Effects of Radical Scavenger and Water on Radiolysis Reaction Velocity of C₁₈ Unsaturated Fatty Acids

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Using C₁₈-unsaturated fatty acids such as methyl oleate, methyl linoleate and methyl linolenate, effect of radical scavenger, antioxidant and water on radiolysis reaction velocity of these substances were investigated.

It was found that the presence of radical scavenger or antioxidant suppress radiolysis of unsaturated fatty acids, on the other hand the presence of water accelerated the reaction.

The velocity constant of the chemical reaction obtained from the ratio of peak dimension of gas chromatograms was the secondary reaction in the former, the primary reaction in the latter; in the both case the constant was found to be proportional to the square of the number of the double bonds in unsaturated fatty acids.

This phenomenon can be explained based on dynamics of chemical reaction.

1. Introduction

In a previous paper the author reported¹⁾ that a mixture of methyl oleate, methyl linoleate, and methyl linolenate was irradiated with over 10⁶R dose in oxygen-free state and the result was examined by gas chromatography. The decomposition rate constant of these unsaturated fatty acids by radioactive rays was calculated as the primary reaction from the result of gas chromatographic analysis and they underwent decomposition in proportion to the square of the number of double bonds in the molecule.

The present paper reports effect of radical scavenger, antioxidants and water on radiolysis reaction velocity of these unsaturated fatty acids.

2. Method

Mixture of highly pure methyl oleate, methyl linoleate and methyl linolenate used as internal standard for methyl myristate, was added with DPPH (1,1-diphenyl -2-picrylhydrazyl) as a radical scavenger, and BHA (3-tert. -butyl-4-hydroxyanisole) as an antioxidant of the unsaturated fatty acids.

The concentration of addition was approximately 10%. These unsaturated fatty acid esters were manufactured by Applied Science Laboratories and result of analysis using gas chromatography showed their fineness was 99%.

Also small amount or large amount of water was added in addition. Since unsaturated fatty acids change themselves rapidly with contact to oxygen, these mixtures were placed into the small glass tubes, and then they were closed in the oxygen free state with vacuum pump. The gamma ray irradiation was performed under the same condition.

A 15 kCi-Co-60 (cylindrical form) in Japan

Atomic Energy Research Institute was used as a radiation source and the mixtures irradiated with the dose of $1.1 \times 10^8 \text{R}$, $1.5 \times 10^8 \text{R}$, $2.03 \times 10^8 \text{R}$, $2.5 \times 10^8 \text{R}$ and $3.17 \times 10^8 \text{R}$ respectively placing 3 cm apart from the radiation source in the cone as the condition of irradiation. After irradiation the mixtures were analyzed with gas chromatography and the ratio of peak dimension was calculated.

3. Results

3.1 Gas chromatography

Gaschromatogram of the mixture of the previously stated unsaturated fatty acids containing BHA, after they were irradiated with the radiation dose of $3.17 \times 10^8 \text{R}$ is shown in Fig. 1.

Fig. 1 Gaschromatograms of MOLLnS containing BHA.

In the area of low retention time small peaks of dissociated products were observed, but addition of DPPH or water to the mixture did not show any difference of peaks.

The ratio of peak dimension were different depending on each case. Result of analysis is shown in Table 1 while irradiation was carried out with the dose of $3.17 \times 10^8 \text{R}$ and the peak dimension of non-irradiated occasion was set as 1.

It is clear from the Table 1 that the presence of DPPH and BHA hindered the dissociation

Table 1 Rate of peak dimension of gaschromatograms under $^{60}\text{Co}-\gamma$ irradiation with the dose of $3.17 \times 10^8 \text{R}$

	MO/C ₁₄	ML/C ₁₄	MLn/C ₁₄
MOLLnS	0.878	0.524	0.296
" + H ₂ O (small amount)	0.840	0.418	0.218
" + H ₂ O (large amount)	0.834	0.415	0.220
" + DPPH	0.892	0.692	0.545
" + BHA	0.908	0.700	0.543

C₁₈/C₁₄; non-irradiated occasion was set as 1.

MO; methyl oleate

ML; methyl linoleate

MLn; methyl linolenate

MOLLnS; Mixture of MO, ML, MLn and methyl myristate (C₁₄).

of the unsaturated fatty acids by irradiation, while the presence of water accelerated it.

In case of the anoxic state, the rate of radiolysis of methyl linolenate was 70%, 45%, respectively in the presence of DPPH and BHA, and 78% with the presence of water.

3.2 Velocity of radiolysis reaction

When the velocity of radiolysis reaction was calculated from the quantity of each dose of radiation while the exposure time of radiation with the dose $1 \times 10^8 \text{R}$ was set to be 1, it was found that in the cases of the unsaturated fatty acids alone and with the presence of water, the constant of chemical reaction was that of primary reaction and in the cases of the presence of DPPH or BHA, the constant was that of secondary reaction. The mean of the constants is shown in Table 2.

Table 2 Rate constants of radiolysis reaction velocity

	order	MO	ML	MLn
MOLLnS	k_1	0.0432	0.204	0.384
" + H ₂ O (small amount)	k_1	0.0558	0.252	0.495
" + H ₂ O (large amount)	k_1	0.0570	0.255	0.490
" + DPPH	k_2	0.0331	0.141	0.264
" + BHA	k_2	0.0325	0.135	0.266

$1 \times 10^8 \text{R}$ irradiation time was set to be 1.

k ; mean value ($1.1 \times 10^8 \sim 3.17 \times 10^8 \text{R}$)

4. Discussion

Radiolysis reaction of C_{18} unsaturated fatty acids is markedly suppressed with the addition of radical scavenger and antioxidant. This is thought to occur due to consumption of free radical by radical scavenger or antioxidant, resulting in interruption of the chain of chemical reactions. As a matter of fact a decrease of the peak of BHA was seen with gamma ray irradiation.

On the other hand, the presence of water accelerates radiolysis reaction of the unsaturated fatty acids regardless of quantity of water, this is thought to occur because ions or radicals take part in the reaction thereby reacting with the unsaturated fatty acids.

The ratio among the constants of reaction velocity of methyl oleate, methyl linoleate, methyl linolenate were approximately 1:4:9, and it was found that they are in proportion to the square of the number of the double bonds in the molecule.

This is considered to be so because radicals of adjacent molecule reacts to each other between themselves, and so the velocity of the reaction is proportional to the multiplication of the concentration of radicals.

In living body there are many oxygen, and this kind of experiment is desired to perform in case of close contact with oxygen, but it is impossible that the radiation source is too small and so requiring longer period of irradiation time in a dose of over 10^8R , for the unsaturated fatty acids are gradually oxidized even in non-irradiated period.

At any rate the velocity of reaction will be much faster in the normal condition than in the anoxic condition, and the order of chemical reaction or the ratios of chemical velocity will be influenced very little.

Reference

- 1) Kitahara, K., Tanaka, Y. and Inoue, F.: *Yakugaku Zasshi*, **85**, 902 (1965)

要 旨

C_{18} 不飽和脂肪酸の放射線分解反応速度に及ぼす

ラジカル捕捉剤、水などの影響

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不飽和脂肪酸として、オレイン酸、リノール酸、リノレン酸の各メチルエステルを用いて、コバルト 60 ガンマ線を 10^8R 以上大線量照射した場合の放射線分解反応速度に及ぼすラジカル捕捉剤 (DPPH)、脂肪酸酸化防止剤 (BHA) および水の影響をしらべた。

その結果、ラジカル捕捉剤、酸化防止剤は不飽和脂肪酸の放射線分解を抑制し、水は促進する。ガスクロマトグラムのピーク面積比から求めた反応速度定数は、前者は二次反応として、後者は一次反応として、いずれの場合も不飽和脂肪酸の二重結合の数の 2 乗に比例することがわかった。これらのことは反応動力学的に説明できる。