FATTY ACID COMPONENTS OF BLACK RIGHT WHALE OIL BY GAS CHROMATOGRAPHY

HIDEO TSUYUKI* AND SHINGO ITOH*

INTRODUCTION

There have been a number of studies on whale oil. However, there are a few studies on black right whale oil. As to the study on black right whale oil, we can find the following reports; Studies on the oil of black right whale in the Northern Pacific Ocean (Tsuyuki and Naruse, 1963), Studies on the lipids in brain of black right whale in the Northern Pacific Ocean (Tsuyuki and Naruse, 1964), Studies on liver oil of black right whale (Tsuyuki, Naruse, Mochizuki and Itoh, 1964).

So far as these works concerned with chemical properties of oils and fatty acid components by fractional distillation, minute examinations of fatty acid components have not been reported yet.

So the purpose of the present work is a minute examination of the fatty acid components of black right whale oil by gas-liquid chromatography using a hydrogen ionization detector.

It is pleasure that we express here our thanks to Dr. H. Omura and Dr. S. Ohsumi in the Whales Research Institute who were kind enough to present us black right whale oils.

MATERIAL AND METHOD

Sample used

Material is the black right whale, *Eubalaena glacialis*, (male, body length: 17.1 m, presumed age: more than 12 years old) which was caught at the southern sea of Kodiak Island in the Northern Pacific Ocean in 1962 (Tsuyuki and Naruse, 1963).

The oils contained in 9 parts of blubbers were obtained by boiling these blubbers with water and those in 3 kinds of organs were extracted with acetone in an atmosphere of nitrogen gas. The oils used in the present report were stocks used in previous reports (Tsuyuki and Naruse, 1963, 1964). The bottles of sample oils have been filled up with nitrogen gas and preserved in refrigerator at low temperature.

Used sample oils are the oils contained in 3 kinds of organs (stomach, liver and tongue), and 9 parts of blubbers (middle back, thoracic, abdominal, brain, hind part of blow-hole, umbilicus, anterior abdominal, fore part of genital aperture and posterior back). The properties of 9 blubber oils and 3 organ oils are shown in Table 1.

^{*} Department of Food Engineering, College of Agriculture & Veterinary Medicine, Nihon University.

Kinds of blubber and organ	Acid value	Saponification value	Iodine value	Unsaponifiable Matter (%)
Stomach	0.8	192.6	131.5	1.02
Liver	2.5	189.5	131.4	1.45
Tongue	2.1	195.4	115.1	1.18
Middle back blubber	1.2	196.6	126.6	0.77
Thoracic blubber	1.1	191.8	123.8	1.05
Abdominal blubber	0.7	187.6	130.5	0.94
Brain blubber	1.1	182.4	134.0	0.80
Blubber of hind part of blow-hole	1.5	194.1	127.1	1.14
Umbilicus blubber	1.6	190.4	128.6	1.05
Anterior abdominal blubber	1.6	190.7	134.9	0.80
Blubber of fore part of genital aperture	1.9	193.7	132.6	0.91
Posterior back blubber	1.8	190.0	129.7	0.70

TABLE 1. PROPERTIES OF THE OILS CONTAINED IN VARIOUS BLUBBERS AND ORGANS OF BLACK RIGHT WHALE

Preparation of methyl ester of fatty acid

The methyl esters of the fatty acids of sample oils were prepared by a semimicro methanolysis adapted to the method of Gauglitz and Lehmann (1963) for use in gas-liquid chromatography analysis. Then, to remove colesterol, coloring materials, impurities and others, the methyl esters were refined by passing through in glass column packed with silicic acid.

Gas-liquid chromatography

The methyl esters of the fatty acids of the black right whale oils were analyzed with a Shimadzu Gas Chromatograph Apparatus, Model GC-1 C. The instrument was equipped with a hydrogen ionization detector. The column used was composed of 3 mm in diameter by 225 cm U shaped stainless steel column containing 20% diethylene glycol succinate polyester (DEGS from Shimadzu Seisakusyo Co.) supported on 60-80 mesh Shimalite. Operating conditions were as follows; column temp. 210°C, injector temp. 260°C, detector temp. 240°C. A column inlet pressure of 1.80 kg/cm² N₂ was used, which measured 70-74 ml/min. at the flow rate.

Each peak was identified by comparing retention time with those in a known mixture of standard fatty acid methyl esters (C_{10} to C_{24} as saturated fatty acids, and C_{16} monoenoic acid and C_{18} mono-, di- and trienoic acids as unsaturated fatty acids), and semilogarithmic plots of carbon number vs relative retention time were used for identification by the method of Nelson and Freeman (1960). Also, for the purpose of identification of odd carbon chain length fatty acids, the hydrogenation was operated as follows; the *n*-hexanate solution of methyl esters was added a pinch of palladium on activated carbon as a catalyst. The mixture was shaken in a small flask for 5–6 hours under hydrogen atmosphere (approximate 1.5 kg/cm²) at room temperature, and then filtered. The hydrogenated methyl esters were operated by gas chromatograph at the same condition.

	Posterior back blubber		0.82	0.28	0.12	6.10	2.04	1.06	0.11	5.20	6.75	1.79	1.05	1.63	18.50	1.72	2.37	1.50	1.53	20.36	1.66	1.94	2.32	16.76	0.98	1.11	2.30
·	Blubber of fore part of genital aperture		0.37	0.17	trace	8.11	0.72	0.34	0.13	6.96	5.03	1.30	1.40	1.54	19.56	1.41	1.99	1.17	1.63	20.90	1.34	1.50	2.21	15.17	1.91	1.26	3.88
S	Anterior abdomi- nal blubber		0.78	0.21	0.12	6.93	1.30	1.08	0.17	6.49	5.81	2.53	1.07	1.15	18.51	1.75	2.18	0.93	1.26	18.91	1.22	2.54	1.76	16.94	1.41	1.89	3.06
VARIOU	Umbilicus blubber		0.81	0.26	0.03	5.83	0.82	0.46	0.33	6.50	4.57	16.1	1.06	1.19	17.96	2.01	2.44	0.97	1.37	21.26	0.97	1.02	1.10	18.67	1.05	3.15	4.26
FATTY ACID COMPONENTS OF OILS CONTAINED IN VARIOUS BLUBBERS AND ORGANS OF BLACK RIGHT WHALE	Blubber of hind part of blow-hole	y acids	0.33	0.15	trace	5.24	0.38	0.45	0.19	5.95	3.60	1.13	1.03	1.81	21.55	1.16	1.61	1.15	2.31	21.15	1.18	1.37	2.01	18.47	2.62	1.37	3.79
ATTY ACID COMPONENTS OF OILS CONTAINED IN BLUBBERS AND ORGANS OF BLACK RIGHT WHALE	Brain blubber	Weight per cent of total fatty acids	0.66	0.53	0.14	6.61	0.76	0.51	0.09	8.37	7.04	1.45	1.36	1.26	12.61	1.64	2.43	1.39	1.25	18.23	1.22	0.57	1.46	17.66	1.06	1.78	4.92
TS OF OI OF BLAC	Abdomi- nal blubber	per cent o	0.54	0.26	trace	5.76	0.68	0.65	0.35	6.26	5.15	1.12	0.81	1.14	21.26	1.30	1.32	0.65	1.81	23.39	1.73	1.30	2.52	15.56	1.17	1.03	4.16
MPONEN	Thoracic blubber	Weight	0.40	0.35	0.18	5.37	1.02	0.64	0.24	6.51	8.40	1.63	1.31	1.92	22.62	1.99	1.80	0.87	2.26	19.39	0.21	0.19	2.01	16.26	1	1.20	3.23
ACID CO	Middle back blubber		0.78	0.19	trace	6.60	0.49	0.17	0.21	8.85	4.05	1.16	1.37	2.93	17.79	1.96	3.12	1.01	2.56	21.44	0.82	0.73	1.46	16.65	1.14	1.68	2.84
	Tongue		0.34	0.12	0.03	6.40	1.52	0.31	0.16	6.90	5.53	2.10	0.80	1.27	18.97	1.33	2.31	1.29	1.07	21.41	0.65	0.00	1.00	17.98	1.25	2.01	4.35
TABLE 2.	Liver		0.63	0.16	trace	6.22	1.39	0.81	0.21	6.69	4.80	2.48	06.0	1.21	18.93	1.34	2.36	1.90	1.82	21.83	0.94	0.73	1.65	16.64	0.85	1.86	3.67
	Stomach		1.21	0.72	0.01	5.48	1.51	1.02	0.26	6.26	60.9	2.66	1.22	1.08	17.74	1.64	2.42	1.11	06.0	19.03	0.75	2.29	1.51	18.62	1.34	1.83	3.30
	Double bond	No. per molecule	0	0	0	0	1	2	0	0	1	2	0	0	1	2	33	0	0	1	2	3	4	5	1	2	9
	Fatty acid chain length	No. C atoms	10	12	13	14	14	14	15	16	16	16	17	18	18	18	18	19	20	20	20	20	20	20	22	22	22

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The each peak was quantitated by the method of Magidmann *et al* (1962). All fatty acids were calculated as weight percentages of the total known fatty acids present.

RESULTS AND DISCUSSION

In this investigation, we confirmed about 25 kinds of fatty acids in the black right whale oil, as the results of the analysis are reported in Table 2. There are as saturated fatty acids, C_{10} , C_{12} , C_{13} , C_{14} , C_{15} , C_{16} , C_{17} , C_{18} , C_{19} and C_{20} , and as unsatturated fatty acids, C_{14} monoenoic, C_{14} dienoic, C_{16} monoenoic, C_{16} dienoic, C_{18} monoenoic, C_{18} dienoic, C_{18} trienoic, C_{20} monoenoic, C_{20} trienoic, C_{20} trienoic, C_{20} trienoic, C_{20} trienoic, C_{20} hexaenoic.

There is no remarkable difference in the fatty acid components of oils contained in various parts of the black right whale body. As the main component fatty acids belong to unsaturated fatty acids, these are C_{20} monoenoic 18.23-23.46% (average 20.7%), C_{18} monoenoic 17.74–22.62% (av. 19.25%) and C_{20} pentaenoic 15.17– 18.67% (av. 17.12%). The next prominent fatty acids are C_{16} saturated 5.20–

Order	Fatty acid chain length No. C atoms	Double bond No. per molecule	Weight par cent of total fatty acids	Average percentages
1	20	1	18.23-23.46	20.70
2	18	1	17.74-22.62	19.25
3	20	5	15.17-18.67	17.12
4	16	0	5.20- 8.85	6.75
5	14	0	5.24- 6.93	6.22
6	16	1	3.60- 8.40	5.57
7	22	6	2.30-4.92	3.65
8	18	3	2.32 - 3.12	2.20
9	16	2	1.12- 2.66	1.77
10	20	4	1.00-2.52	1.75
11	22	5	1.03- 2.01	1.68
12	20	0	0.90- 2.56	1.65
13	18		1.16- 2.01	1.60
14	18	TE OF OCETACE	1.08- 2.93	1.51
15	20	3	0.19- 2.29	1.26
16	22	1	0.85- 2.62	1.23
17	19	0	0.65- 1.90	1.16
18	17	0	0.80- 1.37	1.12
19	20	2	0.21- 1.73	1.12
20	14	1	0.38- 1.52	1.06
21	14	2	0.17- 1.08	1.05
22	10	0	0.33- 1.21	0.61
23	12	0	0.12- 1.21	0.28
24	15	0	0.09- 0.35	0.20
25	13	0	trace- 0.18	0.05

TABLE 3.	EACH	FATTY	ACID	CONTENT	OF	OILS	IN	VARIOUS	BLUBBERS
		AND O	RGANS	5 (in order o	f hig	gh per	centa	ages)	

Fatty acid	Weight per cent of total fatty acids									
chain length No. C atoms	Saturated	Averages	Unsaturated	Averages						
10	0.33-1.21	0.61	_	_						
12	0.12-0.72	.0.28	_	_						
13	trace-0.18	0.05	_	_						
14	5.24 - 6.93	6.22	0.38- 1.52	1.05						
15	0.09-0.35	0.20		_						
16	5.20-8.85	6.75	4.60-10.92	7.34						
17	0.80 - 1.37	1.12	<u> </u>	_						
18	1.08 - 2.93	1.51	10.02 - 27.29	23.05						
19	0.65 - 1.90	1.16								
20	0.90 - 2.56	1.65	34.83-48.16	41.89						
22	—	·	4.05-10.10	6.53						
Total		19.55		79.86						

TABLE 4. A COMPARISON OF SATURATED AND UNSATURATEDFATTY ACID OF BLACK RIGHT WHALE OIL

8.85% (av. 6.75%), C_{14} saturated 5.24–6.93% (av. 6.22%) and C_{16} monoenoic 3.60–8.40% (av. 5.57%). The total of the above mentioned fatty acids holds really 64.18–88.93% (av. 75.61%) of all total fatty acids. (Table 3)

In comparison with saturated and unsaturated fatty acids, the proportions of total saturated fatty acids are 19.55% (average), and those of total unsaturated fatty acids are 79.86% (average) as shown in Table 4. In view of these facts, the principal fatty acids of the black right whale oil are monoenoic and polyenoic unsaturated fatty acids.

SUMMARY

1. Fatty acid components of oils contained in 9 parts of blubbers and 3 kinds of organs of black right whale, *Eubalaena glacialis*, caught in the Northern Pacific Ocean were analyzed by gas-liquid chromatograph using a hydrogen ionization detector on a DEGS column.

2. Fatty acid components of the above mentioned sample oils were as follows;

C ₂₀ monoenoic	18.23-23.46% (av.	20.70%)
C_{18} monoenoic	17.74–22.62% (av.	
C ₂₀ pentaenoic	15.17-18.67% (av.	17.12%)
C_{16} saturated	5.20- 8.85% (av.	6.75%)
C_{14} saturated	5.24– 6.93% (av.	6.22%)
C ₁₆ monoenoic	3.60- 8.40% (av.	5.57%)
C_{22} hexaenoic	2.30- 4.92% (av.	3.65%)
C ₁₈ trienoic	2.32- 3.12% (av.	2.20%)
C_{16} dienoic	1.12- 2.66% (av.	1.77%)
C_{20} tetraenoic	1.00- 2.52% (av.	1.75%)
C_{22} pentaenoic	1.03– 2.01% (av.	1.68%)
C_{20} saturated	0.90– 2.56% (av.	1.65%)

C ₁₈ dienoic	1.16– 2.01% (av.	1.60%)
C_{18} saturated	1.08– 2.93% (av.	1.51%)
C_{20} trienoic	0.19– 2.29% (av.	1.26%)
C_{22} monoenoic	0.85– 2.62% (av.	1.23%)
C_{19} saturated	0.65– 1.90% (av.	1.16%)
C_{17} saturated	0.80– 1.37% (av.	1.12%)
C ₂₀ dienoic	0.21– 1.73% (av.	1.06%)
C14 monoenoic	0.38– 1.52% (av.	1.05%)
C ₁₄ dienoic	0.17– 1.08% (av.	0.63%)
C_{10} saturated	0.33– 1.21% (av.	0.61%)
\mathbf{C}_{12} saturated	0.12-0.72% (av.	0.28%)
C_{15} saturated	$0.09-\ 0.35\%$ (av.	0.20%)
$\mathbf{C}_{{\scriptscriptstyle 13}}$ saturated	trace- 0.18% (av.	0.05%)

3. A substantial part of the saturated acids were hexadecanoic (5.20-8.85%) and tetradecanoic (5.24-6.93%). On the other hand, that of the unsaturated acids were nonadecamonoenoic (18.23-23.46%), octadecamonoenoic (17.74-22.62%) and nonadecapentaenoic (15.17-18.67%).

4. The difference in the fatty acid components of various blubbers and organs of the black right whale was not found clearly.

REFERENCES

GAUGLITZ, E. J. Jr. and L. W. LEHMAN, 1963. The preparation of alkyl esters from highly unsaturated triglycerides. J. Am. Oil Chemists' Soc., 40: 197–198.

- MAGIDMANN, P., S. E. HERB, F. E. LUDDY and R. W. RIEMENSHNEIDER, 1962. Fatty acid cow milk, II. Composition by gas-liquid chromatograph aided by other methods of fractionation. J. Am. Oil Chemists' Soc., 39: 142-146.
- NELSON, G. J. and N. K. FREEMAN, 1960. Phospholipide and phospholipide-fatty acid component of human serum lipo-protein fraction. J. Biol. Chem., 235: 578–583.
- TSUYUKI, H. and U. NARUSE, 1963. Studies on the oil of black right whale in the Northern Pacific Ocean. Sci. Rep. Whales Res. Inst., 17: 171-190.
- TSUYUKI, H. and U. NARUSE, 1964. Studies on the lipids in brain of black right whale in the Northern Pacific Ocean. Sci. Rep. Whales Res. Inst., 18: 173-180.
- TSUYUKI, H., U. NARUSE, A. MOCHIZUKI and S. ITOH, 1964. Studies on liver oil of black right whale. Bull. Coll. Agr. & Vet. Med., Nihon Univ., 18:17-24.