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# On the Sensory Tubercles of Lips and of Oral Cavity in the Sei and the Fin Whale

by

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Knowledges upon the sensory capacity of the cetacea are not only important for the biological science, but must have also some practical meaning for the whalers. Recently a few works pertaining to this field have been published from our laboratory, viz. upon the sinus-hair of the Sei whale (Nakai and Shida), the auditory organ of the Fin whale (Yamada), and the intracerebral acoustic system of the whales (Ogawa and Arifuku); the paper last mentioned contains the authors' summarized observations upon the serially sectioned brain-stems of *Lagenorhynchus*, *Tursio*, *Cogia*, *Physeter*, and *Balaenoptera*.

In the study upon the sinus-hair of the Sei whale, Nakai and Shida were induced to believe that the scanty but very regularly arranged hairs serve not merely as a tactile organ in searching for food, but more probably as the organ to feel the stream of water. They came to this assumption chiefly from findings on the inclination of the hair-shaft and the excentric position of the hair-root within the venous sinus. Huber (1934) seemed also to have considered that the sinus-hair of the baleen whales may afford a mechanism for the appreciation of slight changes in water pressure.

While studying the sinus-hair, one of us, Shida, directed his attention to a large assembly of small granular tubercles on the inner surface of the upper and lower lip in the Sei whale, *Balaenoptera borealis*. The same structure was seen, though less remarkably, not only on the palatal surface sandwiched in between the right and left areas of baleens, but also on the tongue and on the floor of the oral cavity. Microscopically examined, they certainly represent highly sensitive tactile organs, performing no doubt a function of much importance in the sensuous life of this whale. A little later, we ascertaind in the Fin whale, *Balaenoptera physalus*, the existence of structures approximate to those of the Sei whale. Though we have not yet compared other sorts of baleen whales regarding this problem, it seems plausible that this sensory apparatus will be found also in other whales, at least in all members of the genus *Balaenoptera*.

1. Distribution of the granular tubercles on the lips and in the mouth cavity.

The tubercles in question are easily seen with naked eyes (fig. 8 a) on the inner surface of the lips as well as on the mucous membrane of the mouth. The size of each tubercle and their number per unit area are not equal according to the locality. On the upper lip, where their presence is the most prominent, they are seen in a long and broad zone, which extends from the anterior extremity of the lip backwards to the angle of mouth (fig. 1 a). The zone is of a breadth 3-4 cm at the anterior end. 6-7 cm at the middle part of the lip and becomes remarkably wider towards the oral angle. There remains a narrow space of mucous membrane of a breadth about 1 cm, where the tubercles are wanting, between the lateral margin of the bases of baleens and this zone (fig. 2). At the anterior end of the upper lip the tubercles are relatively large, each of them measuring about 1 mm in diameter, in the hinder parts they are smaller but more crowded, where it is difficult to determine their accurate number. The density is ca. 200 per square centimeter at the anterior end (fig. 3), about 550 at the middle part of the upper lip, and nearly equal or a little less at the posterior end.

On the lower lip (fig. 1 b), where in general the presence of the tubercles is not so prominent as in the upper lip, they are the best seen in the posterior area near the oral angle, here about so clearly as in the hinder part of the upper lip. Forewards they become within the lower lip more indistinct, and at the most anterior portion the surface is everywhere smooth, devoid of any remarkable tubercle, but instead of this, many gray spots are visible on the surface, denoting the existence of well developed papillae of the connective tissue underneath.

On the narrow palatal mucous membrane between the right and left areas of baleens, relatively large granular prominences are present only in the anterior part, about 1/5 of the entire length of the palate (fig. 1 a), while in the hinder part the surface looks quite smooth, though here also we can recognize easily through the epithelium tops of many papillae of the connective tissue. On the sensory tubercles of lips and of oral cavity in the Sei and the Fin whale

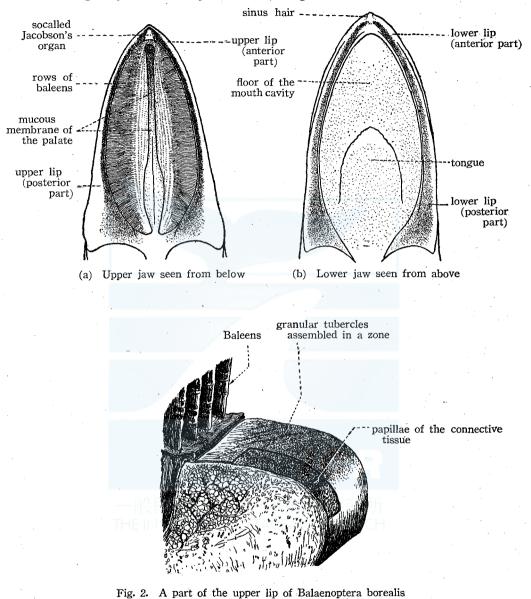


Fig. 1. Jaws of Balaenoptera borealis; the granular tubercles are dotted.

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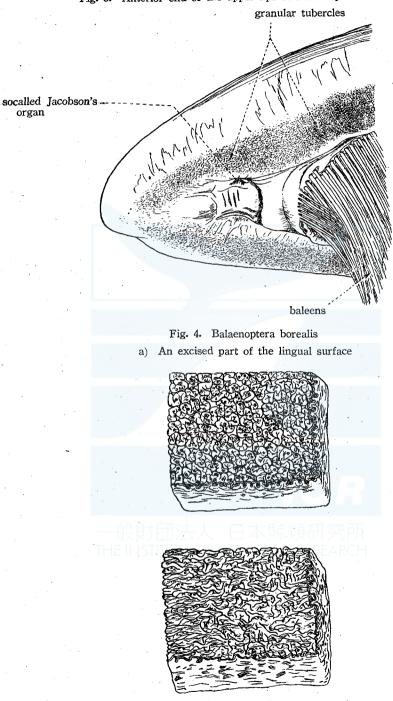


Fig. 3. Anterior end of the upper lips of Balaenoptera borealis

b) Excised mucous membrane from the floor of the oral cavity

# On the sensory tubercles of lips and of oral cavity in the Sei and the Fin whale

In addition, the dorsal surface of the tongue and the floor of the oral cavity have also the granular tubercles nearly all over the surface, except the most anterior parts (fig. 1 b), and backwards on the tongue they become sparse, disappearing completely at the fauces. The tubercles disseminated in the posterior part of the oral floor do not show any clear boundary against those in the hindermost portion of the lower lip. On the dorsal surface of the tongue, where the mucous membrane forms a great number of soft and gyrate folds, the tubercles are seen only on the summits, but not in the valleys between the folds (fig. 4 a). The same circumstance applies also to the floor of the oral cavity (fig. 4 b). In these localities the density is about 70 per square centimeter in the normally contracted mucous membrane. So when the membrane is distended, their number per unit area will be much decreased.

In the Fin whale, compared with the Sei whale, we saw a difference of little significance, only in regard to the upper lip, for the tubercles are within this lip the most easily recognized in the hindermost portion, and forewards they become relatively less distinct, being much smaller at the anterior end than those at the corresponding part of the Sei whale. But as to the tubercles of the lower lip, the palate, the tongue, and other parts of the oral cavity there is no remarkable difference between the two kinds of *Balaenoptera*.

2. Papillae of the connective tissue.

Microscopical examination reveals, that just under the tubercles papillae of the connective tissue are very well developed, protruding deep in the layer of epithelium, and approaching very near the epithelial surface. The form of these papillae is not always equal according to the locality, but usually they are more or less swollen near the summit and show secondary branchings. Especially at the anterior end of the upper lip of the Sei whale the papillae are thick and high, providing with 3-5 elongated branches (fig. 5 a), while at the posterior part of the upper as well as of the lower lip they are in form somewhat like mushrooms, having less branches (fig. 5 b). At the palate and at the anterior part of the lower lip they are more cylidrical (fig. 5 c), and on the tongue as well as on the floor of the oral cavity their form is irregular, though thick and short in most cases (fig. 5 d).

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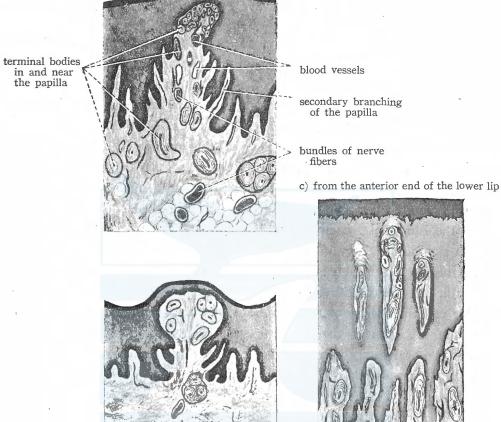
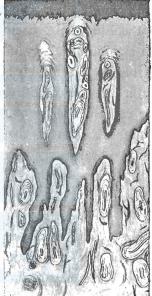


Fig. 5. Balaenoptera borealis; sections showing the sensory papillae a) from the anterior end of the upper lip

b) from a middle portion of the upper lip





d) from the dorsal surface of the tongue

# On the sensory tubercles of lips and of oral cavity in the Sei and the Fin whale

In the Fin whale we see very slender papillae well developed, reaching near the surface of the epithelium, at the anterior end of the upper lip, where as mentioned above the granular prominences are indistinct. As to other places, no remarkable difference was ascertained between the two kinds of *Balaenoptera*, concerning the development of the papillae in question.

3. Nerve endings in and near the papillae under the tubercles.

Within the papillae, especially near the summit of them we see many terminal bodies of very characteristic form, to which sensory nerve fibers are continuous. The intrapapillary terminal bodies are aggregated especially in a large number at the anterior end of the upper lip of the Sei whale. But the apparatuses denoting the nerve ending are also found outside of the papillae near the bases of them in the subepithelial layer. These extrapapillary terminal bodies are the most frequently seen at the anterior end of both the upper and the lower jaws, less in other parts of the lips as well as in the anterior portion of the palate. In these places we see here and there within the depth of 0.4 mm from the base of the epithelium a small group of 2—6 terminal bodies, sectioned in various directions and accompanied by a nerve bundle.

The general form of the terminal bodies within or near the papillae is known easily by examining the serial sections, stained simply with haematoxylin-eosin. But we further tried block-staining by the Bielschowsky's method, and prepared the serial sections and stained them with van Gieson's picrofuchsin, in order to study the finer structures, especially of the axons.

In the basal part of the papilla the terminal bodies are comparatively large in size, but less in number, while near the top of the papilla they are relatively small but more numerous (fig. 5). Calculated at the base, each papilla seems to receive from one to several nerve bundles, each of which contains a number of nerve fibers. Sometimes we see a very small bundle, consisting of only a single nerve fiber. A terminal body near the top of the papilla measures  $15-48 \mu$  in breadth, while its length can not be easily determined, as its very long body is wound in a complicated manner, not unlike the secretory part of a sweat gland. The terminal body has in the interior a relatively thick, eosinophile core and the core

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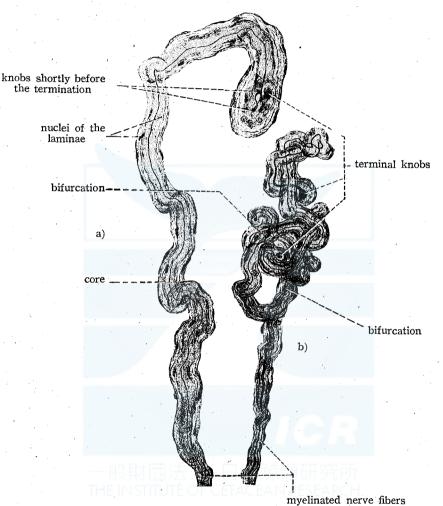


Fig. 6. Terminal bodies, not branched (a) and branched into three (b), both of which are much convoluted near the summit of a papilla ( $\times 500$ ). Reconstructed from sections.

is covered externally by several thin sheets of collagenic nature. This apparatus belongs certainly to the category of the lamellar corpuscles of Vater and Pacini, but the number of lamellae is in this case very few. So they seem to be the nearest to the Golgi-Mazzoni's corpuscles, as described by Dogiel and van de Velde especially in the tactile balls of the cat. But compared with these corpuscles already known, they are surpri-

### On the sensory tubercles of lips and of oral cavity in the Sei and the Fin whale

singly long and much convoluted (fig. 6). Because of the convolution a single terminal body appears on the cut surface several times, which may lead to an estimation of them too much in number. But in reality, even at the anterior end of the upper lip, where the terminal bodies are the best developed, their number is in one papilla a little more than ten.

Each terminal body seems to receive only a single nerve fiber. But we encountered bifurcations of the terminal body, the axon with core and lamellae being divided into two or three equally thick branches, which are proximally united together, but have their ends separately (fig. 6 b). Such a remarkable branching of the terminal body was observed especially at the anterior end of the upper lip of the Sei whale. In the Fin whale bifurcation of the axon occurs within the core very rarely.

The axon forms at the termination a small rounded or more irregularly formed knob, which is not always the same in size (fig. 7). In the knob we see a network or a glomerule-like group of fine fibrils, with which neurofibrils of the axon are directly continuous (fig. 7).

Moreover, we observed the axon in the terminal body a short distance before its real ending to be divided suddenly into several fibrils; the frayed fibrils are conglomerated, forming a knob, and then unite together into a single axon, and advance further to attain its real ending. Such a fraying occurs 1-3 times during the course of an axon. Van de Velde seems also to have seen the same relation in the Golgi-Mazzoni's corpuscles of the cat (1907, fig. 8 of his paper).

The extrapapillary terminal bodies in the subepithelial layer are elongated ovoidal or rod-shaped, provided with a core internally and with a lamellated capsule externally. The number of lamellae is as few as within the papilla. Also there they are wound, though the windings are not so numerous, compared with the intrapapillary terminal bodies. They are various in size; we measured a breadth of  $38-170 \mu$  and a length of 150  $-615 \mu$  (in the relatively straight ones). The core is  $7-23 \mu$  in breadth; this is swollen at the end usually to a diameter of  $30 \mu$ . Within the core a single axis cylinder is contained, and its terminal portion is dilated into a spherical or irregularly formed knob, with a network of fine fibrils.

4. Considerations upon the functional meaning.

The papillae mentioned above, being provided with so well developed

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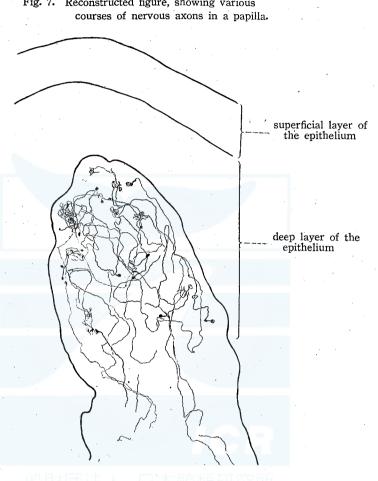


Fig. 7. Reconstructed figure, showing various

terminations of nerve fibers, are certainly an exquisite sensory organ for the oral and labial stimulations. The impulses must be conducted from them to the brain-stem by the maxillary and mandibular nerves of the trigeminus.

Concerning the problem, whether the sensation caused by these papillae has anything to do with the gustatory sense, we suppose, that it can not

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belong in its nature to the sense of taste, for histologically the terminal apparatuses, showing no intraepithelial nerve endings, quite different from the well known structures of the taste-buds with the intra- and intergemmal nerve fibers. From every point of views this labial and oral apparatus does not seem to be a chemoreceptor but merely a tangoreceptor, receiving mechanical impulses, therefore very different from the taste.

On the other hand, we are at present uncertain, whether the cetacea be endowed with the gustatory sense, for we have not yet found the tastebud in them, though Wolf (1911) is said to have seen it in toothed whales. Moreover, considering the meagre development or the complete absence of the olfactory nerves in the whales or dolphins respectively, it is quite possible in our opinion, that the oral sense treated especially in the present paper may substitute for the gustatory and olfactory senses in the cetacea. It may also compensate in a certain degree the insufficiency of the visual sense, for the comparatively small eyes of the whales, situated far behind and directed to the side, do not seem to be a very effective organ for the perception of food entering the mouth.

The sinus-hair might also be effective as a tactile organ for food, but they serve probably more for appreciation of the pressure or stream of water, having functionally much in common with the lateral line system of the fishes. Nearly eighty years ago Malm (1866) reported an interesting experience upon a Blue whale stranded alive at Göteborg, that "Der Mann, welcher den Walfisch erlegte, bemerkte ebenfalls, dass die Lippen unter den zugänglichen Teilen die empfindlichsten waren, da das Tier durch Berührung derselben in die grösste Raserei versetzt wurde." Japha (1910) quoted this story to explain for the high sensitiveness of the hair for touch, but in our opinion the granular tubercles with the richly innervated papillae are far more responsible for the sensitiveness of the lips than the hair. At present one of our eager wishes lies in a repetition of such an interesting experiment as Malm reported.

In the human anatomy it is well known, that the inner surfaces of the lips and of the cheek have in the newborn infants as well as in the advanced foetuses numerous villi, which resemble therefore in localisation the granular tubercles of the whales; they however begin to disappear in the fourth week after the birth (Ramm, Heidsieck). In other mammals, it deserves also noticing, that villi are well developed on the mucous mem-

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brane of the lips and of the cheek of ruminants and of many marsupials (Owen, Immisch, Schulze, Sonntag). Though coincident in a certain degree as to the localisation, the villi in question of these animals seem to have only a mechanical meaning in food-taking or in mastication; they have probably not the meaning of highly sensitive organs as in *Balaenoptera*. Neither can we expect any important function in the labial and buccal villi of the human being, which disappear very soon after the birth.

Lastly we wish to consider briefly the interrelationship between the cetacea and other mammals. Since old time the opinion has prevailed, in the realm of comparative anatomy, that the whales are the most kindred to the ungulates. Relatively recently Anthony (1926) insisted upon the affinity between the whales and the perissodactyla, and later Ommanney (1932), studying the urogenital system of the Fin whale, agreed with Anthony on this point. On the other side, many palaeontologists have been inclined to look for the ancestor of the living cetacea in carnivora, taking chiefly the dental characteristics of *Zeuglodon* in comparison.

Of course we can ourselves say nothing definite upon this important problem, but would only refer to the fact, that the remarkable development of the palatal crests simultaneously with the high villous state of the labial and buccal mucous membrane is met with in ruminants. And the baleens of the whales have been explained since Tullberg (1883) as specially differentiated sort of the palatal crests, an opinion, which seems to have much truth. Moreover, the palatal crests of some ruminants are closely related to the baleens of whales, inasmuch as they are frayed at the margin. This combination of structures, that is, the simultaneous presence of palatal crests and of labiobuccal villi, is found in a very accentuated and specially differentiated form also in the whalebone whales. Naturally it does not mean an important thesis in determining the relationship of the baleen whales, but a comparative anatomical fact not unworthy of being borne in mind at such a discussion. Of course the toothed whales stand here out of the question, but we think there may be phylogenetically a pretty large distance between the toothed and the baleen whales.

# Summary

1. On the lips as well as in most parts of the oral cavity of the Sei whale a great many sensory tubercles are found, especially well developed at the anterior portion of the upper lip. In the Fin whale the circumstance is nearly the same, with only the difference, that in this species the elevations are at the anterior portion of the upper lip comparatively less distinct.

2. Microscopically examined, we see underneath the tubercles well developed papillae of the connective tissue, containing numerous sensory apparatuses, with which the nerve fibers are directly continuous.

3. The terminal bodies in question show the structure the most approximate to the so-called Golgi-Mazzoni's corpuscles. But they are characterized by the remarkable elongation. They have a core with the axon internally and a few sheets of collagenic lamellae externally.

4. Successive bifurcation of a terminal body into equally thick branches occurs in the Sei whale. We found also sometimes the axon before its real ending to be frayed out suddenly into a number of neurofibrils, which after forming a knob unite together into a single axon, to proceed further toward its termination.

5. Because of the presence of the granular tubercles, the lips and the mucous membrane of the oral cavity seem in *Balaenoptera* to be a highly sensitive tactile organ, which is very important for the perception of foods, while the sinus-hair are believed to have the functional meaning more for the appreciation of pressure or stream of water.

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### Fig. 8.

a) Granular tubercles at the anterior end of the upper lip; with naked eyes, Sei whale.



c) Axons seen here and there in the papilla at the anterior end of the upper lip; a terminal knob is visible. Fin whale (×95).

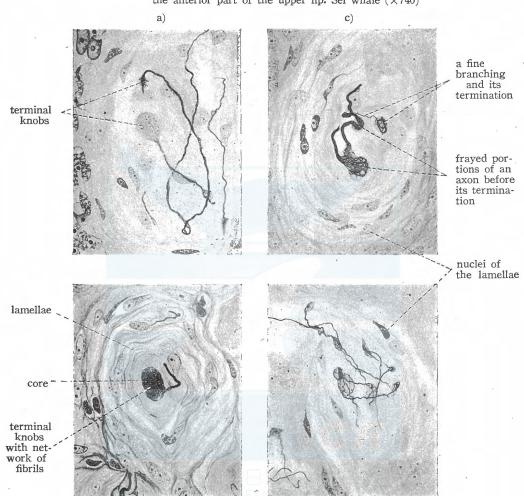




 b) Cross-section of a papilla at the anterior end of the upper lip, containing many terminal bodies. Sei whale (×65).

d) Oblique section of a terminal body, showing core and lamellae, at the anterior end of the upper lip. Fin whale ( $\times 160$ ).

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d)

b)

Fig. 9. Various aspects of axons in the papillae at the anterior part of the upper lip. Sei whale  $(\times 740)$ 

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# Distribution of the Red Marrow in Bones of the Fin Whale

by

# Tadahiro Ohe

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The present investigation was performed at Mombetu, Hokkaido, in the summer of 1948, when I stayed there for ten days. The object of my study was to determine the site of haematopoesis in the body of whales, for then we had in mind a question, whether or not the blood corpuscles of the baleen whales are developed in the organs just as they are in the terrestrial mammals. Except the quite recent paper of Slijper et al (1948), which reached us after the completion of the present work, there seems to have been no detailed report upon the distribution of the red bone marrow in the baleen whales.

Materials:

Four individuals of the Fin whale, *Balaenoptera physalus L.*, were examined. All of them were captured off the northeastern coast of Hokkaido by boats of the Taiyo Fishering Company; by the courtesy and deep understanding of the members of this company the present work could be fulfilled to the success of a certain grade.

Methods:

Of course at the whaling station the scientific research is met with much difficulties, as speedy disposition of the whale body is the most important factor in the whaling industry and in most cases the heavy materials can only be moved by the help of winches. We can examine them not always to our hearts' content. My own experience at that occasion is as follows. The vertebral column, after most of the truncal muscles were cut off, is brought by the winch to the "bone platform"; the huge mass was separated into each segment, by cutting through the intervertebral discs. At that time I endeavoured to arrange all of the vertebral bones in the natural order, and asked one of the workers to break them lengthwise in the oro-caudal direction. Of some vertebrae the transverse and the spinous process were also cut for examination of the bone marrow. The cervical vertebrae were split longitudinally in a mass.

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The dissectors used to remove the ribs altogether with intercostal muscles and pleura as a whole from the vertebral column, then to separate them into each rib and to bring them to the bone platform. At that occasion I arranged the ribs in the natural order on each side and examined their caput, cauda and the middle part, sectioning transversely. Though the ribs of the Fin whale are said to be usually fifteen pairs, there were two cases Nos. 42 and 40, in which I found only fourteen pairs. The problem, whether there were really only fourteen pairs, or one pair, probably the last, was lost at the dissection platform, is not certain. I guess the former assumption may have more truth.

To my regret, I had no chance to examine sternum and pelvis. I did not dare to find out these small bones, not to disturb too much the speedy work at the dissection platform. The chevron bones are carried to the bone platform as a long chain-like block, and I cut some of them to see the marrow.

The large skull is at first divided by the bone saw lengthwise at the middle line into right and left halves, cut then in various directions into many blocks, which are small enough to be easily dropped into the pressboiler. I examined the cut surfaces of them as much as possible. The hyoid consists of one median part (basihyal and thyroyal) and paired lateral parts (stylohyal). The stylohyal was usually not discovered, as it was carried probably to another place. I cut as a rule the hyoid bone in the median plane and further along its long axis.

The scapula was cut into two parts, a cranial and a caudal. The large projected acromion was examined especially. Bones of the pectoral limb were cut in a mass transversely into two pieces, and then in the same direction into four portions. The head of the humerus was especially observed.

I made some microscopical preparations from the red marrow and ascertained that the haematopoesis was certainly taking place in it.

No. of the specimen	44	41	40	42
Body length in feet	50	52	53	67
Sex	Female	Male	Female	Female
Date of the capture	8 a. m. August 14.	5 p. m. August 12.	5 p. m. August 9.	8 a. m. August 12.

**Results**:

# Distribution of the red marrow in bones of the Fin whale

Beginning of dissection		6 p. m. August 14.	3 a.m. August 13.	6 a.m. August 10.	4 p. m. August 12.	
Approximate hours elap- sed between capture and dissection		10	10	11	8	
Vertebral	formula	$7+15+14 \\ +20=50$	$7+15+14 \\ +22=58$	Not especially examined.		
Atlas		The red marrow was found only in the ventral arch.				
Cervical vertebrae 2-7		Entirely filled with red marrow, but in No. 44 not so complete- ly as in three other individuals.				
Thoracic vertebrae 1-15		Also filled with red marrow, but the white marrow <sup>(1)</sup> was present in a wedge-like form <sup>(2)</sup> in the peripheral <sup>1</sup> part of the vertebral body, becoming larger in the caudal direction. <sup>(3)(4)</sup>				
Lumbar a vertebra		Beginning with the 15. (in No. 42 with 14.) thoracic vertebra caudalwards the red marrow becomes irregular in shape and gradually decreases in extent, but in Nos. 41 and 40 it was present in addition along the lines of epiphysis. A red spot was found down to the 2. caudal (No. 44), to the 12. lumbar (No. 41), to the 8. lumbar (No. 40), or to the 2. caudal (No. 42).				
presence of	es examined vertebrae (+: of the red mar- is absence)	6. thor.+ 14. thor 1. lumb	10. thor+ 15. thor	4. thor.+ <sup>(5)</sup> 7. thor.+ 10. thor.+ 14. thor	5. thor.+ 10. thor.+	
Skull		Two small pieces of red marrow, each the size of a hen's egg, we- re present in the basal part, and a larger one in the roof of the brain case.	One red por- tion was pre- sent in the ba- sal part and another in the frontwall of the brain case and one more in the occipital part.	Though I re- member to ha- ve seen the red marrow in the skull, I can not clearly locate it.	The skul had the rec marrow only in its anterior basal part.	
Hyoid		Not examined.	A relatively large red por- tion in the thy- rohyal and scattered red spots in the basihyal. Stylohyal not examined.	The red mar- row was pre- sent only in the thyrohyal. The stylohyal was not obser- ved.	The red mar- row was pre- sent neither in the basi- and thyrohyal, nor in the stylo- hyal.	
Ribs		The red marrow was present only in the caput. But only in the caput of the 10. rib of No. 41, the red marrow was mixed with the white one.				
Glenoid			The red spot	he red spot was seen.		
Scapular plate Acromion		Three very small spots of the red marrow were found.	Here and there red mar- row was pre- sent.	No red r	narrow	
		No red marrow				
		No red marrow				

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Supplementary remarks to the table of the foregoing pages:

- (1) Man says usually "red" and "yellow" as to the bone marrow, but in the whale, especially in the vertebrae of them, the yellow marrow might be better called "white", because in the lumbar vertebrae, for instance, we get an impression, as if red ink were dropped on the snow. So I used in this paper, instead of "yellow", the term "white bone marrow." But in hyoid, scapula, and bones of the skull the yellowish colour predominates.
- (2) By cutting further, the red marrow was seen in form of an hour-glass.
- (3) In the epiphyses of the thoracic vertebrae of No. 42 the red marrow of a patch-like form is present in the center, surrounded by the white marrow; this fact was not seen in the other specimens.
- (4) In the 10. to 13. thoracic vertebra of No. 42 the dorsal wedge of the white marrow was relatively sharp and small, while the ventral white part showed more rounded contour.
- (5) The red marrow in the processes of the 4. and 7. thoracic vertebra was continued to that of the vertebral body, while that in the processes of the 10. thoracic vertebra showed no direct continuation with that of corpus.

# Summary:

(1) In the atlas the red marrow is present only in the ventral arch. From the 2. cervical down to the 14. thoracic vertebra the vertebral bodies are filled nearly completely with the red marrow, and as the whale advances in age, this seems to disappear in the caudo-cranial direction.

Beginning with the 15. thoracic vertebra the red marrow becomes irregular in shape and decreases suddenly in extent, but point-like red places can be found up to lumbar or caudal vertebrae.

As Slipper says, the red marrow seems to disappear earlier in the processes than in the corpus of vertebra. In the latter the white marrow appears at first in a wedge-like form in the circumferential part, and at that time the convertion of the red marrow into the white has not yet finished in the processes.

In the vertebrae of two individuals (No. 44 and No. 41), the red marrow, irregular in shape, was also present along the lines of epiphysis, but this was not the case in the other two (No. 40 and 42).

(2) In scapula, hyoid and skull the red marrow becomes smaller with the

Distribution of the red marrow in bones of the Fin whale

age. Slipper et al seem in the Blue whale not to have seen the red marrow in these bones.

(3) In all of the costae the red marrow was present in the caput.

(4) In bones of the flipper and in the chevrons the red marrow is completely lacking.

# Explanation of the figures:

Except hyoid, lateral views of all of the bones are shown. The processes of vertebrae are omitted; ribs, hyoid and bones of the flipper are moved from their natural positions. black.....red bone marrow.

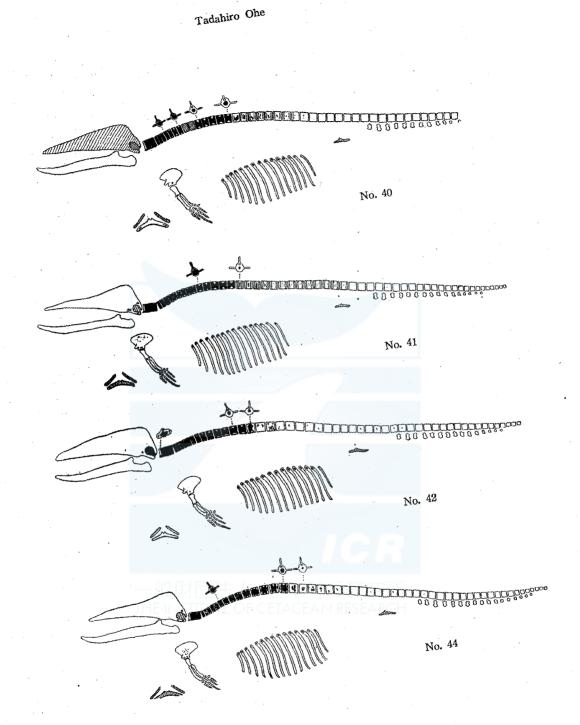
obliquely lined area .....bones not examined.

# Literature :

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# On the Cetacean Larynx, with Special Remarks on the Laryngeal Sack of the Sei Whale and the Aryteno-Epiglottideal Tube of the Sperm Whale

# by

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# Introduction

Since very early the larynx of the whales has, because of its peculiar structure, attracted notice of many anatomists. The most striking peculiarities lies in the tube formed by the upward elongated epiglottic and arytenoid cartilages and dilated at its summit like a knob, protruding into the choana. After Tyson (1680), who observed this tube for the first time in Phocaena communis, a number of anatomists described this structure in various kinds of whales, for instance, J. Hunter (1787) in the harbour porpoise, dolphin, pottfisch and narwhal, Meckel (1833) in Delphinus and Phocaena, Stannius (1846) in Delphinus and Monodon, Vrolick (1848) in Hyperoodon, Huxley (cited from Howes), Burmeister (1867), Howes (1879), Rawitz (1900) and Boenninghaus (1902) in Phocaena communis, Murie (1871, 74) in Grampus rissoanus and Globicephalus, Watson and Young (1878-79) in Beluga leucas, Dubois (1886) in Hyderoodon, Tursiops, Phocaena, Delphinus and Globicephalus, and Benham (1901) in Cogia breviceps. To the last mentioned author we owe probably the name "arytenoepiglottideal tube".

Meanwhile it is worthy of notice that research materials of these anatomists were always toothed whales. The question naturally arises, whether the whalebone-whales have also the same tube in the larynx.

This problem was answered by Eugen Dubois (1886), who compared the larynges of Hyperoodon, Tursiops, Phocaena, Delphinus and Globicephalus with that of a fetus of Balaenoptera Sibbaldii, and asserted the non-existence of the aryteno epiglottideal tube in the latter. Besides there are, according to him, many other differences in the larynx between whalebone and toothed whales.

While engaging recently in the anatomical research upon some whales,

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I was much interested by the striking difference in the larynx between two groups of the whales and was led to make some detailed inquiries about this matter.

The following materials have been hitherto examined by me in this research. Whalebone whales:

- 1. Balaenoptera borealis, 42 feet, female; 46 feet, female (at Ayukawa, Miyagi Prefecture).
- 2. Balaenoptera musculus, fetuses of various lengths (in Antarctic Ocean).

3. Balaenoptera physalus, fetuses of various lengths (in Antarctic Ocean). Toothed whales :

- 4. Physeter catodon, 32 feet, male; 36 feet, female (at Ayukawa).
- 5. Lagenorhynx obliquidens, male fetus of 51 cm length (at Kawana, Shizuoka Prefecture).
- 6. Prodelphinus caeruleo albus, 11 feet, male; 117 cm, female (at Kawana).
- 7. Globicephalus melas, 15 feet, male; 16 feet, female (at Ayukawa).

In this paper I will treat mainly the larynx of the Sei whale (Balaenoptera borealis) and that of the Sperm whale (Physeter catodon); observations on the larynges of the others will be reported later.

# I. Cartilages of the larynx

The larynx of the whales is constructed, as in other mammals, of the cartilagenous framework and several muscles connecting the cartilages. They form as a whole a tubular organ with the laryngeal cavity in it. The inner surface is covered with the mucous membrane continuous with that of the pharynx upwards and of the trachea downwards.

The laryngeal cartilages are five in number, three of which (thyroid, cricoid, epiglottic) are unpaired and the rest two are paired (arytenoid). The corniculate and the cuneiform cartilages do not exist as separated ones. At first the form of each cartilage will be mentioned, compared between the Sei and the Sperm whale.

# The THYROID cartilage

Sei whale: (fig. 1) Two laminae, right and left, of this cartilage are fused together in the anterior middle line, there forming no sharp angle.

## On the Cetacean Larynx

The cartilage is as a whole a plate concave behind and shaped like a flying swallow, providing with four processes; the inferior corun, corresponding to wings of the swallow, is very well developed, projects caudally and makes a joint with the cricoid. The upwards directed superior cornu is not so well developed. Between the superior horns of both sides a deep V-shaped notch exists, separating partly the right and the left lamina (incisura thyreoidea cranialis). The caudal border has in the median line a shallow notch (incisura thyreoidea caudalis).

The outer surface of the laminae, smooth and convex as a whole, shows no remarkable prominence. The thyreohyoid and sternothyreoid muscles are attached to relatively small areas near the superior lateral border of the lamina. The cricoarytenoid muscle inserts partly to the outer surface around the caudal notch, but for the greater part to the inner surface of this cartilage. To the inner surface are attached also three muscles, thyreopharyngicus, thyreoarytaenoideus and thyreoepiglotticus. Moreover the posterior end of the inferior horn is connected with the musculus cerato-cricoarytaenoideus dorsalis.

Sperm whale: (fig. 1') The thyroid is the largest cartilage in the larynx of this whale, differing in shape very little from the homologous structure in most of other mammals. Two laminae, right and left, are fused together at the middle front, forming a tolerably sharp angle, and there we see the prominentia laryngica.

The superior horn is very slight, and the inferior one is also not so well developed as in the Sei whale. Each lamina is irregularly triangular in shape; its outer surface is not smooth, having along its upper border a lineal crest, which courses downward to the middle portion of the lamina, and is divided there in two crests; one of them goes to the dorsal side, slightly inclining to the caudal direction, while the other runs ventrocaudally, and serves as insertion for the thyreohyoid and sternothyreoid muscles. At the point, where the lineal crest changes its course from the upper border to the middle of the lamina, it surrounds a pore, foramen thyreoideum, through which a nerve (the inner branch of the n. laryngicus cranialis) and a vessel (a. laryngica cranialis) penetrate. Existence of such a pore was mentioned already by Stannius in various whales, and by Rawitz in Phocaena communis.

The inner surface of the lamina is nearly smooth and gives attachment

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to the thyreopharyngeal and thyreoarytenoid muscles etc.

The EpigLottic cartilage

Sei whale: (fig. 2) This elastic cartilage is not so hard as other larygeal cartilages. Its upper half, not so dilated widely as in the human larynx, is concave behind and convex in front. The lower half (petiolus epiglot-tidis) is long and thin, and directed to the cranial notch of the thyroid cartilage.

Sperm whale: (fig. 2') This large cartilage is an elongated triangle in shape seen from lateral, with an apex directed ventro cranially, where the hyo-epiglottic muscle is attached. The cranial apex is, seen from behind, triangular and involved forwards.

The hind surface of this cartilage has three lineal longitudinal elevations, one in the median line and two others along the lateral margins; the latter meet with the lateral border of the arytenoid cartilage and are connected with it by the mucous membrane, forming so together the aryteno-epiglottideal tube.

The caudal end of this cartilage has a tubercle, which is connected to the angle between two laminae of the thyroid cartilage, a little below the cranial noch. This connection is strengthened by soild ligamentous fibres.

The CRICOID cartilage

Seiwhale: (fig. 3) The name "cricoid" does not hold good for this whale, for the cricoid cartilage is not shaped like a signet-ring as in other mammals, but its ventral half, corresponding to arcus, is completely deficient and the whole cartilage is merely a plate concave in front. The upper margin of this plate has a slight prominence at the middle, and a little more laterad on each side an articular facet for the arytenoid cartilage.

Caudally it is directly united with the tracheal cartilages, forming together the cricotracheal plate in the dorsal wall of larynx and trachea. Lateral parts of this plate are partially separated by a longitudinal notch from the main medial portion and may be named "processus lateralis"; the inner concave surface of this lateral process gives attachment to a portion of muscles of the laryngeal sack, which will be mentioned later. Moreover, the m. cricothyreoideus inserts to the outer surface of this process, a little above its caudal end.

# On the Cetacean Larynx

Inner and outer surfaces of the cricoid cartilage, nearly smooth all over, have a slight crest along the median part of the outer surface, which runs from the upper border downwards, remarkably dilating. Wide areas on both sides of the crest are for insertion of the dorsal cricoarytenoid muscles. The most caudal part of this area is connected with the criocopharyngeal muscle. A little more mediad than the upper end of the notch, which demarcates the lateral process, there are articular facets for the inferior horns of the thyroid cartilage. This joint is not fully diarthrodial but somewhat syndesmodial, strengthened behind by fibrous bands, while the aryteno-cricoidal joint is diarthrodial, provided with the articular capsule.

Sperm whale: (fig. 3') This whale has a completely ring-formed cricoid cartilage, a very diferent circumstance therefore from the Sei whale. This fact deserves special attention, for, according to Dubois, all of the toothed whales he examined, with the exception of Tursiops, have incompletely ring-formed cricoid cartilage; in them the ventral portion is said to have a fissure, though the fissure is not so broad as in whalebone whales. Sanctis (1879) seems to have found in Physeter macrocephalus the same matter as in Tursiops (cited from Dubois); and Benham found also in an adult male Cogia a completely ring-shaped cricoid catilage. In other toothed whales examined by me the matter is quite the same as Dubois described. Therefore, as to the cricoid cartilage there are two groups in the toothed whales, one of which (Physeteridae etc.) has a completely ring-shaped cricoid cartilage, while the other (Delphinidae and Hyperoodon) an incom-The cricoid cartilage of the sperm whale is not united with plete one. the tracheal cartilages, a distinct space filled with fibrous bands existing between them.

The very thick posterior portion of the cricoid cartilage gives attachment to the dorsal cricoarytenoid muscle, while the middle portion of its cranial border is connected with some fibres of the transverse arytenoid muscle. The anterior part of this cartilage is narrow and somewhat thin, but widens as it approaches the median line.

# The ARYTENOID cartilages

Sei whale: (fig. 4) Each of the paired cartilages has two processes directed up and down (processus cranialis and proc. caudalis). The tongue-shaped cranial one is thin in the transverse direction, while the caudal one

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is dick and rounded. The pointed ends of the caudal processes are connected by fibrous tissue between both sides. The main part of the cartilage, corpus, has a laterally directed large prominence, which corresponds probably to the processus muscularis, giving attachment for the dorsal cricoarytenoid muscle etc. The dorsal surface of this portion has an oblique oval facet to articulate with the upper border of the cricoid cartilage. The corpus has furthermore a crest along the medial border of its ventral surface, probably homologous to the processus vocalis. Upon the relation of various muscles to this cartilage 1 want to refer it to the accompanying figures.

Sperm whale: (fig. 4') The most remarkable difference upon the arytenoid cartilage between Sperm and Sei whale is the absence of caudal process in the former. In the Sperm whale the well developed cranial processes of both sides, standing side by side on the cricoid cartilage, form a concave furrow in front, and their upper ends are fused together into one cartilage. Between the ventrolateral border of this process and the lateral crest of the epiglottic cartilage a fold of mucous membrane (plica aryepiglottica) is stretched and thus the aryteno-epoglottideal tube is completed.

# The CORNICULATE and the CUNEIFORM cartilages

These cartilages could not be found in all the whales I have examined, at least as separated ones. Almost all of the authors who have studied the larvnx of whales did not mention about them. Watson and Young denied the existence of them in Beluga. But Howes remarked in a fetus of Phocaena that the arytenoid cartilage consists of two separated pieces, and the upper portion of the so-called arytenoid, which is prolonged into the nasal passage, must represent an elongated cuneiform cartilage, the Santorinian element being absent. Negus (1928) agreed with Howes; he had examined a reconstruction model of the larynx of a fetal Sperm whale, which was 4.5 inches long, and finding two columns of mesenchymal, cartilaginous cells lying above and slightly ventral to the arytenoids, mentioned confidently that these cell-groups would make the cartilages of Wrisberg and, together with the epiglottis, constitute the peculiar laryngeal tube. Of the Santorinian cartilage he denied its existence. In small fetuses of Blue and Fin whales I examined minutely the cranial process of the arytaenoid, but no separated piece of the cartilage was found, and in a

very small fetus (17 cm of length) of Megaptera nodosa, I saw that only the corpus of the arytenoid consists of cartilaginous element, the cranial process being formed merely of connective tissue.

# The TRACHEAL cartilages

As these are closely related to the larynx, I will give a brief description of them.

Sei whale: (fig. 3) All of the tracheal cartilages are continuous with each other and moreover, as mentioned above, with the cricoid cartilage in their dorsal parts, forming altogether a single plate. In the lateral parts they are however separated from each other by tranverse fissures and the ventral portions are completely wanting. Though Dubois stated that only the upper 4-5 tracheal cartilages are fused with the cricoid in a fetus of B. Sibbaldii, in my observation of the Sei whale all of the eight cartilages up to the bifurcatio trancheae form a single plate.

Sperm whale: (fig. 5') In this whale the tracheal cartilages are complete rings, and the most cranial one is separate from the cricoid cartilage. The upper three cartilages are fused together in the dorsal parts, but the others are all separated from each other. The fact, that all of the tracheal cartilages form complete rings, seems to be an exception in the whales. For, according to Dubois, the uppermost one (Hyperoodon) or two (Delphinus, Phocaena) tracheal cartilages are lacking of its ventral portion, and also Watson and Young mentioned the same relation as to two or three upper cartilages respectively for Beluga and Delphinus.

# II. Muscles of the larynx

Laryngeal muscles of the Cetacea have been studied by a number of anatomists, but their explanations are not always coincident. The difference of opinions come, to my mind, partly from difference of the materials treated. In fact, every species of the whales has more or less its own structure. We must naturally consider seriously the differences between toothed and whalebone whales. In the following my results upon laryngeal muscles of the Sei and the Sperm whale will be mentioned.

# Sei whale:

As in the case of other mammals, the laryngeal muscles are classified also in this whale into two groups, extrinsic and intrinsic. The extrinsic

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muscles, connecting the larynx with neighbouring structures, are:

Musculus sternothyreoideus (innervated by branches from the ansa hypoglossi)

Musculus thyreohyoideus (ditto)

Musculus laryngopharyngicus (innervated by branches of the plexus pharyngicus)

Musculus hypepigletticus or baseohypepigletticus

The STERNOTHYREOIDEUS and the THYREOHYOIDEUS belong to the so-called infrahyoidal muscles and insert to the thyroid cartilage. The well developed LARYNGOPHARYNGICUS consists of THYREOPHARYNGICUS and CRICOPHARYN-GICUS. The former arises from the frontolateral part of the inner surface of the thyroid cartilage and its massive fibres surround the pharynx to meet with that of the opposite at the posterior median part of the pharyngeal wall, i. e. raphe pharyngis. The latter originates from the dorsolateral part of the cricoid cartilage and, running up and medialwards, terminates also at the raphe pharyngis. The unpaired HyoEPIGLOTTICUS was described by Carte and Macalister (1868) in Balaenoptera rostrata, but I could not see it in the Sei whale.

The intrinsic muscles are those, which begin and end within the larynx itself. In the following I will describe them one by one.

(1) M. CRICOTHYREOIDEUS; (fig. 5) This muscle arises from the outer surface of the lateral process of the cricoid cartilage and runs upward and laterad, to insert on the medial border of the inferior horn of the thyroid cartilage. It is not divided into two parts, pars recta and pars obiqua, as in the human larynx. This muscle is the only one in the whole larynx, that is innervated by the cranial laryngeal nerve (fig. 18).

(2) M. CRICOARYTAENOIDEUS DORSALIS; (figs. 6,7) This muscle arises mostly from the posterior medial portion of the cricoid cartilage, partially also from the caudal end of the inferior horn of the thyroid cartilage. Its fibres converge upwards and laterad to reach the muscular process of the arytenoid cartilage. Because of the connection with the inferior horn, it may be called better "m. ceratocricoarytaenoideus dorsalis", the name used already by Dubois.

(3) M. ARYTAENOIDEUS; (fig. 16) Tracing the fibres of m. cricoarytaenoideus dorsalis upwards, we come across muscle fibres running transversally between the arytenoid cartilages of both sides. The fibres arise from the dorsolateral margin of the cranial process of the arytenoid cartilage on one side and attain the corresponding part of the same cartilage of the other side. This muscle (m. arytaenoideus transversus of Dubois) corresponds probably to the transversal part of m. arytaenoideus in the human larynx. The pars obliqua is not found in the cetacean larynx. According to Dubois the most caudal part of the arytenoidal muscle is covered by the upper part of the dorsal cricoarytenoid muscle, and moreover some fibres of the former are attached to the cricoid cartilage. On this point I agree with Dubois, though the boundary between these two muscles is not very distinct.

(4) Muscle of the walls of the laryngeal sack (M. THYREOARYTAENOIDEUS, M. CRICOARYTAENOIDEUS LATERALIS, M. ARYEPIGLOTTICUS, M. THYREOEPIGLOT-TICUS); (figs. 5, 8, 9, 12—17) As will be mentioned later, the Sei whale has a large sack attached to the ventral wall of the laryngotrachea. It communicates with the proper laryngeal cavity through a wide opening which exists between the arytenoid cartilages of both sides. This laryngeal sack has thick muscular walls and its muscles, cross-striated in nature, are arranged so differently from the laryngeal muscles in other animals, that it is not easy to determine the homologies between them.

Carte and Macalister examined for the first time these muscles in Balaenoptera rostrata with the conclusion that they belong to m. thyreoarytaenoideus. Dubois insisted that not only the thyreoarytenoid muscle but also the m. cricoarytaenoideus lateralis takes part in the formation of this muscular wall. Thereafter Benham, studying a newborn female Balaenoptera rostrata, supported Carte and Macalister's opinion and denied the co-existence of the lateral cricoarytenoid muscle.

At any rate these muscles exhibit quite strange arrangement in relation to the cartilages. In the Sei whale we see on the ventral surface of the larynx the m. cricothyreoideus on both sides. Moreover, massive muscle fibres come into sight from behind the caudal ridge of the thyroid cartilage, running downwards and being directed in the lateral parts somewaht laterad. These muscles, which form the ventral and lateral walls of the laryngeal sack, insert partly to the ventral ridges of the cricoid and tracheal cartilages, where these cartilages are defective in the ventral portion. More cephalic and deep fibres of them reach the muscular process of the arytenoid cartilage.

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The comparatively superficial muscular layer of the laryngeal sack courses as a whole in the caudo-lateral direction; I would call it "A-group" in the following description.

A deeper group of muscle fibres arises mainly from the ventral surface of the caudal process of the arytenoid cartilage and, diverging downwards, forms a part of the ventral, lateral and to some extent the dorsal wall of the laryngeal sack ("B-group"). These fibres seem to have no insertion to any cartilage, terminating within the muscular wall itself. The ventral portion of them, running caudally and mediad, contrary to the caudal and lateral direction of the A-group, is interwoven in the median line between both sides!

Such a division of superficial and deep layers is however not so distinct, and they form altogether the very thick wall of the laryngeal sack. For this reason, the muscles were taken as a whole by Carte-Macalister and Benham as the thyreoarytenoid muscle, while Dubois regarded probably the B-group as the lateral cricoarytenoid muscle.

It seems certain from my observation that the walls of the laryngeal sack are mainly formed by the thyreoarytenoid muscle, but this muscle has, because of the extraordinary development of the sack, additional insertions to the cricoid and tracheal cartilages. As to the problem, whether the B-group corresponds to the lateral cricoarytenoid muscle, I can say nothing confidently. The bare fact is that the superficial fibres are directed somewhat differently than the deep fibres.

In addition to A- and B-group, some muscle fibres situated in a more cranial place arise from the inner surface of the thyroid cartilage and course almost horizontally, surrounding the sack from lateral to reach the middle ventral part of the arytenoid cartilage ("C-group"). We see besides other fibres in a more cranial level, connecting the basal part of the upper process of the arytenoid cartilage with the root of the epoglottis ("Dgroup"). These groups are however not very clearly separated from each other, but form as a whole the thick wall of the laryngeal sack. I guess that the D-group in my description may correspond to M. ARYEPIGLOTTICUS mentioned by Dubois and Benham respectively in the larynx of B. Sibbaldii and B. rostrata, or the superior and inferior arytenoepiglottic muscle described by Carte and Macalister in B. rostrata, while the C-group seems not to have been specially remarked by previous authors. They have probably considered this group merely as a part of the thyreoarytaenoideus.

The THYREOEPIGLOTTIC muscle, which extends between the root of the epiglottis and the cranial border of the thyroid cartilage, makes also a part of the muscular walls of the laryngeal sack.

Innervation of the laryngeal muscles (fig.18):

Of all the laryngeal muscles in the Sei whale only the cricothyreoideus receives the external branch of the N. LARYNGICUS CRANIALIS. This nerve descends along the lateral surface of the larynx and, changing its course slightly mediad, crosses over the thyroid cartilage. It comes down along the lateral ridge of the cricothyreoideus, and then terminates there. The internal branch of this nerve runs through the thick laryngopharyngeal muscle and ends in the mucous membrane of the larynx.

The N. LARYNGIGUS CAUDALIS is considerably thick, ascends as the n. recurrens in the furrow between trachea and œsophagus, comes near to the inferior horn of the thyroid cartilage and, crossing over the here arising m. ceratocricoarytaenoideus dorsalis, passes under this horn, to reach the laryngeal sack, where it is divided into several branches, some of which innervate the m. cricoarytaenoideus and others enter into the thick walls of the sack, ending in its muscles. The ending occurs in such a way, that the uppermost branches course under the caudal border of the thyroid cartilage and attain the m. arytaenoideus transversus; the next branches go to the C- and D-group; the lowermost branches, running in the caudal direction, terminate in the superficial and deep layers of A- and B-group, and we can trace their small branches macroscopically up to the half level of the sack.

Thus it is certain that, as all the muscles of the walls of the laryngeal sack are innervated by n. laryngicus caudalis, A-group of muscles, in spite of its insertion to the cricoid and tracheal cartilages, does not belong to the cricothyreoideus.

# Sperm whale:

Extrinsic muscles

M. STERNOTHYREOIDEUS and M. THYREOHYOIDEUS: Connecting places of these muscles to the thyroid cartilage are shown in the accompaning figures (fig. 1'--a, b).

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M. LARYNGOPHARYNGICUS: Of this muscle the thyreopharyngeal part is well developed, but the cricopharyngeal part is not found at all.

M. HYOEPIGLOTTICUS: This unpaired muscle arises from the hyoid bone<sup>•</sup> (probably from its median part) and inserts to the ventral angle of the epiglottic cartilage.

M. PALATOPHARYNGICUS: The aryteno-epiglottideal tube, protruding into the choana, is encircled at the height of its neck by a muscular bundle, which lies within the choanal walls. This bundle, named by von Baer (1826) as the palatopharyngeal muscle, is well developed also in the Sperm whale. It must have an important meaning in relation to the function of the aryteno-epiglottideal tube. For, when it contracts, the communication between the naso-pharynx and the laryngeal cavity is shut out; at the same time the aditus laryngis must be closed completely. The meaning of such anatomical structures will be considered later.

# Intrinsic muscles

(1) M. CRICOTHYREOIDEUS: Stannius (1848) and Rawitz described this muscle in phocaena communis as a small muscular bundle; also Dubois and Macalister (1867) remarked the same in Hyperoodon and Globicephalus svineval respectively. But according to Benham, this muscle was invisible in Cogia. In the Sperm whale, too, I could not ascertain its existence. Whether it is quite absent in this whale, I want to determine by further researches.

(2) M. CRICOARYTAENOIDEUS DORSALIS: (fig. 6') This is very well developed. As in the Sei whale not only the dorsal surface of the cricoid cartilage but also the inferior horn of the thyroid cartilage gives rise to this muscle (m. ceratocricoarytaenoideus dorsalis). Its partial connection with the inferior horn was already reported by Stannius and Dubois, but Rawitz stated nothing about it.

I was much interested by the asymmetrical development of this muscle; namely it is very massive on the right side, but much poorer, though dilated considerably, on the left side. It must have some intimate relation with the asymmetry of the skull and nasal passage of this whale. (3) M. ARYTAENOIDEUS TRANSVERSUS: (figs. 6', 7') This muscle, connecting transversely the arytenoid cartilages of both sides, is well developed. Its lowermost fibres insert to the cranial border of the cricoid cartilage.

The caudal boundary of this muscle is distinct against the upper fibres of m. cricoarytaenoideus dorsalis, the latter covering the former to some extent. This fact coincides well with Dubois' description on B. physalus. As mentioned already, the cricoidal insertion of m. arytaenoideus transversus is also present in the Sei whale: This relation, which according to Fürbringer (1875) never exists in the human larynx, seems to occur in the whales not rarely.

(4) M. CRICOARYTAENOIDEUS LATERALIS: This muscle was described by Stannius in Phocaena, by Murie in Globicephalus melas and by Benham in Cogia. But Meckel and Fürbringer denied the existence of it in Delphinus delphis. Watson and Young did not mention this muscle in Beluga leucas (cited from Dubois). Denying its existence in Hyperoodon and in other toothed whales, Dubois said that the so-called m. cricoarytaenoideus lateralis described by Stannius and Murie is nothing but a ceratal portion of the cricoarytaenoideus dorsalis. In the Sperm whale I too can not find this muscle.

I want to mention here my finding of a probably new muscle, which arises from the INNER surface of the body of the cricoid cartilage and inserts to the INNER surface of the arytenoid cartilage (fig. 9'): Though its attachments to the cartilages are not very firm compared with other muscles, it is certainly a special muscle clearly discernible from the surroundings. Its nature cannot be determined at present, but the fact itself is interesting that such an unusual cricoarytenoid muscle exists, deserving the name, "m. cricoarytaenoideus internus". Moreover there are several muscular fibres scattered on the inner surface of the thyroid cartilage, and I wondered whether the muscle above mentioned would be nothing but a collection of these fibres. But this assumption is not very probable, for the internal cricoarytenoid muscle is a bundle so distinctly isolated.

(5) M. THYREOARYTAEEOIDEUS: (figs. 7', 8') This is a massive muscular bundle, which arises from an area along the caudal notch of the thyroid cartilage and inserts to the ventral and lateral surface of the muscular process of the arytenoid cartilage. As to locality and form, this muscle is not so unusual as in the Sei whale, for the laryngeal sack is absent in this whale.

(6) M. THYREOEPIGLOTTICUS and M. ARYEPIGLOTTICUS: Though Rawitz denied the existence of these muscles in Phocaena communis, Benham

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found them in Cogia. In the Sperm whale I see no muscle fibres, which may deserve the names. The upper portion of the m. thyreoarytaenoideus comes in contact with the petiolus epiglottidis, but shows no direct connection to this. Absence of these muscles might be explained from the very firm ligamentous connection between epiglottis and thyroid cartilage, where almost no mobility seems to remain.

# III. The laryngeal cavity

Sei whale:

The larynx of the Sei whale does not exhibit a tubal structure as in the toothed whales. As shown in fig. 10, the aditus laryngis of the Sei whale differs in its general form not much from that of the human being. Namely, the tongue-shaped epiglottis projects from the ventral surface of the pharyngeal cavity, with its apex directed somewhat upwards (about 12 cm high). From its lateral ridge on each side a fold of mucous membrane (plica aryepiglottica) is stretched, encircling the laryngeal aperture, dorsally and downwards to the arytenoid cartilage. The cranial processes of the arytenoid cartilages are connected between both sides by a part of mucous membrane; the apex, though projecting upwards a little, is situated much lower than that of the epiglottis.

The laryngeal cavity is in this whale a relatively narrow space surrounded by the paired arytenoid cartilages and the single cricoid cartilage behind. The mucous membrane, covering the inner surface, is generally smooth, but has a few fine grooves. This cavity passes downwards behind the caudal processes of the arytenoid cartilages, which are also connected between both sides, and continues further with the tracheal cavity.

The laryngeal sack (figs. 11, 13-17, 18):

As related before, the laryngeal cavity has in the Sei whale a large sack-formed appendage, which is to be named "saccus laryngis ventralis". It is on the ventral wall of the larynx, elongated downwards through the whole length of the trachea, the ventral clefts of cricotracheal cartilages offering a space just fitted for this sack. The constituents of the thick muscular wall of this sack were already mentioned in the precedent chapter. The cavity within the sack is elongated longitudinally and communicates with the proper cavum laryngis through a slit along nearly the whole length

of the arytenoid cartilage. The inner surface of the sack has much folds and grooves, as shown in fig. 11. There are many longitudinal folds in the upper part, while the lower parts show many reticulated grooves and the meshes are especially fine on the dorsal wall. We see many granular prominences here on the mucous membrane. In the ventral parts the folds and grooves are considerably rough. Judging from this structure, it seems doubtless that this sack is liable to extension and contraction in the living whale.

It is naturally important to know the function of this highly contractile laryngeal sack. I will consider this problem at first historically.

The honour of the discovery of the laryngeal sack is ascribed to J. Hunter, as this great anatomist stated in his study on Balaenoptera rostrata, that a sack is present on the ventral surface of the larynx.

Since then the laryngeal sack has been observed by Eschricht and Reinhardt (1866) in Balaena mysticetus and by Carte and Macalister, and Benham in Balaenoptera rostrata. Beauregard and Boulart (1882), who examined the laryngeal sack in Balaenoptera musculus, B. physalus and Balaena antipodum, noticed the difference that the sack in question is far smaller in Balaenidae than in Balaenopteridae. In the Sei whale Schulte (1916) touched this sack briefly.

The fact that this laryngeal sack is present only in the whalebone whales, was indicated for the first time by Eschricht and Reinhardt. Dubois, comparing between B. physalus and several kinds of toothed whales, ascertained this. Rawitz, too, mentioned this fact, when he studied the larynx of Phocaena communis.

In the meantime the problems, what is the anatomical nature of the laryngeal sack and why this sack exists only in the whalebone whales, have been very differently answered. Dubois regarded this sack as homologous with the Morgagni's ventricle. He came to this opinion by comparing the laryngeal structure of toothed whales, because these animals have no laryngeal sack on the ventral wall of the larynx, but have a recess on each side of the median longitudinal membranous fold; and he explained this recess as corresponding to the Morgagni's ventricle. Dubois thought that the laryngeal sack of whalebone whales is formed by the further enlargement of the recess, extension of its walls and by development of the thyreoarytenoid muscle.

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But Benham doubted the homogeneity of the laryngeal sack of whalebone whales with the laryngeal recess of toothed whales, though he accepted the correspondence of the recess with the Morgagni's ventricle. Such a problem is very difficult to decide, for the two structures, both of which are appendages of the laryngeal cavity, differ very much from each other especially in their relation to muscles and cartilages. On the other hand, there are undeniable communities between them, in spite of the differences in size, form and structure of the walls.

A key for settling the problem may probably lie in the fact, that the laryngeal sack for Balaenidae and Megapteridae is not so large as that of Balaenopteridae, showing, so to speak, an intermediate form between the laryngeal recess and the laryngeal sack. I myself ascertained the smallness of the laryngeal sack in a fetus of Megaptera nodosa.

Now, on the function of the laryngeal sack there seem to be three possibilities as follows.

First, it may be possible that this is an apparatus for preventing the entrance of water and food into the respiratory canal, as the contraction of massive muscles of this sack will make the laryngeal sack, at the same time the larynx itself firm and solid, so as to avoid mis-swallowing of a large quantity of food with water into the larynx and trachea. Rawitz, for instance, stated this opinion. Prof. Ogawa has also an opinion near but a little different to this, for he thinks that the blow of air produced by contraction of the laryngeal sack will prevent the entrance of water and food into the respiratory canal.

The second possibility is, that this sack may concern with complete utilization of oxygen in the inspired air. Schulte said in his paper on a fetus of the Sei whale, "that by its contraction and relaxation during submergence, a circulation of air in the wide trachea and bronchi might be set up, which would favor the absorption of oxygen by bringing the air in these passages more rapidly into contact with the respiratory membrane than could be done by the usual diffusion currents". Another very interesting opinion related to the second possibility was stated by Negus in his book titled "The Mechanism of the Larynx", that all the air-sacs communicated with the respiratory canal of animals must be for the "rebreathing of air". To explain the "rebreathing of air", he took up the frog as an example, saying "a frog kept under water for a prolonged period expels air

from its lungs into its mouth and pharynx; the used-up air which has been in contact with the pulmonary epithelium mixes with the relatively unused air which has lain in the centre of the sac-like lungs. This mixed air, when blown back into the lungs, gives up a fresh supply of oxygen and takes up  $CO_2$ , so that respiration can be continued for a time without the intake of a fresh supply of air". According to him, the laryngeal air sacs not only of the aquatic animals including sea-lion, whales etc., but also of the terrestrial animals are for this purpose.

The third possibility is the relation of this sack with the phonation. The Sei whale has no vocal cord in the larynx; its absence is also proved in other whalebone and toothed whales. And so, it is generally believed that the whales produce no voice. Some authors however insisted upon voices of whales (Schneider, 1795; Murie; Rawitz). Turner (1872) explained the mechanism of phonation in Balaenidae as such, that the elongated caudal processes of the arytenoid cartilages are drawn near to each other and vibrate by the strong exspiration of whales. This opinion was accepted by Weber and Dubois, and was, though denied by Rawitz, adopted again by Göppert in Bolk's "Handbuch der vergleichenden Anatomie" (1937). If such a mechanism of phonation be true, we must hold in mind the utility of the laryngeal sack in this function.

At present it is impossible to determine, which of these three be the fact. Moreover the truth may lie outside of them.

Relation of the laryngeal sack with the internal structure of the brain (fig. 19):

While the Sei whale has a large laryngeal sack, the Sperm whale has not such a sack. Probably relating to this difference, a remarkable different structure was found in the medulla oblongata of whales. Namely, the nucleus ambiguus is poorly developed in the pigmy Sperm whale, Cogia breviceps (Fig. 12'), while the same nucleus shows in the Sei whale such a striking development that the name "ambiguus" is very unsuitable for this animal. Prof. Ogawa (1948) has directed his attention for the first time to this fact and ascribed the extraordinary development of this nucleus in the Sei whale to the large laryngeal sack. For the massive muscular sheets of this sack are innervated by the vagus and it is generally believed that the nerves innervating laryngeal muscles arise from the nucleus ambiguus.

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To make this problem more definite, we must examine in the future the brains of Megapteridae and Balaenidae, for the laryngeal sack of them is, as already mentioned, much smaller than that of Balaenopteridae, but larger than the laryngeal recess of toothed whales.

### Sperm whale:

The entrance to the larynx is in this whale a transverse fissure at the top of the aryteno-epiglottideal tube, which protrudes into the choana and is encircled at its neck by circular fibres of the palatopharyngeal muscle. The larynx of the Sperm whale shows therefore the well known characteristic of various toothed whales (figs. 7', 10').

Figure 11' shows the inner surface of the laryngeal cavity. In this sketch the whole larynx except the epiglottic cartilage is cut in the median plane and the right half is removed. The cavity communicates above with nasal cavity through the laryngeal aperture and below with the tracheal canal, while the greater part of this cavity belongs to the aryteno epiglottideal Along the median line of the ventral wall, we see a longitudinal tube. elevation covering the median crest of the epiglottic cartilage. The mucous membrane of the laryngeal cavity has numerous fine furrows and folds. Very fine furrows run longitudinally in the upper half of this cavity and between them many minute points or holes are seen in rows. The mucous membrane, which covers the medial surface of the arytenoid cartilage is quite smooth, lacking of furrows. In the lower parts, near the trachea, there are fine reticular furrows and folds, while the part covering the median crest of the arytenoid cartilage has oblique or looped furrows.

From the caudal end of the median crest a membranous fold (plica mediana) runs to the cranial and ventral border of the cricoid cartilage and we see here a triangular recess on each side of this fold. The furrows are reltively rough in this part and many lacunae are present, which seem to be apertures of the lymphatic nodules.\*

This triangular recess was already referred to in relation to the laryngeal sack. The opinion that this recess would correspond to Morgagni's ventricle in other mammals was stated for the first time by Murie in his

<sup>\*</sup> The inner surface of the laryngeal sack of the Sei whale does not show such lacunae, but I found many relatively larger lacunae on the ventral surface of the membrane connecting the upper ends of the cranial processes of the arytenoid cartilages of both sides. (fig. 12)

paper on Globicephalus melas. Watson and Young published the same opinion for Beluga leucas (cited from Dubois) and Dubois also accepted it.

Physiological meaning of the aryteno-epiglottideal tube:

The opinion has been prevalent, that the toothed whales can swallow and respirate at the same time owing to this peculiar structure, for the alimentary and the respiratory canal can be in this way completely separated from each other. This opinion seems to have its origin in Camper (1820), and was supported by Milne-Edward (1860), Gegenbaur (1891), Zuckerkandl (1898) (cited from Boenninghaus) and Negus (1928). But it had not so definite basis and was opposed to by Boenninghaus from anatomical and physiological reasons. According to him, all the mammals must dilate its pharynx at deglutition and simultaneously the larynx must be shut off as a reflex by contraction of the laryngeal muscles. My observations upon the larynx of the Sperm whale justify more the opinion of Boenninghaus. I want to suggest, that, when the palatopharyngeal muscle is contracted at deglutition to avert the entrance of food and water into the choana, the laryngeal aperture also must be closed, so there would occur no respiratory action. Besides, I suppose, the Sperm whale would have no need to be able to swallow and respirate at the same time, because this whale used to take its food in the depth of sea. Taking in mind the high pressure of water there, it seems very probable that such a tubal apparatus is exclusively to avoid the mis-swallowing of food and water into choana and larynx, rather than to swallow and respirate simultaneously.

Another somewhat different opinion was offered by Huber (1934), who stated in his paper on the nasal passage of Tursiops, that the palatopharyngeal muscle around the aryteno-epiglottideal tube, together with the m. maxillonasolabialis around the blow-hole and the muscular plug in the osseous nasal passage, belongs to the blow-hole mechanism, which performs the task to shut the nasal passage closely, so as to avoid the entrance of water into lungs, when the whale submerges into the depth of sea. At any rate, it is quite reasonable, also from the standpoint of dynamics, to close the laryngeal aperture by contracting the neck of this tube by the palato-pharyngeal muscle.

As an appendix to this work I wish to say briefly about the Phonation

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## of Whales.

Neither the Sperm whale nor the Sei whale has the vocal cord in the larynx. Absence of the vocal cord in whales has been written by many authors, for instance by Meckel for Delphinus and Phocaena, by Cuvier, Stannius, Milne-Edward etc. for various kinds of whales, and Hunter insisted that the whales do not possess the ability of crying. But Schneider stated, in opposite to Hunter, that he heard from a "Grönlandfahrer" about crying of whales and Murie said in his paper on Grampus rissoanus, "that some species of whales utter sound is now unquestioned, notably the species under consideration and the ca'ing whale (Globicephalus melas)".

How can the whale then make sound without the vocal cord? Murie thought, taking the membranous folds above mentioned- two folds are said to exist in the larynx of Grampus- as the substitute for the vocal cord, that the forcible blow or exspiration of the whale would vibrate these folds, producing sound. He added furthermore that in some animals, for instance, Hyomoschus aquaticus and Saiga tartaria, the vocal cords are placed nearly vertically on the sides of the larynx, similarly as in the cetacean larynx.

Concerning the whalebone whales, Milne-Edward (1876) said that they would be dumb, but Turner, Watson, Young and Dubois accepted the possibility of them to make sound. They explained that the caudal process of the arytenoid cartilage would be vibrated by the exblows of the whale, producing thus voices. Dubois added furthermore that perhaps a similar modus of phonation is possible also for Otaria gillespii. Though Rawitz mentioned, opposing to Turner etc., that it is impossible for a cartilaginous substance to vibrate and produce voice, yet he believed the ability of whales to make voice, especially of Megaptera boops. According to him, the organ of phonation is in the whales not the arytenoid cartilage but membranous folds formed temporarily in the larynx or in the soft palate, especially during the rutting season.

So far I have reviewed the opinions and possibilities published by various authors on the phonation of whales, but, regret to say, that the final solution of this problem is far beyond us. But it seems doubtless that some dolphins can make sound, though it might be faint. Of the whalebone whales I have not been told by any whaler that he heard the voice of whales. During my Antarctic whaling trip (1947-1948), I had no

opportunity to experience the voice of whales. Even when they were hurted by the harpoon, they did not cry at all. If they could produce voice, I supposed, judging from the situation, that they would make voices in such cases. Of course it is not impossible that the whales produce sound and communicate between them especially during the rutting season. Even at that time the sound might be so faint that it does not deserve to be called "voice"; but it would be sufficient for the communiction between them; the vibration will be propagated in the water and received by their sensitive auditory organs. If so, there is no need for us to seek for any special vocal organ in the larynx, for such a vibration could be produced without the vocal cord from other parts of the body. At any rate it would be necessary to know the nature of the voice of whales, its relation to the breeding season and so forth.

IV. Comparison of the larynx between the Sei and the Sperm whale From the foregoing description I will point out the remarkable differences in the larynx of the Sei and the Sperm whale.

•	Sei whale	Sperm whale		
Thyroid cartilage	Relatively small, without the median angle.	Of a large size, with a remar- kable median angle.		
Epiglottic cartilage	Relatively small.	Well developed and elongated upwards		
Arytenoid cartilages	Caudal process is well deve- loped and its apex is connected with that of the other side.	Cranial process is continuous between both sides at the upper end. The caudal process is absent.		
Cricoid cartilage	Ventral half is widely lacking and continuous downwards to the tracheal cartilages.	Completely ring-formed.		
Tracheal cartilages	Ventral parts are widely lacking.	Completely ring-shaped.		
Thyreoarytenoid muscle	Long and wide, forming the walls of the laryngeal sack.	An ordinary muscle bundle.		
Laryngeal sack	Of very large size.	Lacking, but provided with a small recess on each side.		
Aryteno-epiglottideal tube	Lacking perfectly.	Well developed, typical for the toothed whales.		
Asymmetry of the laryngeal structure	Almost symmetrical.	Somewhat asymmetrical (for example, development of m. cricoarytaenoideus dorsalis).		

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From this table, it may be concluded that the most important difference lies in the remarkable development of the laryngeal sack of the Sei whale and in the typical aryteno-epiglottideal tube in the Sperm whale. For, almost all of other differences are more or less in relation to these features, which are at the same time the characteristic respectively of the whaleboneand of the toothed whales.

As the last problem, I wish to consider, why such a tubal structure of the larynx exists only in the toothed whales? And why is the laryngeal sack present only in the whalebone whales? These problems, I suppose, cannot be answered merely from the functional standpoint, but are concerned in a high degree also with the phylogenetic relation. In other words such remarkable differences in the larynx between toothed and whalebone whales is not to be explained solely as acquired characteristics during the aquatic life.

The interrelationship between these two groups of whales does not seem to be so close, and the resemblance of their external form might be rather explained as a convergence of organisms; for, the inner structures of them are, in some points, too much different to be regarded as intimately related. Concerning the relationship between the toothed and the whalebone whales, Dubois remarked that "der Larynx der Odontoceti steht dem ursprünglichen Typen näher, während derjenige der Mystacoceti sich weiter Rawitz opposed to this opinion and said, "dass Odontoentfernt hat". und Mystacoceten-larynx sich nicht aus einander entwickelt haben können, dass der erstere nicht die Vorstufe des letzteren darstellt, dass aber das umgekehrte Verhältnis nicht statthat. Und daraus folgt, dass sie aber an der Wurzel nicht einander zusammenhängen, nicht direkter Verwandtschaft mit einander stehen", and I myself agree with Rawitz's opinion. Pütter (1902) also stated approximate ideas in his comparative anatomical work upon the eyes of marine mammals.

## Summary and Conclusions

- 1) As to the laryngeal cartilages and muscles, many remarkable differences are found between the Sei and the Sperm whale.
- 2) The larynx of the Sei whale shows the most noteworthy characteristic in the presence of an enormous muscular sack on its ventral side, saccus laryngis ventralis, and almost all of the peculiarities of its cartilages and

muscles are more or less concerned with this laryngeal sack.

- 3) Four muscle-groups (A, B, C, D) were classified in the muscular walls of the laryngeal sack, with considerations upon their homogeneity with the laryngeal muscles of other mammals.
- 4) These muscle-groups of the laryngeal sack are all innervated by the caudal laryngeal nerve.
- 5) The extraordinarily well developed nucleus ambiguus in the medulla oblongata of the Sei whale seems to be intimately related with the action of this laryngeal sack, which is probably able to contract and dilate in a considerable degree. The corresponding nucleus is not well developed in the toothed whales, which lack of the laryngeal sack.
- 6) The general characteristic of the larynx of the toothed whales, including the Sperm whale, lies in the upward elongated epiglottis and arytenoid cartilage, which form altogether the aryteno-epiglottideal tube.
- 7) Some asymmetrical relations were noticed in the larynx of the Sperm whale, for instance in the development of the dorsal cricoarytenoid muscle and in the relative position of oesophagus to the larynx. These asymmetries may be related with the extraordinarily asymmetrical nasal passages of this whale.
- 8) With the completely ring-formed cricoid and tracheal cartilages, the Sperm whale is, together with Cogia and Tursiops, an exceptional case in the realm of Odontoceti, for these cartilages have a median cleft in the ventral portion in almost all of the toothed whales.
- 9) The lateral cricoarytenoid muscle was not found at all in the Sperm whale, but I saw a muscle bundle, probably not yet known, deserving the name "m. cricoarytaenoideus internus".
- 10) The remarkable differences in the larynx between the whalebone and toothed whales cannot be explained merely from the adaptation of these whale to the aquatic life, but must be rather refered to the phylogenetic relation; namely, I believe, coincidently with some previous authors, that the two groups of whales, whaebone and toothed whales, are not so intimately kindred to each other as generally assumed.

# Explanation of figures:

## Sei whale

- Fig. 1, Thyroid cartilage
- Fig. 2, Epiglottic cartilage
- Fig. 3, Cricoid and tracheal cartilages
- Fig. 4, Arytenoid cartilage
- Fig. 5, Larynx (ventral view)
- Fig. 6, Larynx (dorsal view)
- Fig. 7, Larynx (dorsolateral view)
- Fig. 8, Larynx (the ventral right quarter is removed)

Fig. 9, Median section through the larynx

- Fig. 10, Laryngeal aperture
- Fig. 11, Laryngeal sack (inner surface)
- Fig. 12, Lymphatic lacunae on the ventral surface of the membrane connecting the cranial processes of the arytenoid cartilages
- Fig. 13, Horizontal section through the larynx (1)
- Fig. 14, Horizontal section through the larynx (2)
- Fig. 15, Horizontal section through the larynx (3)
- Fig. 16, Horizontal section through the larynx (4)
- Fig. 17, Horizontal section through the larynx (5)
- Fig. 18, Innervation of the laryngeal muscles

Fig. 19, Nucleus ambiguus

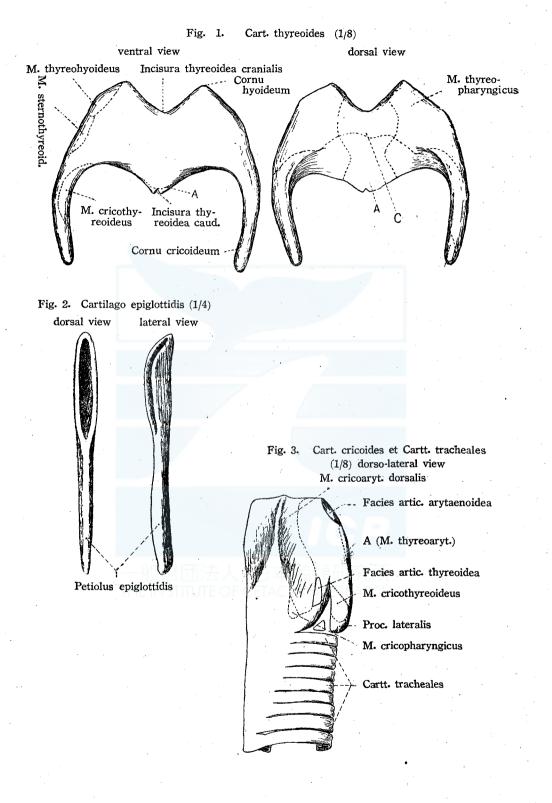
#### Sperm whale

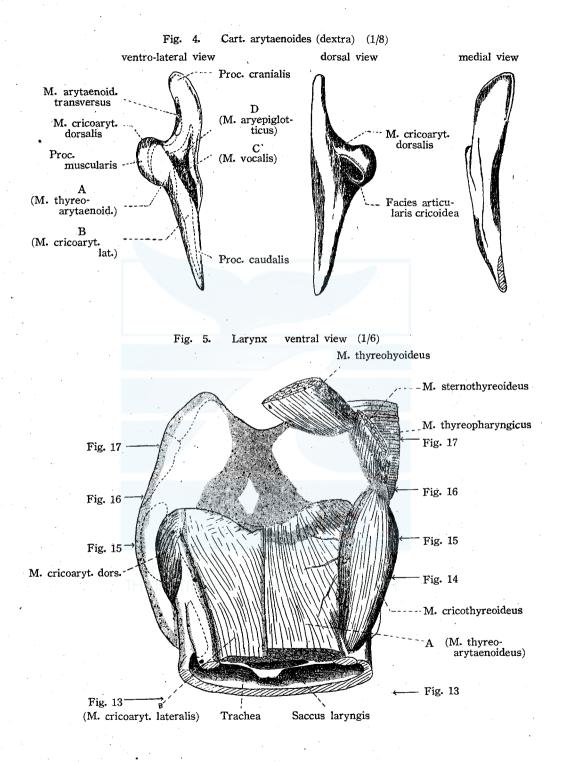
- Fig. 1', Thyroid cartilage
- Fig. 2', Epiglottic cartilage
- Fig. 3', Cricoid cartilage
- Fig. 4', Arytenoid cartilage
- Fig. 5', Tracheal cartilages
- Fig. 6', Larynx (dorsal view)
- Fig. 7', Larynx (dorsolateral view)
- Fig. 8', Larynx (lateral view, the right lamina of the thyroid cartilage is removed)
- Fig. 9', M. cricoarytaenoideus internus
- Fig. 10', Laryngeal aperture
- Fig. 11', Laryngeal cavity
- Fig. 12', Nucleus ambiguus of the pigmy Sperm whale (Cogia breviceps)

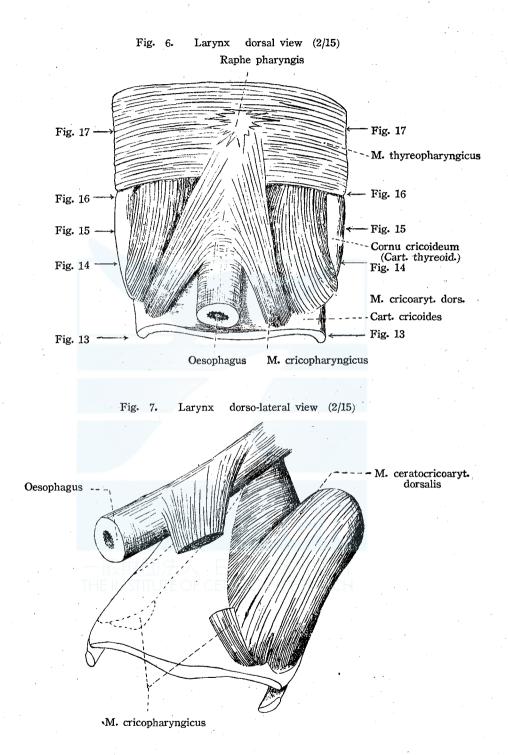
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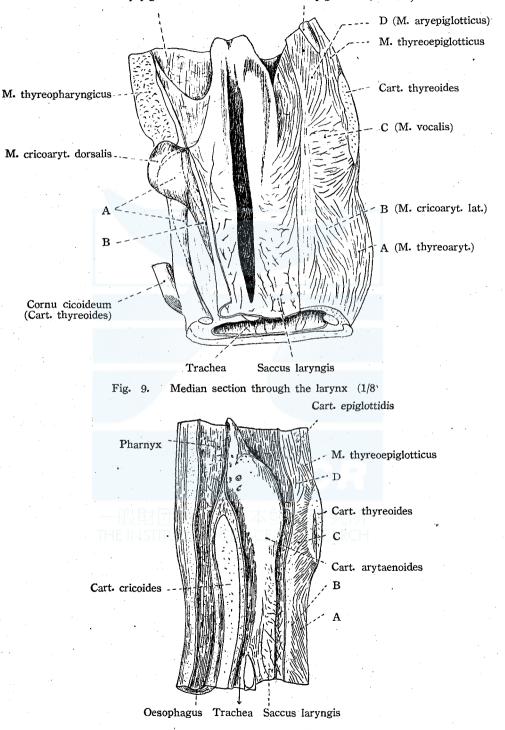
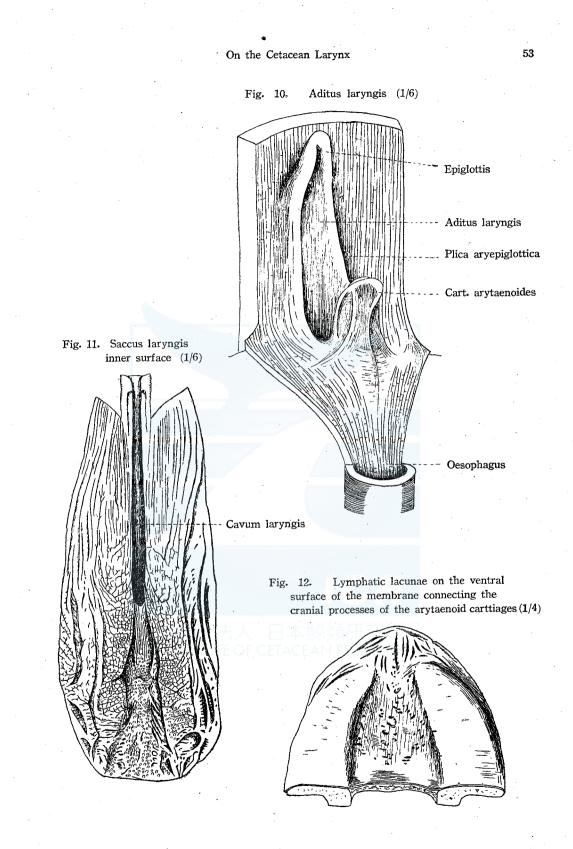
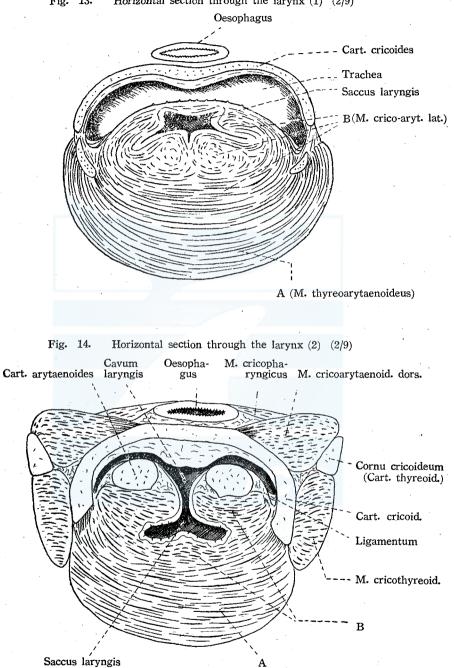
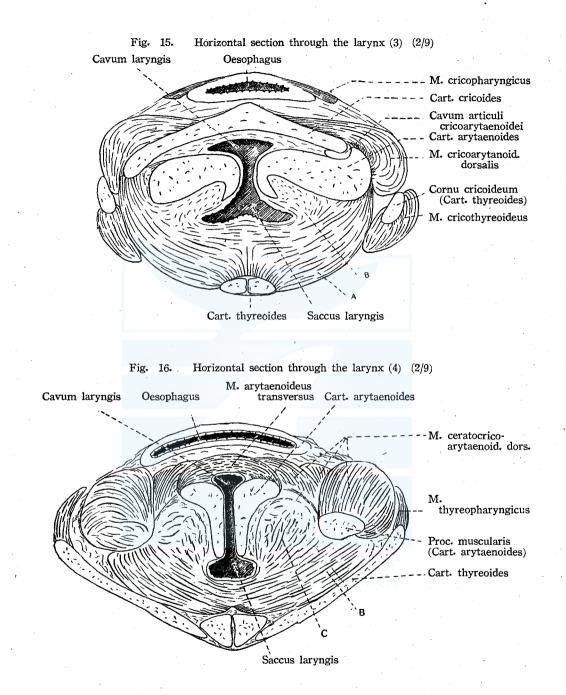


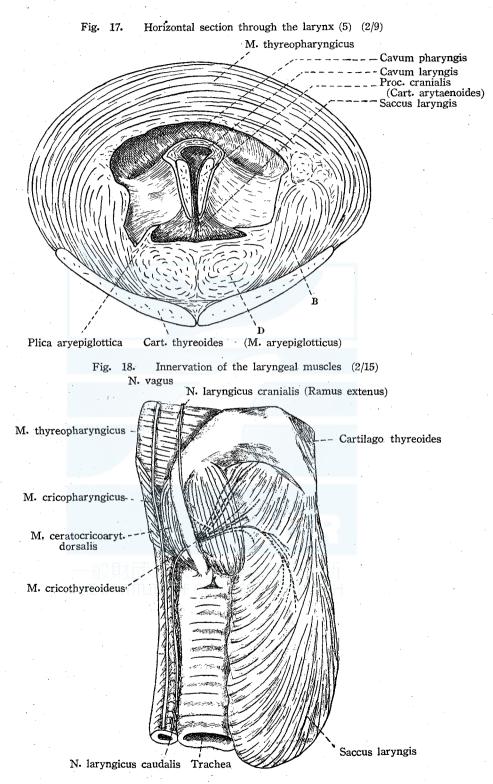
Fig. 8. Larynx (the ventral right quarter is removed) (2/15) Plica aryepiglottica Cart. epiglottidis (Petiolus)

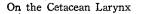


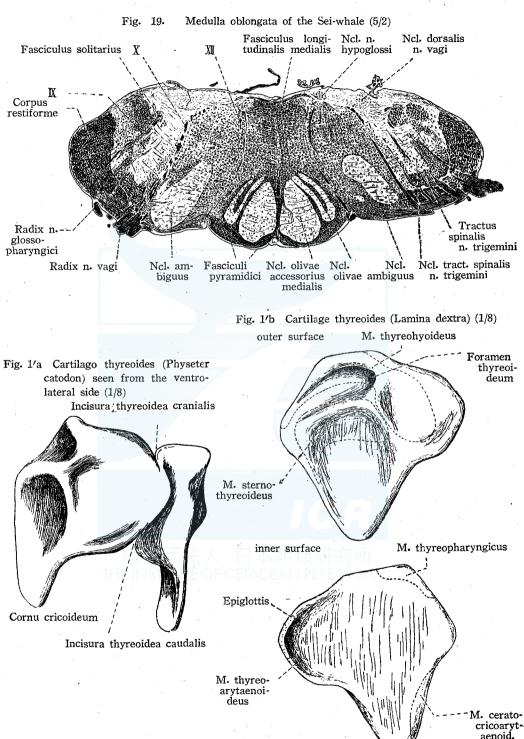


Horizontal section through the larynx (1) (2/9) Fig. 13.



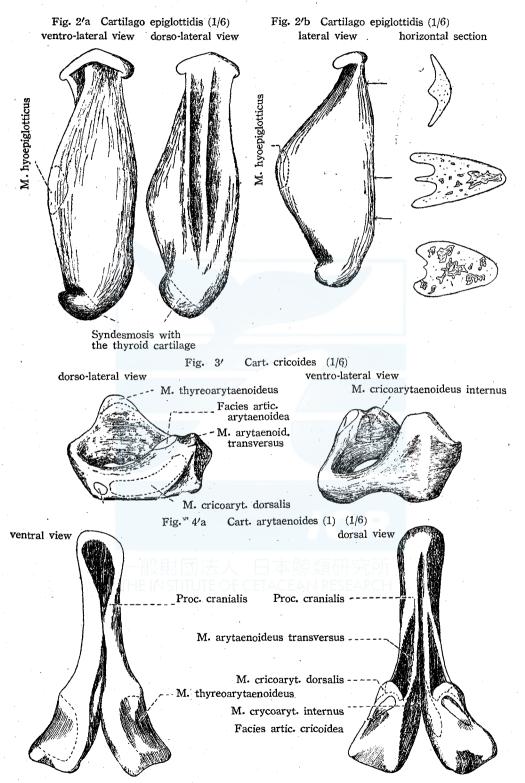


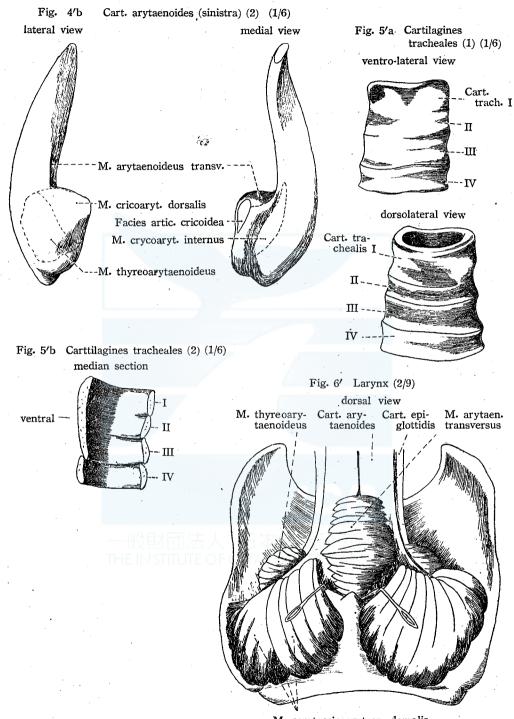




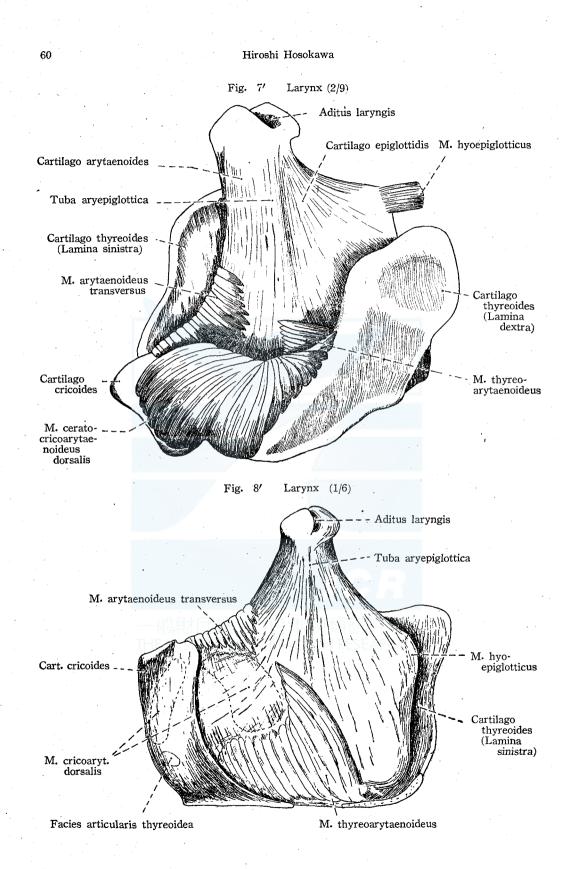
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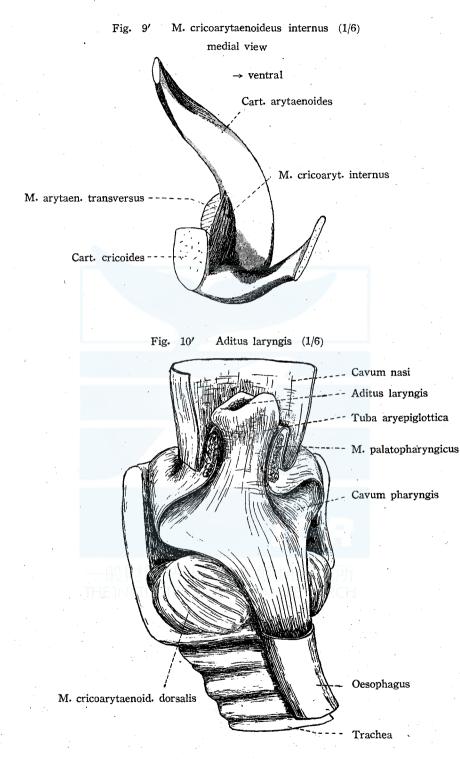
aenoid. dorsalis

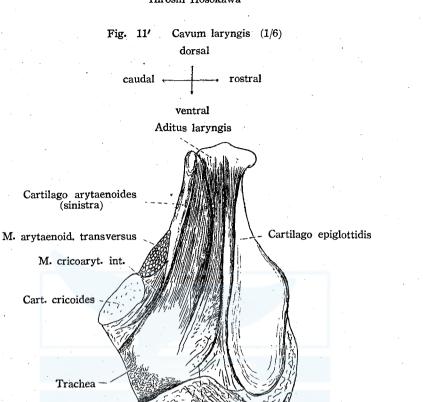




M. ceratocricoarytaen. dorsalis

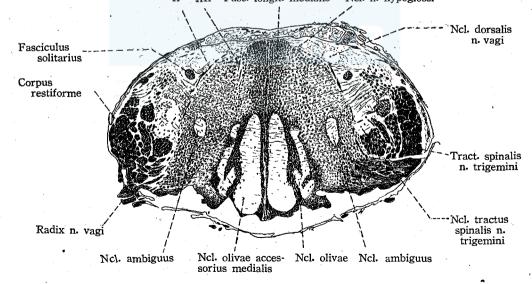






Cart. cricoides // / -- Cart. thyreoides M. thyreoarytaenoideus Plica Recessus mediana laryngis

Fig. 12' Medulla oblongata of the pigmy Sprm-whale (10/3) X XII Fasc. longit. medialis Ncl. n. hypoglossi



#### Hiroshi Hosokawa

# Bacteriological Studies on Freshness of Whale Meat

## (Report No. 1)

# Tomoichiro Akiba, Takeshi Tsuchiya, Makoto Umehara and Yoshiharu Natsume

In using whale meat and organs as food stuff and material of medicaments the value of their utilization depends to a large extent on their freshness. Consequently, it is of great importance to take necessary steps preventing decrease in freshness, and in other words the factors causing deterioration must be clear. However, the research along this line has been insufficient. In order to study these points it is necessary to investigate the various factors such as whether this is due only to self-digestion or autolysis after death of whale, or besides what route bacteria take to be involved in this, or what kind of bacteria invade when and how, if such bacteria are involved in this. We began this research in the summer of 1947 with this object, but our material was very scarce in 1947 due to the passed fishing season, and so these results of 1947 and a part of 1948 will be consolidated and reported as Report No. 1.

## **Experimental** Results

1. Collection of Samples. Trips were made twice, once in August, 1947 and another in July, 1948 to Ayukawa-machi which is located at the tip of Ojika Peninsular. The determination of various kinds of bacteria, putrefaction tests, etc. in this report were carried out on these samples taken from one whale which was dissected in August 1947. As materials for our tests, its blood, muscles, contents of small and large intestines and sea water were used. Blood which flowed out from the heart during dissection, was collected with a sterilized pipette. Consequently, since dissection was not carried on under sterilized condition, the invasion of bacteria from the environments cannot be absolutely refuted. However, since a sterilized pipette was inserted deeply in a copious flow of blood, it may be assumed that bacteria contained in the blood itself were collected. As to meat. about 20 c. cm. of meat lump was cut off, placed in boiling water and boiled for about 3 to 5 minutes. After sterilizing the surface of muscle tissue it

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was cut open with a scapel, and 2 or 4 gm. of muscle tissue was cut off from the center. To this, 8 or 2 cc. of physiological saline solution was added and stirred well. After standing only the meat juice was taken and used in the experiments. As to the contents of the intestines, these were cut open and the contents were collected with a sterilized pipette. This was diluted to five times with physiological saline solution and used in the experiments. The intestines were also swollen due to gas produced by fermentation of the contents at that time. Sea water was collected in front of the works and from the bay.

As culture media sea water agar media, meat infusion agar, Endo's media and cysteine glucose agar were used. Sea water agar media were used for the purpose of not losing specific sea water bacteria, cysteine agar media for anaerobic bacteria, and Endo's media for the isolation of colon type bacteria. Sea water agar media were made according to Zo Bell's method, and 0.5% of peptone, 0.01% of ferric phosphate and 1.5% of agaragar were disolved in 1000 cc. of sea water being aged before the use.

2. Freshness and Number of Bacteria. Number of bacteria detected in blood and muscle was almost proportional to freshness and the number of bacteria also increased as the time elapsed longer after catching. However,

			A 1 Sperm	B 1 Sei	B 2 Sei	B 3 Sei	B 4 Sperm
	1		Whale	Whale	Whale	Whale	Whale
D	Date Whale Caught			July 25	July 25	July 25	July 27
T	ime of elar	se after catching (hours)	30	34	26	68	24 Fresh (80%)
F	reshness		Satis- factory (50%)	Fresh (80%)	Fresh (80%)	Satis- factory (50%)	
ia	Blood per ml.	Meat influsion agar media Sea water agar media	Innume- rable Innume- rable	70,000	12,000	9,360,000	23,400
Bacteria	Muscle per gm	Meat influsion-agar media Sea water agar media	4,575 4,425	9,400	8,200		
Number of	Large In- testine contents per ml.	Meat influsion-agar media Sea water agar media	Several 0		•		
Nu	Small In- testine contents per ml.	Meat influsion-agar media Sea water agar media	Several Several				
pH Blood Muscle			6.2 6.2	6.2 5.8	5.9 6.0	5.6 6.1	

ria 🛛

Note: Freshness was expressed by the visual judgement of experts.

it was not clearly recognized that there was some relation between freshness and hydrogen ion concentration in blood and muscle at the time of dissection, but all the reactions indicate the progressing decomposition of the sugar.

3. Indentification of Isolated Bacteria. In this paper are reported only the results of experiments as to the samples collected from a sperm whale dissected in August 1947. Result of another samples collected in July 1948 shall be reported in the second report. Identification of various bacteria was attempted as to bacteria isolated at the works and also bacteria isolated after bringing samples to our laboratory. Even the bacteria obtained by anaerobic culture grow better by aerobic culture, after successive transfer on culture media, so all the succeeding examinations of biological characters were made by aerobic culture.

Microscopic examinations were made as to the pure culture of the 57 strains to determine their shape, grouping, Gram's stain, size, motility, etc. and furthermore 19 species were selected according to shape, form and color of colonies on the media such as meat infusion agar, sea water agar media and cysteine glucose culture, and gas production, liquefaction of gelatin, hydrogen sulphide production and indol reaction. As to 19 strains the further examinations were made as following table (Table 2).

Of 19 strains another three were same strains, so that 16 species were identified according to Bergey's Manual of Determinative Bacteriology and other literatures. It was possible to determine 9 different Micrococci, 1 Streptococcus foecalis, 2 Flavobacteria and 4 Achromobacters.

These bacteria may generally be found in soil, air, water, etc., and although there may be few cases in which their biological characteristics do not exactly coincide with those found in references, but the principal ones coincide exactly and we could not find new species. In a case as Micrococcus epimetheus, there is a question more or less compared with the place primarily found, but there is not any reason that such bacteria must not be found in places other than where they were found, so that it may be assumed that the presence of such bacteria is probable.

4. Putrefaction Tests. When these bacteria were inoculated on pieces of whale meat and left standing at 30°C, 8 strains produced rancid irritating or light deterioration odor after 18 hours, and 14 strains after 48 hours, and the same time pH trended to alkaline.

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Also, in case several bacteria being grouped according to the origines where they are found, are inoculated on the surface of a piece of meat, the rancid odor is produced faster than in the previous cases and appears after 6-12 hours.

No. of Stra	Time	Before	After 4 hrs.	6	12	18	24	48	26
F	19	6.2	6.1	6.0	6.0	6.3	6.8	6.9	7.7
$\mathbf{F}$	88	6.2	6.2	6.0	6.0	6.4	7.0	7.4	++8.2
F	15	6.3	6.1	6.0	6.0	+6.0	+5.9	#6.6	#8.2
$\mathbf{F}$	17	6.2	6.1	6.3	6.3	6.3	6.4	6.5	<del>++</del> 7.0
F	5	6.3	6.2	6.2	6.2	±6.4	+7.0	+7.4	##8.0
$\mathbf{F}_{i}$	6	6.2	6.2	6.0	6.0	6.0	6.4	#6.8	₩7.6
$\mathbf{F}$	1	6.2	6.2	6.1	6.1	$\pm 6.4$	+6.8	#6.8	++7.2
$\mathbf{F}$	65	6.2	6.2	5.9	5.9	6.2	6.4	$\pm 6.6$	+6.8
F	66 ·	6.2	6.2	6.0	6.0	6.1	6.3	#6.3	+6.4
$\mathbf{F}$	12	6.2	6.2	6.0	6.0	+6.0	+6.4	+6.4	+6.6
С	32	6.2	6.2	6.1	. 6.1	6.1	6.5	$\pm 7.6$	<del>   </del> 8.0
С	33	6.2	6.1	6.3	6.3	6.1	6.0	6.1	++7.0
С	34	6.2	6.2	6.0	6.0	+6.0	+5.7	+6.2	₩6.8
В	24	6.2	6.2	6.0	6.0	$\pm 6.4$	+6.8	+7.4	++8.0
в	82	6.2	6.2	6.0	6.0	6.6	7.0	+7.4	<del>  </del> 7.7
I	43	6.2	6.2	+6.2	#6.3	#6.6	<del>  </del> 6.9	₩7.8	₩8.0
I	44	6.2	6.2	6.2	6.2	6.0	5.8	5.8	6.6
I	45	6.2	6.2	6.0	6.0	6.0	6.2	5.8	+6.0
S	90	6.2	6.2	6.0	6.0	6.0	5.6	±6.7	+7.4
S	97	6.2	6.2	6.0	6.0	6.0	6.3	$\pm 6.5$	++7.0
Con	trol	6.2	6.2	6.2	6.2	6.2	6.3	6.3	6.6

Table	3.	Deterioration	Test,	pH	aod C	)dor
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Note:	+~#	indicate	the	degreee	of	deterioration	odor.
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Time Bacte- rial groups	Before	After 4 hrs.	6	12	18	24	48	Oder
F group	6.2	6.2	6.4	+6.4	+6.8	<del>  </del> 7.8	#6.2	Putrefied odor
C group	6.2	6.2	6.2	6.4	6.5	±6.4	+6.8	Putrefied odor
B group	6.2	6.2	6.3	+6.3	+6.4	#7.0	₩8.2	Ester-like odor
I group	6.2	6.2	+6.6	+6.3	+6.5	+6.8	₩8.0	Putrefied irritating odor
S group	6.2	6.2	6.2	6.2	6.3	+6.8	+7.4	Putrefied odor
Control	6.2	6.2	6.2	6.2	6.2	6.2	6.4	

If 0.2 cc. of bacterial emulsion injected into the pieces of meat, the production of odor is even more conspicuous and also pH becomes conspicuously alkaline.

5. Invading Route of Bacteria. The route of entry of bacteria into blood and muscle has not been clarified as yet, but 4 routes of their entry can be assumed, that is, explosion wound caused by spear, compressed air pumped into the whale after death in order to keep it afloat, respiratory tract such as lung and the intestinal tract.

In the case of entry through the explosion wound, there is considerable negative pressure in the severed vein on account of the bleeding, therefore it is understandable that some bacteria sticking to the part affected will be absorbed and reach to the heart. When pig is slaughtered and bacteria coated on the wound (in such cases, it is obvious that easily detectable bacteria are used), it is reported that the same bacteria can be found in the bone marrow, and so these facts support this theory. Whales being speared are especially different from animals being slaughtered, because the former try to escape with strenuous activity for more than one hour. More opportunity may be given for bacterial invasion due to the laboured breathing during that time. Next, in regards to pumping in high pressure air, special efforts to prevent entry of bacteria are not taken, so it can be assumed that there is a possibility of bacterial invasion. Also the cutting open the abdomen to cool can give the path of entry for bacteria. Thirdly, in the case of respiratory tract, bacteria can be discovered in the lung of human being and slaughtered pig, in which even the scraps of food are also occasionally found, so there are some scholars who say contamination of blood in heart is caused by passing through veins from the lung. If the detection of principally soil bacteria in lung of pig by Hulphers is taken into consideration, this path of entry can be naturally considered. Even with the present experiments, Micrococcus halophilus, which is a specific sea water bacteria, was detected in the sea water and muscle; also bacteria detected in blood and muscle belong chiefly to soil bacteria; and since a long time has elapsed between the time of catching and the dissection, it is considered that the above three routes of entry play considerably important parts.

However, since whales are animals living in the sea, there isn't any reason why the entry of bacteria into blood and muscle by way of intestinal tract cannot be considered. Even in the case of drowned or poisoned human beings or animals, it can be assumed that perhaps the large number of bacteria which originarily live in the intestines, enter into the blood vessels through the mucus membrane of the intestine due to the bacterial permeability of the intestines after death and to the extraordinary increase

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of bacteria. In the case of whale this mode of entry may be accelerated by the air forced into the abdomen, by which the bacteria invade into the blood, and then they will circulate throughout the body and extend from the end of blood vessels into the muscle. Furthermore, from long experience, the staleness of fishes is prevented by removal of the intestine as soon as possible after death, so it may be assumed that intestinal route plays a major role. Also, from our experiments, the facts that 5 kinds of bacteria, i. e. Micrococcus piltonensis, M. epimetheus, Flavobacterium sewanense, Achromobacter ubiquitum and Achromobacter halophilum were found in the contents of the intestine, muscle and blood, and that Streptococcus faecalis which is always found in the intestine was detected in muscles, supports the theory of the intestinal tract route. We were not able to discover Bacterium coli in samples of 1947. This may be due to the fact that Bacterium coli cannot exist for long in the blood as whale is a warm blooded animal similar to pig. However, this requires further study in the future to settle this point.

6. Putrefaction and Bacteria. It was proven that the number of bacteria will increase in blood and muscle as degree of freshness decreases. Next is that three strains of the detected bacteria do not grow at temperatures above 35°C. The majority has optimum temperatures from 27–30°C, so if the temperature of the body of a whale from the time of catching to dissection is within this range, it will be the optimum condition for increase of bacteria and their putrefaction will occur faster.

Furthermore, these bacteria grow even in a media of low salt content (1%), but growth will be better at about 3%. If the concentration is increased to 15%, growth will be greatly reduced, only 4 strains will exist at 20% and growth is checked at 25%. Therefore, putrefaction can be prevented by maintaining salt contents above 25%.

Of the 57 strains of bacteria, 38 decomposed glucose and produced large quantity of gas. The large quantity of foam observed on stale whale meat is probably due to such bacteria. If such bacteria are inoculated on a piece of whale meat and its pH is examined, at first it is about 6.2 and will become lower after 24 hours, but after 48 hours it will become 6.4—8.2 and will give off rancid, irritating or at times a mild putrified odor. These phenomena will become very marked when several strains are inoculated together, but even then the odor is not as foul as in the case of

actually putrefied fish. Also, in case of actual putrefaction proteus bacteria are sometimes detected, but in our cases there was no such advanced case of putrefaction, and the Micrococcus flavus, M. pikowskyi, M. halophilus, Strept. faecalis, Flavobacterium sewanense and Achromobacter halophilum found in our examinations coincided with those detected by other researchers.

Samples examined	Name of Bacteria	Number of strains	Place ordinarily found, according to references		
	Micrococcus epimetheus	2	rubber tree		
Blood	Achromobacter ubiquitum	6	soil, water		
	Micrococcus spheroides	3	manure, soil		
	Micrococcus piltonensis	2	manure, soil		
	Micrococcus flavus	1	air, milk		
	Micrococcus subcitreus	1	air, water		
	Micrococcus saccatus	1	mucous of nose		
Muscle	Micrococcus halophilus	3	sea water		
··· ··	Streptococcus faecalis	5	animal faeces		
	Flavobacterium sewanense	.1	sea water		
	Achromobacter ubiquitum	8	soil, water		
	Achromobacter halophilum	1	sea water		
	Achromobacter lacticum	1	cow's milk		
	Micrococcus piltonensis	1	manure, soil		
	Micrococcus epimetheus	1	rubber tree		
	Micrococcus pikowskyi	1	sea water		
· · ·	Micrococcus luteolus	1	cheese		
Intestines	Flavobacterium fucatum	1	halibut		
	Flavobacterium sewanense	1	sea water		
	Achromobacter ubiquitum	7	·soil, water		
	Achromobacter halophilum	1	sea water		
	Micrococcus halophilum	2	sea water		
Sea watet	Achromobacter cystinovorum	6	soil		

Table	4.	Snecies	of	Bacteria <sup>,</sup>	detected

# Conclusions

1. Freshness is decreased in proportion to the time elapsed from catching to dissection and to the number of bacteria detected in blood and muscle.

2. As shown in Table 4, 15 species of bacteria were detected in whale's blood, muscle and intestines and 2 species were found in sea water. Of the above, 6 species coincides with those reported by other researchers.

3. There are 4 possible routes for entry of bacteria into blood and muscle and it may be assumed that the intestinal tract is the most impor-

Tomoichiro Akiba, Takeshi Tsuchiya, Makoto Umehara and Yoshiharu Natsume

tant route.

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4. Although the bacteria detected may not play the leading role in putrefaction, it is obvious that they contribute to the decrease in freshness.

5. Therefore, the most effective method to prevent decrease in freshness is to dissect and dispose of it as soon as possible after catching.

6. That the previous method of salting and freezing after dissection is effective for prevention of putrefaction, can be proven by our experimental results on arresting increase of isolated bacteria.

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## Protein Digestive Power of Sperm Whale Pancreatic Enzyme. II.

#### Yasutaro Ishikawa

In the 1st report, protein digestive power of pancreatic enzyme from sperm whales was compared with that of a cow and was found much weaker than the latter. Casein was used as the substrate and the digestive powers were measured by the method of Michaelis given in the Japanese Pharmacopoeia.

Even after being activated in various manners, enzyme from whales digested only 2.5 times of casein of its own weight while that from a cow digested 125 times. This large difference was almost unbelievable.

However in the experiment previously reported the samples were collected from too few bodies to discuss generally. And the time-lapse from the caching to the dismembering of these whales was so long under higher temperature of the summer, that there was a suspision that a part of proteases may have been destroyed by autolysis.

The present experiment was undertaken in order to eliminate these suspicions and to determine the true protein digestive power of a sperm whale pancreatic enzyme. The samples, therefore, were collected at the coldest season during whaling (December 1948) and only the very fresh ones were used. It had been desired to collect samples from as many animals as possible in order to eliminate individual differences and samples were collected from six fresh sperm whales during the two weeks at the landstation. Ayukawa, Miyagi Prf.

#### EXPERIMENTAL

No.	Kind Sex Bo		Body length (ft.)	Time elapsed from catching to disme- mber (hrs.)	Freshness %
1	Sperm	male	45	25	85
2	Sperm	male	46	25	85
3	Sperm	male	46	23	90
4	Sperm	male	50	18	85
5	Sperm	male	42	22	85
6	Sperm	male	51	20	85

#### 1) Whales Used

The freshness is the value evaluated by the whaling experts on the station by macro-observations with considerations to the elasticity and color of

#### Yasutaro Ishikawa

the whale meat and others. The freshest state, i. e. that right after death, is taken as 100 and deterioration from that is shown by a rough percentage. Various scientific methods for the determination of freshness have been forwarded but none is completed and procedures are too complicated so that it has to rely on these empirical method.

2) Treatment and Preparation of Samples

As soon as the whale body has been cut open, the pancreas taken out, adipose and connective tissues removed as much as possible and minced by a hand chopper, From this were prepared 3 kinds of samples for reasons as described in (3). These samples were:

- a) One dried immediately with acetone.
- b) The minced pancreas left standing overnight at room temperature with small amount of toluen in a flask, stoppered, which was then dried with acetone.
- c) The minced pancreas mixed with 30% aqueous acetone solution in an equal amount and stored.

Samples (a) and (b) were prepared by adding anhydrous acetone to minced pancreas in an amount about 5 times its volume, well stirred and the supernatant solution decanted. The same amount of acetone is then added and filtered with suction. The residue on the funnel is washed, first with acetone and then with ether, and is dried by spreading it out in the room. This is then well dried in a dessicator over Sulfuric acid. Procedures up to this point were carried out at the station and this was then brought to the laboratory for testing. The dried pancreas was first powdered by a boat-shaped mortar, sieved through a 60 mesh sieve and divided into powders of gland substances and connective tissues. Special note should

No.	Powder (g)	Powder (g) Sieve residue (g)	
1 A	2.8	5.4	
2 A	1.9	11.6	14
3 A	3.0	10.7	22
4 A	3.2	9.4	25
4 B	1.4	4.0	26
5 A	5.3	9.0	37
5 B	6.3	5.1	55
6A	5.3	7.5 .	41

Table I.

A denotes samples dried immediately

B denotes samples dried after being left overnight.

be taken of the fact that the pancreas of a sperm whale contains much larger percentage of connective tissues compared to that of bovines, hogs and baleen whales, so that the yield of gland substance is very poor. The resultant yield of sieving is given in Table I.

The samples immersed in 30% acetone were brought back to the laboratory and stored at room temperature. And the supernatant solution which might contain protease, was taken from time to time to test the digestive power.

The kinds of samples prepared were as follow:

No. 1—A,	C	No. 4—A,	В	
No. 2-A,	С	No. 5A,	В,	С
No. 3—A,	C	No. 6—A,	B (lost),	С

A, denotes samples dried immediately,

B, samples dried after being left overnight, and

C, samples immersed in 30% acetone.

#### 3) Method of Activation

As in other kind of animals, pancreatic protease from sperm whales do not show digestive action in its fresh state. It follows, therfore, that this protease must in some way be activated and the subsequent testing of digestive powers must be carried out at the same degree of activation, in order that a compaison can be made; if possible, at a maximal activated state. For this reason, activation was carried out in following three ways and the digestive powers determined and compared.

- a) The pancreas was dried in the most fresh condition, powdered as has been described before and this was activated by enterokinase from a sperm whale.
- b) Minced pancreas was left overnight at a room temperature and treated in the same way as before. This was then activated with enterokinase.

The above two were made in order to find the effect of autolysis in the fresh condition.

c) Minced pancreas was dispersed in the same amount of 30% aqueous acetone solution and its supernatant solution, assumed to contain protease from the pancreas, was used as a sample for testing.

The reason that this experiment was carried out was that Kleiner and Tauber reports<sup>2</sup>) that the protease from hog pancreas is best extracted with 33% alcohol and is well activated by a long-time storage. Tejima,<sup>3</sup>)

also obtained a successful result by extracting protease from a sei whale pancreas by the use of 30% acetone.

#### 4) Method of Digestive Power Determination

The method of determining digestive power followed the same one as described in the previous report<sup>1</sup>), i. e. the Item 6 of the Japanese Pharmacopoeia for pancreatin which uses Michaelis' method of protease testing.

The substrate used was casein purified by Hammesten's method and stored in a dessicator over sulfuric acid. 5 cc. each of 0.2% solution of this casein (containing 2 cc. 1/10 N-KOH in 100 cc. of this solution) is taken in test tubes and these tubes are stood in a row. A definite amount of enzyme is dispersed in a definite quantity of water to extract the enzyme and a definite amount of this enzymatic solution is added step-wise to the casein solution and the total volume is then brought to 10 cc. each. This solution is kept for 1 hour at  $40^{\circ}$  to effect digestion of casein. After exactly 1 hour, 3 drops of a mixture of 1 cc. glacial acetic acid, 9 cc. water and 10 cc. alcohol is added to such test tube. One that remains clear is taken as digested and the one showing least opalescence is taken as the limit of digestion. Digestive power of enzym-containing powder is found from the amount of the powder used to digest 5 cc. of 0.2% casein solution (i. e. 0.01 gm. casein) in the limit case and is then shown by the ratio of casein to the enzymatic powder.

In the case of 30% acetone extracted solution, a definit of its supernatant solution is taken, suitably diluted and used in determination as above.

#### 5) **Results of the Experiments**

i) Acetone dried powder

a) Digestive power when enterokinase is not added

Digestive power was determined with No. 1A, No. 2A and No. 4B. None of these had the power to digest casein equal to their own weight. It follows, therefore, that the acetone dried powder either lacks digestive power totally or has a very weak action. This in turn may mean that the enzyme is in an inactivated state due to freshness of pancreas or that enzyme had been destroyed altogether.

b) Digestive power when enterokinase is added

0.2 g. each dried powder of No. 1A and No. 2A was taken, 0.05 g. enterokinase added end dispersed in 100 cc. of water. This was warmed at 40°

for 15 minutes, 30 minutes and 1 hour and activated. The result of the determination of digestive power using these extracts was as follows:

No. 1 A (15 min.)	Digest 10 times its weight of casein but cannot digest 12.5 times its weight.
No. 1 A (30 min.)	ditto
No. 1 A (1 hour)	ditto
No. 2 A (15 min.)	ditto
No. 2 A (30 min.)	ditto
No. 2 A (1 hour)	Digests 7 times its weight of casein but cannot digest 10 times its weight.

From these results, it can be seen that the time of warming for activation does not differ much between 15 minutes and 1 hour but is slightly inferior at 1 hour.

Therefore, the conditions were set for an addition of a 1/4 amount of enterokinase and warming for 30 minutes at  $40^{\circ}$  to effect proper activation. The result of determination of digestive power of various samples were as follows:

No. 1 A	 Digests 10 times its own weight but not 12.5 times.
No. 2 A	 ditto
No. 3 A	 ditto
No. 4 A	 ditto
No 4 B	 Digests 8.3 times its own weight but not 10 times.
No. 5 A	 ditto
No. 5 B	 Digests 10 times its own weight but not 12.5 times.
No. 6 A	 Digests 8.3 times its own weight but not 10 times.

Digestive power of each sample made from above results is shown in Table II, which indicates that there are hardly any difference between the samples.

Sample No.	Digestive Power (cas./powder)	Sample No.	Digestive Power (cas./powder)
1 A	10	4 B	8
2 A	10	5 A	8
3 A	10	5 B	10
4 A	10	6 A	8

Table II. Digestive Power of Acetone Dreied Powder

ii) 30% Acetone Extracted Solution

The solution was stored at room temperature for about 2.5 months during which a definite amount of the clear, supernatant solution was taken and its digestive power determined. Table III, shows the relationship between the digestive power and the time lapsed after immersion in acetone. The figures denote the amount of acetone solution (in cc.) necessary to

#### Yasutaro Ishikawa

digest 0.01 g. of casein (shown by the least opalescence manifested) so that the smaller the figures, greater the digestive power.

Lapse of time (days)	1 C	2 C	3 C	5 C	6 C
5				2/100	
7	1/100				-
9				1/100	10/100
11	1/100	6/100		1/100	
13			15/100		
15				1/100	4/100
17	1/100	3/100	15/100		
21	-			1/100	2/100
23	1/100	2/100	12/100		L.
26	1/100	1/100	12/100	1/100	1/100
34	1/100	1/100	9/100	1/100	1/100
54	1/100	1/100	9/100	0.8/100	0.8/100
75	1.5/100	1.5/100	9/100	1/100	1.5/100

Table III. Relationship between digestive power and lapse of time.

As can be seen from Table III, each sample differs greatly in its digestive power at the start of experiment but after about one month, all samples, except No. 3 C, show constant values of 1/100. This level is kept until about 2 months have elapsed after which the power gradually decreases slightly.

#### DISCUSSIONS

1) Direct comparison cannot be made between the value of 8-10 for digestive power obtained by the determination of dried powder and the values obtained with 30% aceton solution. However, if the amount of water included in minoed pancreas is considered and if the whole amount of enzyme present in the pancreas were to have transferred to the solution, then in the case of 30% acetone solution, the digestive power of dry pancreas as a whole can easily be calculated.

If the amount of the dried matter is assumed to be 25% of the minced pancreas, and since the minced pancreas was added with an equal amount of 30% acetone, then 1 g. minced pancreas + 1 cc. 30% acetone = 0.25 g dry pancreas + 1.75 g extracted solution. i. e. 0.25 g dried matter from pancreas should have the same digestive power as 1.75 g. of the extracted solution. Therefore, 1/100 cc of this extracted solution is equal to

 $\frac{0.25}{1.75} \times \frac{1}{100}$  g dried matter of pancreas.

Since this amount had digested 0.01 g. of casein, then the digestive power of the dried matter of pancreas is

$$0.01 / \frac{0.25}{1.75} \times \frac{1}{100} = 7$$

i. e., it has digested 7 times its own weight of casein. Of course, this calculated amount shows the digestive power of pancreas as a whole but in the case of acetone dried power, it has been put through the sieve during which purification of the enzyme may have been effected so that a precise comparison cannot be made as to which is larger. However, it is considered that the order is well adapted in this case.

2) When enterokinase is not added to the dried powder of pancreas, no digestion occurred. From this fact, it can be assumed that these pancreas was stored in a comparatively fresh state until treatment and that protease had not been destroyed and stayed in its natural state.

The digestive power of the dried powder of pancreas when activated by the addition of enterokinase was found to be about the same in each individual and digested 8—10 times its weight of casein. The protein digestive power of 30% acetone extracted solution varies greatly at first but becomes most activated about one month after immersion at which time, with the exception of one sample, the values are exactly the same for all the samples. In this one exception, i. e. 3C the supernatant solution was found to be considerably opalescent due to the inclusion of grease (the others were almost clear). This is apparently the error due to the inclusion of a large amount of adipose tissues at the time of sampling. The value when this same level had been reached in all the sample, i. e. 1/100, shows that the dry matter of the original pancreas would digest about 7 times its own weight of casein.

From these results, it is considered that the digestive power of protease from sperm whale is considerably inferior to that from bovines and hogs. However, this is true only when casein has been used as a substrate and it is doubtful whether the same tendency will be manifested when other proteins are chosen as a substrate.

From the results of these experiments, it can be assumed that there is very little difference between individuals.

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#### SUMMARY

Protein digestive power of pancreas from 6 sperm whales was determined and following results were obtained:

1) Acetone dried powder does not show digestive action as it is.

- 2) When acetone dried powder is activated by the addition of 1/4 its amount of enterokinase from a sperm whale and warmed for 30 minutes at 40°C, it digests 8-10 times its own weight of casein and there were no differenc between individual animals.
- 3) 30% Acetone extracted solution of minced pancreas, reached its maximal activated state after storage of about one month at room temperature and the digestive power was the same for all the samples, i. e. 1/100 cc. of this solution digested 0.01 g. of casein. Calculated from this value, the dry matter of the pancreas used would have digested about 7 times its weight of casein.

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## Properties of Fats and Oils contained in Various Parts of a Sperm Whale Body

#### Takajiro Mori and Masamichi Saiki

#### Introduction

Numerous studies have been made since the olden times on whale oil. Many works have been made on its physico-chemical properties, on the components of its fatty acids and its saponifiable matter and their properties. However, these studies have chiefly been confined to the oils from whale blubber, bones and head cavity and there has been very little evidence of chemical studies made on oils from large and small intestines, liver, kidney and other organs. It seemed that it would not only be interesting to make a study of oils from various organs from the point of fat metabolism but also necessary from the point of their utilization. Therefore, studies in this line were made.

#### **Experimental materials**

The present experiments were carried out with 2 heads of sperm whales (*physeter macrocephalus* L.) which were caught off Kinkazan point (Miyagi Prefecture in the northeastern part of Japan). One was a male (Sperm Whale B). measuring 34 feet long, caught in September 1946, and the other, also a male (Sperm Whale A), measuring 35 feet long, caught on 23rd. September 1947. Each individual animal was dissected in the dissecting room, various parts collected and those from which oil can be obtained by boiling, i. e. skins, large and small intestines, were boiled on the spot. Those difficult to put to this procedure, i. e. liver and large and small intestines of the Sperm Whale A, were frozen and sent to the laboratory.

In the laboratory, these organs were defrosted, dehydrated by treatment with alcohol, carbon dioxide then blown in and dried in vacuum. After this dried organs were pulverized, it was extracted by ether in a Soxhlet extractor. The alcohol used for dehydration was evaporated under reduced pressure and dried and this residue was shaken with ether. This ether and that from the Soxhlet extractor were brought together and after drying with anhydrous sodium sulfate, the ether solution was dropped into dry acetone while stirring constantly and the phospholipid precipitated was filtered off.  $^{\rm 34)}$ 

Acetone and ether were driven off under reduced pressure in carbon dioxide stream and some oil was obtained. The ether extract from the liver of Sperm Whale B amounted to 11.73% of dried matter. The oil from the head cavity was collected when the oil flowed out at the time of dissection. This head oil (oil from the head cavity) is a white solid at a room temperature, forms a pale, yellow liquid at (around) 30° and precipitates about half the amount of white, cloudy crystals at around 25°.

The blubber oil (Oil from the skin) is pale yellow. The oil obtained from large and small intestines is yellow, but that obtained by ether extraction is brown. During the summer, intestinal oil is liquid but pricipitates solid matter during the winter (11°C). Liver oil is brown and remains liquid, although slightly viscous, all through the winter.

These samples were treated with acid clay or activated charcoal and yellow oils were obtained which were used for the following experiments.

#### Experiment

Neutral oils— With the above samples, specific gravity, coefficient of refraction, acid value and saponification value were measured in an ordinary manner, and the iodine value by Wijs' method. Saponification was limited to 2 hours. Vitamin A content was also measured by Beckmann's spectrophotometer. The results obtained are shown in Tables I and II.

Parţs	Appearance	d <sup>25°</sup> 4°	$n_{\rm D}^{40^\circ}$	Acid Value	Sapon. Value	Iodine Value	Non- sapon. matter	Vitamin A (I. U.)
Head oil	White soild	0.8744	1.4500	0.73	140.71	55.01	44.98	103
Blubber	Pale yellow liquid	0.8779	1.4570	0.75	131.08	82.33	35.51	370
Small intestine	Pale yellow liquid	0.8879	1.4570	2.01	156.84	78.88	17.35	865
Large intestine	Pale yellow liquid		1.4522	13.36	154.48	77.71	19.18	- 1
Liver	Brown, viscous liquid		1.4711	2.64	165.34	92.41	19.01	12,655
Table II. Neutral Oils from Sperm Whale B.								
Parts	Appearance	$d_{4^{\circ}}^{20^{\circ}}$		<sup>32°</sup> D	Acid Value	e Sapor	1. Ic alue	dine Value
Head oil	White solid	0.8752	2 1.	4689	0.52	142	.35	57.57
Blubber	Pale yellow liquid	0.8792	2 1.	4696	0.36	127.	.89	82.13
Small intestine	Pale yellow liquid	0.9056	5   1.	4721	1.03	1	-	78.23
Large intestine	Pale yellow liquid	0.9054	1 1.	4724	1.01	-		79.41
Liver	Dark brown semi-solid		1	4861	18.83	155.	.91	82.12

Table I. Neutral Oils from Sperm Whale A.

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Although the whales A and B are different individuals, all values in the two above tables are similar, showing that there are very little difference between the individual animal.

Non-saponifiable matter— The samples were saponified with alcoholic potash, water added and shaken 4 times with ether. combined ether solution was washed with water, dried with sodium sulfate and ether evaporated under blowing in carbon dioxide.

Non-saponifiable matter from all the samples were a pale yellow solid during the winter  $(11^{\circ}C)$  and the properties are as shown in Table III. The melting points were determined in a capillary tube, values being that of the beginning and end of melting.

Parts Appearance		n <sub>D</sub> <sup>40°</sup>	Melting point (°C)	Iodine value (Wijs')		
Head oil	Pale yellow solid	1.4450	35.0-37.0	39.08		
Blubber	Pale yellow solid	1.4500	20.5 - 24.0	71.74		
Small intestine	Pale yellow solid	1.4490	29.5 - 31.8	51.72		
Large intestine	Pale yellow solid	1.4500	29.0-31.8	50.53		

Table	ш.	Properties of	Non-Saponifiable	Matter	from	the	Sperm	Whale	A.
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**Fatty acids**— The ageous solution obtained by the removal of nonsaponifiable matter was evaporated under reduced pressure to remove alcohol, hydrochloric acid added and the freed fatty acid extracted by ether. This was treated in the same way as for non-saponifiable matter. Of the fatty acids obtained:

Mixed fatty acids from the skin oil was a pale brown liquid during the winter (11°C) precipitating some solid fats; That from head oil was pale yellow solid, with a mixture of some crystalline solid fats; and those from large and small intestines were a yellow solid. Properties of these fatty acids are shown in Table IV.

Parts	nD Melting poi (°C)		Iodine value (Wijs')	Neutralization Value		
Head oil	1.4437	16.0-20.0	56.44	246.68		
Blubber oil	1.4511	13.5-18.0	80.57	199.22		
Small intestine oil	1.4530	28.0-31.0	79.54	194.66		
Large intestine oil	1.4518	28.0 - 31.5	81.85	195.27		

Table IV. Mixd Fatty Acids from the Sperm Whale A.

**Solid and Liquid Acids**— Solid and liquid fats were obtained from the mixture of fatty acids using about 10 g of each sample according to Twitchell's method<sup>35)</sup>. The solid acids here obtained were white solids, and liquid acids from intestinal and head oils were yellow, that from blubber

oil, yellowish brown. The properties of these solid and liquid acids are shown in Tables V and VI.

Parts	Appearance	Percentage against total fatty acid	$n_{\mathrm{D}}^{60^{\circ}}$	Melting point (°C)	Iodine value (Wijs')	Neutraliza- tion value
Head oil	White solid	21.31%	1.4289	38-39	3.06	250,63
Blubber	White solid	10.07%	1.4352	50 - 52	14.56	213.62
Small intestine	White solid	18.64%	1.4363	50 - 52.5	15.39	211.36
Large intestine	White solid	19.29%	1.4368	50-52.5	11.48	210.49

Table V. Solid Fatty Acids from the Sperm Whale A.

Tale VI. Liquid fatty acids from the Sperm Whale A.

Parts	Appearance	Percentage against total fatty acid	$n_{\mathrm{D}}^{40^{\circ}}$	Iodine value (Wijs')	Neutralization value
Head oil	Yellow liquid	78.69%	1.4450	73.93	236.12
Blubber	Yellowish brown liquid	89.93%	1.4522	92.26	194.72
Small intestine	Yellow liquid	81.36%	1.4550	95.07	187.25
Large intestine	Yellow liquid	80.71%	1.4542	91.03	187.83

#### Summary and Discussions

1) The blubber oil of the sperm whale, especially the oil from its head cavity, is composed of fatty acids of small molecular weight and is unique to it except the butter fat. As a result of the present experiments, it has been found that the average molecular weight of fatty acids was the smallest in those contained in head oil, that from the blubber oil next and followed by large and small intestine and liver oil, which contained the large molecular weight acids although the values were approximately similar (estimated from the amount of non-asponifiable matter of neutral fats and the saponification value).

Both the solid and liquid acids from the head oil were of small molecular weight compared to other oils from other organs.

2) The degree of unsaturation of mixed fatty acid is the smallest in the head oil, those from blubber, large and small intestines are about the same and are larger. This seems to be due to the fact that the head oil contains a large amount of solid acids and that the degree of unsaturation of its liquid acid is small compared to the oils of other organs.

3) It is known that sperm whales feast largely on quantities of squids proof of which was borne out by the discovery of a large amount of them in the stomachs of the sperm whales used for these experiments. In order

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to find the relationship between the specific nature of sperm whales and the oil of squids devoured as their chief food, reference was made to the analysis of the oil from a certain kind of squids caught in the Hokkaido, Japan, by Mitsumaru Tsujimoto<sup>36)</sup>. This analysis showed that this oil contained a very small amount of non-saponifiable matter and its degree of unsaturation was exceedingly high. Comparison of similar data by E. André and H. Canal<sup>37)</sup> showed that the degree of unsaturation was also high in the French squid oil but contained more non-saponifiable matter than the Japanese squid oil, values being near those of sperm oil. According to these French investigators, the non-saponifiable matter contains, differing from the sperm oil, no aliphatic alcohols, either saturated or unsaturated. From these facts, it is assumed that sperm whales utilize these squid oils to turn into its own oil.

4) As has been shown in the foregoing, sperm whales possess a large amount of wax so that it gives abnormally high amount of non-saponifiable matter. In the present experiments, the amount of non-saponifiable matter in the head and blubber oil was exceedingly large but that from large and small intestine and liver oil was only one-half of the former, being about the medium between the oil of sulfur bottom and that of sperm whale. It is interesting to note that the average moleclar weight of fatty acids and the amount of non-saponifiable matter are in an inverse ratio in the above two.

It would be interesting to study the relationship between these facts and the fat metabolism in whale bodies and if such a relation does exist, what significance it has. These must be left for the future.

5) The degree of unsaturation of the non-saponifiable matter is the smallest in head oil, followed by the oils of large and small intestines which were about the same, and largest in the blubber oil.

Our thanks are due to the Department of Education for grants in aid of this research.

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## Studies on Kitol I. Preparation of Kitol from Whale liver Oil

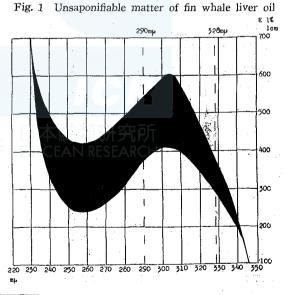
#### Tadashi Tawara and Ryusuke Fukazawa

Willstaedt & Jensen<sup>1</sup>) and Nakamiya<sup>2</sup>) pointed out the presence of a Vitamin A-like substance, differing from the ordinary Vitamin A in its properties, in whale liver oil. Recently, Baxter, et al.<sup>3</sup>) isolated kitol.

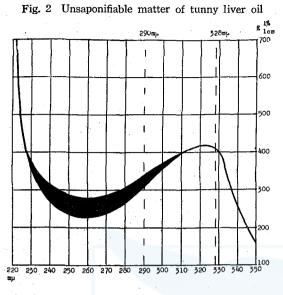
The presence of a foreign substance in whale liver oil had long been predicted by the difference of their spectrum analysis and the Carr-Price reaction from ordinary fish liver oil. The maximum of the absorption spectrum of the unsaponifiable matter of ordinary fish liver oil is in the neighborhood of  $328 \text{ m}\mu$ , but that of whale liver oil is at  $290-325 \text{ m}\mu$ , and the Carr-Price reaction of whale liver oil is bluish-purple whereas that of fish liver oil colors blue. From the proximity of the figures of extinction coefficient,  $\frac{1\%}{1\text{ cm}}$ , of  $265 \text{ m}\mu$  and  $328 \text{ m}\mu$  in the absorption spectrum of the unsaponifiable matter of whale liver oil, it had been believed that the absorption curves would lie close together and overlap each other and an apparent maximum would lie in intermediate point.<sup>2</sup>)

The authors also determined the extinction coefficient  $E_{1cm}^{1\%}$  of the unsaponifiable matter of fin whale liver oil (Fig. 1)\* by the spectrophotometer and were able to prove this point.

This absorption curve apparently differs from that of tunny liver oil (Fig. 2). The greatest difference exists in the position of the absorption maximum and the width of the curve. The Carr-Price reaction of this unsaponifiable matter of



\* The reason that the curve is so wide is because the absorption is so indistinct that the value of extinction coefficient,  $E_{1cm}^{1\%}$ , does not come out clearly.



fin whale liver oil gives bluish purple color while that of tunny liver oil, blue.

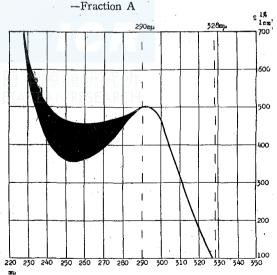
The oxidation product of Vitamin A gives red or reddish brown coloring by this reaction. According to the authors' experiments, majority of this oxidation product transists into saponifiable matter during saponification so that it does not seem to constitute a reason for bluish purple coloration. Nakamiya and others harbored a

doubt that it might be due to the large amount of cholesterol present but the authors take the view that this is due to the presence of kitol.

Kitol as isolated by Baxter, et al., were colorless, elongated prismatic crystals of Fp. 88–90°, with a molecular formula corresponding to  $C_{40}H_{58}$  (OH)<sub>2</sub>, possessing 8 double bonds. It gives an absorption maximum at 290 m $\mu$ , and  $E_{1cm}^{1\%}$  290 m $\mu$ =707.

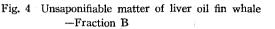
The fact that kitol possessed twice the molecular weight of Vitamin A was utilized in separating the two by the difference of solubility. The unsaponifiable matter of fin Fig. 3 Unsaponifiable matter of fin whale liver oil

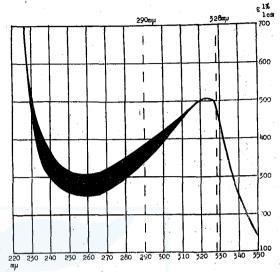
whale liver oil was dissolved in petroleum ether and this was extracted with 90% methanol into which Vitamin A was transited. The substance which remained in the petroleum ether solution to the last was examined by the spectrophotometer and the value of  $E_{1cm}^{1\%}$  at each wave length was shown by a curve (Fig. 3). This substance showed an absorption maximum at 290 m $\mu$ , and the Carr-Price



reaction gives a red coloration. Apparently, it contained a very small amount of Vitamin A.

On the other hand the  $E_{1cm}^{1\%}$  value of the substance that transited to 90% methanol (curve in Fig. 4) gave a maximum absorption at around  $328 \text{ m}\mu$ ,\*\* which is very similar to the curve of tunny liver oil (Fig. 2). It does not give the indistinct curve as in Fig. 1, any more. The Carr-Price reaction is blue, so that this substance must be nothing but Vitamin A.





From these facts, it can be assumed that the substance that reamained in petroleum ether to the last is clearly kitol.

#### EXPERIMENTAL

250 g. fin whale liver oil was warmed with 500 cc of 20% methanolic potash for 1 hour in a water bath. After saponification, this was extracted with ether and the ethereal layer was washed several times with water and dried with anhydrous Glauber's salt. After distilling off ether, 54 g. of unsaponifiable matter was obtained. This substance gives bluish purple coloration by Carr-Price reaction and, according to  $E_{1cm}^{1\%}$  curve (Fig. 1) by spctrophotometer<sup>\*\*\*</sup>, possesses an absorption maximum 303 m $\mu$  although the absorption area is so indistinct that it is hard to obtain a correct value of  $E_{1cm}^{1\%}$ . Accordingly, the curve became one of a wide band. This is apparently due to the fact that two or more substances are mixed in it and gives a complex absorption so that this must be taken as the apparent absorption maximum.

The total amount (54 g) of this unsuponifiable matter was dissolved in 1000 cc. of petroleum ether (Bp. 30-60°), placed in a separating funnel

<sup>\*\*</sup> According to this experiment, the absorption maximum of Vitamin A-like substance isolated from the unsaponifiable matter of tunny liver and whale liver oil is more nearer  $325 \text{ m}/\ell$  than  $328 \text{ m}/\ell$ .

<sup>\*\*\*</sup> Spectrophotometer by Carl Zeiss was used. The solvent was absolute alcohol.

#### Tadashi Tawara and Ryusuke Fukazawa

and extracted 38 times with 500 cc each 90% methanol. The petroleum ether layer was then washed several times with water, dried with anydrous Glauber's salt and distilled under reduced pressure. A reddish yellow, syrupy matter was obtained to the amount of 14 g. 90% Metanol solution was distilled under reduced pressure in carbon dioxide stream and the residue, composed mostly of water and a small amount of oil drops floating on it, was placed in a separating funnel. This was extracted with ether and the ethereal layer was distilled off after dry in with anhydrous Na SO. The residual matter, reddish yellow syrup obtained was ca. 39 g. About l g unsuponifiable matter was lost during this separating process.

The substance which transited to 90% methanol (hereafter designated as fraction B) gives a blue coloration to Carr-Price reaction, extinction coefficient curve as shown in Fig. 4 which approximately coincides with that (Fig. 2) of the unsuponifiable matter from tunny liver oil, i. e. it shows an absorption maximum at around  $328 \,\mathrm{m}\mu$  and does not give the indistinct absorption as before separation. The Carr-Price reaction is not longer bluish purple but blue so that this is a substance devoid of foreign matter, i. e. Vitamin A alone.

The residual matter in petroleum ether (hereafter designated fraction A) gives a red coloratin to Carr-Price reaction and extinction coeffcient curve (Fig. 3) is clearly different from that of fraction B (Fig. 4) and possesses an absorption maximum at 290 m $\mu$ ,  $E_{1cm}^{1\%}$  290 m $\mu$ =500.\*\*\*\* It is apparent that fraction A is kitol.

No presence of kitol were discovered in the oil obtained from the eyeball of sperm and sei whales.

The authors' deepest gratitude is due Dr. Tsutomu Maruyama, Director of the institute, who gave them kind instructions, and to Mr. Tadashi Nakai of this institute and to Dr. Seiichi Ishikawa of the University of Literature and Science for their unfailing assistance and advice.

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\*\*\*\* Extinction coefficient of pure kitol at 290 mµ is 7073) but that of kitol separated by Embree, etal.<sup>4)</sup> at first was 580 at 290 m $\mu$ , which apparently was still quite impure. Frantion A obtained by the author theoretically contains 70% kitol.

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# Studies on Kitol. II. Influence of Kitol Fraction on the Determination of the International Unit of Vitamin A.

#### Tadashi Tawara and Ryusuke Fukazawa

In the previous paper<sup>\*</sup>, the authors showed that the unsaponifiable matter of whale liver oil gave only the apparent absorption maximum due to the presence of a mixture of two or more substances while that of tunny liver oil gave an absorption maximum at around  $328 \text{ m}\mu$ . The Vitamin A fraction (fraction B) of whale liver oil gives a maximum at around  $328 \text{ m}\mu$  but due to the presence of kitol fraction (fraction A), the whale liver oil gives a very complicated absorption. Accordingly, the determination of Vitamin A of whale liver oil by a spectrophotometer does not give the correct extinction coefficient at  $328 \text{ m}\mu$ , only that of an apparent value, so that determination of true Vitamin A content cannot be made by this method. This fact is borne out by the fact that with whale liver oil, results of animal test and spectrum analysis do not coincide.

The authors examined the influence of kitol fraction on Vitamin A by the use of fractions A and B separated by the authors. However, this is an intermediate report and further detailed experiments must follow since there is no direct proof that the kitol fraction obtained does not contain even a minute amount of Vitamin A, or that it does not contain other unsaponifiable matter.

39 g. of Vitamin A fraction and 14 g. of kitol fraction were obtained from 53.5 g. of unsaponifiable matter of the whale liver oil. First, the international unit of Vitamin A was determined with Vitamin A fraction and then the extinction coefficient at  $328 \text{ m}\mu$  of a mixture of 14 parts kitol fraction to 39 parts Vitamin A fraction, which the international unit of Vitamin A was calculated. As a result, it was shown that the value of the international unit of Vitamin A in this case came out 12.1% higher than the true value of Vitamin A content. In other words, the true content of Vitamin A is the value obtained by subtracting 12.1% from the value obtained by spectrophotometric determination.

It follows, naturally, that the determined value of Vitamin A comes

Cf. Report I, p. 85 of this Bulletin.

out larger than the true content of Vitamin A when the amount of kitol content becomes large compared to the amount of Vitamin A. This will show that a presence of kitol has a very great effect on the assay of Vitamin A by the spectrophotometer and therefore, points to the fact that the method of Vitamin A determination of whale liver oil must be improved.

The authors' thanks are due to Taiyo Fisheries Co., for their kindness in furnishing the material for these experiments.

#### **EXPERIMENTAL**

53.5 g unsaponifiable matter were obtained from 250 g. of the liver oil of a fin whale by the method as described in the previous report. From this unsaponifiable matter, 39 g Vitamin A fraction (fraction B) and 14 g. kitol fraction (fraction A) were obtained (loss of 0.5 g.).

1) Carl Zeiss' spectrophotometer was used to determine the international unit of Vitamin A fraction (solvent—absolute alcohol).

B fraction— $E_{1cm}^{1\%}328m\mu=454.5$ therefore, this substance contains 727,000 I. U. of Vitamin A per 1 g.

**2**) Kitol fraction (fraction A) $-E_{1cm}^{1\%}290 \text{ m}\mu = 500$ 

$$E_{1cm}^{1\%}328 m\mu = 108.7$$

therefore, this substance would look as though it contained 174,000 I. U. of Vitamin A per 1 g., but the Carr-Price reaction gives a red coloration and contains virtually no Vitamin A.

**3**) International unit was determined with a mixtue of 39 parts Vitamin A fraction and 14 part kitol fraction (a proportion so arrived because the unsponifiable matter of this whale liver oil contained these 2 fractions in a proportion of 39:14).  $E_{1cm}^{1\%} 328 \text{ m}\mu = 375$ 

therefore, this substance must contain 600,000 I. U. of Vitamin A.

However, 1 g. of this substance contains only  $\frac{39}{39+14}$  g. of Vitamin A fraction and, taking that the kitol fraction does not contain any Vitamin A (by Carr-Price reaction), the true content of Vitamin A can be shown by the following equation:

727,000 I. U.  $\times \frac{39}{53} = 535,000$  I. U.

therefore 600,000-535,000=65,000 I. U.

Actually, 65,000 I. U. Vitamin A had been shown than was actually con-

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The Influence of Kitol Fraction on the Determination of the International Unit of Vitamin A 91

tained, or in other words 12.1% more Vitamin A.

4) The determination of international unit was made with a mixture of an equal portin of A and kitol fraction which gave:

## $E_{1cm}^{1\%}$ 328 m $\mu$ = 35.8

therefore, this substance seems to contain 572,800 I. U. Vitamin A but according to the aforementioned calculations, it actually contains only 363,500 I. U. In this case, the spectrophotometric value gave 30.4% higher content of Vitamin A than was actually present.

5) International unit was determined with the unsaponifiable matter of tunny liver oil which gave:

## $E_{1cm}^{1\%}$ 328 m $\mu$ = 38.5

therefore, 1 g. of this substance contains 61,600 I. U. of Vitamin A.
6) International unit was determined with a mixture of 39 parts unsaponifiable matter of tunny liver oil and 14 parts kitol fraction which gave:

 $E_{1cm}^{1\%}328 m\mu = 50.0$ 

therefore 1g. of this substance seems to contain 80,000 I. U. of Vitamin A but according to the above calculation, the real content of Vitamin A is 45,100 I. U. In this case, ca. 77.4% higher value than the actual content of Vitamin A was obtained by the spectrophotometric determination.

7) International unit was determined of an equal amount of unsaponifiable matter of tunny liver oil and kitol fraction which gave :

 $E_{1cm}^{1\%}328 m\mu = 59.0$ 

therefore 1 g. of this substance shows a Vitamin A content of 94,400 I.U. but, according to foregoing calculation, the actual content of Vitamin A would be 30,800 I.U., if there is no presence of Vitamin A in kitol fraction. In this case, the photometric determination has given 206.5% higher value than the true content of Vitamin A.

## Studies on Kitol. III. The Effect of Sunlight, Air and Heat on the Vitamin A and Kitol Fractions

#### Tadashi Tawara and Ryusuke Fukazawa

In the 1 st Report, the separation of Vitamin A and kitol fractions from unsaponifiable matter of a whale liver oil was described. In order to refine kitol, it became necessary to know its properties, chiefly its stability, and the effect of sunlight, air and heat on these two fractions were examined.

1) The sunlight has a considerable effect on both fractions. Direct radiation of sunlight on 0.1% alcoholic solution of Vitamin A fraction destroyed one-half the amount of Vitamin A in 3 hours and that on kitol fraction destroyed one-half of it in 4 hours.

2) Continuous passage of air through 0.1% alcoholic solution of kitol fraction showed no marked effect on kitol but the same process on Vitamin A fraction resulted in the destruction of about 2/3 the amount of Vitamin A present after 5 hours.

**3**) Boiling 0.1% alcoholic solution of kitol did not show any damage to kitol in 12 hours but the same treatment on Vitamin A fraction resulted in the destruction of ca. 1/7 the amount of Vitamin A.

As a result, it can be stated that Vitami A is very unstable toward sun light and air, and is slightly effected by heat. Kitol, on the other hand, is fairly stable against heat and oxygen but is considerably sensitive to light.

#### EXPERIMENTAL

The Vitamin A and kitol fraction obtained by the method as described in Report I were dissolved in absolute alcohol to obtain a 1% solution, respectively. With this solution, following experiments were carried out the amount of Vitamin A and kitol remaining in the respective solutions were determined by measuring the extinction coefficient at 290 m $\mu$  for kitol and at 328 m $\mu$  for Vitamin A.

#### 1) The Effect of Light

30 cc. each 0.1% alcoholic solution of Vitamin A and kitol fractions were

#### The Effect of Sunlight, Air and Heat on the Vitamin A and Kitol Fractions

placed in 5 graduated test tubes (dia. ca. 1.3 cm, length ca 21.5 cm, colorless, hard glass) each and after being stoppered, were radiated to direct sunlight (on 6 th September 1949. Fine. From 1200 to 1700). One test tube each of Vitamin A and kitol fractions were taken every one hour and the values were determined. After radiation was completed, the content of the test tuve was diluted with absolute alcohol to bring the concentration to 0.002% and the extinction coeffecient was measured with Carl Zeiss' spectrophotometer, that for Vitamin A fraction at 328 m $\mu$ , and for kitol fraction at 290 m $\mu$ . The results of the measurements are shown in Table I and Fig. A.

The Influence of Sunlight

Table I.

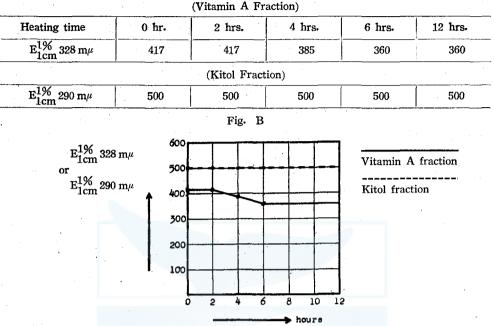
		(Vitami	n A Fraction	<b>1)</b>		
Radiation time	0 hr.	1 hr.	2 hrs.	3 hrs.	4 hrs.	5 hrs.
$E_{1cm}^{1\%}$ 328 m $\mu$	417	312	278	228	156	125
		(Kito	l Fraction)		1	
$E_{1cm}^{1\%}$ 290 m $\mu$	500	417	385	278	243	192
or	6 328 mμ 6 290 mμ	F 600 500 400 200 100 0 1	2 3		nmin A fract	ion 

#### 2) The Effect of Heat (78°C)

500 cc. each of 0.1% alcoholic solutions of both fractions were taken into 1 *l*. Erlenmeyer flask and boiled with a reflux condenser in a boiling water bath (78°C). After each 2 hours period, the solution was slightly cooled and 30 cc. of it was drawn out for testing. This was diluted to 0.002% strength with absolute alcohol and determinations made with spectrophotometer. Results are shown in Table II and Fig. B.

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#### Tadashi Tawara and Ryusuke Fukazawa



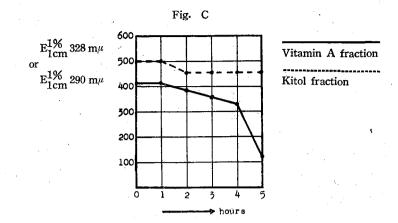
#### Table II. The Influence of Heat (78°C) (Vitamin A Fraction)

#### 3) The Effect of Air

15 cc. each of 0.1% alcoholic solution of both fractions were taken in the graduated test tubes (as described above), and the air was passed through this solution of the extent of its surface is continueing constantly noise by means of a glass tube measuring ca. 0.5 mm. in inside diameter. This procedure was carried out from 1 to 5 hours. When the passage was stopped, each glass tube was washed with little alcohol and this washing was added into the glass tube and the volume brought correctly with absolute alcohol to 15 cc again (alcohol evaporated ca. 1.5 cc. per hour during passage of air). Absolute alcohol was further added to this solution to make it a 0.002% solution and the value were determined as previously. Results are shown in Table III and Fig. C.

Table	m.	The	Influence	of Air
1	(Vitan	nin A	Fraction)	

Passage time	0 hr.	1 hr.	2 hrs.	3 hrs.	4 hrs.	5 hrs.
$E_{1cm}^{1\%}$ 328 m $\mu$	417	417	385	360	333	125
		(Kito	I Fraction)			
$E_{1cm}^{1\%}$ 290 m $\mu$	500	500	455	455	455	455





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## On the Respiratory Pigments of Whale (Studies on Whale Blood II.)

#### Tadashi Tawara

The diving method of whale is very regular and ordinarily this may be divided into the two classes of short diving and long diving. That is. it makes a relatively shallow dive, then breathes by raising its head and back above surface of water and then makes a shallow dive again. The diving and breathing is repeated several times. This repeated dives are After repeating this surface dives several times, it called surface dives. makes a deep dive in the water. This long dive is called "sound". After one "sound", surface dives are repeated several time. The time of "sound" is relaively constant, depending on kind of whale, in case of baleen whale, it is from 7 to 10 minutes, but this exceeds 30 minutes in case it senses danger. It is especially long in case of sperm whale, which is an tooth whale. It is from 30 to 70 minutes and averages about 40 minutes.

Several explanations are given on how oxygen is supplied during such prolonged submergence.

Ommanney<sup>(1)</sup> states that the vascular networks (retia mirabilia) functions as this oxygen supply. That is, a special structure called vascular networks which are abundant in blood vessels and fats act as accessory lung and functions as strage for oxygen. However, Laurie<sup>(2)</sup> states that this is impossible and points out that from the relation between basal metabolism and lung's vital capacity, whale can be submerged for a long time. That is he calculated the lung's vital capacity of a 122,000 kg blue whale at not less than 3050 liters and that the air requirement per minute at 178.75 liters. He states that according to these calculations, it is possible for a whale to be submerged for at least 17 minutes.

As stated above, a whale makes several surface dives and then a long dive. "sound" From this fact, the author considers that the whale breathes several times during the several surface dives, by this stores a large quantity of oxygen in some part of the body and consumes it during a long submer gence. One of the storage organ may be the vascular networks as proposed by Ommanney out the author thinks that it is rather the respiratory pigments. The muscles of mammals which carry on a swiming life such as pinnipedias and cetaceas are darker in color than those of land mammals. Especially in the case of sperm whale, the muscles are almost black in color.

The so-called respiratory pigments are haemoglobin, myoglobin (muscle haemoglobin) and various kinds of cytochromes. All of these are pigment proteins containing iron and conbines readily with oxygen. Of the above, the molecular weight of myoglobine is  $17,500^{(3)}$  or about 1/4 of that of haemoglobin, about the same as that of haemoglobin in iron content, or 0.345%,<sup>(4)</sup> combining power with oxygen and carbon monoxide is much stronger than haemoglobin and also, reaction is much faster.<sup>(5)</sup> There are several kinds of cytochromes and it is believed that they act as carriers of hydrogen by the oxidation and reduction of iron within the molecules. The representative ones are *a*, *b* and *c*.

The author carried on quantitative analysis of iron in haemoglobin, myoglobin and cytochromes a and c of these respiratory pigments and determined the content of the different pigments. As control, the blood and muscle of cow and horse, which are land mammals, were used.

To determine the haemoglobin content in blood, Sahli's method using haemoglobinometer was used concurrently with the method in which iron content is quantitatively determined directly. The result of the determination shows that by Sahli's method, there is no great difference in haemoglobin content between whale and land mammals. However, the direct determination of iron indicated that it is considerably greater in whale than in land mammals. In the case of land mammals the results from Sahli's method and iron determination method coincides rather closely but in the case of whale, the iron content is greater in each case. That is, it can be assumed that there is a greater quantity of iron compounds other than in the haemoglobin state. By Barkan and Schales,(6) it was discovered that in the blood of mammals, desides haemoglobin, iron-containing organic compounds whose iron ion is readily liberated by dilute It is said that this substance has an hydrochloric acid are also present. affinity to oxygen about 4 to 10 times greater than that of haemoglobin. It can be considered that perhaps the presence of these substances is anticipated.

In regards to cytochrome b, the quantity was so small that it was

#### Tadashi Tawara

impossible to determine so it will be omitted. Content in cytochrome a and c did not differ greatly from those of land mammals. Thus it is assumed that it did not have any special meaning relative to oxygen storage.

Myoglobin content in whales is much greater in all cases when compared with land mammals and especially in the case of sperm whale which makes "sound", the content is about 8 times more.

From the above result, it is assumed that one reason why whales can withstand "sound" is that oxygen is stored by myoglobin within the muscles.

The author wishes to think Dr. Tsutomu Naruyama and Mr. Tadashi Nakai for guidance in this experiment and to Profs. Shichiro Akiya and Teizo Ogawa for their instructions. The author is grateful to Mr. Ryusuke Fukazawa for assistance in the experiment and to the people of the Ayukawa Station of the Taiyo Fishing Co., for assistance in providing materials.

#### Experimental

Materials used in this experiment was a sperm whale and a sei whale caught off shore of Kinkazan Island, the former 31 feet 3 and the latter 45 feet 3. In both cases, they were 24 hours after death.\*

Materials from cow and horse used as control was obtained immediately after slaughter from the Shibaura Slaughter House in the case of heart and in the case of muscle, those on the market were used.

#### (1) Quantitative Determination of Haemoglobin

Blood samples in each case was obtained from the heart.

(A) Sahli's Method

According to Sahli's method, haemoglobin was transformed into acid haematin by adding  $\frac{N}{10}$  HCl and colorimetric determination was made with Sahli's Haemoglobinometer.

(B) Quantitative Determination of Blood Iron

In accordance with Pincussen's method,<sup>(7)</sup> blood was decomposed with sulphuric acid and 30%  $H_2O_3$ , and comparative colorimetric determination was made of it, colored with KCNS, with standard concentration solution of Iron Alum (Fe<sub>2</sub>(SO<sub>4</sub>)<sub>3</sub> (NH<sub>4</sub>)<sub>2</sub> SO<sub>4</sub>·24H<sub>2</sub>O) colored with KCNS, using

Materials were obtained in July, 1948. After this, there was a revision in law and cathing of sperm whale less than 35 feet was prohibited,

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	Haemoglobin	Fe (Hb×0.336)	(Pincussen Method)
Sei Whale	15.6 g	52.4 mg	. 84.0 mg
Sperm Whale	15.8g	53.1 mg	95.0 mg
Spem Whale foetus	5.8g	19.8 mg	53.8 mg
Cow	12.4 g	41.6 mg	41.0 mg
Human(29 years old)	13.0 g	43.7 mg	46.5 mg

Duboscq's colorimeter.

Quantity of haemoglobin determined by Sahli's method multiplied by 0.336 should give the quantity of iron in the haemoglobin state. In the case of cow and human the quantity of iron in the haemoglobin state and the total iron content is about the same. However, in the case of whale. iron determined by Pincussen's method is much greater. The difference is great especially in the case of foetus. It is believed that this is because of the presence of larger quantity of iron other than in haemoglobin state. (2) Quantitative Determination of Iron in Cytochrome State

In order to be accurate, the quantitative determination of cytochrome should be made by spectral analysis but in the case, it was refined considerably and iron content determined. The determination figures does not indicate absolute quantity but relative quantity determined under the same conditions. For materials the above mentioned heart and muscles of whale, cow and horse were used.

(A) Quantitative Determination of Iron in Cytochrome a State<sup>(8)</sup>

A solution containing 2% Na-cholat and  $\frac{1}{20}$  mol Na<sub>2</sub>HPO<sub>4</sub> is, for convinience, designated as A-solution. Muscle is washed with water, chopped with a meat chopper and 1 kg taken. 3 liter of A-solution is added to this, extracted for one night at room temperature, strained with gauze and to 3 liter of the filtrate, 750 cc of a weakly ammoniac saturated ammonium sulphate soltion is added (0.2 saturation). This liquid is separated with a centrifugal separator and to the clear liquid is added more saturated ammonium sulphate solution to make it 0.5 saturation. Cytochrome *a* will precipitate out. The precipitate is placed in a centrifugal separator, dissolved in 200 cc of A-solution and made 0.2 saturation with saturated ammonium sulphate solution. This solution is left standing for 2 days in an ice room, precipitate is removed with centrifugal separator, the clear liquid is made 0.33 saturation with ammonium

#### Tadashi Tawara

sulphate, precipitate is dissolved with 100 cc of A-solution, precipitate obtained by 0.2 saturation ammonium salphate is removed, precipitate obtained by 0.33 saturation is collected and this operation is repeated once more. Finally, precipitate obtained by 0.23 saturation becomes a reddish brown, paste-like substance. This is dissolved in A-solution, the total quantity brought to 100 cc., 10 cc. of this is taken, the entire precipitate obtained by saturation with ammonium sulphate is decomposed with  $H_2SO_4$  and  $H_2O_2$  and iron determined by the aforementioned Pincussen's method.

Table 2. Quantitative Determination of Iron in Cytochrome a State (mg/kg)	Table 2.	Quantitative	Determination	of Iron	in	Cytochrome	a	State	(mg/kg)
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	Sei Whale	Sperm Whale	Cow
Body muscle	1.29	1.91	2.28
Heart muscle	2.54	4.03	0.27

In the case of cow, the figure for body muscle was larger than for heart. In this case, it was not possible to obtain body muscle and heart from the same cow so the materials came from different cow.

(B) Quantitative Determination of Iron in Cytochrome c State<sup>(9)(10)</sup>

The muscle sample is washed with water, chopped with a meat chopper, 2.5% trichloracetic acid is added to 1 Kg of this, stirred and extracted for about 2 hours at room temperature. This is then strained with gauze and 1 liter of the filtrate is brought to pH 7.0 with 6N-NaOH solution. 500 g of ammonium sulphate is added to this and left standing for one night in an ice room. This is centrifugally separated and the clear liquid brought to pH 3.7 by adding 5N-H<sub>2</sub>SO<sub>4</sub>. Precip tate preduced from this is separated with centrifuge, 20 cc. of water is added to this and dialyzed (cellophane) against 1% saline solution for 2 days. The liquid thus obtained is centlifugally separated and a dark red, clear cytochrome c solution is obtained. This is diluted to 100 cc., 10 cc. of this is saturated with ammonium sulphate at 60°C, the entire precipitate is decomposed with  $H_2SO_4$  and 30%  $H_2O_2$  solution and quantitative determination of iron made by the aforementioned method.

	Sei Whale	Sperm Whale	Blue Whale	Cow	Horse
Body muscle	0.43	0.47	0.44	0.30	-
Heart muscle	1.29	0.57	. <b>-</b> · ·	0.70	1.17

Table 3. Quantitative Determination of Iron Cytochrome c State (mg/Kg)

Cytochrome c is obtained by multiplying this quantity of iron by  $\frac{100}{0.43}$ . (3) Quantitative Determination of Iron in Myoglobin State<sup>(4)</sup>

Sample muscle is washed with physiological saline solution, chopped with a meat chopper and 1 kg of this is taken. 1 liter of water is added to this, extracted for 1 night at room temperature and strained with gauze. *N*-NaOH solution is added to 1 liter of this solution and brought to pH 7.0, 250 cc of concentrated basic lead acetate solution is added and the precipitate produced from this is separated with centrifuge. Na<sub>2</sub>HPO<sub>4</sub> solution is added to the clear liquid while maintaining it neutral with NaOH and lead is removed. 5 cc. of the clear liquid obtained by centrifuge is taken, saturated with ammonium sulphate, the entire precipitate is decomposed with H<sub>2</sub>SO<sub>4</sub> and 30% H<sub>2</sub>O<sub>2</sub>, and colorimetric determination of iron, in the form of Rhodan-Fe, is made.

Table 4. Quantitative Determination of Iron in Myoglobin State (mg/kg	Table 4.	Quantitative	Determination	of	Iron	in	Myoglobin	State	(mg/kg)
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	Sei Whale	Sperm Whale	Cow	Horse
Body muscle	31.35	151.00	18.08	
Heart muscle	7:62	23.12		7.15

Myoglobin is obtained by multiplying this iron quantity by  $\frac{100}{0.345}$ . Compared to cow, the myoglobin content is 2 times more in sei whale, and 8–9 times more in sperm whale.

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## Research on Methionine in Whale

(On Methionine (CH<sub>3</sub>S--CH<sub>2</sub>--CH<sub>2</sub>-CHNH<sub>2</sub>) Content)

#### Masami Yoshida

#### I. Introduction

Methionine is an amino acid containing sulphur and was discovered in the hydrolyzed product of casein by Mueller in the United States in 1922. This was synthetically produced by Bagar and Coyne in 1928 and named it methionine. After this, a large number of people attempted to synthesize and isolate it.

From the nutrition point of view, methionine, together with cystine, is an important amino acid as a source of sulphur for animals. According to recent researches, it is said that methionine transforms into cystine in a living body. In 1937, Rose<sup>(1)</sup> reported that methionine is a necessity and cystine is not a necessity and explained that it aids growth, and disproved Osborne an Mendel's theory.

Furthermore, in regard to its content, Baernstein in 1932, made determinations of methionine, a characteristic methylthiol group, of various proteins and reported that methionine content is greater in animal protein than in vegetable protein.

Lact albumin	2.3%(3)	Glycinin	1.8%(2)
Egg albumin	4.5%(3)	Gliadin	$2.0\%^{(2)}$
Casein	$3.1\%^{(3)}$	Zein	2.2%(3)
Sardine flesh	3.14%(4)		
Beef	$3.66\%^{(2)}$		\

Whale meat (Kind unknown) 2.92%<sup>(4)</sup>

In regard to methionine content in whale meat, the only report is by Tomiyama.

The auther made determinations of methionine content in whale meat in order to determine its nutritional value, compared with other foodstuff and as a first step in effective separation and utilization of methionine from whale meat; a relatively detailed analysis of its content in various parts of sei whale, sperm whale and fin whale was made.

There are various methods for making this determination, but Baernstein's method was used here. Baernstein made quantitative analysis under the assumption that methoxyl and methylimid bases does not exist in protein molecules and that the quantity of methyl iodide obtained by reaction with HI results only from  $CH_2S$ .

For accuracy, methionine content in beef protein and casein was determined and compared with previous literatures.

#### II. Experiment

(a) Collection of Samples

Heart, tongue, ventral meat (Sunoko) and meat was obtained from a 13.6 meter male sei whale at Ayukawa, Miyagi Prefecture. The length and sex of the fin whale and sperm whale are not known, but both were obtained at Ayukawa. For beef, the round of beef obtained on the market was used.

(b) Preparation of samples

#### Whale and beef protein

Flesh is first sliced thinly and soaked in cold water, which is heated to  $36^{\circ}$ C and changed to new water. The it is boiled once. This is then expressed and water removed as much as possible, chopped in a meat chopper, dried in sun, extracted with boiling alcohol for 4 hours and then with ether for 10 hours and dried for 3 hours at  $105-110^{\circ}$ C.

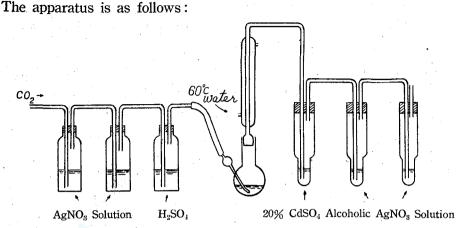
#### Baleen

After washing with water, it is dried at room temperature, cut in thin slices, extracted with ether for 10 hours and dried for 3 hours at 105–110°C.

(c) Outline of Operation

Sample is decomposed with HI and  $CH_3I$  produced, absorbed in alcoholic AgNO<sub>3</sub> solution to form AgI, filtered and the residual Ag determined quantitatively with 2/100 N KCNS using m cro-burette. The methionine quantity is calculated from the difference in Ag quantity in a blank test.

The conditions for the determination were as follows; In the original report, it is stated that about 90% of the CH<sub>3</sub>I come over in the 1 st hour but the remainder usually takes several hours more, and Baernstein adopt about 15 hours, but here the reaction time is 6 hours, about 0.5 g of the sample is decomposed with 10 cc. of HI (s. g. 1.7) and absorbed in 20 cc. of alcoholic AgNO<sub>3</sub> solution.



(d) Experimental results

Table No. 1.

	Content	
Sei Whale	Heart Tongue Ventral meat (Sunoko) Belly meat Head meat Back meat Tail meat	4.80 4.22 4.17 3.42 3.33 2.78 2.71
Fin Whale Meat Sperm Whale Mea Blue Whale Baleen Fin Whale Baleen Sei Whale Baleen		4.82 3.46 0.82 1.92 1.96

The methionine content of beef and casein was determined by the same method and compared with previous literature, it is as follows:

Table No. 2.,

Sample -	HE IN STITUTE OF CETACE Analyst EAPCH			
	Baernstein <sup>(2)</sup>	Baernstein <sup>(3)</sup>	Tomiyama, Hanada <sup>(4)</sup>	Yoshida
Casein	3.53% 3.36 3.25	3.1%	2.54%	3.94%
Beef protein	3.66			3.56
Whale meat protein			2.92	3.06

In Table No. 1, the average methionine content of 3.06% in the various parts of sei whale, which is ordinarily called meat is very close to the value 2.92% by Tomiyama, which was obtained by the difference between

#### Research on Methionine in Whale

total S for Denis Benedicts method and cystine S for Okuda's method.

However, there is something here which require special attention. In the case of casein. several experiments gave the result of  $3.94 \pm 0.02\%$ , but in the case of beef and whale meat, there are some cases when the results of several experiments does not coincide. As to the cause of this discrepancy, on considering Table No. 1, it appears that there is a tendency for the value to be high in parts which are rich in connective tissues. Thus it was thought that there may be some relation between methionine content and the connective tissue contained in the sample, so the following experiment was performed.

The kind of whale is not known but the sample was prepared in the same manner as before, connective tissue was separated as much as possible from the meat and methionine of each was determined. The result is as in Table No. 3.

Table No. 3.

Connective Tissue	Meat Scrap	
3.22%	1.43%	

That is, from the determination results, it is clear that methionine content is greater in the conective tissues than in the meat scraps.

#### III. Summary

- (1) The methionine content of the part ordinarily called meat is 3.06% in sei whale, 4.82% in fin whale and 3.46% in sperm whale.
- (2) Of the different parts of sei whale, content is largest in heart, next in the portion rich in connective tissues (tongue, ventral) and then in the part where there is little connective tissues (meat).
- (3) The difference in methionine content in the different part of whale meat is thought due to the difference in the quantity of connective tissues in the various parts.
- (4) The large content in the heart is thought to be of great interest in relation to physiological action of renewal.

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### Factory Ship Whaling Around Bonin Islands in 1948

#### Kazuhiro Mizue

#### 1. Introduction

The author carried on investigation on board the whaling factory ship Kaiko Maru of the Nippon Marine Products Co., which carried on whaling in the waters around Bonin Islands from February to May, 1948.

Various biological investigations were made on board the factory ship and a general outline will be presented here.

Catcher boats attached to the factory ship were the No. 2 Kyo Maru and No. 3 Kyo Maru of the Nippon Marine Products Co., and those attached to No. 9 ship of the Taiyo Fishery Co., were the Seki Maru, No. 7 Seki Maru, and the No. 7 Seki Maru was replaced by the No. 2 Seki Maru during the operation.

Salting and refrigeration ship was attached to both fleet to act as transporting ships, and in general, the composition of the fleet was as above.

First, a brief history of whaling in the waters around Bonin Islands is a follows. Whaling was begun in Japan before the preceding century but whaling in the waters of Bonin Island was of relatively recent origin. In 1922, an investigation was made to determine latent resources in the waters of Bonin Islands as a whaling ground, and actual operation was begun in the following year, 1923. That is, the Toyo Whaling Co., the predecessor of the present Nippon Marine Products Co., was the first to develop the whale resources in this region. Much later, in 1937. Taiyo Fishery Co., known as Hayashi Kane Shoten at that time, also engaged in this operation.

From then, the so-called Bonin Islands base whaling was continued until 1944. During this period, Nippon Marine Products Co. maintained a base on Chichi Jima and Taiyo Fishery on Haha Jima, competition between the two was removed and operation was carried on in a friendly manner.

At the time when land bases were established and operation begun in 1922, it was a good fishing ground for winter, shore approaching hump back whale, similar to those offshore Galanpi Cape in Formosa, and together with this, the whaling ground was developed year after year, so that catches of Sperm Whale and Sei Whale gradually increased and considerably good whaling record was established, such that after 1939, a total of 200 whales were caught.

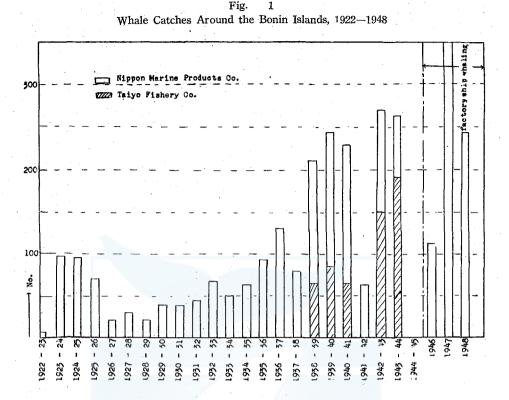
Season	November	December	January	Februaly	March	April	May	Total
1922-23	0	3	0	0	0	0	0 .	3
1923-24	0	3	9	29	28	17	8	94
1924 - 25	0	0	21	17	28	23	6	. 95
1925 - 26	0	0	12	19	20	12	7	70
1926 - 27	0	0	5	5	9	2	0	21
1927 - 28	0	5	8	9	4	4	0	30 ·
1928-29	2	0	6	6	8	0	0	22
192930	0	14	18	3	5	0	.0	40
1930-31	0	1	16	12	11	0	0	40 .
1931 - 32	0	5	16	13	11	0	0	45
193233	0	1	26	25	16	0	0	68
1933-34	.0	11	10	16	9	4	0	50
1934-35	1	13	10	16	23	1	0	64
193536	0	5	11	22	28	25	. 2	.93
1936-37	0	17	. 18	36	42	18	0	131
193738	0	1	23	11	16	22	6	79
1938-39	0	3	44	67	55	34	8	211
1939-40	0	0	9	40	141	50	4	244
1940 - 41	· 0	0	7	72	52	93	5	229
1941-42	0	0	20	37	6	0	.0	63
1942-43	0	0	- 19	36	115	82	18	270
1943-44	0	. 0	11	32	158	67	0	268
1944—45	0	0	0	0	0	0	0	0
1945-46	0	0	0	0	59	54	0	113
Total	3	82	319	523	844	508	64	2343

	Table 1.	
1922 - 1946	Monthly Wahle Catches for the Bonin Islands	

Table 1 shows the yearly catches of whale by month, from 1922 when operation was begun, up to 1948.

In regards to Table 2, only one company was engaged in whaling from the time whaling was begun, up to 1938 so that yearly average number of whales caught was only 62. However, after Taiyo Fishery began actual operation in the following year, the rate of catches increased rapidly for the following three years and the yearly average catches increased to 228 whales. The following year was the year in which war began and with the outbreak of war, operations in the waters were greatly restricted. As the result of this, the catches shows a sharp decline and only 68 whales were caught during this year. Operations were again resumed to normal

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the following year and the year after, inspite of being in the midst of war and with the increase demand for the army and domesticuse, a record number of catches were made.

The following year was the year in which war was terminated and base whaling in the Bonin Islands were stopped with the fall of Saipan and with the occupation of Iwo Jima by the United States Forces. During this period, (22 years), the average yearly catches were 155 whales.

In regards to the whaling season, as can be seen in Table 1, the total catches is greatest in March, followed by February and April.

As to kinds of whale, ordinarily, they are Humpback Whale, Sei Whale, and Sperm whale and sometimes, Blue whale and Fin whale are caught.

In regards to whaling ground, the center of operation is 100 nautical miles east of Haha Jima. (It is needless to say that Humpback Whales have the habit of approaching the shore so this is an exception).

With the termination of war, the Bonin Islands were outside the area in which fishing is permitted. However, due to the zeal of the whaling firms and efforts of the Fishery Bureau, and due to the kind consideration

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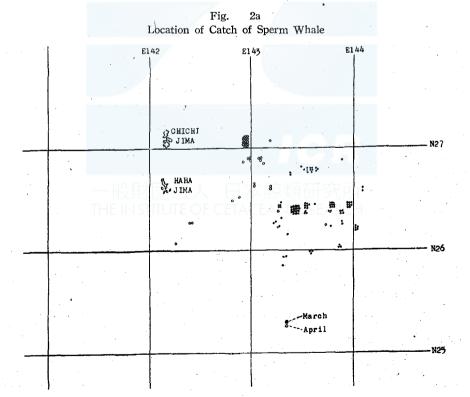
of SCAP, factory ship whaling in the waters of Bonin Islands was permitted with various conditions attached. The result of whaling after the use of factory ship whaling is as shown in Table 2.

Table	2.
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Year Kind	Blue Whale	Fin Whale	Sei Whale	Humpback Whale	SprmWhale	Total
1946		· .	29	12	72	113
1947	4		150	1	195	350
1948	1	3	105	3.	131	243

As mentioned above, many Sperm whales, Sei whales and Humpback whales are found in this fishing ground and Blue whale, Fin whale and Right whales only rarely migrate here. After the termination of war, operation was changed to factory ship whaling and the number of catches of Humpback whales was greatly reduced, as compared with the period when whaling was by base whaling. This is a natural result because Humpback whales likes to migrate close to the reef of the island and since factory ship whaling cannot be carried on within 12 nautical miles from shore.

During the base whaling period, operation was carried on from Nove-



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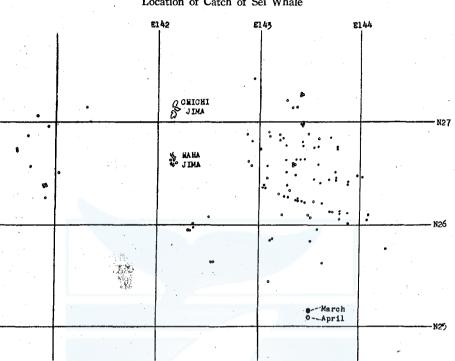


Fig. 2b Location of Catch of Sei Whale

mber to May of the following year, the peak being from February to April. Even with the change to Factory ship whaling, this peak period is taken advantage of.

Originally, the fishing ground in the Bonin Islands is very small, compared to that off shore Sanriku Area. Table 4 is a chart showing the position of catches in 1948 and in looking at this, catches were made between lat. 26°N-27°N, long. 143°E-144°E, or in an area of 60 nautical miles squaee.

That is the principal whaling ground is 50-100 nautical miles E-SE of Haha Jima.

Also, in looking at this chart, the position of catches of Sei whale and Sperm whale gradually approach the islands from February to May. This is related with the water temperature and feed.

The quantity of fuel oil was very small in 1984 so during the 1948 whaling, the factory ship could not be stationed in the middle of the fishing ground and usually was in a drifting condition so in looking at the position of both factory ships in the course chart, it is apparent that they drifted here and there at the mercy of the wind. As a result, both fleets were cosiderably separated from each other and resulted in numerous difficulties in operation.

## 2. Detail

In 1948, besides Sperm whales and Sei whales, only 3 Fin whales. 1 Blue whale, and 3 Humpback whales were caught. This is a very small number, and in the future, it will be necessary to investigate, compare and examine it with those of Antarctic Ocean and investigate it from the resource point of view.

(a) Sei whale-Balaenoptera borealis (Lesson)

Table 3 shows the length of whales caught and of the total of 105 caught by both fleet, sex was about half and half.

Body length in	feet	\$	ę	Total
35			, 1,	1
36		2	3	5
37		2	1	3
38		1	5	6
39		3	3	6
40		2		2
41		7	9	16
·· 42		10	9	19
43			. 3	10
44		6	4 .	10
. 45		3	5	8
46		5	6	11
47		3		.3
48		3		3
49		みまれ ロー	大船市米百万工之中百斤	1
50	川又!			
51	I HE IN	SHIULE OF CEIA	LEAN KELEAKUTI	. 1
Total		54	51	105
Mean lengtl	1	42.65	41.96	42.31

Table 3.

Mean length for  $\circ$  was 42.7 feet and  $\circ$  was 42.0 feet, Mean length for  $\circ$  and  $\circ$  as 42.3 feet.

The largest whale was 48 feet for  $\diamond$  and 51 feet for  $\Diamond$  and the smallest whale was 36 feet for  $\diamond$  and 35 feet for  $\Diamond$ . The peak of body length frequency curve is 42 feet.

In such Sei whales, there is a considerable difference in length with

## Kazuhiro Mizue

sea areas, and average length is greater as it goes northward and gets smaller as it goes southward from Kurile Islands sea area, Sanriku sea area, Bonin Island area, Kinan sea area and Tsushima-Formosa sea area. In viewing former statistics, there is as much difference as 5 feet in those from north sea area and Tsushima-Formosa sea area. The average length of Sei whale caught in the waters around Bonin Islands in 1948 was in between these.

Also, in Sei whales caught in the waters around Japan there are some in which corrugated markings on the dorsal and ventral side are conspicuous and some which are not. Thoese which have corrugated markings are called "totan-iwashi" (corrugated Sei whale) and as a general rule, these are of Northern variety and those without corrugated markings are found in the south western sea area and are of southern variety. It is thought that northern and southern varieties may be differentiated from these corrugated markings and difference in length.

However, during the current investigation, Sei whale with corrugated markings are not limited only to large whales and considering other points, there are no indications that they are of different speice. That is, the so-called corrugated Sei whale are Sei whales with numberous old scars and are not regional difference or sub-specie. Scars which are the cause of this corrugated markings are due to parasitic protozoa and in extreme cases, there are considerable number of scars so that they can be scooped up with a spoon.

Outside of this, the only exterior parasites were small number of pennella whose part exposed from the skin surface, has fallen off. As a general rule, exterior parasite is very few because of sub-tropic area.

Furthermore, as to interior parasite, only a small number of distoma was discovered in the small intestine.

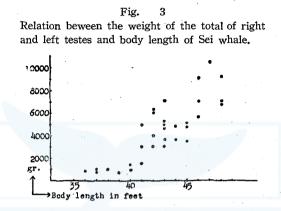
In regards to these parasites, it is planned to report on this after research, together with those in the stomack.

As to thickness of blubber, the vertical thickness was measured for 99 whales at the side below the dorsal fin and back of the head. The mean value below the dorsal fin was 5.2 cm and are of normal nutritional condition in general. In regards to investigation of the mammary gland, it will be omitted here.

As to feed, there were many with empty stomach and for 140 whales

investigated, 40 had empty stomachs, 13 had eaten Euphausia, 34 Calanus and 17 young sardines. As a general rule, kind of feed was of single variety.

As to the investigation of reproduction gland, the weight and volume of the  $\Im$  testes was determined. Table 4 shows the relation between body length and weight of testes.



That is, it is about 1 kg (total of left and right) up to 39 feet and more than 4.0 kg above 42 feet. In general, it is said that the sexual maturity of  $\delta$  Sei whale is 44.6 (13.5 m) but from this investigation, it is estimated to be 42.0 feet. Howeber, there is a very marked difference in the Sei whale in different regions so it is necessary to make comparative studies with those of other regions.

As so the  $\varphi$ , the weight, volume, number of corpora and graafian follicle of the ovary were determined.

According to the corpora Table showing the relation between corpora number and length of body, the body length of minimum sexual maturity of a  $\varphi$  is 38 feet, minimum for pregnant whale is 41 feet and maximum is 46 feet. Even when looking at the frequency curve of corpora number, the number of young whales with little corpora number in very large as a general rule. As to the weight of the ovary, there is considerable difference in accordance with the presence or absence of functional corpora but generally they are matured at 250 gr. It is said that as a general rule, the body length of sexual maturity of a  $\varphi$  Sei whale is 14.5 m (47.9 feet) but according to the present investigation, it is much smaller than this and is completely matured above 46 feet. Also, in case there were those with functional corpora the uterus was always out open, inspected for foetus and

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their body length, sex, etc, were determined. Pregnant whales numbered 11, smallest foetus 2 inches and the maximum 3 feet 5 inches. Their sex were 6  $\Im$ , 5  $\Im$  and no multiple foetus or deformity were noticed.

Next, the adhesion of the cartilagenous layer between the vertebral matter and tip of bone of the vertebrae was determined and the maximum body length of non-adhesion in  $\diamond$  was 46 feet and the minimum body length of adhesion was 42 feet. In the  $\varphi$ , it was 49 feet in the former and 46 feet in the latter. In the  $\diamond$ , there were some which had adhesion at 42 feet.

(b) Sperm Whale (Physeter macrocephalus, L)

Sperm whale is polygamous and has a habit of forming harems. They are distributed widely throughout the world and in the waters around Japan, from offshore Sanriku to Hokkaido and Kurile Islands are good fishing grounds. It is the general rule in all mystacoceti that  $\varphi$  is larger than  $\vartheta$ , but in the odontoceti, this is the opposite. In Sperm whales,  $\vartheta$ is much larger than  $\varphi$ .

Past statistics shows that large sperms of more than 60 feet are not rare but maximum for  $\varphi$  is 31 feet and there are only a few above 40 feet. Therefore, the body length and production of sperm whales differ considerably with sex so that they should be considered as of being of different variety.

Whalers in Japan classify Sperm whales into the three classes of large sperm, medium size sperm and small sperm. Table 4 shows the length of Sperm whales.

		Fig.	*	· · · · · · · · · · · · · · · · · · ·
Body	length in feet	δ	<b>P</b>	Total
	26			1
	27	NSTITUTE OF CETAG	LEAN RESEARCH	1
	28		1	1
	29	1	1	2
•	30		3	3
	31	1	· 5 *	6
	32	1	6	7
	33	2	6	8
	34		3	3
	35	4	27	31-
	36	3	13	16
	37	1	8	9

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Factory	Ship	Whaling	Around	Bonin	Islands	in	1948
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Body length in feet	\$	ę	Total
38	1	3	4 ·
39	1	3	4
40	3		3
41	1		ï
$_{42}$ .	1		1
43	1 .		1
44			
45	1		. 1
46	2	N	2
47	3		3
48	2	· · · ·	2
49	3		3
50	2	• • • • • • •	2
51	3		<b>`</b> 3
52	4		4
53	6		6.
54	1		1
55	1	~	1
56			
57	1		1
Total	50	81	
Mean length	44.64	34.41	38.31

The total catches for both fleet was 131 and of this, 50 were  $\diamond$  and 81 were  $\varphi$ . The size of the Sperm whale harem in the vicinity of Bonin Islands is much smaller than those off shore Sanriku, but even then, it is not a rare thing to see schools of 20-40 small Sperm whales. However, since the catching of whales less than 35 feet by factory ship whaling is prohibited, the catching of  $\varphi$  is restricted to a considerable extent and whaling is done for  $\diamond$ .

Mean body length is 44.6 feet for  $\diamond$  and 34.4 feet for  $\diamond$ .

Maximum body length for  $\diamond$  is 57 feet and 39 feet, for  $\diamond$ .

Minimum body length is 29 feet for  $\diamond$  and 26 feet for  $\diamond$ .

In picking out what appears to be the peaks of body length frequency curve, it is 35 feet, 40 feet and 53 feet for  $\diamond$  and 35 feet for  $\diamond$ , but determination of age school is very difficult due to insufficient information, the same as in the case of Sei whale. In regards to color of body, Mr. Omura is investigating and classifying Antarctic sperm whales but in the case of sperm whale in Bonin Islands it differs with whales, and various kinds of scattered patterns and white spots are present around the stomach

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#### Kazuhiro Mizue

and pudenda. However, these patterns are found not only in the large ô of the so-called isolated Sperm whales but even more in small Sperms. Also these differences in pattern is also recognized from the time of foetus.

Next, Sperm whales have teeth and there are two rows, in the lower jaw but in the case of whales with short body length, these are buried under the skin. Even in considerably matured whales, the innermost 2-3 pairs are not exposed but they appear only as protuberance of the skin when seen from the outside. The upper jaw has holes in which the teeth of the lower jaw fits in but 5-6 layers of traces of teeth can be recognized in it or in its vicinity.

In many cases, the number of teeth on the left and right side differs but as a general rule, there are from 12-25 pairs.

Scars of Sperm whale does not differ very much from that of Sei whale but only in that the number is relatively small. Also, they are present only in the tail portion in case of Sei whale, but in the case of Sperm whale some can be seen at the head.

Furthermore, in most cases, traces of line-like scratches can be seen around the mough, which tells of fierce battles with large cuttlefishs, which are its feed and coin-size traces on the suckers.

In regards to parasites the only thing is a few traces of pennella parasite and 2—3 cases of conchoderma parasites of tooth. In case of whales from northern parts such as White whale (not Delphinaptenes leucus but old bull) Aosa whale (refers to North Pacific Fin whale with diatom film attached), etc, forms diatom film on the body and diatmes is greater in whales in the lowen temperature belt and are parasitric and this is one way of estimating the period of life in low temperature belts. From this, it can be said that whale in the Bonin Islands has very little relation with the northern low temperature belt. As a general rule, there are very few parasites on Sperm whale in this region because of its subtropic water, similar to Sei whale. Also as to internal parasites, only the very common ones are found.

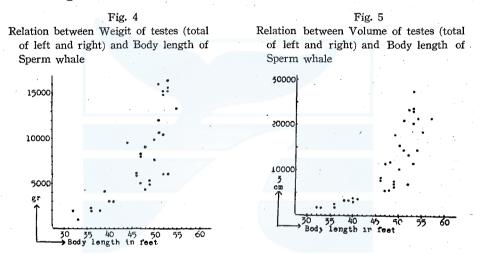
Thickness of blubber was determined by the same method as in the case of Sei whale and it was found that nutrition was ordinary.

Feed is ordinarily cuttlefish and octopus but besides these, there were some which ate jellyfish. Those with empty stomach were 47 out of the 131 investigated but large amount of mouth piece and eye of cuttlefish and Nematoda were found in the stomach.

In regards to reproductive organ, the weight and volume of testes, weight, volume, Graafian follicle corpora, etc were determined, the same as in the case of Sei whale.

The testes of  $\hat{\circ}$  Sperm whale is considerably larger than that of other Mystacoceti, spherical in shape and its position is entirely different from those of Mystacoceti.

Table 4 shows the weight of testes and body length and the curve makes a sharp rise from body length of 40 feet, and there are some winch exceed 15 kg.



The same can be said for Table 9 also which shows the relation between volume of testes and body length.

There is considerable difference for each whale, but from this, it can be estimated that the sexual maturity of  $\diamond$  Sperm whale is body length from 42 to 44 feet, as a general rule. However, more investigation is necessary in order to arrive at a definite conclusion.

As to  $\varphi$ , the body length of minimum sexual maturity is 28 feet, according to the corpora table. However, they are unmatured whales of 35 feet. Since the whaling was by the factory ship method and as a result, catching was limited to whale whose body length is more than 35 feet, it is difficult to estimate the sexually matured body length by this investigation alone because in  $\varphi$  of Sperm whale, many reach sexual maturity below 30 feet. As to corpora, those with 5 were most numerous, centered around body length of 35 feet and maximum number is 14.

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There were 22 pregnant whales, the maximum pregnant whale being 39-feet and the minimum 28 feet. Maximum body length of foetus was 13 feet and the minimum was 2 inches. Ratio by sex was 10  $\circ$  and 12  $\circ$ .

It is reported that the length of pregnancy of Sperm whale is 16 months and can be roughly classified into the two groups of those which were pregnant for less than 1 year with foetus of less than 1 foot and whose which were pregnant for a long time and approaching delivery, with foetus of more than 10 feet.

However, there is a necessity of collecting more data to find out the growing period, accurate period, of pregnancy and body length of foetuss at delivery.

This ends the current report. The author wishes to express sincere grasisude to Mr. Omura and Sakiura of the Fishery Board for direct instructions in this investigation.



# Statistic Study of Foetuses of Whales

# Kazuhiro Mizue and Hisako Jimbo

# Introduction

The material of the present study regarding foetuses of whales is based upon the monthly reports forwarded to the Whaling Society by various whaling companies from 1911 to 1948. Most of such reports were destroyed by the fire during the War. Thanks to Mr. Ohmura's efforts, however, all the numbers issued in the following sixteen years, —1911, 1914, 1919, 1921, 1922, 1926, 1932, 1934, 1941, 1942, 1943, 1944, 1945, 1946, 1947 and 1948— have been recollected so as to be available for us at present. Moreover, six numbers of both 1910 and 1940, too, have been restored.

These whaling reports record particulars of individual animales of the monthly catch with such items as date of the catch, species, sex, body-length, foetus, stomach contents, and locality of the catch.

As for the older records of foetuses, they seem to have been made according to the rough estimation by the eye. Yet there may be no gross forgery about them, as in the case of body-lengths which are subject to body-length limitation and ratio money. Since 1946, however, the records have become authentic, because in that year whaling inspectors began to be employed in each landstation along our coasts, who, as in factory-ships, make biological investigation of the catch as well as superintend whaling operations. As anyone that has been on board a factory ship and at present in the scene of dissection of a carcass, can well suppose, foetueses below 5 inches are very apt to be overlooked. In fact, either for this reason or for the ship in recording, description of such small foetuses are extremely rare before 1945. Neither is it probable that before 1945 each ovaries with functional corpora lutea should have been operated upon so as to ascertain the presence of a foetus. The description of foetuses are confined to such large sized species as Sperm, Sei, Fin, Blue, Hump-back, Right and Grey whales. Of these the foetuses of Blue, Hump-back, Right and Grey whales are so few in number that it is hardly possible to study them with any statistic result.

## Statistic Study of Foetuses of Whales

# Sex Ratio

As for Sperm whales, the ratio of males and females are 46.2% and 53.8% respectively: so it is roughly half and half. Since early days it has been said of Sperm whales, which have the habit of forming Harem, that females greatly outnumber males. This is by no means correct. According to the sex ratio of foetus of the same species, females slightly outnumber males, but this relation is quite reversed in the sex ratio of the caught whale. (males are 52.4%, females are 47.6%)

The sex ratio of the foetuses of Sei whales is 45.5% and 54.5%, showing the ratio of either sex is almost the same. This relation is not changed in the sex ratio of the caught whale. (males are 49.0%, females 51.0%)

								•				
Spe- cies		Foetus	of Fin			Foetus	of Sei			Foetus o	of Spern	1
Year	Male	Female	Unkn- own	Total	Male	Female	Unkn- own	Total	Male	Female	Unkn- own	Total
1910	0	0	• 0	0	1	1	0	2	1	5	0	6
1911	1		0	1	2	2	1	5	3	3	1	7
1914	2	2	2	6	1	- 1	11	13	1	.7	10	18
1919	0	0	0	0	1	, 0	2	3	8	4	0	12
1921	· 0	0	0	. 0	1	1	2	4	7	9	0	16
1922	0	0	0	0	2	0	. 0	2	6	9	· 3	18
1926	0	0	0	0	1	2	0	. 3	0	3	0	3
1.932	4	1	5	10	5	9	2	16	0	2	3 '	5
1934	2	4	0	6	13	14	1	28	20	25	0	45
1940	4	2	0	6	1	1	0	2	8	13	. 0	21
1941	6	2	0	8	5	. 9	· 0	14	28	38	. 0	66
19 <b>42</b>	6	8	0	14	6	· 14	0	20	2	4	0	6
194 <b>3</b>	4	6	0	10	6	6	0	12	17	14	0	31
1944	3	2	0	5	8	7	1	16	19	21	2	42
,1945	. 1	1	0	2	<b>0</b>	3	0	3	4	´ 9	0	13
1946	· <b>1</b> ·	6		ST 7	4	4	DEAON	8	22	29 -	0	51
1947	1	2	0	3	7	12	0	19	45	33	1	79
1948	3	0	0	3	17	11	1	29	56	60	2	118
Total	38	36	7	81	81	97	21	199	247	288	22	557
Sex ratio	51.4	48.6		100	45.5	54.5		100	46.2	53.8		100

Table 1.

So is the sex ratio of Fin whales, males being 51.4% while females 48.6%. According to Mr. Matsuura, female foetuses are less in number than male ones in Mystacoceti. But it cannot possibly be correct, for both the sex ratio of foetuses shown in Table 1 and that of the caught whale, indicate either in Mystacoceti or in Odontoceti the sex ratio is half and

half.

# Relation between Pregnancy and Body-length

Fig. 1 shows graphically the numbers of pregnant whales of the species of Sperm, Sei and Fin whales, respectively, according to their body-lengths. For lack of material, we had to omit the observation of pregnancy in other species. As for Fin whales, the minimum size of pregnant whales is 50 feet and the maximum size 71 feet, while the greatest number of pregnant cases, are seen in the body-lengths of 64—66 feet. In Sei whales, the minimum size is 38 feet, the maximum size 56 feet and the peak of pregnancy is formed between 45—50 feet. In Sperm whales, the maximum size, of pregnant animales is 46 feet but females measuring 30—37 feet, especially 32—35 feet are most frequently pregnant. In the adjacent waters of Japan the catch of Sperm whales below 30 feet is prohibited by body-length limitation, while Sperm whales measuring 30 feet are already sexually mature and therefore liable to pregnancy. For this reason the minimum size of pregnant of Sperm whales cannot be determined.

Fig. 1

In recent years the size of the whales in the catch has been reduced

as is clearly seen in any body-length frequency curve. So in the stock of whales as they are in the sea, this curve of pregnancy may shift a little to the side of larger sizes.

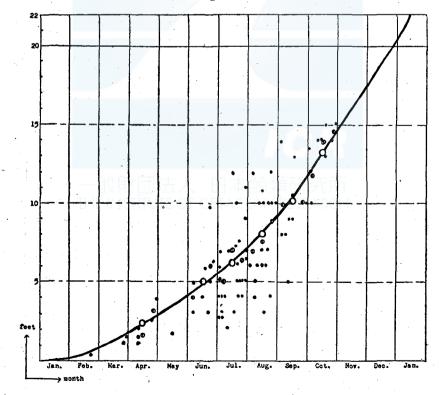
# Growth Curves

Now we are going to determine the pairing period, the duration of pregnancy, the delivery period and the body-length at delivery according to such species as Fin whales, Sei whales and Sperm whales.

(A) Regarding Fin whales, Fig. 2 shows coordinate with the transversal axis indicating months and the vertical axis indicating body-lengths of foetuses. The black dots representing the foetuses in our material, are placed on the coordinates according to their size and the time of the catch. Each signs of () represents an average body-length per ten days. Due to the scantiness of our material, tracing these signs of () would produce too rugged a curve. So we have shown an average body-length per month by small circles, and prepared a growth curve based on them. We have no foetus in our material for the month of November and of December. So

Fig.

2

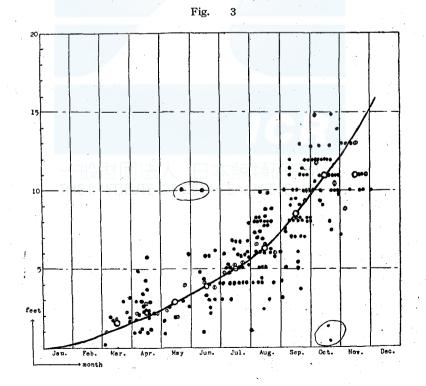


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## Kazuhiro Mizue and Hisako Jimbo

we are obliged to prolong the growth curve which has already shown a clear tendency by the time it reaches the month of July-so as to complete the year's curve. If we are to apply the body-length of Fin whales at delivery, in the South Semisphere, 21—22 feet, determined by Mackintosh & Wheeler and Matthews, to our growth curve, the size will find its place on the curve at the beginning of January next year. The pairing period according to our growth curve comes in January. Therefore, if we are to assume, and we have some reason to believe, that the boby-lengths at delivery in both the semispheres are the same, the duration of pregnancy is twelve months and one decade. There is six months' variation in both the periods of pairing and of delivery between the South and the North Semispheres. Our growth curve has roughly two months' breadth.

(B) The coordinates in Fig. 3 have been prepared in the same way as those of Fin whales, and show the growth of foetuses of Sei whales. The signs of () represent average body-lengths per ten days, and the circles those per month. The records of foetuses in the months of March, April and May are exclusively those of the Bonin Islands sea area because there is no foetus found in other sea areas during that period. The season for



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Sei whales in the Bonin Islands sea area lasts only these three months and the foetuses found then are still very small in size as is shown in the curve. In July, August and September pregnant whales are captured mainly in the Sanriku sea area, and in October, mainly in the Hokkaidō Pac fic sea area. These shifts in seasons and whaling grounds as well as the different growth stages of foetuses of the catch lead us to suppose that the Sei whales in our adjacent waters migrate according to the course and season above mentioned. If we had whaling landstation at the south of the Bonin Islands sea area we should get still amaller foetuses.

During the whaling operations in this season, some marking were discharged in the Bonin Islands sea area. The result of this experiments will clarify the course and the season of migration in several years.

The growth curve of Sei whales was shown in Fig. 3, run smooth upwards as far as the tenth month, after that the dots are hardly traceable. So we have prolonged the curve already drawn as in the case of Fin whales. According to this curve, foetuses measuring 13 feet are seven in number while those of 14 feet and of 15 feet number one and two respectively. From this we assume foetuses at delivery measure 14 to 15 feet and that delivery time is at the end of November. Like Fin whales, Sei whales have their pairing period in January, so that the duration of pregnancy is less than 11 months. Our growth curve of Sei whales has three months' breadth.

Sei whales are to be subdivided into totan whales (there are many designs like the surface of galvanized iron plate in abdomer.) and Sei whales proper. Totan whales are larger sized and supposed to belong to a Northern stock while Sei whales are of a Southern stock and mostly captured off the coast of Wakayama Prefecture. The differences of these two kinds of Sei whale have been a subject of many discussions but not yet put to any morphological investigation. Fig. 3 show four dots placed far apart from the main group. These four foetuses are all found in animals captured off Wakayama Prefecture. This will give rise to the question whether the Sei whales in that neighbouring sea are of different species from that of Totan whales or of the same species taking different course in their migration. The problem will be clarified by the further study of the locality of the catch, of the distribution of body-lengths according to the sea areas and by experiments by marking as well as by morphological investigations.

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(C) In order to ascertain the courses of migration of Sperm whales in the adjacent waters of Japan we marked the locality of the catch of each individual whale on a chart, which has led us to suppose that Sperm whales in those waters live in several different groups. So we have made our study of their foetuses according to the four different sea areas, the Bonin Islands, the Kinan, the Sanriku and the Hokkaidō-Pacific sea areas. In other respects, the method of our study is the same as in the case of Fin and Sei whales; we have drew the growth curves of the foetuses of Sprem whales by the average body length per ten days and that per month.

In Fig. 4 you will find a growth curve of foetuses of Sperm whales in the Kinan sea area though the cruve is imperfect for lack of material. The curves in Fig. 5 shows the growth of the foetuses in the Bonin Islands sea area. The whaling season here lasts only three months, March, April and May, and yet we can clearly discern that foetuses group themselves Figs. 6 and 7 show the growth curves of along the two growth curves. the Sanriku and the Hokkaido-Pacific sea areas respectively, either of which show two groups of foetuses with different tendencies.

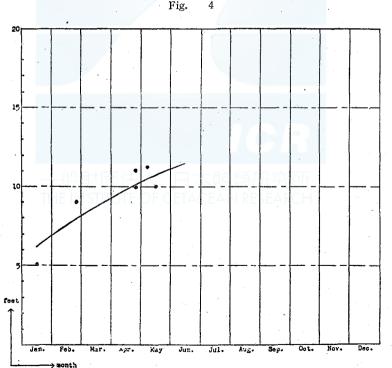
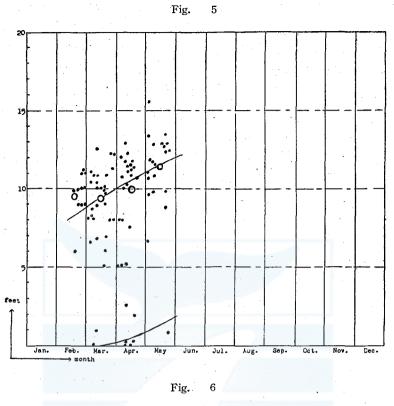
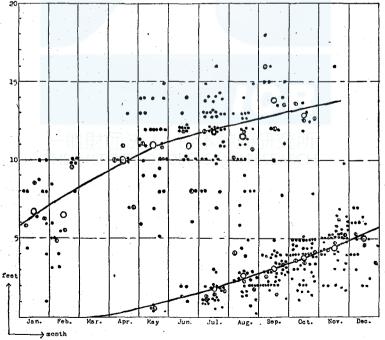


Fig.

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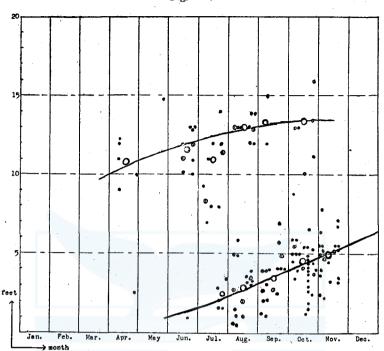
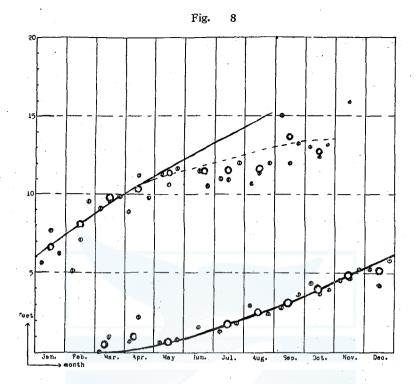


Fig. 7

Comparison of the curves of these four sea areas has proved that there is no variation between the tendencies of each curve. So, upon the combined data of these curves we have formed body-length distribution curves per months. Then tracing the vallegs appearing in the monthly curves, we calssified the foetuses into two groups, upper and lower. We have prepared a growth curve for each group of the foetuses based upon average body-lengths per ten days and per month in the same way as before. When we arrange the curve of the lower group to the left of the curve of the upper one we can complete the growth curve of the foetuses of Sperm whales. (Fig. 8)

According to this curve the pairing period of Sperm whales in our adjacent waters is March. As for the body-length at delivery, Wheeler made a report of a Sperm whale immediately after birth measuring 13.4 feet, captured in the neighbouring waters of Bermuda Island, the Atlantic, in 1933. So did Bennett of the one measuring 14.2 feet in 1840, and reported by Matsuura two measuring 17 feet. Figs 5, 6 and 7, foetuses measuring 13 feet are found in numbers but those of 14 feet are comparatively few while those of 15 feet are rare. From this we assume that the body-length Statistic Study of Foetuses of Whales

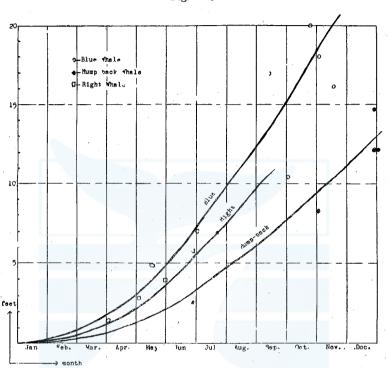


at delivery is 14—15 feet. The growth curve in Fig. 8 shows that the size of 14—15 feet is attained to at August, so that the delivery time of Sperm whales is to be determined as August. However, this growth curve whose composing members are rather homogeneous at first, grows in breadth and at the time of delivery it has the breadth of two months and a half. So the delivery period of Sperm whales may practically be determined as lasting from the end of July to the beginning of October.

If we are to determine the duration of pregnancy of Sperm whales from the delivery period shown on the curve of the foetuses, which is August, it lasts 17 months. The pairing period of Sperm whales differs from that of Baleen whales by two months, and the duration of pregnancy here determined is much longer than theirs, which naturally brings about a different delivery period.

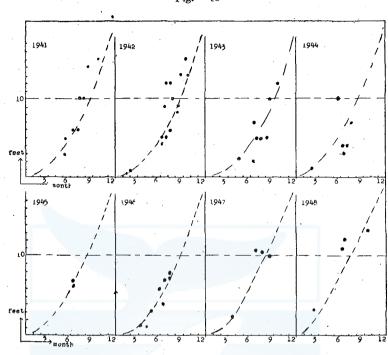
(D) In this present study, we have determined the pairing period, the duration of pregnancy, the delivery period and the body-length at delivery of Fin whales, Sei whales and Sperm whales. Fig. 9 shows the distribution of the foetuses of Right, Blue and Hump-back whales in our material. The growth curves coincide, with six months' variation, with those of Blue and

Hump-back whales in the Antarctic prepared by Mackintosh & Wheeler and Matthews. The pairing period of Baleen whales in the adjacent waters of Japan and perhaps in the North semisphere at large is January.



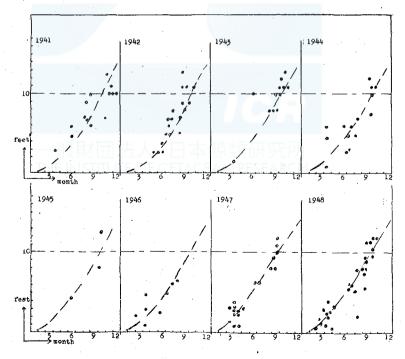
In Figs. 10, 11 and 12, you will find the seasonal distribution of foetus' body-length of Fin, Sei and Sperm whales in successive eight years.

Fig. 9



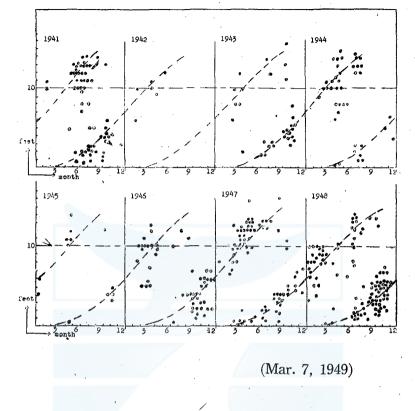






# Kazuhiro Mizue and Hisako Jimbo





# Biological Survey of Fin and Blue Whales Taken in the Antarctic Season $1947 \sim 48$ by the Japanese Fleet

# October 1948

## by

# Masaharu Nishiwaki and Kazuo Hayashi

# CHAPTER I. Introduction

I. Permission of Whaling for the Season 1947-48.

The whaling in the Antarctic water for the season 1947–48, a second one since the cessation of the War, was granted by the GHQ, SCAP. II. Objects of the Whaling Survey.

The present investigations were carried out aboard the "Hashidatemaru", a whaler of the Nippon Suisan K. K. (Japan Marine Products Co., Ltd.), and the "Nisshin-maru", a whaler of the Taiyo Gyogyo K. K. (Taiyo Fisheries Co., Ltd.) which went out on the permission of the above during 8 December 1947 to 10 March 1948. The movement of the vessels are shown in Fig. 1. During this season, the whales caught were: 713 fin whales (Balaenoptera physalus), 608 blue whales (Balaenoptera musculus) and 2 Sperm whales (Physeter catodon), of which 3 heads of fin were lost by the Hashidate Maru and were, therfore, not included in the investigation. Sperm whales were also excluded from survey.

The chief object of the present survey was the classification of whales caught in the Antarctic as part of the world-wide investigations on cetaceae. On the other hand, this report was drawn up in order to present numerical appreciation of the Japanese whaling fleet. At the same time, data which would serve as bases for the biological studies of the Cetaceae were collected as much as possible. In short, this is a statistical data and any phenomena which might accompany them have been included in the Appendix. Physiology of various organs of the whales and anatomical studies of the foetuses have been omitted.

III. Method of Work

1) Body colour: Blue whales— Colour of the skin; size, clarity, number and distribution of pale spots on the skin; size, clarity and number of white flecks; location of ventral grooves; clarity of striation; were all observed by naked eyes. Fin whales— Shading of the skin colour and extension of the pigmentation to the ventral surface from the back were observed by naked eyes.

2) Measuring of the various parts of the body were made by steel tapes on those parts described by Mackintosh and Wheeler in their Discovery Reports, i. e.:

- (1) Total length.
- (2) Lower jaw; projection beyod tip of snout.

(3) Tip of snout to blow-hole.

(4) Tip of snout to angle of gape.

(5) Tip of snout to center of eye.

(7) Eye to ear (centres).

(8) Notch of flukes to posterior emagination of dorsal fin.

(9) Flukes, width at insertion.

(10) Notch of flukes to anus.

(11) Notch of flukes to umbilicus.

(12) Notch of flukes to end of ventral grooves.

(13) Anus to reproductive aperture (centres).

(14) Dorsal fin, ventral hight.

(15) Dorsal fin, length of base.

(16) Flipper, tip to axilla.

- (17) Flipper, tip to anterior end of lower border.
- (18) Flipper, length along curves of lower border.
- (19) Flipper, greatest width.
- (20) Severed head, condyle to tip.
- (21) Skull, greatest width.
- (22) Skull length, condyle to tip of premaxilla.
- (23) Flipper, tip to head of humerus.
- (24) Tail, depth at dorsal fin.
- (25) Flukes, notch to tip.
- (26) Flukes, total spread.

Since (2), (4), (22) and (26) seemed to contain a large amount of measuring errors, these were omitted in the present investigation, and following measurements were added :

- (27) Breadth of the body, back to front (at tip of flipper).
- (28) Height of the body when lying on its side.
- (29) Greatest circumference.

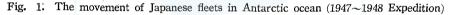
(30) Tail flukes, from tip to tip.

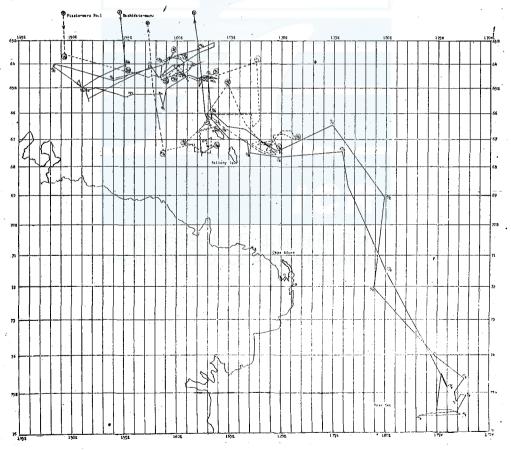
(31) Lower jaw bones, length along curve of border.

3) The determination of the weight of various parts of whale body was carried out with a platform scale, cutting the parts down to the size (about 50 cm<sup>3</sup>) to fit the scale and adding up the result. Loss of blood and body fluids had to be tolerated but small pieces sawed off were collected as much as possible and also added up.

Investigations on testes, overies, mammary glands, food, thickness of blubber, etc., were carried out by biologists apointed by the Japanese Government and several men under their guidance so as to eliminate the difference by individual investigation.

IV. Activities of the Japanese Whaling Fleet in the Whaling Ground (South of Lat,  $60^{\circ}$  S, east of Long,  $90^{\circ}$  E., and west of Long  $170^{\circ}$  W,) granted for the Season 1947-48.



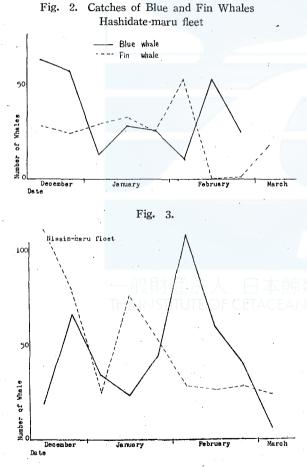


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The Hashidate-maru Fleet left on 6 November 1947, and the Nisshinmaru Fleet left Yokosuka on the same date arriving in the ground on 5 and 7 December 1947, respectively. Their activities after those dates are as shown in Fig. 1. The Nisshin-maru confined her activities to an area enclosed by Lat. 63° to 68° S., and Long. 155° to 172° E., but Hashidatemaru went further into the Ross Sea for operation. The present expedition found that the opening to the Ross Sea was not in the direction of the Balleny Is. as usual but far to the east and the fleet found it difficult to find the opening.

V. The Amount of Catch by the Japanese Fleet inside the Whaling Ground granted for Season 1947–1948.

The Amount of whales caught by the expedition, is shown in Figs. 2 and 3 by month. As can be seen from these Figures, both the Hashidate-



maru and Nisshin-maru caught many blue whales at the beginning of the season but these animals began to grow less by January and the fleets decided to go after fin whales. Nisshin-maru continued to operate in the same area and encountered the largest catch of fin whales during February. Hashidate-maru, however went into the Ross Sea and operated for blue whales. Fig. 4 shows the amount of whales caught according to different areas which is shown by the same scale as used in the Whaling Ground of the Antarctic Ocean, 6th Series (1) of Whale Resurce Data published by the Japan Association of Whaling Industry.

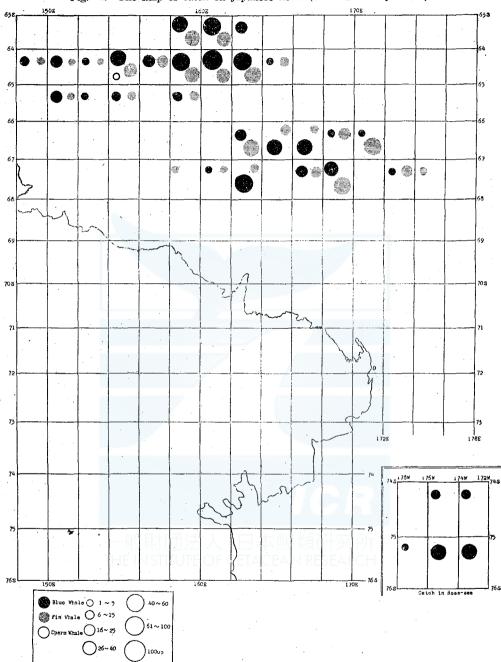


Fig. 4. The map of catch on Japanese fleets (1947~1948 Expedition)

# VI. Material and Data.

The number of whales examined during the present investigations on the Hashidate-maru and Nisshin-maru was as follows:

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		Hashidate-maru	Nisshin-maru	Total	
	Male	138	200	338	
Blue Whale	Female	136	236	372	
	Totel	274	436	710	
	(Male	87	176	263	
Fin Whale	Female	124	221	345	
	Total	211	397	608	

Grand total of whales caught 1318 (1014 B. W. U.)

Investigation data on individual whales are to be published by the Marine Products Bureau aside from the investigation report.

Average time elapsed between the killing and treating of whales was as follows:

Fleet	Hashidate-maru			Nisshin-maru				
Species	Bule whale Fin whale Bule wh		whale	Fin whale				
Sex	Male	Female	Male	Female	Male	Female	Male	Female
Average time interval	7' '36''	7' 06''	4' 57''	5' 47''	6' 16''	5' 51''	3' 55''	4' 23''

Unless otherwise stated, all data used in Figures and Tables refer to the total figures and data obtained by both fleet, i. e. results obtained by the Japanese Whaling Fleet.

VII. Abbreviations used.

Abbreviations used in this report and in the appendix are as follows: In the "Ossification of vertebrae":

1. Number and state of epi. in thoracic (or lumber) refer to the number and state of epiphyses in thoracic series (or in lumber series) of vertebrae.

2. In the column for state, ank. refer to anklosed, those not stated refer to "not ankylosed".

On stomach contents:

1. Size: L=Large, ca. 5.0 cm. and over (from rostrum to tail).

M = Medium, from ca. 4.0 cm. to 5.0 cm.

S=Small, up to ca. 4.0 cm.

X=Mixture of conspicuously different sizes.

?=Sizes indistinct due to high degree of digestion.

2. Quantity: 0 = Empty.

r=Very small amount of krill

rr=Small amount of krill

## Biological Survey of Fin and Blue Whales Taken

## rrr=Moderate amount of krill

R=Large amount of krill

3. Degree of freshness: f = Almost digested

ff=Half digested

fff=Fresh

F=Very fresh

On the thickness of blubber:

- Point 1— The point on the horizontal cut side of the body (at the position of lateral line in fish), where it intersects a vertical line from the dorsal fin.
- 2. Point 2— The point on the vertical cut near the earhole, where it intersects a mid-dorsal line.

External parasites: #=Much infected or fully developed (diatom)

#=Moderately infected or partially devloped (diatom)

+=Scarcely infected or developed (diatom)

No notation=Not infected in naked eye.

White scar:

Number - 0 = None

1 = Few

2 = Scarce

3 = Normal

4=Numerous

5 = Very numerous

Colour of the blue whale: Pale spots, whits flecks and striation.

Number— 0=None State of distinction— I=Indistinct

1=Few 2=Scarce II=Distinct

III=Very distinct

3=Normal

4=Numerous

5 =Very numerous

Colour of the fin whales:

Body colour- N=Normal

B = Blackish

Extension of pigmentation on ventral grooves-

U=Upper

(11–13 stripes of ventral grooves upward from navel line)

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N=Normal

L=Lower

Tongue of pigmentation behind anus-

+=Present

-=Absent

Meeting of pigmentation in front of tail flukes-

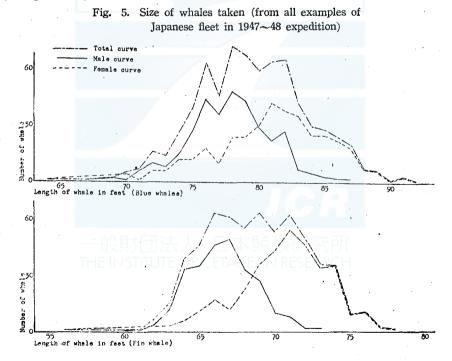
+ =Fused

-=Not fused

## CHAPTER II. Composition of Whales

I. Composition of Whales Taken.

The formation of whales taken by the present expedition is shown in Fig. 5. The modes of blue whale male are 76, 78 and 82 feet while that of female is 81 feet. Those in fin whale male and female are 67 and 71 feet, respectively.



# II. Sex Ratio.

In blue whale, the sex ratio was 47.6% male while that in fin whale was 43.3% male. The sex ratio of the foetuses was 48.8% male in blue whales and 57.4% male in fin whales.

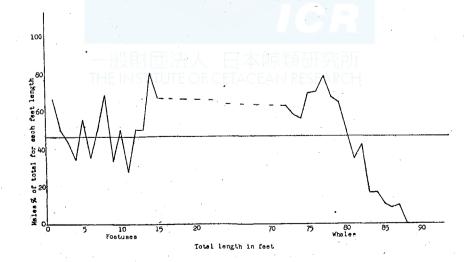
Figs. 6 and 7 show the sex ratio according to body length. In blue whales, there were about 60% of male in whales of under 78 feet where as there are none in those over 88 feet. In other words, male blue whales do not grow to over 88 feet in length, at least, under the present investigations.

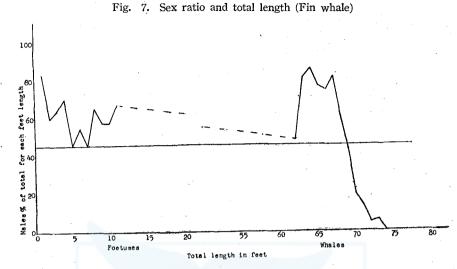
In fin whale, the sex ratio of foetuses was 57.4% male (ca. 60%). About 60% of whales under 69 feet were male, but the number dropped suddenly in those over 70 feet in length.

The reason for this sudden fall in the number of males is that the females normally reach a greater length than the males, as is shown graphically in Fig. 5, in which the number of whales examined is plotted against their length.

The whalers naturally select the largest whale to catch, which so often happens to be a female so that the figures as a whole are not representative of the whale population in that region. However, the sex ratio worked out from data obtained on whales up to 78 feet in total length, which may be expected to show the proportion of males and females more correctly, show that the sex ratio is about 60% of males and not 43.3%, as would appear if the total catch alone is considered.

These figures show the sex ratio among whales of similar length but not necessarily of those of similar age. The proportion of males among those of similar age is probably less than 90%. The males stop growing after reaching a length of about 80 feet and consequently, are removed Fig. 6. Sex ratio and total length (Blue whale)





from the length classes of their contemporary females which grows to more than 80 feet. The length classes containing those of 80 feet and less, must, therefore, contain the males of an age equal to that of the females over 80 feet in length. Among whales of similar age, therefore, the primary sex ratio found in the foetuses is probably steadily declining from the high proportion of males to something nearer equality as the whales grow older. III. Composition of Whales taken as Classified by the "International

whaling Statistics Board".

Female

The size of whales when reaching sexual maturity was taken from the works of Mackintosh and Wheeler, the blue and fin whale generally reaching their maturity at the following length:

Blue whale- Male

63 feet, 8 inches (63 feet)

65 feet, 7 inches (65 feet)

Classification	Number	RCH Ratio (%)
Group 1 (70 feet and less)	9	1.27
Group 2 (71-85 feet)	639	90.00
Group 3 (86 feet and over)	62	8.73
Total	710	
Immature male	28	8.58
Mature male	310	91.42
Immature female	61	16.40
Mature female	311	83.60 '
Total No. of immature animal	89	12.54
Total No. of mature animal	621	87.46

Table I. Composition of Blue whales

Biological Survey of Fin and Blue Whales Taken

Fin whale- Male

Female

74 feet, 2 inches (74 feet) 77 feet, 0 inches (77 feet)

77 feet, 9 inches (77 feet)

Classification	Number	Ratio (%)
Group 1 (55 feet and less)	0	0
Group 2 (56-65 feet)	110	18.09
Group 3 (66 feet and over)	498	81.91
Total	608	
Immature male	6	2.28
Mature male	257	97.72
Immature female	13	3.77
Mature female	332	96.23
Total No. of immature animal	19	3.12
Total No. of mature animal	589	96.88

Table II. Composition of Fin whales

GHAPTER III. Composition of Male Whales

I. Sexual Maturity of Male Whales.

The sexual maturity of male whales are determined by the following:

1. Size of the penis

2. Size of the testis

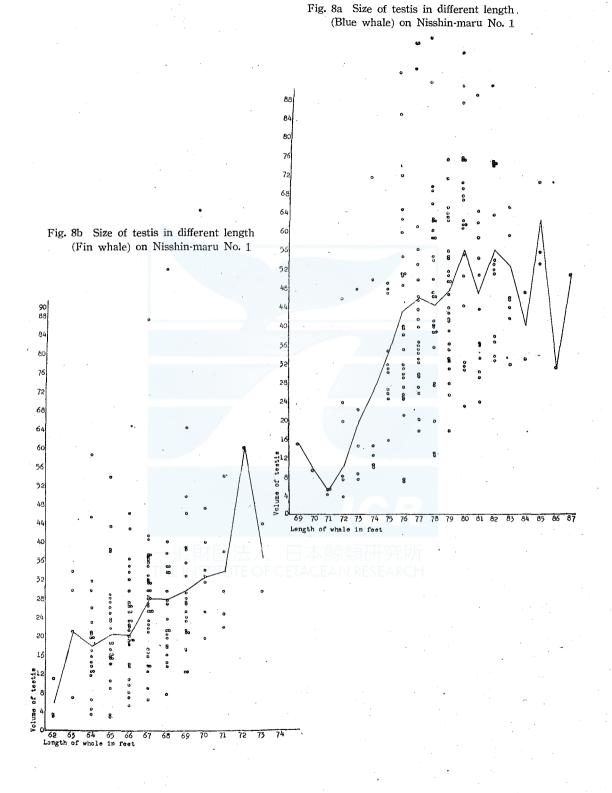
- 3. Weight of the testis
- 4. Histology of the testis.

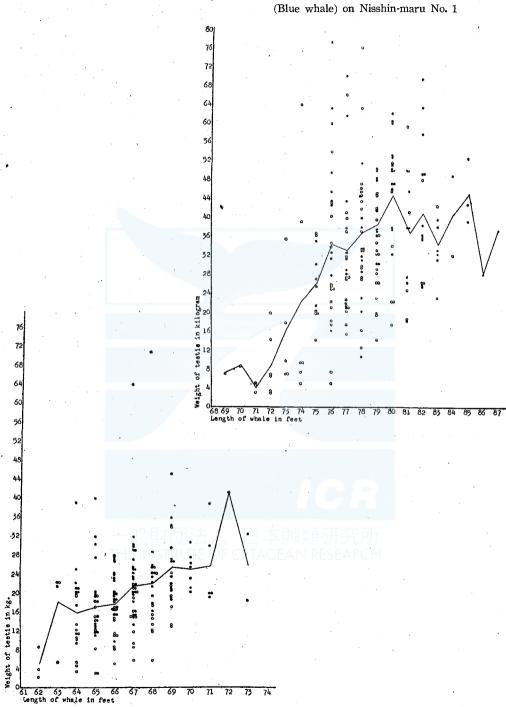
The present investigations took the weight of the testis as a chief indication of maturity with additional considerations on other points.

II. Relationship beween the size and weight of testis.

In their Discovery Report, Mackintosh and Weeler classified male whales according to the size of the testis ("the size of the testis represented by a number obtained by multiplying together the length, breadth and depth measured in centimetres").

In order to obtain the relationship between the size and weight of the testis of male whales taken by the Nisshin-maru, various parts were plotted against body length and the average values obtained are shown graphically in Figs. 8a, 8b, 9a and 9b. Figs. 10 and 11 show the comparison of their size and weight. Naturally, these Figures show that both weight and size curves have the same tendencies and seem to show that the weight method in which values taken from fresh ones can be used, is much more exact than the size method which necessitates calculations for tabulation.





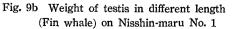
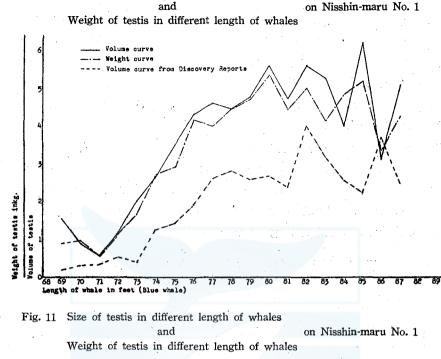
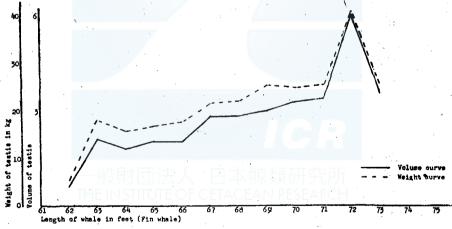


Fig. 9a Weight of testis in different length (Blue whale) on Nisshin-maru No. 1

Fig. 10 Size of testis in different length of whales





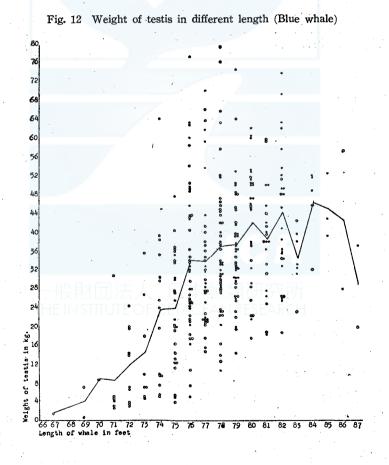
III. Weight of the Testis.

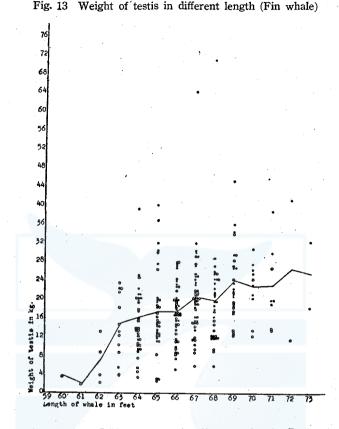
The weight of the testis of all whales taken by the Japanese fleet in the present whaling season were taken to observe overall tendencies.

As shown in Fig. 12, the development curve of testis in blue whales. is very gradual at first but becomes rapid when the body length reaches. 73-75 feet, and again becomes slower thereafter. In male blue whales.

of over 83 feet in length, decrease of sexual function can generally be seen. This point constitutes one of the biggest difference from the curve of the number of corpora lutea in female whales. These tendencies observed in the present investigations coincides well with the reports of Makintosh and Wheeler as found in p. 406 of their Discovery Report, Vol. 1. This evidence is shown by the average curve taken from their Fig. 139 (p. 406) plotted on Fig. 10.

The same tendencies can also be seen fin whales. As can be seen from Fig. 13, the develoment curve makes a sudden upturn around a body length of 62 feet and takes a natural course of development thereafter. The fact that a remarkable decrease phenomenon cannot be seen must be due to the failure to catch large, aged whales.



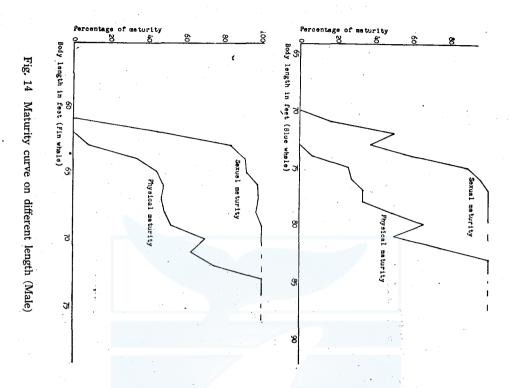


IV. Sexual and Physical Maturity according to Body Length.

The maturity of males according to their body length is shown in Fig. 14. The sexual maturity was classified only by the weight of testis as is done and not by macro- or microscopic observations of the testis. If the combined weight of both testis was less than 10 kg in blue whale and 5 kg in fin whale, they were considered immature.

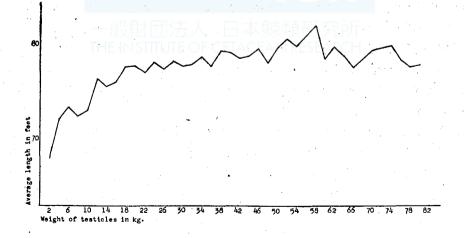
The physical maturity of males was considered complete when all their epiphyses of vertebral column were fully ankylosed. They were considered immature if thoracic epiphyses were not ankylosed even if caudal and lumber ones were.

Considered from these points, the sexual maturity of Blue whales is reached when the body length is about 74 feet, and the physical maturity at about 80 feet. The sexual maturity in Fin whales is reached at about 63 feet of body length and the physical maturity at about 70 feet.



V. Average Length of Whales according to the Weight of Testis.

Fig. 15 and 16 show the average length of whales according to the weight of testis from which it can be seen that in blue whales the body length of whales having 10 kg testis is about 73 feet but the length suddenly increases to 76 feet in those having 12 kg testis. In fin whales, Fig. 15 Average length of whales in each class of weight of testicles (Blue whale)



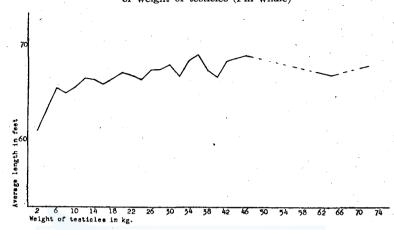
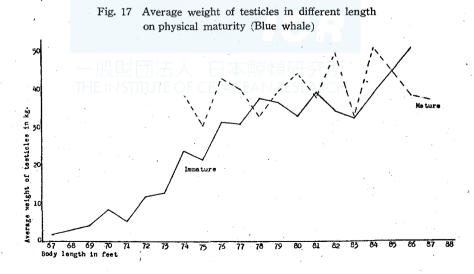


Fig. 16 Average length of whales in each class of weight of testicles (Fin whale)

the curve for whales having testis weighing up to 6 kg take a sudden upward trend which then becomes gradual. The body length of whales having 4 kg testis is about 63 feet, and that of 6 kg testis about 66 feet.

The foregoing is a good evidence that the classification of maturity by body length is based on the classification by testicular weight.

Figs. 17 and 18 show the curves for average testicular weight at different body length according to physical maturity. The physical maturity curve in Fig. 14 thow the ratio of the mature and immature in the former data. These figures also furnish good evidence of the various factors as stated above. It seems correct to observe that the body length of male whales at its maturity, as seen from Fig. 14, is 79 feet in fin whales.



Biological Survey of Fin and Blue Whales Taken

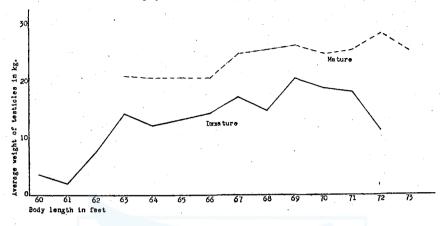


Fig. 18 Average weight of testicles in different length on physical maturity (Fin whale)

## CHAPTER IV. Composition of Female Whales

I. Factors decisive in Determining Sexual Maturity of Female Whales.

In order to investigate the sexual maturity in cow whales, following points must be considered :

- 1. Presence of a foetus
- 2. Presence of corpora lutea in the ovaries
- 3. Size of the uterus
- 4. Size of the ovaries
- 5. Weight of the ovaries
- 6. Condition of the ovaries follicle
- 7. Condition of the mammary glands.

However, the present researches took chief account in the presence of copora lutea in the ovaries with additional considerations on 1, 5, 6 and 7. II. Relationship between Body Length and the Number of Corpora Lutea.

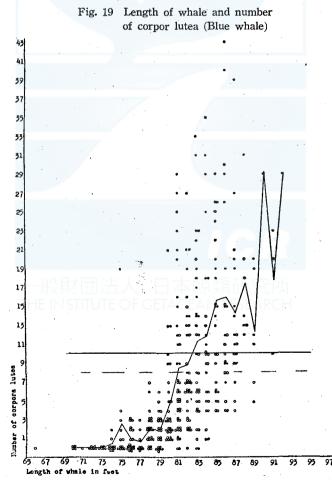
Figs. 19 and 20 show the distribution of the number of corpora lutea according to body length. In Blue whales, the curve is gradual up to body length of 79 feet (uncer 3 corpora) but takes a sudden upward swing after it reaches 80 feet in length. The same is seen in Fin whales, the curve being gradual up to 65 feet body length (under 2 corpora) but becomes suddenly high at 66 to 67 feet length.

III. Average Length of Whales according to the Number of Corpora lutes. Figs. 21 and 22 show the curves for average length of whales accor-

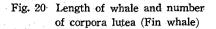
ding to the number of corpura lutera observed. In blue whales, the average length of body for those having 3 corpora lutea is 79 feet but those having 4 corpora suddenly goes up to 82 feet. In fin whales, those having 3 corpora lutea is 68 feet and those with 4 corpora, 70 feet. From Figs. 21 and 22 it can be seen that the average body length according to the number of corpora lutea is 84 feet in blue whales and 72 feet in fin whales. In other words, the standard body length in female blue whales is 84 feet and that in fin whales, 72 feet.

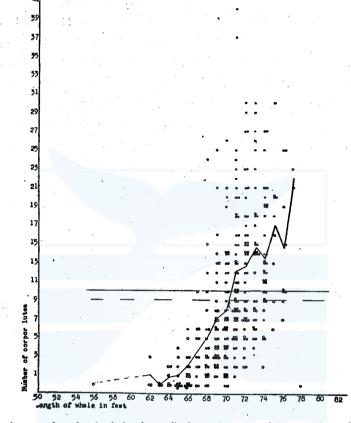
IV. Relationship between Sexual Maturity and body Length.

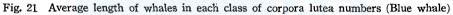
F g. 23 shows the percentage curve of sexually mature (possessing corpora lutea) animals according to body length from which it can be seen that sexual maturity in blue whales is reached at about 78 feet and that in fin whale females at 67 feet.

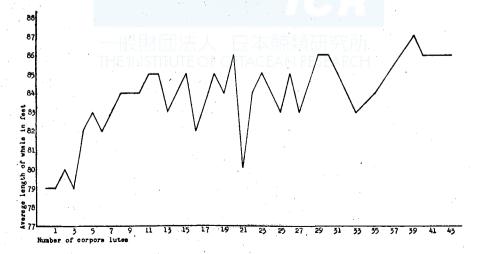


Biological Survey of Fin and Blue Whales Taken









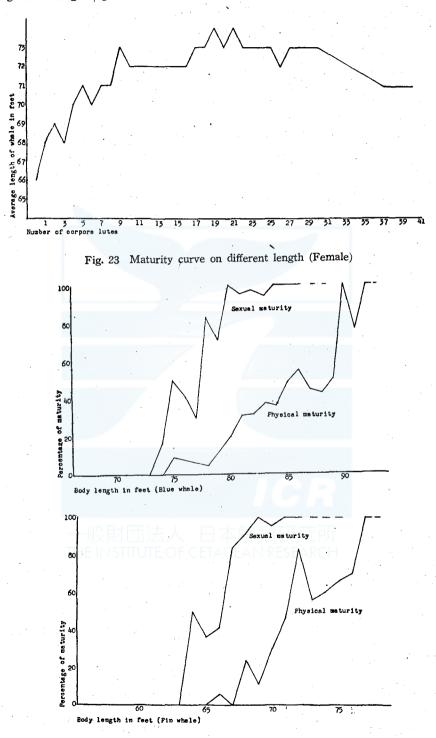


Fig. 22 Average length of whales in each class of corpora lutea numbers (Fin whale)

Fig. 24 shows the classification of female whales, indicated in the composition of whales taken, according to sexually immature, pregnant, resting and lactating by their body lengths.

In blue whales, the height of curves for the immature and the mature (resting and pregnant) are at different places. The trend of the curves for resting and pregnant match well and show that approximately half of the mature whales are pregnant.

In fin whales, there are a very small number of immature females due, probably, to the fact that whalers generally aim to catch large sizes. Approximately one-third of the mature females are pregnant and the trend of the curve by their body lengths is similar to the above.

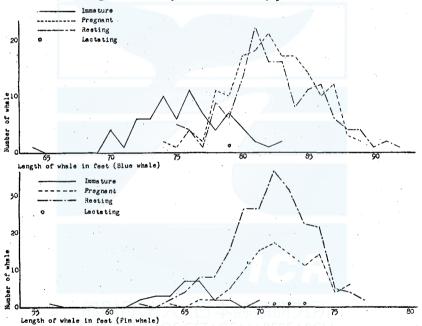


Fig. 24 Maturity of female in all Japanese fleet

V. Monthly Ratio of Pregnant Whales.

Fig. 25 shows the monthly ratio of pregnant whales and endorses the views of previous workers in the pregnancy rate decreases monthly from December to March. Following figures show the pregnancy rate as a whole:

Table III

Whale speces	Pregnant	Resting	Lactating	Total Mature Female
Blue whales	$162 \\ 53.29\%$	131 46.38%	1 0.33%	304
Fin whales	$101 \\ 32.17\%$	210 66.88%	3 0.95%	314

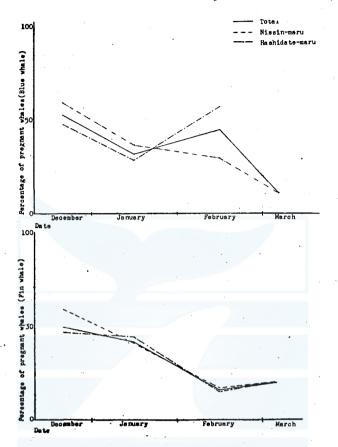
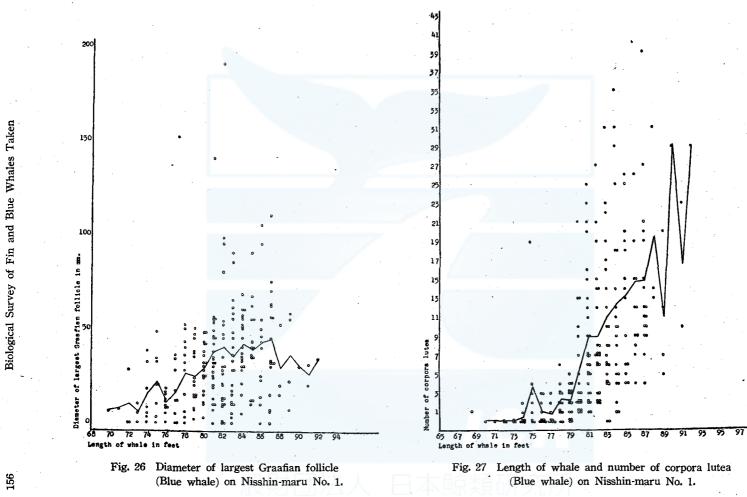


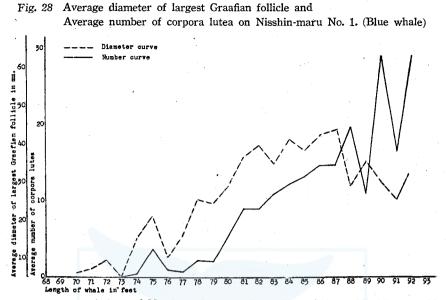
Fig. 25 Percentage of pregnant whales in the catch by month (contain the whale, which has function corpora lutea & no foetus)

VI. Relationship between the Number of Corpora Lutea and the Largest Graafian Follicles.

Figs. 26 to 31 show the results of observations on the relationship between the largest Graafian follicles and the number of corpora lutea. Due to the time allowed for handling, this work was carried out on 236 heads of blue whales and 221 heads of fin whales taken by Nisshin-maru.

Figs. 26 and 29 show the diameter of the largest Graafian follicle in each animal and the average in each body length class. Figs. 27 and 30 give the relation of the number of corpora lutea according to the body length (same as Figs. 19 and 20 but only on data from those taken by the Nisshin-maru). Although they fail to show, at first glance, any presence of a relationship between them, their average curve alone plotted on the same diagram show, as in Figs. 28 and 31, that there is a tendency of coinci-







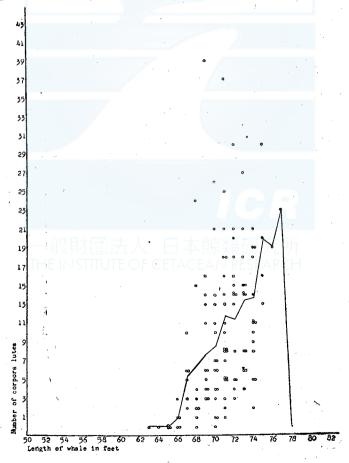
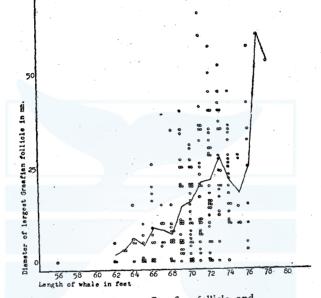
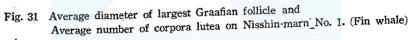
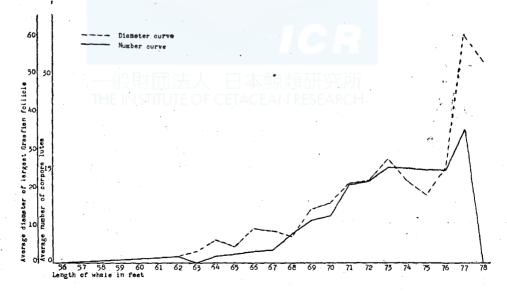


Fig. 30 Diameter of largest Graafian follicle (Fin whale) on Nisshin-maru No. 1.





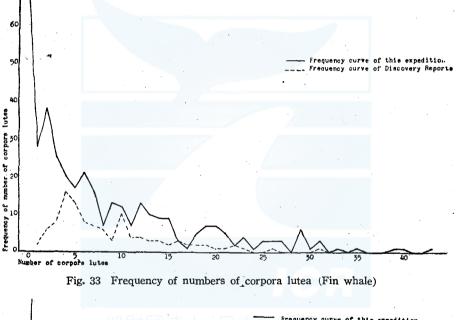


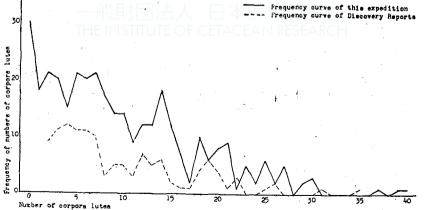
dence. In other words, the appearance of the largest diameter in Graafian follicles appear with the increase in the number of corpora lutea, i. e. with advancing age.

VII. Frequency Curve for the Number of Corpora Lutea.

Figs. 32 and 33 show the frequency curve for the number of corpora lutea the peaks for which in blue whales occur at 2, 6, 9, 12, 15 and 19 (dotted lines show the value given in the Discovery Report). These peaks in fin whales occur at 2,, 5 7, 10, 14, 18 and 21, which coincide well with the peaks for fin whales of 1, 5, 7, 10, 13 and 15 obtained by summari-

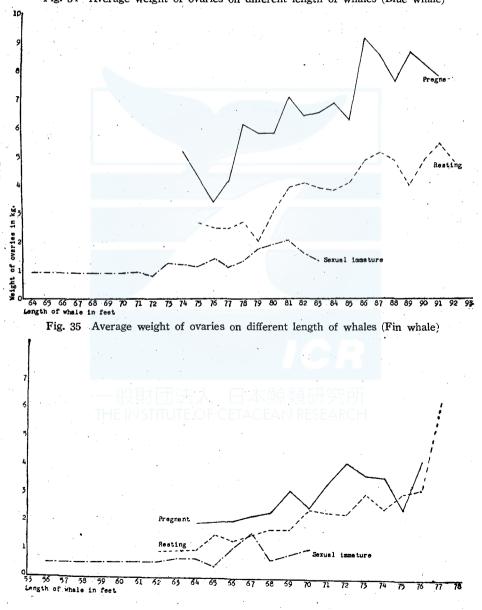
Fig. 32 Frequency of numbers of corpora lutea (Blue whale)





zation of various values obtained for Antarctic fin whales. These curves for fin and blue whales show that, especially in fin whales, there are small number of comparatively aged whales with over 13 corpora lutea. On the whale, there are more number of aged animals which seems to indicate the fact that the whaler go after larger animals when the composition of whales in general is getting younger.

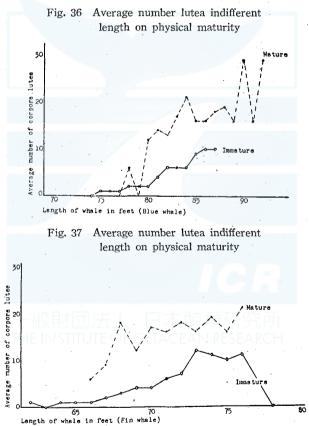
VIII. Relationship between Sexual Maturity and the Weight of Ovaries. Fig. 34 Average weight of ovaries on different length of whales (Blue whale)



Figs. 34 and 35 indicate the weight of ovaries in sexually immature, resting and pregnant whales classified according to their body lengths. This coincides will data funrnished in the Discovery Report and needs no further comment.

IX. Average Number of corpora lutea according to Body length.

The number of corpora lutea averaged by body length in physically mature whales is shown in Figs. 36 and 37 (Cf. Figs. 19 and 20), Fig. 25 shows the physical maturity curve given by the percentages of the above. From these figures, it can be said that the body length of physically mature animals is about 85 feet for blue whales and about 71 feet for fin whales.

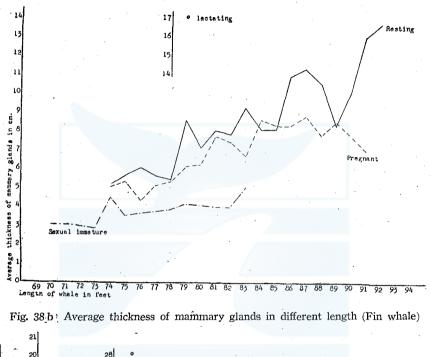


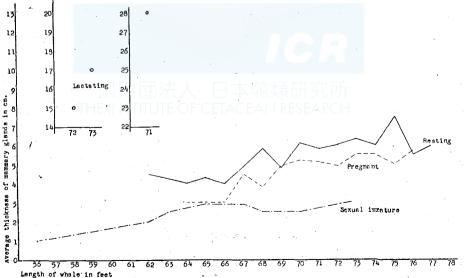
## X. Mammary Glands.

The present investigations were carried out on the colour and thickness of mammary glands on 372 heads of blue whales and 345 heads of Fin whales, values taken being the average of them all. Those classified as

#### Biological Survey of Fin and Blue Whales Taken

lactating are animals from which even a drop of milk was observed during treatment, others being classified as "not lactating." The latter was again divided into pregnant, resting and immature according to the condition of the ovaries; e. g. those possessing functional corpora lutea classed as the "pregnant" (functional corpora lutea of pregnancy and of ovulation were Fig. 38 a Average thickness of mammary glands in different length (Blue whale)





not classified), those possessing no corpora lutea in the ovaries as the "immature" and others as "resting". Figs. 38 and 39 show these averages according to body length. It is noted that both the immature and other curves increase with the increase in body length while that for lactating is in a place by itself with no relation to the former.

There are always a great variations in colour of mammary glands but they have been averaged as follows:

As a whole, the colour of mammary glands in young whales is pale which increases in tone as the whales grow older. In aged animals, the colour becomes deep brown. The colour of mammary glands in lactating whales is orange tinted, irrespective of their age. This tendency is in keeping with the fact that the thickness of mammary glands and that of blubber always change irrespective of the age of whales.

Following table shows the percentage rate in colour of mammary glands in blue whales.

Walls W. Calana of management aloude in blue inholes

Nisshin-ma	aru							
		White	Pink	Ivory	Cinnamon	Reddish Yellow	, Tawny	Brown
mmature	No. %	10 28.6	7 20.0	7 20.0	6 17.1	3 8.6	2 5.7	
Resting	No. %	2 2.1	3 3.1	3 3.1	22 22 <b>.</b> 9	$25 \\ 26.1$	34 35.4	7 7.3
regnant	No. %	2 2.1	$2 \\ 2.1$	5 5.2	23 23.7	22 22.7	38 39.2	5 5.2
actating	No. %						$1 \\ 100$	· ,
Hashidate-	maru		,, ,		•			· · · · · · · · · · · · · · · · · · ·
	• [		White	Pinl	k Cinna	amon	Tawny	Brown
Immatu	re	No. %	14 42.4	3 9.1	. 15 45		1 3.1	
Resting		No. %	1 5.9	4 23.3	3		9 52.9	3 17-3
Pregnan	it	No. %	2 3.0	10 15.2	29 42		11 16.7	$\begin{array}{c} 14\\ 21.2 \end{array}$
Lactatin	g	No. %				1		

Following table shows the percentage reate in colour of mammary gland in fin whales.

Table V. Colour of mammary glands in Fin whales

Nisshin-maru

		White	Pink	Ivory	Cinnamon	Reddish Yellow	Tawny	Brown
Immature	No. %	13 62.0	4 19.0		4 19.0		•	
Resting	No. %	5 3.8	4 3.0	3 2.3	46 34.6	40 30.2	31 23.3	4 3.0
Pregnant	No. %	$\begin{array}{c}1\\1.6\end{array}$	2 3.1	$\begin{array}{c}1\\1.6\end{array}$	26 42.6	$\begin{array}{c} 17\\ 26.6\end{array}$	15 23 <b>.</b> 5	$2 \\ 3.1$
Lactating	No. %						$\begin{smallmatrix}&1\\100.0\end{smallmatrix}$	

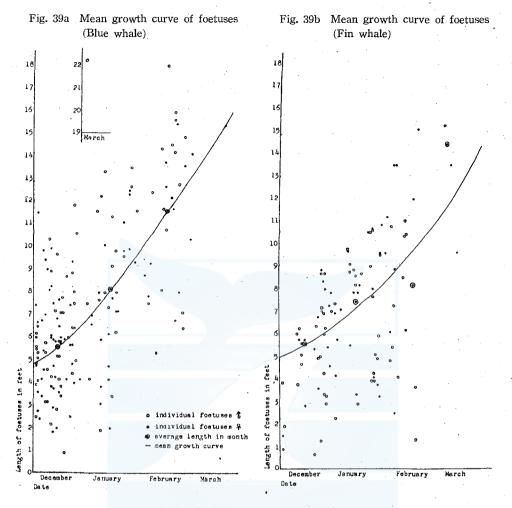
Hashidate-maru

-		White	Ivory	Cinnamon	Tawny	Brown
Immature	No. %	4 44.5	1 11.0	4 44.5		•
Resting	No. %	2 2.9	9 12.9	35 50 <b>.</b> 0	14 20.0	$\begin{array}{c} 10 \\ 14.2 \end{array}$
Pregnant	No. %	2 5.3	6 15.8	$\begin{array}{c}14\\36.8\end{array}$	10 26.3	6 15.8
Lactating	No. %				2 100.0	•

## CHAPTER V. Foetuses

Main growth curves for 162 heads of blue whale foetus and 101 heads of fin whale foetus, classified according to the date their mothers were caught and by body length, were taken. There were nothing new to be learned from them, only endorsing the data contained in the Discovery Report. The monthly mean body length on the curves, especially, coincided with the older data.

The sex ratio of the foetuses is as described in previous chapter. On the whole, there were 48.8% male in blue whales and 57.4% male in fin whales and 57.4% male in fin whales. The smallest foetus was that measuring 10.6 inches in the blue whale and 7.9 inches in the fin whale, while the largest obtained was 22' 2'' in the blue whales and 15' 3'' in fin whales.



CHAPTER VI. External Characters

## I. Colour.

There are many reports on the colour of whales. Observations on the colour of blue whales during the present expeditions followed the notations made by Mackintosh and Wheeler on pale spots, i. e. "most of the body is covered with a pale mottling which consists of small, roughly oval marks of colour which is similar to, but lighter than the blue-grey background", on their clarity and number, and on white flecks on which the two authors say that "those pale spots may also be seen here and there on the ventral grooves". The clarity on the appearance of striation on the ventral surface of tail flukes was also shown by percentages. These results are shown in Table VI. The name of the fleet was noted in because of the fear that

Sex	•	MA	LE			FEM	IALE	· ·
Fleet	Hashid	ate-maru	Nissh	in-maru	Hashid	ate-maru	Nissh	in-maru
Grading of Nos.	No.	%	No.	%	No.	%	No.	%
0	0		0	- 1	0		0	
1	7	52.0	39	19.5	4	3.1	40	18.1
2	17	12.6	2	1.0	26	19.8	. 8	3.4
3	58	43.0	40	20.0	52	39.7	45	19.2
4	44	32.6	84	42.0	43	32.8	93	39.8
5	9	6.6	. 35	17.5	6	. 4.6	48	20.5
Clarity		·	·	. <u>.</u>				··
0	0		0		0		0	
· I	19	16.1	40	20.0	10	8.3	29	12.4
II	34	28.8	139	69.5	.24	19.8	177	75.6
III	15	55.1	21	10.5	87	71.9	28	12.0
White Flee	ks	·					· ·	<u>.</u>
0	0	] _	1	0.5	0		4	1.7
1	17	12.7	71	35.5	20	15.1	63	26.8
2	39	29.1	13	<b>`8.</b> 5	26	19.7	15	6.4
3	48	35.8	35	17.5	53	40.2	64	27.2
. 4	27	20.1	55	27.5	24	18.2	48	20.4
5	3	2.3	25	12.5	9	6.8	41	17.5
Striation								
0	0	·	3	1.5	0	-	4	1.7
I	36	26.9	64	, 32.0	42	31.3	75	31.0
п	43	32.1	102	51.0	39	29.1	115	48.9
III	55	41.0	31	15.5	53	36.6	41	17.5

## personal view may differ in individual cases. Table VI.

On the colour of fin whales, Mackintosh and Wheeler says in their Discovery Report, that "the most obvious feature is that pigment covers the whole of the back and flanks, while the ventral surface remains unpigmented". This shading of body colour over the back and flanks (not darkened due to light and air), extention of pigment over the ventral surface, as follows, were examined:

- i) Extention of the blackened area of the back over the ventral groove as though by brush;
- ii) Extention of the blackened area of the back and flanks in tonguelike form towards anus from the tail; and
- iii) Whether the blackened area of back and flanks meet form either

Sex			MA	LE			FEM	IALE	
Fleet		Hashidat	e-maru	Nisshir	ı-maru	Hashidat	æ-maru	Nisshir	ı-maru
Classifica	tion	No. of whales	%						
Normal co	lour	67	81.7	154	87.5	. 83	68.6	205	92.7
Blackened	colour.	15	18.3	22	12.5	38	31.4	16	7.3
Extention	High	13	15.1	49	27.8	25	20.2	36	16.3
	Norm.	50	58.1	102	58.0	61	49.2	130	58.8-
of Pigm.	Low	23	26.8	25	14.2	38	30.6	55	24.9
Tongue of	+ -	70	81.4	102	92.0	89	72.9	183	82.8
Pigmentation beh. anus		16	18.6	14	8.0	33	27.1	38	17.2
Meeting of Pigmentation	+	73	85.9	99	56.3	90	73.8	110	49,8
in front of flukes	-	12	14.1	77	43.7	32	26.2	111	50 <b>.2</b>

side just in front of the tail flukes.

Table VII shows the number of whales in percentage on above points.

Table VII

As a result of foregoing data, the normal body colour of whales can be summarized as follows:

In blue whales, pale spots do not appear collectively but is dispersed; there are 6 or 7 spots, normally, of a size about  $(4 \times 6)$  cm 2 in an area of 1 m<sup>2</sup>. The clarity of these spots is distinct. The white flecks are also dispersed in an area about 1/3 behind the ventral grooves. The striation is normally distinct and there can be seen no difference between sexes.

In fin whales, the normal body colour is slate-gray, and the extention of pigmentation to the ventral groove is on the 11th or 13th from the navel line, neither higher nor lower. The tongue of pigmentation behind anus is normally present. The pigmentation also normally meets in front of the tail flukes. Here, also, it is hard to distinguish the sexes according to body colour.

Although the distinct white flecks seen in blue whales seem to be due to old age, the difference in body colour due to age is very slight. II. Proportion of Body Length.

At the time of the weighing of whales on the Hashidate-maru, measurement of body proportion was also carried out (Ct. Table IX). According to Mackintosh and Wheeler, the proportion of anterior part of the body becomes larger with the increse of body length, both in blue and fin whales. However, since the measurements made were small and the range narrow in the present investigations, no calculations were made on the mean values and their distribution according to body length group. There were evidence, however, of the proportional increase of anterior portion of the body and decrease in the posterior portion with the increase in age, not necessary with body length. From what measurements that were made during this expedition, no distinctive relationship can be seen between the body length and its proportions.

From the relationship between the number of corpora lutea and body proportion in female whales, as shown in Fig. 40, it can be seen in Nos. 3, 5 and 6, especially in 3 and 5, that the proportionate percentages of anterior portion increase with the increase in the number of corpora lutea. In the measurement of posterior portion, as in Nos. 8, 10, 11 and 12, the proportionate percentage apparently decrease with the increase in the number of corpora lutea. (Numbers used here refer to the investigation Nos. as explained in the "Method of work" in the Introduction).

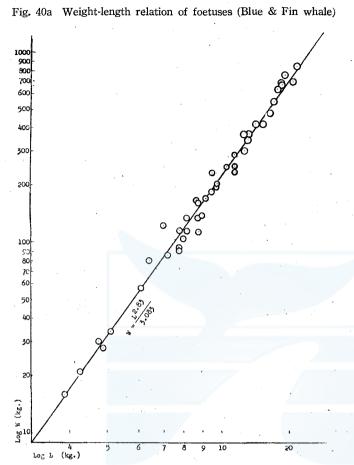
III. Proportions of Body Weights.

Total weight of foetus was measured on 30 heads of blue whales and 16 heads of fin whales. Foetuses were cut into blocks of about 30 to 50 kg. each and weighted on a platform scale of 200 kg. capacity. Some of the body fluid has naturally been lost during the process so that the weight cannot be taken as the true weight.

The relationship between body length, L (ft.), and body weight, W (kg.), obtained by the least squares, from all the measured, values on blue and fin whales, is as shown in Fig. 40a, the regression line in these samples being

for the foetusess of blue and fin whales.

Weight of variously dissected parts were weighed in a box placed on the said platform scale on 4 heads of male and 10 heads of female blue whales and 2 heads of male and 3 heads of female fin whales on the Nisshin-maru, and on 7 heads of male and 9 heads of female blue whales, and 4 heads of male and 7 heads of female fin whales on the Hashidatemaru (Cf. Table X). At the same time, oil production of these various parts were measured by the readings on oil tank gauge from part of these material. Since the total weight of various parts could not be obtained



due to loss of body fluids, excreta, urine and stomach contents. overall weight of a whale could not be taken. If on the other hand, the proportion of were the same in whales, their relationship to each other would be similar to the total body weight. Fig. 40 b shows the total weight plotted according to body length.

Although the process of dissection of the two fleets differed slightly, there would be no effect on the measured values with

the exception of one or two points. In grand total, especially, same items have been inculuded on values from both fleet.

**Total weight:** Measurements were made on 30 heads of blue and 16 heads of fin whales. Their regression line, measured as for foetus, obtained from the specimens by the least squares, are

Blue Whales ...... 
$$W = \frac{L^{3.5}}{5.27 \times 10^4}$$
 ..... (2)

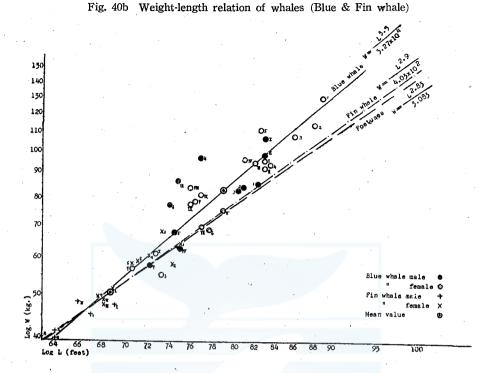
Fin whales...... $W = \frac{L^{2.9}}{4.30 \times 10^2}$  .....(3)

## CHAPTER VII. Thickness of Blubber

### I. Sites of Measurement

Thickness of blubber was measured in following places:

Point 1-- The point on the horizontal cut side of the body (at the



position of lateral line in fish), where it intersects a vertical line from the dorsal fin.

- Point 2— The point on the vertical cut near the earhole, where it intersects a mid-dorsal line.
- II. Average Thickness of Blubber according to Sexes and Body Length measured on the Nisshin-maru Fleet.

Figs. 41 to 44 show mean values measured by the Nisshin-maru Fleet on the thickness of blubber according to sexes and body length. Figs. 41 and 42 show curves for male whales and indicate that the thickness approximately increases in proportion to body length. It is interesting that in blue whales, thickness becomes less with age when the animals attain sexual maturity. Curves in Figs. 43 and 44 represent those for females which peculiar modes appear every 3 feet in blue whales and every 4 feet in fin whales.

III. Monthly Change in the Thickness of Blubber.

Average percentage monthly change of the thickness of blubber by body length are shown in Figs. 45 and 46. From these Figures, it can be seen that the thickness of the blubber increases during December to March,

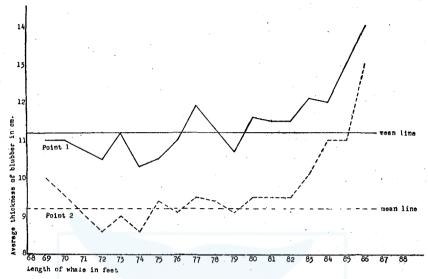
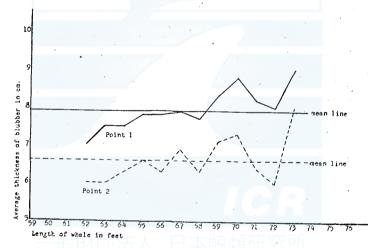


Fig. 41 Variations of thickness of blubber in length of whales (Male Blue whale)





i. e. the summer months in the Antarctic, irrespective of the sexual state of the whales. In other words, the state of nutrition of the whales become better during these months. This is particularly remarkable in blue whales.

Due to the lack of whales in lactating stage, no curve for such animals was obtained but this has been noted in places. As can be seen from the Figures, the thickness is remarcably small which seems to have a relative connection with the fact that the thickness of mammary glands in lactating whales is conspicuously thick.

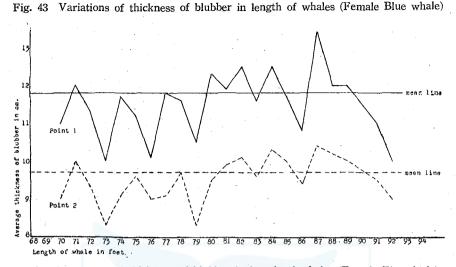
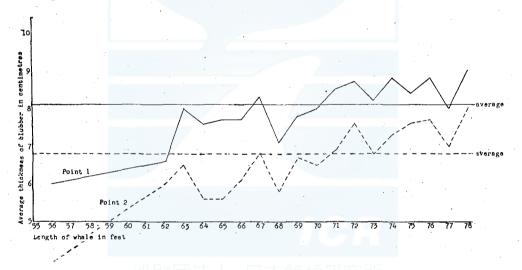


Fig. 44 Variations of thickness of blubber in length of whales (Female Fin whale)



IV. Relationship between the Thickness of the Blubber and Body Length as measured on the Hashidate-maru Fleet.

The coefficient of the thickness of blubber is shown by  $D/L \times 10$  where D denotes the thickness of blubber in cm., and L, the body length in feet. Distribution of its mean value at Point 1 (at the side of the body) is shown in Figs. 47 and 48. In general, both the male and female blue whales coefficient of from 0.09 to 2.0, the peaks being presentat 3 points between 1.1-1.5. In Fin whales, the coefficient is distributed between 0.7 to 1.5, peaks being present in 1 to 3 places between 0.9 and 1.3.

V. Relationship between the Thickness of Blubber and Body Length by

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# Biological Survey of Fin and Blue Whales Taken

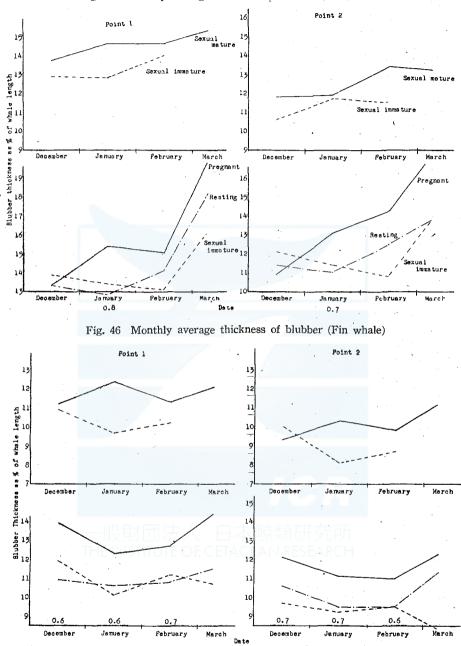
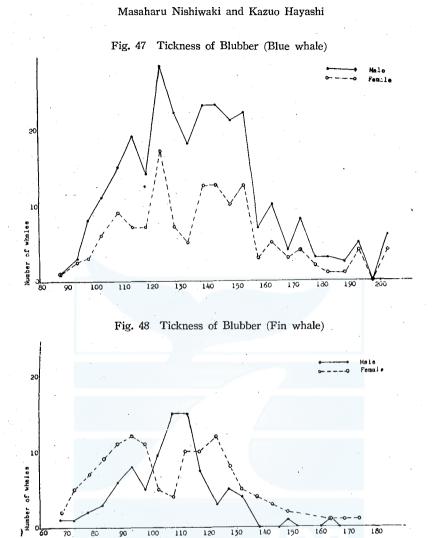


Fig. 45 Monthly average thickness of blubber (Blue whale)

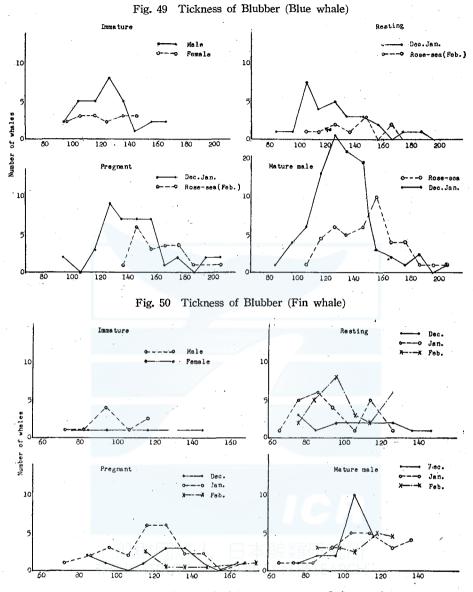
the Degree of Sexual Maturity, as measured on the Hashidate-maru. The distribution of mean values as shown in Figs. 47 and 48 can be divided into several groups by the mature and immature in males, and immature, pregnant and resting in females. These are as shown in Figs.



49 and 50 which indicate that in immature whales both male and female, the average value is distributed between 1.0-1.4 and seems to have a uniform mode, although the trend is not remarkable probably due to the small number of whales measured. The mode in mature males is at 1.3, especially that in whales from the Ross Sea being at 1.5, showing clearly the difference in the thickness of blubber. The distribution is broader both in the pregnant and resting, although the distribution in the pregnant whales is slightly higher. In both cases, whales from the Rose Sea contains larger proportion of higher coefficients.

The coefficient for immature fin whale males is situated around 0.9-1.0. There is no remarkable difference in females according to month, but

### Biological Survey of Fin and Blue Whales Taken



in the pregnant and resting whales larger coefficients increase as the month advances. On the whole the difference in the coefficients between blue and fin whales seem to be essential.

## CHAPTER VIII. Food

### I. Krills and Other Fish

The present investigation also disclosed the fact that the chief food of the blue and fin whales in the Antarctic Ocean is the krill. Of the 710 heads of blue and 608 heads of fin whales examined, fish other than krill found in their stomach were as follows:

Fish of large Harpodontidae: 2 cases (1 in blue whale, 1 in fin whale) Squids: 4 cases (1 in blue whale, 3 in fin whale)

Small type of Harpodontidae: 24 cases (10 in blue whale, 14 in fin whales)

Kind of Iniomi: 1 case in blue whale.

Jelly fish: 3 cases in fin whale.

II. Euphausia as Whale Food

Results of investigations on Euphausia, especially E. superba, are shown Tables VIII and IX, in which the empty spaces are mostly those in which the krill was found almost digested as reddish-brown colour in the intestines. It is interesting to note that the semi-monthly percentage rate of stomachs with krill coincide wholly with the catch curves as shown in the foregoing Figs. 2 and 3. In other words, th's shows how the whales move their positions according to the presence of krill or that the investigation of krill would enable judgement on condition of the catch.

					1.000									
Name of fleet	Half-months	No. of stomach examined	No. of stomach with krill	No. of stomach empty	% of stomach with krill	No. with much krill (R)	No. with mod krill (rrr)	No. with little krill (rr)	No. with very little krill $(r)$	L	M	S 'Type of krill	X	0.
Hash	December	41 79	26 53	15 26	63 66	73	3 15	2 17	14 18	5	14 21	1 3	5 26	1 3
idate	January	27 39	12 11	15 28	44 28		13	43	7 5		8 8	-1	4 2	_
Hashidate-maru	Feburary	36 52	23 31	13 21	64 60	1	10 .9	10 7	$\begin{array}{c} 2\\ 12 \end{array}$	2 8	10 14	-	9 8	2 1
fleet	March		=			-		-			-		1 <u>-</u>	
Nis	December	112 78	99 58	13 20	88 74	44 27	24 10	14 11	17 10	24 5	41 19	31 33	$3 \\ 1$	-
Nisshin-maru	January	66 87	83 51	33 36	50 59	$\frac{3}{10}$	9 11	9 17	12 13	1 5	8 11	24 35		-
	Feburary	31 38	16 13	15 25	51 34	$\frac{1}{3}$	7 6	$\frac{2}{2}$	$\frac{6}{2}$	1 	6 1	9 17		
fleet	March	24 —	9	15 —	37 —	1	4	3	1		4	5	_	-

Table VIII. Stomach contente (Blue whale)

## Biological Survey of Fin and Blue Whales Taken

Name of fleet	Half-months	No. of stomach examined	No. of stomach with krill	No. of stomach empty	% of stomach with krill	No. with much krill (R)	No. with mod krill (rrr)	No. with little krill (rr)	No. with very little krill (r)	· · ·	, M	S Type of krill	X	5
Hash	December	5 47	$\begin{array}{c} 2\\ 22 \end{array}$	3 25	40 47		5	7	29	2	1 8		1 7	-2
Hashidate-maru	January	44 43	19 23	25 20	43 54	4	3 4	6 5	10 10	1	8 9	$\frac{1}{2}$	7 10	2 1
maru	Feburary	. 53 1	25 0	28 1	47 0	4	1	6	14 —	4	8	1	11 —	1
ı fleet	March	13	4	14 —	22 —	-	2	=	2	-	$\frac{2}{-}$	-	2 	
Nis	December	18 67	16 38	$\frac{2}{29}$	89 57	4 15	4 5	5 8	3 10	5 14	5 11	$5 \\ 12$	1 1	-
Nisshin-maru	January	44 56	28 28	$\frac{16}{28}$	64 50	10 9	6 8	6 9	6 2	2 4	6 9	20 15		_
	Feburary	$\begin{array}{c}143\\63\end{array}$	85 35	58 28	59 56	$\frac{20}{9}$	25 11	26 10	14 5	6	$\begin{array}{c} 22\\ 14 \end{array}$	56 21	1 	-
fleet	March	6	5	1	83 —	3	1	1		_	2	2	1	-

#### Table IX. Stomach contente (Fin whale)

Symbols used in tables are as follows:

L=E. superba. Large, 5.0 cm. and over (from rostrum to tail).

M = E. superba. Medium, from ca. 4.0--5.0 cm.

S=E. superba. Small, up to ca. 4.0 cm.

X=E. superba. Mixture of conspicuously different sizes.

?=E. superba. Sizes unknown due to high degree of digestion.

R=Stomach with large amount of krill.

rrr=Stomach with moderate amount of krill.

rr=Stomach with small amount of krill.

r=Stomach with very small amount of krill.

## CHAPTER IX. Prasites

### I. External Parasites

Following external parasites have been collected from blue and fin whales apart from certain more or less minutes form found on the baleen :

Cirripediae- Coronula sp. (C. regina)

Conchoderma sp.

Coppepodae— Pennella sp. Anphipodae— Cyamus sp. Diatomes— Cocconeis sp.

The incidence of these parasites are shown in Tables X. and XI. Seasonally, there is no great change according to whaling seaosn with Cirripediae, but Pennela is found more in the earlier part of the whaling season, one example being a clue whale on which about 30 full grown specimen had been found hanging. Cyamus was found in the latter part of the whaling season. Almost all of the diatomes found were of the Cocconeis sp., which began to increase, both in incidence and density, as the whaling season advanced.

	Infe	cted	Not In	fected	Percentage	of Infection
	Hashidate Maru	Nisshin Maru	Hashidate Maru	Nisshin Maru	Hashidate Maru	Nisshin Maru
Whale lice (Cyamus Sp.)	17	32	176	402	6.32%	7.37%
Barnacles (Coronula Sp.)	7	17	262	417	2.60%	3.92%
Conchoderma Šp.	1	4	268	430	0.37%	0.92%
Parasitic coppepedae (Pennella Sp.)	2	8	267	426	0.74%	1.84%
Diatomes	72	109	197	325	26.76%	25.12%

Table X. Percentage Incidence of External Parasites in Blue Whales.

Condition unknown: 2 whales used as fenders in the Nisshin-maru Fleet. 5 whales missed from the Hashidate-maru Fleet.

	Infe	cted	Not In	nfected	Percentage of Infection		
	Hashidate Maru	Nisshin Maru	Hashidate Maru	Nisshin Maru	Hashidate Maru	Nisshin Maru	
Whale lice	24	19	184	378	11.53%	4.79%	
(Cyamus Sp.) Barnacles (Coronula Sp.)	24	30	184	367	11.53%	7.56%	
Conchoderma Sp.	HE INISTITI	JTE (4F. C	207	RES 393 CH	0.48%	1.01%	
Parasitic coppepedae (Pennella Sp.)	1	4	207	393	0.48%	1.01%	
Diatomes	77	131	131	266	37.02%	33.00%	

Table XI. Percentage Incidence of External Parasites in Fin Whales.

Conditions unknown: 3 whales missed from the Hashidate-maru Fleet.

The incidence of white scars was as shown in Table XII which indicates that no whale is free from it.

The proportion of scars obtained in the previous year and prior to it were found to be, in the maiority of cases, more numerous for the older one, the values being:

About 92% had more scars obtained earlier than the previous year; about 2% had equal number of scars obtained in the previous year and earlier, about 6% had more scars obtained during the previous year.

There were no difference between male and female, or between blue and fin whales.

It was thought possible to tell the age of whales by the white scars but there are some remarkable exceptions that they would not constitute a very good facter in assuming age. There is a tendency, however, of larger number of scars in aged animals.

Sex	Male			Female				
Name of fleet	Hashidate-maru		Nisshin-maru		Hashidate-maru		Nisshin-maru	
Classification	Number of whale	%	Number of whale	%	Number of whale	%	Number of whale	%
none	0	0.0	0	0.0	0	0.0	0	0.0
few	5	3.8	39	19.5	6	4.5	70	29.8
scarce	40	30.0	2	1.0	46	34.3	8	3.4
normal	25	18.8	40	20.0	27	20.2	26	11.1
numerous	57	42.9	84	42.0	51	38.0	90	38.3
very numerous	6	4.5	35	17.5	4	3.0	41	17 <b>.</b> 4

Table XII. White scars on Blue Whales.

Sex	Male				Female			
Name of fleet	Hashidate-maru		Nisshin-maru		Hashidate-maru		Nisshin-maru	
Classification	Number of whale	%						
none	0	0.0	0	0.0	2	0.0	0	0.0
few	0	0.0	58	33.0	5	4.1	63	26.8
scarce	36	42.4	5	2.8	26	21.5	9	3.9
normal	13	15.3	14	8.0	30	24.8	27	11.5
numerous	32	37.6	81	46.0	45	37.2	108	45.9
very numerous	- 4	4.7	18	10.2	15	12.4	28	11.9

Table XIII. White scars on Fin Whates.

# **II.** Internal Parasites

Internal parasites are found more commonly than the external Crustaceae and they are often present in great number in whales. However, since not all of the intestines were cut open, no percentage of incidence were obtained. The species found were tapeworms (Tetrabothrius sp.?) or Acanthocephala.

Numerous encysted tissues, due probably to nematode worms, were fuond near the body cavity, especially in the ventral muscles. Since calculation of its incidence would be difficult, no attempt was made to obtain percentage rates.

# CHAPTER X. Summary

The results of various observations made the catch by the Japanese fleet during the season 1948—1949 can be summarized as follows:

1) Composition of Whales Taken

There is no denying that the body length of whales taken is decreasing year by year. However, as far as the present expedition is concerned, the results showed a slight tendency to improve in this respect. This is an evidence that the Japanese whalers have come to recognize the fact and are taking great care to catch large-sized whales. It is regrettable that four of the blue whales taken were under-sized and is therefore in violation of the international law. However, it is hoped that everyone will recognize the fact that the Japanese whalers are doing their best to comply with the international code of ethics. Average body length were as follows:

	· .	Male	Female	Animal
Blue w	hale—	77.86 ft.	81.09 ft.	79.55 ft.
Fin wh	ale—	66.52 ft.	70.53 ft.	68.80 ft.

2) Body length of mature whales according to their sexes came out as follows:

At sexual maturity:		Male	Female	
	Blue whale—	74 ft.	78 ft.	
•	Fin whale—	63 ft.	68 ft.	
At	physical maturity:	Male	Female	
	Blue whale—	79 ft.	85 ft.	
	Fin whale—	68 ft.	74 ft.	

3) Pregnancy rate, especially in fin whales, during the present expedition seemed to be lower than the pre-war records, values being :

Blue whale— 53.29% of total mature animals.

Fin whale— 32.17% of total mature animals.

4) It has been found that it is impossible to tell the sex of a whale from its body colour and that only a very inaccurate data can be obtained in telling the age of whales from their body colour.

5) Although it is still uncertain, judging of age from the condition of white scars and the amount of parasites on whales would be a good

subject of study in knowing migratory habit of whales.

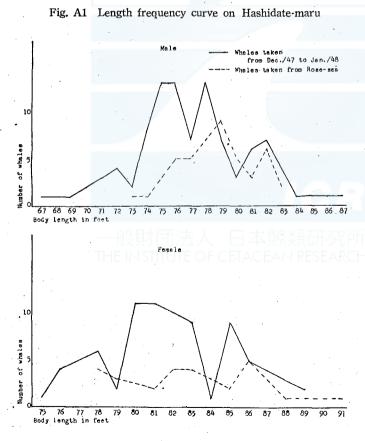
6) It is to be regretted that no investigations were made on the baleens.

### APPENDIX I.

## Number of Whales in the Ross Sea

I. Blue Whales in the Ross Sea

During the present season, the Hashidatemaru operated around the Ross Sea (Lat.  $74^{\circ}$ — $76^{\circ}$ S, Long.  $172^{\circ}$ — $178^{\circ}$ W) from 11 to 25 February 1948. During this time, 77 heads of blue whales alone (Nos. 393 to 471) were taken and distribution of their body length is shown in Fig. A1. As a result of investigations, there were no immature whale among 47 heads of males and only one immature whale among the 30 heads of females. 19 of the mature females, about 66%, were pregnant, 10 or 34%, resting.



This percentage seems to be rather high for February. The ratio of male to female was 100 males to 64 females, incidence of males being remarkably higher than in other waters. Average body length of whales taken in the Ross Sea was 78.66 ft. for males. 82.77 ft. for females, the total average being 80.62 ft. This is slightly above the average but the percentage of aged males was

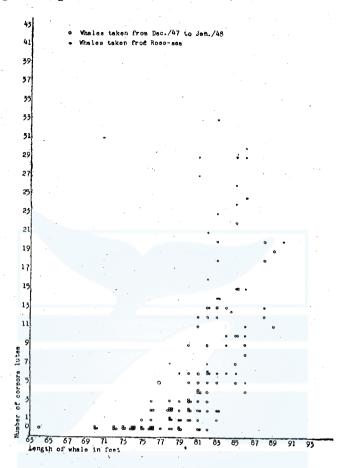


Fig. A2 Length of whales and Number of corpora lutea (Blue whale)

higher than in other water, individuals in which ossification of vertebral column had been ankylosed was 55% in males and 20% in females. Immature animals were few in both male and female. There were also a very low incidence of external parasites, Cyamus being found only in one case each in male and female. There was no evidence of Coronula sp., one each in male and female of Pennela sp. Diatome film infection was found in 15% of the male and 30% of the female. As was explained in the Chapter on the blubber, its thickness per unit body length was larger the whales being fatter. This was not only seen in external appearance but also borne out by the fact that the production of o'l per B. W. U. during operation in the Ross Sea was extremely high.

No particular difference from other waters was seen in the relationship

between body length and number of corpora lutea (Cf. Fig. A2). This is an evidence that there were a small number of large-sized whales in the Ross Sea during the present operational season when considering the facts that the average body length of whales taken in the same region by the 2nd Tonan-maru in 1941 were 79 ft. for males and 82 ft. for females, in average, and that a school of quite a large-sized blue whales had been taken in the Kerguolen sector during the season 1923 — 1933 (A. H. Laurie: Discovery Reports. Vol. XV, pp. 223—284). However, due to a lack of biological data, no explanation can at present be given.

# APPENDIX II.

# Copulation of Humpback whales

There has been many tables told by whalers on the copulation of whales but very few description has been given by biologists. As far as is known by the auther, the only report of such detail is the one by D. C. Lilie published in 1910 (Cf. except at the end of this chapter).

During the present expedition, the auther witnessed following actions by the humpback whales.

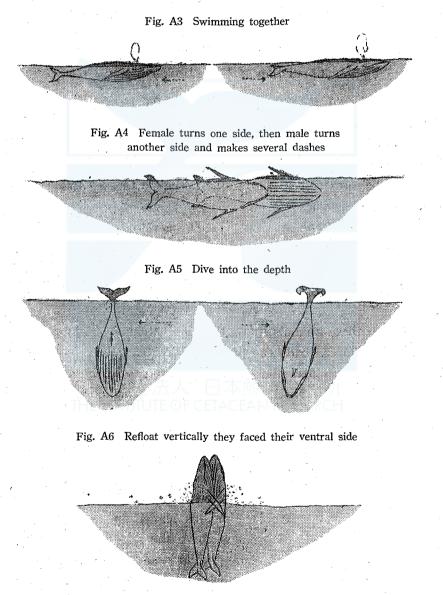
On 13 January 1948, at about 21–00, operation was being carried out on the Nisshin-maru drifting about 1 mile off the so-called pack line. There were many cracks and inlets into the pack ice near the ship and many humpback whales (about 15 to 16 heads) were swimming around there.

As though these animals know that the catching of humpback whales in the Antarctic Ocean had been prohibited by the International agreement, they often came very near the ships. Just then, some humpbacks started to come nearer to the Nisshin-maru, making tremendous spashings. At first, one stood watching without any thought but there were something peculiar in their movement and the author began to watch with intent.

There were two heads of humpbacks and, as shown in Fig. A3, they at first swam in single file. Shortly later, as shown in Fig. 4, the two whales began to roll and romp together with each one of their fippers well above the water. At one time, as shown in Fig. A5, one of the whales flipped its peculiar tail flukes above the water and then dived deep into the water, followed immediately by its mate. For a few seconds afterwards, the surface of the water remained calm and then the quiet of the

### Masaharu Nishiwaki and Kazuo Hayashi

water was broken by the sudden surfacing of the two whales vertically, as shown in Fig. 6, with their ventral surfaces in close contact and appearing above water almost to the middle of their bodies, just below their flippers. Then the whales fell back into the water, sometimes together, sometimes separately. After that, the same movements started all over again. Although there were some pauses, the romping continued well over three hours from the time it was first noticed until the whales were lost to sight. It may be that the objective had not been attained, but the so-called



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"standing up on the flippers", as shown in Fig. A6, were witnessed more than to or three times. The author was of the opinion that this was the copulation of humpback whales, the fact of which was borne out by the experienced crews of the ship. The author had heard previously from a whaler that the whales jumped above the water, their ventral surfaces together, during copulation but was very doubtful if this were feasible. However, the present expedition showed that this is possible although the copulation itself may be performed underwater. The whole scene was a grand sight if slightly garish. It was a great pity that the time was at sundown and did not permit photographing. The nearest that whales came to the ship was about 200-300 metres. Some whalers said that the surface of the water became was not witnessed at this time. It seems that the whales moved away before the acts were completed but there were no evidence that the male whale heaved a great sight or become exhausted after the act. Even during copulation, the whales rested a while, sometimes swimming quiently for 15 to 30 minutes.

# ABSTRACT

# Observations on the Anatomy and General Biology of Some Members of the Larger Cetaceae

by D. G. Lilie, Hutchinson Research Student of St. John's College, Cambridge.

(Taken form the Proceedings of the Zoological Society of London, 1910)

## VIII. Miscellaneous Observations

3. Copulation, period of gestation and rate of breeding.

The Balaenoptera are said by whalers to copulate at the surface of the sea. The pair swim towards each other and turn slightly on the sides so that their ventral surfaces face one another. The male makes several dashes at the female to insert the penis. When the pair first rush together, the long axes of their bodies are paralel with the surface of the sea; but they curve up vertically at the end of the act. After copulation, the male is said to be exhausted and easily caugt.

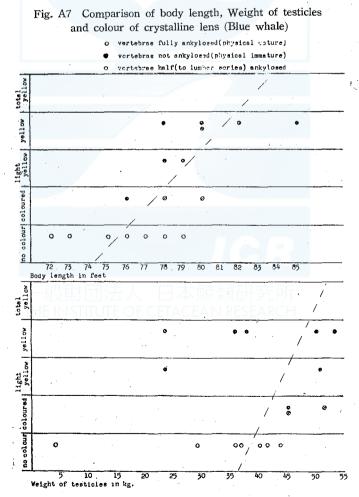
# APPENDIX III.

Age of Whales and Colour of Crystalline Lens

Wheeler's statement in his Discovery Report says that "the determination of age on whales is a question of much scientific research and economic importance".

During the expedition, I have stood at the side of the slipway watching the eyes of whales as they came up. Some looked as if they were peacefully sleeping, whilst others had a hunted look. Amongst these, the colour was of a golden shine. But I understand the colours vary.

When I was a boy, I used to be to told that there were witches (an old cat, an old fox or an old badger) who could change themselves into men, then some hero would have to go out and dispose of them. These



animals (wiches) were always old, I was informed, and their eyes shone a bright golden colour.

Looking at the eyes of whales, I wondered if this story could not be connected with them.

In dissecting, first, cut the vertex corneae in a V shape manner without scratching the lens and then take the lens out with the corpus vitreum with two fingers. Secondly, wash the corpus vitreum and the iris off with fresh water. It is then ready for observation in the shade. It is best to look at the lens in the shade and not in direct sunshine (though the sun in the Antarctic was not strong).

Lastly, I have found at the crystalline lens, some to be yellow, others light yellow, and still others without colour or transparent.

It was interesting to discover that the colour of the eyes had a very close resemblance to that shown in the chart. There was no colour by

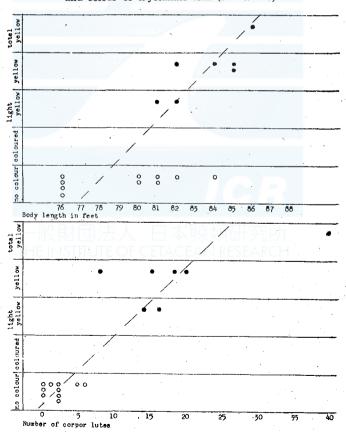
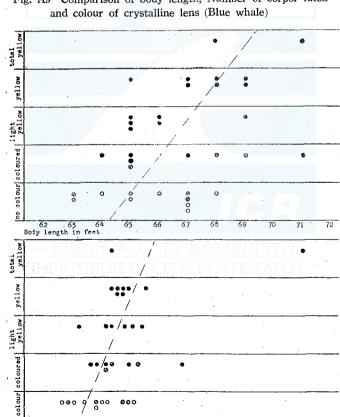


Fig. A8 Comparison of body length, Weight of testes and colour of crystalline lens (Fin whale)

naked, eye, in the lens of the eyes especially, if the epiphyses of the lumbar series was not ankylosed. As ankylosis progressed, the colour became vellowish, and when the ovaries had a number of corpora lutea (much as 30) and complete ankylosis had set in, they were found to be completely Depending upon individual whale, the gradual change of colour vellow. was also noticed with the number of corpora lutea or weight of testicles and state of ankylosis.

In order to gauge the change in colour, it would have been an advantage to have used the lovibond tintometer (the British Drughouses pattern) or some electric method, but I came upon this instrument too late to obtain full advantage of its use.

The classified colour of lens in the chart has been done only through The five colours as classified are no colour (transmy naked eves. parent), coloured, light yellow, yellow, total yellow.



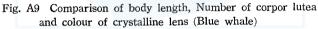
20

30

50

10

Weight of testicles in kg.



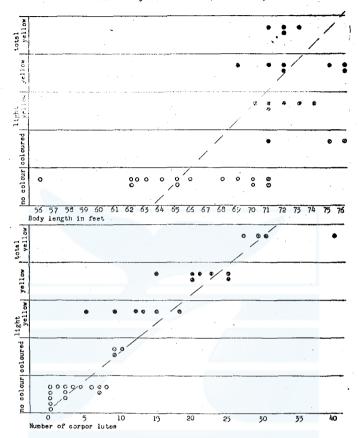


Fig. A10 Comparison of body length, Number of corpor lutea and colour of crystalline lens (Fin whale)

The data gathered in this Report may be of some help in determining the age of whales through the colour of the crystalline lens.

It is hoped to include colourimeter and electric ammeter in the next expedition in order to complete this research but the present report will only suffice by preliminary descriptions. (Cf. Figs. A7–A10).

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