Mass Spectra of 3-Methylcholanthrene and Some Related Compounds

GONYA TAKAHASHI* AND AKIRA KATO**

(Received 19 July 1972)

Data are given of the mass spectra of 3-methylcholanthrene, one of the strongest carcinogenic hydrocarbons, and some related compounds. The structures of their fragments are shown speculatively.

1. Introduction

Many studies on the metabolism of polyaromatic hydrocarbons have been reported from interest in carcinogenicity. In the previous investigation¹⁾ on the metabolism of 3-methylcholanthrene (3-MC), one of the strongest carcinogenic hydrocarbons, we detected some metabolites of this chemical substance in the feces of mice. These compounds have been detected with conventional methods. Recently, a combining system of gas chromatography and mass spectrometry has been used widely in other studies but rarely in polyaromatic hydrocarbons. As a preliminary investigation of the detection of these compounds using this system, we have studied the mass spectrometry of 3-MC and some related compounds.

This paper gives mass spectral data of 3-MC and related compounds, and speculative structures of their fragments.

2. Results and Discussion

The mass spectra and speculative structures of the fragments are shown in Figs. 1 to 11. When the metastable ions

in fragmentation were observed, they were shown as m^* in these figures.

2.1 3-MC (Figs. 1 and 2)

The mass spectrum is very simple with a few fragment ions and the strong molecular ion which is the base peak. The peak at m/e 267 may be caused by the tropylium ion formed by ring expansion as shown by the fact that an analogous peak was found at m/e ($M-1$) for methyl acenaphthylenes.²⁾ In the higher mass range, one of the main peaks at m/e 253 (m^* 238.7, calc. 238.8) is due to an ion formed with loss of the methyl radical from the parent ion, and the other at m/e 252 (m^* 250.8, calc. 251.0) does from loss of the hydrogen radical. In the intermediate mass range all peaks are small, and in the lower mass range there are prominent series of doubly charged ion peaks.

2.2 1-Hydroxy-3-MC (Figs. 3 and 4)

The peak at m/e 284 is due to the molecular ion, the relative abundance of which increases when the energy of the electron beam is reduced to 12 eV. The base peak at m/e 266 (m^* 249.1, calc. 249.1) is due to mainly to an ion formed

* Chest Disease Research Institute, Kyoto University (Sakyo-ku, Kyoto, Japan).

** Faculty of Pharmaceutical Sciences, Kyoto University (Sakyo-ku, Kyoto, Japan).

Fig. 1. Mass spectrum of 3-methylcholanthrene.

Fig. 2. Speculative structures of fragments in 3-methylcholanthrene.

by dehydration from the parent ion. Ions containing tropilidene and dehydrotropilidene structures are apparent to recognize. The peaks at m/e 241 and 239 are probably due, respectively, to ions formed with loss of acetylene and ethylene from the cyclopentene ring of the cholanthrene structure. In the lower mass range, doubly charged ion peaks appear prominently.

2.3 2-Hydroxy-3-MC (Fig. 5)

The mass spectra of 1- and 2-hydroxy-3-MC show almost the same fragmentation pattern, except for some differences between relative abundance of peaks. The base peak is the molecular ion peak at m/e 284. The speculative fragmentation is the same as that of 1-hydroxy-3-MC.

2.4 1-Keto-3-MC (Figs. 6 and 7)

In the higher mass range, the molecular

Mass Spectra of 3-Methylcholanthrene and Some Related Compounds

Fig. 3. Mass spectrum of 1-hydroxy-3-methylcholanthrene.

Fig. 4. Speculative structures of fragments in 1-hydroxy-3-methylcholanthrene.

Fig. 5. Mass spectrum of 2-hydroxy-3-methylcholanthrene.

ion gives the strong peak at m/e 282. The peak at m/e 254 may be due to an ion containing tropilidene. The peaks at m/e 252 and 239 are probably due to ions containing dehydrocyclooctatetraene and dehydro-tropilidene, respectively. The peak at m/e 141 must be due to a doubly charged ion of the parent because there is a peak at m/e 141.5, which is due to a doubly charged ion of the $(M+1)$, and the ratio of the heights of m/e 141 to 141.5 is the same as the ratio of the parent ion to the $(M+1)$ ion.

2.5 1, 2-Dihydroxy-3-MC (Figs. 8 and 9)

The base peak is the molecular ion peak at m/e 300. The peak at m/e 282 (m^* 264.8, calc. 265.1) is due to an ion formed by dehydration from the parent ion. The peak at m/e 269 (m^* 241.0, calc. 241.2) is due to an ion formed with loss of the methoxy radical which was caused by rearrangement of the methyl radical into the 2-hydroxy radical. The tropilium and its benzyen type ions also appear at m/e 253, 239, 228. The peaks at m/e 228 (D, m/e 229) and 226 (without containing deuterium) are probably due,

respectively, to ions formed with loss of acetylene from one of the aromatic nuclei and cyclopentene.

2.6 11, 12-Dihydro-11, 12-dihydroxy-3-MC (Figs. 10 and 11)

As shown in Fig. 11, the three cases are possible in rearrangement of the deuterium-labeled ions. The peak at m/e 302 is due to the molecular ion. The base peak at m/e 284 (m^* 267.1, calc. 267.1) is due to an ion formed by dehydration from the parent ion. The peak at m/e 269 (m^* 254.7, calc. 254.8) is due to loss of the methyl radical from the fragment at m/e 284. The peak at m/e 241 (D, m/e 242; m^* 215.6, calc. 215.9) is due to an ion containing fluorene which is formed with loss of CO from the hydroxy group of the fragment at m/e 269 (D, m/e 270). It is impossible to recognize an ion containing biphenylene arising from fluorene as shown in Fig. 12. The peak at m/e 242 is expected to be an ion of biphenylene derivative which is formed with loss of ethylene diol, and it decomposes finally to the tropilium ion and its benzyen type ion, taking their places at m/e 239 and 226.

Mass Spectra of 3-Methylcholanthrene and Some Related Compounds

Fig. 6. Mass spectrum of 1-keto-3-methylcholanthrene.

Fig. 7. Speculative structures of fragments in 1-keto-3-methylcholanthrene.

Fig. 8. Mass spectrum of 1,2-dihydroxy-3-methylcholanthrene.

Fig. 9. Speculative structures of fragments in 1, 2-dihydroxy-3-methylcholanthrene.

Fig. 10. Mass spectrum of 11,12-dihydro-11, 12-dihydroxy-3-methylcholanthrene.

Mass Spectra of 3-Methylcholanthrene and Some Related Compounds

Fig. 11. Speculative structures of fragments in 11, 12-dihydro-11, 12-dihydroxy-3-methylcholanthrene.

Fig. 12. Mass spectrum and speculative structures of fragments of fluorene.

3. Experimental

3.1 Apparatus

Mass spectra were measured by a modified Hitachi RMU-6C mass spectrometer equipped with the direct inlet system. Ionizing voltage: 70 eV; total emission: 80 μ A; ion source temperature: 200°C.

3.2 Reagents

3-MC was obtained from Fluka (CH-9470 Buchs, Schwizerland).

1-Keto-3-MC was prepared by dichromate oxidation of 3-MC by the method of Fieser *et al.*³⁾ The crude product was purified and finally crystallized from ether to yield yellow needles, m. p. 261.7°C, according to the method of Sims.⁴⁾

1-Hydroxy-3-MC—1-Keto-3-MC was reduced with lithium tetrahydroaluminate according to Sims.⁴⁾ The crude product was purified by chromatography on silica

gel in benzene and crystallized from ethanol to yield 1-hydroxy-3-MC white needles, m. p. 210.5°C (decomp.).

1, 2-Dihydroxy-3-MC was prepared by acetylation of 3-MC by lead tetraacetate in hot acetic acid and by hydration of the product by potassium hydroxide according to the method of Sims.⁴⁾ The crude product was chromatographed in benzene-acetone (9:1) on silica gel. The two violet fluorescent bands were evaporated and crystallized from ethanol to yield *cis*- and *trans*-1, 2-dihydroxy-3-MC both in microscopically visible brown needles. The melting point was uncertain because of decomposition into 2-keto-3-MC at 190–200°C. The substance obtained from the fast moving band was proved to be the *cis* type from production of the isopropylidene derivative with acetone, and the one obtained from the second band was proved to be the *trans* type from no

production of this derivative. In our experiment the *trans* type was used.

2-Hydroxy-3-MC—According to the method of Sims,⁴⁾ 2-hydroxy-3-MC was obtained by the reduction of 2-keto-3-MC in the same manner as preparation of 1-hydroxy-3-MC from 1-keto-3-MC. The melting point of this product was uncertain because it seemed to change to another compound at 180°C which melted at 219.6°C (decomp.).

11, 12-Dihydro-11, 12-dihydroxy-3-MC was prepared by OsO₄ oxidation according to the description by Cook and Schoental.⁵⁾ The crude product was purified by chromatography on silica gel in benzene-acetone (9:1) and was crystallized from ethanol to yield 11, 12-dihydro-11, 12-dihydroxy-3-MC in white powder which turned to red at 190°C and melted at 202°C (decomp.).

The deuterium-labeled compounds were obtained by mixing deuterium oxide and the chemicals.

All compounds prepared were identified with the standards described by Sims⁴⁾ by UV absorption spectra and *R_f* values of thin layer chromatography on silica gel, and their purities were confirmed by disappearance or decrease of the peaks except for the molecular ion when the energy of the electron beam is reduced to 12 eV.

References

- 1) G. Takahashi and K. Yasuhira, *Cancer Res.*, **32**, 710 (1972).
- 2) I. D. Entwistle and R. A. W. Johnstone, *J. Chem. Soc.*, **1968**, 1818.
- 3) L. F. Fieser and E. B. Hershberg, *J. Am. Chem. Soc.*, **60**, 2542 (1938).
- 4) P. Sims, *Biochem. J.*, **98**, 215 (1966).
- 5) J. W. Cook and R. Schoental, *J. Chem. Soc.*, **1948**, 170.