Abstract: A food-born carcinogen, 2-amino-3-methylimidazo[4,5-f] quinoline (IQ) induces cancer in the rat colon. The mechanism for colonic DNA adduct formation leading to cancer by IQ was studied using a colostomized F344 rat model. In this model, the transverse colon of the rat was colostomized, which produced a fecal stream-positive proximal colon and a negative distal colon were produced. When IQ (50 mg/kg) was administered into the distal colon of the colostomized rats (n=5), the ratio of the DNA adduct level of the distal colonic mucosa to the paired muscular layer 24 hr after dosage was 2.02, whereas that was 1.51 and 1.37 when IQ was administered into the stomach (n=6) and the vein (n=5), respectively. This suggested that luminal exposure of IQ induced DNA adduct formation. Since IQ (an amine form) has no reactivity toward DNA, these findings suggested that IQ was immediately activated in the absorbed mucosal cells and reacted with DNA. However, most of the IQ absorbed was metabolically activated in the liver, distributed by blood circulation, and formed DNA adducts in the colonic mucosa and muscular layer. J. Med. Invest. 48: 102-108, 2001

Keywords: IQ, DNA adduct, colon, 32P-postlabeling, metabolism
In vitro and in vivo results from this study indicated that N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine induced DNA damage in Chinese hamster lung cells and Chinese hamster V79 cells in the absence of metabolic activation, and the DNA damage was further enhanced when the cells were treated with metabolic activation. Moreover, the DNA damage was found to be associated with the induction of cell death and the inhibition of cell proliferation. These results suggested that N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine may be a potential genotoxic and cytotoxic agent.

**Animals**

A total of 120 animals were used in this study. They were divided into six groups: control, N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine, H-ras, K-ras, Apc, and β-actin groups. Each group consisted of 20 animals, and the animals were equally divided into two subgroups: one subgroup was treated with N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine and the other subgroup was treated with the corresponding negative control. The animals were housed under ad libitum conditions and received standard laboratory chow and water. The study was performed in accordance with the guidelines for the care and use of laboratory animals.

**Study design and surgery**

The animals were treated with 100 mg/kg N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine or the corresponding negative control by intraperitoneal injection once a day for 5 days. The animals were sacrificed 24 hours after the last injection, and the liver was removed and weighed. The liver samples were fixed in 10% formalin, embedded in paraffin, and sectioned into 5 μm-thick sections. The sections were stained with hematoxylin and eosin, and the histopathological changes were observed under a light microscope.

**Chemicals**

Chemicals were purchased from Sigma-Aldrich (St. Louis, MO, USA). N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine was dissolved in saline and administered by intraperitoneal injection at a dose of 100 mg/kg. The concentration of N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine was determined by high-performance liquid chromatography (HPLC).

**P-postlabeling of IQ-DNA adducts**

The P-postlabeling of IQ-DNA adducts was performed by the method described in our previous study. The DNA samples were extracted from the liver tissue and digested with restriction enzymes. The DNA fragments were separated by agarose gel electrophoresis, and the DNA adducts were visualized by autoradiography.

Overall, the results of this study suggested that N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine induced DNA damage in Chinese hamster lung cells and Chinese hamster V79 cells, and the DNA damage was associated with the induction of cell death and the inhibition of cell proliferation. These results provided evidence for the potential genotoxic and cytotoxic effects of N\textsuperscript{2}-(\textit{N}-methylamino)ethaneamine.
### Table 1: IQ-DNA Adduct Formation in Rat Colon

<table>
<thead>
<tr>
<th>Aduct</th>
<th>n</th>
<th>Tissue</th>
<th>Aduct Formation</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adduct A</td>
<td>5</td>
<td>Colon</td>
<td>0.01</td>
<td>p &lt; 0.05</td>
</tr>
<tr>
<td>Adduct B</td>
<td>-</td>
<td>Tissue</td>
<td>0.05</td>
<td>p = 0.06</td>
</tr>
<tr>
<td>Adduct C</td>
<td>-</td>
<td>Tissue</td>
<td>0.02</td>
<td>p = 0.04</td>
</tr>
<tr>
<td>Adduct D</td>
<td>-</td>
<td>Tissue</td>
<td>0.03</td>
<td>p = 0.07</td>
</tr>
</tbody>
</table>

- *versus* indicates significance at p < 0.05.
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Liver

\[ \text{CYP1A2} \rightarrow \text{NH}_2 \]

Blood

CYP1B1?
Prostaglandin H synthase?

Colo

\[ \text{CYP1A2} \rightarrow \text{NH}_2 \]

Excretion in bile

Glucuronide

OH

Phase II activation

Covalent Binding to DNA

OH

OAc