Assessment of Oxidative Deterioration of Salted Dried Fish by Nuclear Magnetic Resonance

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Nuclear magnetic resonance (NMR) was used to evaluate the oxidative deterioration of oil in salted dried fish. The ratios of olefinic protons (Ro) and divinylmethylene protons (Rm) to aliphatic protons in the oil of salted dried fish, as determined by NMR, decreased steadily during storage. On comparing Ro and Rm with data for peroxide value (PV) and acid value, Ro was shown to serve as an index of the oxidative deterioration of the oil of salted dried fish, particularly in the case of oil whose PV had decreased.

KEY WORDS: Acid value, carbonyl value, dried products, fish oil, lipid oxidation, nuclear magnetic resonance (NMR), olefinic proton, oxidative deterioration, peroxide value, salted dried fish.

Recent epidemiological, clinical and nutritional studies on animals and humans have shown marine fish oils, rich in polyunsaturated fatty acids of the n-3 series, to be useful for reducing the risk of coronary heart disease and atherosclerosis, as well as preventing certain forms of cancer. With growing recognition of consumers and industry of the beneficial uses of dietary fish oil, the seafood market is expanding considerably (1–3).

Fatty fish, such as mackerel, is an important source of dietary fish oil and is generally preserved by salting and drying after being caught. Salted dried fish has been consumed for generations, and constitutes an important part of diets of populations in Japan and developing countries in Southeast Asia.

The oil in dried fish products is highly susceptible to atmospheric oxidation, because it contains high proportions of n-3 polyunsaturated fatty acids, and the salt may act as a catalyst of oil oxidation (4). Some oxidation products of lipids are toxic, and cured products obtained from the marketplace are occasionally unsatisfactory. An appropriate method for assuring satisfactory quality of salted dried fish has long been needed. However, such methods are not presently available. Peroxide value (PV) and acid value (AV), currently are used as quality indices (5,6).

PV and AV are useful indicators of the deterioration status of vegetable and animal oils but are not always useful for marine lipids because PV increases at the initial stage of oxidative deterioration, peaks, and then decreases during storage. AV does not change much in dried fish during storage, and therefore is not useful for assessing oil deterioration.

According to a previous study (7), the ratios of olefinic protons (Ro), (δ 5.1–5.6 ppm) and divinylmethylene protons (Rm) (δ 2.6–3.0 ppm) to that of aliphatic protons (δ 0.6–2.5 ppm) in nuclear magnetic resonance (NMR) decrease continuously during oxidative reaction. Thus, the ratios should be useful for measuring oxidative deterioration of fish oil, even after PV has reached a maximum value. NMR for evaluating oxidative deterioration of several different processed fish products has been studied, and the results are discussed.

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MATERIALS AND METHODS

Scomber japonicus, Trachurus japonicus and *Cololabis saira*, commonly called mackerel, Japanese horse mackerel and Pacific saury, respectively, were purchased fresh from a market at Tsukiji in Tokyo, Japan. The fish were cleaned and filleted prior to salting. The fillets were immersed in 20% brine for 15 min at 25°C. After the water was drained off, the salted fillets were air-dried at 35°C for 3 h. Ordinary salted dried fish (mackerel, horse mackerel and saury) on the market were purchased from retail outlets at Tsukiji and used for the storage experiment.

Storage conditions. The experimental models and ordinary market products were allowed to stand at −10 and −5°C, respectively, and oil in each sample was extracted with a mixture of chloroform and methanol (2:1) according to the procedure of Folch *et al.* (8).

Analytical methods. PV and AV were determined by *The Official and Tentative Methods of the American Oil Chemists' Society* (5,6). Carbonyl value (CV) was determined according to Kumazawa and Oyama (9). Spectra were recorded with a Jeol GSX-270 NMR spectrometer (Japan) in the pulsed Fourier transform mode at 270 MHz in deuterochloroform solution. Tetramethylsilane was the internal standard (frequency, 3001.2 Hz; resolution, 0.18 Hz; pulse, 90°). A sample solution containing oil (about 20.0 mg) from salted dried fish in deuterochloroform was kept in a 5-mm NMR sample tube, and spectra were obtained from a total of 16–64 readings of 10-s pulse delays for accurate equalization and integration. Because the concentration of fish oil (20.0 mg in 0.4 mL of deuterochloroform) was high, the noise signal was negligible. Error in integral calculation was below 0.2% for each Ro, even though integration was done more than ten times.

RESULTS AND DISCUSSION

The PV of oil of salted dried fish (experimental models) was periodically measured during one year of storage at −10°C. As shown in Figures 1–3, PV of oil extracted from each salted dried fish (mackerel, horse mackerel and saury) increased gradually during the first 200 days of storage, reached a maximum (about 600–700 meq/kg) and then gradually decreased. PVs of the oil of salted dried mackerel were 48.3 meq/kg at 0 d, 389.0 meq/kg at 72 d, 586 meq/kg, maximum value, at 132 d, and 456.7 meq/kg at 377 d of storage.

Ro and Rm values steadily decreased. Ro values were 13.43% at 0 d, 10.92% at 72 d, 9.21% at 132 d and 7.00% at 377 d, showing steady decreases with time. Thus, Ro reflects oxidative deterioration of fish oil in dried fish.

As shown in Figure 4, the change in AV of the oil of the ordinary market products was too small to permit assessment of the oil deterioration. The AV reflects the amount of fatty acids, which were mainly enzymatically

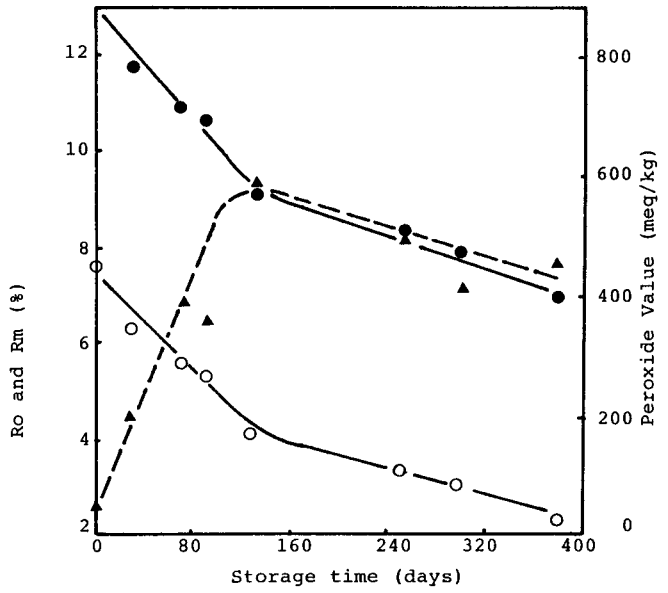

FIG. 1. Changes in the ratios of olefinic protons (Ro) and divinylmethylene protons (Rm) to aliphatic protons, and peroxide value (PV) for the experimental model (dried mackerel) during storage at −10°C. ●, Ro; ○, Rm; and ▲, PV.

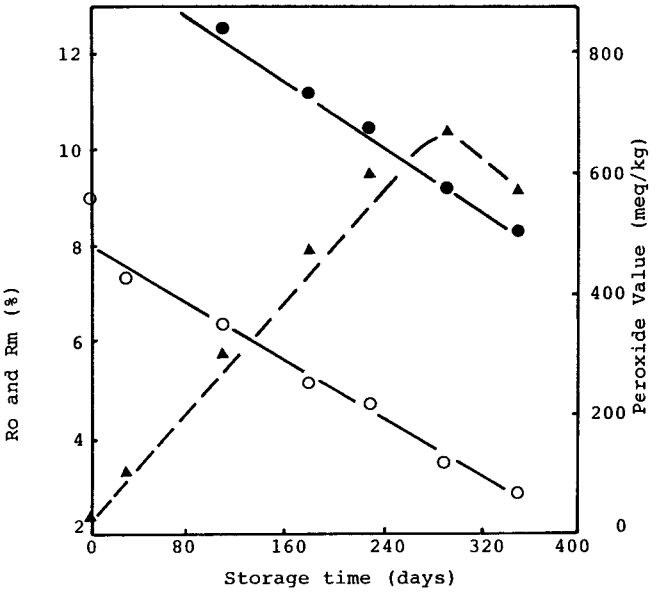

FIG. 3. Changes in the ratios of olefinic protons (Ro) and divinylmethylene protons (Rm) to aliphatic protons, and peroxide value (PV) for the experimental model (dried saury) during storage at −10°C. ●, Ro; ○, Rm; ▲, PV.

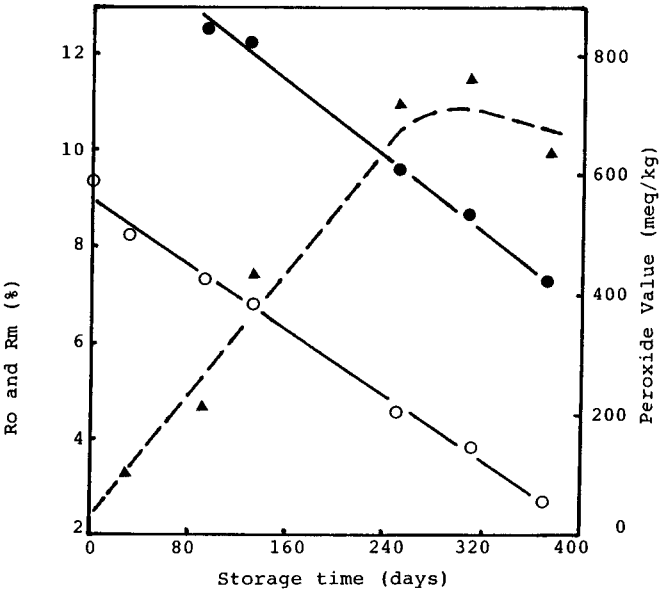

FIG. 2. Changes in the ratios of olefinic protons (Ro) and divinylmethylene protons (Rm) to aliphatic protons, and peroxide value (PV) for the experimental model (dried horse mackerel) during storage at −10°C. ●, Ro; ○, Rm; and ▲, PV.

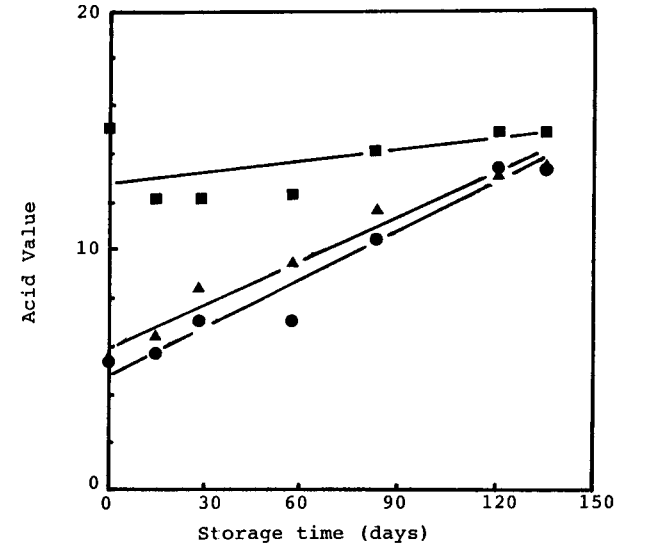

FIG. 4. Changes in acid value for ordinary market products during storage at −5°C. ●, Mackerel; ▲, horse mackerel; and ■, saury.

hydrolyzed. However, enzymatic hydrolysis does not proceed rapidly at low temperatures. Figures 5–7 show variation in PV of the three ordinary market products to be essentially the same as that observed with the experimental models (Fig. 1–3). Ro and Rm of oil of the market products decreased linearly with storage time as in the experimental models. The data for the two experiments indicate that the increase in PV does not always reflect

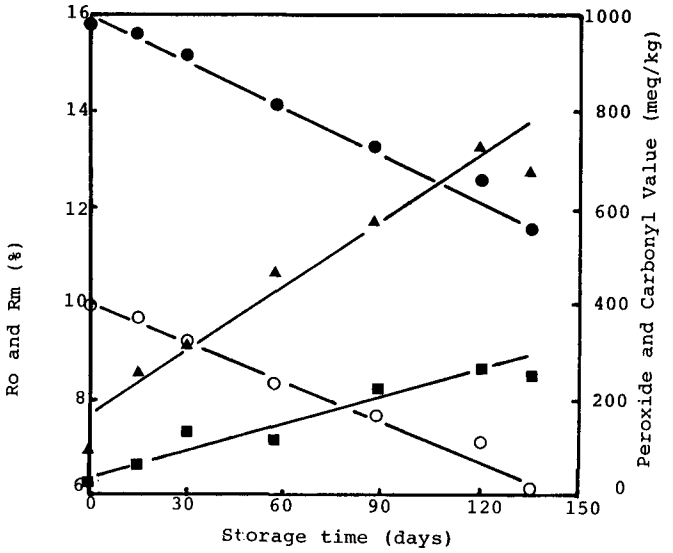

FIG. 5. Changes in the ratios of olefinic protons (Ro) and divinylmethylene protons (Rm) to aliphatic protons, and peroxide value (PV) and carbonyl value (CV) for ordinary market products (dried mackerel) during storage at −5°C. ●, Ro; ○, Rm; ▲, PV; and ■, CV.

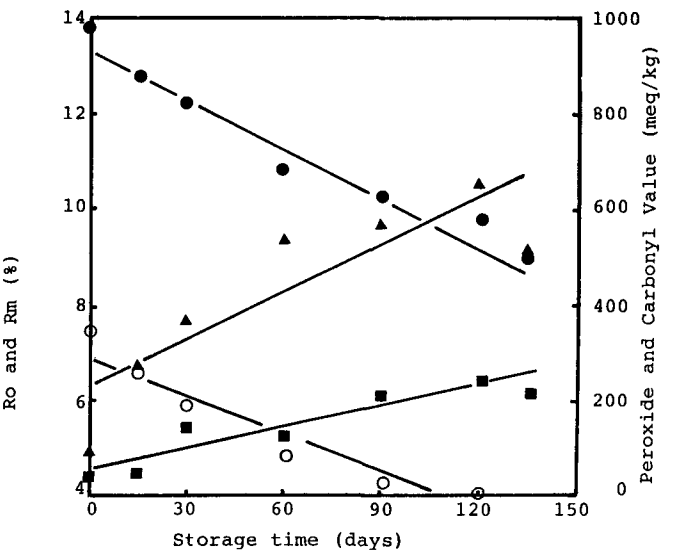

FIG. 7. Changes in the ratios of olefinic protons (Ro) and divinylmethylene protons (Rm) to aliphatic protons, and peroxide value (PV) and carbonyl value (CV) for ordinary market products (dried saury) during storage at −5°C. ●, Ro; ○, Rm; ▲, PV; and ■, CV.

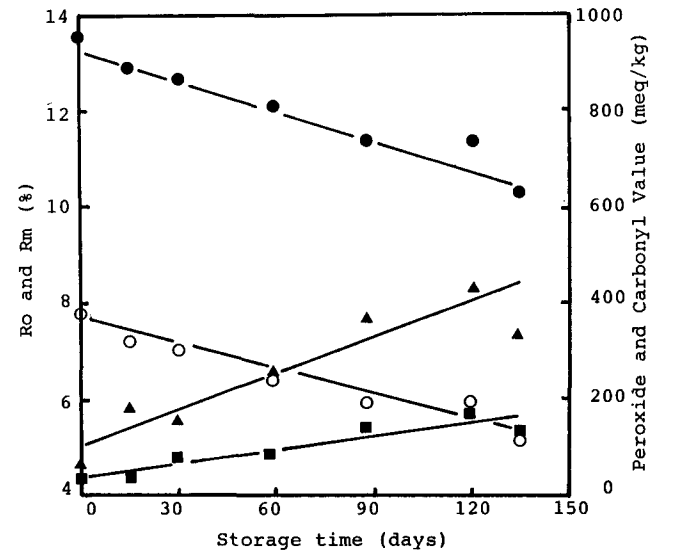

FIG. 6. Changes in the ratios of olefinic protons (Ro) and divinylmethylene protons (Rm) to aliphatic protons, and peroxide value (PV) and carbonyl value (CV) for ordinary market products (dried horse mackerel) during storage at −5°C. ●, Ro; ○, Rm; ▲, PV; and ■, CV.

the progress of oil deterioration of dried fish. A simple PV measurement may possibly lead to error in judging the quality of oil in dried fish.

Although the applicability and imitations of our NMR method for the evaluating of oil deterioration in many types of dried marine products should be examined further, it appears promising.

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[Received March 3, 1992; accepted July 29, 1992]