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INTERNATIONAL WHALING COMMISSION

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ABSTRACT

This paper reports the results of biological sampling of the Antarctic minke whale during the first New Scientific Whale Research Program in the Antarctic Ocean (NEWREP-A) survey conducted in Area V (130°E-170°W, south of 60°S) during the 2015/16 austral summer season. The paper also reports the results of the sighting surveys and Photo-ID and biopsy sampling of large whales by the sighting sampling vessels (SSVs). Two SSVs and one research base, were engaged in the survey for 65 days. A total of 335 primary sightings (involving 915 individuals) of Antarctic minke whale were made during 2,394n.miles of searching distance. A total of 333 Antarctic minke whales (230 females and 103 males) were sampled, and a number of biological samples and data required for the two main objectives of NEWREP-A were obtained from each whale taken. Earplugs for age determination were collected from all whales. A total of 13 large whales were photo-identified: blue whale (3), humpback whale (9) and killer whale (1). A total of seven biopsy samples were collected from blue (1), humpback (5) and killer (1) whales. The samples and data collected in this survey are available for interested national and international scientists under the guidelines for research collaboration posted at the home page of the Institute of Cetacean Research (ICR): http://www.icrwhale.org/NEWREP-AProtocol.html.

KEYWORDS: SCIENTIFIC PERMITS; ANTARCTIC; FEEDING GROUNDS; ANTARCTIC MINKE WHALE; BIOPSY SAMPLING; PHOTO-ID

INTRODUCTION

The first survey of the New Scientific Whale Research Program in the Antarctic Ocean (NEWREP-A) was carried out in Area V during the 2015/16 austral summer season, after the review of the research plan was completed by the International Whaling Commission Scientific Committee (IWC SC) following the guidelines in Annex P (IWC, 2015a; see GOJ, 2016 and Matsuoka *et al*., 2016).

Research under NEWREP-A requires the collection of various types of samples and data which are important for addressing main Objectives I and II of the program. For example, under objective I (ii) samples and data for obtaining the age, sexual maturity and body length of the whales are required. Under objective I (iii) data and samples for studying morphometric, morphological and genetic differences among whales are required. All that information together with other obtained by non-lethal means (e.g. abundance under objective I (i)) is necessary for the specifications of RMP *ISTs* for Antarctic minke whales (objective I (iv)).

Under objective II (iii), stomach contents of the whales are required to estimate prey composition and consumption by Antarctic minke whales; and blubber thickness, fat weight and girth data are required to study the nutritional condition of the whales. All that information together with other obtained by non-lethal means (e.g. whale abundance under objective II (ii) and krill biomass and oceanographic information under objective II (i)) is necessary for the ecosystem modelling work (objective II (iv)).

Age data at the annual scale is required by the Statistical Catch-at-Age Analysis (SCAA) under objective I (ii). Age information can be obtained only from internal earplugs and therefore only through lethal sampling methods. The NEWREP-A review workshop agreed that at present, the technique commonly used for the determination of the biological parameters used in the SCAA model require earplug for age determination (IWC, 2015b).

Calculation of sample size of Antarctic minke whale in NEWREP-A was based on the biological parameter Age at Sexual Maturity (ASM). ASM is of great importance not only for contributing information on the proportion of matured animals in the SCAA (related to the main objective I) but also as an important indicator of changes in the nutritional condition of the whale population (related to main objective II). The age-at-50% sexual maturity (ASM 50) was used to set the annual sample size of 333 Antarctic minke whales (see GOJ, 2015 for details).

The first multidiscipline survey of the NEWREP-A was conducted in Areas IV and V during the austral summer season 2015/16. An overview of the survey is presented by Matsuoka *et al*. (2016). The report of the dedicated sighting survey is presented by Isoda *et al*. (2016) and that of the krill and oceanographic survey is presented by Wada *et al.* (2016).

The objective of this paper is to present the results of the biological survey of 333 Antarctic minke whale sampled during the first field survey of NEWREP-A in Area V. Sighting data obtained by the Sighting Sampling Vessels (SSVs) are also presented in this paper.

SURVEY DESIGN

Research area

IWC Antarctic Management Area V, south of 60°S, was surveyed during the 2015/16 season. The area was divided into the East and West sector at 165°E. Each sector was divided into the South and North strata. The boundary between the South and North strata in the West sector was defined by a line 45n.miles from the ice-edge. In the East sector, 69°S was the latitudinal boundary line. The East-South stratum (Ross Sea) was further divided into the West and East sector at the 180° longitudinal line (Figure 1a). Within the Ross Sea, the area comprised east of 170°W (enclosed within pack ice) was also included in the East-South stratum.

Estimated pack-ice line (ice-edge) was obtained from direct observation from the vessels, from the Defense Meteorological Satellite Program (DMSP; Maslanik and Stroeve, 1999) and from the Advanced Microwave Scanning Radiometer 2 (AMSR2; JAXA, 2016).

Research vessels

Two research vessels *Yushin-Maru* (*YS1*) and *Yushin-Maru No.2* (*YS2*) were engaged in sighting and sampling (SSVs). They were equipped with a top barrel platform (TOP), upper bridge platform (UBP), and a whaling cannon. Biological research of the whales sampled was carried out on board the research base vessel, *Nisshin-Maru* (*NM*). One researcher was on board each *YS1* and *YS2*. A total of eleven researchers, included the cruise leader and vice cruise leader, were on board the *NM*. Specifications of the research vessels were shown in Table 1 and Figure 2.

Cruise track-line

Survey courses were established in offshore and ice edge waters of the research area by the line transect method. Two SSVs advanced along parallel track-lines 7n.miles apart (Main course and sub-course). Each of the SSVs changed the track-line order every day to avoid possible sighting bias by fixed position. Starting point of the day was set at the position where one of the vessels ended the surveys on the previous day in the most advanced position. The other vessel moved to the starting position of the next day after the end of the daily survey.

The predetermined track-line of the sampling survey was shown in Figure 3a. Track-line for each vessel consisted of two legs in the northern stratum at 3°20' longitudinal degree intervals, and six legs in the southern stratum at 1°40' longitudinal degree intervals in a 10° longitudinal band in the research area (Nishiwaki *et al.*, 2014) except for the East-South stratum (Ross Sea). A latitudinal zig-zag line was set for the East-South stratum. Track-lines were decided based on the original longitude line, which was selected at random in the western and eastern part of the stratum, respectively. The interval of legs and number of legs in each stratum could be changed in consideration of delay caused by bad weather conditions and other factors. Survey course angle and distances were calculated using a Tamaya Navigator NC-2100G (Ver.1.2.1; Tamaya Instrument Co., Japan).

Sighting protocols

Survey mode

Survey mode of sampling survey for Antarctic minke whale was conducted using Normal Closing mode (NSC). Sighting activities of the survey mode were classified into two types according to 'On-effort' or 'Off-effort'. Both survey activities followed the protocol endorsed for the IWC/SOWER surveys (e.g. Matsuoka *et al.*, 2003).

Sighting teams and observation time

Sighting protocols were the same as those in IDCR/SOWER. Research effort began 60 minutes after sunrise and ended 60 minutes before sunset, with a maximum 12 hour per day (approximately 06:00–18:00). Time-zone change was recorded at 30 minute intervals, effective from 01:00h.

Time was adhered to local 'ship' time ranging between +9.0 and +12.0 GMT. Data collected throughout the survey and all associated reporting were in accord with the local 'vessel' time. Searching activity was conducted when the weather conditions were suitable for observations: minke whale visibility better than 1.5n.miles and the wind

speed less than 21knots (northern strata) or 26knots (southern strata). Vessel speed during the sighting survey was 11.5knots with slight adjustment to avoid vibration of the vessels. Sighting effort was conducted by the boatswain and top-men from the TOP (there were always two primary observers in the TOP) and the upper bridge where the helmsman, captain or officer-on-watch, researchers, and the chief engineer (or second engineer) were also present.

Sighting distance and angle experiment was examined in each SSV for evaluating the accuracy of sighting distance and angle given by observers. Observers on each vessel were required to assess eight sets of angles and distance estimation from two platforms (TOP and UBP). The experiment was conducted using a buoy with a radar-reflecting transponder, using angle board and scope (7x) equipped with estimate scales on eye lens. All trials were conducted under good sighting conditions.

Whale sampling

Sample size for Antarctic minke whales was 333 animals (GOJ, 2015). Whales were sampled using a random sampling method (Kato *et al*., 1989). One or two whales were sampled randomly from each primary sighted school using harpoon with 30g penthrite grenade. Sampled whales were immediately transported to the research base vessel, where biological measurements and biological sampling were carried out.

Biological measurements and sampling

Morphometric and body weight

After photographing the lateral side of each whale, a series of standard measurements was taken, including body length to the nearest 1cm and body proportion at 7 different points (to the nearest 1cm; Figure 4). Skull measurements (length and greatest breadth to the nearest 0.5cm were taken for most whales using a large pair of vernier calipers. Blubber thickness measurements were taken at two points on the lateral side of the body.

Body weights of each whale were measured using an electronic track scale (EDP-1801 and EDI-801, Yamato Scale Co., Ltd., Japan) on board the NM to the nearest 0.01tons. Body parts and organs weight of five whales were weighted for studying energy storage, using an electronic hanging scale (Kubota) and a marine scale (M1100 and S-182, Marel, Iceland).

Definition of sexual maturity

The maturity of the females was determined by the presence of at least one corpus luteum or corpus albicans in either ovary. In the case where no corpus luteum or corpus albicans was observed, the female was categorized as immature. The definition of male sexual maturity was defined preliminary based on the weight of one testis. If the testis was over 0.4kg, the whale was determined as sexually mature (Kato, 1986). Reproductive status (of female whales was classified into four categories (ovulating, pregnant, resting and pregnant and lactating), based upon observation of ovary, uterus, and mammary gland. Pregnancy of the animal was defined based on conceptus with placental development in the uterus. Body length and weight of foetus was measured in the same way as in adult whales. Sex of the foetus was classified into three categories (female, male or sex unidentified).

Earplug survey

Left and right earplugs were collected for age determination by the routine procedure (Omura, 1963). After removing the mandibles, the proximal part of the earplug was exposed along the surrounding the external part of the ear canal from the tympanic bulla using a knife, for subsequent incision. The external part of the ear canal was carefully cut open so as not to incise the earplug, and then the earplug was collected with glove-finger using a scalpel. Earplugs were fixed and stored in 10% formalin solution. As a supplement for age determination studies, the largest baleen plates were collected from whales of either sex smaller than 7.0m long.

Stomach contents

Conventional stomach content records were obtained from all sampled whales. Prey species from the fore- and main-stomach contents are weighed for each whale sampled (47 whales) and stored at −20°C or in 10% formalin solution.

Other biological samples

Ocular lenses of each adult whale and foetuses with body length less than 10cm were collected, and stored at −80°C for age estimation purposes. Ovary, mammary gland and endometrium from female animals were dissected from the uterus and stored at −20°C for reproductive study. Tissue samples of testis for the histological observation were collected and fixed using 10% formalin solution. After measurements of blubber thickness (two points), blubber samples were taken from all specimen for the study of feeding ecology. Muscle and liver samples were sampled and stored at −20°C for pollutant studies. Skin samples were collected and fixed in ethanol solution (70%) for genetic studies.

RESULTS AND DISCUSSION

Narrative of the cruise

SSVs (YS1 and YS2) departed from Shimonoseki (Japan) on 1 December 2015. NM departed from Innoshima (Japan). Transit sighting surveys to the research area were conducted from 14 December 2015 to 21 December 2016. The total research period of the sampling survey was 65 days from 22 December 2015 to 25 February 2016. Transit sighting surveys from the research area were conducted from 26 February 2016 to 8 March 2016. SSVs and NM arrived at Shimonoseki on 24 March 2016.

Ice concentration

Figure 1b shows the spatial distribution of representative sea ice concentration during NEWREP-A 2015/16 cruise. There were several large polynya along the continent in the western sector during the earlier period. These area could not be surveyed because the sea ice prevented access.

Area coverage

Figure 3b shows the main cruise tracks of the SSVs. The searching distance of the SSVs in each stratum is shown in the Appendix. The total searching distance during the 65-days research period was 2,394n.miles. The percentage of actual coverage in the whole research area was 11.2% (main course). The actual coverage was less than half compared with predetermined track-line due to bad weather, especially in the northern part of the research area. The survey in the western part of the Ross Sea was cancelled to avoid a high vessel traffic zone and to avoid meeting a vessel from an anti-whaling NGO.

Geographical distribution of sampled whales

A total of 333 Antarctic minke whales were caught from a total of 335 primary sightings (involving 915 individuals). All whales were sampled in a random manner. Geographical distribution of sighting and sampling were shown in Figure 3c and 3d, respectively. Of the total, 230 whales were females and 103 males (male sex ratio was 0.31). Table 2 show the mean body length and weight of the sampled whales, by sex, reproductive status and stratum.

Sampling efficiency

A total of 333 individuals was sampled from 317 targeted school. The technical sampling efficiency (number of whales sampled per number of individuals targeted) was 0.95, and did not change significantly between number of whales targeted (e.g. one and two whales targeted) (0.93 and 0.97, respectively; $P > 0.05$: Chi-squared test). The main reason for missing a targeted whales (18 cases) was the quick movement of the whale (12 cases out of 18). Two struck and lost cases occurred.

Biological measurements and sampling

External measurements

Table 3 summarizes the information on external body proportions of the sampled whales, and provides the percentage of each measurement in relation to body length. The female and male external body measurements had nearly identical percentages. Table 4 summarizes the information on body parts and organ weights (kg) for the sampled whales. Figure 5 shows the body length distribution of the sampled whales, by sex. The maximum body length of males and females was 9.00m and 10.06m, respectively. The range for females was 5.27–9.00m and for males 5.17–10.06m.

Sexual maturity of sampled whale

Table 2 shows that mature animals were dominant in the East-South stratum, which is similar to results of previous surveys. Mature males were more frequent (66.7% male sex ration) in the North strata. Maturity rate of both sexes was high (93.9 % in female and 71.4 % in male) in the South strata (e.g. the maturity is higher than 50% for all strata in both sexes: 51.7–91.5%). Two lactating females with pregnancy were sampled in the West-South (24 December 2015) and East-South strata (13 February 2016).

The occurrence of lactating females was recorded in the early period of the survey, as in previous surveys. Secretion was observed in the mammary glands of the early-pregnant animals. These observations suggested that pregnant females migrated to the feeding ground after parturition. The animal with maximum testicular weight was also observed in the early period (December). These results reflect seasonal changes in the reproductive activity of the adult Antarctic minke whales.

Stomach contents

A total of 309 stomachs were examined (in 24 cases the stomach was broken). Table 5 shows the frequency of appearance of dominant prey species in the forestomach, by strata. Antarctic krill (*Euphausia superba*) was the dominant prey species (92.8% of the whales examined) in Area V. In the eastern part of Area V, south of 76°S, the dominant prey species was ice krill (*Euphausia crystallorophias*).

Other sampling

Age information is important for the main objective 1 of NEWREP-A. During the survey earplugs were collected from all animals sampled. Eye lenses were also collected for the purpose of age determination based on the ratio of aspartic acid enantiomers in the lens nucleus.

Some other samples collected such as baleen plate, prey species from stomach contents, blood, muscle, and biopsy skin would be useful for analysis of stable isotopes analyses to estimate the duration of Antarctic minke whale in the feeding grounds.

Data and samples obtained from this survey will be validated and stored at the Institute of Cetacean Research (ICR), Japan, and they are available to national (Japan) and international scientists under established guidelines (see http://www.icrwhale.org/NEWREP-AProtocol.html).

Catch data record will be submitted to the IWC secretary, as in the previous surveys.

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Table 1
Specifications of the research vessels.

	Yushin-Maru	*Yushin-Maru No.2*	*Nisshin-Maru*
Call sign	JLZS	JPPV	JJCJ
Length overall [m]	69.61	69.61	129.58
Molded breadth [m]	10.40	10.80	19.40
Gross tonnage [GT]	724	720	8,145
Barrel height [m]	19.5	19.5	–
Barrel height (IO) [m]	13.5	13.5	–
Upper bridge height [m]	11.5	11.5	–
Bow height [m]	6.5	6.5	–
Engine power [PS/ kW]	5,280/ 3,900	5,280/ 3,900	7,320/ 5,383
Captain	Yasuaki Sasaki	Nobuo Abe	Hiroshi Eguchi

Table 2
Mean (±SD) Body length (m) and weight (t) of Antarctic minke whale by the stratum in the NEWREP-A 2015/16.

Stratum	Sex	Sexual Maturity	Reproductive status	N	%	Body length (Mean±SD)	Body weight (Mean±SD)
West-North	Male	Immature		0	–	–	–
		Mature		0	–	–	–
	Female	Immature		1	100.0	6.03	2.59
		Mature	Maturing	0	–	–	–
			Resting	0	–	–	–
			Pregnant	0	–	–	–
			Pregnant & Lactation	0	–	–	–
East-North	Male	Immature		1	12.5	7.5	4.87
		Mature		6	75.0	8.41±0.33	6.90±1.02
	Female	Immature		1	12.5	5.19	1.98
		Mature	Maturing	0	–	–	–
			Resting	0	–	–	–
			Pregnant	0	–	–	–
			Pregnant & Lactation	0	–	–	–
West-South	Male	Immature		10	16.7	6.88±1.02	3.87±1.63
		Mature		15	25.0	8.29±0.33	6.46±0.82
	Female	Immature		19	31.7	6.42±0.90	3.16±1.36
		Mature	Maturing	1	1.7	9.22	7.54
			Resting	1	1.7	9.30	8.86
			Pregnant	13	21.7	8.92±0.33	8.15±0.97
			Pregnant & Lactation	1	1.7	10.06	11.21
East-South (Ross Sea)	Male	Immature		6	2.3	7.49 ±0.56	4.47±0.90
		Mature		65	24.6	8.23±0.35	6.47±0.80
	Female	Immature		35	13.3	7.45±0.75	4.92±1.23
		Mature	Maturing	5	1.9	8.88±0.41	7.85±0.64
			Resting	10	3.8	8.64±0.49	7.69±0.87
			Pregnant	142	53.8	8.76±0.37	7.66±1.02
			Pregnant & Lactation	1	0.4	8.58	7.73
Total				333		8.26±0.87	6.64±1.77

Table 3
External body proportions of Antarctic minke whales in the NEWREP-A 2015/16.

Measurement Point[1]	Male			Female		
	No of Measured Samples	Mean (cm)	%BL	No of Measured Samples	Mean (cm)	%BL
Total length (body length)[2]	230	835	100.0	103	806	100.0
Tip of snout to centre of eye[2]	229	164	19.6	103	154	19.1
Tip of snout to tip of flipper[2]	223	381	45.6	100	363	45.0
Tip of snout to end of ventral grooves[2]	230	428	51.2	103	406	50.4
Tip of snout to umbilicus[2]	229	458	54.8	103	439	54.5
Tip of snout to anus[2]	229	620	74.3	103	599	74.3
Girth of chest, half[2]	222	227	27.2	101	215	26.7
Girth of abdominal, half[2]	227	204	24.5	100	199	24.7
Skull length, condyle to tip of premaxilla[3]	224	202.6	24.3	101	192.2	23.8
Skull, greatest width[3]	221	110.7	13.3	94	105.6	13.1

1: Measurement point was described in Figure 5; 2: measured by 1cm intervals; 3: measured by 0.1cm intervals.

Table 4
Body parts and organ weight (kg) of Antarctic minke whales in NEWREP-A 2015/16.

Catch date	Sex	Body length (m)	Total parts	Blubber	Meat	Bone	Viscera and others
30 Dec 2015	F	8.74	6,736	1,451	3,578	1,002	705
5 Jan 2016	F	6.03	2,507	540	1,338	358	270
3 Feb 2016	F	5.19	1,889	510	905	270	205
8 Feb 2016	M	8.07	6,357	1,565	3,231	899	662
11 Feb 2016	M	8.53	6,501	1,463	3,501	923	613

Table 5
The Frequency of the appearance of dominant prey species found in forestomach contents of Antarctic minke whales sampled in the NEWREP-A 2015/16.

Stratum	Antarctic krill		Ice krill		Empty or trace		Broken	
	N	%	N	%	N	%	N	%
West-North	1	0.3	0	0	0	0	0	0
East-North	5	1.5	0	0	2	0.6	1	0.3
West-South	35	10.5	0	0	20	6	5	1.5
East-South	163	48.9	11	3.3	72	21.6	18	5.4
Total	204	61.3	11	3.3	94	28.2	24	7.2

N: number of whales examined; Broken: animals with broken stomach by harpoon.

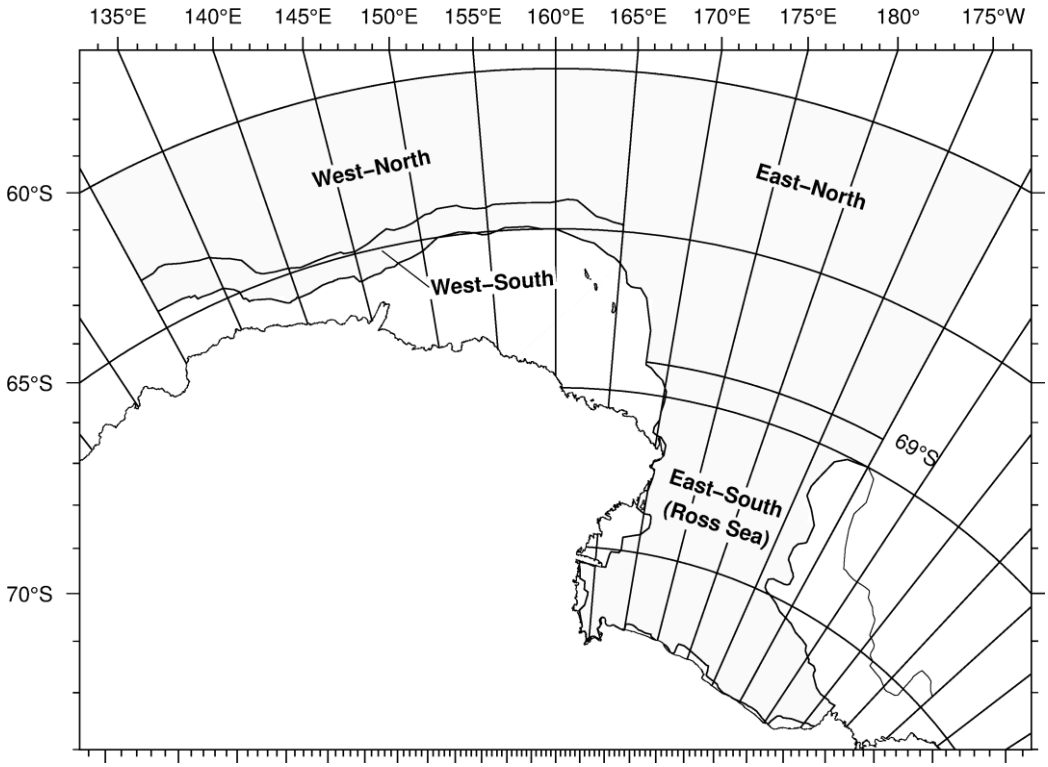

Figure 1a. Stratification of sampling survey for Antarctic minke whales in the NEWREP-A 2015/16 research area. Gray shade; research area of the NEWREP-A 2015/16. The boundary between North and South strata of each research area was the 45n.mile lines from estimated ice-edge line except for the east strata of Area V. The boundary between North and South boundary of east strata of Area V (from 165°E to 170°W, in longitude) was 69°S in latitude. The map using Lambert conic conformal projection.

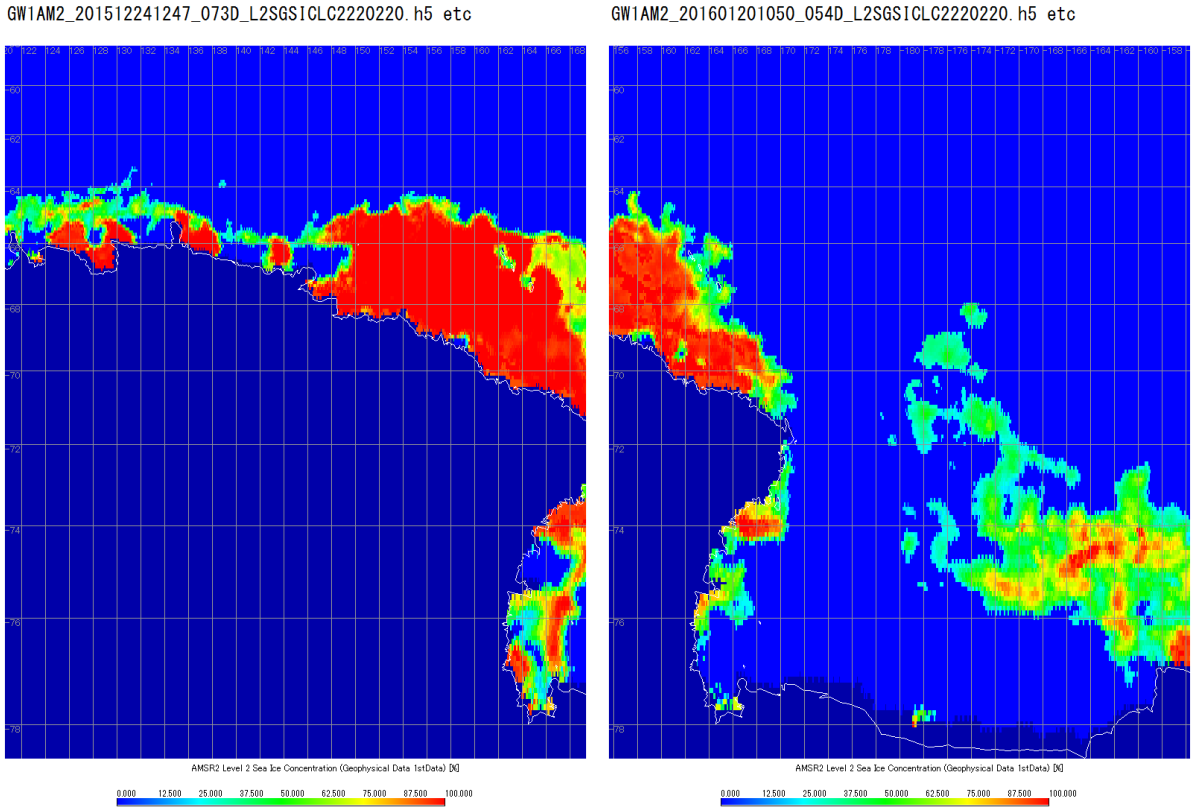

Figure 1b. Spatial distribution of representative sea ice concentration of AMSR2 data during NEWREP-A 2016 cruise. Right: 24 Dec 2016 in West sector. Open water areas (Antarctica polynyas) existed along the continent's coastline. Left: 20 Jan 2016 in East sector. Colour bar denotes sea ice concentration of each map.

Figure 2. Research vessels which were used in NEWREP-A 2015/16. Upper left: *Yushin-Maru*; upper right: *Yushin-Maru No.2*; lower: *Nisshin-Maru.*

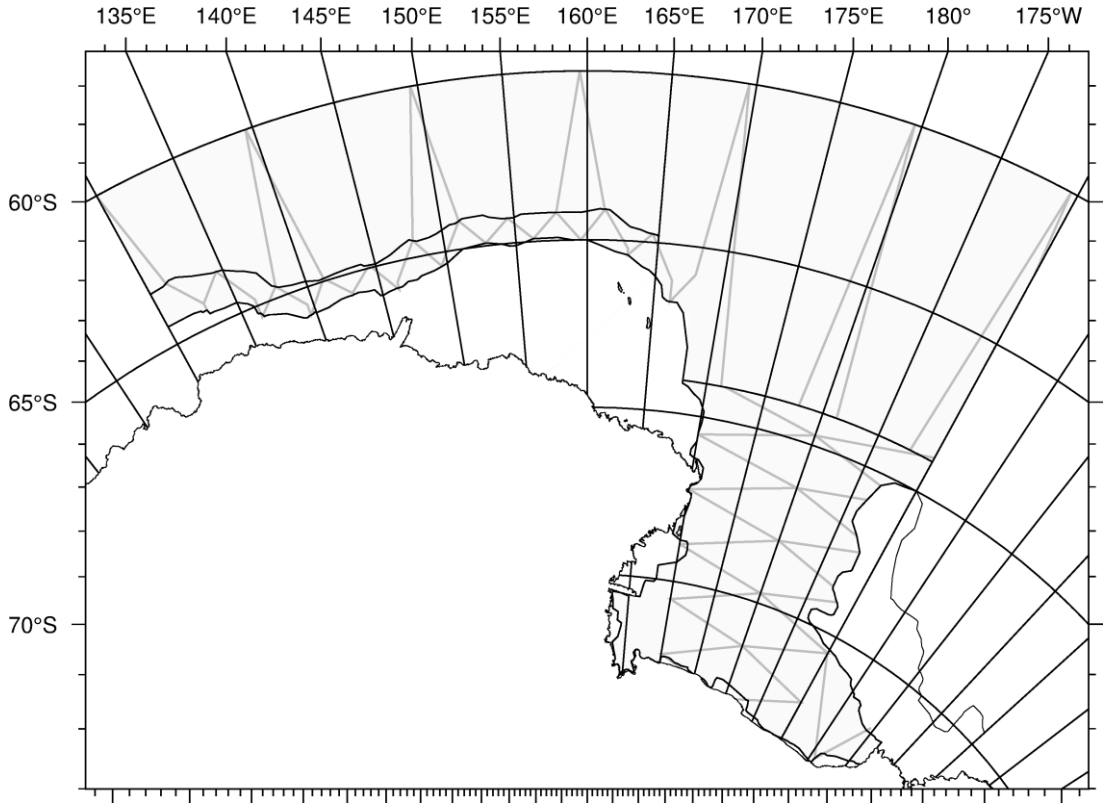

Figure 3a. Predetermined track-line (Gray lines: main course) of sampling survey for Antarctic minke whales in the NEWREP-A 2015/16. The map using Lambert conic conformal projection.

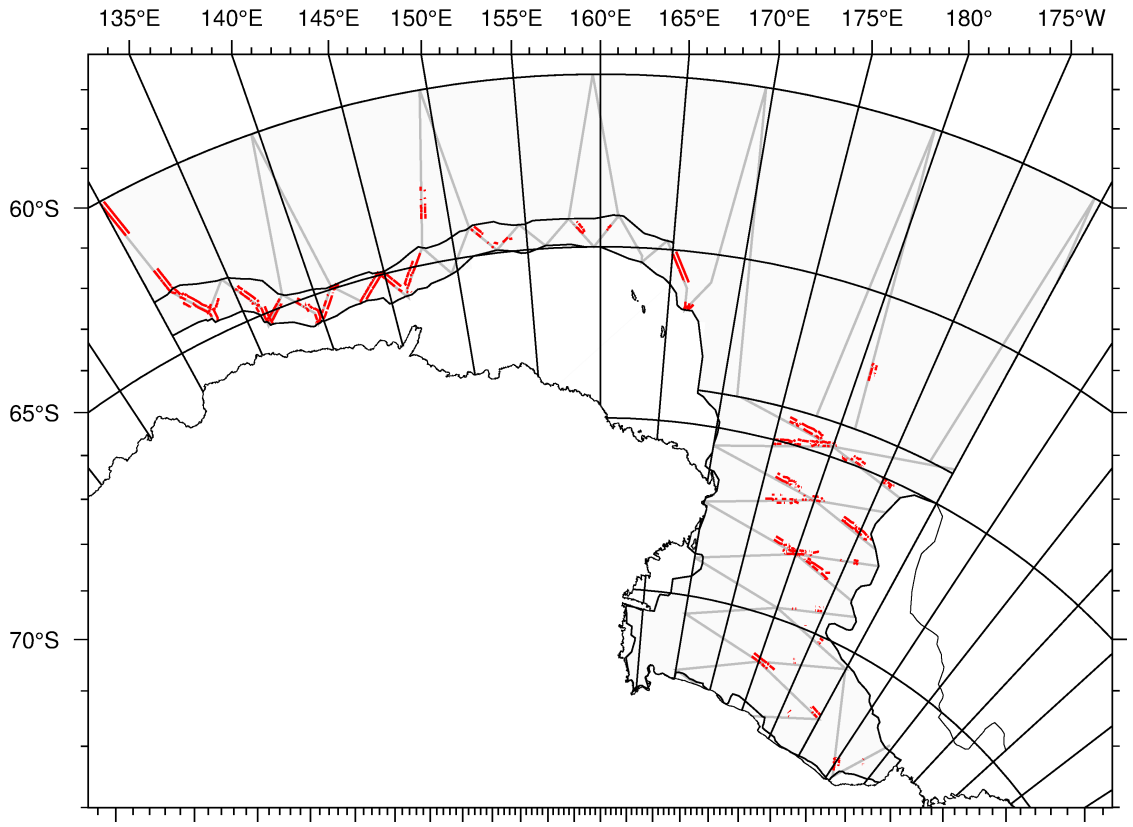

Figure 3b. The portion of the vessel's track-line (red line) that was surveyed on the effort of sampling survey for Antarctic minke whales in the NEWREP-A 2015/16. Two parallel lines show the main- and sub-courses.

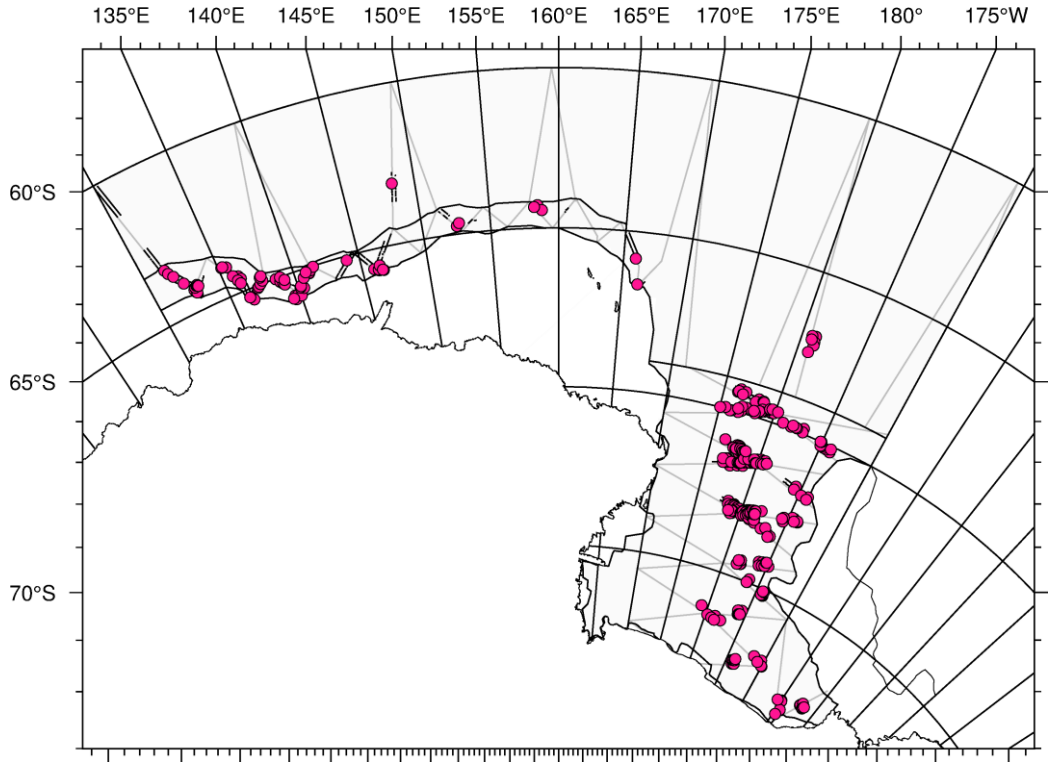

Figure 3c. Geographical distribution of primary sighting position of Antarctic minke whales (pink closed circle) sighted in the NEWREP-A 2015/16 sampling survey. Black bold line of the map was the boundary line for a stratum of the research area. Gray lines were the predetermined track-line. The black line was the track-line of the searching effort. The map using Lambert conic conformal projection.

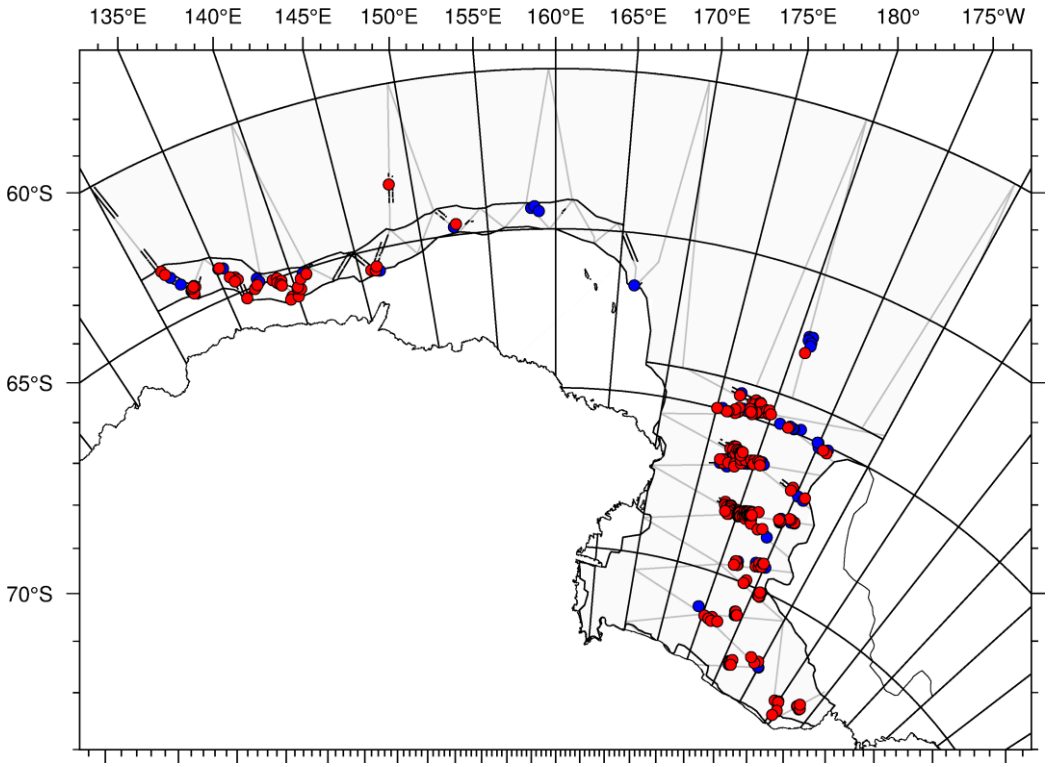

Figure 3d. Geographical distribution of sighting position of Antarctic minke whale sampled in the NEWREP-A 2015/16. One plot showed the sighting position of the one school of the primary sighting. In cases where the school consists of multiple individuals, some samples were sampled less than two individuals/school. Red closed circle: female; blue closed circle: male. Black bold line of each map was the boundary line for a stratum of the each research area. Gray lines were the predetermined track-line. The black line was the track-line of the searching effort. The map using Lambert conic conformal projection.

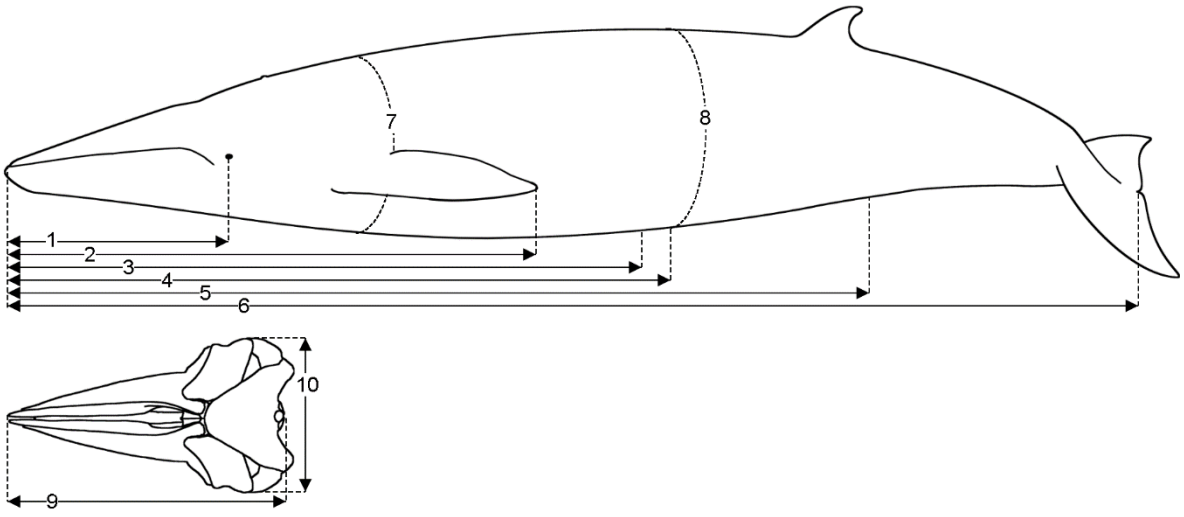

Figure 4. Measurement points of external proportions for Antarctic minke whale in the NEWREP-A 2015/16. The number corresponds to row number is below the keys. 1: tip of snout to centre of eye; 2: tip of snout to tip of flipper; 3: tip of snout to end of ventral grooves; 4: tip of snout to umbilicus; 5: tip of snout to anus; 6: total length; 7: girth of chest, half; 8: girth of abdominal, half; 9: skull length, condyle to tip of premaxilla; 10: skull, greatest width.

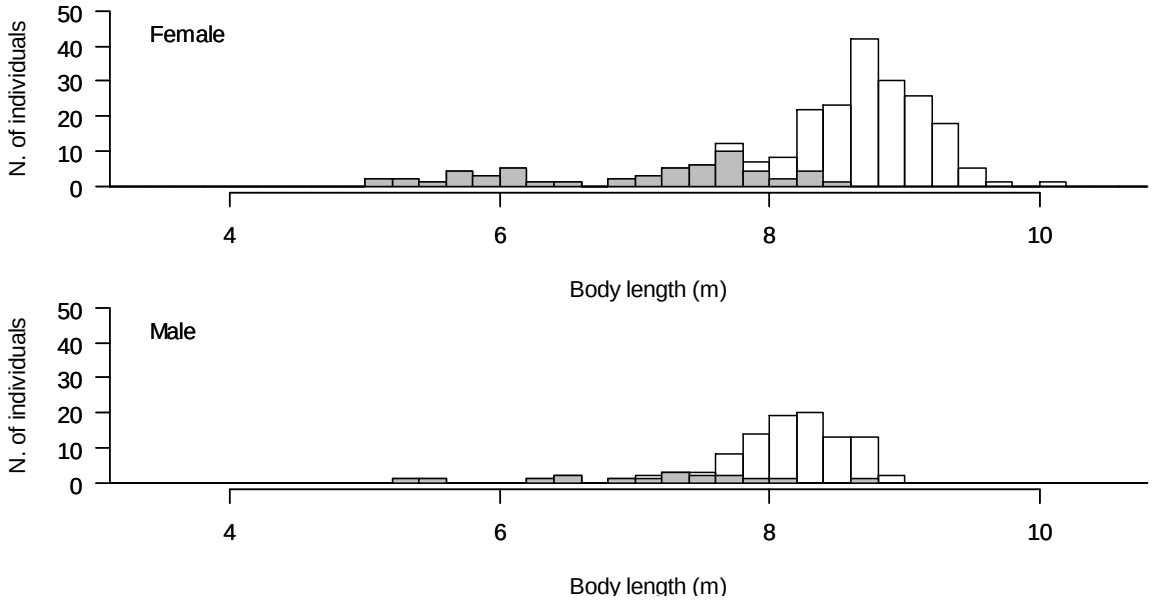

Figure 5. Histogram of Antarctic minke whales body length in the NEWREP-A 2015/16. Upper: female; lower: male. Two vectors colours mean the value of the stature in maturity. White: mature individuals; gray: immature individuals. The breakpoints between histogram cells were 0.20m intervals.

APPENDIX

Sighting results by the sighting and sampling vessels during NEWREP-A 2015/16 in Area V

IDENTIFICATION OF SPECIES

Guidelines for species identification were based on the IWC-SOWER methods for classification of identification (IWC, 2008): 'Positive identification of species was based on multiple cues and usually required clear observation of the whale's body. Occasionally, repeated observations of the shape of the blow, surfacing, and other behavioural patterns were sufficient; this judgement was made only by the Cruise leader or other designated researcher. Identification of species was recorded as 'probable' based on multiple cues, which were nevertheless insufficient to be absolutely confident of identification. This usually occurred when blows and surfacing patterns could be confirmed, but the whale's body could not be clearly seen. Details of recording procedures during sightings can be found in 'Information for Researchers''. From initial observations of morphologic characteristics, killer whale data were divided into three ecotypes (Pitman and Ensor, 2003).

DETERMINATION OF GROUP SIZE

Following guidelines were used in determining group size (IWC, 2008): 'Schools where the number of animals or an accurately estimated range of the number of animals was determined, were classified as confirmed schools. Data from the confirmed schools can be used to determine a mean school size. Therefore, it is critical that the confirmed schools accurately represent the size of schools in the survey area. Normally, schools believed to be confirmed for school size are approached to within 1 n. mile for large whales and to within 0.3n. miles for minke whales. Allowing for context-specific differences (i.e. environmental conditions and animal behaviour), every effort was made to be consistent with regard to the maximum time spent on the identification of species and confirmation of numbers. Normally, if the sighting was thought to be minke whales, no more than 20 minutes (after the closure has been completed) should be spent on confirmation. This reduces the potential for confusion with other sightings in the vicinity. Counts of individuals provided by the sighting summary represent best estimates of school sizes in the research area, except when indicated otherwise'.

OTHER RESEARCH ACTIVITIES

Photo-ID

Photo-ID data of individual whales was collected for stock structure analyses as well as examining mixing and movements during the research time. Data were captured by digital photograph using Digital camera (300mm lens, Canon Co., Ltd., Japan). Target species for Photo-ID were blue whale (*B. musculus*), humpback whale (*Megaptera novaeangliae*), southern right whale (*Eubalaena australis*), and killer whale.

Biopsy sampling for large whales

Samples of skin biopsy were collected by biopsy from the target whales. The target species were blue whale, fin whale (*B. physalus*), sei whale (*B. borealis*), southern right whale, sperm whale (*Physeter macrocehpalus*), killer whale, pygmy right whale (*Caperea marginata*), and southern bottlenose whale (*Hyperoodon planifrons*) sighted in the research time. The system for biopsy sampling was a Larsen gun (Larsen, 1998). The open sight was replaced with an electronic aiming device (red-dot-sight), which allows faster aiming and thus faster shooting. The biopsy darts consisted of a carbon fibre shaft, which is high-pressure moulded to a polyethylene float that also functions as a stop to limit penetration into the tissue. In the float end of the dart, a threaded insert is used for attaching the screw-on biopsy-sampling tip. The biopsy tip is a stainless steel cylinder with a 9mm outer diameter, an internal diameter of 7mm and three internal barbs for sample retention. All collected samples were stored at −80°C.

RESULTS

Sightings

The searching effort and experimental times are summarised in Table 1. The portion of the SSV's track-line that was surveyed on the effort of sampling survey is shown in Table 2. Total searching distance in the research area was 2,394n.miles in Area V (main and sub course). The sighting records are not suitable for design-based abundance estimation as the sighting records of the sampling and survey vessels were made by NSC mode using TOP and UBP except for examination of the distribution of specified species using model-based estimation. The sightings recorded in the sampling survey are shown in Table 3. Figures 1a–b show the geographical distribution

of baleen whales sighted in sampling survey (Figure 1a: blue and fin whales; Figure 1b: humpback whales). Figure 2 shows the geographical distribution of toothed whales (sperm whale, Southern bottlenose whale, Arnoux's beaked whale, and killer whales) sighted in sampling survey. In Area V, Antarctic minke whale was the most abundant species followed by humpback whales. Distribution of Antarctic minke whales was narrower than that of humpback whales, mainly found in East-South strata (Ross Sea) and West-South stratum, which were concentrated at the ice edge. As for observations of vomit in Antarctic minke whales, during a total of 284 school 825 individuals (7 hours 22 minutes) observations, no vomit behaviours were recorded during the sampling survey.

From the differences in morphologic characteristics of killer whales, the effort was made to identify them into three ecotypes (types A, B and C). A total of 13 schools 101 individuals were sighted and the most sighted schools (12 schools) were 'not identified ecotypes (undetermined)' due to the difficulty of approaching the whales, and recorded as 'killer whales (undetermined)'. One individual was identified as type C (1 school/ 1 individual), and the whale was photographed as photo-ID data. One biopsy sample was collected from this species. Further observation, experiments, and analysis of this species will be continued during the next cruise based on this programme.

After examination of sighting distance and angle experiments, total experiment times were 7 hours and 47 minutes in total survey hour by three vessels during the sampling survey and dedicated sighting survey. The results of this experiment will be used for the calculation of abundance estimates.

Photo ID and biopsy

A total of 13 Photo-ID data was obtained from 3 species (blue, humpback and killer whales). In terms of biopsy skin sampling, a total of 7 individuals were sampled from 3 species (blue, humpback and killer whales). The number of Photo-ID and biopsy samples are summarised in Tables 4a–4b. Distribution of the sighting position of photo-ID data and biopsy sampling are shown in Figure 3a and 3b, respectively. Table 5a summarises the details of the Photo-ID and biopsy sampling. Table 5b summarises the details of the encounter duration of the biopsy sampling.

Photo-ID records were collected for four blue whales, nine humpback whales and one killer whale. The records will provide useful data for understanding the stock structure, mixing and movements of these species. These data will be submitted to the IWC secretary and will be analysed comparing previous IWC and other relevant catalogues (e.g. Olson, 2012, Matsuoka and Pastene, 2009).

Furthermore, biopsy skin samples were obtained from a total of 13 individuals in one research season. These biopsy samples obtained during surveys of the NEWREP-A 2015/16 will be used for microsatellite DNA loci analysis for studies of stock structure in the Antarctic feeding ground. Biopsy samples may also be used for other research (e.g. chemical markers as body condition indicator, stable isotope, or hormones).

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Table 1
Summary of research effort in NEWREP-A 2015/16.

Strata	Date		Days	NSC		NSP		Photo-ID Biopsy	Estimated angle and distance training
	Start	End		Time	Dist.	Time	Dist.	Time	Time
West-North	23 Dec 12:00	6 Jan 8:26	4	23:29	274.5	0:00	0.0	0:00	0:00
East-North	9 Jan 6:00	3 Feb 13:23	8	17:29	187.4	0:00	0.0	0:00	0:00
West-South	24 Dec 10:05	7 Jan 11:09	12	96:09	987.0	0:00	0.0	2:10	7:47
East-South (Ross Sea)	12 Jan 6:00	25 Feb 18:00	41	91:27	945.5	0:00	0.0	0:26	0:00
Total	23 Dec 12:00	25 Feb 18:00	65	228:36	2,394.3	0:00	0.0	2:36	7:47

Table 2
Predetermined waypoints (WP) and the actually covered ratio of sampling survey in Area V.

Strata	Area Code	Mode	WP No.	Leg. No.	Latitude	Longitude	Course	Plan	Effort	Covered (%)
West-North	53	NSC	601	601	60°05'S	130°00'E	169°	193.4	40.9	21.2
			602	602	63°15'S	131°14'E	—	—	—	—
			606	606	64°40'S	137°54'E	009°	284.2	0.0	0.0
			607	607	60°00'S	139°34'E	171°	299.0	0.0	0.0
			608	608	64°55'S	141°14'E	—	—	—	—
			612	612	64°30'S	147°54'E	009°	274.3	29.3	10.7
			613	613	60°00'S	149°34'E	169°	254.6	0.0	0.0
			614	614	64°10'S	151°14'E	—	—	—	—
			618	618	64°30'S	157°54'E	009°	274.3	0.0	0.0
			619	619	60°00'S	159°34'E	169°	254.6	0.0	0.0
			620	620	64°10'S	161°14'E	—	—	—	—
East-North	54	NSC	623	623	65°06'S	165°00'E	152°	66.6	49.7	74.6
			624'	624	66°05'S	166°14'E	180°	38.0	22.3	21.3
			624	624	66°43'S	166°14'E	037°	66.7	0.0	0.0
			625	625	65°50'S	167°54'E	141°	64.3	0.0	0.0
			625	625	65°50'S	167°54'E	007°	353.4	0.0	0.0
			626	626	60°00'S	169°34'E	175°	542.5	0.0	0.0
			627	627	69°00'S	171°14'E	—	—	—	—
			631	631	69°00'S	177°54'E	004°	542.5	0.0	0.0
			632	623	60°00'S	179°34'E	175°	542.5	25.6	4.7
			633	633	69°00'S	178°46'W	—	—	—	—
			637	637	69°00'S	172°06'W	004°	542.5	0.0	0.0
			638	638	60°00'S	170°26'W	175°	158.7	0.0	0.0
			639	639	62°38'S	170°00'W	—	—	—	—

Table 2
Continued.

Strata	Area Code	Mode	WP No.	Leg. No.	Latitude	Longitude	Course	Plan	Effort	Covered (%)
West-South	51	NSC	502	502	63°15'S	131°14'E	144°	76.3	51.9	68.0
			503'	503	64°17'S	132°54'E	180°	13.0	18.8	22.5
			503	503	64°30'S	132°54'E	038°	70.4	0.0	0.0
			504	504	63°35'S	134°34'E	149°	85.1	37.9	44.5
			505'	505	64°48'S	136°14'E	180°	34.0	70.8	75.5
			505	505	65°22'S	136°14'E	045°	59.7	0.0	0.0
			506	506	64°40'S	137°54'E	139°	65.5	19.0	29.0
			507'	507	65°30'S	139°34'E	180°	20.0	54.2	60.8
			507	507	65°50'S	139°34'E	037°	69.1	0.0	0.0
			508	508	64°55'S	141°14'E	133°	58.0	9.7	16.6
			509	509	65°35'S	142°54'E	046°	58.0	50.2	86.6
			510	510	64°55'S	144°34'E	142°	69.1	43.7	63.3
			511'	511	65°50'S	146°14'E	000°	13.0	59.3	64.3
			511	511	65°37'S	146°14'E	032°	79.3	0.0	0.0
			512	512	64°30'S	147°54'E	141°	68.8	0.0	0.0
			513	513	65°24'S	149°34'E	029°	85.5	0.0	0.0
			514	514	64°10'S	151°14'E	136°	62.4	19.2	30.8
			515	515	64°55'S	152°54'E	043°	62.4	11.7	18.7
			516	516	64°15'S	154°34'E	132°	58.8	0.0	0.0
			517	517	64°55'S	156°14'E	043°	62.4	0.0	0.0
			518	518	64°10'S	157°54'E	139°	66.0	16.7	25.3
			519	519	65°00'S	159°34'E	038°	69.9	9.7	13.9
			520	520	64°05'S	161°14'E	148°	81.3	0.0	0.0
			521'	521	65°14'S	162°54'E	180°	11.0	0.0	0.0
			521	521	65°25'S	162°54'E	022°	108.4	0.0	0.0
			522	522	64°45'S	164°34'E	152°	23.8	0.0	0.0
			523	523	65°06'S	165°00'E	152°	66.6	49.7	74.6
East-South (Ross Sea)	52	NSC	701	701	69°00'S	171°34.5'E	103°	183.7	55.5	30.2
			702	702	69°42'S	180°00'E	265°	210.7	95.0	45.1
			703	703	70°32'S	170°00'E	104°	202.7	29.8	14.7
			704	704	71°22'S	180°00'E	255°	194.7	44.8	23.0
			705	705	72°12'S	170°00'E	105°	186.6	26.7	14.3
			706	706	73°02'S	180°00'E	253°	178.6	17.4	9.8
			707	707	73°52'S	170°00'E	107°	170.5	0.0	0.0
			708	708	74°42'S	180°00'E	252°	162.5	0.0	0.0
			709	709	75°32'S	170°00'E	108°	154.4	13.6	8.8
			710	710	76°22'S	180°00'E	249°	146.4	0.0	0.0
			711	711	77°12'S	170°00'E	111°	138.4	0.0	0.0
			712	712	78°02'S	180°00'E	—	—	—	—
			713	713	78°02'S	180°00'E	068°	138.4	0.8	0.5
			714	714	77°12'S	170°00'W	290°	146.4	23.3	15.9
			715	715	76°22'S	180°00'E	071°	154.4	0.4	0.2
			716	716	75°32'S	170°00'W	287°	162.5	1.7	1.1
			717	717	74°42'S	180°00'E	072°	170.5	10.0	5.9
			718	718	73°52'S	170°00'W	286°	178.6	39.5	22.1
			719	719	73°02'S	180°00'E	074°	186.6	25.2	13.5
			720	720	72°12'S	170°00'W	284°	194.7	50.0	25.7
			721	721	71°22'S	180°00'E	075°	202.7	8.1	4.0
			722	722	70°32'S	170°00'W	283°	210.7	23.6	11.2
			723	723	69°42'S	180°00'E	076°	181.8	0.0	0.0
			724	724	69°00'S	171°40'W	—	—	—	—
			731	731	75°35'S	170°00'W	155°	187.3	21.7	11.6
			732	732	78°25'S	164°15'W	025°	114.5	0.3	0.3
			733	733	76°42'S	160°26'W	—	—	—	—

Table 3
Number of whales sighted in Area V during sampling survey by each stratum (school/ individuals).

Species	West-North				East-North				West-South				East-South				Sub total				Total	
	Prim.		Second.		Prim.		Second.		Prim.		Second.		Prim.		Second.		Prim.		Second.			
	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.	Sch.	Ind.
Blue whale	0	0	0	0	0	0	0	0	3	4	0	0	0	0	2	3	3	4	2	3	5	7
Fin whale	0	0	0	0	4	11	1	1	2	4	1	3	1	2	1	2	7	17	3	6	10	23
Antarctic minke whale	1	1	1	1	8	14	0	0	59	84	7	10	267	816	27	95	335	915	35	106	370	1021
Like minke whale	0	0	0	0	1	1	0	0	2	2	0	0	4	4	0	0	7	7	0	0	7	7
Humpback whale	13	30	0	0	7	22	1	1	120	187	2	4	4	6	1	1	144	245	4	6	148	251
Baleen whales	0	0	0	0	1	3	0	0	5	6	0	0	0	0	0	0	6	9	0	0	6	9
Sperm whale	0	0	0	0	0	0	0	0	12	12	1	1	0	0	0	0	12	12	1	1	13	13
Southern bottlenose whale	0	0	0	0	0	0	0	0	2	4	0	0	0	0	0	0	2	4	0	0	2	4
Arnoux's beaked whale	0	0	0	0	0	0	0	0	0	0	1	7	2	20	0	0	2	20	1	7	3	27
Unidentified beaked whales	1	1	0	0	2	2	1	1	6	7	0	0	0	0	0	0	9	10	1	1	10	11
Killer whale	1	4	0	0	0	0	0	0	5	37	0	0	7	60	0	0	13	101	0	0	13	101
Unidentified whale	2	2	0	0	1	1	0	0	14	14	0	0	19	23	0	0	36	40	0	0	36	40
Unidentified dolphin	1	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	4	0	0	1	4

Prim: primary sightings; Second: secondary sightings; Sch: school; Ind: individuals.

Table 4a
Number of individuals photo-identified in the NEWREP-A 2015/16.

Species	No. of individuals photographed
Blue whale	3
Humpback whale	9
Killer whale	1
Total	13

Table 4b
Number of biopsy samples collected in the NEWREP-A 2015/16.

Species	No. of individuals collected
Blue whale	1
Humpback whale	5
Killer whale*	1
Total	7

*: Type C.

Table 5a
Details of the Photo-ID and biopsy sampling

Date	Sight No.	Species	Sighted position		School size	Est. body length of target Ind. [m]	N. of marked Ind.	N. of shoots	Opportunity of shoot	Position of shoot	Biopsy sample No.	Notes.
			Lat.	Long.								
YS1												
24 Dec 2015	11	Humpback	63°32.89'S	132°03.41'E	1	12	1	2	P	LD,FL,	J15YS1H001	-
24 Dec 2015	12	Humpback	63°35.29'S	132°03.93'E	2	12.7	2	1	P	LD,	J15YS1H002	-
4 Jan 2016	6	Humpback	65°14.31'S	146°44.08'E	2	12.7, 13.0	2	4	G	LD,RD,	J15YS1H003	-
4 Jan 2016	7	Humpback	65°10.16'S	146°51.07'E	3	10.9, 11.3, 11.7	3	5	G	LD,RD,	J15YS1H004	-
4 Jan 2016	8	Humpback	65°08.04'S	147°44.00'E	1	12.6	1	2	G	LD,RD,	J15YS1H005	-
14 Feb 2016	5	Killer	78°01.76'S	164°33.13'W	1	5.8	1	3	P	LD,RD,HD	J15YS1K001	TypeC
YS2												
26 Dec 2015	7	Blue	64°09.20'S	133°40.66'E	1	24.3	1	26	P	RD	-	-
26 Dec 2015	8	Blue	64°05.58'S	133°45.26'E	2	23.2, 18.1	1	48	G	RD	-	Mother & calf pair
27 Dec 2015	6	Blue	64°02.53'S	135°12.13'E	1	22.1	1	40	P	HD	J15YS2B0001	-

LD: left dorsal; RD: right dorsal, HD: head.

Table 5b
Details of the encounter duration in biopsy sampling.

Date	Sight No.	Species	School Size	BX time			Est. body length of target Ind. [m]	N. of shoots	N. of hits	Struck position	N. of samples	Sample No.	Shooting Equipment	Notes.
				Start	End	Duration								
YS1														
24 Dec 2015	11	Humpback	1	13:14:39	13:25:54	0:11:15	12	1	1	LB1a	1	J15YS1H001	Larsen	-
24 Dec 2015	12	Humpback	2	13:40:49	13:44:18	0:03:29	12.7	1	1	C1	1	J15YS1H002	Larsen	-
4 Jan 2016	6	Humpback	2	13:16:55	13:41:48	0:24:53	12.7	2	1	RC2	1	J15YS1H003	Larsen	-
4 Jan 2016	7	Humpback	3	14:22:40	14:30:12	0:07:32	11.7	2	2	RC1, RB1a	1	J15YS1H004	Larsen	-
4 Jan 2016	8	Humpback	1	14:55:45	14:59:35	0:03:50	12.6	1	1	C2	1	J15YS1H005	Larsen	-
14 Feb 2016	5	Killer	1	9:41:09	10:07:10	0:26:01	5.8	4	2	C1	1	J15YS1K001	Larsen	Type C
YS2														
26 Dec 2015	7	Blue	1	14:23:40	14:52:26	0:28:46	24.3	1	0	-	0	-	Larsen	-
26 Dec 2015	8	Blue	2	15:23:03	15:56:26	0:33:23	23.1, 18.1	0	0	-	0	-	Larsen	Mother & calf pair
27 Dec 2015	6	Blue	1	6:24:14	6:40:43	0:16:29	22.1	1	1	Bla	1	J15YS2B001	Larsen	-

Larsen: Larsen biopsy system.

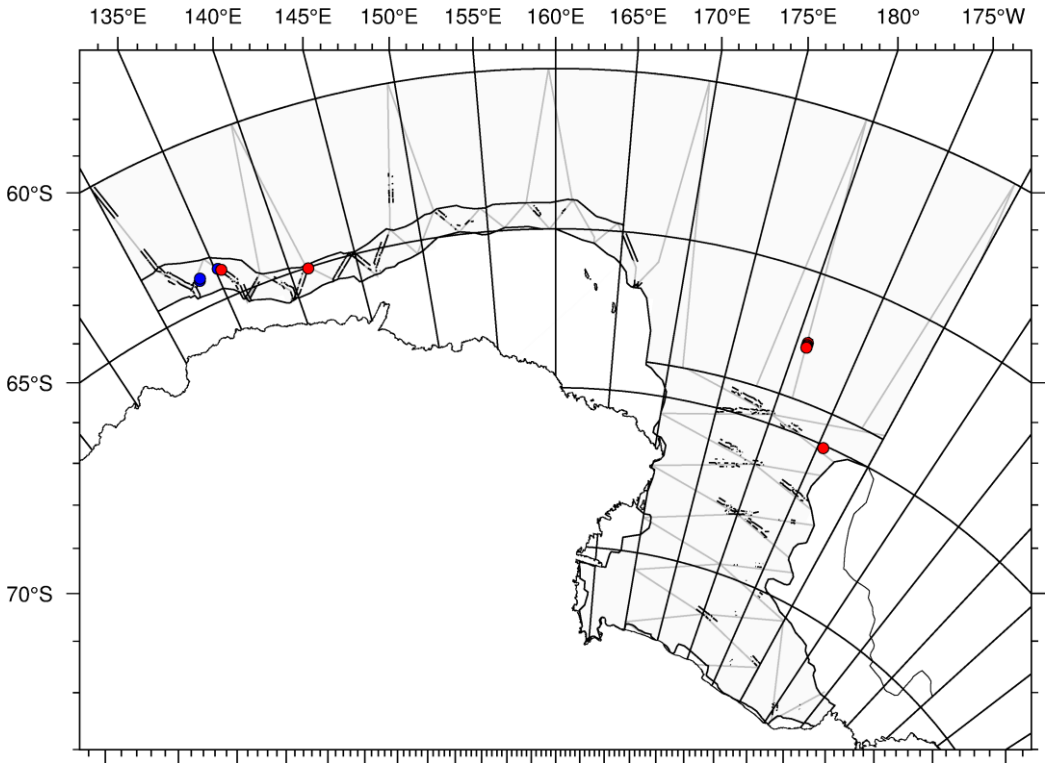

Figure 1a. Geographical distribution of primary sighting position of blue (blue closed circle) and fin (red closed circle) whales sighted in the NEWREP-A 2015/16 sampling survey. Gray line of the map was the predetermined track-line. The black line was the track-line of the searching effort. The map using Lambert conic conformal projection.

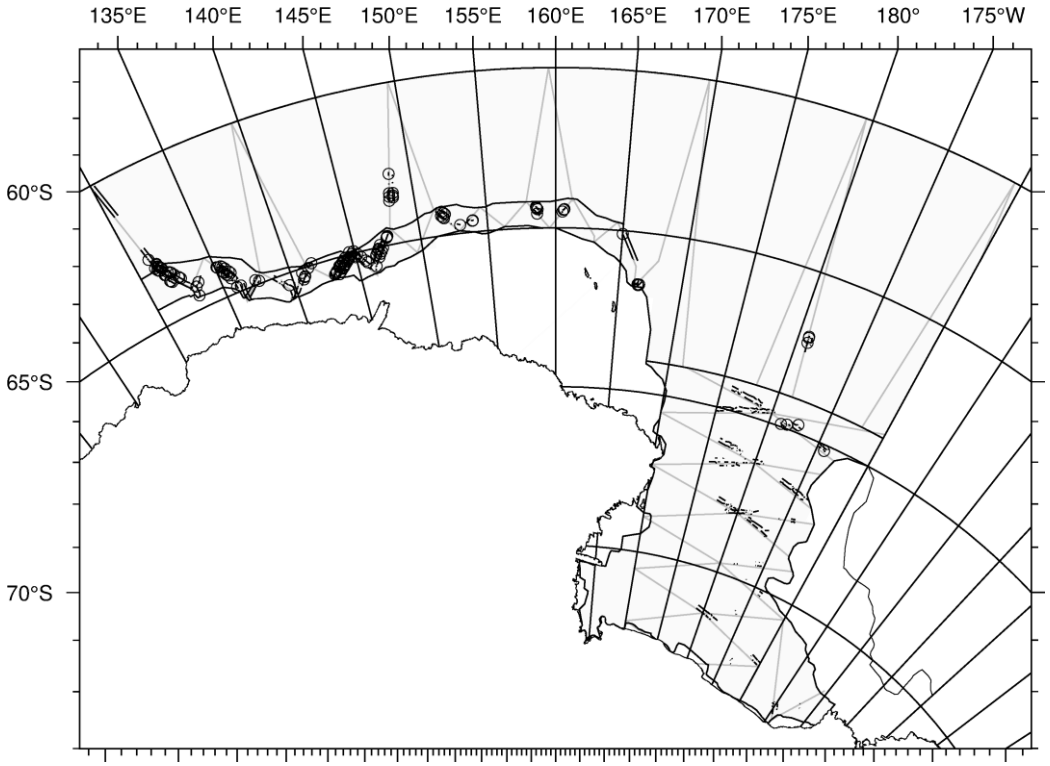

Figure 1b. Geographical distribution of primary sighting position of humpback whales (black open circle) sighted in the NEWREP-A 2015/16 sampling survey. Black bold line of the map was the boundary line for a stratum of the research area. Gray line was the predetermined track-line. The black line was the track-line of the searching effort. The map using Lambert conic conformal projection.

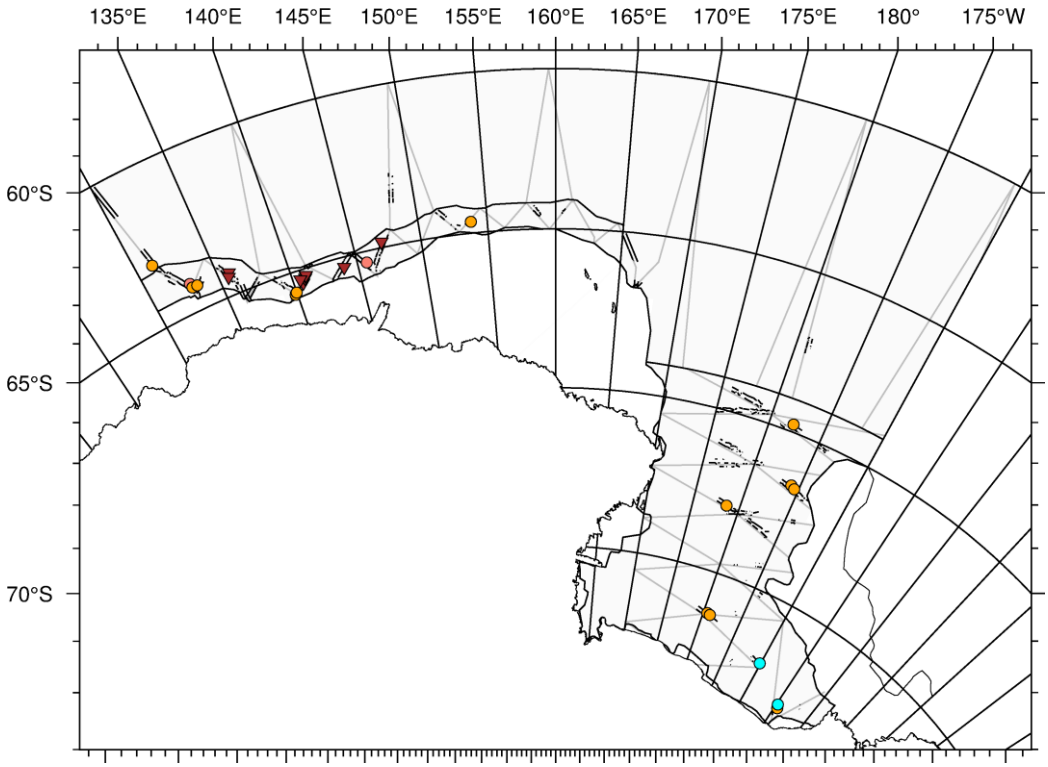

Figure 2. Geographical distribution of primary sighting position of sperm (brown closed reverse triangle), Southern bottlenose (light red closed circle), Arnoux's Beaked Whale (cyan closed circle) and killer (orange closed circle) whales sighted in the NEWREP-A 2015/16 sampling survey. Black bold line of the map was the boundary line for a stratum of the research area. Gray lines were the predetermined track-line. The black line was the track-line of the searching effort. The map using Lambert conic conformal projection.

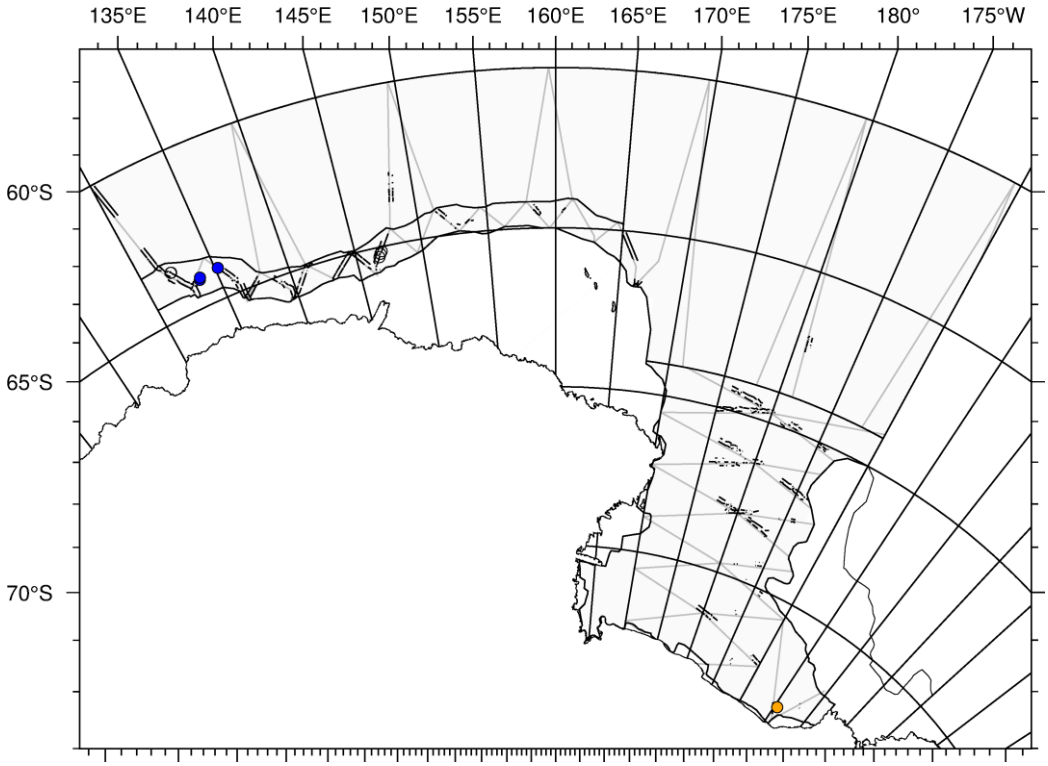

Figure 3a. Geographical distribution of Photo-ID data from blue (blue closed circle), humpback (black open circle) and killer whale (orange closed circle) collected during the NEWREP-A 2015/16 sampling survey. A single symbol could represent more than one photograph data. Black bold line of the map was the boundary line for a stratum of the research area. Gray lines were the predetermined track-line. The black line was the track-line of the searching effort. The map using Lambert conic conformal projection.

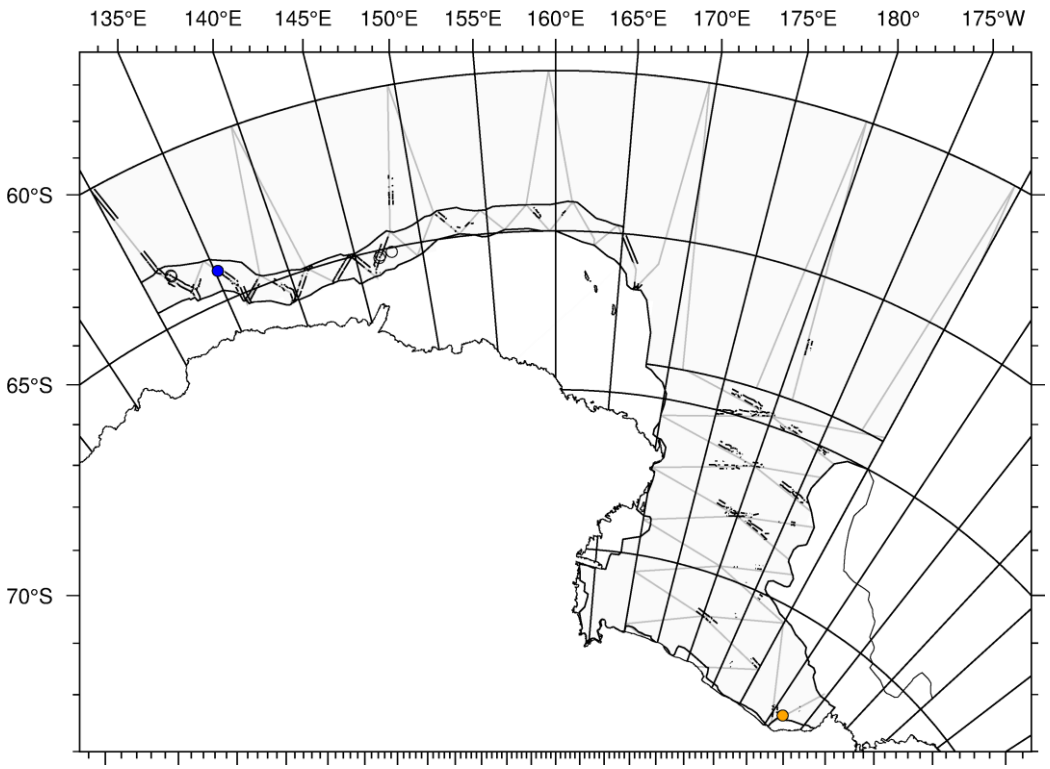

Figure 3b. Geographical distribution of biopsy samples of blue (blue closed circle), humpback (black opened circle) and killer whales (orange closed circle) collected during the NEWREP-A 2015/16 sampling survey. Black bold line of the map was the boundary line for a stratum of the research area. Gray lines were the predetermined track-line. The black line was the track-line of the searching effort. The map using Lambert conic conformal projection.