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### On the extrinsic eye muscles of the whale, with special remarks upon the innervation and function of the musculus retractor bulbi

### By

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While I was engaged in the anatomical study on the visual organs of the whale, special attention was directed to the musculi palpebrales, which are in the cetacea, as some anatomists said, of an unique existence throughout the animal kingdom. Next to that, my special interest was rendered to the well-developed musculus retractor bulbi, the existence of which is never characteristic for the cetacea, but is rather common for almost all vertebrates except the cyclostoma and fishes (Haller v. Hallerstein, 1934; Franz, 1934 etc.)<sup>1</sup>. Meanwhile I encountered in the whale a peculiar feature concerning the innervation of the retractor bulbi and tried to explain it by various means of researches, which will be related in order in the following paragraphs. As the result, I came to the conclusion that the innervation of the cetacean retractor bulbi is never essentially different from that in other vertebrates, though it exhibits a noteworthy peculiarity at first sight.

Relating to this problem, I tried to know the functional meaning of this muscle and studied also its original nucleus in the brain-stem, experimenting on the cat.

At the end, my observations on the sensory nerves of the retractor and of other extrinsic eye muscles in the cetacea will be mentioned.

### MATERIALS

**MYSTACOCETI:** 

- 1) Sei whale (Balaenoptera borealis, Lesson): adult 1 (45 feet, female), fetus 1 (7 feet, male)
- 2) Blue whale (B. musculus, L.): fetuses 2 (6 feet, female; 7 feet, male)
- 3) Fin whale (B. physalus, L.): fetuses 3 (3 feet, male; 8 feet, female; 13 feet, male)

1) Even among the fishes some teleostei are said to have slight indication of this muscle.

**ODONTOCETI:** 

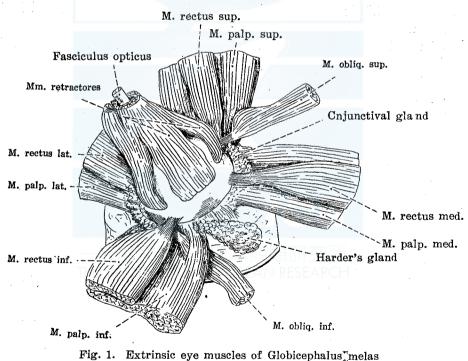
- 4) Sperm whale (Physeter catodon, L.): fetuses 2 (3 feet, female; 5 feet, male)
- 5) Pilot whale (Globicephalus melas, Traill): adult 1 (17 feet, male)
- 6) A dolphin (Prodelphinus caeruleo-albus, Meyen): adults 2, fetuses 3.

Other mammals used for comparison:

1) cats 5 2) dogs 2 3) rabbits 2

### 1. The extrinsic eye muscles of the whale

In the cetacea the extraocular muscles consist of two oblique, four straight ones and of the retractor bulbi. Of them, it is noteworthy that the greater part of each of four recti muscles extends forwards over the eyeball and inserts into the eyelid, delivering only a small muscle branch to the eyeball (Fig. 1, 2). While the ocular portions



(17 feet, male) (left eye)

of the recti muscles are relatively well preserved in the Globicephalus, they are very rudimentary in the Sei and other baleen whales. For

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this reason, these muscles of the cetacean eye do not deserve well the name "Mm. recti bulbi", but the name "Mm. palpebrales" is for them more preferable, as Weber (1886) and Pütter (1903) called them.

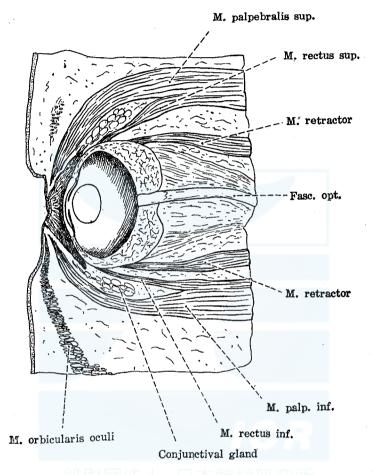


Fig. 2. Median section through eye and palpebrae of Globicephalus melas

The peculiarity of the musculi palpebrales of the cetacea has been well known since Weber, but the meaning of it is not yet fully determined. Weber was of the opinion that they might serve for the mechanical protection of the eyeball against water pressure, while Pütter ascribed them a hydrodynamic function, assuming that they might efficiently work against cooling down of the eyeball producing heat by their contraction.

The smallness of the ocular attachments of the recti muscles is

generally thought to have much to do with the decline of the oculomotor function in the whale.

Concerning the obliqui muscles nothing special is found in the cetacean eye, except that the ocular insertion of the inferior obliquus is sometimes divided into two portions, as shown in Fig. 3.

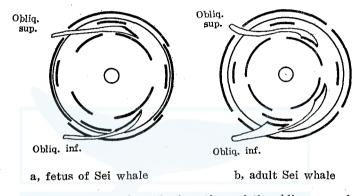


Fig. 3. Diagram to show the insertions of the oblique muscles (right eye, posterior view)

The retractor bulbi will be described at length later on.

THE INNERVATION of the obliqui and recti muscles is in the whale intrinsically the same as in other animals; namely the inferior obliquus and all the recti except the lateral rectus are supplied by the oculomotorius, while the superior obliquus and the lateral rectus are innervated respectively by the trochlearis and the abducens. These muscles and nerves are illustrated in Fig. 4 (a-g). Fig. 5 (a-e) shows the intramuscular distribution of nerve fibres in each muscle. Fig. 6 is a diagram showing the innervations.

GANGLION CILIARE. In old times, more than a century ago, Burns (1832) and Rapp (1837) could not find the ciliary ganglion in Phocaena communis, but Stannius (1842) saw it in the same porpoise (cited from Schwalbe, 1879). Weber (1886), not finding it in Hyperoodon and other toothed whales, thought that this ganglion might be missing or at least very rudimentary in them. In the whales treated in the present work, both in the baleen and toothed ones, the existence of this ganglion was ascertained, though they are usually very small in size (Fig. 5, c). The radix brevis, connecting this ganglion with the trunk of the oculomotor nerve, consists of one or two thin bundles, sometimes very short, while in other cases it is considerably longer.

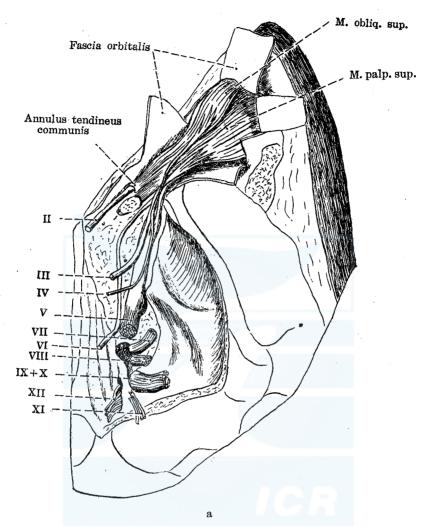


Fig. 4. Extrinsic eye muscles and their innervations of the Sei whale (fetus, 7 feet) (right eye)

The radix longa, the connection with the ophthalmic nerve, is always present, while the sympathetic root could not be seen.

The size of this ganglion is as follows;

1)	Blue whale	(fetus,	6 feet, left)	$3 imes$ 1. $8 imes$ 1. $5~\mathrm{mm}$
2)	,, <u>,</u> ,	( ,,	7 feet, l.)	$1.1 \times 0.8 \times 0.8$ mm
3)	Fin whale	( ,,	8 feet, l.)	3 imes 2 imes 1.2 mm
4)	,, ,,	( ,,	13 feet, righ	t) $1.7 \times 1.3 \times 1.0 \text{ mm}$
5)	Sperm whale	( ,,	3 feet, r.)	$1.5 \times 1.0 \times 0.8$ mm

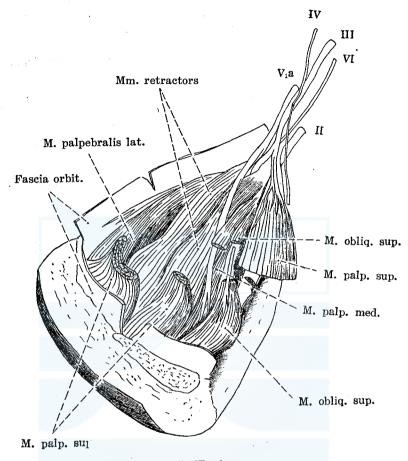


Fig. 4. b

Comparison with the ciliary ganglion of the cat  $(3.5 \times 1.9 \times 1.5 \text{ mm};$  $3.0 \times 1.8 \times 1.5 \text{ mm})$ , dog  $(3.0 \times 1.7 \times 1.5 \text{ mm})$  and of the rabbit  $(0.8 \times 0.6 \times 0.5 \text{ mm})$  shows how small it is in the whale.

### 2. General view of the retractor bulbi

The retractor bulbi is in the whale, as illustrated in Figs. 1 and 2, composed of not very distinctly separated four portions. They insert to the eyeball, as shown in Fig. 7, nearer the posterior pole of the eyeball than the recti muscles, and each portion is situated between two of the latters. So I would call them respectively Pars superior medialis, Pars superior lateralis, Pars inferior medialis and Pars inferior lateralis. As generally known, the retractor is well developed

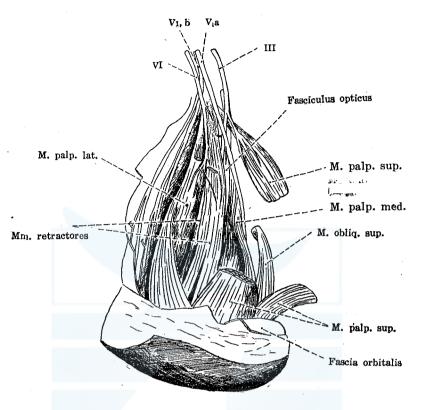


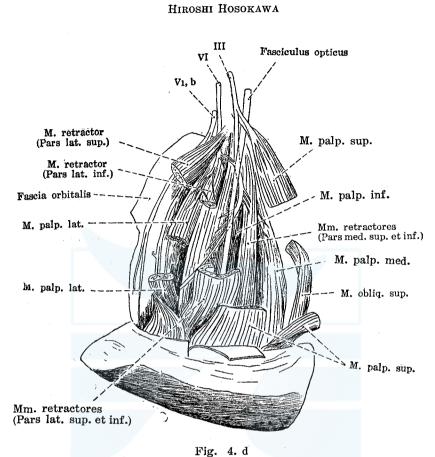
Fig. 4. c

in amphibia and reptilia, while in birds the musculus pyramidalis and musculus buralis s. quadratus are probably homologous to it. Also in mammals the retractor has a wide distribution; only a few of them including man are devoid of this muscle. According to Haller v. Hallerstein, these exceptional mammals are Orycteropus, Pteropus and Primates<sup>1</sup>.

As in the whales, the mammalian retractor is often divided into four portions. But it is sometimes not readily divisible into portions, for example in some domestic animals such as horse, ox, sheep and pig.

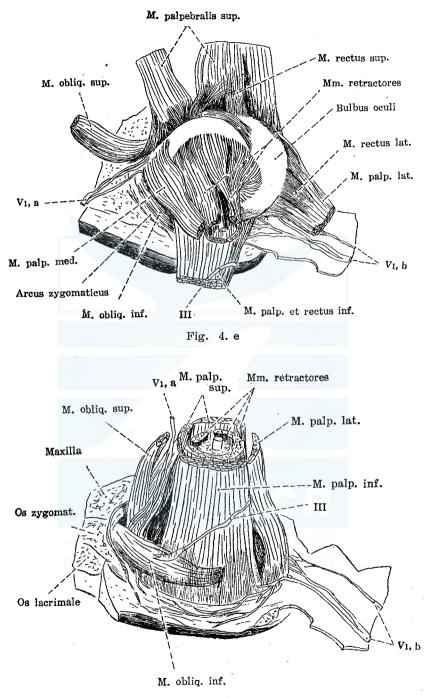
Many authors have studied the retractor, calling it with various names, for instance, m. retractor bulbi s. oculi, m. suspensor[ius] oculi, m. choanoides, Grundmuskel, muscle en étonnoir, posterior rectus etc.

1) Nussbaum (1893) and Fleischer (1907) described as anomalous cases a rudimentary retractor in the human being.

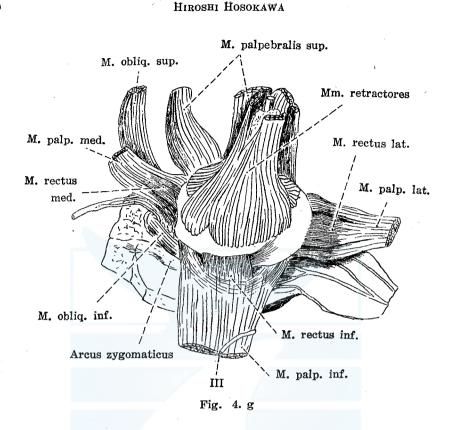


### 3. Historical review on the motor innervation of the retractor bulbi

Before mentioning the nerves of the retractor in the whale, I will review briefly the literature of the problem for various kinds of animals. Concerning lower vertebrates, previous authors said unanimously that it is supplied by the abducent nerve (Corning, 1902; Nishi, 1922, 1938). As to the mammals, however, remarkable discrepancies have prevailed among the authors, of which the details were already reported by Hopkins (1919) and Cords (1924). Table 1 represents the brief summary of them. As shown in this table, the authors might be classified into two groups, the one saying that the retractor is innervated merely by branches from the abducens, and the other attributing two or more nerve sources for this muscle. It is interesting to note that many of veterinary anatomists belong to the latter group. Most of them insisted on the dual innervation by the abducens



### Fig. 4. f



and the oculomotorius.

This remarkable disagreement attracted naturally the attention of some anatomists, and especially Hopkins and Cords, in order to decide this problem, dissected many kinds of mammals repeatedly, using the binocular microscope. They found then no indication of filaments either from the superior or from the inferior division of the oculomotor nerve to the retractor, and so concluded that the mammalian retractor is supplied exclusively by the abducent nerve just like in lower vertebrates. They said with confidence that all the statements attributing the oculomotorius or other motor source were incorrect for this muscle.

### 4. Motor innervation of the cetacean retractor bulbi

As shown in Table 1, Rapp (1837) and Stannius (1842) stated that the retractor of Phocaena communis is supplied by the abducent nerve<sup>1</sup>.

<sup>1)</sup> They described also a nerve supply from the ophthalmic nerve. About such a sensory innervation I will state later in Chapter 8.

Weber (1886) mentioned the same for Hyperoodon and other whales, and Cords (1924) was also of the same opinion, when he dissected a young specimen of Delphinus phocaena.

In the present study, however, an interesting fact was seen that the retractor of the whale, especially its medial portions receive decidedly branches from the inferior division of the oculomotor nerve, while the lateral portions of this muscle are supplied without doubt by branches of the abducens (Figs. 5, 8 and 9 a). I confirmed this fact in all the whales treated in the present paper. There exists, concerning this point, no exception, although the branches are quantitatively variant to some extent according to individuals or to species.

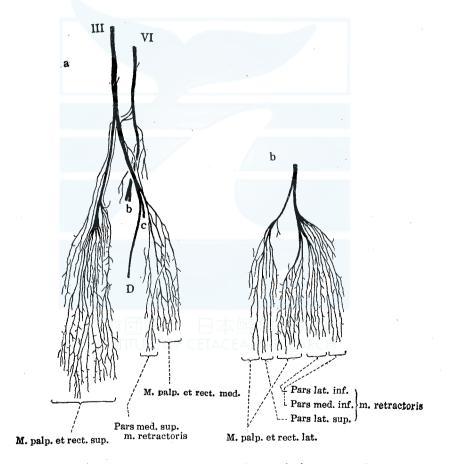


Fig. 5. (1) Motor innervation of the extrinsic eye muscles (fetus of the Fin whale, 13 feet, male) (right eye)

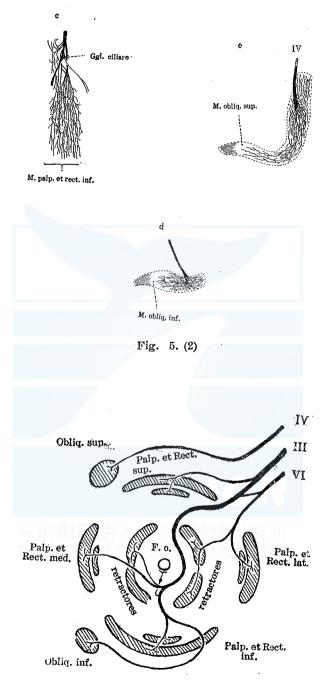


Fig. 6. Diagram to show the innervation of the extrinsic eye muscles in the whale (left eye, anterior view)

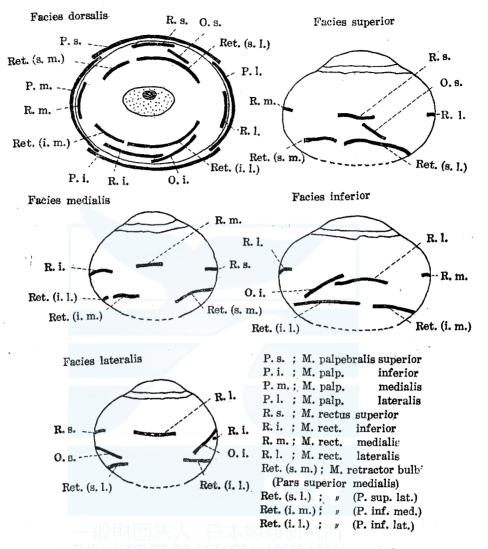


Fig. 7. Insertions of the extrinsic eye muscles of the whale (Fetus of Sei whale, 7 feet) (right eye)

Moreover, in all cases an anastomsis is existent between the trunk of the abducens and the inferior division of the oculomotor nerve near the top of the orbital cone. The anastomosing nerve bundle comes proximally from the abducens and passes distally into the oculomotorius. It is illustrated in Fig. 5, 8 and 9a. Fig. 9 b is a simplified diagram to show the relation.

### Table 1.

Opinions of the previous authors upon the innervation of M. retractor bulbi in mammals

VI (N. abd	lucens) only	VI and other nerves			
Authors	Materials	Authors	Innervation	Materials	
Stannius (1846)	(comparative anatomy)	Rapp (1838) Stannius (*42)	VI + V <sub>1</sub> " + "	Phocaena comm.	
Foltz ('62)	horse, rabbit	Chauveau ('57)	VI + III	domestic animals	
Owen ('68)	(comp. anat.)	Krause ('68)	{ III only { VI only	rabbit etc. cat, young ox	
Milne-Edwards ('76)	(comp. anat.)	Gurlt ('73)	VI + III	(comp. anat.)	
Wilder & Gage ('86) Weber ('87)	domestic animals Phocaena comm.,	Schwalbe ('79) Leisering ('85)	VI + III " + "	sheep, ox, dog, rabbit etc. domestic animals	
. ,	Sei whale				
Motais ('87) Mivart ('89)	cat	Ellenberger & Baum ('91, '96)	III only	dog	
Nussbaum ('93)	(comp. anat.)	Frank ('94) Reute ('97)	VI + III // + //	domestic animals fetus of pig	
Gegenbaur ('98)	(comp. anat.)	Bradley ('97) Varaldi ('99)	" + " " + "	(veterinary anat.) ( // // )	
		Du-Bois Reymond & Silex ('99, '07)	" + "	dog, cat, rabbit	
Corning ('02)	dog, cat	Reighard etc. (1901)		cat horse	
Wiedersheim ('06)	(comp. anat.)	M'Fadyan ('02) Struska ('03) Martin ('04)	VI + III " + " " + "	domestic animals	
		Share-Jones ('06) Fleischer ('07)	III+IV+VI III only	horse man (anomaly)	
Montane-	domestic animals	Ellenberger & Baum ('08, '12)	VI + III	domestic animals	
Bourdelle ('13) Hopkins ('17)	horse, ox, sheep, pig, dog, cat, rabbit, etc.	Strangeway ('09) Zimmerl ('09) Bensley ('10)	VI + x (III ?) VI + III " + "	(veterinary anat.) ( " " ) rabbit	
Cords ('24)	many kinds of mammals includ- ing Phocaena comm.	Bradley ('12) Sisson ('14)	III only VI + III	dog domestic animals	
Key-Åbergs ('34)	rabbit				
Imai ('34, '36)	monkey, cat				

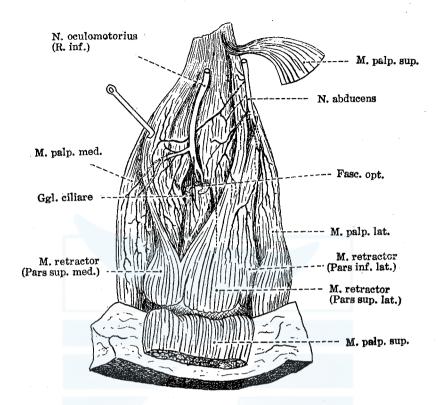
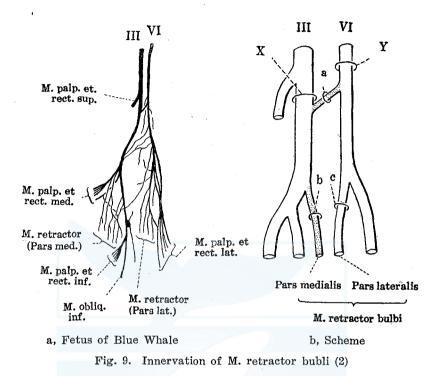


Fig. 8. Innervation of M. retractor bulbi (1) Fetus of Blue whale (7 feet) (left eye)

Now, it occurs naturally the question whether the nerve fibres of the anastomosis (a in Fig. 9 b) continue directly into the branches of the oculomotor nerve to the medial portions of the retractor (b in the same figure). In other words, the branches in question may belong intrinsically to the abducens, like the branches to the lateral portions, passing only temporarily via the oculomotorius. But because of the close adhesion of fiber bundles, it was impossible for me to separate and trace the distal extention of the bundle a macroscopically. So, I tried to settle the problem from various viewpoints.

### A. CALCULATION OF NERVE FIBERS

First of all, the nerve fibres were calculated in the anastomosis and in the branches of the oculomotorius going to the retractor. The result upon a fetus of the Blue whale is shown in Table 2. The medullated fibers of the bundle a amount to 768, while those of the bundle b are 806, exceeding thus the former by about 5%. If Kölliker



(1867) and Sherrington (1894) be right in their remarks that the nerve fiber does not bifurcate during its course in relatively thick nerve trunk, which is macroscopically recognizable, one could admit that at least some of the fibers in b are probably derived from the oculomotor nerve itself. But my further calculation of fibers in the trochlear nerve in the same whale, showed that the nerve trunk contains in the distal part more fibers than in the proximal part, the difference amounting to 10% approximately.

Moreover, I found in a cat that a distal portion of the abducens possesses about 20% more fibers than a proximal portion. And as between the two points examined no addition of nerve fibers from outside to the nerve trunk occurred, this increase of nerve fibers in number is very probably due to bifurcation or dichotomizing of a nerve fiber within the nerve trunk. Eccles and Sherrington (1930) insisted statistically, on the branching of both the afferent and efferent fiber within the nerve trunk with unequivocal illustrations of bifurcating fibers. Other authors such as Dunn (1902), Björkmann and Wohlfart (1936), Takagi (1948) etc. have also suggested or confirmed the increase of nerve fibers distalwards in the nerve trunk.

### Table 2.

Fiber-analysis of N. III and N. VI of a Blue whale fetus (6 feet, left eye)

Parts as shown in Fig. 9, b	total number of nerve fibres	thickness* of a fiber	number of fibres of each thickness	% to the total number
	9247	thick	401	4
x		medium-sized	5791	63
		fine	3055	33
	7041	thick	199	3
Y		medsized	4512	64
		fine	2330	33
		thick	34	4
a	769	medsized	549	71
		fine	186	25
	806	thick	18	2
b		medsized	548	72
		fine	204 ·	26
	1160	thick	18	2
c		medsized	901	78
		fine	241	20

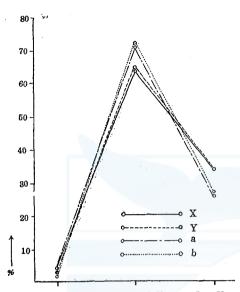
\* thick fibres: larger than  $5.0 \mu$  (in diameter) medium-sized f.: between 5.0 and  $2.5 \mu$  (") fine fibres: smaller than  $2.5 \mu$  (")

Anyway, now that the distal increase of nerve fibers in number along the nerve trunk is certain, all the nerve fibers in the bundle bof the whale may possibly be derived from the anastomosis a, though at the same time we cannot deny also the possibility that some of the fibers in b may belong originally to the oculomotorius. Thus it became clear that the numerical calculation of nerve fibers does not afford any reliable basis to decide the problem.

### B. FIBER-ANALYSIS OF THE NERVE BUNDLES IN QUESTION

As the second step, fiber-analysis was undertaken upon the nerve bundles in question. Namely, all of the fiber-constituents were clas-

sified into three classes according to their diameter; thick, mediumsized and thin fibers, each being respectively more than  $5 \mu$ , between 5 and 2.5  $\mu$  and less than 2.5  $\mu$  in diameter. I determined the number



thick fibres medium-sized fibres fine fibres Fig. 10. Fiber-analysis of N. III and N. VI of a Blue Whale fetus

of fibers belonging to each class in the bundles a, b, Yand X as indicated in Fig. 9 b. My data are given in Table 2. Fig. 10 shows the result diagrammatically. Thus. the bundle b displays nearly the same fiber-composition as the bundle a and also the same as the trunk of the abducens (Y). If otherwise the trunk of the oculomotor nerve (X) exhibited a different fiber-composition from these three, it would tell almost undoubtedly that the bundle b is nothing but the distal extention of the bundle or anastomosis a.

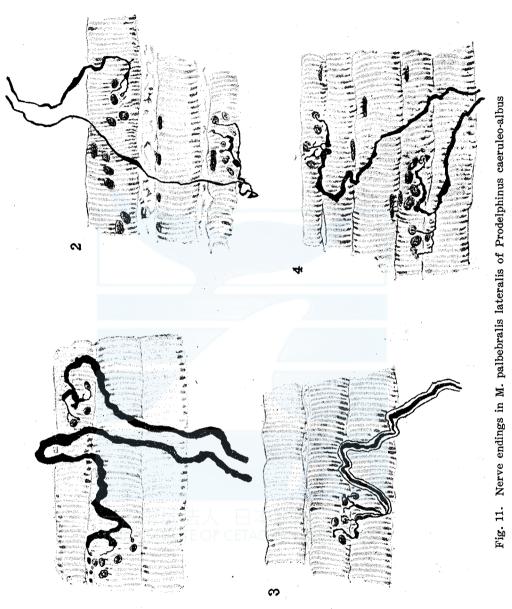
In reality, however, a striking resemblance was found to exist as to the fiber-composi-

tion between the abducens (Y) and the trunk of the oculomotorius (X), and therefore, it became impossible for me to know the relation between the bundles b and a by means of the fiber-analysis<sup>1</sup>.

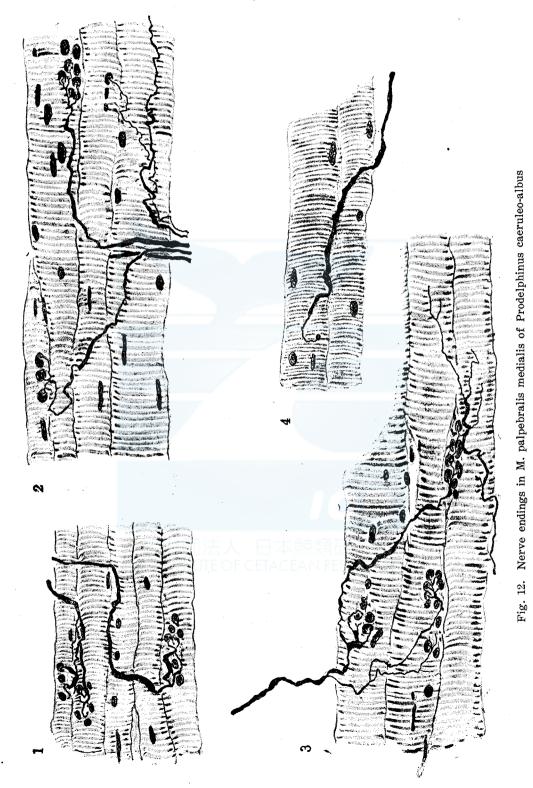
### C. NERVE ENDINGS OF THE ABDUCENS, COMPARED WITH THOSE OF THE OCULOMOTORIUS

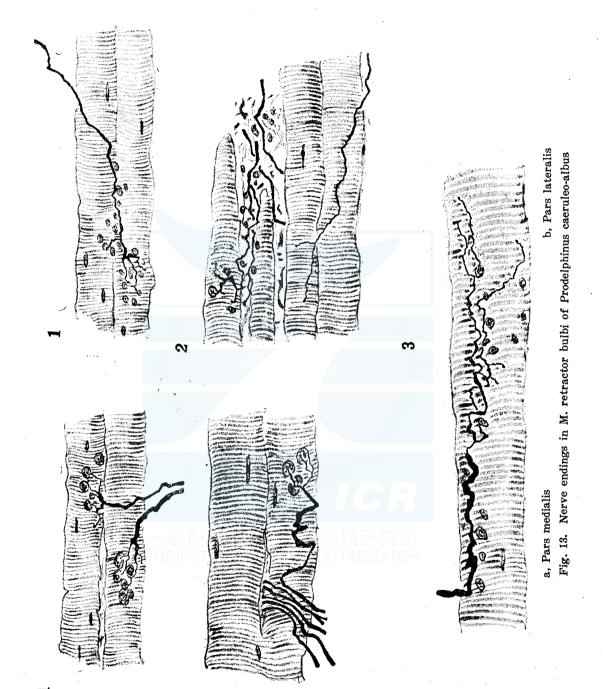
Thirdly, nerve endings were examined in each of the eye muscles in the whale. For, if there be, concerning this point, any noticable difference between the abducens and the oculomotorius, though this assumption is rather unplausible, it might be helpful for determining the nature of the bundle b, which innervates the medial portions of the retractor. Some of the nerve endings stained by the Bielschowsky method are illustrated in Fig. 11 (m. palpebralis lat.), in Fig. 12 (m.

<sup>1)</sup> Fig. 10 shows that the bundle b contains somewhat less fibers of larger diameter than the bundle a. It may be caused by the distal caliber-decrease of nerve fibers testified by Eccles, Sherrington (1930), Björkmann and Wohlfart (1936).



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palp. med.) and in Fig. 13 (m. retractor bulbi). A precise description of these endings and the comparative anatomical discussions upon them are not needed here, for we find no remarkable difference or peculiarity, compared with them in other animals.

The motor nerve endings in the extrinsic eye muscles have been studied by many authors, viz. Retzius (1892), Hamada (1928), Hines (1931), Woollard (1931), Yokomatsu (1932) etc., for various kinds of animals, but never for the whale. In the huge amounts of literature. however, no remarkable difference has ever been reported as to the anatomy of nerve endings between the trochlearis, the abducens and the oculomotorius. For, all of the previous authors who have studied this problem treated always the eye muscles as a whole, without paying attention to each muscle separately. I myself compared the terminal branchings, terminal buttons etc. between the abducens in m. palpebralis lateralis and pars lateralis m. retractoris bulbi and the oculomotorius in m. palpebralis medialis. But I failed to see any noteworthy difference between the two. All of these muscles exhibited very similar nerve endings, notwithstanding whether the examined muscle bundle belongs to pars medialis of the retractor, the medial palpebralis, to the lateral palpebralis or to pars lateralis of the retractor. The same histological research was performed also on the extraocular muscles of the cat, but with the similar results.

### D. MICRODISSECTION OF THE NERVE BUNDLES IN A DOL-PHIN, PRODELPHINUS CAERULEO-ALBUS

A kind of the sharply snouted dolphin, Prodelphinus caeruleo-albus Meyen, is very common to the sea near Izu Peninsula. And a good opportunity was given me to dissect the extrinsic eye muscles of this dolphin, and I found that, though the relations of nerves and muscles in question are quite the same as in other whales, branches from the oculomotorius to the retractor are relatively simple in this dolphin and the distance between the anatomosis a and the branching of bundle b is considerably shorter than in other whales. So I endeavoured to separate and trace the distal course of the bundle a along the oculomotor nerve. Using sharp pointed pincettes and needles under the binocular microscope, the bundle a was dissected distalwards. The result is shown in Fig. 14, which indicates clearly that the branches innervating the medial portions of the retractor are really continuous with the abducent nerve via the anastomosis. Thus finally I believe to have succeeded in proving that all portions of the retractor are innervated also in the whale by the abducens, though some of the nerve fibers supplying the medial portions of this muscle run temporarily in the trunk of the oculomotorius and look like branches from the latter apparently.

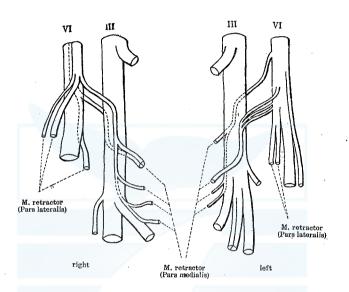


Fig. 14. Innervation of the M. retractor bulbi of Prodelphinus caeruleo-albus

# 5. Comparative anatomical study upon the innervation of the retractor bulbi

Then it occurred to me the question, whether the same relation holds good also in other animals. If so, the divergent statements of the previous authors reviewed in Chapter 3 might be easy to understand. In other words, if the anastomosis a, being situated in the depth of the orbita, be missed in observation, the peripheral branches would be taken very possibly for branches of the oculomotor nerve itself. For this sake the eyes of cats, dogs and rabbits were dissected with greatest caution. But, as shown in Figs. 15 and 16, such relations as seen in the whale were never present in these animals. Neither the anastomosis between the abducens and the oculomotorius nor branches which, arising from the latter, innervate the retractor were found. Hence this type of innervation of the retractor is for

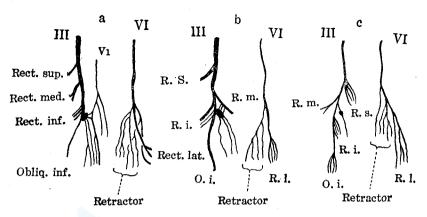


Fig. 15. Innervation of the M. retractor bulbi of cat (a), dog (b) and rabbit (c)

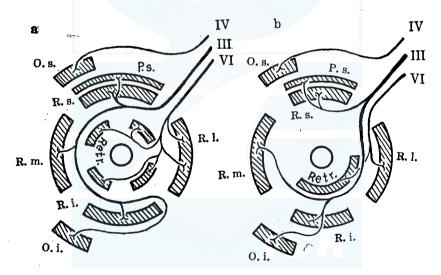


Fig. 16. Diagram to show the innervation of the extrinsic eye muscles of cat (a) and rabbit (b) (left eye, anterior view)

the present to be considered as peculiar to the whales<sup>1</sup>.

Additionally I wish to mention a bold assumption upon the cause why a number of authors such as Schwalbe (1879), Du-Bois Reymond (1907) etc. have incorrectly thought of the dual innervation of the retractor from the abducens and the oculomotorius. In the mammals

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<sup>1)</sup> For me it is difficult to understand, why Rapp, Stannius, Weber and Cords did not find this peculiarity in the whale. Phocaena and Hyperoodon which they used as the materials may lack in this peculiarity. But I can hardly surmise such a possibility.

examined by me, cat, dog and rabbit, some of the nervi ciliares breves arising from the ciliary ganglion go winded around the retractor or penetrate this before they reach the optic nerve. Because of this somewhat confusing course, they might have been taken as terminating in the retractor itself<sup>1</sup>.

### 6. Physiological studies upon the retractor bulbi of the cat

### A. INNERVATION OF THE RETRACTOR

According to Hopkins and Nishi, Foltz (1862) and Key-Abergs (1934) examined physiologically the innervation of the retractor respectively in the horse and in the rabbit. I myself carried out a few physiological experiments upon the retractor bulbi of the cat. The result is shown in Fig. 17. When the trunk of the abducens is stimulated with electri-

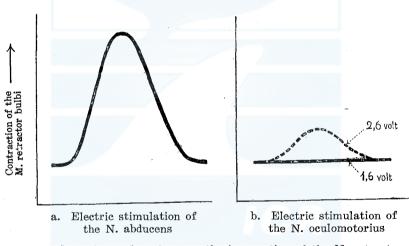


Fig. 17. Experimental study upon the innervation of the M. retractor bulbi (cat)

city, the retractor contracts intensely, while it does not show any contraction by stimulation of the oculomotor nerve. This observation is quite consistent with the macroscopic dissection illustrated in Fig. 15. In Fig. 17 *b* one sees a slight contraction of the retractor by stimulation of the oculomotorius. Perhaps, this may be due to collateral current, presence of which could be proven with the tester.

1) Cords said the same thing, too, though briefly (p. 243, foot-note 2).

### B. THE FUNCTION OF THE RETRACTOR BULBI

Together with the above mentioned experiments, I attempted to know the real function of the retractor muscle, for the name "m. retractor bulbi s. oculi" is one which was given by the anatomists such as Stannius, Benz, Gegenbaur etc. probably from purely morphological standpoint, and we have had seemingly no definite, physiological proof to show the appropriateness of this name. According to Cords, some authors assumed other functions for this muscle; for example, Milne-Edwards, Stannius, Straus-Durkheim, Le Double, Ellenberger-Baum thought that it might merely cooperate with the recti muscles. Motais and Martin supposed, on the other hand, that it might serve to support the eyeball for the purpose to allow the higher activity of the straight and oblique muscles (m. suspensorius oculi).

To answer this problem, I stimulated the nerves and muscles individually with the electrode, examining the movements of the eye-

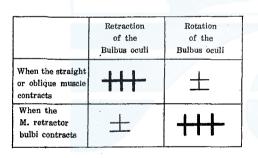


Fig. 18. Experimental study upon the function of the M. retractor bulbi (cat)

ball. The details of my results are omitted here. The conclusive remarks are represented by Fig. 18; that is, the recti and obliqui muscles rotate the eyeball by their contraction, but do not retract the eyeball at all, while the retractor muscle pulls backward the eyeball strikingly, without causing any rotation.

Thus, it became doubtless

that the retractor bulbi serves almost exclusively for the retraction of the eyeball; in other words, it deserves the name quite adequately also from the physiological aspect.

### 7. Motor nucleus of the retractor bulbi in the brain-stem

The intramedullar location of the motor nucleus for the retractor bulbi has not yet been determined. According to Kappers, Huber and Crosby (1936), some authors have accounted the so-called accessory nucleus of the abducens for it. But this nucleus, which generally exists in mammals including man (Terni, Preziuss, Addens etc.), and on the other side interpreted as the "dorsal facial nucleus" (van Valkenberg, Kappers), does not seem in reality to send fibers either in the facial or in the abducent nerve. Furthermore, if we recollect that the retractor bulbi is absent in the primates including man, while this cell group is well developed in them, it is quite unreasonable to regard those nerve cells as the nucleus innervating the retractor. In my opinion, such a nucleus may exist somewhere in the brain-stem, probably occupying a portion in the nucleus nervi abducentis. The reason, why I supposed so, is thus:

The fibers of the abducens going to the retractor amount to about 2,000 in a fetus of the Blue whale, making nearly one third or one fourth of all the fibers of this nerve (Table 2), and they can be followed proximalward as relatively definite parts within the trunk of the abducens<sup>1</sup>. In other words, the nerve fibers supplying the retractor are well localized within the abducens. I guess, such a localization may also exist in its original nucleus.

So I studied at first the comparative anatomy of the abducens nucleus by serial sections stained with the Pal-Weigert method, paying special attention to the morphological difference of this nucleus between animals with and without the -retractor muscle. But no remarkable difference could be found, which is available for this purpose<sup>2</sup>.

Secondly, I performed degeneration-experiments using a few cats as the material. Because of technical difficulty, however, the operative removal of the retractor bulbi was not successful. So the lateral rectus muscle of one side was extirpated together with its nervous insertions; the retractor with its nerves remained intact. After the lapse of ten or fourteen days the cat was killed and the brain-stem was fixed in alcohol,  $20 \mu$  thick sections were prepared serially and stained with thionin. At the same time, utilizing the osmic acid, the retrograde degeneration of the lateral rectus fibers of the abducens and the intactness of the retractor fibers were examined. The serial sections of the brain-stem revealed, however, no marked change either in the root fibers of the abducens or in the nerve cells of the original nucleus of the abducens.

It seems, injuries on such peripheral extremities of root fibers do not bring forth any remarkable tigrolysis in their original nerve cells. But the cells in the rostral portion of the abducens nucleus showed a

<sup>1)</sup> In the abducent nerve of the cat, I found the retractor supplying fibers numbered about 300, occupying approximately one fourth of the total fibers of about 1200.

<sup>2)</sup> Concerning the comparative anatomical study of the nucleus n. abducentis, Fuse's detailed work published in 1912 teaches us very much.

slight tencency of tigrolysis on the operated side, whereas the caudal portion looked perfectly intact. At present I am inclined to think, that the rostral part of this nucleus may supply fibers to the lateral rectus, while the caudal portion may send them to the retractor muscle. Naturally I can have no definite opinion upon this problem.

# 8. Sensory fibers of the retractor bulbi and of other extrinsic eye muscles in the whale

Though the retractor bulbi is said to have no sensory innervation (Huber, 1899, 1900, etc.), I observed in this muscle of the whales as well as of the cat many nerve endings, which are very much like the sensory ones. Some of them are illustrated in Fig. 13 (b, 2, 3). These endings, especially that shown in Fig. 13, b, 3 resembles to a high degree the grape-like ending (terminaison en grappe) described by Tschiriew (1879), who explained them as a young, undeveloped form of the motor end-plate. Retzius (1892) too called such endings as "atypical" motor ones. On the other hand, Bremer (1882), Huber (1899), etc, took them, being situated epilemmally, for sensory endings in the skeletal muscles, and also in Dogiel's excellent work on the sensory endings in the extrinsic eye muscles (1906) similar endings were described as sensory and illustrated very precisely. Though some recent anatomists, Hines (1931) etc., were of the opinion that those endings, lying sometimes hypolemmally or coming from an axis cylinder which constitutes a medullated nerve fiber continuing to motor end-plates, might be motor in nature, it is more generally believed that at least some of those grape-like endings are of the sensory character (Kulchitsky, 1925; Hinsey, 1927; Woollard, 1931 etc.)<sup>1,2</sup>.

My macroscopice] observations upon the sensory nerves to the retractor of the whale, are shown in Fig. 19; the ophthalmic nerve  $(V_1 \text{ in this figure})$  is seen supplying thin branches to the retractor<sup>3</sup>. In

3) The ophthalmic nerve of the whale is composed of two main branches. One of them takes the course similar to the nasociliary nerve in other mammals and man  $(V_{1a} \text{ in Fig. 19})$ , while the other appears to correspond to the lacrimal nerve  $(V_{1b} \text{ in Fig. 19})$ .

<sup>1)</sup> Boeke (1927) said that, though the most of grape-like endings are either motor or sensory, some of them are of the sympathetic nature.

<sup>2)</sup> In relation to the sensory endings in the extrinsic eye muscles, Cooper, Daniel and Whitteridge's recent paper (1949) is very important. By recording impulses in the nerve from the inferior obliquus of goats during stretch or active contraction of the muscle, they obtained physiological evidence for the presence of proprioceptors in this muscle. They ascribed however such a proprioceptive function to the muscle spindles.

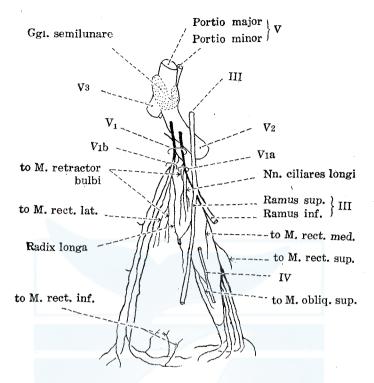
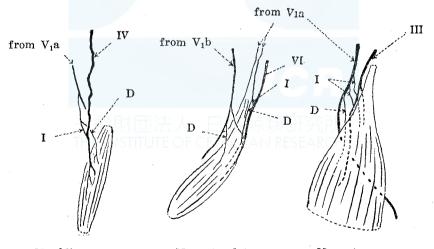


Fig. 19. Sensory innervation of the external eye muscles of the whale (1) (Fetus of the Sperm whale)



M. obliq. sup.M. rectus lat.M. rectus sup.D: direct insertionI: indirect insertionFig. 20. Sensory innervation of the external eye muscles of the whale (2)

this figure, furthermore, the sensory branches to other extrinsic eye muscles are illustrated. Examining these nerves, a remarkable feature quite different from the case in man is noticed. For, as described well in Schwalbe's textbook (1881), in man the sensory fibers to the extrinsic eye muscles, branching from the trunk of the ophthalmic nerve near the sinus cavernosus or at the entrance of the orbita, go immediately into the trunk of the oculomotorius, trochlearis or of the abducens and run together with those motor nerves until their destinations. In the whale, on the contrary, the sensory nerves course independently for a long distance until very near their insertions to the muscle. Though sometimes they run along with the motor nerves, the adhesion or union takes place only in the very vicinity of their terminations (indirect insertion in Fig. 20). Even in such cases, some of the sensory nerves reach the muscle quite independently from the motor nerves (direct insertion in Fig. 20).

Concerning the sensory fibers of the extrinsic eye muscles of rabbit, cat and monkey, Tozer and Sherrington (1910) stated that nearly all of them run from beginning to end of their ocurse within the motor nerves such as trochlearis, oculomotorius and abducens, while the trigeminal nerve has nothing to do with the sensory innervation of these muscles. Woollard (1931) and Hines (1931) also said the same for rabbit, dog, cat, rat etc. Their reason for this conclusion is, that almost all of the intramuscular nerve endings, both of motor and sensory nature, disappeared, after the motor nerves were severed at their exits from the brain-stem. According to Hines, those sensory nerves have their origin in the upper portion of the nucleus mesencephalicus of the trigeminal nerve.

In the present work no definite evidence was obtained to settle the problem, whether the extraocular muscles of the whale receive also such sensory fibers other than the above mentioned branches issuing from the ophthalmic nerve.

### Summary

1) The musculi palpebrales of the cetacean eye are of an unique existence throughout the animal kingdom. The motor innervation of them is quite the same as that of four straight muscles, which insert to the eyeball; namely, the lateral palpebralis is supplied by the abducens, while the other three are innervated by the oculomotorius.

2) The musculi obliqui and the innervations of them show nothing

peculiar in the cetacea, with one exception that the ocular insertion of the inferior obliquus is sometimes divided into two portions.

3) The ciliary ganglion, the presence of which has not yet been clarified in the cetacea, exists in all of Mystaco- and Odontoceti treated in the present work, though always rather in vestigial conditions.

4) The retractor bulbi is remarkably well developed in the whales, and is divided into four portions: Pars superior medialis and lateralis, Pars inferior medialis and lateralis.

5) The retractor bulbi of the whale is apparently innervated doubly by the abducens and by the oculomotorius, each nerve supplying the lateral and medial portions of this muscle respectively. But the branches innervating the medial portions are, though they appear to arise from the oculomotorius, nothing but fibers which belong essentially to the abducens and have coursed only temporarily in the trunk of the oculomotorius. Namely, all the portions of the retractor are supplied in the whale exclusively by the abducens just like in all other animals, whose retractor receives direct branches from the abducens only. Such a temporary course of the abducens fibers via the oculomotorius trunk is very peculiar for the cetacea.

6) The name "musculus retractor bulbi" is appropriate for this muscle, as my experiments on the cat's eye resulted, that its contraction caused a striking retraction of the eyeball, while the recti and obliqui muscles brought forth merely rotating movements of the eyeball.

7) There seems to be some possibility that the nerve cells supplying motor fibers to the retractor are localized in the caudal part of the nucleus n. abducentis.

8) The retractor bulbi as well as the other extrinsic eye muscles receives in the whale sensory innervation from the ophthalmicus.

9) The sensory nerves of the extrinsic eye muscles show in the cetacea a different feature from those in man. In the former they run from the ophthalmic nerve quite independently until their insertions to the muscles, while in the latter the sensory nerves course nearly through the whole length united with the motor nerves, viz. oculomotorius, trochlearis, and abducens.

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### Histological Studies on the Respiratory Portions of the Lungs of Cetacea

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### INTRODUCTION

The histology of the lungs of aquatic mammals have been studied by a number of previous authors, FIEBIGER (1916), NEUVILLE (1928), WISLOCKI (1929), BÉLANGER (1940) etc. In the present paper my own histological studies on the pulmonary divisions from the bronchoile to the air spaces in several kinds of the cetacea will be mentioned. The whales treated here are three species of Odontoceti, Prodelphinus caeruleoalbus MEYEN, Berardius bairdii STEJNEGER and Catodon physeter L. (Sperm whale) and three kinds of Mystacoceti, Balaenoptera physalus L. (Fin whale), Balaenoptera borealis LESSON (Sei whale) and Balaenoptera musculus L. (Blue whale).

The Prodelphinus caeruleo-albus was caught near the coast of Izu Peninsula, the Berardius near Shirahama in Tiba Prefecture and the Sei whale near Kinkazan in Miyagi Prefecture, while the Sperm, the Fin and the Blue whale were all captured by the boats of Nisshin-Maru in the Antarctic Ocean.

Blocks cut from the lungs were fixed in 10% formalin, embedded then in celloidin and made into serial sections of  $20 \mu$  and  $100 \mu$  thickness. The  $20 \mu$  sections were stained alternately with haematoxylin-'eosin and WEIGERT'S resorcin-fuchsin, while the  $100 \mu$  sections were stained only by the latter method for the study of elastic fibers.

### MICROSCOPIC OBSERVATIONS

### Toothed whales:

### A) Prodelphinus caeruleo-albus MEYEN

The bronchiole is 1.5–2.0 mm in diameter, and is lined by a continuous ciliated columnar epithelium stratified in two or three layers. In the connective tissue two elastic layers are present; the inner of them, longitudinally directed, pertains to the tunica propria, while the outer elastic layer, surrounding and connecting the cartilages together, is situated in the submucous layer. There are no bronchial glands. Many blood vessels are seen between the two elastic sheets. A few smooth muscle fibers exist in the subepithelial connective tissue in the parts of the bronchiolar wall, where the hyaline cartilages are lacking.

As WISLOCKI described in the porpoise, the terminal bronchiole gives off several branches, which undergo sudden change in the structure of the wall, when they have decreased in thickness to a diameter of 1 mm, such that well developed circular bands of the smooth muscle cause remarkable eminences of the mucous membrane into the lumen. These circular eminences follow one after another at short intervals. On the at first appearing eminences, the epithelium changes from the high columnar to the non-ciliated, cuboidal type and the latter is altered here and there into the very thin respiratory epithelium, to which blood capillaries are directly attached.

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The circular muscular bands exist only at the intercartilagenous portion, between which the lumen forms small rounded chambers in a successively continuous row. This part corresponds probably to the main stem of the respiratory bronchiole. The proximal chambers of the row are from 0.5 to 0.7 mm in diameter. The main stem gives off usually several side branches at right angles and is divided at its end dichotomously into two side branches. Each side branch is subdivided repeatedly like the main stem (fig. 1).

The branches are also segmented in the row of successively continuous, rounded chambers bordered by the circular eminences. The main stem is 4-6 mm long, provided with 5-10 chambers, and gives off 5-7 side branches. Each side branch is 0.3 mm in breadth, 1.0-1.5 mm in length, and possesses 5 or 6 small chambers. The circular muscular bands lie between the inner and the outer elastic membranes, and correspond certainly to the sphincters already reported by FIEBIGER, WISLOCKI and BÉLANGER. Radial elastic fibers, traversing the circular muscles and being entangled with circular elastic fibers contained within the musculature, connect the two elastic membranes together. Here and there the cuboidal epithelium is replaced by a very thin membrane, to which blood capillaries are closely applied. That is to say, it is very characteristic for the dolphin's lung that the respiratory epithelium appears already in the bronchiolar wall provided with car-In other words, the cartilage reaches in this animal much tilages. deeper up to the respiratory bronchiole.

The side branches are in structure just the same as the main stem. Each of them communicates with 6 separate alveolar ducts through cleart-cut openings. Especially near the opening the cuboidal epithelium becomes fewer in distribution and numerous blood capillaries approach the nearly naked surface. The cartilages, which are of the hyaline nature, extend down to the opening, in the form of irregular outlined rings or of isolated pieces.

At the opening the inner and outer elastic membranes are joined together, and form a large myoelastic band containing a small amount of muscle fibers. From the edge of the terminal cartilages this band descends to the alveolar duct and supports its wall. Other large elastic bands arising from the outer membrane of the side branch reach also the alveolar duct. They encircle orifices of the alveolar sacs and of the alveoli which are projected from the wall of the alveolar duct. Each alveolar duct is cylindrical in form, 0.4 mm in diameter and 1 mm in length, and its wall is occupied by many round orifices encircled by the elastic band.

An alveolar duct sends off at the beginning two alveolar sacs simultaneously and is divided at the end dichotomously into alveolar sacs. At this terminal portion the duct has a spherical cavity which corresponds presumably to the atrium of MILLER. Each sac is an elongated canal of the length about 0.5 mm, its orifice is 0.2–0.3 mm broad and encircled by a large elastic band, from which fine elastic fibers are detached towards the alveoli belonging to the sac. The group of alveoli, which belong to an alveolar duct, is surrounded by relatively much connective tissue containing small blood vessels, and never communicates with the alveoli belonging to other alveolar ducts. An alveolar sac has nearly 20 alveoli.

An alveolus of the dolphin's lung is 0.1-0.15 mm in diameter and 0.15-0.2 mm in depth. Fine elastic networks surrounding the alveolus anastomose with elastic fibers of adjacent alveoli. On the surface of the alveoli very thinly extended membrane, probably a cytoplasmic part of the so-called cuboidal epithelial cells is seen over blood capillaries of the wall. Nuclei of the cuboidal cells are aggregated at certain places, which correspond to meshes of the capillaries (Nischenzellen).

The visceral pleura is about 0.1 mm thick and has the inner and outer elastic layers, in which elastic fibers run in various directions. Between the layers is a thick connective tissue with relatively large blood vessels. Slender elastic bundles arise from the inner elastic tissue and attain the wall of subpleural alveoli, uniting with the elastic network belonging to the alveoli.

### T. MURATA

# B) Berardius bairdii STEJNEGER

The interlobular bronchiole, having a high columnar ciliated epithelium, forms many longitudinal folds. There the bronchiole is about 1 mm thick and accompanied closely by a large blood vessel (branches of the pulmonary artery) running in a comparatively large amount of connective tissue. Between the hyaline cartilages of its wall circular bands of the smooth muscle are well developed.

When the bronchiole enters a lobulus, the columnar epithelium is changed to the cuboidal, non-ciliated one, and the cartilages already disappear. This early disappearance of the cartilage characterizes the bronchiole of this beaked whale. The circular muscles, the so-called sphincters, cause remarkable eminences of the mucous membrane into the lumen. By them the lumen is divided into several (4-9) chambers of various size which are serially arranged in a row (fig. 2). The respiratory epithelium is seen here and there in these chambers. Thispart makes possibly the main stem of the respiratory bronchiole. In its proximal portion the chambers are small and spherical, while more distally they are more enlarged and resemble the alveolar ducts. From the main stem many side branches are sent off at right angles. Each of them has also several chambers divided by the sphincters, and moreover it is repeatedly subdivided into smaller branches, all consisting of a series of chambers of various size. Each side branch leads to several alveolar ducts which constitute a mass of pulmonary substance well limited by the connective tissue against the alveolar ducts belonging to other side branches. Further detailed structures of the alveoli. especially of the respiratory epithelium are not clearly observable in this Berardius, as the present material was not sufficiently fresh at the fixation.

# C) Sperm whale

The bronchiole, about 1 mm in diameter, has a continuous layer of columnar, ciliated epithelium. The relatively thick tunica propria forms many longitudinal folds, while ring-shaped cartilages and a few circular smooth muscles encircle the wall.

By branchings the bronchiole becomes smaller and its columnar epithelium is changed to a cuboidal one. Here the lumen is very much narrowed by extraordinarily well developed sphincters (fig. 3). This portion, only ca. 1 mm long, corresponds probably to the respiratory bronchiole, from which the alveolar ducts are given off. The cartilages and the circular muscular bands are present up to orifices of the al-

# Histological Studies on the Lungs of Cetacea

veolar ducts. From the edge of the terminal cartilage myoelastic bands extend to the wall of the alveolar ducts, and make a chief constitueut there.

The alveolar duct, about 0.7 mm wide, is soon dichotomized. The interalveolar septa are well developed, nearly  $60 \mu$  thick, and have two sheets of blood capillary beds, each one on the internal surface of two neighbouring alveoli. Very thick connective tissue builds the wall of the alveoli, which do not communicate directly with the alveoli belonging to other ducts.

# Baleen whales:

# D) Fin whale

Smaller bronchioles, the internal passages of which are 1.5-2.5 mm wide, are branched off from the larger stem at acute angles. The epithelium is a high columnar ciliated one with polygonal basal cells, which are in contact to the basement membrane. Networks of fine elastic fiber are adjacent to this membrane. There are two elastic layers with relatively large blood vessels between them; the inner layer exists in the tunica propria, while the outer one surrounding and connecting the cartilages lies in the submucosa. Ring-shaped cartilages enclose the bronchiolar wall, and a few circular muscular bands are seen in the intercartilagenous portions.

The high columnar ciliated epithelium, after the bronchiole is repeatedly branched, is altered to a non-ciliated one. The bronchiolar wall has, between circular muscles, large outpocketings, which are covered by the respiratory epithelium (fig. 11). This part might be taken for the main stem of the respiratory bronchiole (fig. 4). It is about 1 mm thick and 7-8 mm long; it gives off 4 or 5 side branches at right angles and is at the end without diminishing the caliber dichotomized into two terminal branches. Each side branch measures nearly 0.5 mm in diameter and 1.5 mm in length. Blood capillaries attached to the respiratory epithelium are well developed (fig. 12).

In the main stem as well as in its branches a small amount of circular muscular bands are present between the two elastic layers, and cause very slight eminences into the lumen. But these eminences are much lower and more insignificant in comparison with the corresponding structure seen in the Odontoceti. Between them wide hollow alveoli are observed. The muscular band is traversed by radial fine elastic fibers, which mix complexly with circular elastic fibers contained in the band.

As the side branch has more muscles, the eminences appear here a little more remarkably, but anyhow they are not so high as in the dolphin's lung, and a row of chambers is not seen in the Fin whale. One side branch communicates at the terminal with 6 alveolar ducts through clear openings. In the terminal portion the ordinary cuboidal epithelium is found only here and there and numerous blood capillaries reach, nearly naked, just beneath the respiratory epithelium. Muscular bands encircle the entrance of the alveolar duct, where large myoelastic bands descend from the terminal cartilage to the wall of the alveolar duct. The irregular ring-shaped cartilages exist up to the entrance of the alveolar duct.

In the Fin whale confluence of the branched alveolar ducts occurs very frequently. The primary alveolar duct, 0.8 mm in diameter, gives off at first three secondary ducts and then another secondary one. The last one anastomoses with the more proximally dispatched one and the thus formed union of ducts sends off further other ducts which again unite with alveolar ducts given off from the other side branch (fig. 6). In this way the alveolar ducts themselves form an extensive reticulum. And we see at some places, where several ducts meet together, relatively wide chamber, which seems to correspond to the atrium of MILLER. The smaller alveolar duct shows the caliber of about 0.5 mm. The ducts are surrounded by large myoelastic bands coming from the edge of the cartilage at the entrance and also from the outer elastic layer of the side branch. This band encircles the orifices leading to the secondary ducts, and then those to the alveolar sacs (fig. 5). The wall of the alveolar ducts has many orifices, which are all bordered by large myoelastic bands. From existence of the smooth muscle fibers this portion must be called difinitely as "alveolar duct."

The alveolar sac is about 0.5 mm broad and 0.4-0.6 mm long. Around its orifice there is a large elastic band, from which fine elastic fibers go to the walls of alveoli. An alveolus is 0.2-0.4 mm in diameter and 0.2-0.4 mm in depth. Alveoli with small orifices are also relatively shallow. The alveolar wall'is built by fine elastic fibers arisen from large elastic band at the orifice. No large assembly of the connective tissue is found, bordering between the alveoli which belong to one alveolar duct and those belonging to other ducts. The interalveolar septum is about  $40 \mu$  thick, and have capillary beds separately on each surface (fig. 7). Round nuclei of the cuboidal epithelial cells are seen, especially gathered in corners of the alveolus. They are crowded also

# Histological Studies on the Lungs of Cetacea

corresponding to meshes of the blood capillaries, in various numbers according to the size of the mesh (fig. 8). A thin membrane, which is very probably nothing than the expanded cytoplasmic portion of these cells, covers continuously the capillary beds of the alveolar surface (fig. 9 and 10).

# E) Sei whale

The bronchiole, whose diameter is pretty large (ca. 2 mm), has a high columnar ciliated epithelium. In the submucosa there are two layers, inner and outer, of the elastic tissue and only a small quantity of the smooth muscles. The ring-shaped cartilages are all hyaline. With branching of the bronchiole the epithelium is changed to cuboidal, flattened one. These branches are only 1 or 2 mm in length and correspond possibly to the respiratory bronchiole. The tunica propria has fine elastic nets together with many blood capillaries and a few smooth muscles between the two elastic membranes. No eminences into the lumen are produced by the muscles. The respiratory bronchiole is divided at the terminal either into two parts which lead to each one alveolar duct, or continues directly without division to an alveolar duct.

Near the entrance of the alveolar duct the cuboidal epithelium suddenly disappears, and the apparently naked surface is occupied by numerous blood capillaries. At the same time two elastic membranes join together and form a large elastic band, with which muscle fibers are intimately mixed. The compact myoelastic band descends along the wall of the respiratory bronchiole to the entrance in question, where it is annular shaped. The cartilages extend also down to the same place. The alveolar duct is about 1 mm in width. The branched ducts are united directly with each other, forming a remarkable reticulum. The histological structure of alveolar ducts, alveolar sacs and alveoli resembles much that mentioned already in the Fin whale.

The about 2 mm thick pleura pulmonalis has in its middle greater portion elastic nets, large meshes of which are filled with the connective tissue and blood vessels. The outermost layer of the pleura is a thin collagenous plate, containing fine elastic fibers. The deepest part of the pleura has also a thin layer of fine elastic fibers, which are united with elastic fibers pertaining to the alveolar wall.

# F) Blue whale

The pulmonary portions, from the bronchiole down to the alveolar sac, show histologically no noticeable difference from that in other

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Balaenoptera. The bronchiole, about 2 mm in diameter, has a high columnar ciliated epithelium, and there are a few circular smooth muscles in the submucosa. It leads directly to the respiratory bronchiole having a cuboidal epithelium, between which here and there the respiratory epithelium appears. The tunica propria shows relatively a large amount of smooth musculature. The cartilage exists down to the end of the respiratory bronchiole, which is only 1 or 2 mm long. A large myoelastic band comes from the terminal cartilage and constitutes the wall of the alveolar duct. The alveolar ducts form an extensive reticulum by frequent branching and confluence.

# CONSIDERATIONS

One might be interested in knowing what characteristics the cetacean lung shows for adaptation to the aquatic life, considering especially that the whales can dive under water so long a time. And I myself, upon comparative studies of six species of the whales, was much impressed by some differences of pulmonary structure between them. Besides, I believe to have obtained a few important findings upon the much discussed problem of the respiratory epithelium of mammals.

1) With the remarks of previous authors that in the cetacean lung the bronchiolar cartilage is well developed and extends down to unusually deeper portion, I agree, so far as five kinds, the dolphin, Sperm, Fin, Sei, and Blue whales are concerned. In them the cartilages of the respiratory bronchiole persist down to the entrance into the alveolar duct. But it is worthy of notice, that only in Berardius the hyaline cartilage is present in the interlobular bronchiole and as soon as this enters a lobulus, the cartilages disappear from the wall. WISLOCKI and FIEBIGER described in porpoise and dolphin that the cartilages extend as a complicated, but well developed armature down to the openings into the air sacs, NEUVILLE also mentioned in Steno and Delphinus that the cartilages persist up to the end of the bronchiole, and BÉLANGER said in the larger cetacea that the cartilagenous armatures extend down to the openings into the respiratory sacs.

2) Concerning the bronchiolar smooth muscles, I observed, that circular muscles are remarkably well developed in the respiratory bronchiole. But there is a great difference between Odonto- and Mystacoceti. In Prodelphinus and Berardius they are the best developed, forming the sphincters, and in the Sperm whale well developed circular muscles extend down to the distal end of the short respiratory bronchiole, narrowing very much its lumen. But in the baleen whales, the development of the circular muscles is in general very poor; only the Fin whale has relatively much of them, while in the Sei and the Blue whale they are quite rudimentary. So it cannot be said simply that the cetacean lungs have always very well developed smooth muscles.

In the literature we read no clear description about such difference. FIEBIGER said in the dolphin that the circular muscles extend from the bronchiole of less than 0.5 mm in diameter to the entrance of the air sacs, and WISLOCKI remarked in the porpoise that the sphincter muscles occur up to the termination of the respiratory bronchiole. NEUVILLE reported in Delphinus and Steno that the sphincters extend to the distal end of the bronchioles, while BÉLANGER said in the larger cetacea that from the muscle fibers encircling the bronchiole, large bundles descend along the first portion of the air sac.

3) I can completely agree with the opinion of some previous authors (FIEBIGER, WISLOCKI, BÉLANGER), that in cetacean lungs elastic fibers are abundant. In the present study, in the parts below the alveolar duct large elastic bands build the wall. But here is also a difference, as in the baleen whales the bands contain a great amount of muscle fibers and might be well called "myoelastic", while in the toothed whales the muscles contained in the bands are very few.

4) FIEBIGER, WISLOCKI and BÉLANGER did not say about the existence of the alveolar ducts in the toothed cetacea, such as dolphin, porpoise and the white whale, while BÉLANGER described it in the baleen whale. I recognized definitely that also the toothed whales have in their lungs the alveolar ducts, built by large elastic bands together with a few smooth muscles. In the dolphin, an alveolar duct gives off four air sacs. The number of alveolar ducts, to which an end portion or a side branch of the respiratory bronchiole leads, is different according to the species; viz. in Prodelphinus and in the Fin whale a side branch has 6 openings, and in Berardius several openings to continue into each one alveolar duct, while in the Sei, Blue and Sperm whales a respiratory bronchiole has only one or two openings to lead directly into each one alveolar duct.

In my opinion, there exists another great difference between Odonto- and Mystacoceti, that is the difference concerning the reticular formation of the alveolar ducts. J. HUNTER reported for the first time the presence of communicated passages between alveoli in the cetacean lungs. Since then many authors have studied upon this problem. Recently, NEUVILLE remarked that in Delphinus the interalveolar com-

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munication is present, but not in Steno. In all of the toothed whales examined by me, well developed connective tissue indicates usually a sharp boundary between the alveoli belonging to an alveolar duct and those of the other neighbouring duct, with no direct passage between them. On the other hand, it is a characteristic for the baleen whales, that the alveolar ducts communicate with one another to a high degree, forming a reticular canalsystem. BÉLANGER, who saw the alveolar ducts in the baleen whales, said nothing about such communication.

5) For the much discussed problem on the alveolar epithelium the cetacean lungs deserve attention, though the previous researchers mostly have not noticed this point, except BéLANGER who mentioned that the subpleural alveoli are lined with a continuous layer of cuboidal epithelial cells with deeply stained nuclei. In my study, not only in the subpleural alveoli, but also in the alveoli adjacent to large blood vessels or to bronchioles a continuous layer of cuboidal epithelial cells is frequently seen, moreover in the alveoli nuclei of the epithelial cells are densely crowded in corners, or in capillary meshes, and over the blood capillaries the cytoplasmic portion of these cells is expanded as a very thin, continuous layer, representing seemingly the so-called non-nucleated plates of the respiratory epithelium.

The discussions upon the respiratory epithelium started with ELENZ (1864) and EBERTH (1884), when they injected silver nitrate solution through trachea into lung of an animal, and made their classical pictures of what KOELLIKER later called the respiratory epithelium. KOELLIKER described for the human lung a continuous epithelium consisting of small nucleated cells and large non-nucleated plates. OppeL (1905) took the latter for a part of the former, denying the independency of the non-nucleated plates. Later OGAWA (1920) from studies upon several sorts of laboratory animals reasserted in general the view of ELENZ, EBERTH, and KOELLIKER. But LANG (1925) found in the rabbit's lung by tissue culture combined with vital staining, that the small nucleated cells arise in the connective tissue of the interalveolar septa. The hypertrophied and into the alveolar lumen migrated cells are phagocytes, taking up a large quantity of the dye. He insisted that the nucleated cells in meshes of the blood capillaries are of mesenchymal origin, and called them "Septumzellen." Also POLICARD (1926) denied the presence of the respiratory epithelium. Questionings have been expressed by some other authors on the epithelial character of it and the opinion has more or less prevailed that the cells might be histiocytes derived from the connective tissue and that blood capillaries of the alveolar wall might be directly exposed to the air.

SEEMANN (1925) found by vital staining in the lung of mouse that the histiocytes in the septa take up grosser particles of the dye, but the epithelial cells take up only finer ones, and thought that the cells which perform phagocytosis in the alveoli are of the epithelial nature. calling them "Nischenzellen." CLARA (1936) observed the same and discriminated between histiocytes and epithelial cells. He was of the opinion, that the alveolar wall has small nucleated cells, the so-called epicytes which are derived from the epithelial cells. **AKAZAKI** (1943) asserted on embryological or pathological studies of the human lung the views of SEEMANN and CLARA and concluded that the alveolar wall has dispersed epithelial cells. MILLER reported that lining the alveolar wall a continuous epithelium exists, made of thin, flattened, nucleated lamina. But LOOSI (1935) said that during the inflammatory process, cells exuded from blood or from connective tissue might assume an epithelium-like arrangement, and that such a cytoplasmic membrane, as MILLER found, could not be a proof for the existence of a membrane in the healthy alveoli.

Upon my findings, the so-called non-nucleated plate is a continuous membrane, nothing than a very thinly outstretched part of the cuboidal epithelial cells. So I can not agree with the views that blood capillaries and the connective tissue of the septa are naked, viz. in direct contact with air. At the same time I am inclined to deny an indepedent existence of the non-nucleated plates in the respiratory epithelium. But my observation is concerned only to the cetacean lungs, nevertheless it seems important it is well coincident with the photographs published by MILLER upon the alveolar epithelium of the human lung.

# SUMMARY

From histological studies upon the lungs of six species of the cetacea, Prodelphinus caeruleo-albus, Berardius bairdii, Catodon physeter, Balaenoptera physalus, B. borealis and B. musculus I reached the following conclusions.

1) The bronchiolar cartilages are well developed and extend down to the respiratory bronchiole, to the places, where the alveolar ducts begin. But exceptionally in Berardius they disappear far up, already at the end of the interlobular bronchiole.

2) Between Odonto- and Mystacoceti there is a remarkable diffe-

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rence upon the development of the smooth muscular fibers in the respiratory bronchiole. In Prodelphinus and Berardius the circular muscles are very well developed, causing high eminences of the mucous membrane inwards ("sphincters"), and the lumen of the respiratory bronchiole is divided into a series of chambers. In the Sperm whale the respiratory bronchiole is short, but has much of circular muscles, narrowing its lumen. In the baleen whales, the muscles are in this part of the bronchiole very poor, only in the Fin whale they are relatively much. The serially arranged chambers do not exist in all of the Mystacoceti examined by me.

3) The elastic fibers are also well developed in the cetacean lungs. By staining of them the configuration of the alveolar ducts could always be seen very clearly, though previous authors have not mentioned the existence of the alveolar ducts in Odontoceti. As to the numerical relation between the respiratory bronchiole and the alveolar ducts, six kinds of the cetacea are classified into two groups, Prodelphinus, Berardius and the Fin whale on one side, the Sei, the Blue and the Sperm on the other side. In the former the respiratory bronchiole is relatively long and opens into several (in the dolphin and Fin whale about six) alveolar ducts, while in the latter it is short and communicates with only one or two alveolar ducts.

4) In the toothed whales the reticular anastomosis of the alveolar ducts between themselves was not ascertained, but in all of the baleen whales examined it occurs to a high degree. That is to say, a reticular formation of the alveolar ducts is very remarkable at least in Balaenopteridae.

5) In all of Balaenoptera and Catodon, the interalveolar septum is thick, having much connective tissue, covered by separate beds of blood capillaries on each surface, while in Prodelphinus and Berardius it is thin and has blood capillaries, which work probably commonly to both neighbouring alveoli.

6) The respiratory epithelium is stretched over the septum in a continuous membrane. Round nuclei of the cuboidal epithelial cells are observed in groups of various size at corners of the alveoli or at places corresponding to meshes of the capillary nets, while the thinly extended portions of their cytoplasma form a membrane covering directly the blood capillaries, and is probably nothing than the so-called "non-nucleated plates."

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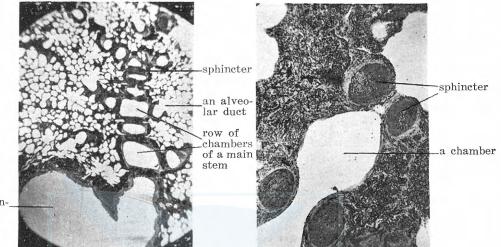
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a bronchiole

Fig. 1. (Prodelphinus) Divided chambers of the respiratory bronchiole. (van Gieson) ×10

Fig. 2. (Berardius) A row of chambers and sphincters of the respiratory bronchiole. (H. E.)  $\times 50$ 



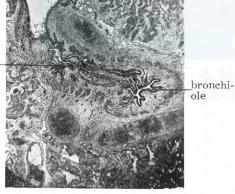
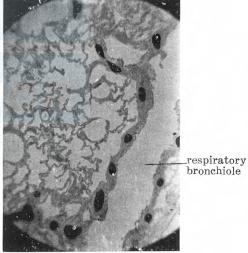


Fig. 3. (Sperm whale) Narrow portion of the respiratory bronchiole. (H. E.)  $\times 30$ 



bronchiole

Fig. 4. (Fin whale) Not narrowed lumen of the respiratory bronchiole. (H. E.) ×10



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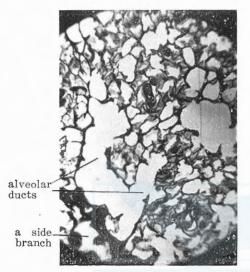


Fig. 5. (Fin whale) Alveolar ducts (Elastic staining) ×10

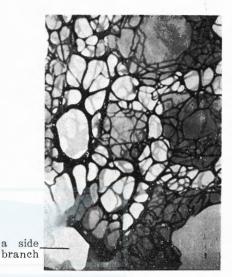
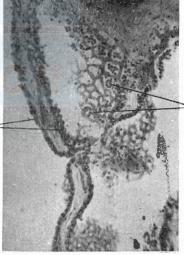


Fig. 6. (Fin whale) Reticular anastomosis of alveolar ducts, which are light coloured in this model reconstructed from serial sections by drawing only large elastic bands.  $\times 12$ 



Fig. 7. (Fin whale) Alveolar septa with separate beds of blood capillaries on both surfaces. (H. E.)  $\times 35$ ,

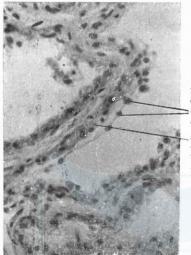
blood capillaries on each = surface of the septa



"Nischenzellen" (cuboidal epithelial cells)

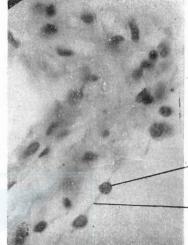
Fig. 8. (Fin whale) Nuclei of cuboidal epithelial cells densely crowded in meshes of the blood capillaries. (H. E.)  $\times 95$ 





nuclei of cuboidal epithelial cells

cross section of a blood capillary

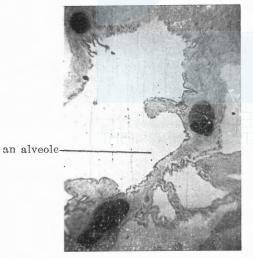


a round nucleus of cuboidal epithelial cell

the thin membrane

Fig. 9. (Fin whale) A thin membrane covers continuously the sheet of blood capillaries. (H. E.) ×190

Fig. 10. (Fin whale) A part of fig. 9, higher magnified. (H. E.)  $\times 450$ 



respiratory epithelium blood capillaries

Fig. 11. (Fin whale) An alveole belonging to the respiratory bronchiole. (H. E.)  $\times 35$ 

Fig. 12. (Fin whale) Respiratory epithelium (higher magnification of a part of fig. 11). (H. E.)  $\times 70$ 

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# On the Brain of the Sperm Whale (Physeter Catodon L.)

# By

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During the Antarctic expedition on board the Japanese whaling factory ship "Nissin Maru No. 1" in the season of 1949-50, I have undertaken to investigate the brain of the Sperm whale upon the following points.

- (I) Brain weight
- (II) Macroscopical observation on the cerebral hemispheres especially on the fissures and the gyri
- (III) Cytoarchitecture of the cerebral cortex

The brain weight (I) was surveyed on the deck, while the other studies (II and III) were later performed at the Brain Institute, University of Tokyo, upon materials fixed in formalin.

In the Antarctic Ocean we captured none of female and infantile Sperm whales but only males of over 40 feet length. During the expedition the Nissin Maru could get 172 Sperm whales, in which only 7 (4.1%) were sexually immatured.

# (I) Brain weight

The brain was taken out from the huge cranium, using such tools as chisel, gimlet or hatchet, after the bone was cut by a bone-saw into a block properly shaped for extraction of the brain, or after sagittal sectioning of the cranium into halves, two separated parts of the brain were gathered.

The brain weight was measured with the steelyard (the measure limit: 100 g), without stripping off the pia mater, because the brain substance is too soft to isolate it safely from the pia mater.

As the spinal cord was usually transected at the occipital condyles, its upper part, more rostral than the place of the IV cervical nerve roots, was attached to the brain.

The brain weights of 16 Sperm whales, of which the average body length is 49.8 feet, are shown in Table 1. This average body length is nearly equal to that (49.9 feet) of the total (172) Sperm whales captured. The brain of No. 3 (\*1) weighed very light (6.4 kg); as the

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		· · · · ·		1.1 L L L	
Date Measure		No. of the Sperm whale	Brain weight (kg)	Body length (feet)	Brain weight / length (kg) / (feet)
5/XII,	1949	1	8.0	50	0.160
11	, <i>11</i>	2	7.0	49	0.142
IJ	"	3	6.4 *1	54	0.118
"	"	4	7.3	49	0.148
7/XII,	11	5	7.7	50	0.154
Ņ	"	6	7.0	52	0.134
9/XII,	"	7	8.7	54	0.161
11/XII,	11	98	9.2 *2	49	0.187
IJ	"	99	8.6	46	0.186
12/XII,	11	108	9.0	51	0.176
"	11	110	8.0	49	0.163
14/XII,	"	122	7.0	49	0.142
15/XII,	11	123	8.0	54	0.148
IJ	IJ	125	7.0	51	0.137
"	"	153	8.0	51	0.156
"	11	154	8.2	49	0.167

Table 1. Brain weights, measured fresh, of 16 Sperm whales

cranium of this individual was fragile, the whale was estimated as a very old one, while the most heavy brain of No. 98 (\*2) belonged to an individual, which seemed to be in full maturity, as the brain substance and the cranium were felt very hard.

Naturally I had an interest in knowing the ratio of the brain weight to the body weight, but it was impossible to determine the latter; moreover, even during the short season from beginning till end of December the Sperm whales increased considerably in the body weight, as they eat much food in the meantime; therefore the body weight varied to a high degree.

On the other hand, the ratio of the brain weight (kg) to the body length (feet) was easy to determine. Its individual difference was great, between 0.118 as minimum and 0.187 as maximum, the average being 0.154. We know that the brain weight bears no direct ratio to the body length. Considering the fact that the Sperm whales caught in the Antarctic are all males and almost all of them are fully matured, the difference in question seems to have some relation to the degree of senility.

It should be noted here that scarcely any reference has ever been made to the brain weights of the Sperm whale, though those of some Mystacoceti and of other Odontoceti have been sometimes studied. But even in these cases they were surveyed under different conditions as to fixation, its duration, the meninges contained or not, and the age, often including embryos.

GULDBERG reported the brain weight of a Blue whale of 60 feet length, measured fresh inclusive the pia mater, arachnoides, and blood, as 6700 g, and the dura mater and rete mirabile of it as 3050 g. He said also that the brain fixed a few weeks in alcohol of a Blue of 64 feet length weighed 4673 g, and that the brain of a Fin whale together with the dura mater and rete miriable was 13680 g, after 8 weeks' fixation in alcohol. According to him, the brain weight had been measured by KNOCH and HUNTER in Balaenoptera rostrata, by SCORESBY and RUDOLPHI in Balaena mysticetus, and by ESCHRICHT in Megaptera boops. After all, we know in the literature GULDBERG's only one case. when the fresh brain was measured in a whale of over 50 feet length: other reports concerned always the weights after fixation, or the brain of smaller whales, mostly of dolphins and porpoises. The brain weights measured by me in Physeter seem to be, though they are quite difficult to compare, heavier than those of Mystacoceti of over 50 feet length.

According to GULDBERG, the brain weight loses about 1/3 after fixation in alcohol. I myself fixed pieces of the cerebral cortex of the Sperm whale in 10% and 20% formalin and followed how they change in weight by fixation; the results are shown in Table 2. I reached

·	in 10% f	ormalin	in 20% formalin		
Weight before fixation	21.5g	15.0g	9.8g	39.4g	
After 24 hours' fixation	29.2g	21.0g	13.2g	50.0g	
After 29 hours' fixation	31.2g	21.0g	13.2g	51.2g	
After 98 hours' fixation	31.0g	22.0g	13.2g	51.0g	
Ratio of increase	44%	46%	34%	29%	

Table 2.	Changes of th	e weight o	of brain	substance	during			
fixation in formalin solution (200 cc)								

the opinion that the ratio of increase is smaller as the size of the fixed material is larger and the increase stops nearly after 29 hours' fixation. In cases of such large materials as the cetacean brain the permeation of the fixing fluid occurs quite uncertainly and incompletely, for example we found that after 7 months' fixation in 10% formalin the brain of the Sperm whals showed very few infiltration of the fluid even in the relatively small outlined brain stem; therefore I think the weight after fixation is quite unreliable.

Now that the brain weights reported in the present paper include pia mater, blood vessels and the cranial nerves of various lengths are attached, they can never be said to represent the genuine weight of the brain substance itself. There can be errors of about 500 g.

# (II) Macroscopical observation on the brain especially on the gyri and the fissures of the cerebral hemisphere

# 1) Weight, volume and dimensions

The brain of a Sperm whale of 50 feet length was examined in details. It weighed 8.2 kg inclusive of the pia mater, which was attached to medial and basal parts of the hemisphere and to the brain stem.

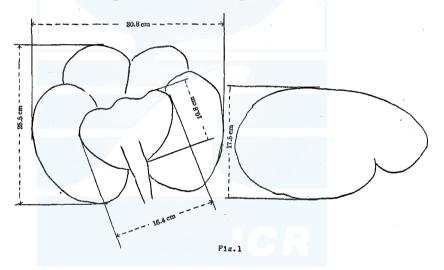


Fig. 1. Size of the brain of a Physeter.

The volume of the whole brain measured by the displacement of water is 8050 cc. The specific gravity as the whole is therefore 1.01. The brain was deformed to a certain degree during fixation in formalin. Especially the basal surface was pushed by the bottom of the vase and the vertical axis was shortened, while the transversal and sagittal axis became larger. Because of the deformation the following data must be a little different from the size of the brain in the living state (fig. 1).

Sagittal axis: 25.5 cm. This is the longest distance between the

frontal and occipital extremities. The difference between both hemispheres is very slight (left 25.4 cm, right 25.5 cm).

Transversal axis: 30.8 cm, with some asymmetry. The right hemisphere 15.8 cm; the left one 14 cm. It was already pointed out by GULDBERG and RAWITZ in Mystacoceti that the right hemisphere is broader than the left.

Vertical axis: 17.5 cm. This must be smaller than in the living animal, as mentioned above.

The ratio between the sagittal and the transversal axis is 0.822: 1, the latter surpassing the former. In the Mystacoceti (Balaenoptera sibbaldi, B. rostrata and B. musculus etc.) the ratio comes very near to 1:1. Therefore, the Sperm whale is extraordinarily brachycephalic.

The cerebellum is 10.8 cm sagittally and 16.4 cm transversally.

# 2) Brain stem and cranial nerves (fig. 2, 4 and 8)

In addition to the above mentioned specimen, I could use the brain of another Sperm whale for the morphological observation of the brain stem and cranial nerves. Viewed from antero-basal (fig. 4 and 8) the

telescoping of the brain is quite remarkable; that is, the axis of corpus callosum is directed at a right angle to the axis from midbrain to pons and the latter axis forms again nearly a right angle with the axis from pons to the upper part of the spinal cord. The spinal cord shows in its upper part a very unusual spatulate bending. It runs at first along the occipital edge of the cerebral hemisphere, reaching far above the sur-

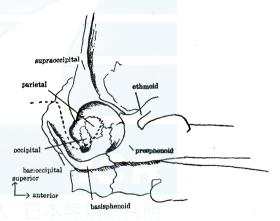


Fig. 2. Peculiar bending of the axis of brain stem (dotted line) projected to the lateral surface.

face of the hemisphere (fig. 2).

Cranial nerves:

The olfactory bulb and tract could not be seen. But in the other brain of the same species we noticed a mere trace of the nerve, which begins from the olfactory area and runs together with a very small vein along the olfactory fissure. The oculomotor nerve appears as a few small roots, ventro-medial to the cerebral peduncle and is immediately gathered into one bundle running ventrolateralwards.

On the basal surface of the brain:

The olfactory area (=substantia perforata anterior), relatively large and swollen, has a shallow transversal furrow, and lies between the under surface of the frontal lobe and the optic nerve. Its frontal edge, differently than in Mystacoceti, is not divided into ecto- and entorhinal fissures and borders mediad on the frontal lobe by a very shallow sulcus.

The cerebral peduncle is composed by two masses of fibers, the one running from dorso-lateral to ventro-medial, the other from dorsal to ventral, a furrow showing the border between the two. The size of them are different between both sides; on the right the ratio is 1 to 3, on the left on the contrary the former protrudes like a boal occupying about 2/3 of the whole peduncle. Besides on the left side the tractus peduncularis transversus of a transversal Y-form is seen, but no such a tract exists on the right side.

On the ventral surface of medulla oblongata we see a pair of olivary eminences, along the outside of which one or two small groups of longitudinal fibers (pyramidal tract) run uphead and seem to cross at the caudal end of medulla oblongata.

Up to the present the brain stem and cranial nerves of the Sperm whale have not been reported by any authors, though many works have been published on those of Mystacoceti and other Odontoceti (HUNTER, v. BAER, BRUNS, RAPP, MAJOR, HERBERT, BEAUREGARD, HASWELL, WILLIAM, GULDBERG, RAWITZ, MILLER, LANGWORTHY and ADDISON etc.). Among them the works of GULDBERG and RAWITZ on Balaenoptera rostrata and that of LANGWORTHY on Tursiops truncatus are especially interesting for comparison with the Sperm whale.

All of the Mystacoceti are said to have small olfactory bulb and tract, while the Odontoceti can be grouped into two; one group (embryo of Beluga, Hyperodon and Phocaena communis) having mere traces of them, and the other (Globicephalus melas, Delphinus delphis) showing no residue of them. The Sperm whale belongs from my observations to either of them, but more possibly to the latter group.

In Mystacoceti the oculomotor nerve comes out from the interpeduncular fossa, while in the Sperm whale it is a little apart from this fossa.

The two division of the cerebral peduncle above mentioned and

the existence of tractus peduncularis transversus have never been heard of the cetacean brains.

3) Gyri and fissures (fig. 3-10)

The configuration of the cerebral hemispheres of the Sperm whale looks at first very much complicated, but after a closer inspection, we recognize in the gyri and fissures a certain regularity which bears some definite resemblances to that in carnivores or ungulates.

Since GULDBERG published his classical work on the brain of the whalebone whales (1885), almost all of the workers have adopted his nomenclatures. On the dorsal surface of the hemisphere pretty parallel and more lateral to the interhemispherical fissure (Fit) the lateral fissure (F 1) runs and on the inner and outer sides of this we see F. endolateralis (F enl) and the F. ectolateralis (F ectl). About at middle of the lateral surface the F. sylvia is surrounded semicircularly by the the F. ecto- and F. suprasylvia and the anterior surface has F. olfactoria, F. coronalis and F. praesylvia.

My own observations will be described in the order of anterior, posterior, inferior, dorsal and medial surfaces of the hemisphere and such terms as "frontal", "occipital", "temporal", will be limited only to the places quite near to each pole, because boundaries between the lobes can scarcely be determined in this whale.

A) Fissures

a) Dorsal surface (fig. 3)

Nearly all of the fissures run here in sagittal direction without any large transversal fissures.

i) F. praesylvia (F pr)

Though it is not quite sure whether this fissure corresponds to that of carnivores and ungulates, I adopted this name considering its situation in relation to the whole hemisphere.

This fissure is asymmetrical, for on the right its anterior end lies lateral to the olfactory fissure, while on the left it is medial to this and moreover, on the right its posterior end does not reach the medial surface, while on the left it reaches almost the F. splenialis.

ii) F. cruciata (F cr)

In carnivores and ungulates this fissure courses quite transversely. But in the whales nobody has remarked about it, except LANGWORTHY, whose reference is but very insufficient. In the Sperm whale the F. cruciata runs almost parallel to the posterior part of the F. praesylvia and enters the medial surface. Its frontal part is not directed transversally, but compared with other fissures, it is the most oblique

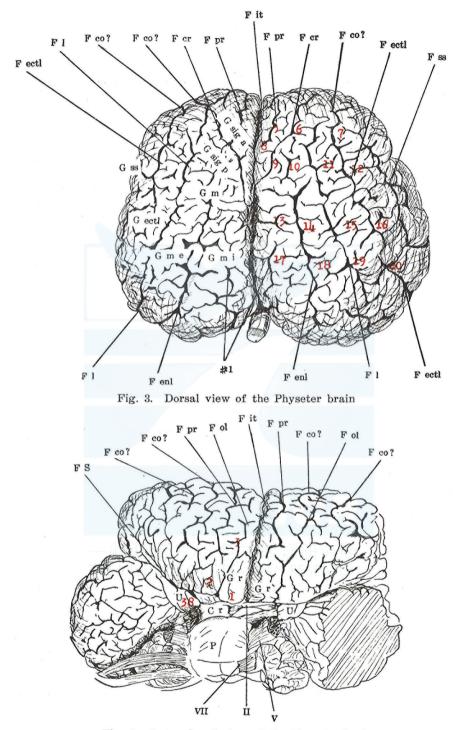


Fig. 4. Antero-basal view of the Physeter brain

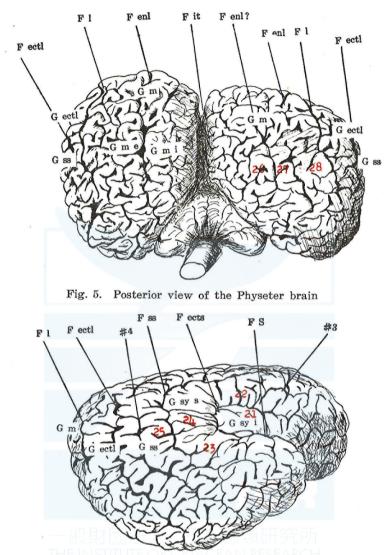


Fig. 6. The right cerebral hemisphere of Physeter, seen obliquely from postero-lateral

to the median plane, when it enters the medial surface, and continues to the F. suprasplenialis (fig. 9 and 10). RICHARD named a fissure probably corresponding to it "scissure endolatérale" in Globicephalus melas, but I take it for certain that this name should be given to a fissure, which lies along the F. lateralis and is other than the one now in question.

iii) F. endolateralis (F enl)=F. confinis (GULDBERG)

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This is a large nearly straight fissure between the pallial margin (Mantelkante) and the F. lateralis. On the right side it is interrupted at one place and runs around the occipital lobe to reach the basal surface after a long course, while on the left side it goes without interruption to the neighbourhood of the occipital pole, assuming there a complicated figure.

According to GULDBERG, this fissure is always present in Phocaena communis, either combined with F. splenialis or bent into the medial surface, but no such combination or bending was seen in the Sperm whale.

iv) F. lateralis (F 1)

This very large and uncrooked fissure is located about at middle between the ecto- and endolateral fissures. It runs quite symmetrically for a long distance along the lateral occipital part up to the lateral temporal portion. Though GULDBERG and RAWITZ pointed out in B. musculus and B. rostrata that its frontal end often continues to F. coronalis and F. praesylvia, as this continuance occurs in carnivores, in the Sperm whale there are no direct connections between them.

v) F. coronalis. (F. co)

We can locate this fissure only with much difficulty. It is very asymmetrical. On the left two crooked fissures run to the frontal surface after being interrupted by F. lateralis and F. ectolateralis, while on the right a fissure which is continuous with F. cruciata and ectolateralis reaches the anterior surface after being cut on the way as on the other side. TURNER mentioned some variations of the F. lateralis: it is continuous either with F. cruciata (Dictyles, a kind of Ungulata) or with F. lateralis (cats etc.); in some other animals it continues to neither of them. There are also individual differences.

In addition there are between the F. endolateralis and the interhemispherical fissure short fissures which run symmetrically (fig. 3, #1). Other small quite asymmetrical fissures do not seem to be of much significance.

b) Lateral surface (fig. 6 and 7)

Around the F. sylvia three semicircular fissures are arranged concentrically in the order from outside F. ectosylvia, F. suprasylvia and F. ectolateralis. Their forms are rather near to a circle than to a key, as this is the case in carnivores and ungulates.

vi) F. ectolateralis (F. ectl)

This is a quite symmetrical, large, very clearly defined sulcus, running from antero-medial of the frontal lobe to postero-basal of the temporal lobe, nearly parallel to F. lateralis.

vii) F. suprasylvia (F ss)

On the left, the frontal F it end of this fissure continues directly to about the middle of F. sylvia, while on the right it starts from a lateral part of the frontal lobe and runs parallel to F. sylvia, from which it is completely apart (fig. 6 and 7).

viii) F. ectosylvia (Fects)

This also is different between both halves; on the left it is directly connected with F. sylvia. Quite a long and distinct fissure, a little asymmetrical, runs along with F. suprasylvia and F. ectosylvia parallel to them. On the left a short sulcus connects F.

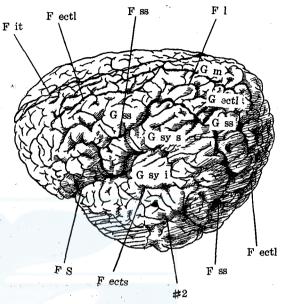


Fig. 7. The left cerebral hemisphere of Physeter: Lateral view

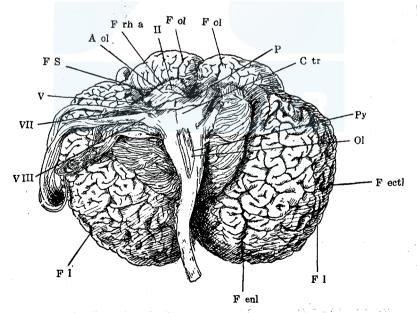
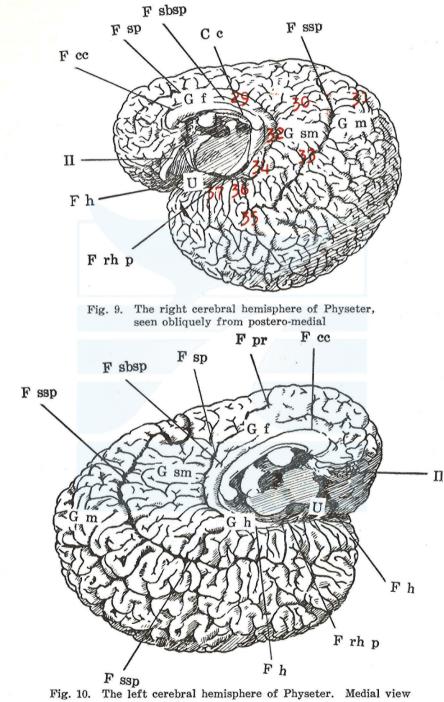


Fig. 8. Basal view of the posterior part of the Physeter brain



suprasylvia with F. ectosyivia in the temporal lobe (fig. 7, #2), while on the right in the lateral part of the frontal lobe a short sulcus runs between these two fissures (fig. 6, #3) and besides a long distinct sulcus courses from the frontal lobe to the temporal lobe (fig. 6, #4).

iv) F. sylvia (F S).

This is nearly symmetrical on both sides.

c) Posterior surface (fig. 5)

No symmetry is seen here. Especially the right shows very irregular sulci of very complicated form. On the left we see parallel to the pallial margin the endolateral, the lateral, and the ectolateral fissures run side by side, while on the right the endolateral, fissure is not found, but the lateral and the ectolateral fissures are present in the lateral border. On both sides sulci of the region between the pallial margin and the lateral fissure show very complicated curving and bending, except the left F. endolateralis, which courses very distinctly as mentioned above.

d) Medial surface (fig. 9 and 10)

x) F. suprasplenialis (F ssp)

This long fissure shows no great difference between both sides. It starts from F. cruciata and runs to about middle of the medial temporal surface in a large semicircular form. It gives off many small sulci at right angles. As discussed before, this corresponds in my opinion to F. suprasplenialis observed by GULDBERG in the Fin whale and differs from "scissure endolatérale" mentioned by RICHARD in Globicephalus melas. TURNER reported this fissure in Balaenoptera rostrata. But nobody seems to have found its continuance to F. cruciata. In carnivores and ungulates it is connected with F. splenialis or intercepted from this by a gyrus; so there is a possibility that the fissure in question corresponds to F. splenialis, but as a more probable F. splenialis is observed in a more basal region, I identified it to F. suprasplenialis.

xi) F. splenialis (F sp)

On both sides it begins from near the frontal pole and runs on the medial surface as if encircling the corpus callosum up to the gyrus hippocampi, which is at the medio-anterior end of the temporal lobe and united there with F. rhinica posterior.

xii) F. corporis callosi (F cc)

It demarcates the upper margin of the corpus callosum and its hinder part runs from the splenium along the medial border of the temporal lobe to the uncus, where it continues to F. hippocampi.

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xiii) F. subsplenialis (F sbsp)

This nearly straight fissure lies between F. splenialis and F. corporis callosi, and shows at the frontal end some asymmetry; on the right it is bent semicircularly around the genu corporis callosi, but on the left it begins from dorsal of the genu. On the medial surface no fissure, which seems to correspond to the calcarine, was observed.

e) Anterior and basal surface (fig. 4 and 8)

xiv) F. olfactoria (F ol)

It runs from the medial part of the anterior F. rhinalis almost parallel to the pallial margin, and behaves somewhat asymmetrically; on the right it lies medial to F. praesylvia, having few branchings, while on the left it is lateral to this fissure and shows a longer course with many side branches. As previously mentioned, owing to absence of the olfactory tract, the anterior F. rhinalis is not divided into two parts, in- and outside.

The uncus is subdivided into three portions by F. hippocampi and another shallow sulcus in the medial part of the posterior F. rhinalis.

Till now I have related only comparatively well defined fissures, while many others of more irregular, very complicated forms are present on the basal temporal and occipital surface.

B) Gyri

On the dorsal and lateral surface, bordered by the above named fissures, broad gyri are arranged concentrically around the posterior end of F. sylvia in the following order, from the median margin of the pallium to the Sylvi's fissure: G. medialis interna, G. medialis externa, G. ectolateralis, G. suprasylvius, G. sylvius inferior. Generally speaking, those gyri show at their terminal parts very complicated figures pari passu with irregularity of the fissures. They show also some notable asymmetries. The gyri, which border the obliquely directed F. cruciata from anterior and posterior, must be called G. sigmoideus anterior and G. sigmoideus posterior in analogy to the cases in carnivores. In the left hemisphere the G. medialis is divided on the posterior surface into internal and external portions owing to the presence of a distinct F. ectolateralis, while in the right such a division does not take place.

On the medial surface there are G. submedialis and G. fornicatus demarcated by F. suprasplenialis, F. splenialis and F. corporis callosi etc. The G. fornicatus is narrowed remarkably when it extends around the splenium and continues then to G. hippocampi, terminating in a swollen form (uncus). Besides we can see also G. dentatus and Fimbria hippocampi behind the medial part of uncus.

Medial to the olfactory fissure lies on the anterior surface G. rectus, which shows some differences between both sides owing to the asymmetry of F. praesylvia. The configuration in the lateral frontal surface is quite irregular.

A number of anatomists have hitherto studied the fissures and gyri in the cetacean brains, especially GULDBERG worked on Balaenoptera musculus, Balaenoptera sibbaldi and Phocaena communis, RAWITZ on Balaenoptera rostrata, RICHARD on Globicephalus melas, and TURNER on Monodon and Balaenoptera rostrata, and the similarities to carnivores and ungulates have been very often acknowledged.

According to my own observation the fissures and gyri of the Sperm whale are also similar to those of Mystacoceti, and of other Odontoceti. But I met with following several points, somewhat different from the mentioning of the foregoers.

As to F. cruciata, LANGWORTHY slightly referred to this in Tursiops truncatus, but no one has remarked its continuance to F. suprasplenialis. In the Sperm whale I found F. suprasplenialis in direct connection with F. cruciata.

The F. coronalis is very differently connected with other fissures according to the kinds of animals, sometimes with F. suprasylvia, with F. lateralis, or with F. cruciata, while its continuance to F. cruciata has been reported neither in Cetacea nor in carnivores. Also in my Sperm whale one hemisphere had two fissures probably corresponding to it, but their direct connection with F. cruciata was not seen.

The presence of F. olfactoria in the Sperm whale is interesting, for it has not been reported in Odontceti, carnivores and ungulates. It is likely that this fissure is present only in such animals as Primates, and Mystacoceti, which have the largely developed hemisphere and are at the same time microsmatic.

The direct continuance of F. corporis callosi to F. hippocampi is not known in pig, cow, horse and baleen whales, though it occurs in the elephant seal and man etc.; from this fact, considering also the relations previously mentioned of F. cruciata and F. coronalis, the Sperm whale seems to have more similarity to carnivores rather than to ungulates.

Two divisions of the uncus has been mentioned in some ungulates and it forms wrinkles divided by several fissures in the polar bear, while three divisions of it were found only in this whale.

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# (III) Cytoarchitecture of the cerebral cortex

1) Method. Materials fixed in formalin were cut into pieces of 1-3 cm thickness. Then by freezing  $20-40 \mu$  sections were prepared, put in 90% alcohol or acid alcohol, containing 10% of concentrated acetic acid and preserved about 24 hours in 37-38°C. After washing in water they were stained with 1% thionin for about half an hour at 40°C. Then they were washed in water again, and differentiated in alcohol.

2) Results

i) General observation

From various parts of the right cerebral hemisphere 38 pieces of the cortex with underlying white matter were cut off at regular and about equal intervals (1-8 of fig. 3, 4, 5, 6, 9 and photo A, B, C).

Compared with the cortex of other mammalia it is noteworthy in this whale that the nerve cells are scarce and their differentiation in 6 layers is not fully developed.

Especially the development of the layers IV, V and VI is comparatively poor and in most places the layers IV and V are not well distinguishable from each other and so the cortex seems to have only 5 cell-layers.

Each of the layers will be described in the following.

Layer I, Lamina zonalis:

This layer is very inconstant in thickness (0.24–0.80 mm); the thickness is different also between top and sides of a gyrus.

Here a number of glia cells are dispersed and though scarce, small round nerve cells are observed. Besides, though it is of great rarity, we see sometimes large nerve cells of  $10/30 \mu$  (the largest diameter along the cortical surface, the largest diameter vertical to the cortical surface) (9, 13). This layer is sharply distinguished from the next deeper layer.

Layer II, Lamina granularis externa:

Of all layers this is the narrowest (0.08–0.20 mm), but very densely populated with cells and clearly distinguishable; most of the cells are either round granular or pyramidal, of the size  $5/15-18/20 \mu$ , and are arranged regularly. This layer is present in all places examined.

Layer III, Lamina pyramidalis externa:

This is well discerned from the more superficial layer but its border against the deeper layer is indistinct. Generally it is thick (0.4-0.6 mm), has scattered granular cells of various size  $(9/15-24/18 \mu)$ and pyramidal cells exist in a larger number. The deeper within this layer, the larger become the cells, the form of which is mostly polygonal or triangular, though sometimes large round cells are also seen and the density of them is different according to the places observed.

Layer IV, Lamina granularis interna:

The granular cells, whose presence characterizes this layer in other mammalia, are not especially gathered here. Though in the deeper portion comparatively large cells are flocked together, I am not sure whether they really belong to this layer.

Layer V, Lamina pyramidalis interna:

The thickness (0.2-0.4 mm) is nearly constant in all places. There are comparatively large cells (about  $18-24/30 \mu$ ) mostly of polygonal or pyramidal form, and they are much crowded. In some places this layer has giant pyramidal cells constituting a very clear layer, while in other places, where no such cells are present, it is difficult to determine the border against the more superficial layers.

Layer VI, Lamina multiformis:

It is 0.8-1.0 mm thick. Here are scattered relatively large pyramidal, round or stellate cells of  $18-24/30-24 \mu$  and many other forms.

ii) As the next step I tried to know the structural localization of the cortex, examining, if any, the peculiarity of each place.

a) Anterior and dorsal surface (1-4 of fig. 4, 3-20 of fig. 3, photo. A and B)

Giant pyramidal cells are present from the lower end of G. rectus to the anterior part of G. medialis on the dorsal surface and up to the frontal part of F. ectolateralis on the lateral surface. Especially on and near G. sigmoideus (5, 9, 8, 14) most of them are gathered. It reminds us of area 4 (praecentralis gigantopyramidalis) of BRODMANN. Further backward as well as foreward they become sparse.

At G. sigmoideus posterior (9) cells increase in number and form a clear layer in the deeper portion of the layer III (IV?). Further backward at G. medialis interna (13) this layer becomes indistinct. Still further back near the occipital pole (17, 18) no giant pyramidal cells are found in the part corresponding to the layer IV (V?) but relatively large granular cells are present forming a sheet.

At G. rectus (1) and near the frontal pole (3, 4) the layer IV can be observed only with difficulty and the layer II becomes thinner. On the whole, nerve cells of the cortex are few in number and giant pyramidal cells are shrunken to angular forms.

The anterior part of G. ectolateralis (3, 4) is similar in structure

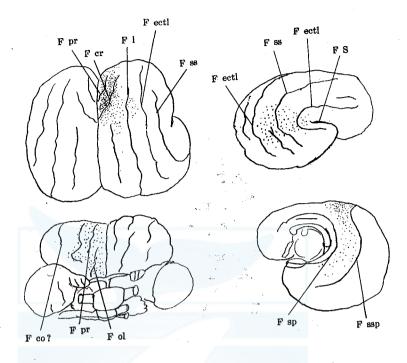


Fig. 11. Distribution of giant pyramidal cells in the right cerebral hemisphere of Physeter: The dots represent the cells in question

to G. sigmoideus, though the number of nerve cells is here somewhat less. G. suprasylvius contains no giant pyramidal cells.

From these observations I came to the opinion that the places, where giant pyramidal cells are present, probably representing the motor area, are in the Sperm whale not restricted within narrow limits but extend widely in the hemisphere.

They are most abundant on the anterior and posterior sides of G. sigmoideus, and dispersed both towards frontal and occipital. Also in the layer IV (V?) cells tend gradually to become dense from F. cruciata backward. The layer II becomes thinner in the frontal part and cells decrease in number, as we go more frontal. But further back than F. cruciata scarcely any change occurs. Generally speaking, cells in all layers tend to increase in number as we examine further backwards. b) Posterior surface (26, 27, 28 of fig. 5, photo. B 27)

Three places near the occipital pole were studied, with the result that cells are numerous here, especially in the layer IV (or V) they are abundant and large in size. This finding reminds us of the striate area, though it was impossible to observe macroscopically neither the intracortical white stria nor the calcarine fissure.

c) Lateral surface (21-25 of fig. 6, photo. B 23)

Both of the superior and inferior G. sylvius and a part of G. suprasylvius were studied. In the antero-dorsal part of G. sylvius superior no giant pyramidal cells are present, while they appear on the lateral basal part of this gyrus and the cells are generally larger.

In the superior and inferior G. sylvius giant pyramidal cells are seen here and there, and cells of the layer III are also comparatively numerous.

d) Medial surface (28-37 of fig. 9, photo. B 29, C 30, 32)

G. submedialis, G. fronicatus and G. hippocampi were examined. The upper frontal part of G. submedialis (30) has densely arranged giant pyramidal cells, and the layers II and III are thick, having many cells. This part resembles G. sigmoideus posterior.

In the postero-basal part (33) many large round cells are assembled in the layer IV (V). Further lower (35) this layer shows middle sized pyramidal cells.

In G. fornicatus (29) each layer is narrow and in the layer V large round nerve cells are gathered densely, forming a special sheet.

G. hippocampi (32, 34, 37) has a quite particular structure; that is, the layers II and IV are thick and crowded with cells. Most cells of the layer IV are polygonal or pyramidal. In the extremely thin layer III small cells are scattered.

Uncus (38 of fig. 4, photo. C 38) is of an extraordinary cytoarchitecture; here only 3 layers are discerned, the first layer of which resembles the layer I of other regions and the second layer, though not very much different from the layer II of other regions, is comparatively thick and has many cells. The third layer contains very few cells of round or pyramidal form.

Considering the colossal bulk of the cortex and the very complicated fissures and gyri of the cetacean cerebral hemisphere, it must be of much interest to know the cytoarchitecture especially its difference according to regions. Though I desired at first to make a map upon the structural differences of the whole cortex, the fulfillment had to be postponed to a later occassion, for the present study was limited to only 38 places in one hemisphere. Meanwhile my object was directed to the determination of cortical regions, which might correspond to the so-called motor area, to the striate aera, or to the superior tem-

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poral area of the human brain, but the result is yet far from success, for the macroscopical orientation of these places is very difficult and moreover the cytoarchitecture of the cetacean cortex is very insufficiently differentiated.

Till today the cerebral cortex of the cetacea has been histologically studied by several authors, among whom MAJOR's work on Balaenoptera musculus, RAWITZ's on Balaenoptera rostrata and Phocaena communis and LANGWORTHY's on Tursiops truncatus are worthy of special reference.

MAJOR observed quite in details the cytoarchitecture at several places chosen from Gyri sylvius, suprasylvius, ectosylvius and medialis, and stated consequently that the frontal lobe is quite similar to the temporal lobe, while the occipital lobe only is a little different in structure as the cells are here smaller. Besides, the cells are pyramidal in the layers I, II, III and the layer IV is not clearly discriminated, while the layer V consists of spindle cells. Chronologically MAJOR's report is previous to BRODMANN's famous work on the localization of the cerebral cortex, and so it did not tell about the formation of 6 cell-layers.

In my opinion, the layers IV and V of MAJOR, are of much doubtful existence, and as giant pyramidal cells were seen in the deeper portion of the layer III in his classification, which I believe is corresponding to our layer V, I can not agree with MAJOR, so far as he said that the layer III has pyramidal cells, as I observed there round and polygonal cells too. As to the fact that in the frontal lobe and in the temporal lobe giant pyramidal cells are extensively observable, I agree with him (fig. 11).

RAWITZ examined only three parts of the cetacean cerebral cortex and stated that it is of a primitive, quite unusual structure, and that there is no differentiation of cell layers as seen in other mammals.

LANGWORTHY researched 16 places quite minutely. He discussed much on the thickness of the layer I, though he did not refer to the conditions of fixation. Deeper than the layer II he named each layer as supragranular, granular, infragranular, and polymorphic. He tried to determine motor area, sensory area, striate area and temporal area as in other mammalia, but he did not seem to have minded much about the complicatedness of the gyri, and identified F. cruciata, F. calcarina and other fissures relatively easily, and spoke even the physiological function of these areas. I endeavoured to compare LANG-WORTHY's observations on the "striate area" of his nomination to my findings in the cortical region, which looks macroscopically like corresponding to his (36 of fig. 9) and further on another place of the occipital pole (27), and found that cells of the layer IV are increased the most noticeably in (27) than in the "striate area" of LANGWORTHY.

One point, in which I agree completely with him, is as also with MAJOR, that giant pyramidal cells exist quite in wide areas beginning from the anterior end of the frontal lobe.

It is a much discussed problem, which fissure in other mammalia the central fissure of Primates corresponds to. And most of the previous authors have identified it either with F. cruciata, with F. praesylvia or with F. coronalis, etc. BYCHOWSKY took F. coronalis for it on the histological basis, that in front of this fissure giant pyramidal cells are found while they are absent in the posterior. In the Sperm whale I can not find such a fissure as showing a clear borderline upon the histological structure. I want to say only that giant pyramidal cells are present the most densely in F. cruciata; it seems to be the centre of their existence (fig. 11).

As to the division of the whole cortex into granular and agranular ones which ECONOMO tried and recently BAILEY reported in the human brain, I can't say anything definite in the Sperm whale, because the layer IV is not well differentiated and moreover my microscopical observations are limited to only a relatively few cortical localities. But so far as my own results are concerned, the granular cortex exists near the occipital pole, on G. sigmoideus posterior and on the laterobasal portion of G. suprasylvius and on G. sylvius etc., while the agranular cortex is seen anterior to F. cruciata and in the antero-dorsal portion of F. ectolateralis. And further to determine the gigantopyramidal area within the agranular cortex was impossible as the gian pyramidal cells are so extensively distributed in the cortex of the Sperm whale, as I stated above.

#### Résumé

Following observations were made on the brain of the Sperm whale caught in the Antarctic Ocean.

1) The average brain weight of 16 male whales (body length of 46-54 feet) is 7.8 kg, that is heavier than that of Mystacoceti (over 50 feet).

The ratio of the brain weight (kg) to the body length (feet) is 0.154 as average and this ratio seems to decrease in senility.

2) Measurement and macroscopical study on the brain of a Sperm whale (50 feet long)

a) The ratio of the sagittal axis to the transversal axis is 0.822:1; that is extremely brachycephalic.

b) The olfactory bulb and tract do not exist.

c) The cerebral peduncle is divided in two parts and the tractus peduncularis transversus is present.

d) On the ventral surface of medulla oblongata small pyramidal tracts are seen.

e) F. cruciata continues directly to F. suprasplenialis.

f) Differently from Mystacoceti, F. rhinalis is not divided into two, medial and lateral, parts.

g) F. corporis callosi is continuous with F. rhinalis posterior.

h) F. olfactoria is present.

i) Uncus is divided into three parts by two shallow sulci.

3) Cytoarchitecure of the cerebral cortex

a) The formation of 6 layers is not clearly observed. As it is difficult to discriminate between the deeper portion of the layer III and the layers IV and V, it seems that only 5 layers exist.

b) Giant pyramidal cells are found quite extensively from the lower end of the frontal lobe till the anterior part of G. medialis, spread on the outside from the anterior part of G. ectolateralis and on the inside till G. submedialis. Especially in G. sigmoideus anterior and posterior they are the most densely gathered.

c) In the vicinity of the occipital pole the layer IV (V?) becomes thicker, having more and larger cells.

d) In the frontal lobe cells of each layer decrease in number, as one observes more forewards.

e) Though it is difficult to divide the cortex into granular and agranular ones, in general F. cruciata seems to show the borderline, for further anterior from this the cortex becomes more agranular, while further posterior it gets more granular.

It is a pleasure to record my indebtedness to Prof. T. Ogawa for his constant guidance and encouragement. My thanks are also due to Ass. Prof. T. Kusama for help and advice on various aspects of this subject. On the Brain of the Sperm Whale

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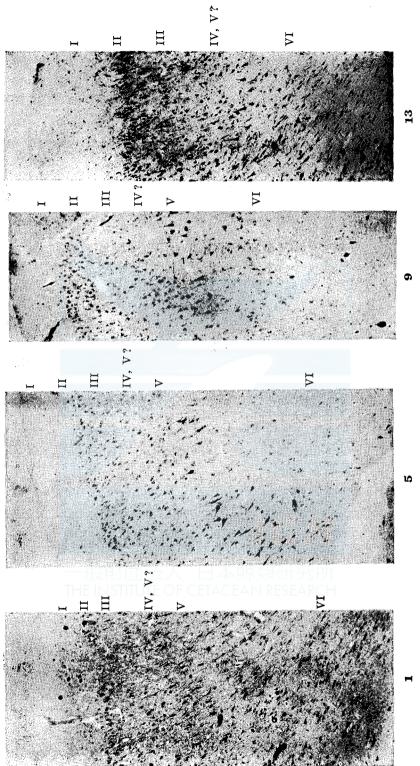
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### Т. Којіма

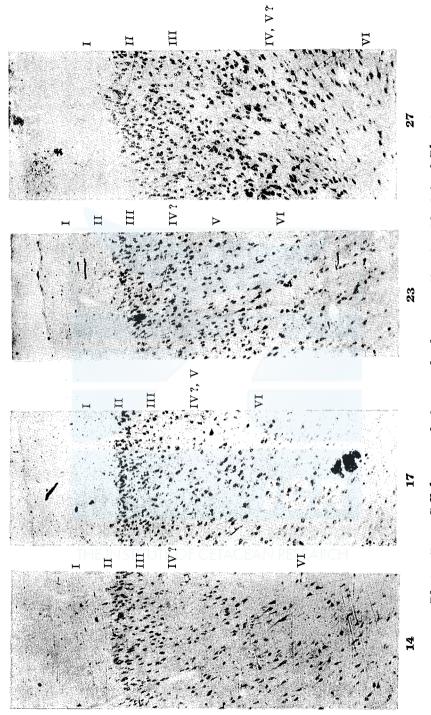
### Abbreviation for all figures

A olArea olfactoria	F prF. praesylvia
C trCrus cerebri	F rh aF. rhinica anterior
Ge ccGenu corporis callosi	F rh pF. rhinica posterior
OlOlive	F SF. sylvia
PPons	F spF. splenialis
PyPyramidal tract	F sspF. suprasplenialis
Sp ccSplenium corporis callosi	F ssF. suprasylvia
UUncus gyri hippocampi IIFasciculus opticus	G ect1Gyrus ectolateralis G f
VN. trigeminus	G hG. hippocampi
VIIN. facialis	G mG. medialis
VIIIN. statoacusticus	G m iG. medialis interna
F coFissura coronalis	G m eG. medialis externa
F crF. cruciata	G rG. rectus
F ccF. corporis callosi	G sig aG. sigmoideus anterior
F ectlF. ectolateralis	G sig pG. sigmoideus posterior
F ectsF. ectosylvia	G smG. submedialis
F hF. hippocampi	G ssG. suprasylvius
F itF. interhemisphaerica	G sy sG. sylvius superior
F 1F. lateralis	G sy iG. sylvius inferior
F olF. olfactoria	

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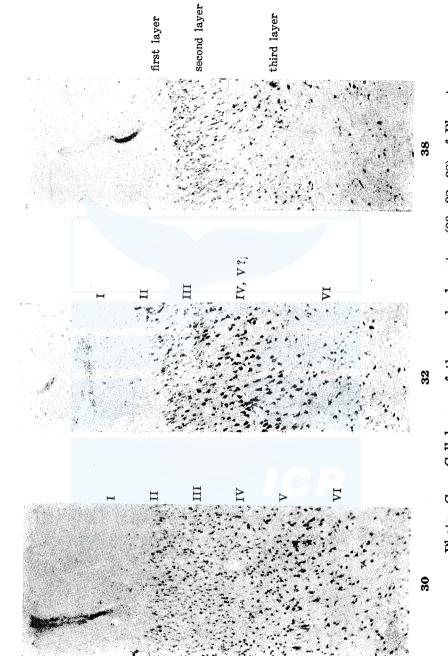


Photo. C....Cell layers of the cerebral cortex (30, 32, 38) of Physeter.

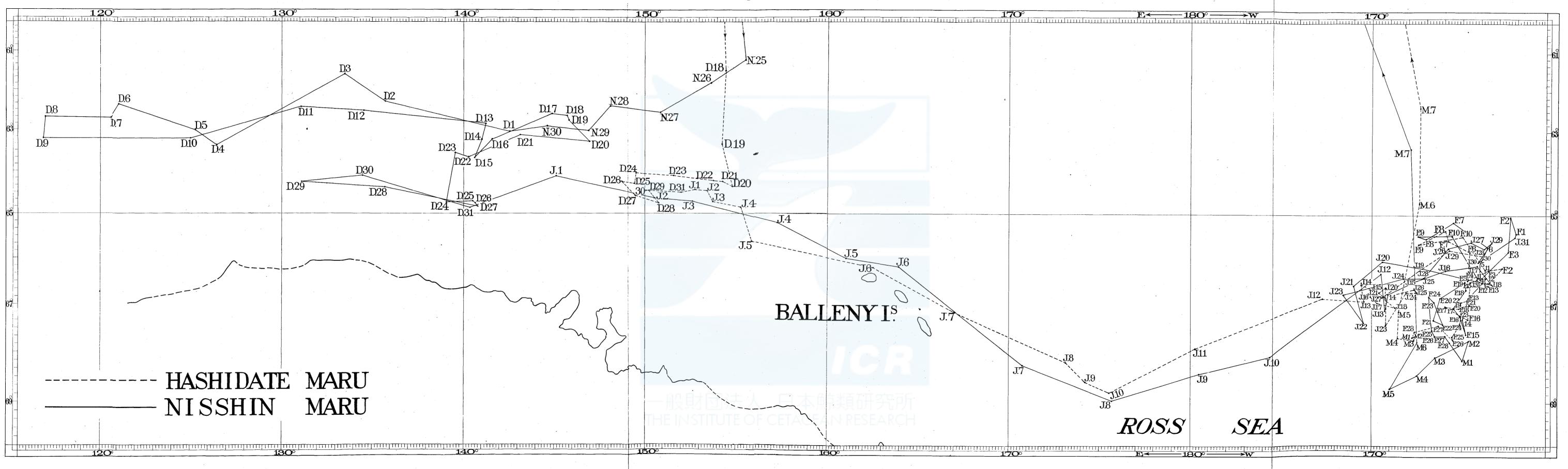
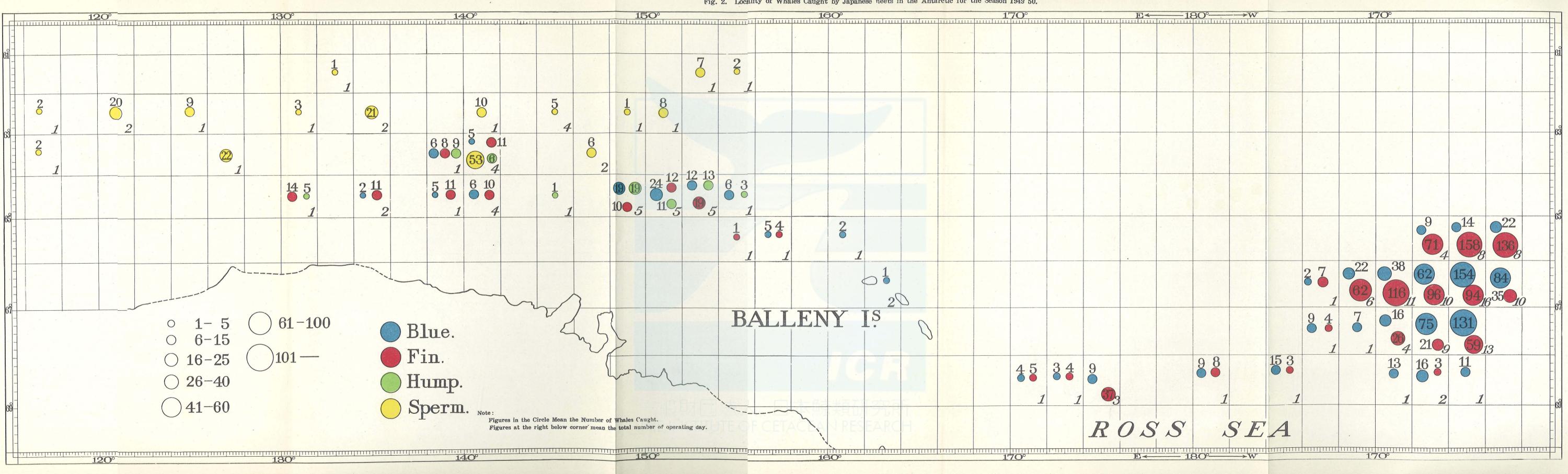


Fig. 1. The Movement of Japanese fleets.



### Fig. 2. Locality of Whales Caught by Japanese fleets in the Antarctic for the Season 1949 50.

### Biological Investigation on the Whales Caught by the Japanese Antarctic Whaling Fleets Season 1949-50

### By Kazuhiro Mizue Tadashi Murata

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#### Introduction

As in the preceding post-war seasons, Japan participated in the Antarctic whaling expedition in 1949-50 with two whaling fleets, namely the Hashidata Maru fleet and the Nisshin Maru fleet. Each fleet consisted of one floating factory, seven catcher boats and one reconnaissance vessel, and was accompanied by one tanker and several cargo vessels.

The floating factory Hashidate Maru, accompanied by the attached catcher boats, sailed from Yokohama on November 10, 1949 and arrived on the Antarctic whaling grounds on December 19, 1949. The Nisshin Maru and her catcher boats sailed from Yokosuka on November 1, 1949 and arrived on the whaling grounds as early as November 25, 1949. This fleet was engaged in the sperm whaling for the following 25 days until the baleen whaling season was opened. The production from this operation is summarized in Table 1.

The grounds for sperm whales are located to the west of those for baleen whales, or between latitudes 61-64 °S and longitudes 116-156 °E (Fig. 2).

The baleen whaling of this season, which was commenced just at

#### K. MIZUE & T. MURATA

Whales caught	172	
Processed products a/	(metric tons)	
Sperm oil	1,651.0	
Liver oil	1.821	
Frozen red meat	660.0	
Gelatine material	86.4	
Leather material	79.4	
Flukes	34.7	
Total	2,513.321	

## Table 1. Catch and processing of sperm whalesby Nisshin Maru fleet, 1949

a/ The amounts of processed products shown are the estimates in the field.

0:00 a.m. on December 22, 1949 by both fleets and closed on March 3 and 5, 1950 respectively by the Hashidate Maru and the Nisshin Maru fleet, have yielded a total production as shown in Table 2. The figures

	Hashidate Maru	Nisshin Maru	Total
Whales processed :			
Blue	384	433	817
Fin	459	597	1,056
Humpback	49	22	. 67
Total	888	1,052	1,940
Blue whale units	631.5	740.3	1,371.3
Processed products $a/:$	(metric tons)	(metric tons)	(metric tons)
Whale oil	12,200.0	14,810.0	27,010.0
Red meat, frozen	10,118.7	13,259.0	23,377.7
Other frozen products	344.8	529.0	873.8
Red meat, salted	3,795.3	3,281.4	7,076.7
Ventral grooves, salted	2,924.1	3,143.0	6,067.1
Other salted products	431.6	314.0	745.6
Baleen	113.2	100.2	213.5
Bone meal	102.5	None	102.5
Liver oil	21.601	21.24	42.841
Blood powder and glue	6.443	None	6.443
Total	30,058.244	35,457.84	65,516.684

Table 2.Catch and processing of baleen whales by the twoJapanese fleets in the 1949-50 season

a/ The amounts of processed products shown are the estimates in the field.

	Season			
•	1946-47	1947-48	1948-49	1949-50
Whales processed:				
Blue	690	710	631	817
Fin	474	608	1,012	1,056
Humpback				67
Total	1,164	1,318	1,643	1,940
Blue whale units	927.0	1,014.0	1,137,0	1,371.8
Processed products $a/:$	(metric tons)			
Whale oil	12,260.0	17,830.0	20,350.0	27,010.0
Frozen and cold-stored products	1,832.9	18,205.3	17,620.1	24,351.5
Salted products	20,385.4	9,048.1	16,535.0	13,889.4
Others $b/$	10.8	301.3	522.7	365.2
Total	34,489.1	45,384.7	55.027.8	65,516.1

## Table 3. Catch and processing of baleen whales by Japanese fleets in the post-war seasons

a/ The amounts of processed products shown are the estimates in the field. b/ Includes liver oil, bone meal, blood powder, baleen, etc.

of this table are summarized and shown in Table 3 in comparison with the production in other post-war seasons. Throughout the period covered, the catch, and consequently the production of whale oil as well as the total amount of products have increased year after year, as the equipments of the floating factories and other various conditions of operation have been improved. The output of the salted products has dropped annually, while that of the frozen products risen sharply.

Catch of humpback whales was permitted in the 1949-50 season on condition that the total catch should not exceed 1,250 whales in the whole Antarctic grounds. But it was only for the first fortnight of the season that this permission was really effective, for the ban was again placed on this operation at the beginning of the third week. Japanese fleets caught 67 humpbacks during the two weeks.

In the 1949-50 season the Japanese fleets operated in the waters between parallels 61°S and 69°S from 116°E longitude eastward to 162°W longitude, which is an area much elongated east and west. Both fleets passed a large part of the season in the part east of 180° longitude.

Being assigned to either of the factory vessels in two groups of three, the writers investigated, throughout the season, every whale carcass when it was hauled up on to the dismembering deck. The main items investigated follow:

Species, sex and length of whale; date and time of capture; date and time of processing.

Body colour.

Scars.

External parasites.

Thickness of blubber.

Kind, amount and degree of digestion of food.

Thickness and colour of mammary glands.

Sex and length of foetuses.

Ovaries (Weight; functional corpora lutea; old corpora lutea; Graafian follicles).

Testes (Weight and volume).

Vertebrae.

Teeth (in the case of sperm whales).

The result of the investigation has been compiled by these items and is presented in this report. In every section of this report, that deals with each of these items, the result is discussed for each species of whales. It should be mentioned here that a satisfactory result has not been reached with humpback whales, because the number caught was so small and their body lengths were limitted to such a small range on account of the short period of operation.

The writers would like to acknowledge most gratefully the matchtess cooperations of the Japan Marine Products Co., Ltd. and the Taiyo Fishing Co., Ltd. in the present investigation as well as in the preparation of this report. Hearty thanks are due to Dr. Hideo Omura, who has kindly helped and guided the writers all through the course of this study. They are also much grateful to Miss Hisako Jimbo for her assistance in the compilation of the data. A particular mention should be made of the exertions of Mr. Hiroshi Ando and Mr. Atae Ihara, who cooperated as the assistants in the laborious measurement of whale carcasses on board the factory vessels.

#### Catch

In Fig. 2 are shown the localities where the stated number of whales were caught by the Japanese fleets during the 1949-50 season. This chart indicates that far more whales were taken in the eastern half of the waters where these fleets operated than in the western half, and that such a large catch as 674 blue and 877 fin whales were made in a period of 51 days from January 12-March 4, 1950 in that relatively small area of the eastern waters bounded by 65°S and 68° 30'S latitudes and by 162°W and 172°W longitudes. This catch accounts for 82.5% and 83.0% of the total catch of the respective species in the present season, and corresponds to an average catch of 1.04 blue whale units per catcher day. In comparison, the same average for other grounds was 0.45 blue whale units in this expedition. Furthermore, a higher average would have been recorded for this particular area, unless the catching operation had been curtailed, in order to meet the processing capacity of the factory ships, so frequently as Though this area was found to be a good whalwas in this season. ing ground by the Japanese fleets during the previous expedition, the catch was not so great in that season as in the present season, because the operations were confined to the part east of 170°W and not continued for so many days.

The length frequency of the catch in this season is graphed in Figs. 3-8 for each species of whales, in comparison with the catch in the preceding three post-war seasons. The two sexes of blue and fin whales are shown in separate figures.

In the length frequency of either sex of the blue whale, the mode for this season has slightly shifted towards large from those for the other seasons. But the length frequencies have remained

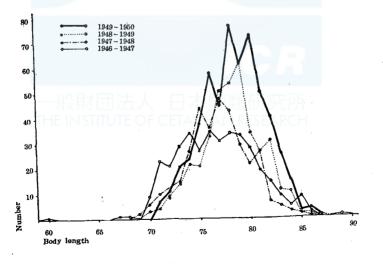
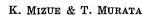
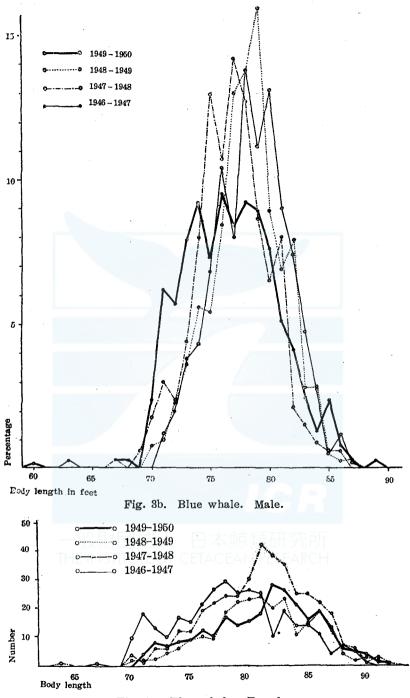
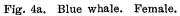
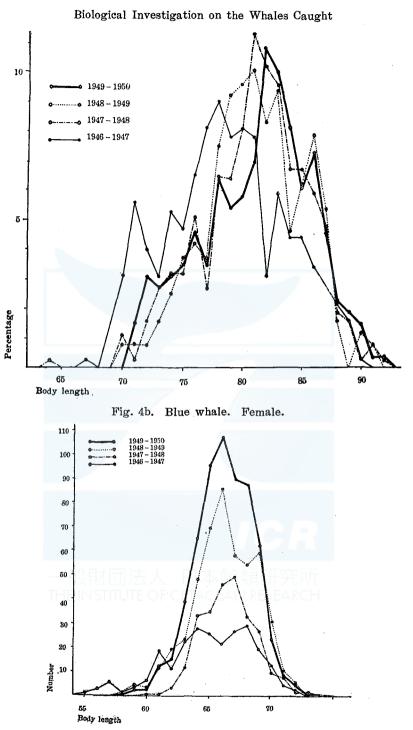


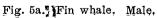
Fig. 3a. Blue whale. Male,











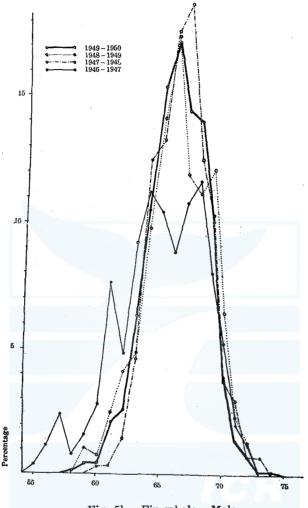
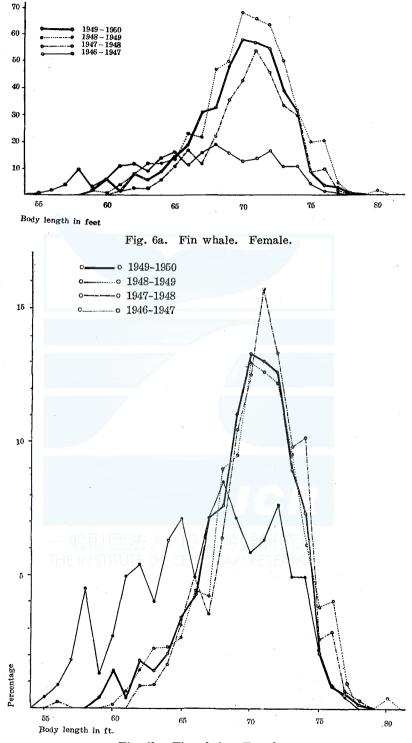


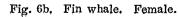
Fig. 5b. Fin whale. Male.

almost unchanged in both sexes of the fin whale throughout the four seasons.

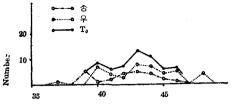
As for the humpback whale, the number caught was so small that any conclusion may not be justified before more data are added through future investigations.

As is obvious from Summary No. 1 through No. 4 (see Appeedixes), in the blue and fin whales the length frequencies of monthly catches show the approximately same tendencies as those in Figs. 3-6. The average lengths of the catch of this season are listed below by species by sexes.



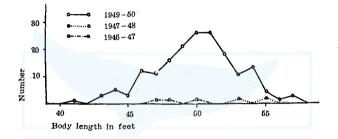


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Body length in feet

Fig. 7. Humpback whale.



#### Fig. 8. Sperm whale.

Species	Male	Female
Blue whale	78.4 ft.	81.2 ft.
Fin whale	66.2 ft.	69.8 ft.
Humpback whale	42.2 ft.	43.3 ft.
Sperm whale	49.9 ft.	(No data)

And the average lengths of the monthly catches hardly differ from these (see Summary No. 1 in the Appendix).

These figures for the blue and fin whales are plotted in Figs. 9 and 10 respectively, in comparison with the corresponding averages in other post-war seasons.

Fig. 9 indicates that in either sex of the blue whale the average length has been gradually increasing throughout the period in question. On the contrary, the same of the fin whale seems to have been decreasing gradually in both sexes (Fig. 10).

As for the humpback whale, of either sex, the average length of the catch by the Japanese fleets in this season is far above the averages for the seasons prior to World War II, which were computed for the catch from the entire Antarctic grounds (Table 4).

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Biological Investigation on the Whales Caught

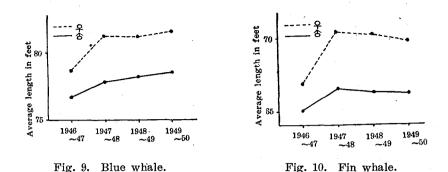
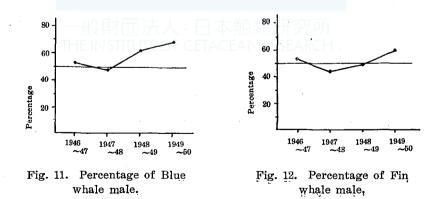


Table 4. Average length of humpback whales taken bythe floating factory whaling in the Antarctic,1934-35 through 1938-39

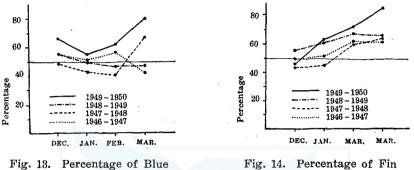
G	Average length in feet			
Season -	Males		Fer	nales
1934-35	39.43	(900)	42.06	(1,027)
1935-36	40.20	(1,256)	42.26	(1,862)
1936-37	39.77	(2,204)	41.39	(2,256)
1937-38	39.61	(781)	41.97	(1,289)
193839	37.69	(266)	40.47	(617)

Note. Numbers of whales are shown in parentheses. Source: International Whaling Statistics.

Figs. 11 and 12 show the percentage of male whales in the blue and fin whale catches in the post-war seasons. In the both species, and especially in blue whales, the percentage of males is higher in the 1949-50 season than in any other post-war seasons.



The sex ratio in the monthly catches of blue and fin whales are graphed in Figs. 13 and 14 for the post-war seasons. In blue whales (Fig. 13), the percentage of males increase as a season advances. This



whale male.

Fig. 14. Percentage of Fir whale male.

tendency is most conspicuous in 1949-50, and 84.7% was reached in March, 1950. Since the Japanese fleets moved toward east with the advance of the season in 1948-49 as well as in 1949-50, this tendency may be regarded as indicating a predominance of males among the blue whales distributed in the eastern grounds. A similar tendency is also observable to some extent in the fin whales caught in this season. In the catch of the humpback whales of this season females exceeded males in number, as was usual in other previous seasons, with the ratio of 64.2% (females) against 35.8% (males). Whether this predominance of females in the catch of humpback whales is caused by chance or is rooted in a preponderance of females in the stock itself, as have been suggested by various authors, is to be decided through the studies in future.

#### Body Colour

Body colour of the blue whales has been investigated with regard to the following three points:

Abundance and distinctness of the pale spots.

Abundance of the white flecks.

Distinctness of the white striations.

The data have been analyzed to study the correlations of these characteristics with length of the whale, with sex and with whaling grounds. In the last case the grounds have been subdivided into two parts along the 180° longitude. But any of these correlations have been found. This suggests that these characteristics of the body colour of blue whales vary from individual to individual, independent from the factors listed above.

The result of observations is summarized in Tables 5-8. These observations are, in their nature, subject to a certain size of personal errors of the observers.

Class and description	Number of whales	Percentage to total
1 (Very few)	17	2.1
2 (Few)	245	30.1
3 (Normal)	394	48.4
4 (Many)	145	17.8
5 (Very many)	13	1.6
Total	814	100.0

Table 5. Abundance of pale spots on blue whales, 1949-50

Table 6. Distinctness of pale spots on blue whales, 1949-50

Class and description	Number of whales	Percentage to total
I (Not distinct)	68	8.3 62.3
II (Distinct) III (Very distinct)	507 239	29.4
III (very distinct)	209	
Total	814	100.0

Table 7. Abundance of white flecks on blue whales, 1949-50

Class and description	Number of whales	Percentage on total
0 (None)		0.6
1 (Very few)	99	12.2
2 (Few)	273	33.5
3 (Normal)	317	38.7
4 (Many)	100	12.3
5 (Very many)	22	2.7
Total	816	100.0

Class and description	Number of whales	Percentage to total	
0 (None)	8	1.0	
I (Not distinct)	390	44.1	
II (Distinct)	372	45.6	
III (Very distinct)	76	9.3	
Total	816	100.0	

Table 8. Distinctness of white flecks on blue whales, 1949-50

Body colour of the fin whales has been investigated with regard to the following five points;

Shade of the dorsal colour.

Which side, right or left, of the lower jaw is black? Distribution of the dorsal colour over the thoracic body surface. Presence of the coloured part protruding toward annus. Unification of the dorsal colour at the base of flukes.

The first item, viz. the shade of the dorsal colour, has been subject to such large personal errors of measurers that the result of the measurements is not presented here.

The second item was studied in the present investigation, though it had been well established in fin whales that the left side of their lower jaw is black and the right side white, because a photograph in a plate of the Discovery Report Vol. I, Plate XXXIV, Fig. 1 had seemed showing a specimen whose lower jaw is black on the right side. Since none of the 1,056 fin whales (619 males and 437 females) examined in this investigation has possessed a white left side of lower jaw, it is most likely that the aforementioned plate shows the negative print of the photograph.

The results of the investigations into the other three items have been analyzed, as in the case of blue whales, respecting their correlations with body length, sex and the whaling grounds as subdivided into two parts along the 180° longitude. Any of these correlations, however, were not found. Consequently, the variation in these characteristics of the body colour of fin whales seems to be nothing more than the individual variation. The results of the observation are summarized in Tables 9–11.

Body colour of humpback whales has been recorded according to Lillie's classification, and the result is summarized in Table 12.

#### Biological Investigation on the Whales Caught

Type of distribution	Number of whales	Percentage to total
L (Dorsal colour reaching lower than normal)	344	32.6
N (Normal)	392	37.1
U (Dorsal colour confined to regions upper than in normal cases)	320	30.3
Total	1,056	100.0

## Table 9. Distribution of dorsal colour on the thoracicbody surface in fin whales, 1949-50

a/ The "normal" type of distribution is defined as such that the dorsal colour reaches down to the 11th to 13th ventral grooves from the umbilicus.

## Table 10.Presence or absence of the protrusion of dorsalcolour towards the annus in fin whales, 1949-50

Presence or absence of protrusion	Number of whales	Percentage to total
+ (Present)	819	77.6
- (Absent)	237	22.4
Total	1,056	100.0

## Table 11. Unification of dorsal colour at the baseof flukes in fin whales, 1949-50

Presence or absence of unification	Number of whales	Percentage to total
+ (Present)	611	57.9
- (Absent)	445	42.1
Total	1,056	100.0

None of the humpback whales dealt with in this investigation belongs to Class 4 (black all over the body), though many individuals of this Class have been reported among the catch from the waters of South Africa and South Georgia. The result of the present investigation almost agrees with the findings made by Matsuura as well as Omura, in their investigations of Area V, for the approximately same waters covered by the present study. Such Agreement seems quite natural, even allowing for the considerably large personal errors which are necessarily connected with the measurements by different investigators.

Body colour class $a/$	Males	Females	Total
1	12.5	11.6	11.9
Intermediate between 1 and 2	12.5	16.3	14.9
2	16.7	2.3	7.5
Intermediate between 2 and 3	54.2	44.2	47.8
3	4.1	11.6	8.95
Intermediate between 3 and 4	0.0	14.0	8.95
4	0.0	0.0	0.0
Whales examined	24	43	67

## Table 12.Percentage frequency of body colour classesin humpback whales, 1949-50

a/ According to Lillie's classification. Descriptions of classes follow: "1"-dorsal colour not protruding onto lateral body sides; "2"-dorsal colour protruding onto lateral body sides (The protrusions of dorsal colour generally occur between base of flipper and posterior end of ventral grooves, on the lateral sides opposite the posterior end of dorsal fin, and at the base of flukes; "3"-paired (right and left) protrusions of dorsal colour uniting on the ventral body side; "4"-black all over the body.

On the other hand, the present result is also analogous to Matthews' observations in the New Zealand waters. This fact implies the humpback whales in the latter waters belong to the stock that have been exploited by the Japanese whalers in the Antarctic, and at the same time confirms the northsouth migration of this species which have been established by marking experiments. Body colour of the whale generally have little bearings on the taxonomy of whales. Nevertheless, the body colour of humpback whales seem worth the further investigation, because it may possibly be utilized as a key characteristic in discriminating the races, if any at all, of this species.

Analysis of the data of the present investigation does not show that there is any correlation between the body length and body colour of the humpback whales, but indicates that the intermediates between Class 2 and Class 3 are prevalent in all length classes of both sexes.

#### Whitish Scars

Whitish scars have been investigated respecting their abundance and their old or new. The result obtained implies nothing but the well established tendency that more scars are found on larger whales than on smaller ones. It is advisable to examine the method now in use critically before resuming the investigation on this characteristic in next season.

Table 13 shows the abundance of white scars in the heigher and

# Table 13Number of blue and fin whales in each abundance<br/>class of whitish scars, 1949-50

a. Blue whales, males.

T () C ) ]			Abundan	ce class $a/$	,	
Length of whale	0	I	II.	III	IV	Total
78 feet or under	0	42	139	96	4	281
79 feet or over	• 0	14	105	153	3	275
Total	0	56	244	249	7	556

#### b. Blue whales, females.

T			Abundan	ce class $a/$		
Length of whale	0	I	II	III	IV	Total
81 feet or under	0	17	82	21	1	121
82 feet or over	0	4	46	81	8	139
Total	0	21	128	102	9	260

#### c. Fin whales, males.

Tomath - Combal			Abundan	ce class $a/$		
Length of whale	0	I	II	III	IV	Total
66 feet or under	0	30	163	140	8	341
67 feet or over	0	5	110	149	14	278
Total	0	35	273	289	22	619

#### d. Fin whales, females.

anoth a Cambola	Abundance class $a/$					
Length of whale	0.	I	II	III	IV	Total
70 feet or under	0	29	149	55	4	237
71 feet or over	0	5	83	103	8	199
Total	0	39	232	158	12	436

a/ The abundance classes of whitish scars are defined as follows: "0"-none; "I" very few; "II"-few; "III"-many; "IV"-very many.

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lower length classes of the two sexes of blue and fin whales.

Not so many white scars were found in humpback whales as in blue or fin whales, and they were mostly round in shape, unlike those found on the latter two species.

#### **External** Parasites

In Table 14 are shown, by species of whales, the number and percentage of the whales which have been recorded in the present

Species of whale	XX71 - 1			Infect	ed
	Whales examine		umber of	whales	Percent
Blue	817	•	5		0.6
Fin	1,056		6		0.6
Humpback	67		0		0.0
Sperm	172		( 3		1.7

 Table 14. Infection with Pennella spp., 1949-50

investigation as being infected by the copepod, *Pennella balaenopterae*. The percentage infection in the blue and fin whales has been very small, being slightly exceeded by that of the sperm whales. Percentage infection has been zero in the humpback whales. But the figures in this table are probably somewhat lower than the true figures, for the parasites as small as this species may have been overlooked at night. The shoulder and the back are the major body parts infected by this species. Head was infected in one individual.

A lower percentage infection has been recorded for the blue whales in this season than in Omura's survey over Area V. For the fin whales, however, the percentages are almost same in the two investigations. *Pennella* is commonly found on the whales captured in the temperate waters, but it gradually falls off the host in the Antarctic. Now that this parasite occurs in the Antarctic waters on the schooling sperm whales more frequently than on the old lone bulls as well as on the blue and fin whales, we may conclude that these schooling sperm whales are migrating over a considerable range of waters.

Only one individual infected by this parasite has been reported by Matthews, out of 38 sperm whales which he studied at Durban.

Table 15 shows the number and percentage of the whales infected by the cirripeds, *Coronula* spp. and *Conchoderma* spp. In the hump-

			Infect	ed with	
Species of whale	Whales examined	Coronul	a spp,	Conchoder	ma spp.
		Number of whales	Percent	Number of whales	Percent
Blue	817	3	0.4	1	0.1
Fin	1,056	15	1.4	4	0.4
Humpback	67	67	100.0	67	100.0
Sperm	172	0	0.0	4	2.3

Table 15. Infection with cirripeds, 1949-50

back whales, the percentage infection has been 100% for either of the genera. The percentages are much lower in the blue and fin whales, and the blue whales show a little smaller percentages than the fin whales. In sperm whales, the percentage infected by *Coronula* has been zero, while the same by *Conchoderma* 2.3%.

Both of Coronula diadema and C. reginae have been found on C. regniae on blue and fin whales.

Table 16 shows the percentage of the whales infected by the

Species of whale		Infected		
	Whales examined	Number of whales	Percent	
Blue	817	20	2.4	
Fin	1,056	52	4.9	
Humpback	67	56	83.6	
Serm	172	47	27.4	

Table 16. Infection with Cyamus spp., 1949-50

amphipods, *Cyamus* spp. Though the table gives 38.6% as the percentage infection of humpback whales, it seems most probable that all the humpbacks handled in this study have been infected, for *Cyamus* are very easily overlooked.

For the same reason, it is also very likely that the percentages of infection of blue and fin whales are underestimated in Table 16. In the blue and fin whales, *Cyamus* are hidden so deep in the posterior parts of the ventral grooves that they may be easily left unnoticed, unless these parts are examined closely.

The percentage infection has been 27.4% in the sperm whales. Various types of *Cyamus*, probably involving a considerable number of new species, have been collected in this investigation. A detailed report will follow on this subject.

Diatoms, as external parasites upon whales, differ from the foregoing animals particularly in that their attachment properly occurs in the Antarctic waters. The diatoms, once attached on the whale, rapidly multiply to form a yellow-brown "skin film" over the body surface of the host, as long as the host remains in those waters. But "skin film" falls off the whale in the waters of low latitudes. It has been estimated that a period of about one month elapses between the arrival of whales within the Antarctic Zone and the formation of visible diatom film upon them.

Karcher has already reported that the percentage infection with diatoms differs from one area of the Antarctic Ocean to another. The percentage infection for the waters presently covered by the Japanese fleets has been lower than Karcher's figure for Area II, but higher than the same for Area III; and it resembles the former figure in the general trend. Karcher's data indicate the percentage infection is higher in the adult whales than in the immature. His result also shows, in agreement with the present investigation, that the fin whales are more liable to infection with diatom film than the blue whales.

We may conclude from these evidences that male whales arrive in the Antarctic Zone earlier than females, and the mature whales earlier than the immature.

The very low percentage infection in the humpback whales is probably due to the fact that this species was fished for only in the earlier part of the season. It is noteworthy, however, that an infection as high as 75.5% has been recorded for the sperm whales, in spite that the operations for this species was confined within the period prior to the baleen whaling season, viz. from November 25-December 19, 1949.

Species		Infe	ected	Ma	les	Fem	ales
of whale	Whale examined	Number of whales	Percent	Whales examined	Percent infected	Whales examined	Percent infected
Blue	816	370	45.3	557	47.6	259	40.5
Fin	1,055	528	50.0	619	54.6	436	<b>43.6</b>
Humpback	67	1	1.5	24	0.0	43	2.3
Sperm	172	129	75.5	172	75.5	-	-

 Table 17. Infection with the diatom film, 1949-50

#### Biological Investigation on the Whales Caught

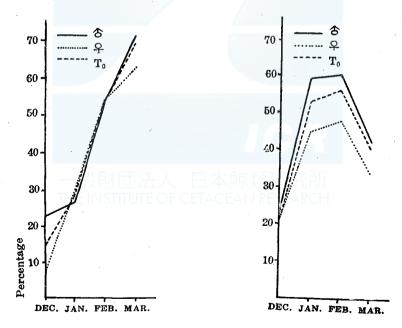
Now, assume the blue and fin whales caught in December and sperm whales caught in November showing thick diatom film on their bodies to have remained in the Antarctic all the previous winter. Table 18 gives, by species of whales, the number and percentage of such whales in the catch of this season.

Species of whale	Whales examined	Whales spending 1949's winter in the Antarctic		
	Whates examined	Number	Percent	
Blue	66	2	3.0	
Fin	101	5	5.0	
Humpback	57	0	0.0	
Sperm b/	22	2	9.1	

#### Table 18. Occurrence of the whales which have spent the 1949's southern winter in the Antarctic a/ in the catch in December b/, 1949

a/ The definition appears in the text.

b/ In the case of sperm whales, the catch in November. 1949 was examined in stead of that in December, 1949.



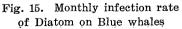
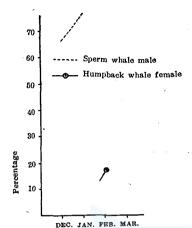


Fig. 16. Monthly infection rate of Diatom on Fin whales



The figures in this table are smaller than Karcher's figures for Area II, but larger than his figures for Area III. Much interesting results will be obtained by investigating the diatomaceous infection on the whales in the Ross Sea, provided that the entrance to that sea is so open as to permit the operations of the whaling fleets therein.

#### Thickness of Blubber

Fig. 17. Monthly infection rate of [Diatom on Humpback and Serm whales.

The thickness of blubber has been measured at the following two parts of the body of every whale caught, viz.: Point 1; the point on the mid-lateral

line, where it intersects the vertical line

passing posterior end of dorsal fin.

Point 2; the point on the dorsal line, where it intersects that cross-section of the body passing ears and perpendicular to the body axis.

Since Point 1 can be located in each whale pretty accurately, and the thickness of the blubber around this point is uniform, the measurements at this point are well comparable among the whales. On the contrary, a slight deviation from Point 2 will result in a fairly large error of the measurement; this is especially true in the sperm whales. Once the whale carcass is laid upon its back on the dismembering deck, it is hardly possible to measure the thickness of blubber at Point 2 accurately. So that, it seems advisable to discontinue the measurement at this point.

In Figs. 18-21 are shown, by species, the average thicknesses of the blubber at Point 1 and 2 of male whales of stated body lengths. In blue and fin whales, the thickness of blubber at Point 1 is larger than the same at Point 2, and both are increasing gradually with body length (Figs. 18 and 19). Data on the humpback whales have not been many enough to justify any conclusions (Fig. 20). The increase in thickness of blubber, which is associated with the increase in length of the whale, is not so conspicuous in the sperm whales as in the foregoing species (Fig. 21). In Fig. 21 the measurements at Point 1 show an increase, though slight, with increasing body length, Biological Investigation on the Whales Caught

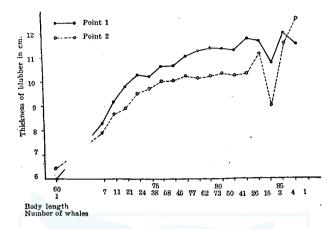


Fig. 18. Average blubber thickness in Blue male whales.

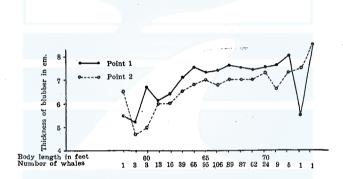


Fig. 19. Average blubber thickness in Fin male whales.

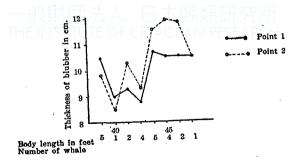


Fig. 20. Average blubber thickness in male Humpback whales,

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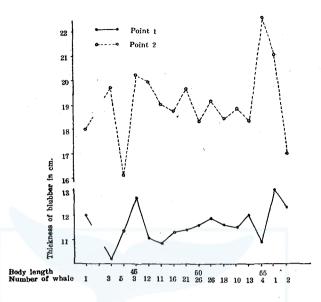


Fig. 21. Average blubber thickness in Sperm whales.

but the values at Point 2 appear to decrease toward larger body lengths, though the result regarding Point 2 are not always reliable on account of the large errors of the measurements. In general, the thickness of the blubber in the Antarctic male sperm whales does not increase with the length of the whale so conspicuously as in the baleen whales.

In contrast with blue and fin whales, the sperm whales show larger thickness of blubber at Point 2 than at Point 1; and the humpbacks, too, show the same tendency.

The thickness of the blubber of female whales differs considerably according to whether the whale is pregnant or not. In Figs. 22-24 are shown the average thicknesses of the blubber of female blue, fin, and humpback whales of stated body lengths, and distinction is made between pregnant and not pregnant females.

These figures clearly indicate that the blubber of female whales, like that of males, adds in thickness as the length of the whale increases.

Humpback whales of the two sexes are well matched with blue whales of the corresponding sexes in the thickness of blubber. In the females of blue and fin whales, like in their males, the blubber is thicker at Point 1 than at Point 2.

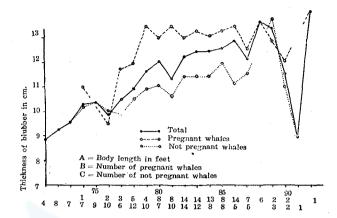


Fig. 22 a. Point 1. Average blubber thickness at Point 1, Blue female whales.

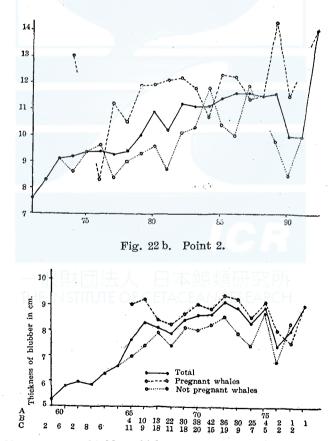


Fig. 23 a. Average blubber thickness at Point 1, Fin female whales.

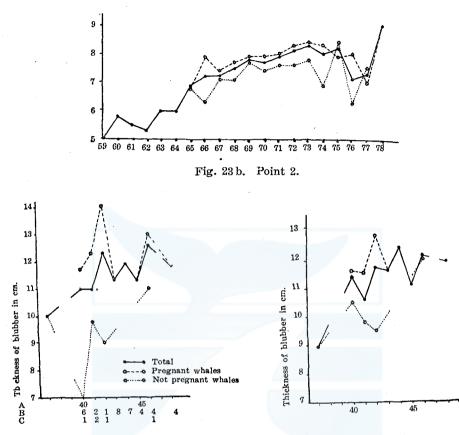
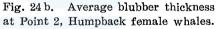


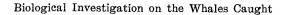
Fig. 24 a. Average blubber thickness at Point 1, Humpback female whales.

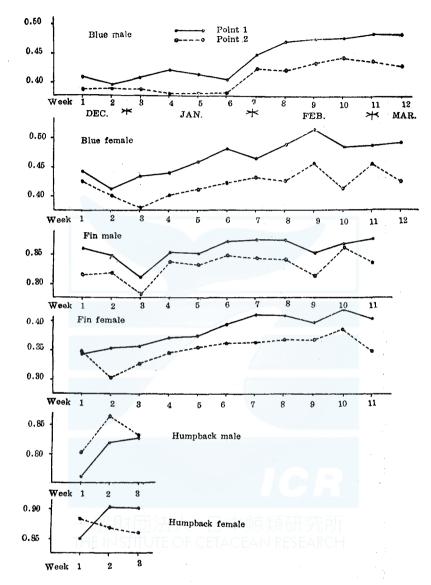


In Fig. 25 is shown the change in the relative thickness of blubber during the season, for different species of whales. The relative thickness of blubber has been calculated for each week by species, and is defined as the ratio, (average thickness of blubber for the weekly catch)/(average body length for the same catch), expressed in percentage. The first week as shown in this figure corresponds to the period from December 22-24, 1949.

Every curve in Fig. 25, though not smooth on account of the small samples, generally tends upwards as the season advances.

Most of the curves in the figure show a decline in January, and tend upwards thereafter. The same tendency has been pointed out in the preceding investigations, though the cause of the decline in January has not been explained.





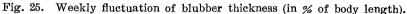


Fig. 25 also indicates that, in every species, the blubber is thicker in females than in males.

Thickness of blubber, as expressed in the percentage to the length of whale, is larger in blue whales than in fin whales.

Humpback whales have thick blubbers for their body lengths.

Cami monthlu												-						
	一 //达 -   E	Blu	Blue whales	ales				Ē	Fin whales	ales			н	Hump	back	Humpback whales	es	
beriod	Whales ex- amined	0	<u>1</u>	LL.	LLL	8	Whales ex- amined	0	H	II	rr	24	Whales ex- amined	0	H	Ľ	rr	24
2nd half, December	68	36.8	36.8 13.2 20.6 19.1 10.3	20.6	19.1	10.3	94		14.9	54.3 14.9 13.8 12.8	12.8	4.2	56	12.5	16.1	28.6	21.4	21.4
lst half, January	112	58.9	58.9 16.1	11.6 10.7	10.7	2.7	191	55.5	20.9	14.1	8.4	1.1	10	20.0	10.0	50.0	20.0	0.0
2nd half, January	92	32.9	32.9 21.7	19.6	15.2	10.9	390	62.6	62.6 16.7	8.7	8.0	4.1	l	1	l	1	l	I
lst half, February	179	33.0	33.0 18.4 25.1 11.2 12.3	25.1	11.2	12.3	284	66.2	66.2 14.4 10.6	10.6	3.5	5.3	1	l	I	1	1	I
2nd half, February	293	28.3	12.3	19.4	17.1	22.9	82	20.7	12.2	19.5	20.7	26.9	I	l	l	I	l	Į
1st half, March	72	37.5	ۍ م	18.1	18.1 11.1 25.0	25.0	15	13.3 13.3	13.3	20.0	20.0 26.7 26.7	26.7	I	1	1	1	1	I
Total	816	35.5	35.5 15.0 19.6 14.3 15.9	19.6	14.3		1,056 57.6 16.3 11.6 8.5	57.6	16.3	11.6	8.5	6.0	99	13.6	15.2	31.8	13.6 15.2 31.8 21.2	18.2
Note. Except for the columns "Whales examined", the figure in this table represents the percentage of whales show- ing the indicated feeding condition to the indicated semi-monthly or total catch. The feeding condition (i. e., empty or filled of the stomach) is classified as follows: R-stomach being full to the utmost (about 75-100% filled); rrr-stomach containing large quantity of food, i.e., about 60-75% filled; rr-stomach containing moderate quantity of food, i.e., 25-50% filled; r-stomach containing small quantity of food, i.e., less than 25% filled; 0-stomach being empty.	r the columns "Whales examined", the figure in this table represents the percentage of whales show- feeding condition to the indicated semi-monthly or total catch. The feeding condition (i.e., empty or nach) is classified as follows: R-stomach being full to the utmost (about 75-100% filled); rrr-stomach quantity of food, i.e., about 60-75% filled; rr-stomach containing moderate quantity of food, i.e., stomach containing small quantity of food, i.e., less than 25% filled; 0-stomach being empty.	ins "V aditior sified food, ntaini	Whale A to t as fo i.e., ng sn	s exa he int llows abou	mined licate : R- t 60- l	l", th d sen stom 75% f	e figure ni-month ach bein illed; r food, i.	in the intervention of the	nis ta total 1 to tl mach	ble re catch he utr t cont an 25	prese 1. Th nost ( ainin % fill	nts the fee he fee about about about about fee ; 0	ie percer ding con 75-100% derate c	ntage dition fille fuanti ch be	of w i.e. d);rr fity of ing e	hales em] rr - st food mpty.	show pty on omach , i.e.	

Table 19. Feeding condition of the captured whales by semi-monthly periods, 1949-50

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#### Stomach Contents

In the Antarctic Ocean the baleen whales are feeding almost exclusively upon *Euphausia superba*, and are not polyphagous as they are in the Japanese waters. Fish were found in the stomachs of some baleen whales handled in this investigation, but such were very rare cases. In Table 19 is summarized the feeding conditions of the whales which were caught by Japanese fleets during this season.

This table refers only to the conditions of the first stomach. Though this table lists a considerable number of whales which showed empty first stomach, many of these whales had food in their second and or further posterior stomachs. And the intestine was filled with faeces in every whale.

Table 19 also shows that the feeding condition of whales improved as the season advanced.

Notes were made on the size of E. superba found in the first stomach, and the data are summarized in Table 20. It is seen from

	Wil-1-	Size	class of <i>Eup</i>	hausia super	rba a/
Species of whate	Whales examined	L	M	S	X
Blue	524	2.5	88.9	6.5	2.1
Fin	441	5.0	73.2	18.1	3.6
Humback	56	5.3	66.1	26.8	1.8

Table 20.Size of Euphausia superba containedin whale stomachs, 1949-50

Note. Figures in the last four columns represent the percentages of those whales to the whales examined, whose stomachs have contained *E. superba* of stated size classes. a/L=over 5 cm.;  $M=4\sim5$  cm.; S=under 4 cm.; X=all of L, M and S mixed.

this table that in most of the cases E. superba in the first stomach belonged to Size Class M (4 to 5 cm. in length). Further analysis of the data shows that relative occurrences of various size classes of E. superba were not correlated with the advance of the season.

From late December through January most of the larger females of E. superba were seen carrying eggs.

Spermatophores had gone from the thelyca of females by early February.

As was mentioned before, fish were contained in whale stomachs in rare occasions, besides E. superba. In the stomachs of 11 blue and 8 fin whales were found the fish which Japanese whalers call "Mizu-tengu", which means the "watery long-nose" in Japanese. This species, which is slender in shape and less than one feet in length, has not been described in the Discovery Reports.

Six blue and 7 fin whales had Myctophum sp. in their stomachs, besides Euphausia.

Fishes of Family Nototheniidae were found, together the Euphausia, in the stomaches of 3 blue whales.

The fish called "Kori-tengu" (which means the "icy long-nose") by our whalers was found in the stomachs of one blue and one fin whale. This species hardly exceeds 2 feet in length, and the Japanese whalers have learned nothing of its scientific name.

Apart from the fishes, *Paratemisto gaudichaudi* was found in one occasion.

These food animals other than *Euphausia*, are usually less than 10 in number per whale, if they happen to be present in the whale stomach. Therefore, their bearing on the nutrition of whales is almost negligible.

However, their occurrence may have been underestimated in the foregoing descriptions, for a considerable number of them has probably passed unnoticed when they came out of the dissected whale stomach mingled in a mass of *Euphausia*.

### Thickness and Colour of Mammary Glands

In the female whales mammary glands were examined respecting their thickness and colour. The thickness was measured at the thickest part of the organ. Colour of the mammary glands was determined by assigning it to one of the colour ranks listed in Tables 21-23, in

	1	Number of whale	s
Colour of mammary glands	Blue	Fin	Humpback
Light pink	50	34	1
Greyish yellow $a/$	9	. 10	0
Reddish yellow	4	3	0
Brownish yellow	2	2	0

# Table 21. Colour of mammary glands in sexually immature whales, 1949-50

a/ The commonest colour in the mammary glands of sexually mature whales.

the investigator's judgement. As this method of determining the colour was considerably subjective, the result may have been biassed to a considerable extent. Consequently, the observations by different investigators do not seem readily comparable with one another, though they are all combined and presented in this section.

Mammary glands are very thin in the sexually immature females. 65 female blue and 49 female fin whales before sexual maturity were examined. The thicknesses of their mammary glands have averaged 2.8 cm. and 2.4 cm. respectively. Maxima of the thickness were 5.5 cm. and 3.5 cm., and minima were 1.5 cm. and 1.0 cm., respectively. There was no sign that the thickness of the mammary glands increases with body length in these sexually immature females. As for colour, the mammary glands of these females were mostly

light pink (Table 21). Only one of the female humpbacks caught was sexually immature, whose mammary glands measured 3.0 cm. in thickness.

Sexually mature females were divided into two groups, the pregnant females and those being In the not pregnant. pregnant females of the blue and fin whales, there was observed a definite increase of the thickness of mammary glands with increasing body length (Fig. But such correlations 26).were not studied in the sexually immature females of the humpback whales, since the data had been scarce for this group.

The averge thickness of mammary gland in the pregnant females of each species was as follows:

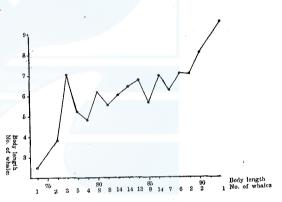


Fig. 26 a. Average thickness of mammary gland of pregnant whales in Blue whale.

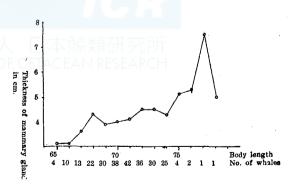


Fig. 26 b. Average thickness of mammary gland of pregnant whales in Fin whale.

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Species of Whale	Average Thickness of Mammary Glands	Number of Individuals Examined
Blue whale	6.2 cm.	112
Fin whale	4.1 cm.	258
Humpback whale	5.1 cm.	87

It is noteworthy that the average thickness for pregnant humpbacks was larger than that for pregnant fin whales. Maximum thickness was 12.0 cm. in these blue whales, 9.5 cm. in these fin whales, and 9.0 cm. in these humpback whales. Minimum was 1.5 cm., 1.5 cm. and 1.0 cm. respectively. The result of observations on the colour of the mammary glands of these pregnant females is given in Table 22.

	· ]	Number of whale	s
Colour of mammary glands	Blue	Fin	Humpback
Light pink	6	32	3
Greyish yellow $a/$	52	123	30
Reddish yellow	41	74	4
Brownish yellow	5	12	0
Brown	7	17	0
Red	1	0	0

Table 22. Colour of mammary glands in pregnant whales, 1949-50

a/ The commest colour in the mammary glands of sexually mature whales.

The table shows that greyish yellow and reddish yellow were the most prevalent among the colours. Pink was almost exclusively associated with the female whales in their first pregnancy.

The data were analized to determine the correlations of the thickness and colour of the mammary glands of pregnant female whales with the size of their foetuses, i.e., with the time of pregnancy, but such correlations have not been established.

Sexually mature but not pregnant female whales have comprised two physiologically different groups, namely the resting females and the females having weaned recently. On account of this heterogeneous composition, possible correlations of the thickness and colour of mammary glands with body length and the time of pregnancy have not been adequately represented for these not pregnant females.

Average thicknesses of mammary glands in these not pregnant females are listed below by species. These figures are larger than those for the pregnant females.

Species of Whale	Average Thickness of Mammary glands	Number of Individuals Examined
Blue whale	8.1 cm.	83
Fin whale	5.1 cm.	127
Humback whale	7.4 cm.	4

#### Biological Investigation on the Whales Caught

In these not pregnant females, the maximum thickness of mammary glands was 25.0 cm. (measured on a lactating female) for blue, 13.0 cm. for fin and 15.0 cm. (measured on a lactating female) for humpback whales. The minima were 2.0 cm., 1.5 cm. and 3.5 cm. for the respective species.

The observations on the colour of mammary glands of the not pregnant females are summarized in Table 23. The tendency illustrated

Table 23. Colour of mammary glands in not pregnanta/ whales, 1949-50

	1	Number of whale	es
Colour of mammary glands	Blue	Fin	Humpback
Light pink	6	. 17	0
Greyish yellow b/	43	56	3
Reddish yellow	21	39	1
Brownish yellow	5	7	0΄
Brown	8	8	0

a/ Not includes immature whales.

b/ The commonest colour in the mammary glands of sexually mature whales.

by this table is very similar to the one shown by Table 22 for pregnant females.

It seems that there is some general relationship between colour and thickness in the mammary glands of whales. Light pink is generally associated with thicknesses less than 4 cm. in the blue whales and with those less than 3 cm. in the fin and humpback whales. The mammary glands of this colour and with these thicknesses are found only in such female whales which have not paired, or are in their first pregnancy, or have ovulated but not have conceived.

Greyish yellow and reddish yellow are usually associated with moderate thickness of mammary glands, namely 5-8 cm. in the blue, 4-6 cm. in the fin, and 5-7 cm. in the humpback whale. Mammary glands showing these characteristic colours and thicknesses are found in the major part of pregnant females as well as in the resting females. A considerable portion of the mammary glands which were

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reddish yellow in colour showed thicknesses larger than what was stipulated above. These are considered to have referred to those females which had weaned relatively long before their capture.

Such colours as yellowish brown, brown and red generally occurred in the mammary glands of large thickness, sometimes as large as 25 cm. These mammary glands may be regarded as from suckling females or those females which had weaned very recently.

#### Foetuses

In this section are dealt with the results of observations on the foetuses and on the pregnant female whales.

In Table 24 are shown the frequencies that the foetuses were found in the right and the left cornu uteri.

Table 24.	Occurrence of foetuses in the right and	
	the left cornu uteri, 1949-50	

. . .

Species of whale	Number of cases examined	Left cornu uteri percent	Right cornu uteri percent
Blue	107	59.8	40.2
Fin	250	48.0	52.0
Humpback	32	56.2	43.8

The frequencies that the heads of foetuses were directed toward the vagina and to the reverse direction are given in Table 25.

Species of w	hales	Number of cases examined	L	head towards	positions in percent Foetal tail towards
				vagina	vagina
Blue		87		96.6	3.4
Fin	60.1	205	- 77 - L N/2	91.7	8.3
Humpback	一版	20 24	人思究到	85.0	15.0

Table 25. Position of foetuses in cornu uteri, 1949-50

Table 26 gives the foetal sex ratio for each species of whales.

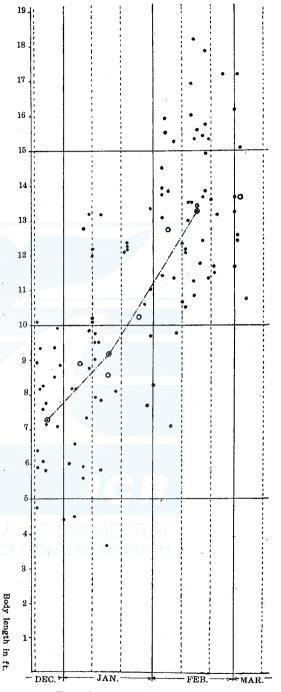
Table 26. Foetal sex ratio, 1949-50

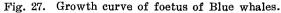
Species of	Ma	les	Fen	nales	Sex not deter-	Total
whale	Number	Percent	Number	Percent	minable (Number)	(Number)
Blue	50	44.6	62	55.4	U	112
Fin	136	52.9	121	47.1	1	258
Humpback	19	52.8	17	47.2	1	37

The lengths of foetuses are plotted against the date of capture of mother whales in Figs. 27-29, espectively for the blue, fin and humpback whales. Average foetus lengths for ten days and monthly periods are also shown in these figures with the marks  $\circ$  and  $\odot$  respectively. The tendencies appearing in these figures are similar to those found Mackintosh bv and Wheeler.

Maximum foetus length was 19'0'' for the blue, 16'1'' for the fin, and 3'0'' for the humpback whales. The minimum was 2'2'' in blue and  $0'3'_{2}''$  in humpback whales. The smallest fin whale foetus was at the stage immediately after the conception.

Table 27 gives, by body length classes, the proportions of the pregnant and the not pregnant females to the total female catch. Pregnant females have accounted for 43.1%, 59.0% and 86.0% of the female catches of blue, fin and humpback whales respectively.





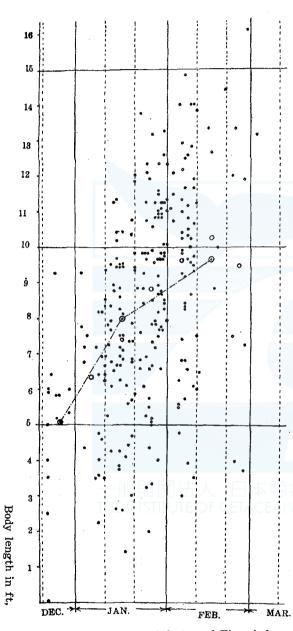


Fig. 28. Growth curve of foetus of Fin whales.

The proportion of the pregnant and the females not pregnant to the catch of the sexually mature females are shown in Table 28. The females showing one or more corpora lutea in their ovaries were considered as being sexually mature. Pregnant females have amounted to 57.7%, 66.8% and 88.1% of the captured sexually mature females of the blue, fin and humpback whales, respectively.

These percentages of pregnant females, which have been derived from the analyses of the catch, are probably overestimating the percentage of the pregnant females in the stock of whales in the sea to a considerable extent, because the mother whales which are accompanied by calves and are seemingly suckling are never hunted, thus preventing the not pregnant females from being represented in the catch as fully as the pregnant individuals.

In both of Tables 27 and 28 the females of blue whales give a higher percentage pregnancy than those of fin whales. And the percentage pregnancy is very high in humpback whales, implying that this species propagates more rapidly than the other species.

Table 28, which refers to the sexually mature females, gives a relatively constant percentage pregnancy throughout the body length classes of each species. Hence, there are found no such length classes in which the female whales are more liable to pregnancy than in others.

#### **Ovaries**

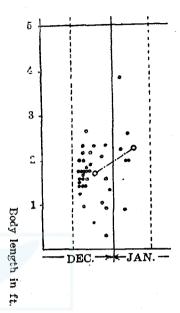
The measurements made on the ovaries of every female have included the weights of the right and left ovary, the number of corpora lutea, and the

Fig. 29. Growth curve of foetus of Humpbackwhales.

Table 27.	Pregnant	and not pregnant whales in the catch	
	of females	a/, by length groups, 1949-50	

Length in	Preg	Pregnant		regnant	Total
feet	Number	Percent	Number	Percent	number
71	0	0.0	4	100.0	4
72	0	0.0	8	100.0	8
73	0	0.0	7	100.0	7
74		12.5	667777	87.5	8
75		0.0	9	100.0	9
76	2	16.7	10	83.3	12 .
77	3	33.3	6	66.7	9
78	5	29.4	12	70.6	17
79	4	28.6	10	71.4	14
80	8	53.3	7	46.7	15
81	8	44.4	10	55.6	18
82	14	50.0	14	50.0	28
83	14	53.8	12	46.2	26
84	13	61.9	8	38.1	21

#### a. Blue whales.



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Length in feet	Pregnant		Not pregnant		Total
	Number	Percent	Number	Percent	number
85	8	50.0	8	50.0	16
86	14	73.7	5	26.3	19
87	7	58.3	5	41.7	12
88	6	100.0	0	0.0	6
89	2	40.0	. 3	60.0	5
90	2	50.0	2	50.0	4
91	0	0.0	1	100.0	1
92	1	100.0	0	0.0	
Total	112	43.1	148	56.9	260

a/ Includes sexually immature whales.

Length in	Preg	nant	Not pr	egnant	Total
feet	Number	Percent	Number	Percent	number
59	0	0.0	2	100.0	2
60	0	0.0	6	100.0	6
61	0	0.0	2	100.0	2
62	0	.0.0	8	100.0	8
63	0	0.0	6	100.0	6
64	0	0.0	9	100.0	9
65	4	26.7	11	73.3	15
66	10	52.6	. 9	47.4	19
67	13	41.9	18	58.1	31
68	22	66.7	11	33.3	33
69	30	62.5	18	37.5	48
70	38	65.5	20	34.5	58
71	42	73.7	15	26.3	57
72	HE 36	65.5	CEA 19RESE	ARC 34.5	55
73	30	76.9	9	23.1	39
74	25	78.1	7	21.9	32
75	4	44.4	5	55.6	9
76	2	50.0	2	50.0	4
77	1	33.8	2	66.7	3
78	1	100.0	0	0.0	1
Total	258	59.0	179	41.0	437

# b. Fin whales.

Length in	Pregnant		Not pr	Not pregnant	
feet	Number	Percent	Number	Percent	number
37	0	0.0	1	100.0	1
38	0	0.0	0	0.0	0
39	0	0.0	0	0.0	0
40	6	85.7	1	14.3	7
41	2	50.0	2	50.0	4
42	2	66.7	1	33.3	3
43	8	100.0	0	0.0	8
44	7	100.0	0	0.0	7
45	4	100.0	0	0.0	4
46	4	80.0	1	20.0	5
47	0	0.0	0	0.0	0
48	4	100.0	0	0.0	4
Total	37	86.0	6	14.0	43

### c. Humpback whales.

Table 28. Pregnant and not pregnant whales on the catch of sexually mature females, by length groups, 1949-50 a. Blue whales.

Length in	Preg	gnant	Not pr	egnant	Total
feet	Number	Percent	Number	Percent	number
74	1	100.0	0	0.0	1
75	0	0.0	0	0.0	0
76	2	66.7	1	33.3	3
77	3	75.0	1	25.0	4
78	5	45.0	6	54.5	11
79	4	40.0	6	60.0	10
80	8	61.5	5	38.5	13
81	8	50.0	8	50.0	16
82	14	51.9	13	48.1	27
83	14	53.8	12	46.2	26
84	13	E 65.0 A	CEAN 7RESE	35.0	20
85	8	50.0	8	50.0	16
86	14	73.7	5	26.3	19
87	7	58.3	б	41.7	12
88	6	100.0	0	0.0	6
89	2	40.0	3	60.0	5
90	2	50.0	2	50.0	4
91	0	0.0	1	100.0	1
92	1	100.0	0	0.0	1
Total	• 112	57.4	83	42.6	195

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Total	Not pregnant		nant	Preg	Length in
numbe	Percent	Number	Percent	Nümber	feet
1	100.0	1	0.0	0	64
8	50.0	4	50.0	4	65
14	28.6	4	71.4	10	66
28	53.6	15	46.4	13	67
31	29.0	9	71.0	22	68
47	36.2	17	63.8	39	69
57	33.4	19	66.7	38	70
57	26.3	15	73.7	42	71
55	34.5	19	65.5	36	72
39	23.1	9	76.9	30	73
32	21.9	7	78.1	25	74
9	55.6	5	44.4	4	75
4	50.0	2	50.0	2	76
3	66.7	2	33.3	1	77
1	0.0	0	100.0	1	78
386	33.2	128	66.8	258	Total

#### b. Fin whales.

#### c. Humpback whales.

Length in	Preg	nant	Not pregnant		Total
feet	Number	Percent	Number	Percent	number
40	6	85.7	1	14.3	7
41	2	50.0	2	50.0	4
42	2	66.7	1	33.3	3
43	8	100.0	0	0.0	8
44	7	100.0	0	0.0	7
45	4	100.0	0	0.0	4
46	HEINATTU	80.0	DEANIRESEA	20.0	5
47	0	0.0	0	0.0	0
48	4	100.0	0	0.0	4
Total	37	88.1	5	11.9	42

diameters of the largest Graafian follicle and corpus luteum.

The females which showed any corpora lutea on their ovaries were regarded as sexually mature, and those showing no corpus luteum as immature. Figs. 30 to 32 show the percentage of the sexually Biological Investigation on the Whales Caught

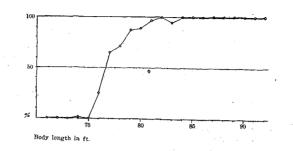


Fig. 30. Maturity of Blue female whales.

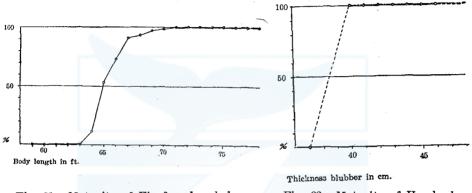


Fig. 31. Maturity of Fin female whales.

Fig. 32. Maturity of Humback female whales.

mature females to the female catch in each length class, respectively for the blue, fin and humpback whales.

Sexually immature females have accounted for the following percentages of the female catch:

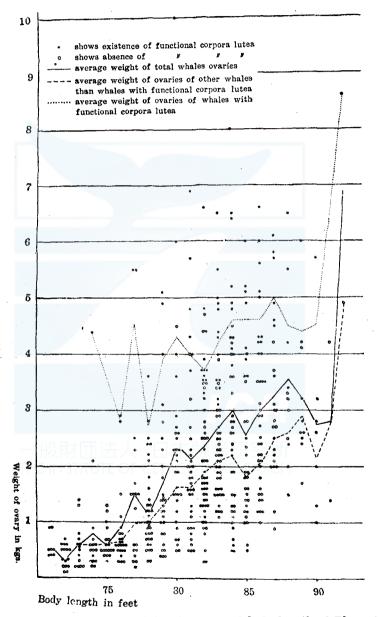
Species of Whale	Percentage of Sexually Immature Females	Total Females Caught	
Blue whale	25.0%	260	
Fin whale	11.7%	437	
Humpback whale	TEOFCE2.4%EAN RES	EARCH 42	

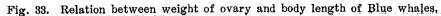
Table 29.	Total number of corpora lutea for all the whales
	examined, by ovaries, 1949-50

Species of	Number of	Right ovary		Left ovary	
whale	whales examined	Total number of c. lutea	Percent	Total nnmber of c. lutea	Percent
Blue	137	750	51.7	701	48.3
Fin	282	1,744	49.5	1,776	50.5
Humpback	26	97	50.5	95	49.5

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In Table 29 are shown the total numbers of the corpora lutea which were found on the right and left ovaries of all the female whales examined. Since the totals for the right and the left ovaries are approximately equal in every species, we may well consider that





# Biological Investigation on the Whales Caught

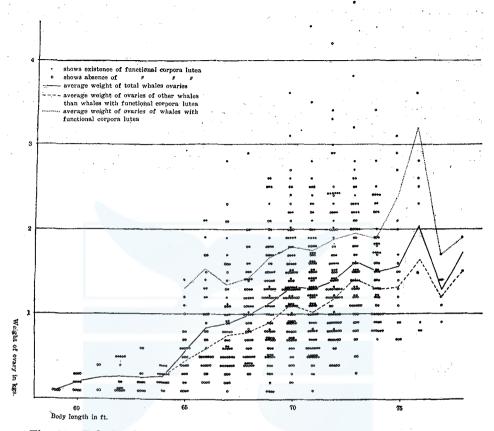


Fig. 34. Relation between weight of ovary and body length of Fin whales.

corpora lutea develop equally on both ovaries, though it does not necessary follow that the right and left ovaries ovulate in exact turns.

In Figs. 33-35 are illustrated the correlation between the weight of the single ovary and the body length in the female blue, fin and humpback whales, respectively. On these figures the weights of the right and left ovary were plotted individually, and the discrimination was made between

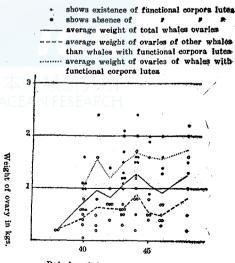




Fig. 35. Relation between weight of ovary

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the ovaries showing the functional corpus luteum (black dots) and those not showing them (white dots). Though the correlation considered has been obscured in these figures by the occurrences of functional corpus luteum in some ovaries, we can still see the general tendency that the weight of ovary increases with the body length.

In Figs. 36-38 are shown the numbers of corpora lutea in the ovaries of the female whales of stated lengths.

Minimum length in the sexually mature females measured 74 ft. in blue, 64 ft. in fin and 40 ft. in humpback whales. (Measurements were not made so many on the sexually mature female humpbacks.)

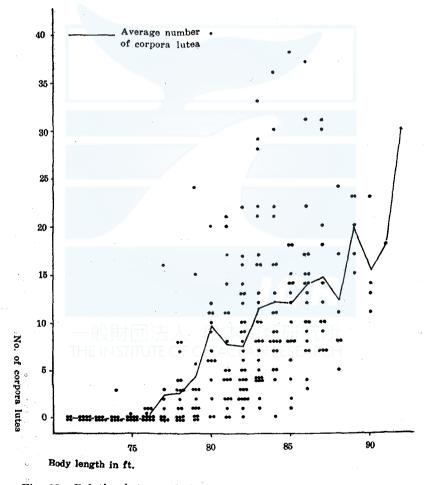


Fig. 36. Relation between body length and number of corpora lutea in Blue whales. Maximum lengths in the sexually immature females were 84 ft. for the blue, 70 ft. for the fin, and 37 ft. for the humpback whales. (There caught only one immature female humpback whale.)

Figs. 36-38 show that there is a general increase in numbers of corpora lutea with increasing body length, and Figs. 33-35 evidence

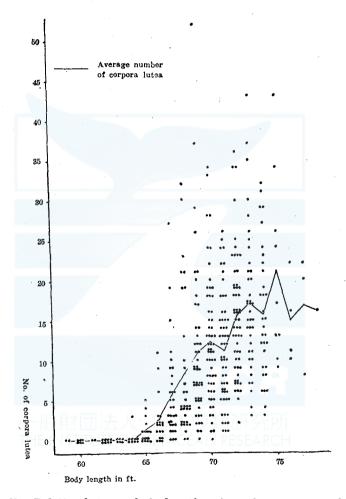


Fig. 37. Relation between body length and number of corpora lutea in Fin whales.

the increase in the weights of ovary with increasing body length. Consequently, we may say that the weights of ovary increases with the numbers of corpora lutea.

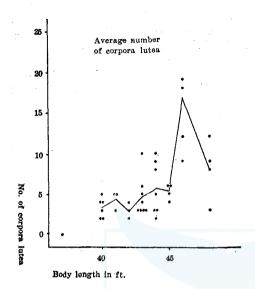


Fig. 38. Relation between body length and number of corpora lutea in Humpback whales.

In Figs. 39-41 are plotted the diameters of the largest Graafian follicle and the functional corpora lutea of the pregnant females against the lengths their foetuses. Since the length of foetus gives an accurate estimate of the time of the last ovulation, we can also study in these graphs the correlations of these diameters with the time that has elapsed since the last ovulation.

These figures illustrate that the diameters of the largest Graafian follicles as well as of the functional corpora lutea remain fairly constant throughout the foetal lengths, that is, irrespective of the time from the

last ovulation. In these analyses, the foetal lengths have ranged between 2-19 ft. in blue whales, corresponding to  $3^{1}/_{2}-9^{1}/_{2}$  months after ovulation, and from less than 1 ft. to 16 ft. in fin whales, representing the period from immediately after ovulation to 10 months later.

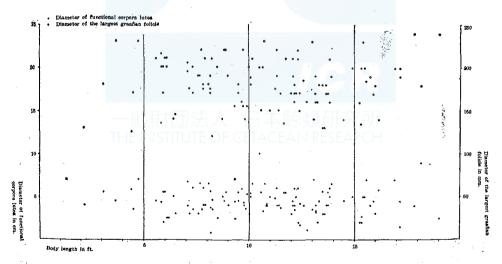


Fig. 39. Relation between body length of foetus and diameters of the largest Graafian follicles and functional corpora lutea in Blue whales.

Biological Investigation on the Whales Caught

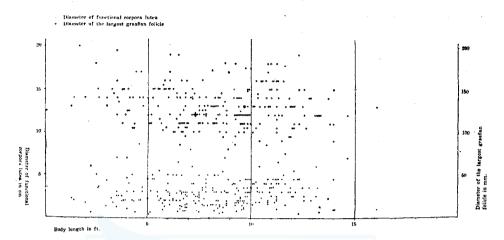
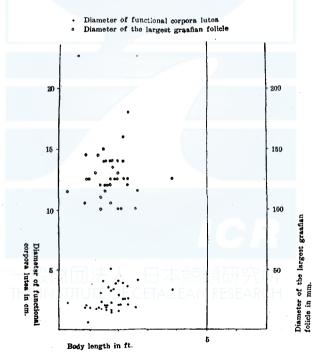
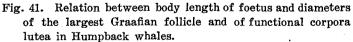


Fig. 40. Relation between body length of foetus and diameters of the largest Graafian follicle and of functional corpora lutea in Fin whales.





It is likely from the above analyses that the functional corpora lutea and the largest Graafian follicles of the pregnant fin and blue females remain at constant sizes throughout the period of pregnancy, neither swelling nor shrinking. (The conditions in the pregnant humpback whales have not been confirmed, because the observations were not enough many.)

The diameters of the largest Graafian follicles and the functional corpora lutea of pregnant females have averaged as follows:

	Average	Number of Indivi-		
Species of Whale	Functional Corpora Lutea	Largest Graafian Follicles	duals Examined	
Blue whale	17.9 cm.	45.8 mm.	112	
Fin whale	12.9 cm.	28.4 mm.	258	
Humpback whale	12.6 cm.	25.0 mm.	37	

The data for the largest Graafian follicles of the not pregnant whales were analyzed in a similar way, and have led to the almost same conclusion as was reached concerning the pregnant whales. There was found no correlation in the sexually immature females between the diameter of the largest Graafian follicles and the time of capture; in other words, there was no sign that the largest Graafian follicles in these whales had shown smaller diameters in the earlier part of the season than they did in the later part of it. The diameters of the largest Graafian follicles of sexually immature whales have averaged as follows:

Species of Whale	Average Diameter of Largest Graafian Follicles	Number of Immature Females Examined
Blue whale	16.8 mm.	. 29
Fin whale	5.0 mm.	26
Humpback whale	8.0 mm.	1

In the sexually mature females which were not pregnant, the diameters of the largest old corpora lutea were measured. The data were analyzed to determine the possible correlation of these diameters with the time of the season. But the result shows that there is no such correlation, and that the sizes of the largest old corpora lutea remain fairly constant throughout the season without any sign of shrinkage in the later part of the season. As for the largest Graafian follicles of these whales, their sizes have not changed appreciably all through the season, just as was the case in the same organ in the sexually immature females. The diameters of the largest Graafian follicles of the sexually mature, not pregnant females have averaged as follows:

Species of Whale	Average Diameter of Largest Graafian Follicles	Number of Not Pre- gnant, Mature Females Examind
Blue whale	25.0 mm.	37
Fin whale	17.0 mm.	63
Humpback whale	22.0 mm.	2

It should be mentioned here that any analyses of the data that are made in this study with reference to time have much limitations to their scope, because the data used have only covered a single season, or a period of two and a half months. And the conclusions drawn on basis of such analyses are mostly tentative ones; this holds true particularly in the discussions in this section.

One of the pregnant fin whales examined showed two functional corpora lutea in spite of being with a single foetus. Data pertinent to this case follow:

Length of Mother	Location Captured	Date & Time Captured	Sex & Length of Foetus
72 ft.	162°44′W 66°11′S	February 2, 1950. 5:00 a.m.	Male; 12'1"; found in the left cornu uteri.
Functional Corpora Lutea		Number of Old	Weight of Oregon
Number	Diameter	Corpora Lutea	Weight of Ovary
(Left) 2	11.0 cm., 10.5 cm.	8	2.4 kg.
(Right) 0		6	1.3 kg.

The correlation between the numbers of corpora lutea and the ossification of vertebrae was also investigated, with a result as illustrated in Fig. 42. Exactly median vertebrae of the thoracic and lumbar series were examined. The determination of the degrees of ossification was subject to a considerable personal error, and the numbers of corpora lutea were considerably divergent in each ossification class. And so, the average value was taken in each class, and are plotted in Fig. 42.

From this figure we may state that, in both blue and fin whales, the ossification of both thoracic and lumbar series is not completed at corpora lutea numbers less than 20, and the physical maturity begins

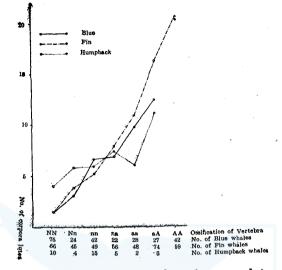


Fig. 42. Relation between number of corpora lutea and ossification of vertebrae.

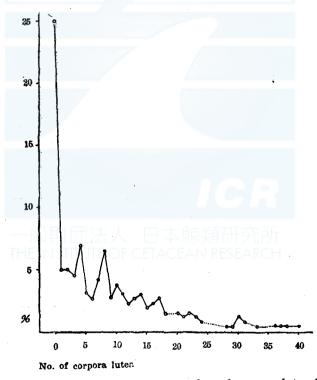


Fig. 43. Frequency curve of number of corpora lutea in Blue whales.

#### Biological Investigation on the Whales Caught

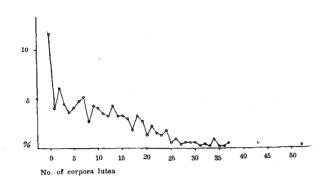


Fig. 44. Frequency curve of corpora lutea in Fin whales.

at corpora lutea number of 3 or 4. The humpback whales examined were so few that the correlation has not been clearly represented for this species.

The percentage frequencies of numbers of corpora lutea are given in Figs. 43-45 for the blue, fin and humpback whales respectively.

#### Testes

The weight and volume of the testes excluding deferent ducts were measured in every male whale captured.

The weights and volumes of the testes of blue whales are plotted against the lengths of whale in Figs. 46 and 47. The distribution of the plots in both these figures generally agree with the records for the usual years. If we consider the male blue whales with the testes weighing 10 kg. upwards as being sexually

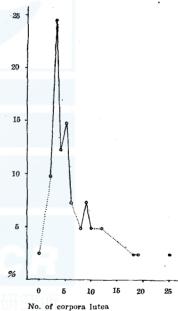


Fig. 45. Frequency curve of number of corpora lutea in Humpback whales.

mature, only 33 whales or 6% of the male blue whales taken were sexually immature.

The data for the fin whales are plotted in Figs. 48 and 49 in same fashions. These figures also give just same features as have been recorded in the preceding seasons. Only 30 or 5% of the male fin whales that were caught in this season were sexually immature, if

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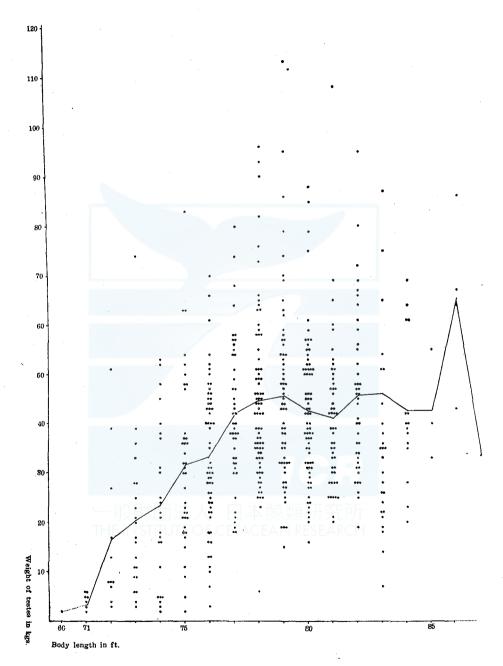
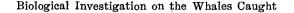


Fig. 46. Relation between total weight of right and left testes and body length in Blue whales.



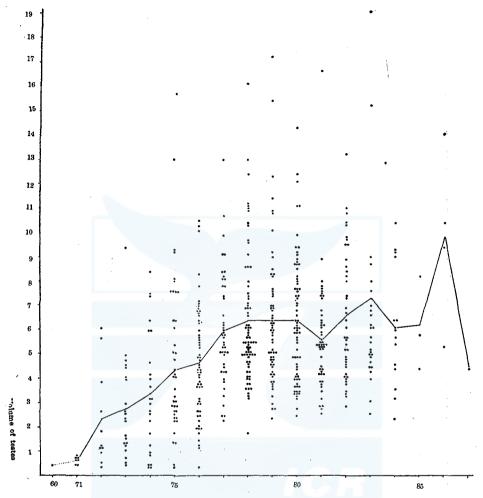


Fig. 47. Relation body length and volume of testes in Blue whales.

5 kg. is taken as the minimum testes weight of the sexually mature fin whales.

The writers are of opinion that both of the weight and the volume of testes need not be measured, as one is to give the same result as the other. And it seems more advisable to choose the weight, for weighing can be done more easily and accurately than measuring the volume, and, in addition, the data of weight are far easier to handle than the volume data.

The measurements on the humpback whales are plotted in Figs. 50 and 51 in same manners. But the data are too scarce to justify

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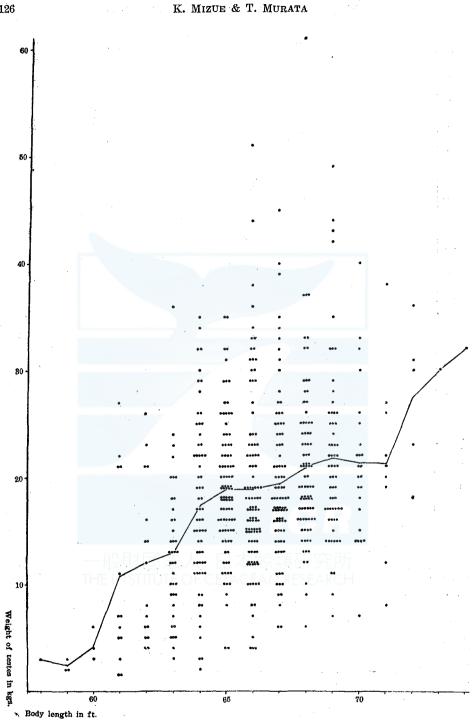


Fig. 48. Relation between weight of testes and body length in Fin whales.

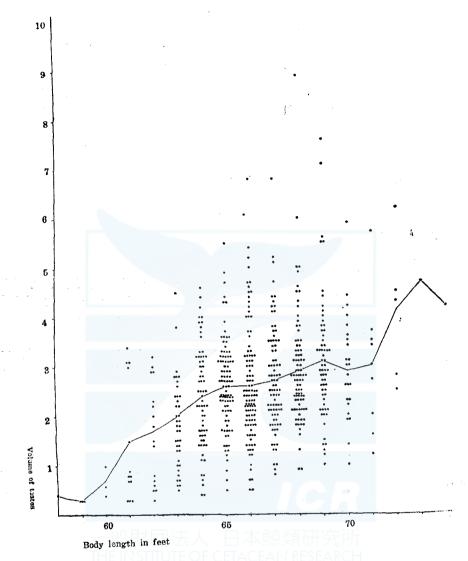


Fig. 49. Relation between body length and volume of testes in Fin whales.

any statement concerning the correlation of these measurements with the length of whale. Such biological characters as the body length at the sexual maturity and others, that have already been clarified in the Antarctic blue and fin whales, remain obscure in the case of humpback whales. It is necessary to throw light upon these problems by repeating the investigations in future.

The correlation of the weight of testes with the ossification of

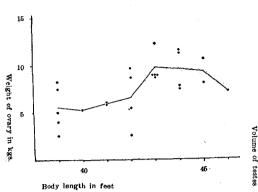
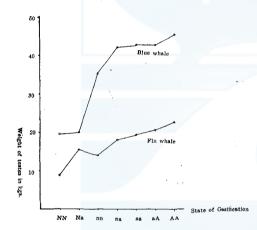
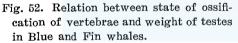


Fig. 50. Relation between body length and weight of testes in Humpback whales.





### Vertebrae

The ossification of the vertebrae was examined by chipping off the cartilage layer between epiphys and centrum. But it seems somewhat doubtful that the data obtained by different investigators are perfectly comparable with one another.

The correlations of the ossification with the numbers of corpora lutea and with the weights of testes have been dealt with respectively in the sections of "Ovaries" and "Testes". The correlation with body length of the whale will be discussed here. Figs. 53 and 54 show

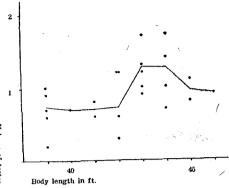
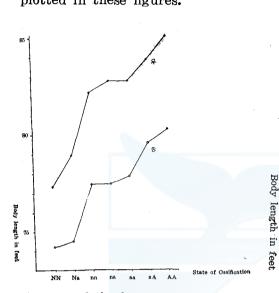


Fig. 51. Relation between body length and volume of testes in Humpback whales.

vertebrae is shown in Fig. 52. Humpback whales are omitted because their number examined are so small.

The weight of testes is so divergent in each ossification class, just as the number of corpora lutea was, that the average weight was calculated for each class, and was plotted in Fig. 52. But the obtained correlation curve is far difficult to interprete, compared with the one or the number of corpora lutea. this correlation in the blue and fin whales. Body lengths are also divergent in each ossification class, just as the numbers of corpora lutea and the weights of testes were. And so, the average length is computed for each class, and is plotted in these figures.



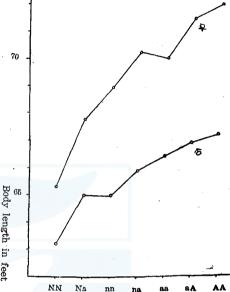


Fig. 53. Relation between state of ossification of vertebrae and body length in Blue whales.

Fig. 54. Relation between state of ankylosis of vertebrae and body length of Fin whales.

According to Figs. 53 and 54 the ankylosis of the vertebrae is completed (i.e. ossification class "AA") at the following body lengths:

Blue whales, males 80 ft.; Blue whales, females 85 ft.

Fin whales, males 67 ft.; Fin whales, females 72 ft. Then, the body lengths at which the ankylosis of vertebrae is completed differ by 5" between the two sexes in both of blue and fin whales.

If we regard the whales showing the ossification of vertebrae of "aa" or further as physically mature, such whales have accounted for 58% or 470 whales of the blue whale catch of this season, and 57% or 597 whales of the same fin whale catch.

\* \* \* \* \* \*

On closing this report, mention should be made of such phases of the present investigation that were not dealt with in this report.

General biological data, as were presented in this report respect-

#### K. MIZUE & T. MURATA

ing the baleen whales, were also collected on all the sperm whales caught in the expedition. But they had not been finally compiled by the time this report was prepared. They are to be dealt with in another report.

In parallel with the general biological investigation into the whale carcasses, the writers carried out further specialized studies on the biology of the whale. The projects of such studies follow:

Study on the blood-group in the whale.

Study on the whale food.

Study on the lung of the whale.

Identification of the fishes found in the whale stomachs.

Identification of the whale lices.

Until present the writers have not had time to compile and analyse the data of these studies. Results of these studies will be published when the necessary treatments of data are finished.

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# On the Periodic Mark on the Baleen Plates as the Sign of Annual Growth

## BY

### MASAHARU NISHIWAKI

The Whales Research Institute

(5. Dec. 1950)

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### Introduction

It was concluded in the previous report (Nishiwaki: 1949b) that one can not trace the life of a whale back to its birth by the study of the sculptures on the surface of its baleen plates, because not merely the tips of the plates are chipping away gradually and incessantly after the whale has attained a certain age, but also it is very difficult to determine whether or not, and how much the tips of the plates of a given whale have been chipped away after its birth. At the same time, however, it was also pointed out that these sculptures were worth of further investigation, because the structure of the baleen plates relates fairly correctly the life history of the whale during a certain number of years prior to its catch and slaughter. And the necessity of such studies was also emphasized at the same time in order to know whether the interval zone between the two successive sculptures shows the annual growth of the baleen plate, or the said zone shows the growth cycle for more than one year; or moreover to know the number of the periodic cycles per year, if more than one cycle are completed annually.

From this point of view, the present study is attempted to know the growth rate of the baleen plates for a certain length of time and then, being based on the result of that study, to settle the problem that the annual growth of the baleen plate really corresponds to the

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periodic cycle of the sculpture, i.e., the interval zone between the neighbouring two main sculptures. The baleen plates of the Antarctic whales were used as the material, because they were available in large numbers and in the most reliable condition. As was in the previous study, the present materials of the baleen plates were collected from all the whales caught by both the two Japanese whaling fleets, the Hashidatemaru Fleet of the Japan Marine Products Co., Ltd., and the Nisshinmaru Fleet of the Taiyo Fishing Co., Ltd.

The collection of the material was carried out through the cordial cooperation of the crews of the two fleets, under the direction of the inspectors-on-board of the Japanese Government, Messrs. Haruyuki Sakiura, Yoshiro Teraoka, Koji Ishizuka, Yasuo Usukura, Kazuhiro Mizue and Tadashi Murata. The Japan Whaling Association rendered much cooperation and gave facility to the writer in preparing the special apparatus for measuring the sculptures of the baleen plates. Mr. Minoru Kubota offered much assistance in the calculation of the data. These cooperations and assistances, which made this study possible, are all heartily appreciated.

#### Chapter I

## The Material

On board the factory ships, the largest left and right baleen plates were collected as the sample from each whale. On this sampling, the line of insertion of the baleen plate into the gum (the intersection of the baleen plate with the contour of the gum) was clearly marked on the baleen plate with a knife-cut, as designated by "a" in Fig. 1. There are in the baleen plate several number of thin zones which are recognizable superficially by rough sculptures. Among them, that nearest to the said knife-cut is designated by "b" in Fig. 1, and is called the first main sculpture" in the present paper. At this part of the baleen plates of some whales, there are recognizable some distinct pockmark-like spots. The next thin zone to that of the mark "a" is denoted by "c" (the "second main sculpture"), and the base of the baleen plate by "d" (Fig. 1.).

The growth of the baleen plate takes place at the base "d", not at the line of the insertion into the gum, "a". Now, assume that the baleen plate grows in such a way as human nails do. Then, "a", "b" and "c" will go apart from "d" at an equal rate as the baleen plate grows. Therefore, in the study distances "ab" and "bc" were measured to estimate the annual growth of the baleen plates from the change of the ratio ab/bc during the whaling season. This ratio was chosen as the index of the growth of the baleen plates, and not the actual length of ab, which varies much from whale to whale, because the rate of the growth of the baleen plate during the formation of the interval zone between the main sculptures, and consequently the width of that interval zone, seemed to be considered as constant for each individual whale. (Speaking more precisely, however, the width of the interval zone becomes smaller, though slightly, toward the younger part of the plate, i. e., toward the gum.)

The percentage value fo the ratio ab/bc is called the "ab-percentage" in the following discussion. The average of the ab-percentages for the right and left plates of the individual whale was defined as the ab-percentage for that individual. The ab-percentages for the individuals caught in each one- and three-week period were averaged, thus yielding the weekly and tri-weekly mean of the ab-percentage respectively. The tri-weekly mean was adopted instead of the mean for four-week period or the monthly mean, because the former seemed to be compared with each other more fairly than the latter in the case of the 1949-50 whaling season, which lasted for only ten weeks. In averaging three weekly means to obtain the tri-weekly mean, the formers were weighted with the number of the whales caught in the respective weeks.

Following discussions are primarily based on the data for all the fin

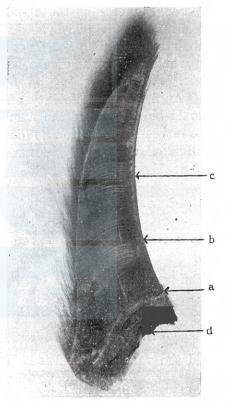


Fig. 1.], Growth Index of Baleen Plate (ab-Percentage) measuring point.

whales (211 famales and 248 males, totaling 459) and all the blue whales (114 females and 270 males, totaling 384) that were caught by the Hashidate-maru Fleet. On the course of reasoning, the data for all the fin and blue whales, 597 fin and 433 blue whales, that the

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Nisshin-maru Fleet caught, were also referred to. The baleen plates of 37 fin and 18 blue whales, on which the collectors failed to cut the "a" marks, were inevitably excluded from the data.

#### Chapter II

## The Periodic Cycle of the Sculptures on the Baleen Plates of Fin Whales

The ab-percentages for individual whales as defined in the preceding chapter are plotted against the date of catch in Fig 2-a for the female and in Fig. 2-b for the male fin whales. Daily, weekly and tri-weekly means of the ab-percentage are computed from the values of all the individuals caught during the respective periods, and are tabulated in Table 1-a for the female and in Table 1-b for the male fin whales. Growth of the baleen plates can be estimated from these data.

At first, the reliability of the tri-weekly means of ab-percentage should be tested. If the number of the sample whales caught during the three-week period is denoted by N, and the ab-percentage of individual whale by  $y_i$ , *i* standing for each of the N whales, then the tri-weekly mean  $\bar{y}$  is

$$\bar{y} = \frac{\Sigma y_i}{N}$$
,

the variance, V, is

$$V{=}rac{\Sigma(y_i{-}ar{y})^2}{N{-}1}$$
 ,

the standard deviation, S. D., is

$$S. D. = \sqrt{V}$$
,

and the variation coefficient is

$$v = \frac{(S.D.)(100)}{\overline{y}}$$

To determine the fiducial limits of the unknown population mean m from the sample mean  $\overline{y}$ 

$$F \ge N(\bar{y}-m)^2/V$$

Taking the square roots of both members,

$$-\sqrt{\frac{F.V}{N}} \leq (\bar{y}-m) \leq +\sqrt{\frac{F.V}{N}}$$

This formula indicates the range of the tri-weekly mean. The values computed with these formulae are entered in the columns right to the tri-weekly means in Table 1's.

If an approximately constant growth rate is postulated for the baleen plates, the relation between the accumulated growth of the baleen plate and the number of weeks that have elapsed can be considered as being linear. Now, assume a linear relation between the number of weeks, symbolized by the variable x, and the accumulated growth of the baleen plate, the variate y. Then, based on the points  $(x_1, y_1), (x_2, y_2), (x_3, y_3), \ldots, (x_N, y_N)$ , the regression equation which is expressed in the following form is obtained:

 $y = \overline{y} + b(x - \overline{x}) = A + B_x$ 

where  $\overline{x} = \frac{\sum x_i}{N}$  and  $\overline{y} = \frac{\sum y_i}{N}$ , A and B being the constants. If the

variances and the covariance are defined as

$$S_{z} = \sum (x_{i} - \overline{x})^{2},$$
  
$$S_{y} = \sum (y_{i} - \overline{y})^{2},$$

and

$$C = \sum (x_i - \overline{x})(y_i - \overline{y}) ,$$

then the constant B is written

 $B = C/S_x$ 

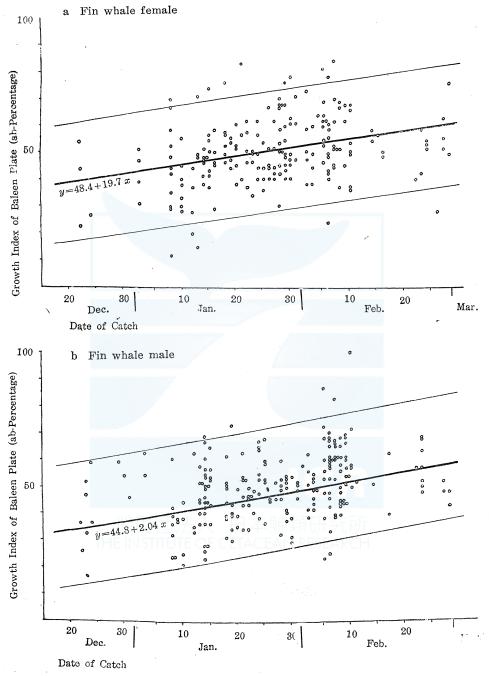
And if  $(C^2/S_x)(\Delta/(N-2)) > F'_{N-2}$ , where

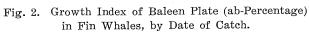
$$\Delta \equiv (y'_{i} - y_{i})^{2} - S_{y} - C^{2}/S_{x},$$

then much of the variation in y is attributable to the variation in x, that is, the accumulated growth of the baleen plate varies much from week to week.

To simplify the calculation, the week January 19-25, 1950, is taken as the origin of x, that is, for this week x=0. This week is the fifth and middle week in the 1949-50 whaling season which lasted for ten weeks from December 22, 1949.

Then, the values are computed with the formulae above and are listed in Tables 2-a and -b for the females and for the males respectively.





n accumulated Growth of their Baleen	eek and 3-Week Period.
ht (N) and Mean a	e) for each day, Week and 3-Week
of Fin Whales caug	Mean ab-Percentag
able 1. Number of	Plates (Mean

									a. Fin	Ň	Fin Whale Female	emal	e							
-	Week					Ĕ	For Each	ch Day	of	the	the Week					Fo	For Each	ЧОЧ	Flach 2.1	Flach 2 meals Danied
Waak		Ē	hurs.		Fri.		Sat.	_	Sun.		Mon.		Tues.	2	Wed.	-	Week	5	המרוח ה-א	
Number	Date	z	Mean ab-%	z	Mean ab-%	Z Z	Mean ab-%	Z U	Mean ab-%	Z	Mean ab-%	z	Mean ab-%	z	Mean ab-%	4	Mean ab-%	Mean ab-%	Range of Mean	Level of Sig- nificance in
Ι	12/22-12/28	4	44.6	T		1	21.5									ũ	35.6			
II	12/29-1/4									01	35.1	01	48.8			4	42.0	39.7	$\pm 5.20$	5.26
III	1/5-1/11	_		XU 4	<u>h 9</u>			11	42.2			2	38.4			18	40.3		_	_
IV	1/12-1/18	4	32.4	4	45.3	3 10	46.7	4	51.8	ю	45.6			9	55.9	8	46.3			_
Λ	1/19-1/25	4	49.8	20	49.1	°°	59.2	5	50.2	4	44.6	õ	49.9	4	43.3	30	49.4	48.9	$\pm 6.30$	5.17
IΛ	1/26-2/1	10	49.3	10	47.5	II	54.5	6	52.7	2	54.3	<del>ന</del>	58.7			40	52.8			-
IIΛ	2/2-2/8	ŋ	52.9	4	47.5	1	63.2	6	60.5	10	49.6	00	56.9	ŝ	55.7	42	55.2			
VIII	2/9-2/15	ရာ	57.5	2	51.0	7	55.9		-			6		Ч	56.3	13	54.9	54.6	人士5.98	4.39
ΙX	2/16-2/22	01	49.1	C F	-6										39.6	က	44.4			-
X	2/23-3/1	01	50.1	က	52.7			н	28.4	ന	56.2	67	61.9			11	54.2			
				ÊF					b. Fin	M	Whale Male	ale								
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Number	Date	z	Mean ab-%	Z	Mean ab-%	Z	Mean ab-%	Z	Mean ab-%	z	Mean ab-%	Z	Mean ab-%	z	Mean ab-%	z	Mean ab-%	Mean ab-%	Mean Range ab-% of Mean	Level of Sig. nificance in
Ι	12/22-12/28	01	31.6	4	37.2	67	47.8									00	38.4			
II	12/29–1/4		59.0	-	54.1	-	41.3					C1.	32.8			10	40.0	37.9	$\pm 4.75$	5.73
III	1/5-1/11			L.			•	5.	36.5	01	37.8	4	31.9			13	35.1	_	_	<u> </u>
IV	1/12-1/18	က	43.3	2	46.2	19	45.5	9	50.4					00	45.3	43	46.1			
Δ	1/19-1/25	9	43.0	ം 	41.1	80	46.9	2	45.2	01	44.1	2	54.9	9	53.7	39	47.9	46.5	$\pm 2.91$	2.52
Ν	1/26-2/1	01	44.5	01	48.0	9	47.5	7	47.6	4	37.1	4	48.7	H	56.8	36	45.1	_	_	-
ΛII	2/2-2/8	ŋ	49.1	8	47.5			6	55.2	16	53.5	10	58.3	12	59.1	66	52.7		_	(
IIIΛ	2/9-2/15	10	57.3	Ð	65.9	<del>,</del>	51.2					Η	49.8			17	59.0	54.0	$\pm 6.42$	4.78
IX	2/16-2/22			07	49.9									ч	56.4	က	52.1	_	_	
X	2,23-3/1	Ľ-	56.8							¢1	49.1	୍ୟ 	44.4			Π	53.1			

## On the Periodic Mark on the Baleen Plates

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Formula	(a) Valu	e for	Females	(b) Va	lue fo	r Males
Say		-79	.8	·	27	73.1
$\Sigma ay^2$		5498	8.52		759	01.23
Saxy		1658	3.20		188	32.6
$S_x = \Sigma a (x_i - \overline{x})^2$			869			1010
$S_y = \Sigma a(y_i - \bar{y})^2$		5466	5.52		759	1.23
$C = \Sigma a(x_i - \overline{x})(y_i - \overline{y})$		1709	.20		206	51.68
$\bar{y} = \Sigma a y / N$		-0	.40			1.21
$\bar{x} = \Sigma a x / N$		0	.65			0.96
$N = \Sigma a$			199	-		225
$B=C/S_x$		1	.97	2.04		2.04
<b>A</b> .		48	3.4		4	4.8
$C^2/S_x$	3164.12	1	3164.13	3509.09	1	3509.09
$\Delta = S_y - C^2 / S_x$	2302.39	197	11.69	4082.14	223	18.35

Table 2Formulae and Values of Calculation for Fin Whales.

As the summary, the growth of the baleen plates of fin whales is expressed in the general formula

## y = A + Bx,

where y stands for the weekly mean of ab-percentage (the index of the accumulated growth), and x for the week number as taken to be zero for the week, January 19-25, 1950. B, being defined as  $C/S_x$ , is the mean weekly growth rate of the baleen plate. Substituting the values of Table 1's in the general formula,

for females:

y = 48.4 + 1.97x

and for males:

#### y = 44.8 + 2.04x

From these equations, it can be seen that by the week January 19-25, 1950 for which x=0, the baleen plates of female and male fin whales have already grown by 48.4% and 44.8% of the width of the interval between the two successive main sculptures, respectively.

If it is assumed that the main sculptures are formed at that period of the year when the whales are most poorly nourished, such period can be estimated by substituting y=0 in the growth equations. Then, the results are On the Periodic Mark on the Baleen Plates

for females:

$$y=0=48.4+(1.94)(-25)$$

and for males:

y=0=44.8+(2.04)(-22),

indicating that the nutrition of the female and the male fin whales would have been worst 25 and 22 weeks before the origin week respectively, i.e., in late July and in middle August.

Again assume that the length of ab would have attained the width of the interval of the two successive main sculptures in the next malnutritious period, had the whales survived. By definition, y=100for that period. Substituting this in the equations,

for females:

 $y = 100 \div 48.4 + (1.94)(+27)$ 

and for males:

$$y = 100 \Rightarrow 44.8 + (2.04)(+26)$$

Accordingly the fin whales of both sexes would have met the malnutritious period 27 weeks after the origin week or in early August.

Furthermore, 52, which is the number of weeks in a year, times of the mean weekly growth rate B is approximately equal to 100% for both sexes, proving that the interval zone between the two successive main sculptures corresponds to the annual growth of the ballen plate. (More precisely, the products 52B exceed 100% slightly. But this seems quite natural because the material baleen plates were sampled during the feeding season of the whales, when the growth rate of the baleen plates, which varies from one week to another, was probably at the maximum level of the year.)

From the foregoing estimates of the malnutritious periods, it can be deduced that the nutrition of fin whales is at the lowest level between late July and middle August. This presumption perfectly coincides with the findings hitherto made on the life history of fin whales. It was also found, at 5% level of significance, that the interval zone between the two successive main sculptures on the baleen plates of fin whales corresponds to the annual growth of the baleen plate. In other word, the periodic cycle of the main sculptures on the baleen plates of the fin whales is one year.

#### Chapter III

## The Periodic Cycle of the Sculptures on the Baleen Plates of Blue Whales

The baleen plates of blue whales were also treated by the same method as those of fin whales. In Figs. 3-a and -b are plotted the ab-percentages for the female and the male individuals, respectively, against the date of catch. Daily, weekly and tri-weekly means of abpercentage are listed in Tables 2-a and -b for the females and for the males respectively. In these tables are also listed the range and the level of significance of the tri-weekly means. Both of these were computed by the same statistical procedures as in the case of fin whales. The process of the calculation is tabulated in Table 4.

From these data, the equation of the growth of the baleen plates of the female blue whales (Fig. 3-a) is doubtless

y = 41.0 + 2.04x

which is similar to the equation for the fin whales.

Substitute 0 and 100 for y as in the case of fin whales, yielding

y=0=41.0+(2.04)(-20)

and

 $y = 100 \div 41.0 + (2.04)(+29)$ 

Accordingly both of the estimates of two malnutritious periods are around the middle and late August. And 52 times of the mean weekly growth rate approximates to 100%, also as in the case of fin whales.

For the male whales, however, the equation of growth of baleen plates is deduced as being

y = 41.0 + 0.68x

from Fig. 3-b (regardless of black and white of the dots) and Table 3-b. Accordingly the time required to complete the interval zone between two successive main sculptures is, by solving  $y=100\div41.0+0.68x$ , 96 weeks or about 1.8 years. This result is contrary to those for the fin whales and for female blue whales.

Now, examine Fig. 3-b carefully but regardless of black and white of the dots: then, it is noticed that the dots are concentrating between

20 and 50% of the ab-percentage value for the period from the middle (about the 14th) of February to the beginning of March. The same fact is also observable in Table 3-b: the weekly mean of ab-percentage is 29.4% and 32.7% for the first and second week of the season respectively, and thereafter increases with the week number, till 48.4% is reached in the seventh week; then the value turns to decrease; 48.3% for the eighth week, 40.5% for the ninth and 41.8% for the tenth. For the first seven weeks, when the mean ab-percentage increases steadily, the growth of the baleen plates of male blue whales does not differ much from that of female blue or fin whales. It is for the later part of the season that the growth of baleen plates disagrees between the two groups. And this disagreement is the cause of the difference between the growth equations of the two groups.

The only possible explanation for this disagreement seems to postulate some unknown factor which might have come into effect in the later part of the whaling season.

Along this line of reasoning, the male blue whales were divided into two groups: those caught on and before February 13, 1950 and those caught thereafter. Regarding the former group, the equation of the growth of the baleen plates is

y = 43.4 + 2.01x

The constants in this equation hardly differ from those in the equations for female blue whales and fin whales of both sexes.

The whales killed after February 14, 1950 were classified on basis of the rate of diatom infection; but this analysis did not lead to any helpful conclusion. Then the thickness of the blubber was considered. The thickness of blubber of each whale, as measured at the body part  $P_1$  (the point on the horizontal cut side of the body, where it intersects a vertical line from the dorsal fin), was expressed in the percentage to the body length and was plotted against the date of catch (Fig. 4.). The straight line appearing in that figure is the standard trend of the change of the thickness of blubber throughout the whaling season, as computed with the data published in September 1949 (Nishiwaki: 1949a).

Such dots are stained black in Fig. 4, that are located above the standard trend line in that figure and correspond to the dots situated below the straight line y=43.4+2.01x in Fig. 3-b. It seems to be one way of thinking to consider the individuals represented by these black dots as such whales that arrived to the Antarctic Ocean comparatively

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late and grow fat thereafter pretty fast. In other word, we can consider them as being engaged in a different migration from that of the normal whales. The calculation with these (black dots) whales omitted will yield Fig. 3-c and, for and after the eighth week of the season, Table 3-c in place of Fig. 3-b and Table 3-b.

From Table 3-c the mean ab-percentage for the last three-week period of the season (from the seventh to the ninth week) is 48.7%, which agrees, at 1.5% level of significance, with the mean computed for the same period with the equation we obtained for the whales caught before February 13 (y=43.3+2.01x), 49.4%.

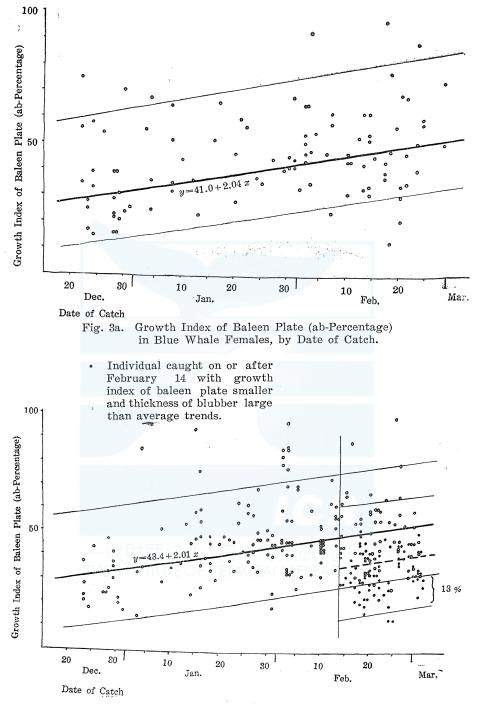
Therefore, it is possible to conclude that the growth of the baleen plates of male blue whales is expressed as

#### y = 43.4 + 2.01x

From this, the periods of undernourishment are estimated around early and middle August, and the interval between the two successive main sculptures is proved to correspond to the annual growth of the baleen plate.

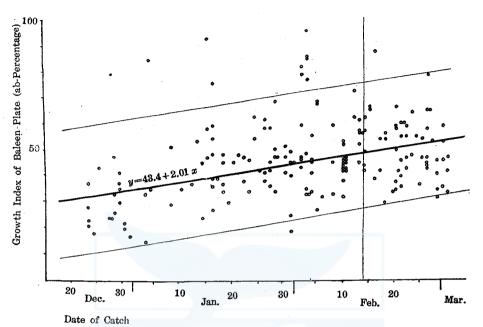
If the growth of the baleen plates is influenced by the feature of the migration of the whale, as we have assumed for the male blue whales, the baleen plates of the female must be affected in a similar way. So, the data of female blue whales were treated by the same method. But it was found that the equation of the growth of their baleen plates is hardly affected by this factor. This proves that the migration of the blue whales differs by sexes. A very similar conclusion was reached regarding the migration of this species by Hideo Omura in his study on the diatom infection on blue and fin whales in the Antarctic Whaling Area V (Omura: 1950).

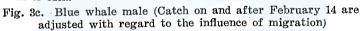
Finally, some references will be made to the variation in growth of baleen plates among those individuals which were considered as arriving comparatively late to the Antarctic Ocean in the foregoing discussions. As indicated in Fig. 3-b, there is the variation as much as 13% in terms of ab-percentage or about 6 weeks in terms of time in the growth of the baleen plates among these late-arriving male blues. This implies that there is as much variation in the nutritious period of these whales. Then, there may be as much variation in their migration, in the growth of their bodies and, probably, in the period of their births. These subjects, however, will be studied in other occasions, for such studies are not the purpose of the present work.



## Fig. 3b. Blue whale male







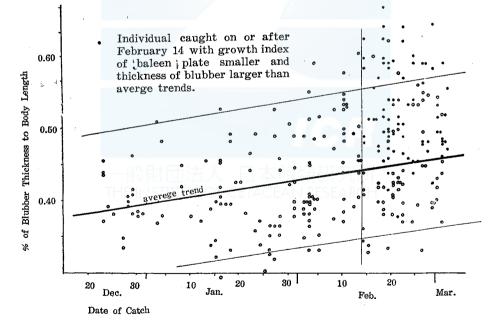


Fig. 4. Thickness of Blubber at Body Part  $P_1$  in Male Blue Whales.

Table 3. Number of Blue Whales caught (N) and Mean accumulated Growth of their Baleen Plates (Mean ab-Percentage) For Each Day, Week and 3-Week Period.

a. Female

-	Week					Foi	For Each Day of the Week	Da	v of th	le W	reek					For	For each		Flach 2 n	Tow Pach 9 maak Panind
Week		F	Thurs.	1 H	Fri.	20	Sat.	Ś	Sun.	Z	Mon.	Ę	Tues.	M	Wed.	A	Week	5 4		notro y voor
Number	Date	z	Mean ab-%	Z	Mean ab-%	z	Mean ab-%	N	Mean ab-%	z	Mean ab-%	Z	Mean ab-%	Z	Mean ab-%	z	Mean ab-%	Mean ab-%	Mean Range ab-% of Mean	Mean Range Level of Sig. ab-% of Mean nificance in
П	12/22-12/28	Ч	35.0	ಣ	23.4	က	28.9		7					2	26.3	14	26.9			
II	12/29–1/4	01	26.0	는	34.3	-	26.2					1	55.0	2	30.3		31.1	30.3	+±11.06	14.65
III	1/5-1/11							ന	39.4				44.0			4	40.5			_
νī	1/12–1/18		35.7	<b>–</b>	22.5	-	36.1			П	51.3					4	36.4		~	
Δ	1/19–1/25			67	36.9	Ч	59.2		55.8		2	Ч	37.1	Ч	34.6	9	43 4	40.9	$\downarrow \pm 6.58$	6.45
IΛ	1/26-2/1		ESE/	-	43.8			Η	40.0	21	43.0			1	33.4	<b>C•</b> .	41.2			
IIV	2/2-2/8	9	53.1	က	41.9					н	45.7	ಣ	47.3			13	48.6			
IIIΛ	2/9–2/15			4	43.4			r-4	51.5	ရာ	45.4	ъ	45.9	-	45.2	14	44.8	45.8	$\pm 6.45$	4.17
XI	2/16-2/22			ന 	39.4		46.5	-	56.1	27	27.5	ы	49.6			12	43.8	<u> </u>		
X	2/23-3/1	01	45.0	۵۱ 	57.7				-			21	61.4			9	54.7			

## On the Periodic Mark on the Baleen Plates

For Each 3-week Period		Mean Range Level of Sig- ab-% of Mean nificance in		8.68		_	1.86	_							·	Poch 2 most Davied	DOLLA L MOD	Mean Range Level of Sig- ab-% of Mean nificance in		2.12			·
lach 3-w		Mean Range ab-% of Mean	_	±6.85	_	~	$\pm 2.04$				_					Toob 2 m		Range of Mean		±2.56	_		
Hor I		Mean ab-%	_	-31.7	_	_	1.44	_								ц Ц С Ц	5	Mean ab-%	_	48.7	_		
For Each	Week	Mean ab-%	29.4	32.2	38.3	43.0	44.5	44.8	48.4	48.3	40.5	41.8	34.1			For Each	Week	Mean ab-%	48.4	44.9	47.9	48.4	
For	M	z	Ц	10	က	17	10	33	32	36	68	43	14		als	For	₿	z	25	31	31	22	
	Wed.	Mean ab-%	34.9	35.1		40.9	41.5		45.9	54.1	39.7	40.8			regarding the Migration by excluding the Individuals which arrived late to the Antarctic Ocean.		Wed.	Mean ab-%		65.3	50.1	48.3	
	5	z	က	-		4	-1		က	က	10	8		:03	le Ir ean.		Δ	z		61	4	4	
	Tues.	Mean ab-%	24.4	24.0	35.3	38.6	58.1	37.1		48.3	41.8	40.2		14, 19	ling th tie Oce		Tues.	Mean ab-%		51.8	46.6	48.4	
	Ļ	z	61	\$		1	61	9		6	10	9		ary	tare		H	Z		Ð	2	4	
Week	Mon.	Mean ab-%				43.9	39.9	46.2	32.4	51.3	46.3	45.1		Febru	t by each	eek	Mon.	Mean ab-%			56.6	52.8	
Day of the We	M	z				9	01	က	ч	2	57	က		fter	ation to t	le W	Z	z			2	1	
	Sun.	Mean ab-%	43.1		39.8	52.7	42.4		54.1	67.4	38.3	43.9		Males caught after February 14, 1950:	Migra d late	For Each Day of the Week	Sun.	Mean ab-%			42.2	46.1	<u> </u>
		z	٦		67	07	01		က	01	13	12		s cal	the trive	Day	Ś	z			က	7	
Each		Mean abl%	18.3	16.7		34.6	48.2	49.4	44.3		40.2	33.9			egarding the Migration by excluding the I which arrived late to the Antarctic Ocean.	Each	Sat.	Mean ab-%			45.2	40.7	
For	S	z	1	-		21	-	ŋ	10		13	4		ల		For	S	z			9	က	
	Fri.	Mean ab-%	27.2	21.2		53.4	44.5	45.1	55.8	44.4	35.6	31.1	31.2		adjusted		Fri.	Mean ab-%			42.3	40.6	35.7
		z	4	64	<u>д</u> β	-	Ч	4	50	15	9	5	က	京类	ñ	h		z			ന 	61	
	urs.	Mean ab-%		35.9		36.6	30.3	49.4	58.8		47.6	45.3	34.9		RESE		hurs.	Mean ab-%			87.0	63.0	46.9
	F	z		4		н	٦	4	9		4	со 	11				L	z					<i>ი</i> ,
Week		Date	12/22-12/28	12/29-1/4	1/5-1/11	1/12-1/18	1/19-1/25	1/26-2/1	2/22/8	2/9-2/15	2/16-2/22	2/23-3/1	3/23/8			Week		Date	2/2-2/8	2/9-2/15	2/16-2/22	2/23-3/1	3/2-3/8
		w eek Number	I	II	III	IV	Λ	IΛ	ΝI	IIIA	IX.	X	IX			Δ	Waal	Number	ΝII	IIIV	IX	X	IX

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Formula	(a) Value	s for H	Temales	(b) Val	ues fo	r Males
Σαγ		+83	.5		+24	6.5
$\Sigma ay^2$		5843	.15		965	8.29
$\Sigma axy$		1795	.19		412	9.7
$S_x = \Sigma a (x_i - \overline{x})^2$			834			2029
$S_y = \Sigma a (y_i - \bar{y})^2$		5713	.01		658	9.49
$B = \Sigma a(x_i - \bar{x})(y_i - \bar{y})$		1704	.5		408	3.7
$\bar{y} = \Sigma a y / N$		+0	.95	•	+	1.34
$\bar{x} = \Sigma a x / N$		+0	.74		+	1.96
$N = \Sigma a$			87	198		
$B = C/S_x$		2	.04	2.01		2.01
A		41	.0		4	3.4
$C^2/S_{2}$	390.21	1	390.21	843.19	1	843.19
$\Delta = S_y - C^2 / S_x$	5372.80	85	62.47	5746.30	196	29.12

Table 4.Formulae and Values of Calculation for Blue Whales.

#### Summary

In this study the periodic cycle of the sculptures on the baleen plates of whales were investigated. Through this investigation, it was intended to estimate the growth of the baleen plates and the time required to complete the interval zone between the successive two main sculptures.

The ab-percentage, namely, the percentage of the distance between the insertion of the baleen plates (into the gum) and the first main sculpture (as counted from the gum) to the distance between the first and the second main sculptures, was chosen as the index of the growth of the baleen plates.

It was found that the growth of the baleen plates can be expressed by the straight line which can be written

y = A + Bx

where x denotes the week number and y the weekly mean of abpercentage, A and B being the constants. The constant B is the mean weekly growth rate of the baleen plates. The fifth week (January 19-25, 1950) of that whaling season which lasted for nine weeks from December 22, 1949 was taken as the origin for x, i.e., x=0 for that week. Consequently the constant A gives the mean accumulated growth of the baleen plates at the fifth week, in terms of ab-percentage,

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Values of A and B were computed for each sex of fin and blue whales with the data of the measurements. Substituting these values in the foregoing formula, the following equations of the growth of the baleen plates were obtained:

for female fin whales:	y = 48.4 + 1.97x
for male fin whales:	y = 44.8 + 2.04x
for female blue whales:	y = 41.0 + 2.04x
and for male blue whales:	y = 43.4 + 2.01x

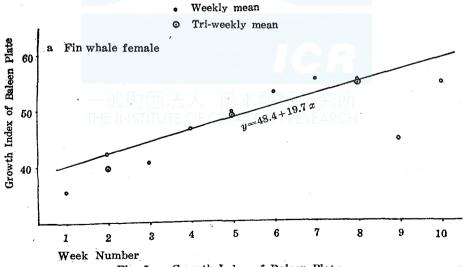
These equations were graphed together with the weekly and tri-weekly means of ab-percentage in Fig. 5.

From these equations it can be deduced that the nutrient of the whales is at the lowest level from late July to late August. This result is in line with the knowledges on the life history of whales so far obtained.

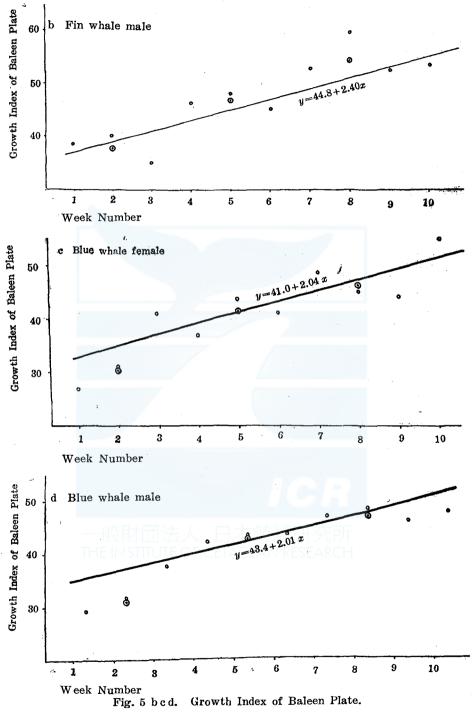
Also from the same equations the growth of baleen plates in each week is about 2% of their annual growth. This proves that the interval zone between the two successive main sculptures is formed in one year.

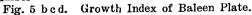
As to the migration, it was deduced that the fin whales arrive to the Antarctic whaling grounds earlier than the blue whales.

Now that the interval zone between the successive two main sculptures on the baleen plates is proved to correspond to the annual growth of the baleen plates, and these sculptures can be utilized as the age marks in the studies on such various important biological charactors









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of the whales, as the age at sexual maturity, the number of ovulation in each breeding season, and many others.

Accordingly, the investigation on these sculptures seems to be an effective and essential tool for the study of the life history of Mystacoceti. These prospects will be examined in the following reports. Here the discussion will be confined to the conclusion reached in the present study.

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STITUTE OF CETACEAN

# On the Sexual Maturity of the Sperm Whale (Physeter catodon) found in the Adjacent Waters of Japan (I).

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(27. May. 1951)

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## Introduction

Since 1948 the ecology of the whales found in the waters adjacent to Japan has been studied by a group of biologists of the Fisheries Agency, Japanese Government and of the Whales Research Institute. Such biological charactors of the various whale species have been surveyed in this study as body length composition, body length at the sexual maturity, geographical distribution, migration, age and others.

While mature female whales can easily be distinguished from those

sexually immature by the presence of yellow bodies in their ovaries, whether a male whale is sexually mature or not can hardly be determined exactly without examining its testes histologically.

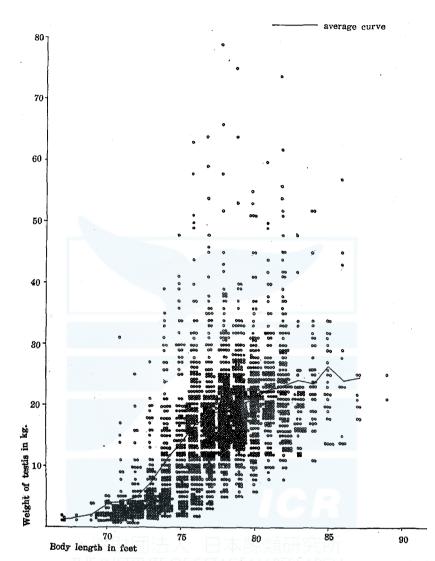
Mackintosh and Wheeler, however, correlated the results of their histological observations on the testes of the Antarctic blue and fin whales to the volumes of their testes, and concluded that the degree of the sexual maturity of these whales can be determined on basis of the volume of their testis (Mackintosh and Wheeler: 1929). Matthews applied an alike treatment to the sperm whales from the Southern hemisphere, and estimated their body length at the sexual maturity at 38 to 41 Eng. ft. (Matthews: 1938).

Aforesaid Japanese group has been measuring the volume and weight of testis of the whales as a tentative means of studying the body length of male whales at the sexual maturity. As far as the data hitherto collected by this group concern, both the ratio "testis volume / body length" and the ratio "testis weight / body length" show the same trend, and the latter ratio correlates better to the degree of sexual maturity than the former.

As an illustration, we shall quote the case of the Antacrtic male blue whales which were caught by two Japanese whaling fleets during the three seasons 1946–47 through 1948–49. This case has been studied by the senior author of this paper (Nishiwaki: 1950).

In Figure 1 are plotted the weight of testis of each individual blue whale against the body length. Inspecting the distribution of the plots in the figure, one can segregate the sexually mature whales from the immature individuals with a considerable accuracy. The average weight of testis for the body length class increases with the body length, gradually for lower body length classes but rapidly for the body lengths 72 Eng. ft. and over. Consequently the curve depicting these averages is discontinuous at the point "72 Eng. ft. (body length)-5 kg. (weight of testis)." This seems to show that both the volume and weight of the testis increase rapidly as the result of the vigorous multiplication of testis contents. The formation of spermatozoa was actually observed in the histological samples of the testes of this stage. Beased on these data and reasoning, it was concluded that 10 kg. is the average minimum weight of the pair of testes of the Antarctic male blue whales at the time of the sexual maturity, and that the average body length at that stage measures 75 ft. since the average weight of a pair of testes attains 10 kg. at this body length.

Sei and fin whales found in the waters adjacent to Japan have been



#### On the Sexual Maturity of the Sperm Whale-

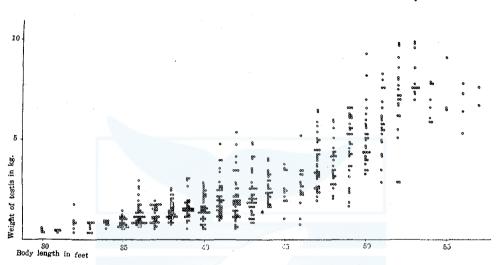
Fig. 1. Weight of testis and body length in Antarctic blue whales caught by Japanese fleets, 1946-47 through 1948-49 (from Nishiwaki: 1950)

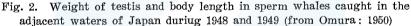
treated in the same method as above. The results, though they are not yet conclusive on account of the scarcity of the data, are usable in estimating the sexual maturity of the males in rough.

As to sperm whales, however, the difference in the weight of testis between mature and immature groups is not so conspicuous as in the foregoing species, nor any discontinuous points have been detected on their "body length-testis weight average curve." As an illustration,

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the relation between body length and weight of testis of the sperm whales caught in Figure 2. Original data were taken from Omura (Omura: 1950).





This figure hardly suggests any standard that can be used for estimating the sexual maturity of these male sperm whales. Thence it was decided to invetigate the testis of sperm whales histologically, and we have been in charge of that investigation.

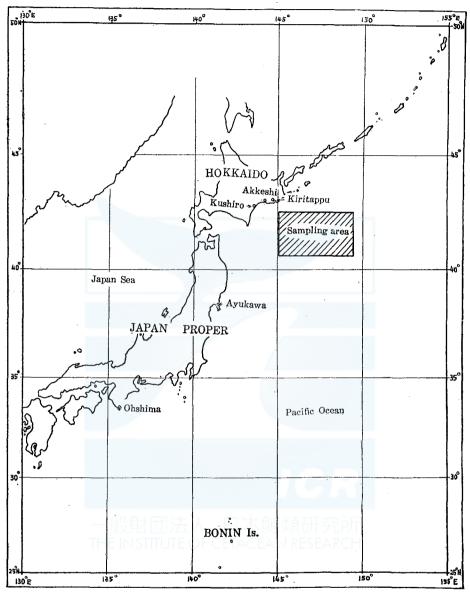
The joint author, Takashi Hibiya, has been participating in this study in order to handle a mass of histological samples in a short period of time and to supply more specialized histological knowledges, ever since such assistance became necessary in 1951. Therefore, he did not take part in such phases of this study as planning and collection and fixing of the histological samples.

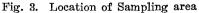
#### Chapter I.

#### Method of Investigation and the Materials

The materials used for this study are the sperm whales caught in the waters south-east off Hokkaido, Japan (Figure 3) during the period July 19-October 6, 1950. They consist of 90 males of the body lengths between 35 and 53 Eng. ft. and 80 females between 34 and 39 Eng. ft.

On the Sexual Maturity of the Sperm Whale





These whales were towed to and dissected at the land stations 14 to 24 hours after they were captured. When they were dissected, the measurements were made in the ordinary manner, their testes or ovaries were taken out, and the right and left testis were weighed separately. Thereafter, in the case of male, each small sample piece of the right and left testis was cut off and was immediately fixed in the alcohol-formalin mixture. A few samples were fixed in formalin as the former solution was not available. The fixed sample pieces were cut into the sections 5 to 7 micron thick in the paraffin method, double-stained with haematoxylin-eosin, and subjected to the microscopic examination. In the case of female, yellow bodies (corpla lutea) were counted in the usual method.

We express our heartfelt thanks to the Japan Marine Products Co., Ltd., the Taiyo Fishing Co., Ltd. and the Kyokuyo Whaling Co., Ltd. for their immense cooperations in collecting the materials and data for this study. We are grateful also to Messer. Keijiro Maeda, Haruyuki Sakiura, Yoshio Teraoka, Setsuo Nishimoto and Shigeo Miyamoto, government inspectors from the Japanese Fisheries Agency, and to Mr. Kazuo Fujino, member of the Whales Research Institute who all directed the collection of the materials in the field. Our thanks are also due to the Japan Whaling Association to which we owe much of the research fund for this study.

## Chapter II.

## Sexual Maturity of the Male and its Relation with Body Length and Weight of Testis

Since the spermatogenesis generally takes place in the convoluted seminiferous tubules ("Tubuli seminiferi contorti") and not in the straight semineferous tubules ("Tub. semini. recti") in mammals, we examined the former tubules closely and determined whether the spermatozoa were present or not.

All the male sperm whales that were dealt with in the present study measured more than 35 Eng. ft. in body length, and needless to say, none of their testes were found in such an immature stage where the spermatogonia, mingled by Sertoli's cells, are arranged on the inner wall of the convoluted seminiferous tubules in a simple manner. The earliest stage we observed was such where numbers of spermatocytes were observable. We met with a variety of more advanced stages, including the one at which spermatocytes were in division, the one where spermatids were already observable, or those where the development was further advanced. The last include the stage where spermatozoa are clearly seen. (See the photographs in the Appendix.)

The well developed spermatozoa of the sperm whale as we observed was of the shape typical to that of the mammal.<sup>1</sup> Its head was pos-

## On the Sexual Maturity of the Sperm Whale

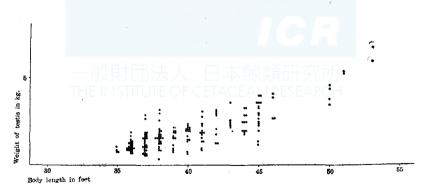
## Table 1.

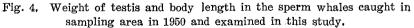
Body Leng	th Number	of Testes E	xamined	Percentag	e to Total
in English Fe	eet Total	4	_	÷	-
35	4	4	0	100	0
36	26	21	5	82	18
37	28	24	4	86	14
38	26	24	2	92	8
39	12	11	1	92	8
40	16	15	1	94	6
41	14	13	* 1	100	0
42	8	8	0	100	0
43	- 6	6	0	100	0
44	12	12	0	100	0
45	16	16	0	100	0
46	4	4	0	100	0
47	0	0	0	0	0
48	0	0	0	0	0
49	0	0	0	0	0
50	4	4	0	100	0
51	2	2	. 0	100	0
52	0	0	0	0	0
53	2	2	0	100	0

## Maturity of testis of the male sperm whale at various body length

+: Spermatozoa were found in the histological sample.

-: Spermatozoa were not found in the histological sample.





O Sexually immature whale

Sexually mature whale

sessed of a distinct acrosome and was rather elongated. The middle piece, approximately as long as the head, followed the head and terminated in a long tail.

At first, all the section preparats of testes that were prepared in the aforesaid method were examined for the presence spermatozoa. The result is summarized for each body length class and is presented in Table 1 and Figure 4.

We had two examples of the body length of 35 Eng. ft., in the testes of both which the spermatozoa were detected. In each body length class from 36 to 40 Eng. ft. there were few individuals from which spermatozoa were not detected, and the percentage of such whales gradually decreased towards higher body length classes. As we shall discuss later, weights of the right and the left testis of an invidual whale were not always equal: in general they were unequal rather than equal. In some individuals the spermatozoa were found from only one of their pair of testes, and not from the other. Such cases counted three in the body length class 36 Eng. ft., two in both of the 37 and 38 Eng. ft. classes and one in both of the 39 and 40 Eng. ft. classes. In two of these cases the spermatozoa were found only in the right testis; in the other seven cases they were detected only in the left testis. There were two individuals in all from the testes of which the spermatozoa were not found. One of them belonged to the 36 Eng. ft. class and the other to the 37 Eng. ft. class.

There was little room for confusion so far as the spermatozoa were observed distinctly in the section preparats. But, if they failed to be detected, all we could say was that spermatozoa were not present in that part of the testis we examined histologically; they might be present in some other part of the testis.

In the consequence it came to be necessary to investigate not only the presence or absence of spermatozoa in the convoluted seminiferous tubules, but also the conditions of various cells therein. In other words, we had to have informations as to the spermatocytes, the presence or absence of spermatids, the degree of abundance of spermatids if they were present, and the presence or absence of such spermatids that were undergoing the transformation into spermatozoa.

Investigating these aspects, we found that the two individuals, of 39 and 40 Eng. ft., from which we failed to detect any spermatozoa, had sexually developed to a considerable extent and were not to be called immature. That individual of 38 Eng. ft., from only the left of whose testes the spermatozoa were detected, is considered as an abnormal individual, because the weights of its two testes differed from each other so conspicuously.

After all, it is proved by our data that the sperm whales, like many other animals, attain the sexual maturity not simultaenously at a definite body length, but individually over a certain range of body length. And it seem quite natural that they should do so. Since we can take an individual as being sexually mature if we find spermatozoa at least in one of its pair of testes, we have only to consider the body length frequency of those whales whose spermatozoa are not found in either of its testes, so far as we are concerned about the body length at the sexual maturity for practical purposes. Such individuals were not found in the body length classes 38 Eng. ft. and over, as we have described; in other words, 37 Eng. ft. was the upper range of the body length of such whales.

The result of our histological examination is summarized in Table 1 and Figure 4, where the right and left testis of each whale are counted individually, and those testis from which no spematozoa were found are classified into the (minus) or immature group, irrespective of the feature of development of the testis tissue. Being based on these data, we can conclude that the body length of the sperm whales found in the adjacent waters of Japan at the sexual maturity is likely to be 35 to 37 Eng. ft., instead of 38 to 41 Eng. ft. or 42 Eng. ft. as was claimed by Matthews or Matsuura and Maeda (Mattews: 1938, and Matsuura and Maeda:1942).

Then we shall refer to the relation between the sexual maturity and the weight of testis of sperm whales. In 10 of the 90 individuals we examined the weights of the right and left testis were equal; in 35 of them the right testis was heavier than the left; in the other 45, the left was heavier than the right. Therefore, neither of the right and left testis showed any such tendency that one was always heavier than the other.

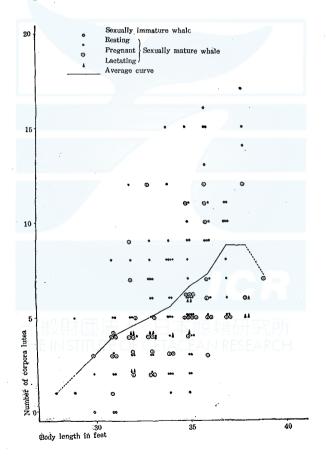
So far as our materials concern, those testis in which no spermatozoa were found always weighed less than 1.6 kg. But it did not follow that all the light testis lacked spermatozoa. We found spermatozoa in a testis that weighed only 0.6 kg. The heaviest testis that we met on the course of this study was the right testis of a male of 53 Eng. ft. in body length, and weighed 6.8 kg.

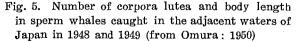
#### Chapter III.

### Sexual Maturity of the Female

As was described in Chapter I, we distinguished mature female sperm whales from immature individuals on basis of the presence of the yellow bodies (corpora lutea) in the ovaries of the mature individuals, as in the case of the baleen whales.

Figure 5, which was cited from Omura (Omura: 1950,), shows the relation between the body length and the number of yellow bodies of the female sperm whales caught in the Japanese waters in 1948 and 1949. In those years the body length limit of 30 Eng. ft. was applied to the castal operations. Three immature individuals (about 2% to





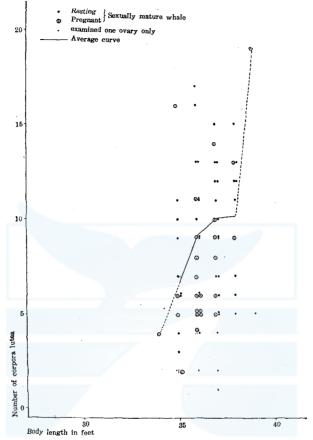


Fig. 6. Number of corpora lutea and body length in sperm whales caught in sampling area in 1950 and examined in this study.

the total) are included in Omura's data quoted here; one of them measured 30 Eng. ft. in body length and the other two 31 Eng. ft.

In 1950 the body length limit was raised to 35 Eng. ft. As the result, a few individuals below 34 Eng. ft. were caught during that year. And one of them were deat with in our study. And no immature individual was caught in 1950 as shown in Figure 6. The numbers of the individuals we examined are shown by body length classes in Figure 7. These figures show that the average number of yellow bodies was 6 to 7 at the body length of 35 Eng. ft. Excluding those whales one of whose pair of ovaries was not examined, eight or 5.6% of the examined whales of the body lengths of 35 Eng. ft. and over were possessed of three or less yellow bodies. The average numbers of yellow

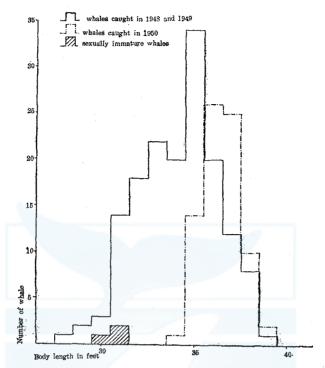


Fig. 7. Length distribution of examined female sperm whales.

bodies at various body lengths are shown by the curves in the figures.

Though our data on the immature or smaller individuals are too short in order to estimate the body length of the female sperm whale at the time of their sexual maturity, we are rather of opinion that Matthew's estimate is almost correct. It is probable that the body length at the sexual maturity of the female is subject to smaller personal errors of estimation than that of the male, because the former can be estimated more easily and with a more distinct result than the latter.

### Chapter IV.

#### Conclusion

Being based on the foregoing evidences, we estimate that the male sperm whales found in the adjacent waters of Japan mature at the body length from 35 to 37 Eng. ft.

As to the body length at the sexual maturity of the female, we are not able to make any definite estimate at present, because the

#### On the Sexual Maturity of the Sperm Whale

data on the immature individuals are short. But we think that the estimates previously made by various authors are almost correct. And all the females of the body length of 35 Eng. ft. and over (the body length limit defined by the existing International Whaling Convention) were considered as being sexually mature. It was only 5.6% of them that were possessed of three or less yellow bodies. In other words, the great majority of the females over 35 Eng. ft. had bred several times.

Our conclusion in the foregoing lines is to be verified by the studies in future. We are not contented with the number of our samples. And we shall have to study how the present conclusion applies to other seasons or to such other grounds as the Bonin waters, the waters around Oshima and off Tohoku and others. These subjects will be discussed in a series of papers that will appear under the same title as this.

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## Appendix

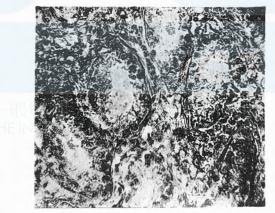
# Microphotographs of the Sections of Testis Tissues

## of the Sperm Whale

## PLATE I.



A. A section of inactive testis tissue. The youngest stage among all the samples examined. (No. N. 375. Body length: 36 Eng. ft.) (× 250)



B. Spermatocytes are shown. This testis is classified in the — (minus) group in Table 1 and Figure 4. (No. N. 327. Body length: 38 Eng. ft.) (× 260).

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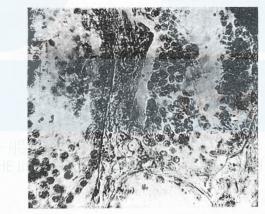








A. Spermatids before undergoing metamorphosis are shown. Well developed spermatozoa are found in other parts of this section than presented here. (No. H. 358. Body length: 46 Eng. ft.) (× 260).



B. Various stages of the metamorphosis of spermatids are shown. (No. H. 276. Body length: 41 Eng. ft.) ( $\times$  260).

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PLATE III.

A. Well developed spermatozoa are shown. (No. K. 83. Body length : 37 Eng. ft.) ( $\times$  200)



B. Spermatozoa. (No. K. 83. Body legth: 37 Eng. ft.) (× 950)





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### Chemical Studies on Freshness of Whale Meat. IV.

# Some Informations of Achromobacter ubiquitum isolated from Whale Carcass

### Βy

### TADASHI NAKAI

(This was reported in Japanese on pp. 152–156, Vol. 71, Journal of the Pharmaceutical Society of Japan (1951))

Prof. Dr. T. Akiba and his co-operators<sup>1)</sup> isolated various kinds of bacteria from sperm whale carcass low in freshness, among which many strains of *Achromobacter ubiquitum* were isolated from any of muscle, blood and intestines. In order to study what part this bacterium played in the decrease of freshness in the whale carcass, I took over a strain of this bacterium from him and investigated the decomposing state of the extract of whale meat by it. Then I searched the acids produced by the decomposition of glucose by this bacterium. Some informations obtained are reported hereon.

### Experiment and Study

### I. Decomposition of the extract of whale meat

Achromobacter ubiquitum (abbreviated to A. u. bacterium) was inoculated on the extract of whale meat and the decomposing process of the extract was traced by determination of volatile basic nitrogen with the lapse of time.

Preparation of the extract of whale meat :—The refrigerated meat of a whale (*Balaenoptera*) was minced, to 500 g of which 1l of physiological salt solution was added and after 3 hours' standing under occasional stirrings, it was heated in the steam-kettle for 1 hour and filtered. 1l of the filtrate was sterilized for 1 hour. (Per 100 cc of the extract, total nitrogen was 237.4 mg, non-protein nitrogen 226 mg and volatile basic nitrogen 10.2 mg.)

To the extract obtained in such a way, 0.1 cc of suspension of one platin loopful of A. u. bacterium for 5 cc of physiological salt solution was added and incubated at  $26 \sim 28^{\circ}$ C.

Determination of volatile basic nitrogen:—It was determined in aeration method. To 5 cc of cultural fluid 0.5 cc of saturated  $K_2CO_3$  solution, 0.1 g of NaF and 5 drops of octyl alcohol were added, where to

T. NAKAI

air washed through dilute sulfuric acid was sent so violently that the volatile base was expelled into 0.02 n H<sub>2</sub>SO<sub>4</sub>. Then the surplus acid was titrated back with 0.02 n NaOH.

The result is shown in Fig. 1. For the first 250 hours, the amount of volatile basic nitrongen (abbreviated to V-N) showed little

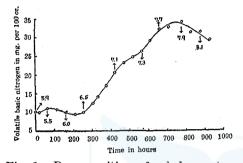


Fig. 1. Decomposition of whale meat extract. Figures marked with \mathcal{mean} the value of pH of the solution at that time. increase. After that, about till the 750th hour, it kept the steady increase and then decreased suddenly. After the decrease from 5.9 to 5.5 in pH, increasing again, it was 6.5 instantly before V-N showed the clear increase. Perhaps the first decrease in pH is due to the acid brought from the small amount of saccharide contained in the extract by the acid productivity of A. u. bacterium. Inspite of no increase

of V-N amount, the value of pH increased in the period about from 60th to 200th hour. This probably shows the formation of non-volatile amine. Clearly the generation of gas was recognized after 24 hours of inoculation. The cultural fluid smelled rather sourish and of its own, not bad.

### II. Comparison of decomposition between suspension and extract of whale meat

The state that whale meat was being eroded by A. u. bacterium was inferred from the decomposing state of whale meat suspension by this bacterium, and at the same time it was compared with that of the extract. The whale meat used was of the same kind as I.

Preparation of whale meat suspension: -50 g of whale meat minced with the meat-chopper was heated in the steam-kettle for 30 minutes and brayed in the mortar. To it, 1l of physiological salt solution was added and after 1 hour's heating in the steam-kettle, its supernatant was put in another vessel. Its residue was fully brayed again and sterilized for 1 hour after dispersing into the supernatant. Total nitrogen per 100 cc was 171.1 mg.

Preparation of the extract: -60 g of the above-mentioned whale meat was heated in the steam-kettle for 30 minutes, and brayed in the mortar. To it, 1.2 *l* of physiological salt solution was added. After

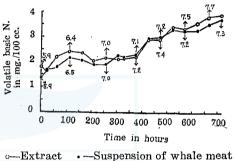
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2 hours' heating in the steam-kettle and filtering, 1050 cc of the filtrate was sterilized for 1 hour. Total nitrogen per 100 cc was 36.3 mg.

5 drops of the above A. u. bacterium suspension were added to thus made suspension and extract respectively, which were kept at  $27^{\circ}$ C. The amount of V-N was measured by the aeration method, with 20 cc of each liquid.

The result is shown in Fig. 2. Fluctuation of V-N amount of the suspension agrees nearly quite with that of the extract. This is, it is not influenced by the existing

amount of muscle protein. This fact can be easily understood from the known fact that A. u. bacterium has no gelatine decomposing power. Therefore, on whale meat eroded by A. u. bacterium, not muscle protein but mainly nonprotein matters will be decomposed. The change of pH in both liquors agreed about till 300th





hour (pH 7.1) and after that with an increase of the amount of V-N, these liquors showed the different value in pH and its difference became larger and larger. And the then value of pH was smaller in the suspension than in the extract. Probably this is due to protein's buffer action.

### III. Effect of the initial value of pH upon volatile base formation

From the above experiments and the general fact that optimum pH for deaminase of bacteria is alkaline, I imagined this bacterium deaminated substrate in neutral and alkaline, but scarcely about pH 6. Therefore, if pH of the extract in the beginning of the experiment was neutral or alkaline, this bacterium would deaminate the substrate at once and the amount of V-N would show a clear increase from the beginning, I imagined. So inoculating A. u. bacterium on the different extracts in pH, I examined the change of the amount of V-N. The whale meat used for the experiment was of the same species as in the above one.

1.6 l of physiological salt solution was added to 800 g of minced whale meat and after 3 hours' standing under occasional stirring, boiled

### T. NAKAI

for 10 minutes and filtered. After steam-sterilizing the filtrate for 1 hour, there was inoculated a platin-loopful of A. u. bacterium and kept standing for one night at 27°C. Thus, fermentable matter (carbohydrate) in the extract was consumed, to preventing as much as possible from decreasing pH instantly after the commencement of the experiment as in experiment I and II. And through the layer of kieselguhr, bacteria were sucked off. Each 480 cc of the filtrate was made at 6, 7 and 8 in pH and after 30 minutes' heating in the steam-kettle, changed pH was adjusted with NaOH and added physiological salt solution made its amount 500 cc. It was put in a 1*l* Erlenmeyer's flask and sterilized for 30 minutes every day for the following three

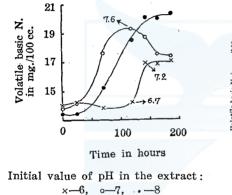
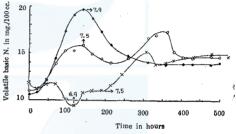
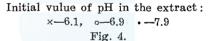


Fig. 3.





days. To each of them, 0.1 cc of the above mentioned suspension of bacteria was added and kept at  $27^{\circ} \sim 28^{\circ}$ C. Often 10 cc of this solution was taken out aseptically to measure the amount of V-N.

The experiment was carried out twice and its result is shown in Figs. 3 and 4. In the extracts of 7, 6.9 and 8, 7.9 in pH at the beginning of the experiment, the amount of V-N increased from the outset but in the extracts of 6, 6.1 in pH it did not increase in the early stage of the experiment with even a decrease until towards 7 in pH. This result seemed to coincide fully to my expectation before the experiment. But in case of peptone

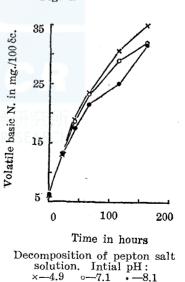


Fig. 5.

solution (1% of Polypepton "Takeda", 0.5% of NaCl) used as cultural medium, in any of 5.9, 7.1 and 8.1 in the initial pH, the amount of V-N increased from the outset and the increase of the amount of V-N at 5.9 in pH was the most till 162nd hour (Fig. 5). Therefore, we can consider that peptone contains some substance that can give the volatile base by A. u. bacterium's action even at 6 in pH but the extract of whale meat does not contain it.

In the extract of whale meat, at a certain stage the amount of V-N decreased in sudden with state of equilibrium in conclusion. In this state the amount of V-N was nearly same, with no relation to pH in the beginning of the experiment (Fig. 4).

### IV. Decomposition of autolyzate of whale meat

In the decreasing process of freshness of whale carcass, prior to or at the same time with the decomposition by bacteria, autolysis of muscle takes place and intermediary products of protein decomposed increase gradually and the amount of the extractive matter also. To find out the relation between it and the A. u. bacterium's action, the extracts of autolyzed and normal whale meat were made and their decomposing state by A. u. bacterium was compared.

The preparation of the extract of normal whale meat (control solution):—To 500 g of minced sperm whale meat, 1l of physiological salt solution was added and after 3 hours' standing, it was boiled for 10 minutes. The filtrate was sterilized for 1 hour and a platin-loopful of A. u. bacterium was inoculated on it and kept at 28°C for a night. Thus, after consuming the acid-fermentable matters, the solution was sucked through the kieselguhr layer and the filtrate was neutralized with NaOH, of which 500 cc was put in a 1l Erlenmeyer's flask and sterilized for 30 minutes every day for the following 3 days. Total nitrogen was 117 mg per 100 cc.

Preparation of the solution of autolyzate:—To 500 g of minced meat of sperm whale, 1l of physiological salt solution saturated with chloroform and 5 g of chloroform were added and kept at  $27^{\circ} \sim 28^{\circ}$ C for 5 days. Then, after 10 minutes' boiling, the filtrate was treated as mentioned above. The total nitrogen was 190.3 mg per 100 cc.

0.1 cc of A. u. bacterium suspension was added to thus obtained two kinds of solutions and kept at  $27^{\circ} \sim 28^{\circ}$ C. The result was shown in Fig. 6.

The amount of V-N was always more in the solution of autolyzate than in the control solution but both solutions showed nearly same

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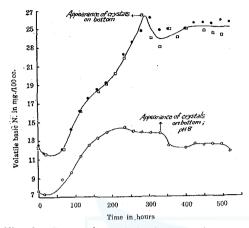


Fig. 6. Comparison of the decomposing process of the extract between autolyzed whale meat and normal whale meat. ., □—Autolyzed, ~—Normal

changing state till about 200th hour. However, after the 200th hour, the control solution showed little increase of the amount of V-N and then gradual decrease, but the solution of autolyzate showed the remarkable increase again. Can't we interprete this phenomenon as follows? In the solution of autolyzate, kinds of substances decomposed by A.u. bacterium were different between till and after about the 200th hour ("The substance decomposed by A. u. bacterium'' is limited to the substance forming the volatile base by the action of A.u. bact-

etrium, in this case). Till about 200th hour, the substance decomposed by A. u. bacterium was of same kind in both solutions and consumed almost all by this time. After this time, the control solution had no substance decomposed by A. u. bacterium, so the amount of V-N did not increase. In the solution of autolyzate, A. u. bacterium found another kind of substance to be newly utilized in the product of autolysis and began to decompose actively and the amount of V-N increased again.

As in the preceding experiments, both solutions showed the sudden decrease of the amount of V-N in a certain stage.

## V. Effect of A. u. bacterium upon the decreasing freshness of whale carcass

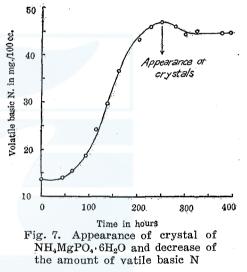
The results of the above experiments can make us imagine as follows:—No A. u. bacterium decomposes protein and gives out the bad smell. So if this bacterium only acts on the whale carcass, it gives no remarkable effect on freshness. However, generally speaking, in the decreasing process of freshness, only one species of bacterium acts rarely, and simultaneously various kinds of bacteria work. So in such a case, A. u. bacterium decomposes actively various intermediary decomposition products of protein which are produced by other bacteria, especially bacteria with protein decomposing power, and gives abundantly the last products of protein decomposed. Namely it can be safely said that A. u. bacterium can not become the main cause of decreasing freshness on whale carcass but plays a part by participating in the last process of protein decomposing.

### VI. The cause why the sudden decrease in the amount of V-N takes place in a certain stage of decomposing process of extract

In most of the above experiments, the sudden decrease of the amount of V-N was found in a certain stage. The pH was always bigger than 7.5. From the further shape of the curve we can judge that this decrease is not caused by volatilization. After my incessant

observation, I found that in this stage fine sand-like or short prismal crystals appeared on the bottom of the vessel without fail and they increased their number with the decrease of the amount of V-N (Fig. 6 and 7). Wondering if this crystal had any relation with the decrease of the amount of V-N, I investigated to find that this crystal contained ammonia as my expectation.

This crystal is an inorganic substance, hardly soluble in water and alkali carbonate solution and readily in dilute mineral



acids. Its dilute mineral acid solution, if it is alkalified with NaOH, gives the gelatinous precipitate and generates ammonia. This crystal was confirmed to be magnesium ammonium phosphate from the fact of containing Mg<sup>••</sup> and  $PO_4^{'''}$  as well as ammonia and the content of phosphorus. The determination of phosphorus was done by Bell-Doisy's<sup>2)</sup> colorimetry with solution of the crystal in 0.01 n HCl as test solution.

Sample :	1.63 mg,	$1.51 \mathrm{mg}$
P:	0.204 mg,	<b>0.1</b> 88 mg
$\mathrm{NH}_{4}\mathrm{MgPO}_{4}{\cdot}6\mathrm{H}_{2}\mathrm{O}$ :	calculated	12.63
	found	12.52, 12.45

Accordingly, the cause for decrease of the amount of V-N was found on the crystallization of magnesium ammonium phosphate. It was certificated by its crystallization even in the extracts in Fig. 1 and Fig. 7 which were made without the treatment through kieselguhr. VII. Acids as the metabolic products of Glucose by A. u. bacterium

A. u. bacterium produces acids from saccharide. Acids produced aerobically from Glucose were searched.

Preparation of cultural medium :—The solution of 1l of whale broth, 10 g of Glucose and 5 g of NaCl were neutralized with NaOH and aftr sterilization, added 5 g of CaCO<sub>3</sub> separately sterilized. A 2l Erlenmeyer flask was used as vessel.

A platin loopful of A. u. bacterium was inoculated on this cultural medium and incubated at  $28^{\circ} \sim 30^{\circ}$ C for 3 days under occasional stirrings. 2l of thus fermented solution was treated as the following table and divided into the volatile acid solution, ether-soluble non-volatile acid solution and ether-insoluble acid solution.

2 l of fermented solu	tion
Precipitate         decomposed with         dilute $H_2SO_4$ Filtrate         extracted         with ether	Filtrate slightly alkalized, concen- trated into about 150 cc on water-bath, decompsed with dilute H <sub>2</sub> SO <sub>4</sub> and centri- fuged. Supernatant
Etheric soluion Ether distilled	Residual solution (Ether-insoluble acid solution)
Residue Steam-distillation	
Distillate (Volatile acid solution)	Residual solution (Non-volatile acid solution)

I tried to isolate acids from the above each solution.

Volatile acid:—To volatile acide solution, PbO was mixed in excess ond evaporated to dryness on the water bath and the residue was infused with about 35 cc of tepid water. The filtrate was heated in the violently boiling water-bath but no basic lead propionate was found. Therefore, after removing lead by adding dilute  $H_2SO_4$  to the filtrate, ZnO was added and evaporated to dryness on the water-bath. The residue was infused with absolute alcohol after two hours' desiccation at 150°C. To the residue, phophoric acid was added and distilled with steam. The distillate showed negative reaction against formic acid. Alcohol distilled off the infusion and after adding phosphoric acid to the residual solution, it was distilled with steam. Though white substance was found in the distillate, its scarcity made the further investigation impossible. Then it was filtered off. The filtrate was neutralized with NaOH and concentrated into about 3 cc on the waterbath and added saturated  $AgNO_3$  solution. Instantly after, white substance appeared, which was filtered out and recrystallized from warm water. About 0.2 g of needle crystal of silver acetate was obtained.

Sample	68.7  mg	$\mathbf{A}\mathbf{g}$	$43.8~\mathrm{mg}$		
CH <sub>3</sub> COO	Ag	calculated	64.64	found	63.75

Non-volatile acid:--Non-volatile acid solution was neutralized with saturated Ba(OH)<sub>2</sub> solution and concentrated on the water bath. 25 gr of the concentrated viscous substance was dissolved into 250 cc of 80% alcohol under warming and after cooling, it was filtered. The residue was inorganic. After distilling alcohol off, the filtrate was diluted with water and decomposed with dilute H<sub>2</sub>SO<sub>4</sub> and filtered. The filtrate was extracted with ether and then it was distilled off. After the volatile substance which somewhat seemed to be contained in the residue was expelled off through steam-distillation, some water and ZnCO<sub>3</sub> in excess were added and heated on the water-bath and filtered. The filtrate was treated with active carbon and concentrated on the water bath. About 10 g of crude zinc lactate was obtained. After recrystallizing from hot water and desiccating at the room temperature, the yield was 5.4 g. Some more crystals were yielded from the mother liquor.

Sample 298.2 mg; 215.5 mg (yielded from the	mother liquor)
Loss on heating at 115°C 53.7 mg. 39.0 mg	
ZnO 81.7 mg; 58.7 mg	
$Zn(C_3H_5O_3)_2 \cdot 3H_2O$ calculated crystal water	18.16
ZnO	27.35
found crystal water 18.01;	18.1
ZnO 27.43;	27.24

According to this analysis value, the lactic acid produced by A. u. bacterium is racemic mixture. Therefore, A. u. Bacterium is imagined to contain racemiase.<sup>3)</sup>

Ether-insoluble acid:—It was searched with newly made fermented solution. It was concentrated on the water-bath and decomposed with dilute  $H_2SO_4$  and centrifuged. The supenatant was extracted with ether for removing the ether-soluble acids and the residual solution was boiled with excess CaCO<sub>3</sub>. In the filtrate and the residue, acid

### T. NAKAI

was searched but no organic acid could be found.

Accordingly, organic acids found as metabolic products of glucose were considerable amount of lactic acid and minute amount of acetic acid.

In conclusion, sincere thanks are expressed to Dr. T. Maruyama for his kind advice and to Prof. Dr. T. Akiba for transfering the precious bacterium and to Mr. H. Okuda for revision.

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### The Effects of Electric Shock and Fatigue on Post-mortem Changes in Muscle\*

#### By

### TADASHI NAKAI and HOKOTO ONO

## (This was reported in Japanese on pp. 358-362, Vol. 71, Journal of the Pharmaceutical Society of Japan (1951).)

This experiment was made with use of albino-rats, as a preliminary test for learning the effects of electrocuting method upon post-mortem freshness of whale carcass, which Japanese whalers are now making trial of. The electrocuting method is a method which gives electrical shock to a whale through a harpoon shot into its body in order to kill it in a short time. When this method is used, there is no longer struggle before death such as seen in the hitherto used method. It is an original purpose to learn the effect of this struggle and electric shock upon the post-mortem freshness of whale carcass. In order to get some key authors made an experiment with use of albino-rats instead of whale body.

They have to thank Dr. T. Maruyama and Prof. Dr. S. Akiya for their kind guidance and Mr. T. Murata for affording facilities in the experiment.

### Method

### 1. Method to give rats fatigue.

Rats were forced to swim for one hour in an enamelled cylindrical tank about 37 cm in diameter and in depth, at about 37°C.

### 2. How to kill them.

To electrocute rats, alternating current of 50 cycles was sent for about one minute into the tank filled with water with copper plates as electrodes. As mechanical shock, rats were heavily struck on their heads with a wooden hammer.

### 3. Sampling.

After a certain hours' storage of five albino-rats carcasses, each about 120 g. in body weight, in an incubator at 37°C, the same quantity of muscle was sampled out of hind leg of each rat for the following observations.

\* Chemical studies on freshness of whale meat. V.

### T. NAKAI and H. ONO

### 4. Measurement of pH.

Sample was brayed with a small amount of sea sand in the mortar and mixed with distilled water four times as much as the sample and its pH was electrically measured with antimony electrode.

## 5. Measurement of the amount of amino-nitrogen and volatile basic nitrogen.

10 g. of sample with some sea sand was well brayed in the mortar and mixed with 30 cc of distilled water. After 15 minutes' heating in the steam-kettle, supernatant was separated by a centrifuge and the residue was washed again and again with warm distilled water, of which the above supernatant was made to 100 cc. 1 cc of phenolphthalein solution, 20 cc of 10 % BaCl<sub>2</sub> and by 5 cc excess of saturated Ba(OH)<sub>2</sub> solution enough to neutralize the mixture were added to 50 cc of the above solution to precipitate phosphate and carbonate, and then through vacuum distillation at 45°C volatile base was collected in 0.02 n-H<sub>2</sub>SO<sub>4</sub>. Excess of the acid was titrated back with 0.02 n-NaOH to determine volatile basic nitrogen. The distilled water was added so as to make the remnants 100 cc. Amino-nitrogen was determined by Sörensen's formol titration with 50 cc of the filtrate.

### 6. Measurement of number of bacteria.

All treatments were aseptically done. With some emery powder the sample was well brayed. Then the physiological salt solution diluted it and the number of bacteria was counted in the ordinary way.

### 7. Lactic acid determination.

The sample was treated by Tanaka and Endo's method<sup>1)</sup> to get a testing solution and lactic acid in it was determined by Friedemann and co-workers' method.<sup>2)</sup>

### Results obtained

### I. Comparative study of post-mortem changes in muscle between normal and fatigued rats electrocuted.

As see in Table I, both groups showed the lowest pH from 12th to 18th hour after death. Normal group showed always more acid than fatigued one.

Although small till 12th hour, the amount of volatile basic nitrogen was suddenly increased after that time. Till 12th hour there was

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Lapse of time		N	ormal*	<i>i</i>	Fatigued *			
after death	р <b>*</b> *	Volatile basic N.	Amino N.	Number of bacteria	p#*	Volatile basic N.	Amino N.	Number of bacteria
(hrs.)		(mg/100 g)	(mg/100 g)	(in 1 g)		(mg/100g)	(mg/100 g)	(in 1 g)
0	6.4	12.6	51.9	0	6.7	12.4	53.0	0
6	5.9	13.0	53.6	0	6.5	13.2	61.8	0
12	5.8	15.9	68.4	170	6.1	16.3	71.9	130
18	5.7	28.9	86.8	560	6.0	21.6	103.8	2720
"	5.7	52.8	114.4	740	6.2	40.0	110.8	4320
24	6.2	107.9	186.2	$166.4 \times 10^{3}$	6.3	132.0	193.4	$27695 \times 10^3$
//	6.4	143.6	245.9	countless	<b>6.</b> 6	114.8	184.0	*** countless

Toble I. Electocution.

Intensity of electric current through the water-tank :---- Normal group-96~ 100 V, 77~79 mA; Fatigued group-96~100 V, about 160 mA (because water in the tank was soiled with excrement). Carcasses were kept at 37°C.

\* Every time 5 rats of each group were used for measuring.

\*\* About an hour passed from their dissection to measurment.

\*\*\* It was macroscopically obvious that there were more bacteria in the fatigued group than in the normal group, though impossible to account for them.

little difference between two groups, while at 18th hour, normal group showed larger amount of volatile basic nitrogen than fatigued one as seen in two instances. At 24th hour, there was no clear trend.

As for amino-nitrogen, till 12th hour a little larger amount of it was found in fatigued group and then it became irregular.

Bacteria were remarkably numerous in number in fatigued group.

No special feature was found by macroscopic observation of rat carcass till 6th hour. At 12th hour rather with offensive smell, gas generated in its intestine and swelled the abdomen. The difference between two groups was difficult to find. At 18th hour, both groups gave out a putrid smell and their muscle increased its viscosity and degree of grey colour and lost its elasticity. In both instances, however, fatigued group was seemingly better than normal group and kept still slight reddish colour and exudation on the side of carcass on the floor was never found or a little less than the normal group. At 24th hour, in the first experiment, the apparent difference between two groups was difficult to find. While, in the second one the fatigued group was distinctly better.

In short, in comparison between two groups on post-mortem changes, the results of comparison of macroscopic state of putrefaction

agreed quite well with that of the amount of volatile basic nitrogen but not with that of the number of bacteria. From pH and the number of bacteria, it was naturally expected that macroscopic putridity appeared stronger in fatigued group than normal one but the truth was not so. It is, however, too premature to draw the conclusion, for only a few experiments were made.

Rigor mortis observed in all experiments will be mentioned later.

## II. Comparative study between normal and fatigued rats killed by mechanical shock.

Lapse of time		Normal*		Fatigued *			
after death (hrs.)	pH N. (mg/100 g)		Amino N. (mg/100 g) pH **		Volatile basic N. (mg/100 g)	Amino N. (mg/100 g)	
0	6.9	13.8	57.2	6.8	12.3	60.1	
12	5.9	19.6	80.2	6.1	19.4	82.5	
18	6.0	75.0	156.7	6.5	34.8	138.5	
IJ I	6.2	110.0	166.0	6.8	132.8	188.5	

#### Table II. Ceating to death.

\* Same as in Table I. \*\*  $30 \sim 40$  minutes passed from their dissection to measurment. Carcasses were kept at  $37^{\circ}$ C.

As for pH, in both groups, at 18th hour already the turn towards alkalinity was found. Initially in the normal group, it was nearer neutral than the fatigued group and then it changed to more acid side. The increasing ratio of pH was rather smaller in the normal group than the fatigued group. This was probably due to the fact that the normal group contained larger amount of glycogen, so lactic acid was made more abundant in post-mortem glycolysis than the fatigued one. The amounts of volatile basic nitrogen and aminonitrogen suddenly increased at 18th hour as seen in Exp. I. No definite difference could be, however, found between these two groups. As stated in I, comparison of the amount of volatile basic nitrogen agreed always with that of degree of rottenness in macroscopic observation.

## III. Comparison between electric and mechanical shocks as method to kill normal rats.

As seen in Table III, the initial pH was in far more acid side in electrically shocked group than in mechanically shocked one. At 12th hour and 18th hour, their pH were approximately equal.

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Lapse of time		Electrocutio		Beating to death *			
after death (hrs.)	pH **	Volatile basic N. (mg/100 g)	Amino N. (mg/100 g)	рН <sup>**</sup>	Volatile basic N. (mg/100 g)	Amino N. (mg/100 g)	
0	6.3	12.3	54.9	6.9	12.3	57.2	
12	5.9	19.5	76.1	5.9	24.3	81.9	
18	6.0	55.1	123.3	6.1	134.1	238.2	

Table III.

\* and \*\* same as in Table II. Carcasses were kept at 37°C.

The volatile basic nitrogen and amino-nitrogen increased their amounts faster in mechanically shocked group than in electrocuted group. The comparative results between I and II can make it clear. This agreed with the result of macroscopic observation. Namely the mechanically shocked group was decomposed earlier than the electrocuted one.

This is probably due to heavy bruise on the head and to the change of pH in muscle to acid side through electric shock (Confer IV).

### IV. pH change of muscle through electric shock.

### (a) Comparison between normal and fatigued conditions.

In the above experiments, normal rats electrocuted gave smaller pH in muscle than fatigued, electrocuted rats and normal, mechanically shocked ones. It is in general, however, that fatigued muscle showes more acid than not fatigued one. So the above result was probably due to the electric shock. This experiment was carried on for the purpose of assurance of it.

In order to measure pH as soon as possible after death, the sample, without braying, was closely attached to the electrodes in the shape of chop. About 5 minutes passed from death to measurement. Sample was obtained from the same part of hind leg as in the previous experiments.

Forced to swim in the above mentioned tank, rats were thrown into a small tank full of clean water and charged with alternating current. They were fatigued rats. Normal rats were directly thrown into the small tank. The current intensity was, therefore, different from that of the above experiments.

Probably due to the chop used, sensibility of the pH-meter was so bad and unconstant that it was difficult to measure pH with it. It could be affirmed, however, that electric shock changed pH in muscle

Norr	Normal, Electrocuted			Fatigued, Electrocuted			Normal, Beaten		
Sex	Weight (g)	pH	Sex	Weight (g)	pH	Sex	Weight (g)	$_{\rm pH}$	
m	160	6.4	m	175	6.9	f	120	7.1	
m	185	6.2	m	215	7.0	f	220	6.9	
$\mathbf{f}$	90	6.6	f	90	7.0	f	80	7.5	
f	75	6.7	f	75	7.5	f	80	7.4	
f	75	6.7	f	70	7.5	f	80	7.3	
m	120	6.7	m	195	6.9	f	80	6.8	

Table	IV.	

to acid side and normal rats showed smaller value in pH than fatigued one.

(b) Effect of electric current upon pH of muscle cut off.

After the normal rats were killed by mechanical shock, muscle of one side of their hind legs was cut off and it was hung in the water charged with the alternating current of 95 V and 45 mA for 30 or 60 minutes. Then brayed swiftly and fully with some quantity of sand. Distilled water four times as much as the sample was added to it and its pH was measured. For comparison, it was made with the use of the same part of muscle of another leg with no electric current flown. This result showed that the electric current gave no effect upon pH of muscle which was cut off carcass. So the change in pH to acid side by electric shock might take place only in the living body.

Lapse of time after death (hrs.)	Electrifying time (seconds)	pН	pH (control)
	FOF 30 ACEAN	7.1	7.3
0	60	7.0	7.0
0	60	7.0	6.8
1	60	6.4	6.4
2	60	6.1	6.1

Table V.

Carcasses were kept at 37°.

(c) Any constant relation between the current intensity, sex, body weight and pH change was not found.

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### (d) Cause for pH change.

As the cause for pH change in electrically shocking, the authors first expected lactic acid and measured the amount of lactic acid of male rats normal and fatigued which were killed by electric or mechanical shock, instantly after death.

		Electrocution			g to death
	Weight (g)	$\mathbf{p}\mathbf{H}$	Lactic acid (mg/100 g)	Weight (g)	Lactic acid (mg/100 g)
Normal	$\left.\begin{array}{c}175\\200\end{array}\right\}$	6.2	84.5	$\left. \begin{smallmatrix} 190\\210 \end{smallmatrix} \right\}$	52.1
	110 110 }	6.3	125.5		
Tatigued	$\left.\begin{array}{c}175\\200\end{array}\right\}$	6.5	77.8	$\left. \begin{smallmatrix} 190\\ 210 \end{smallmatrix} \right\}$	75.9
guou	$\left.\begin{array}{c}125\\135\end{array}\right\}$	6.9	95.0		

Table VI.

As shown in the above table, the amount of lactic acid ranks as follows.

normal rats electrically shocked > fatigued rats electrically shocked > fatigued rats mechanically shocked > normal rats mechanically shocked

Consequently the cause for pH change is nearly sure to lie in the growth of lactic acid.

Electric stimulus inspires the muscle and is followed by fatigue. So the increase of the amount of lactic acid in muscle is naturally conjectured. Rats are gradually fatigued through swimming and muscle glycogen is consumed and lactic acid accumulates. Grown lactic acid is however not all accumulated but a part of it is gradually excreted even in the process of fatigue. Now, when electric impulse stimulates very intensely the muscle of normal and fatigued rats, the former rats produce the considerable amount of lactic acid in a very short time and accumulate all of it without excreting. We can be ready to imagine that the amount is larger than the total amount of lactic acid accumulated before the electric shock and lactic acid newly produced through it in the latter. So pH is to be smaller in the former than in the latter. This is the authors' interpretation for the phenomenon that pH of the muscle of normal rats is smaller than that of the fatigued rats in electrically shocking.

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### V. Rigor mortis.

*Rigor mortis* observed in the above experiments is summerized as follows. Carcasses were kept at 37°C.

	Electrocution		Beating to death	
	Normal	Fatigued	Normal	Fatigued
Appearance (hrs. after death)	1~1.5	0.5~1	1.5~2	1~2
Beginning of Loose (hrs. after death)	4~4.5	3.5~4	3.5~4	

Table VII.

*Rigor mortis* began earlier and stronger and later got loose in electrically shocked rats than in the mechanically shocked ones.

*Rigor mortis* began earlier and got loose earlier in fatigued rats than in the normal ones.

### Résumé

1. Either through electric shock or through mechanical shock the condition of rats instantly before death (normal or fatigued) did not give any definite effect on the post-mortem change of the amounts of volatile basic nitrogen and amino-nitrogen. The increasing speed of them is however larger in rats mechanically shocked than in the rats electrocuted. Namely the former rotted faster than the latter. From this result only, putrefaction seems to depend more on the killing method than on the condition instantly before death.

2. Number of bacteria on the electrocuted rats was always larger in fatigued group than in the normal group. The result of the comparison of number of bacteria, however, did not agree with that of macroscopic observation. On the contrary, the result of the comparison of the amount of volatile basic nitrogen agreed quite well with that of macroscopic observation.

3. In all experiments pH changed swiftly to acid side immediately after death. When rats were killed by mechanical shock, it began to turn towards alkalinity around 12th hour after death and when electrocuted, it began at 12th to 18th hour. About that time rot increased its degree. pH moved in more alkaline side in the fatigued group than the normal group.

4. The electric shock changed pH of muscle to acid side. In this case, pH of muscle of normal group was smaller than that of fatigued

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group. This is probably due to the sudden rise of the amount of lactic acid caused by sudden and intense stimulus by electric shock.

5. Rigor mortis lasted longer and stronger in the electrocuted group than in the mechanically shocked group.

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### Complete Recovery of Vitamin A from Molecular Distillation Residue of Whale-liver Oil

### By

### YOSHIMORI OMOTE

When the whale-liver oil is treated with molecular distillation, more than 100% of vitamin A (to be abbreviated A hereafter) can be recovered in a distilled concentrate, as the thermal decomposition of kitol produces A during distillation. The residual oil has an absorption maximum at 290 m $\mu$  and gives a bluish violet color with antimony tricholride. The absorption maximum which is identical with that of kitol indicates the presence of kitol, while the color reaction is considered due to the blue color by A and red by kitol. Now it is probable that the thoroughgoing molecular distillation can recover not only the remaining A in the residue, but also the newly produced A from kitol. The presumption is discussed with several fundamental experiments and the pratical thoroughgoing distillation, with the result of which this paper is concerned.

Analysis of Absorption Spectrum of Residue Oil.-Table I shows an example of absorption spectrum of molecular distillation residue of

whale-liver oil. For the sake of convenience, Oser's correction<sup>1)</sup> was directly applied to Table I, though the method strictly must be used upon the unsaponifiable fraction. The corrected coefficient,  $E (1\%, 1 \text{ cm}, 325 \text{ m}\mu)$  was much lower than that of the original. The authors' so-called AK method, which is grounded on the assump-

Table	I.	Absorption		Spectrum	
	of	the	Residue	Oil	

wave length,	Absorption coefficient,
(mµ)	E (1%, 1 cm.)
280	49.65
290	52.05
310	48.1
325	37.75
328	35.1
330	33.55
334	29.95

## Table II. Comparison of Corrections about the Absorption Spectrum of Residue

Method	E (1%,	1 cm)
Non-corrected	E (325) 37.75	E (328) 35.1
Oser's	E (325) 6.8	
AK	-	E (328) 22.5

tion that the absorption spectrum of sample is mainly due to A and kitol, was also applied (see Table II). Both corrected values showed

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a considerable difference, the true A value probably being between them, and their mean value of ca. 15 for E  $(328 \text{ m}\mu)$  was taken as that of A. Kitol content was calculated with AK method to be nearly five times as much weight as A.

Distillation Time and Recovery of A.—The residual oil was distilled in a semi-micro molecular pot still at a constant temperature of 200°, the relation between distillation temperature and A recovery being studied (Table III). As Table III shows that one hour heating was able to distil A, but not enough to decompose kitol, heating time was extended (Table IV).

## Table III.Distillation Time and Recovery of A(in case of short time)

Time (min.)	A Unit*	Time, total (min.)	Unit, total
02	17.9	2	17.9
2-5	17.9	5	35.8
5-10	11.2	10	47.0
10 - 15	8.97	15	56.0
15 - 20	3.58	20	59.6
20 - 25	3.58	25	63.2
25 - 55	7.16	55	70.4

\* Cod Liver Oil Unit, cf. Experiment 2.

### Table IV. Distillation Time and Recovery of A (in case of long time)

Time (hr.)	A Unit	Time, total (hr.)	A Unit, total
0.0-0.5	71.7	0.5	71.7
0.5 - 1.0	8.96	1.0	80.7
1.0 - 1.5	7.17	1.5	87.9
1.5 - 2.0	2.39	2.0	90.3

By making distillation curves based on data from Table III and IV one can recognize the discontinuous point near 10 minutes' heating, where A is almost recovered and kitol begins to decompose to form new A. The recovery of A, however, was about 100% after 30 mins., and 130% after 2 hrs., so the decomposition rate of kitol seemed very small at a temperature 200°.

Distillation Temperature and Recovery of A.-Using the same apparatus as above the recovery of A at discontinuously elevated temperature was measured (Table V).

As shown in Table V, the recovery of A was about 60 units at 200°, about 220 units at 270°, so the difference of 160 units may be considered to represent newly produced A from kitol pyrolysis. This

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		-		
Temperature °C*	Time (min.)	Recovered A**	Time, total (hrs.)	Recovered A total
200	30	57.8	0.5	57.8
220	30	76.1	1.0	133.9
250	40	71.6	1.67	205.5
270	40	10.8	2.33	216.3
* Cf. ex	periment 4 **	• Cod Liver Oil	Unit for the so	ource oil.

Table V. Relation between Distillation Temperatureand Recovery of A

corresponds to about 2.7 times as much as first contained A. If it is assumed that kitol content in the sample was about five times as much as A and one mole of kitol can produce two moles of  $A^{2}$ , five times as much A as first A must be newly obtained theoretically. On the other hand, 2.5 times of A must be produced under the assumption of one mole A being obtainable from one mole kitol<sup>30</sup>. Considering the destructions of A during distillation the result of above experiments seems accorded with theoretical value of 5 times, nevertheless it is close to 2.5 times in case of neglecting the destruction of A.

Thoroughgoing Molecular Distillation in Falling-film Still.-Being clarified fundamental conditions about the complete distillation in a molecular pot still, two hundred grams of the sample was distilled in a falling-film still. The' sample was somewhat different from that used by the foregoing fundamental experiments, so that assays were conducted again in detail. The result of the distillation was that about two times as much as original A was obtained (Table VI).

	Temperature,	Weight,	7	vitamin A U	nit (U.S.P.)*	
	_₀C	g	Oshima's	Whole-oil	Oser's	AK
Original		(200)	30,400	31,200	15,800	26,000
Fr. 1	190 - 205	90	185,000	124,000	93,700	121,800
Fr. 2	205 - 215	40	46,800	50,000	-41,200	49,200
Residual	<b>.</b>	70		8,500	133	5,500
	· ·	Rec	covery of V	itamin A		•
	Oshima's	Wh	ole-oil	Oser's	AK	
Original	(100)	. (1	00)	(100)	(100)	
Fr. 1	266	1	.80	268	211	
Fr. 2	30.8		32.0	52	37.8	$(1,\ldots,k_{n-1})$
Residual			9.5	0.3	7.4	

## Table VI. Recovery of A from Residual Oil with Complete Molecular Distillation in Falling-film Still

\* Conversion factor, Oshima's Cod Liver Oil Unit was multiplied by 380, others by 1900.

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Further discussions may be described below. Contents of A and kitol in the original residue was calculated with AK method, A being ca. 0.76% from  $(328 \text{ m}\mu)=13.7$ , kitol ca. 1.5% from K  $(290 \text{ m}\mu)=10.6$ . Assuming that kitol changes into A completely, and one mole kitol forms two moles A, the content of A must be about three times as much as that of original A, the result of the experiment corresponding with the calculation. Furthermore, the calculation on this experiment led to the conclusion that one mole kitol produced 1.69 moles A considering the distillate only, and 1.53 moles A considering both the distillate and residue.

### **Experimental Part**

1. Analysis of Absorption Spectrum Curve.—Oser's correction<sup>1)</sup> was made through the following revised equation, employing data from Table I.

f = 7 - 2.625 E (310/325) - 4.375 E (334/325)

Also AK method was adopted as stated in the previous paper,<sup>2)</sup> A(328)=22.5 and K(290)=43.4 being obtained.

2. Distillation Time and Recovery of A (Short Time).—The semimicro molecular still was of great value, in which 279 mg. of the sample was taken. The distillation was effected under the vacuum of  $10^{-3}$  to  $10^{-4}$  mm. and a constant oil bath temperature of 200°. As the distillate was cooled, little or no A was destroyed. At first, after two minutes' heating, the distillation was stopped and the condenser was taken out to be washed in an aliquot of benzene, which, after suitable dilution with chloroform was treated with Oshima's colorimetry to determine the blue value. Cod liver oil unit was tentatively calculated using 279 mg as the weight of oil. Similarly the process was carried on as follows (see Table II). Cod liver oil unit of the original sample was found to range 80 to 90, though the violet color prevented exact colorimetry.

3. Distillation Time and Recovery of A (Long Time).—The same conditions as Experiment 2 were adopted except for the range of heating time extended (Table III). The final residual of this experiment gave a reddish color with antimony trichloride, while the original residual oil showed bluish violet.

4. Distillation Temperature and Recovery of A.-Also the same apparatus was appreciated, but the temperature was raised step by

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step (cf. Table V). In spite of its strong fluorescence, the yellow distillate at 270° had a low potency of A. And the gray black final residue gave a reddish brown color with antimony trichloride.

5. Complete Distillation with Falling-film Still.—The sample was the residual oil obtained from the Antarctic whale-liver oil, distilled in the molecular still to concentrate A in a distillate, about 30% of the total oil being distilled (The sample used in Experiment 1 to 4 was about the same as this). The still was falling-film cyclic type, the temperature was of falling liquid (Table VI). The process of distillation was not so smooth because of occasional sudden bubbling on the distilling surface.

6. Calculation of A Formation Ratio from Kitol.-Mole of A formed from one mole of kitol is expressed as follows:<sup>2)</sup>

A mole =  $[A(328 \text{ m}\mu) \text{ increased}/\text{K} (290 \text{ m}\mu) \text{ decreased}] \times 0.786.$ 

A (328) and K (290) were calculated by AK from the absorption spectrum data of each fraction, the result of which already appeared in the preceding explanation.

Grateful acknowledgment is expressed to Dr. S. Ishikawa for his advice and encouragement during the course of this research.

#### Summary

Molecular distillation residue of whale-liver oil, containing vitamin A and kitol of about 1% and 5% respectively, was distilled in a semimicro molecular pot still. At a constant temperature of 200°, about 100% of A was recovered through thirty minutes' heating, yet only 130% of A by two hours' heating; while, by raising the temperature up to 270°, approximately 400% of A could be recovered.

Furthermore, the practical thoroughgoing distillation showed that kitol in the residue was completely converted into A, the absorption curve analysis proving that one mole kitol produced two moles A.

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### Chemical Structure of Kitol (I) Double Bonds and Hydroxyl Groups

#### By

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In 1943 Embree and Shantz<sup>1)</sup> postulated the molecular formula  $C_{40}H_{58}(OH)_2$  for kitol isolated from whale liver oil, and in 1947 Baxter and co-workers<sup>2)</sup> succeeded to obtain kitol in crystalline form, but since that time no research about the chemical structure of kitol has been The author<sup>3)</sup> studied on thermal decomposition of kitol with reported. absorption spectra, yet definite conclusion has not been obtained. The decomposition of kitol has smoothly occurred only in case of molecular distillation, different from the pyrolyses of common substances. This interest reaction must be connected with the chemical structure of kitol, so that the author planned to confirm its structure and the study is now going on. This paper is concerned with results from preparatory experiments on double bonds, especially conjugated double bonds, and hydroxyl groups in kitol molecule.

Hydrogenation of Kitol Concentrate.-Through the hydrogenation of kitol concentrate having  $E(286 \text{ m}\mu)$  of 580, Embree and Shantz determined eight double bonds in a molecule. The author tried to

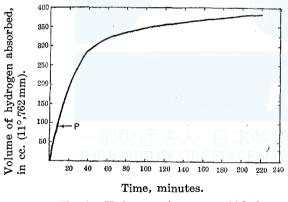


Fig. 1. Hydrogenation curve of kitol. P: end point of the reduction of platinum oxide catalyst. purify kitol with the method similar to that of their report, obtaining the concentrate having  $E(290 \text{ m}\mu)$  of 482, which was used in the study to trace the hydrogeneration curve. while the number of double bonds was not confirmed.  $\mathbf{As}$ shown in Fig. 1, it was remarkable that hydrogen absorption proceeded rapidly during the former two third period, but

very slowly during the remaining one third period.

Conjugated Double Bonds.—The absorption maximum of kitol was reported 290 m $\mu$  (no solvent being mentioned) by Baxter etc.<sup>2)</sup>, 286 m $\mu$ 

(in ethanol) by Embree etc.<sup>1</sup>, while the author obtained 290 m $\mu$  (in isopropanol) in previous paper<sup>30</sup>, and 284 m $\mu$  (in isopropanol) in this report. These differences seem to be due to steric isomerization of kitol, besides to its change during purification.

From the absorption spectrum, Barua and Morton<sup>5)</sup> proposed four

conjugated double bonds in kitol. considering five ones in vitamin A. The author has independently had the same opinion as theirs for the last two years. The data of absorption spectra about hydrocarbons<sup>6)</sup> and alcohols<sup>7)</sup> with a conjugated system shows that the addition of each new -CH =CH- group causes the absorption bands to shift toward the longer wave length region by some 40  $m\mu$  in the region of  $250 m\mu$  to 350 mµ.

Therefore, kitol having an absorption maximum at 285-290 m $\mu$  must have one less conjugated double bonds, which furthermore situated in the chain bond, not in the ring one, than vitamin A having a maximum at 325-8 m $\mu$ .

It was, moreover, proved through absorption spectra that conjugated double bond in kitol reacted with maleic anhydride to decrease its extinction as shown in Fig. 2. As clarified in the figure about 30% of kitol reacted with maleic anhydride in toluene at 100° for thirty minutes.

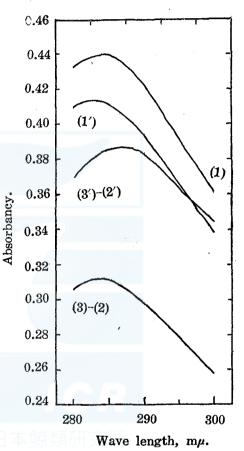


Fig. 2. Reaction of kitol with maleic anhydride, spectrophotometric curve of: (1) kitol, (1') kitol after heating, (3)-(2) kitol immediately after mixing with maleic anhydride, and (3')-(2') kitol after heating with maleic anhydride solution.

Position of Hydroxyl Groups in Perhydrokitol.—Hydrogenated kitol, which will be called "perhydrokitol" was obtained as white solid through hydrogenation of kitol. The carbon hydrogen analyses of perhydrokitol concentrate about agreed with the formula  $C_{40}H_{76}O_2$ , this

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sample being used in the following experiments. Though kitol itself is unstable, perhydrokitol, being a saturated compound, seemed to be stable. As the position of hydroxyl groups in perhydrokitol is same as that of groups in kitol, one can clarify the position of hydroxyl groups in kitol based on the experiments with perhydrokitol.

From this standpoint perhydrokitol was examined by Murahashi's method<sup>8)</sup> and was proved to be primary alcohol: 44-69% of the hydroxyl group in perhydrokitol were esterified after heating at 155° for one hour with phenylacetic acid, under the same conditions 39-59% in case of cetyl alcohol (primary), and in case of cholesterol (ring and secondary) 16-34%.

In order to ascertain the fact the esterification rate with phthalic anhydride was measured. The percentage of esterified hydroxyl group after heating for one hour to that after heating for thirty hours was observed with perhydrokitol, cetyl alcohol and cholesterol, 25.4%, 29.5% and 0.0% being respectively obtained, so that perhydrokitol was proved to be primary alcohol. Thus, the hydroxyl groups in perhydrokitol, accordingly in kitol, was found to be primary, just same as that of vitamin A, from which the author may presume that two moles of vitamin A polymerize into kitol without any change in the position of hydroxyl groups.

### **Experimental Part**

1. Kitol Concentrate.—The unsaponifiable matter from Antarctic whale liver oil was distilled in a molecular still to distil out vitamin A, the remaining portion<sup>4</sup>) was chromatographed four times on alumina in petroleum ether to produce kitol concentrate, which was yellowish white solid having an absorption maximum at  $284 \text{ m}\mu$ , E ( $284 \text{ m}\mu$ ) of 489 and E ( $290 \text{ m}\mu$ ) of 482. Compared with a crystalline kitol having E ( $290 \text{ m}\mu$ ) of 707, the concentrate contained only 68% of pure kitol, yet appeared to be chromatographically pure. It melted at  $87-90^{\circ}$ , giving bloodish red color with antimony triohloride reagent, deep red with trichloracetic acid, and red then yellow with concentrated sulfuric acid and acetic anhydride reagent.

2. Hydrogenation of Kitol.—1.0387 g. of above concentrate was dissolved in 20 cc. glacial acetic acid, being hydrogenated with 0.500 g. of platinum oxide catalyst to produce the hydrogenation curves shown in Fig. 1. The total volume of absorbed hydrogen was 277 cc., so the 271 cc. (at normal state) of hydrogen for one gram of the sample. Assuming the molecular formula  $C_{40}H_{60}O_2$  and eight double bonds one gram of kitol must consume 313 cc. of hydrogen, then the found volume absorbed corresponds 87% of the calculated volume.

3. Reaction of Kitol with Maleic Anhydride.—Three samples were prepared: (A) kitol solution (80 mg. kitol per 100 cc. toluene), (B) maleic anhydride solution (5 g. maleic anhydride per 100 cc. toluene), and (C) an equivolume mixture of (A) and (B). (A), (B) and (C) were heated for thirty minutes at 100°. Before and after heating,

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exactly 1 cc. of the sample solution was pipetted, diluted with isopropanol into 50 cc., with which the absorption spectrum was measured. Correcting the absorption by toluene, and adjusting the dilution same, the absorbancy of each sample was calculated and summarized in Table I.

Table	I.
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#### **Reaction of Kitol with Maleic Anhydride**

mμ	(1)	(2)	(3)	(1)'	(2)'	(3)'
280	0.4325	0.2715	0.640	0.410	0.270	0.576
284	0.440	0.231	0.615	0.412	0.237	0.549
290	0.423	0.1733	0.5565	0.394	0.191	0.4895
300	0.361	0.1113	0.4555	0.339	0.138	0.3965

Absorbancy with same dilution, of (1) kitol solution (A), (2) maleic anhydride solution (B), and (3) equivolume mixture (C): and of (1)', (2)' and (3)', corresponding to (1), (2) and (3) after heating for thirty minutes at  $100^{\circ}$ .

From Table I, (3)-(2) represents the absorption only by kitol immediately after mixing with maleic anhydride in the solvent, while (3)'-(2)' means that of kitol after heating in maleic anhydride solution. These values are plotted in Fig. 2, (above shown).

4. Perhydrokitol.—The colorless transparent solution of hydrogenated kitol was filtered to separate from catalyst, added with excess volume of water, and extracted with ether. Ether solution was evaporated, the residue being saponified with methanolic potash. The nonsaponifiable matter was extracted with ether. After evaporating ether in vacuo the remaining portion became white solid which could not easily be crystallized. The elementary analyses of the compound were as follows:

		Found	Required for
	(1)	(2)	$C_{40}H_{76}O_2$
С	81.11	80.78	81.54
$\mathbf{H}$	12.50	12.75	13.02
O (diff.)	6.40	6.47	5.44

Molecular weight was determined by cryoscopic (camphor) method :

Sample,	Camphor,	Temperature	Molecular
mg.	mg.	difference	weight
10.02	121.17	ACEA 9.3 SEARC	355
5.48	104.32	4.5	469
	Ree	quired for C <sub>40</sub> H <sub>76</sub> O <sub>2</sub>	589

5. Determination of Hydroxyl Group in Perhydrokitol.—Customary methods <sup>9) 10)</sup> to determine OH groups were semimicronized as follows.

Reagent: 2g. of phthalic anhydride (sublimed) to 12cc. pyridine. Standard aqueous potassium hydroxide solution, 0.01026n (to benzoic acid).

Method: A weighed sample in a small thin glass test-tube is added with exact volume (0.210 cc.) of the reagent. Sealing the test-tube, it is kept at  $100^{\circ}$  for seventy-five minutes. After the reaction the sealed tube was broken in the flask, 1cc, water

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added, then a few drops of phenolphthalein indicator dropped into, and the flask contents are titrated with standard alkali solution. A blank is run in the same manner as the sample, and the difference of alkali cc. used for the sample and that for a blank shows the alkali cc. corresponding to the hydroxyl group.

The data with perhydrokitol, cetyl alcohol and cholesterol are summarized in the following. In the table, corr. OH% means found OH% minus quantity of free acid in the sample, expressed in OH%. Free acids in perhydaokitol, cetyl alcohol and cholesterol were respectively found to be 0.13, 0.09 and 0.42 OH%.

	Sample,	Alkali,	OH%	corr. OH%	theor. OH%
	mg.	cc.			•
Perhydro-	22,48	11.60	8.99	8.86	5.76
kitol	13.30	7.82	10.25	10.12	5.76
Cetyl	19.98	12.30	10.72	10.6	6.95
alcohol	12.91	9.80	13.23	13.1	6.95
Chole-	29.90	7.70	4.49	4.07	4.40
sterol	23.51	8.30	6.15	5.73	4.40
	OH% = (0.0)	$1026 \times 17.0 \times c$	$c./mg.) \times 10$	0%	

6. Rate of Esterification of Perhydrokitol with Phenylacetic Acid. — Murahashi's method <sup>8)</sup> was used, except that 10 cc. methanol plus 1 cc. chloroform were added before titration in order to dissolve the solid which was insoluble in aqueous alkali solution. A blank, of course, was carried with the solvent. Data with cetyl alcohol and cholesterol are also quoted for a comparison. A standard was 0.01025n. KOH aq. solution. A.G. means % of acid esterified after one hour at 155°.

	Sample,	Phenylacetic	Alkali,	A.G.
	mg.	acid, mg.	CC.	
Perhydro-	9.04	6.38	3.27	43.7
kitol	10.73	6.07	1.91	68.9
Cetyl	14.82	6.89	2.62	39.0
alcohol	12.41	7.00	2.10	58.8
Chole-	12.09	6.94	4.50	15.8
sterol	12.36	7.09	4.04	33.6

7. Rate of Esterification of Perhydrokitol with Phthalic Anhydride.—After so many preparatory experiments on cetyl alcohol and cholesterol, the author found the following method to discriminate primary alcohol from secondary one: almost same method as the determination of hydroxyl group in experiment 5, except the reaction temperature kept at  $106^{\circ}$  for one and thirty hours.

	ŝ	Sample, A	Alkali, O cc.	H (corr.) %
	hydrokitol	10.60	1.00 2.10	$1.45 \\ 2.61$
	tyl alcohol olesterol	$13.00 \\ 15.30$	0.07	0.0
	chydrokitol tyl alcohol	12.00 15.50	4.00 7.90	5.72 8.85
$\mathbf{Ch}$	olesterol	16.70	3.67	3.44

From above data the percentage of OH% after 1 hr. to OH% after 30 hrs. was calculated as follows: perhydrokitol 25.4, cetyl alcohol 25.5, cholesterol 0.0.

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### Summary

From the expriments on the hydrogenation curve and the addition with maleic anhydride of chromatographically pure kitol, the author presumed four conjugated double bonds containing one in the ring and proved that they reacted with maleic anhydride.

Esterification of perhydrokitol prepared from kitol through hydrogenation, when treated with phenylacetic acid at 155° for one hour, showed that perhydrokitol is a primary alcohol. The fact was also confirmed by the method to measure the esterification rate of an alcohol with phthalic anhydride in pyridine at 100°.

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### Experimental Investigation on Flattened Head Harpoon An Attempt for Restraining Ricochet

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### 1. Introduction

It is well known that the harpoon with the usual pointed head ricochets when it strikes the water surface aslant, jumping over the body of whale even when the point of incidence falls within a few feets from the aim. The present writer also witnessed in several occasions such wastefull missings of aims in the course of an experimental test of electrocution of whales, carried out by the whaling boat, Taiheimaru No. 1 in the vicinity of Hokkaido between the period of August to October, 1949, under the auspices of the Committee for the Improvement of Equipments of Whaling Vessels. It was considered very desirable to improve the harpoon so as to prevent such ricochet and to hit the whale even if the harpoon passed some distance through the water. A brief account of the experimental investigations made in connection with this problem will be reported in the following lines.

### 2. Mechanism of ricochet and its counter-measure

The phenomenon of ricochet is not characteristic one of harpoon, but also holds for any high speed rigid body impinging obliquely against the surface when its angle of incidence is smaller than some critical value which is determined by both the shape and the impacting velocity of the body. E. de Jonquiére<sup>1)</sup> showed that when a spherical body, initially immersed in water, was shot in the direction parallel to the free surface, its path turned upwards gradually and at last emerged out of the surface. As for the pressure distribution in front of the sphere, the point of maximum pressure was slightly shifted to the side opposit to the free surface, thus exerting some driving force towards the free surface. C. Ramsauer<sup>2)</sup> also studied the path of a sphere shot into the water with small incident angle. It followed

<sup>1)</sup> E. de Jonquiére, C. R. 97 (1883), 1278.

<sup>2)</sup> C. Ramsauer, Ann. d. Phys. 84 (1927), 721.

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nearly a circularly curved path in water and the angle of emergence was nearly equal to that of incidence. T. Isobe<sup>3)</sup> determined the critical angle of incidence for occurrence of ricochet for ordinary pointed bullet. In this case, the axis of bullet, after the immersion into water, started rapid rotation in the direction such that the pointed side turned upwards, thus at the first stage, the bullet received the lifting force of greater magnitude than the case of sphere, resulting to some complicated path in water. As an example of such a case, a moment photograph of bullet moving in water taken by the present writer is shown in Fig. 1., in which the region of great pressure along the front surface of the bullet revealed by the change of index of refraction of water will be noticeable.

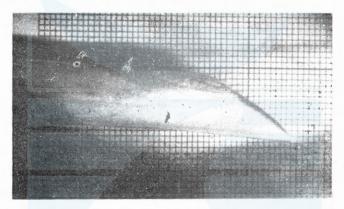


Fig. 1. Moment photograph of 6.6 mm bullet moving in water.

These investigations indicate clearly that the ricochet is not the phenomenon occurring only at the surface but is closely associated with the behaviour of the body in water. When the shape of the body is oblong, if we want to prevent its ricochet and to extend the path straight forewards into the water as far as possible, it is necessary to keep its longer axis always in the direction parallel to that of motion.

The rudder action of a flat plate moving in fluid should be noticed as the most effective measure for this purpose. Namely, the most stable orientation of the flat plane moving in water is attained when the normal to the surface coincides with the direction of motion. Various reports have already been published concerning the bullet with flattened head for the purpose of hitting the object under

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<sup>3)</sup> T. Isobe, Journ. Ordn. Soc. Japan 36 (1942), 237.

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water<sup>4)</sup>. Now in the case of harpoon, however, with such a long stem trailing heavy rope, it is questionable in what extent the flattening the head is effective in comparion with the case of bullet. Great difficulties will be expected to solve the problem only from the side of the mathematical calculations. In Fig. 2, a schematic diagram of expected formation of cavitation in water due to the head of harpoon.

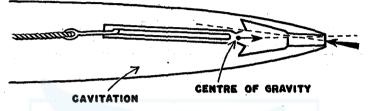


Fig. 2. Harpoon moving in water. In case of slight inclination of the axis of harpoon, the force acting at the flattened head surves the restoring effect.

So, in the course of the test work of Taiheimaru No. 1 mentioned above, following preliminary experiment was made in the harbour of Kusiro, Hokkaido, in September, 1949.

### 3. Experiment on flattened head harpoon

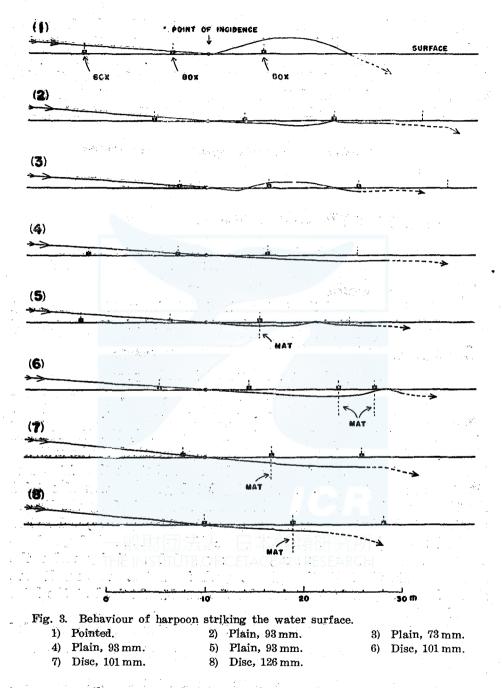
The tip of the ordinary grenade of harpoon was cutted off and the inner cavity was filled closely with hard wood making a flattened head. Two kinds of such grenades were prepared, the diameters of the flattened areas being 93 mm and 73 mm respectively. The ordinary harppons equipped with these grenades were shot at the distance of about 50 m from the gun. An usual pointed grenade was also tried for the comparison. Three small boxes were floated along the line of shooting at the distances of 25 ken (45.5 m), 30 ken (54.5 m) and 35 ken (63.6 m) respectively as the markings of distances for naked eye observation. A straw-mat  $(2 \times 2 \text{ m}^2)$  was suspended vertically under the second box for the indication of the path in water. The results are shown in Fig. 3, which contains also the results obtained at the later experiments.

The flattened head harpoon with the diameter of 93 mm proceeded

<sup>4)</sup> T.F. Framantle, "The Book of the Rifle", (1901) 304. Scientific American, 68 (1918), 125.

T. Isobe made a detailed experiment on 6.6 mm bullet with flattened head in 1943, though not published officially, of which the writer was told by his kind personal communication.





in water trailing a straight streek of cavitation as long as 20 m in one case (Fig. 3 (4)), while in the other case (Fig. 3, (2), (5)), the harpoon

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exposed a part of its head out of the surface only a moment after diving into water as long as 14 m from the point of incidence and again continued its way into water. In the case of 73 mm in diemeter, the duration of the first divinig into water was much reduced as shown in Fig. 3, (3).

Consulting on these results, the pointed head of electric harpoon which had been used at that time was decided to be replaced by the flattened one. On board of Taiheimaru No. 1, however, any suitable technical facility was not available, so a thick circular disc of steel was bolted on the tip of the harpoon as a temporary measure (Fig. 4). Three discs of different diameters, 89 mm, 101 mm, 114 mm, were prepared, thickness being the same, 19 mm.



Fig. 4. Electric harpoon equipped with a circular disc, 101 mm.

### 4. Thrusting power of flattened head into blubber

It was feared originally that the resistance of flattened head should be so great that the harpoon could not penetrate the blubber. But, at the first practical application of this electric harpoon equipped with 101 mm disc for a Sei whale (13.25 m) at the distance of 50 m, the harpoon hit the back of the whale after passing through the water as long as 2 m, and this harpoon was found by dissection lodging in the meat, 1.5 m deep from the point of intrusion. In the second case, when we met a Sperm whale (13.45 m) the same harpoon was used which struck aslant the blubber and lodged in the meat 2 m beneath the surface. It was also revealed that at the point of intrusion of harpoon the surface of blubber was scooped out in the form of a conical groove gradually increasing its depth as long as 80 cm. The angle of incidence of the harpoon to the surface was estimated from the dimensions of this groove as only  $5^{\circ}$ , thus it was ascertained that the flattened head was also very efficient for restraining the slipping at the surface of blubber.

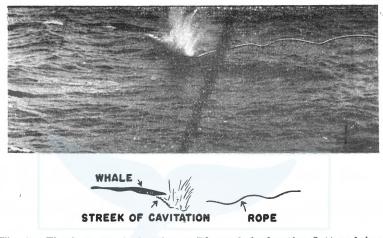


Fig. 5. The instant of shooting a Blue whale by the flattened head harpoon, which is seen making its way into water trailing a white streek of cavitation.

In many examples obtained later, involving the cases of explosive harpoons, the thrusting power of flattened head, 101 mm in diameter, proved satisfactory (Fig. 6), except when the distance was greater than about 70 m, especially in the cases of Sperm whales.

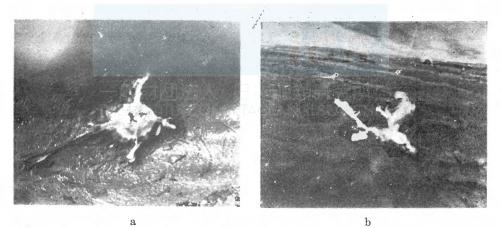


Fig. 6. Marks of intrusions of harpoon into blubber.a) Flattened head, 101 mm, Sperm whale.b) Pointed head, Sperm whale.

The relations between the penetrating depth into blubber (x), the diameter of head plane (d), the mass of harpoon (m) and its striking velocity (v) were obtained by the model experiment with the whalemarking darts<sup>5)</sup> and the following expression proved applicable approximately,

$$x = \frac{Km}{ad^2 + A^2} v^2,$$

where, K and  $\alpha$  being both a constant respectively, determined by the physical properties of blubber, A, an effective diameter of the maximum sectional area of harpoon containing hooks. This relation can also be derived theoretically with some plausible assumptions, that the decrement of momentum of harpoon, after its intrusion into blubber, is equall to the sum of the momentum newly given to the mass which is pushed away according to the progress of harpoon and the work done against the resistance due to the flattened head, while this resistance is supposed to be nearly the same to the one obtained when the harpoon is pushed in statically.

If we want to get much deeper penetration of harpoon into blubber at a given shooting distance, or by a given striking velocity, the

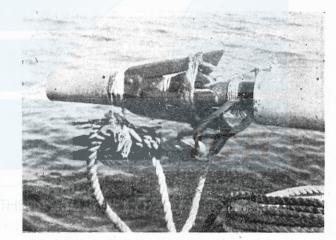


Fig. 7. Flattened head grenade, 90 mm.

diameter of the flattened head should be decreased suitably as derived from the above equation, though the efficiency of restraining ricochet will lessen inevitably. Thus, as for the size of flattened head,  $80 \sim 90$ 

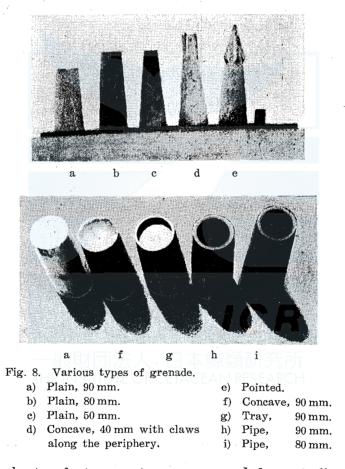
<sup>5)</sup> S. Nishimoto, M. Hirata, Report of the Committee for Improvement of Equipment of Whaling Vessels, Feb. 24, 1950.

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mm in diameter seemed suitable under usual conditions, and such grenades of explosive harppon have been tried in practice by some of the gunners with good results in the neighbouring waters of Japan as well as in Antarctic ocean since the winter of 1949 (Fig. 7).

### 5. Supplementary experiment

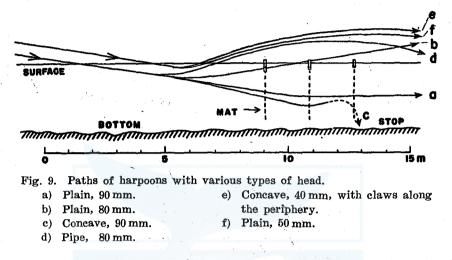
An additional experiment was carried out in August, 1951, with the gun placed on the coast of Ayukawa concerning various modified types of grenades such as shown in Fig. 8.



Three sheets of straw-mat were suspended vertically in water fixed by wooden frame, separating 2 m with each other, for the purpose of determining the path of harpoon in water. At the same time, high speed cinematographies were taken with the rate of 130 frames

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per second. The velocity of harpoon at the instant of incidence into water was about 75 m per sec., while the angle of incidence being 8°. Some of the results are depicted in Fig. 9.



It is revealed that the efficiencies of restraining ricochet of the head with concave surface as well as with thin walled pipe, which are originally planned for the purpose of prevention of slipping at the surface of blubber, are inferior to that of flat surface with the same diameter, while their velocities in water drop more quickly.

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