Esophageal and Hepatic Microsomal Metabolism of \( N \)-Nitrosomethylbenzylamine and \( N \)-Nitrosodimethylamine in the Rat

George E. Labuc and Michael C. Archer

Department of Medical Biophysics, University of Toronto, Ontario Cancer Institute, Toronto, Ontario, Canada M4X 1K9

ABSTRACT

The metabolism of the rat esophageal carcinogen \( N \)-nitrosomethylbenzylamine (NMBzA) was studied using microsomes prepared from liver and esophageal mucosa of untreated male Sprague-Dawley rats. NMBzA was extensively metabolized to benzaldehyde, benzyl alcohol, and formaldehyde by hepatic microsomes. The rate of metabolism at the benzyl moiety was 10-fold higher than that at the methyl moiety. Mucosal microsomes metabolized NMBzA to benzaldehyde and formaldehyde at rates one-fifth and one-sixtieth of those in the liver, respectively; benzyl alcohol formation was undetectable. Esophageal metabolism of NMBzA was exclusively located in the mucosa, preferentially in the microsomal fraction, was reduced nicotine-amine adenine dinucleotide phosphate dependent, and was inhibited by CO and 2-diethylaminoethyl-2,2-diphenylvalerate. A low level of cytochrome P-450 was detected in the mucosal microsomes. Whereas hepatic metabolism of NMBzA was inducible by phenobarbital pretreatment, mucosal metabolism was not altered by either phenobarbital or 3-methylcholanthrene pretreatment. The hepatocarcinogen \( N \)-nitrosodimethylamine was extensively metabolized by hepatic microsomes to formaldehyde, but demethylation was not detected in the microsomes from esophageal mucosa, a nontarget tissue. The results indicate that rat esophageal mucosa contains an enzyme system which metabolizes NMBzA at a high rate and exhibits properties typical of cytochrome P-450. This enzyme may play a role in determining which compounds induce tumors in rat esophagus.

INTRODUCTION

The rat esophagus is a primary target organ for many carcinogenic \( N \)-nitrosamines (1, 11). High incidences of basal cell and squamous cell tumors are induced by these nitrosamines, independent of the route of administration (1, 11). Apart from the nitrosamines and some nitrosamides, few other chemicals have been reported to induce esophageal tumors (6, 20). However, the mechanism of this remarkable sensitivity of the rat esophagus to nitrosamines is not known.

In general, tissues that are sensitive to tumor induction by indirect acting chemicals exhibit the capacity to metabolize such compounds into electrophilic intermediates, which are capable of interacting with tissue macromolecules (9, 13). However, little is known concerning the metabolism of nitrosamines by the rat esophagus. A number of studies have demonstrated the alkylation of esophageal macromolecules when nitrosamines are incubated with intact cell preparations, such as esophageal slices (2) or cultured explants (23), providing indirect evidence for the metabolic activation of these compounds in the esophagus. Rat and hamster esophageal slices have also been reported to oxidize NDMA3 and \( N \)-nitrosodimethylamine to CO\(_2\) (14). However, direct studies on the formation of nitrosamine metabolites using cell-free systems have provided contradictory results. Whereas one report indicated a higher rate of nitrosamine metabolism by esophageal microsomes than by hepatic microsomes (22), another study reported little if any activity in the esophagus (21).

NMBzA is a potent esophageal carcinogen and toxin in the rat but produces no liver tumors (1); NDMA is a potent hepatocarcinogen that gives no esophageal tumors in the rat (1, 11). The aim of the present study was to investigate the metabolism of NMBzA and NDMA in both target and nontarget tissues. The predominant pathway for nitrosamines is considered to be hydroxylation at the \( \alpha \)-carbon, an activation step which ultimately yields an alkylating agent and an aldehyde in stoichiometric amounts (9). The alkylating agent can react with tissue nucleophiles or with water to form the corresponding alcohol. Thus, NDMA is known to yield formaldehyde and a methylating agent (9). NMBzA is expected to yield formaldehyde and a benzylating agent from hydroxylation at the methyl carbon, or benzaldehyde and a methylating agent from hydroxylation at the benzyl carbon of the benzyl moiety. NMBzA was found to be metabolized by esophageal microsomes via a cytochrome P-450 pathway almost exclusively at the benzyl carbon. In contrast, hepatic microsomal metabolism of NMBzA occurred at both \( \alpha \)-carbons. NDMA metabolism was detected only in the liver.

MATERIALS AND METHODS

Chemicals. NMBzA (b.p. 60–62°/0.01 mm), synthesized by the method of Druckrey et al. (1), was >99% pure as determined by gas chromatography and high-performance liquid chromatography. NDMA was purchased from Eastman Kodak Co., Rochester, N. Y. \([\text{methyl-}1^4\text{C}]\text{NMBzA}\) (5.46 mCi/mmol) and \([\text{methyl-}1^4\text{C}]\text{NDMA}\) (54.2 mCi/mmol) were obtained from New England Nuclear, Boston, Mass. and \([\text{methyl-}1^4\text{C}]\text{NMBzA}\) (30 mCi/mmol) was obtained from Amersham–Searle Corp., Arlington Heights, Ill. NADP\(^+\) (sodium), G-6-P (sodium), G-6-P dehydrogenase (type XV), \( \beta \)-NAD\(^+\) (Grade III), arachidonic acid (type I), phenazine ethosulfate, and bovine serum albumin (Fraction V) were purchased from Sigma Chemical Co., St. Louis, Mo. SKF 525-A was a generous gift from Smith, Kline & French Laboratories, Mississauga, Ontario.

Animals. Male Sprague-Dawley rats (21 to 23 days old) were purchased from Charles River Canada, Inc., Laprairie, Quebec, Canada. Rats were maintained on Teklad 6% fat rat-mouse diet (Teklad Mills, Iowa) ad libitum for 5 to 10 days prior to sacrifice. Where specified, ---

1 This work was supported by the Ontario Cancer Treatment and Research Foundation and Grant MT7025 from the Medical Research Council of Canada.
2 To whom requests for reprints should be addressed.

Received November 18, 1981; accepted April 1, 1982.

AUGUST 1982
animals were pretreated with phenobarbitone (0.1% in drinking water for 5 days) or either corn oil (0.8 ml/100 g body weight) or 3-MC (80 mg/kg body weight) in corn oil, injected i.p. 40 hr prior to sacrifice.

**Enzyme Preparation.** Rats were sacrificed by a blow to the head. Liver and esophagus were excised and rinsed in cold homogenizing buffer [1.15% (w/v) KCl-50 mM Tris-HCl, pH 7.40]. All subsequent steps were carried out at 4°. Esophageal mucosa was obtained by physically shearing the outer muscle-submucosa layers away from the mucosa. The whole esophagus was griped centrally with 2 pairs of forceps; holding one pair firmly in place, the second was gently moved towards one end of the tissue, such that the outer muscle-submucosa peeled away from the mucosa. This process was repeated at the other end. The resulting mucosa was completely intact and contained no muscle or submucosa contamination, as determined by routine histology. The mucosa was slit longitudinally, blotted dry, weighed, and cut into small pieces. Mucosa of an appropriate number of rats (typically 7 to 10) to give the required amount of microsomal protein were combined and homogenized in 9 volumes of homogenizing buffer, with 30 passes of a motor-driven Duali all-glass homogenizer (20-sec stoppages after each 10 passes). Where specified, whole esophagus or the muscle-submucosa was similarly homogenized. Liver was homogenized in 3 volumes of homogenizing buffer with 8 passes of a motor-driven Potter Elvehjem homogenizer with Teflon pestle. Homogenates were routinely centrifuged at 9,000 x g for 20 min, and the postmitochondrial supernatants were centrifuged at 100,000 x g for 60 min (an initial spin at 2,000 x g for 10 min was undertaken for the desired subcellular fractionation study). The 100,000 x g pellet, referred to henceforth as the microsomal fraction, was resuspended to a final concentration of approximately 5 mg protein per ml in homogenizing buffer for metabolism studies. A yield of 4.49 ± 0.17 (S.E.) mg of microsomal protein was obtained per g mucosa, each mucosa weighing 40 to 50 mg. For cytochrome P-450 assays, the microsomes were washed with 1.15% KCl and resuspended in 50 mM Tris-HCl (pH 7.40).

**Incubation Conditions.** Unless stated otherwise, incubation mixtures contained 5 mM substrate (NMBzA; [methyl-14C]NMBzA, 0.33 mM/mmol; [methylenedioxy-14C]NMBzA, 0.33 mM/mmol; or [methyl-14C]NDMA, 0.33 mM/mmol), 20 mM semicarbazide, 2 mM NADP+, 10 mM G-6-P, G-6-P dehydrogenase, 2 units/ml, 10 mM MgCl2, 50 mM Tris (pH 7.40), and 0.1 ml of microsomal suspension (approximately 0.5 mg of protein) in a final volume of 0.6 ml. Reactions were started by the addition of microsomes to the other components which had been preincubated at 37° for 5 min. Samples were incubated at 37° under air with shaking for 20 min (NMBzA metabolism) or for 40 min (NDMA metabolism). Reactions were terminated by placing the mixtures on ice. Under these conditions, reaction rates were linear with respect to both incubation time and protein concentration. Formation of benzyl alcohol from benzaldehyde as substrate (16.7 μm) was measured using identical incubation conditions.

**Metabolite Analyses.** Incubation mixtures containing unlabeled NMBzA were processed for determination of benzaldehyde, benzyl alcohol, and benzoic acid. Samples were deproteinized with 0.3 ml of 20% (w/v) ZnSO4·7H2O and 0.3 ml of saturated Ba(OH)2 solution. Following centrifugation of the precipitates, supernatants were stored in the dark at −20° overnight. Freezing was essential for minimizing nonenzymatic production of benzaldehyde prior to analysis. Supernatants were thawed, and immediately chromatographed on a C18 reversed-phase column (Waters Associates, Inc., Milford, Mass.), eluted with 15% (v/v) methanol-3% (v/v) acetic acid, pH 4.20, at a flow rate of 2.0 ml/min. The eluate was monitored at 260 nm, and metabolites were quantitated by peak height analysis. A typical chromatogram is shown in Chart 1. Under these conditions, benzaldehyde semicarbazone exists in equilibrium with small quantities of free benzaldehyde. The identities of the metabolites that cochromatographed with authentic benzyl alcohol and benzaldehyde semicarbazone were confirmed by gas chromatography-mass spectroscopy. The fraction containing benzyl alcohol was collected and extracted directly into dichloromethane. The fraction containing benzaldehyde semicarbazone was treated with HNO3 (3) to generate free benzaldehyde, which was then extracted into dichloromethane.

In some studies, incubations containing [methyl-14C]NMBzA or [methylenedioxy-14C]NMBzA were deproteinized and subjected to chromatography as above. Fractions (0.5 min) were collected and counted for 14C in 10 ml ACS (Amersham).

Incubation mixtures containing [methyl-14C]NMBzA or -NDMA were analyzed for [14C]formaldehyde production by quantitative precipitation as the dimedone derivative by the method of Paik and Kim (19), except that the deproteinized supernatants were extracted 3 times with dichloromethane (1.0 ml) prior to dimedone formation in order to remove residual substrate.

Controls were mixtures incubated at 37° in the absence of tissue or mixtures incubated at 0° in the presence of tissue. Both gave similar background levels. Low levels of benzaldehyde (0.005%) and benzoic acid (0.05%) were present as contaminants in the NMBzA, and [14C]formaldehyde (0.01 and 2.3%) was detected in the [methyl-14C]NMBzA and [methyl-14C]NDMA, respectively.

**Electron Microscopy.** The 100,000 x g pellet obtained from esophageal mucosa was fixed using 3.5% glutaraldehyde in cacodylate buffer, followed by 1% osmium tetroxide. The pellet was dehydrated in acetone and then embedded in Epon. Thin sections were stained with uranyl acetate and lead citrate and examined in an Elmiskop 1 electron microscope.

**Other Analyses.** Cytochrome P-450 was determined spectrophotometrically by the method of Jakobsson and Cinti (7), using dithionite as the reducing agent. Protein was analyzed by the method of Lowry et al., as described by Munro and Fleck (15), using bovine serum albumin as standard. Statistical analyses were carried out using 2-tailed Student's t tests.

**RESULTS**

Microsomes prepared from either liver or esophageal mucosa metabolized NMBzA to benzaldehyde and formaldehyde.
(Table 1). In addition, benzyl alcohol was a major hepatic metabolite but was not detectable in incubations containing mucosal microsomes (Table 1). Benzyl alcohol formation from the reaction of a benzylation intermediate with water theoretically cannot exceed the rate of formaldehyde formation. However, hepatic microsomes extensively reduced benzaldehyde to benzyl alcohol (0.73 nmol/min/mg protein), using a benzaldehyde concentration (16.7 μM) typically measured in the NMBzA incubation mixtures. Thus, the high rate of benzyl alcohol formation from NMBzA by hepatic microsomes results primarily from benzaldehyde reduction. Preliminary experiments demonstrated that 20 mM semicarbazide in the incubation medium minimized benzaldehyde reduction; higher concentrations lacked further effect. Since semicarbazide did not alter nitrosoamine metabolism, it was routinely included in all subsequent assays. In contrast to hepatic microsomes, mucosal microsomes lacked benzaldehyde reductase activity (≤0.14 nmol/min/mg protein). In neither microsomal fraction was oxidation of benzaldehyde to benzoic acid detectable, although benzoic acid was present as a minor contaminant of NMBzA. Furthermore, formaldehyde was stable with both tissues under the incubation conditions, indicating minimal underestimation of NMBzA demethylase activities. Taking these factors into account, it was calculated that microsomes from esophageal mucosa metabolized NMBzA at the methylene carbon of the benzyl moiety to yield benzaldehyde, at about one-fifth the rate of hepatic microsomal metabolism (allowing in this case for reduction of benzaldehyde). In contrast, mucosal metabolism of NMBzA at the methyl carbon to yield formaldehyde occurred at a rate 60 times lower than the rate of hepatic metabolism.

Radioactivity profiles of the chromatograms revealed the absence of metabolites of [methyl-14C]- or [methylene-14C]-NMBzA other than those described above in either hepatic or mucosal incubations. In particular, enzymatic formation of methylbenzylamine (eluting at 3.4 min) was not detected (≤0.08 nmol/min/mg protein), indicating an absence of denitrosation.

Under incubation conditions identical to those used for NMBzA metabolism, NDMA was extensively metabolized to formaldehyde by hepatic microsomes (Table 1). In contrast, microsomes prepared from esophageal mucosa lacked detectable NDMA demethylase activity (Table 1). The high detection limit (0.03 nmol/min/mg protein) was due to the presence of relatively high levels of [14C]formaldehyde as a contaminant of the [methyl-14C]NDMA.

The characteristics of the metabolizing system in the mucosa were then examined. Incubation of NMBzA in the presence of mammals (100,000 × g pellet) obtained from whole esophagus, muscle-submucosa, or residual muscle-submucosa indicated that the mucosa contained the entire esophageal metabolizing activity, as determined by benzaldehyde production (Table 2). Microsomes prepared from whole esophagus exhibited lower activity per esophagus compared to those prepared from the mucosa, although the difference was not significant. The lower activity probably reflects a lower efficiency of homogenization of whole esophagus, resulting in a lower yield of microsomes. An identical tissue distribution of NMBzA metabolism was obtained when using whole homogenates rather than microsomes.

Subcellular fractionation of the mucosa was carried out using a differential centrifugation method commonly used for hepatic subcellular fractionation. Although the "nuclear" (2,000 × g pellet) and "mitochondrial" (9,000 × g pellet) fractions contained some capacity to metabolize NMBzA to benzaldehyde, the microsomal (100,000 × g pellet) fraction was markedly enriched in this activity compared to the other fractions (Chart 2) and contained approximately 40% of total homogenate activity. No activity was detected in the "cytosol" (100,000 × g supernatant). Electron microscopy of the microsomes from esophageal mucosa indicated the presence of some endoplasmic reticulum vesicles, with masses of free ribosomes and low levels of contaminating fragments of other organelles (Fig. 1). Thus, based on the preferential localization of NMBzA metabolizing activity and the electron microscopic appearance, microsomes prepared from esophageal mucosa appear to be similar to hepatic microsomes.

---

### Table 1

<table>
<thead>
<tr>
<th>Substrate</th>
<th>Metabolite</th>
<th>Hepatic microsomes (nmol/min/mg protein)</th>
<th>Mucosal microsomes (nmol/min/mg protein)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NMBzA</td>
<td>Benzaldehyde</td>
<td>0.978 ± 0.106 (4)</td>
<td>0.549 ± 0.055 (18)</td>
</tr>
<tr>
<td></td>
<td>Benzyalcohol</td>
<td>2.0 ± 0.3 (4)</td>
<td>&lt;0.1</td>
</tr>
<tr>
<td></td>
<td>Formaldehyde</td>
<td>0.289 ± 0.087 (4)</td>
<td>0.005 ± 0.001 (6)</td>
</tr>
<tr>
<td>NDMA</td>
<td>Formaldehyde</td>
<td>0.871 ± 0.102 (4)</td>
<td>≤0.03</td>
</tr>
</tbody>
</table>

* Mean ± S.E.

Numbers in parentheses, number of replicate assays.

---

### Table 2

<table>
<thead>
<tr>
<th>Tissue layer</th>
<th>Benzaldehyde production (pmol/min/esophagus)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole esophagus</td>
<td>81.0 ± 9.8*</td>
</tr>
<tr>
<td>Mucosa</td>
<td>111.1 ± 17.4</td>
</tr>
<tr>
<td>Muscle-submucosa</td>
<td>2.2 ± 1.1</td>
</tr>
</tbody>
</table>

* Mean ± S.E. of 4 replicate assays.
NMBzA metabolism to benzaldehyde by mucosal microsomes appeared to be enzymatic in nature. Metabolism was dependent on both the presence of tissue and incubation at 37°C, whereas boiling the microsomes for 5 min prior to incubation completely abolished activity (Table 3). Furthermore, this metabolizing enzyme appears to be a typical cytochrome P-450-dependent system. Metabolism was dependent on the presence of a NADPH-generating system, and it was markedly inhibited by the classical cytochrome P-450 inhibitors, CO or 0.6 mM SKF 525-A (Table 3). Metabolism of NMBzA was not detected when arachidonic acid (0.18 μmol in 2.7 μl ethanol) was used in place of the NADPH-generating system.

Analogous studies examining NMBzA metabolism to formaldehyde also indicated this metabolic pathway to be preferentially located in the mucosal microsomes and to be cytochrome P-450 mediated.

To further examine the cytochrome P-450 dependency of NMBzA metabolism, rats were pretreated with either phenobarbitone or 3-MC. Phenobarbitone pretreatment greatly increased hepatic microsomal metabolism of NMBzA to benzaldehyde (3.4-fold), benzyl alcohol (6.0-fold), and formaldehyde (4.2-fold), and a significant production of benzoic acid was detected (0.40 nmol/min/mg protein). In contrast, phenobarbitone pretreatment did not alter NMBzA metabolism by microsomes from esophageal mucosa. 3-MC pretreatment did not alter NMBzA metabolism by either hepatic or mucosal microsomes.

Cytochrome P-450 could not be detected in the mucosal microsomes using the conventional reduced CO difference spectrum method of Omura and Sato (18) due to the presence of contaminating cytochrome oxidase. However, when the method of Jakobsson and Cinti (7) was used, which involved preincubating the microsomes with succinate to remove mitochondrial interference, reduced CO difference spectroscopy revealed a typical cytochrome P-450 spectrum, with λ<sub>max</sub> at 450 nm (Chart 3). The peak at 423 nm is likely to represent cytochrome b<sub>5</sub>, which would also exhibit a difference spectrum under these conditions (7). Incubation in the presence of ascorbate and phenazine ethosulfate did not alter the height of the peak at 423 nm, indicating a lack of heme contamination (8). Using an extinction coefficient of 91 mM⁻¹ cm⁻¹ (18), the level of cytochrome P-450 in the mucosal microsomes was calculated to be 0.096 ± 0.012 nmol/mg protein (n = 4), approximately 15% of that detected in hepatic microsomes (0.66 ± 0.10 nmol/mg protein, n = 4).

**DISCUSSION**

The present results demonstrate that both rat esophageal mucosa and rat liver are able to metabolize the esophageal...
carcinogen NMBzA. However, whereas hepatic microsomes oxidized NMBzA at the methylene carbon at a rate approximately 10-fold higher than at the methyl carbon, this differential was 100-fold in the case of mucosal metabolism. Schweinsberg and Kouros (22) have reported similarly a high rate of NMBzA metabolism to benzaldehyde by microsomes prepared from the whole esophagus of the rat, whereas formaldehyde was not detected. In that study (22), formaldehyde was assayed by the colorimetric method of Nash (16), which is much less sensitive than is the dimesone assay (19). In contrast to the present results, Schweinsberg and Kouros (22) did not detect hepatic metabolism of NMBzA to formaldehyde and reported that the specific activity of the debenzylase in the whole esophageal microsomes was greater than that in the hepatic microsomes. Schweinsberg and Kouros (22), however, did not use an aldehyde-trapping agent, such as semicarbazide, in their incubation mixtures nor did they measure formation of benzyl alcohol. Under the present conditions, both formaldehyde and benzaldehyde were further metabolized when they were incubated with hepatic microsomes in the absence of a trapping agent, but they were considerably more stable in the presence of mucosal microsomes. Thus, the hepatic metabolism of NMBzA in the study of Schweinsberg and Kouros (22) is probably underestimated.

In contrast to the present findings and those of Schweinsberg and Kouros (22) that indicate a high level of NMBzA metabolism by rat esophageal microsomes, Scanlan et al. (21) were unable to detect metabolism of another esophageal carcinogen, 2,6-dimethyldinitrosopiperazine, by microsomes and cytosol obtained from the combined esophagus and forestomach of the rat. However, N-nitrosopyrrolidine, which is not an esophageal carcinogen, was metabolized at a low rate (21). The reason for the lack of metabolism of 2,6-dimethyldinitrosopiperazine by the esophagus is not known at this stage.

Since the esophageal mucosa metabolizes NMBzA to benzyaldehyde at approximately 100 times the rate of metabolism to formaldehyde, formation of a methylylating intermediate is expected to occur similarly at a 100-fold faster rate than is the formation of the benzylating intermediate. Studies by Hodgson et al. (4, 5) have in fact demonstrated that, following a single i.v. dose of NMBzA to rats, high levels of esophageal DNA methylation, including the promutagenic adduct O\textsuperscript{2}-methylguanine, are produced, whereas benzylolation of DNA is undetectable. Thus, although other factors such as differences in stability or nucleophile affinities (24) may exist between the methylylating and benzylating intermediates, the present study suggests that the relative lack of esophageal DNA benzylolation is primarily due to a low rate of formation of benzylating intermediate.

The metabolism of NMBzA within the esophageal mucosa appears to involve a typical cytochrome P-450 pathway. Like typical mixed-function oxidases in the liver, NMBzA metabolism in the mucosa is NADPH dependent, is preferentially located in the microsomal fraction, and is inhibited by CO and SKF 525-A. A low level of cytochrome P-450 is also detectable in the mucosal microsomes. A number of studies have demonstrated that extrahepatic xenobiotic metabolism may be mediated via prostaglandin synthetase rather than cytochrome P-450 (12). However, no evidence for such a pathway existing in the esophageal mucosa could be obtained in the present study, since arachidonic acid was not found to support the metabolism of NMBzA.

Two important differences between hepatic- and esophageal mucosa-mediated metabolism of NMBzA were noted in this study. (a) The ratio of oxidation at the 2 α-carbons of NMBzA differs by 10-fold in the 2 tissues. (b) Hepatic metabolism of NMBzA at both α-carbons is induced by phenobarbitone pretreatment, in contrast to mucosal metabolism which is not altered. Since the liver contains multiple forms of cytochrome P-450, each having different, although broad, substrate specificities (10), the results of the present study suggest that the esophageal mucosa may contain only a subset of hepatic cytochromes P-450, different ratios of forms, or different form(s) altogether. Attempts to examine this question by subjecting the microsomes to sodium dodecyl sulfate-polyacrylamide gel electrophoresis and staining for heme-containing proteins have been unsuccessful to date, due to the low levels of cytochrome P-450 in the mucosa.

The present study thus demonstrates that rat esophageal mucosa contains an enzyme which can metabolize the esophageal carcinogen NMBzA at a high rate and shows properties typical of a cytochrome P-450. In contrast, NDMA, which is not an esophageal carcinogen, is a poor substrate for this enzyme. Furthermore, the esophagus is deficient in other typical carcinogen-metabolizing enzymes, such as aryl hydrocarbon hydroxylase (25, 26) and epoxide hydrlase (17). The enzyme system in the mucosa may thus play a crucial role in determining which chemicals are carcinogenic in the rat esophagus. Since rat liver can metabolize NMBzA and NDMA to yield methylating agents at similar rates (present study; Ref. 2), other factors must be responsible for the inability of NMBzA to induce liver tumors.

ACKNOWLEDGMENTS

The authors thank Dr. A. Howatson for performing the electron microscopy, Dr. K. Jain for assistance with the gel electrophoresis, and Kwan Leung and Louis Marai for performing the gas chromatography-mass spectroscopy.

REFERENCES

Esophageal and Hepatic Microsomal Metabolism of \( N \)-Nitrosomethylbenzylamine and \( N \)-Nitrosodimethylamine in the Rat

George E. Labuc and Michael C. Archer


Updated version
Access the most recent version of this article at:
http://cancerres.aacrjournals.org/content/42/8/3181

E-mail alerts
Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions
To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions
To request permission to re-use all or part of this article, contact the AACR Publications Department at permissions@aacr.org.