. Induction of Tumors in Rats with N-Nitrosamines Related to N-Butyl-N-(4-hydroxybutyl)nitrosamine and N,N-Dibutylnitrosamine, and the Histological Findings

<table>
<thead>
<tr>
<th>Compound</th>
<th>Period for drinking water (weeks)</th>
<th>Target organ, incidence (%), and histology of tumors</th>
<th>Urinary bladder</th>
<th>Liver</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>with compound</td>
<td>Hyperplasia</td>
<td>Papilloma</td>
<td>Cancer</td>
</tr>
<tr>
<td>MHBN</td>
<td>20</td>
<td>2/3(67)</td>
<td>2/3(67)</td>
<td>1/3(33)</td>
</tr>
<tr>
<td>MHBN</td>
<td>20</td>
<td>6/6(100)</td>
<td>6/6(100)</td>
<td>6/6(100)</td>
</tr>
<tr>
<td>EHBN</td>
<td>20</td>
<td>19/19(100)</td>
<td>19/19(100)</td>
<td>19/19(100)</td>
</tr>
<tr>
<td>BBN</td>
<td>20</td>
<td>3/3(100)</td>
<td>2/3(67)</td>
<td>0/3(0)</td>
</tr>
<tr>
<td>BBN</td>
<td>20</td>
<td>7/7(100)</td>
<td>7/7(100)</td>
<td>2/7(29)</td>
</tr>
<tr>
<td>AHBN</td>
<td>20</td>
<td>3/3(100)</td>
<td>0/3</td>
<td>0/3(0)</td>
</tr>
<tr>
<td>AHBN</td>
<td>20</td>
<td>6/7(86)</td>
<td>3/7(43)</td>
<td>0/7(0)</td>
</tr>
<tr>
<td>t-BBN</td>
<td>20</td>
<td>0/3</td>
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<td>0/3(0)</td>
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<td>20</td>
<td>0/7</td>
<td>0/7</td>
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</tr>
<tr>
<td>ECPN</td>
<td>20</td>
<td>9/9(100)</td>
<td>9/9(100)</td>
<td>9/9(100)</td>
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<td>20</td>
<td>0/3</td>
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<tr>
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<td>20</td>
<td>0/3</td>
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</tr>
<tr>
<td>BHPN</td>
<td>52</td>
<td>0/8</td>
<td>0/8</td>
<td>0/8(0)</td>
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<tr>
<td>PBN*</td>
<td>16–19</td>
<td>2/10(20)</td>
<td>0/10</td>
<td>0/10(0)</td>
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<tr>
<td>DBN</td>
<td>20</td>
<td>5/9(56)</td>
<td>1/9(11)</td>
<td>0/9(0)</td>
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<tr>
<td>BHBN-3</td>
<td>20</td>
<td>0/7</td>
<td>0/7</td>
<td>0/7(0)</td>
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<tr>
<td>BOBN-3</td>
<td>20</td>
<td>0/10</td>
<td>0/10</td>
<td>0/10(0)</td>
</tr>
<tr>
<td>BOBN-2</td>
<td>20</td>
<td>0/9</td>
<td>0/9</td>
<td>0/9(0)</td>
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</table>

* Six out of 10 rats developed esophageal papillomas.
CARCINOGENICITY OF BBN-RELATED COMPOUNDS

logically in a few of the treated rats. Large liver tumors developed by treatment with BOBN-2 and BOBN-3 for 20 weeks in all the rats and 2 out of 10 rats, respectively. These tumors were diagnosed as poorly or well-differentiated hepatomas (Photo 6). Metastasis of the tumor into the lung was observed in the rats treated with BOBN-2 (Photo 7). Numerous hyperplastic nodules were seen in nearly all the rats treated with BOBN-3 or BOBN-2 (Photo 5).

**DISCUSSION**

Induction of urinary bladder tumors in rats by the oral administration of an N-nitroso compound, DBN, was first reported by Druckrey et al.8) The principal target organ of DBN, however, was not the urinary bladder but the liver or the esophagus. Subsequently, selective induction of bladder tumors in rats was demonstrated by Druckrey et al.7) by the oral administration of the \( \omega \)-hydroxylated derivative of DBN, namely BBN. Based on their extensive studies6) on the carcinogenicity and target organs of numerous N,N-dialkyl-nitrosamines in rats, it was presumed that dibutyl structure is responsible for the induction of bladder tumors since neither symmetric N,N-dialkyl-nitrosamines other than DBN nor asymmetric ones having a butyl group induced bladder tumors.

In order to elucidate a possible relationship between the metabolism and organotropic effect of BBN and DBN on the urinary bladder, the metabolic fate of these compounds in the rat was investigated by Okada and his co-workers.16,19,20,22) They demonstrated that the principal urinary metabolite of BBN as well as of DBN was BCPN which was also a potent and selective bladder carcinogen as BBN.10) Then the carcinogenic effect of several compounds structurally related to BBN, which had alkyl and \( \omega \)-hydroxyalkyl groups, was investigated.15) Among these compounds only a BBN homolog, N-propyl-N-(4-hydroxybutyl)nitrosamine (PHBN), which had a propyl and 4-hydroxybutyl groups, was found to be selectively effective on the urinary bladder. The principal urinary metabolite of PHBN, on the other hand, was identified by Okada et al.17) as N-propyl-N-(3-carboxypropyl)nitrosamine (PCPN), a homolog of BCPN. It seemed reasonable, therefore, to assume that an essential structural requirement of the N-nitrosamine for the selective induction of urinary bladder cancer may not be to possess a dibutyl structure but may be to have a 4-hydroxybutyl group which undergoes metabolic transformation to a 3-carboxypropyl chain.

To verify this assumption, carcinogenicity of other BBN homologs, MHBN, EHBN, and AHBN, was studied. As presumed, selective induction of bladder tumors was demonstrated with these compounds similarly with BBN and PHBN, although AHBN having a relatively long alkyl chain did not induce cancers but only papillomas under the present experimental conditions. Metabolic study13) on these BBN homologs in the rat, on the other hand, demonstrated that the principal urinary metabolite of MHBN and EHBN was the corresponding BCPN homolog, N-methyl-N-(3-carboxypropyl)nitrosamine (MCPN) and ECPN, respectively, while that of AHBN was not a BCPN homolog but a BCMN homolog, N-amyl-N-(carboxymethyl)nitrosamine. As was reported earlier9,10) and as described below, the BCPN homologs may be regarded as the active metabolite of the original BBN homologs, while BCMN having a carboxymethyl group was found to be noncarcinogenic. Consequently, the weak carcinogenic effect of AHBN on the urinary bladder could be explained by the urinary excretion of a small quantity of the active metabolite.

\( t \)-BBN which is also a homolog of BBN induced neither bladder tumors nor any tumors in other organs. The principal urinary metabolite of \( t \)-BBN was identified as N-tert-butyl-N-(3-carboxypropyl)nitrosamine, a BCPN homolog. In this connection, N,N-dialkyl-nitrosamines having a tert-butyl group
were reported\textsuperscript{6,12} to be non-carcinogenic. Absence of a hydrogen atom at α-position of the one alkyl group of N,N-dialkynitrosamines may be associated with their non-carcinogenicity.

ECPN, the principal urinary metabolite of EHBN,\textsuperscript{13} selectively induced bladder cancer as effectively as the original compound, so that ECPN is responsible for the organospecific carcinogenicity of EHBN to the urinary bladder as in the case of BCPN and BBN.\textsuperscript{10} The carcinogenicity of BCPN homologs, MCPN and PCPN, has not yet been examined but it seems quite reasonable to assume that they should be selectively carcinogenic to rat urinary bladder by analogy of the relationship demonstrated between BBN and BCPN as well as between EHBN and ECPN.

BCMN was first obtained as one of several minor urinary metabolites of BBN or DBN in rats,\textsuperscript{20} and then as the major metabolite of a potent carcinogen, N-butyl-N-(2-hydroxyethyl)nitrosamine (BHEN),\textsuperscript{17} which induced hepatomas as well as papillomas of the esophagus but no bladder tumors in rats. This finding indicated that BCMN would not be tumorigenic to the urinary bladder.\textsuperscript{15} As predicted, BCMN induced neither bladder tumors nor any tumors in other organs.

In order to confirm our previous noteworthy finding\textsuperscript{15} that BHPN, a BBN analog having a 3-hydroxypropyl group, was not tumorigenic, the effect of BHPN was again examined together with that of EHPN, a homolog of BHPN. Both compounds were found to be nontumorigenic, thus confirming the previous finding. In the present experiment, BHPN was given to a group of rats for a much longer period (52 weeks) as compared with the previous experiment (20 weeks). The principal urinary metabolite of BHPN\textsuperscript{17} and EHPN\textsuperscript{23} was identified as the corresponding N-alkyl-N-(2-carboxyethyl)nitrosamine, the carcinogenicity of which has not yet been tested. However, in the light of the above results, N-alkyl-N-(2-carboxyethyl)nitrosamine should not be involved at least in the induction of bladder tumors.

It is quite evident from the results obtained in the present experiment that the dibutyl structure in N,N-dialkynitrosamines is not an indispensable requirement for inducing bladder tumors as presumed earlier.\textsuperscript{7} Furthermore, it can be mentioned that the structural and metabolic requirements for the selective induction of bladder tumors are to possess a 4-hydroxybutyl chain which undergoes metabolic transformation to the 3-carboxypropyl group, resulting in a considerable excretion of a metabolite having this group into urine. The presence of a 4-hydroxybutyl chain, however, is essential but not sufficient enough for the selective induction of bladder tumors, as illustrated with t-BBN and N-(α-hydroxyalkyl)- N-(4-hydroxybutyl) nitrosamines.\textsuperscript{14}

Druckrey \textit{et al.}\textsuperscript{6} examined the carcinogenicity of several asymmetric N,N-dialkynitrosamines having a butyl group but PBN was not included among them. Hepatomas were induced in all the rats treated with PBN. Simultaneous development of papillomas in the esophagus was observed with PBN, but the incidence was lower than that of hepatomas (Table II). In contrast, it was reported\textsuperscript{5,6} that asymmetric N,N-dialkynitrosamines such as N-methyl-N-butyl- and N-ethyl-N-butyl-nitrosamines preferentially induced esophageal tumors in rats. Based on our metabolic study of PBN in the rat,\textsuperscript{18,21} it was expected that PBN might induce bladder cancer but all the rats treated with PBN died due to hepatomas before possible development of bladder cancer.

In view of our previous finding\textsuperscript{15} that hepatocarcinogenic N-nitrosamines such as BHEN and N-butyl-N-(2-oxopropyl)nitrosamine (BOPN) markedly enhanced the activities of serum enzymes (GOT, GPT, alkaline phosphatase) associated with the impairment of liver function while urinary bladder carcinogens (BBN, PHBN) as well as noncarcinogenic compound (BHPN) did not
affect the enzyme levels, the activities of these enzymes were assayed with EHBN, BBN (as the control), AHBN, BCMN, and PBN. Significantly higher enzyme levels were noticed only with PBN which has been found to be a potent hepatocarcinogen.

In contrast to the previous finding of Druckrey et al.,\textsuperscript{6,40} DBN induced hepatomas but neither esophageal tumors nor bladder cancers under our experimental conditions, although development of papillomas and focal hyperplasia in the urinary bladder was observed. The metabolic fate of DBN in the rat, on the other hand, was elucidated by Okada and his co-workers\textsuperscript{19,20,22} and by Blattmann and Preussmann.\textsuperscript{2,3} DBN underwent metabolic transformation in at least three ways. First it was metabolized via BBN. Secondly, it underwent ($\omega$-1)-oxidation of the one butyl group to give BHBN-3, and thirdly ($\omega$-2)-oxidation of the one alkyl chain. BHBN-3 and the ($\omega$-2)-hydroxylated derivative of DBN were conjugated with glucuronic acid to form glucuronides or further metabolized through the corresponding oxobutyl compounds, namely BOBN-3 and BOBN-2, respectively.\textsuperscript{4,22}

Tumors were not induced by BHBN-3 under the present experimental conditions, and only degenerative changes in the liver was observed histologically. In the light of potent hepatocarcinogenic effect of N-alkyl-N-(oxoalkyl)nitrosamines, such as BOPN\textsuperscript{18} and N-propyl-N-(2-oxopropyl)nitrosamine\textsuperscript{1} in rats, carcinogenicity of BOBN-3 and BOBN-2 was examined. Both compounds induced hepatomas as expected, but no esophageal tumors. BOBN-2 was more effective than BOBN-3 (Table II).

Among a number of N,N-dialkylnitrosamines only DBN induced tumors of the urinary bladder as well as tumors of the liver and the esophagus. From the metabolic point of view, it seems quite reasonable to assume that $\omega$-oxidation of DBN to form BBN is responsible for the induction of bladder tumors, while ($\omega$-1)- or ($\omega$-2)-oxidation may be responsible for inducing tumors of the liver and/or the esophagus, although the induction of esophageal tumors by any metabolites of DBN has not so far been demonstrated. Multiple carcinogenic effects on different organs of an “indirect” carcinogen may be due to the diversity in its metabolic activation.

The ACI/N rat used in the present experiment seems to be an extremely relevant strain, so far as the tumorigenic response to N,N-dialkylnitrosamines and related compounds is concerned. The results of carcinogenicity tests summarized in Table II clearly indicate all-or-none response of the rat to the compounds tested with few exceptions, particularly in the induction of urinary bladder cancers. Under our present experimental conditions, calculus formation in the urinary bladder of the rat was hardly noticeable.

The authors are grateful to Drs. S. Odashima and A. Maekawa, National Institute of Hygienic Sciences, for histological examinations. They are also indebted to Mr. M. Iiyoshi for his help in preparing the N-nitrosamines and Dr. H. S. Kitagawa and Mrs. K. Miyajima for their assistance in animal experiments and enzyme assays, respectively.

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\textbf{REFERENCES}


EXPLANATION OF PLATES

Photo 1. (a) Papilloma of the urinary bladder of a rat treated with MHBN for 20 weeks. Cauliflower-like growth which consisted of papillary benign proliferation of the epithelium is seen. (b) Marked keratinizing metaplasia in a papilloma of the urinary bladder of a rat given EHBN for 20 weeks. Besides the papillomas large and small carcinomas were developed in the urinary bladder. H-E. × 20.


Photo 3. Downward proliferation of epithelial cells (upper dark area) and basal cell carcinoma cells in the urinary bladder of a rat treated with EHBN for 20 weeks. H-E. × 100.


Photo 6. Hepatoma in a rat treated with BOBN-2 for 20 weeks.

Photo 7. Metastatic foci of the lung of the rat in Photo 6. Artery filled with tumor cells is seen in the upper right. H-E. × 100.


H-E=Hematoxylin and Eosin stain
CARCINOGENICITY OF BBN-RELATED COMPOUNDS