IDENTIFICATION OF PRODUCTS OF THE INITIATED OXIDATION OF AMIDOPYRINE BY CHROMATO-MASS SPECTROMETRY

S. T. Kozlov, T. V. Ushakova, L. F. Sokolova, and V. I. Gol'denberg

It was shown previously [1] that amidopyrine and dipyrone (Analgin) are acceptors of peroxy radicals and may be used as antioxidants for the stabilization of drugs.

The oxidation products of amidopyrine have been investigated in the present work by chromato-mass spectrometry with the aim of studying further the mechanism of the reaction.

EXPERIMENTAL

The initiated oxidation reaction of amidopyrine in methyl ethyl ketone was carried out in the presence of azoisobutyronitrile as donor of peroxy radicals on bubbling air through them at a temperature of 60°C. The kinetics of the oxidation were followed by the change in intensity of the chemiluminescence [2]. Test samples for analysis were taken after initiation of the oxidation process from the induction period in the presence of added amidopyrine which corresponded to complete conversion of amidopyrine in the reaction [1].

Experimental investigations of the oxidation products of amidopyrine were carried out on an LKB 2091 chromato-mass spectrometer (Sweden).

The operating parameters of the mass spectrometer were: accelerating voltage, 3.5 kV; energy of ionizing electrons on recording the mass spectrum, 70 eV, and on recording the chromatogram, 20 eV; emission current, 50 μ A; ion source temperature, 250°C, separator, 250°C.

The conditions of gas chromatographic analysis were: borosilicate glass column of length 150 cm and internal diameter 2 mm packed with diatomite adsorbent with 2% SE-30; carrier gas, helium, 30 ml/min; column temperature, 200°C; evaporator, 250°C.

RESULTS AND DISCUSSION

Figure 1 shows a chromatogram of the products of the initiated oxidation of amidopyrine. It is seen that three main compounds (1), (2), and (3) are formed as a result of the reaction. A chromatographic peak of amidopyrine taken under identical conditions is shown graphically with dotted lines. Mass spectra were recorded corresponding to the maxima of chromatographic peaks. The mass spectrum of amidopyrine (Fig. 2A) was studied in order to interpret the mass spectra of the peaks being analyzed. The presence of five intense lines of m/e 56, 97, 111, 112, and 231 was a characteristic of the spectrum. The molecular ion of m/e 231 taken as 100% was the most intense line. Peaks were also present in the spectrum at m/e 72, 121, 122 which were also observed in the mass spectra of the substances being analyzed.

The mass spectrum of the compound corresponding to chromatographic peak (1) is shown in Fig. 2B. The spectrum was characterized by seven sufficiently intense peaks of m/e 43, 92, 93, 107, 121, 122, and 164.

Let us consider the region of molecular ions. It follows from the spectrum that the ions with m/e 164 and 165 are the ions with the highest mass numbers. The ion peak of m/e 165 has an insignificant intensity and is a carbon isotope of the ion of m/e 164. The suggestion that the peak of m/e 164 is an ion of mass $[M-1]^+$ formed as a result of ion-molecular reactions is not very likely [3].

The molecular mass of compound (1) is therefore 164. The intensity of the ¹³C isotopic peak of m/e 165 was 9.4% of that of the molecular ion of m/e 164. The natural content of

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Fig. 2

Fig. 1. Chromatogram of the mixture of oxidation products (1, 2, 3) of amidopyrine (4).

Fig. 2. Mass spectra of A) amidopyrine and of its oxidation products; B) (1); C) (2); D) (3).

the ¹³C isotope is 1.08%; therefore, 9.4:1.08 = 8.7. Thus compound (1) may contain nine carbon atoms [4]. The substance has an even-numbered molecular weight and must therefore have an even number of nitrogen atoms [5]. The suggestion that there are four nitrogen atoms in the molecule is not very likely.

The presence in the spectrum of peaks of m/e 77 and 65 indicates the aromatic character of the compound. The intense peak of m/e 43 $[CH_3CO]^+$ and the base peak of m/e 121 $[M-43]^+$ formed by elimination of the CH₃CO particle indicated the presence of an acetyl group in the compound. The peak of m/e 92 is the $[C_6H_5-NH]^+$ ion [4].

Proceeding from the above and in view of the structure of the initial reaction products it has been proposed that the reaction product being investigated has the structural formula

 $C_8H_5 - NH - N(CH_8) - CO - CH_8$.

The formation of all the characteristic peaks in the mass spectrum and the corresponding routes of fragmentation are readily explained from the assumed structure. The peak of m/e 122 is formed by migration of a hydrogen atom from acetyl and subsequent detachment of a COCH₃ particle from the molecular ion which is typical of N-acetyl compounds [6]. Subsequent detachment of a methyl group (M-COCH₂-CH₃) led to the ion of m/e 107.

The mass spectrum of substance (2) is shown in Fig. 2C. Its molecular weight proved to be 192. If the mass spectra of substances (1) and (2) are compared it is possible to see much similarity. However, compound (2) has a molecular mass 28 units greater which corresponds to a CO group in the case in question. The obtained mass spectral data indicate that the most probable structure of compound (2) is

$$C_{6}H_{5} - N(CHD) - N(CH_{3}) - CO - CH_{3}$$

Detachment of a CO particle from the molecule leads to the formation of an ion of m/e 164, the subsequent breakdown of which is analogous to that of compound (1) and is evident from both mass spectra.

The mass spectrum of compound (3), shown in Fig. 2D, is characterized by the presence of five intense lines of m/e 72, 121, 191, 221, and 235. The molecular weight is 263. The intensity of the molecular ion peak was small, which indicates the weak stability of the molecule towards electron impact. This phenomenon may be explained by the presence in the molecule of substance (3) of a sufficiently long unconjugated chain. The mass difference between the molecules of amidopyrine and its derivative is 32, i.e., the mass of two oxygen atoms. The intense peak of m/e 72 corresponds to a $[(CH_3)_2N-CO]^+$ ion [7]. It may be proposed that in the course of the initiated oxidation of amidopyrine a peroxy compound is formed at the double bond with subsequent cleavage of the 0-0 bond.

On the basis of the available data the following structure is proposed for compound 3:

 $C_{6}H_{5} - N$ $C_{6}H_{5} - N$ CO - C = O CO - C = O $N (CH_{3})_{2}$

The formation of all the characteristic peaks of the mass spectrum is in good agreement with the proposed structural formula.

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