STUDIES ON THE OILS CONTAINED IN BLUBBER OF A SOUTHERN ELEPHANT SEAL

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INTRODUCTION

The Southern elephant seal, *Mirounga leonina*, is found off the Antarctic Ocean and thinking approximate species, the Northern elephant seal, *Mirounga angustirostiris*, is mostly distributed over the coast of California and Mexico. They are the largest of all the marine carnivores and belong to the seal family. They grow up body length 5.5–6.7 metres and body weight about 3 tons.

As to the study on the seal family oil, we find the following reports; Saghalien seal oil (Tsujimoto, 1916), Antarctic seal oil (Ueno & Iwai, 1939), common seal oil (Bauer & Neth, 1942), common seal, *Phoca vitulina*, oil (Williams & Makhrov, 1935), commercial Newfoundland seal oil (Burke & Jasperson, 1944), blubber and liver oil of Grey Atlantic seal, *Halichoerus grypus* and common seal (Hilditch and Pathak, 1947, 1949), milk oils of Grey Atlantic and common seals (Meave, 1952), blubber oils of Leopard seal, *Hydrurga leptonyx* and Crabeater seal, *Lobodon carcinophagus* (Winter & Nunn, 1950, 1953).

Reviewing the works ever reported on the elephant seal oil, Tsuyuki studied on the properties of the oils contained in various blubbers of the Northern elephant seal caught at the coast of Mexico (1957) and its component fatty acids (1958), and Winter and Nunn studied on the fatty acid composition of the blubber oils from a wide range of specimens of the Southern elephant seal caught at Macquarie and Herald Island in the Antarctic (1950, 1953).

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MATERIALS AND METHODS

Sample used

The Southern elephant seal, *Mirounga leonina*, caught off the South Georgia Island on the 15th of November, 1964 was used in this experiment. It was immediatly frozen within an hour after the catch. The frozen body was brought to Japan on the 28th of January, 1965. The details of the Southern elephant seal used in this experiment are shown in Table 1 and Fig. 1. Oils were extracted by boiling with water from various blubbers which are shown in Fig. 1 and Table 2. The properties of various blubber oils were examined by ordinary methods (Table 3).

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Sex	Age	Body weight (except blood)	Body length	Girth at the tip of flipper	Girth at the shoulder	Girth at anus
Male	6	$906.9~\mathrm{Kg}$	2.73 m	2.50 m	2.47 m	0.75 m

TABLE 1. DETAILS OF SOUTHERN ELEPHANT SEAL (Cited from ICHIHARA & NISHIWAKI, 1966)

Sample	Kinds of blubber	Thickness of blubber (cm.)	Oil content in blubber (%)	Appearance (at 30°C)
А	Dorsal blubber of abdominal cavity	4–6	67.2	Yellowish orange liquid
В	Blubber of frontal (part between eyes)	2–3	53.8	Yellowish orange liquid
С	Dorsal blubber of thoracic cavity	4–6	57.1	Yellowish orange liquid
D	Dorsal blubber of abdominal cavity	5.5–6	76.9	Yellowish orange liquid
E	Ventral blubber of thoracic cavity	5-6	52.1	Yellowish orange liquid
\mathbf{F}	Ventral blubber of neck	3-4	69.5	Yellowish orange liquid
G	Ventral blubber of thoracic cavity	4–5	62.9	Yellowish orange liquid
Н	Ventral blubber of abdominal cavity	3-4	57.7	Yellowish orange liquid
I	Ventral blubber of pelvis	2-3	71.8	Yellowish orange liquid
J	Ventral blubber of hindmost part	2	56.9	Yellowish orange liquid
K	Blubber of tongue	1	66.3	Yellowish orange liquid

TABLE 2. BLUBBERS AND OILS

TABLE 3. PROPERTIES OF BLUBBER OILS

Sample	N_{D}^{30}	Acid value	Sapon. value	Iodine [®] value	matter (%)
Α	1.4691	1.67	191.4	127.3	0.23
В	1.4689	1.32	197.5	127.6	0.26
С	1.4690	1.73	189.5	117.6	1.10
D	1.4699	1.55	190.7	130.7	0.58
Е	1.4691	1.29	193.1	118.2	0.69
F	1.4684	1.59	195.8	121.5	0.55
G	1.4690	1.49	196.2	126.5	0.24
H	1.4690	1.47	196.1	120.5	0.19
Ι	1.4689	1.42	191.6	132.2	0.23
J	1.4692	1.28	190.3	132.4	0.26
K	1.4691	1.21	192.7	129.7	0.23



Fig. 1. Various blubbers of Southern elephant seel

Preparation of the methyl esters of the fatty acids

The various blubber oils were saponified by refluxing them with excess 2N alcoholic potash for 2 hours on a steam bath. The alcohol was evaporated, the soap residue dissolved in water and excess dilute sulphuric acid added to liberate the fatty acids. These were washed with successive portions of water to remove mineral acid. The fatty acids were dissolved in ether and this solution was dried by adding anhydrous sodium sulphate. The ether was evaporated under reduced pressure. The properties of conjugated fatty acids are shown in Table 4.

Sample	Appearance (at 30°C)	${ m N_D^{80^o}}$	Iodine value	Neutralization value
А	Reddish orange liquid	1.4599	129.6	193.2
в	Yellowish orange liquid	1.4590	129.5	199.0
\mathbf{C}	Yellowish orange liquid	1.4518	127.0	195.8
D	Yellowish orange liquid	1.4598	133.5	191.0
Е	Reddish orange liquid	1.4600	124.8	194.1
F	Reddish orange liquid	1.4598	128.2	198.1
G	Orange liquid	1.4595	131.5	198.5
н	Reddish orange liquid	1.4597	128.6	199.2
I	Yellowish orange liquid	1.4582	137.3	193.3
J	Yellowish orange liquid	1.4589	136.7	192.7
K	Yellowish orange liquid	1.4561	ESEA 131.7	194.1

TABLE 4. PROPERTIES OF MIXED FATTY ACIDS

The fatty acid residue was refluxed for 10 hours on the steam bath with four to five times its weight of a 2% solution of sulphuric acid in methanol under N_2 gas.

After removal of the bulk of the methanol, the residue was dissolved in ether and washed first with water to remove mineral acid, then with 5% sodium carbonate solution to remove unesterified fatty acid, and finally with water to remove carbonate.

The ether solution of the methyl esters was dried over anhydrous sodium sulphate and the solvent evaporated under reduced pressure. The methyl esters were colorless liquid and slightly ester-smell.

Gas chromatographic examination of the fatty acids methyl ester

Gas chromatographic apparatus used in this study was a Shimadzu Model GC-1B. A three metres stainless steel column of 4 mm. internal diameter was packed with 60/80 mesh succinate polyester. Helium was used as the carrier gas at flow rate of 50 ml./min. at 2.5 kg./cm². and chart speed 0.5 cm./min. A sample size of 7 μ l gave a good general chromatogram of all the fatty acids. The identity of the peak was established by reference to relatively pure standard samples of methyl ester of the various fatty acids and other peaks on the chromatograms were presumed by reference to retention time of each peak and carbon numbers.

RESULTS AND DISCUSSION

Gas chromatogram operated with above mentioned condition obtained the peak more than 20 as shown in Fig. 2. After the fatty acid methyl ester was hydrogenated by a catalyzer of platinum black, stirring with a magnetic-stirrer as 10% solution of methanol for 24 hours at room temperature, gas chromatography was operated about it (shown in Fig. 3), and the following results were obtained.

Peak	Fatty					Sa	mple oil	s				
No.	acid	А	В	С	D	\mathbf{E}	F	G	н	Ι	J	К
1	12:0	0.49%	0.64%	1.52%	0.46%	0.56%	0.46%	0.51%	0.41%	0.57%	1.16%	0.69%
2	13:0	trace	trace	trace	trace	trace	trace	trace	trace	trace	trace	trace
3	14:0	5.07	4.80	4.93	5.06	4.70	4.32	5.00	5.16	4.46	4.17	5.41
4	14:1	0.81	1.08	1.07	1.02	1.18	1.33	1.52	1.23	1.59	1.16	1.34
5	15:0	0.16	0.70	0.15	0.63	0.68	0.81	1.00	0.76	1.02	0.47	0.89
6	14 2	0.08	0.25	trace	0.40	0.25	0.11	0.13	trace	0.10	trace	trace
7	16:0	11.60	10.85	12.63	12.16	11.77	10.51	11.60	12.24	10.43	10.22	11.63
8	16:1	11.11	12.00	11.68	11.83	11.94	11.37	11.03	11.19	11.56	12.68	11.42
9	${17:0 \\ 16:2}$	0.94	0.89	1.07	0.85	0.79	0.93	1.12	0.88	1.59	0.47	1.84
10	16:3	1.71	1.46	1.52	1.54	1.69	1.79	1.82	1.64	1.87	1.47	1.89
11	18:0	2.91	1.40	1.75	2.39	1.30	1.80	1.43	1.64	1.42	1.32	3.18
12	18:1	38.30	41.47	40.46	38.92	39.76	39.66	37.10	41.13	37.50	36.27	35.40
13	18:2	2.10	2.29	2.05	1.90	2.48	2.67	2.50	2.17	2.53	2.64	2.49
14	19:0	0.10	0.25	trace	trace	trace	0.21	0.11	0.10	0.12	trace	0.37
15	18:3	trace	0.70	trace	1.14	1.30	1.10	1.84	0.65	1.99	1.80	1.04
16	20:0	0.35	0.24	0.26	0.15	0.31	0.40	0.22	0.26	0.14	0.34	0.31
17	20:1	12.20	11.77	10.65	11.07	11.43	11.35	11.23	10.75	10.86	12.35	11.19
18	20:2	1.26	1.00	1.07	1.10	1.21	1.11	1.01	1.11	1.06	1.21	1.13
19	22:0	trace	trace	trace	trace	trace	trace	trace	trace	trace	trace	trace
20	22:1	6.60	4.04	4.55	4.56	5.17	5.05	6.72	5.39	5.87	6.77	4.78
21	22:5	2.00	2.17	2.54	2.56	1.43	2.38	2.11	1.36	2.71	2.86	2.64
22	22:6	2.21	2.00	2.10	2.26	2.05	2.64	2.00	1.93	2.61	2.74	2.36
Т	otals	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00

TABLE 5. FATTY ACID COMPOSITION OF BLUBBER OILS OF SOUTHERN ELEPHANT SEAL

It was comfirmed that the component fatty acid of the blubber oil of Southern elephant seal was the saturated fatty acids as follows; C_{12} , C_{13} , C_{14} , C_{15} , C_{16} , C_{17} , C_{18} , C_{19} , C_{20} , C_{22} and other peaks were estimated by the relation of straight line in the numbers of double-bond of carbon and the logalism of retention time as the unsaturated acids of following; C_{14} monoenoic, C_{14} dienoic, C_{16} monoenoic, C_{16} dienoic,



Fig. 2. Gas chromatogram of fatty acid metyl esters of blubber oils



Fig. 3. Gas chromatogram of fatty acid methyl ester of blubber oil before and after hydrogenation A. before hydrogenation B. after hydrogenation

 C_{16} trienoic, C_{18} monoenoic, C_{18} dienoic, C_{18} trienoic, C_{20} monoenoic, C_{20} dienoic, C_{22} monoenoic, C_{22} pentaenoic, C_{22} hexaenoic.

According to the weight method of each peak area, the percentages of total fatty acids were shown in Table 5.

The acid values of blubber oils extracted from various parts of Southern elephant seal were low as shown in Table 2, therefore it proved that the free fatty acids were less in blubber oils.

On the other hand, both acid value and unsaponification material content are comparatively low, therefore these facts seem to show that the blubber oils consist of nearly glycerides.

As shown in Fig. 2, the component fatty acids of oils obtained from various blubbers are nearly similiar.

According to the results, the component fatty acid is unsaturated C_{18} monoenoic (35.40–41.73%), the next prominent is unsaturated C_{16} monoenoic (10.83– 12.68%). Other component fatty acids are as follows; saturated C_{16} (10.22–12.63 %), unsaturated C_{20} monoenoic (10.03–12.35%), unsaturated C_{22} monoenoic (4.04– 6.77%), saturated C_{14} (4.17–5.16%), unsaturated C_{18} dienoic (1.90–2.67%), unsaturated C_{22} hexaenoic (1.51–2.74%), unsaturated C_{22} pentaenoic (1.36–2.86%), saturated C_{18} (1.29–3.18%), unsaturated C_{16} trienoic (1.20–1.89%), unsaturated C_{14} monoenoic (0.81–1.59%), unsaturated C_{20} dienoic (0.98–1.26%), unsaturated C_{18} trienoic (0.65–1.24%), unsaturated C_{16} dienoic + saturated C_{17} (0.47–1.21%), saturated C_{19} (0.15–1.02%), saturated C_{12} (0.41–1.00%), saturated C_{20} (0.14–0.42%), saturated C_{19} (0.10–0.37%), unsaturated C_{14} dienoic (0.08–0.40%), saturated C_{13} and C_{22} (respectively trace), as shown in Table 6.

After all, the total saturated fatty acids content is 16.95-24.99%, while the total unsaturated fatty acids content is 68.54-90.09%.

SOUTHERN ELEPHANT SEAL OIL

Order	Fatty acid	Percentages	Average percentage
1	C18:1	35.40-41.73	38.67
2	C16:1	10.83-12.68	11.61
3	C16:0	10.22-12.63	11.37
4	C _{20:1}	10.03-12.35	11.27
5	C22:1	4.04-6.77	5.19
6	C14:0	4.17-5.16	4.83
7	C _{18:2}	1.90- 2.67	2.36
8	C22:6	1.51- 2.74	2.21
9	C22:5	1.36- 2.86	2.05
10	C18:0	1.29- 3.18	1.94
11	C16:3	1.20- 1.89	1.67
12	C14:1	0.81- 1.59	1.21
13	C20:2	0.98- 1.26	1.11
14	C18:3	0.65 - 1.24	1.06
15	$C_{17:0}+C_{16:2}$	0.47-1.21	1.03
16	$C_{15:0}$	0.15- 1.02	0.66
17	C12:0	0.41- 1.00	0.43
18	C20:0	0.14 - 0.42	0.27
19	C19:0	0.10 - 0.37	0.17
20	C14:2	0.08-0.40	0.09
21	C13:0	trace	
22	C22:0	trace	

TABLE 6. EACH FATTY ACID CONTENT (%) IN VARIOUS BLUBBER OILS (in order of large value)

TABLE 7. A COMPARISON OF THE FATTY ACID COMPOSITION OF THE SOUTHERN ELEPHANT SEAL AND THE NORTHERN ELEPHANT SEAL OIL

Fa	atty acid	Southern elephant seal	Northern elephant seal	
Saturated acid	C_{12}	0.41- 1.00%	-%	
	C_{13}	trace		
	C ₁₄	4.17-5.16	3.52	
	C_{15}	0.15- 1.02		
	C_{16}	10.22–12.63	12.82	
	C_{17}	0.47- 1.21		
	C ₁₈	1.29- 3.18	3.61	
	C19	0.10- 0.37	_	
	C20	0.14- 0.42	0.41	
	-C ₂₂	trace	0.01	
Total saturated	acid	16.95–24.99%	20.37%	
Unsaturated aci	d C ₁₄	0.89-1.99(-2, -4H)	0.96 (-2.0H*)	
	C_{16}	12.50-16.41 (-2, -4, -6H)	10.02 (-2.5H*)	
	C_{18}	37.95-45.64 (-2, -4, -6H)	33.22 (3.0H*)	
	C_{20}	11.05-13.68 (-2, -4H)	24.50 (-4.4H*)	
C		6.15–12.37 (–10, –12H)	10.25 (-7.1H*)	
	C_{24}	_	$0.59 (-6.6 H^*)$	
Total unsaturate	ed acid	68.54–90.09%	79.63%	

* average unsaturation

Comparing with the component acids of Northern elephant seal, Mirounga angustirostiris, oil studied by Tsuyuki and Southern elephant seal, Mirounga leonina, oil in this experiment (Table 7), there is remarkable difference in C_{18} monoenoic acid content.

The total unsaturated C_{18} acid content (37.95–45.64%) of the Southern elephant seal oil is higher than that (33.22%) of the Northern elephant seal oil, on the contrary, the total unsaturated C_{20} acid content (11.05–13.68%) of the Southern elephant seal oil is lower than that (24.50%) of the Northern elephant seal oil. In other fatty acid contents, there is no remarkable difference between both seal oils.

In this study, the existance of the saturated acids with odd carbon atoms (13, 15, 17, 19) was comfirmed, but the unsaturated C_{24} acid was not comfirmed.

SUMMARY

The oils contained in various blubbers of the Southern elephant seal, 1. Mirounga leonina, were studied.

The fatty acid composition of the Southern elephant seal oil was studied by gas chromatography on a polyester column.

The results obtained are as follows;

Total saturated fatty acids 16.95-24.99%;

lauric acid	0.41- 1.00%
tridecanoic acid	trace
myristic acid	4.17- 5.16%
pentadecanoic acid	0.15- 1.02%
palmitic acid	10.22-12.63%
heptadecanoic acid	0.47- 1.21%
stearic acid	1.29- 3.18%
nonadecanoic acid	0.10- 0.37%
arachidic acid	0.14- 0.42%
behenic acid	trace
stal unsaturated fatty acids 68 54-90 09%.	

Total uns

C ₁₄ monoenoic acid	0.81- 1.59%
C ₁₄ dienoic acid	0.08- 0.40%
C ₁₆ monoenoic acid	10.83-12.68%
C ₁₆ dienoic acid	0.47- 1.21%
C_{16} trienoic acid	1.20- 1.89%
C ₁₈ monoenoic acid	35.40 - 41.73%
C_{18} dienoic acid	1.90- 2.67%
C_{18} trienoic acid	0.65- 1.24%
C_{20} monoenoic acid	10.03-12.35%
C_{20} dienoic acid	0.98- 1.26%
C_{22} monoenoic acid	4.04 6.77%

C_{22} pentaneoic acid	1.36 -	2.86%
C_{22} hexaenoic acid	1.51 -	2.74%

2. The component acids of blubber oil of the Southern elephant seal and the Northern elephant seal were nearly similar, except that there is remarkable difference in C_{18} monoenoic acid content.

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