Research Plan for New Scientific Whale Research Program in the Antarctic Ocean (NEWREP-A)

The Government of Japan

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1. INTRODUCTION

The original proposal of the research plan was drafted and submitted to the Secretary to the International Whaling Commission (IWC) and the Chair of its Scientific Committee (SC) on 18 November 2014, in conformity with paragraph 30 of the Schedule to the ICRW and Annex P (IWC, 2013b). Subsequently it was reviewed by the Expert Panel in February 2015 (NEWREP-A Review Panel), and then additional materials (in the form of 'preliminary response' and 'addendum') was reviewed by the IWC SC at its 2015 annual meeting from May to June, 2015.. The research plan has been now finalized taking into account the comments and suggestions on scientific aspects provided by experts inside and outside the IWC SC, in particular those from the NEWREP-A Review Panel, and from the IWC SC members. This research plan forms the basis for the Government of Japan to issue a special permit, in accordance with Article VIII, paragraph 1, of the International Convention for the Regulation of Whaling (ICRW). Even after the research plan is finalized, the proponent always welcomes comments from outside on this plan that are based upon scientific consideration to which it will give due regard. Furthermore, this research plan could be revised in relation to the lethal sample size, depending on the results of the analysis of non-lethal research methods detailed in Section 4 below.

1.1 Importance of scientific research in the Antarctic Ocean

The Antarctic Ocean has its unique marine ecosystem and has the potential of its abundant living resources that could be sustainably exploited for food and other purposes. In recent years, the surrounding Antarctic region has been substantially affected by climate change and the resultant fluctuations in the oceanographic environment are known to have influences on the global environment.

The Antarctic marine ecosystem is defined in Article 1, paragraph 3, of the Convention on the Conservation of Antarctic Marine Living Resources (Convention-CAMLR) as "the complex of relationships of Antarctic marine living resources with each other and with their physical environment". This effectively means that it is composed of and made from complex interactions among the Antarctic Ocean, its circumpolar currents, sea ice formations and biota including 17 species of cetaceans. Among these, blue (*Balaenoptera musculus*), fin (*B. physalus*), humpback (*Megaptera novaeangliae*), and Antarctic minke (*B. bonaerensis*) whales are the top krill predators. Other components of the ecosystem include pinnipeds (*e.g.* crabeater seal), birds (*e.g.* penguins), and fish (*e.g.* toothfish) all of which are directly or indirectly supported by the massive krill resources. For more than one hundred years, mankind has been utilizing Antarctic marine ecosystem resources, including whales, seals, toothfish, and krill. Some of these have been severely overexploited in the course of history. The ICRW and Convention-CAMLR were promulgated in order to conserve and manage these important resources and to maintain their harvest potential for future generations.

In order to achieve conservation of its resources while pursuing their sustainable utilization and to understand and predict the effects of factors such as climate change, it is scientifically imperative to obtain an accurate understanding of many aspects of the Antarctic marine ecosystem including its animals and their dynamics through collection, accumulation, and analysis of scientific data.

Long term research surveys in the Antarctic are scarce. Systematic sighting surveys were conducted during the International Decade for Cetacean Research/Southern Ocean Whale and Ecosystem Research (IDCR/SOWER) under the auspices of the IWC SC for decades (Matsuoka *et al.*, 2003). These surveys produced important sighting data to study the abundance and abundance trends of large whales in different IWC Management Areas. The Japanese whale research programs in the Antarctic (JARPA/JARPAII) were the only comprehensive long-term systematic surveys that collected biological information from whales and data from its environment, using lethal and non-lethal approaches. The comprehensive biological and environmental data (including abundance trends for several whale species) collected by these programs for around 27 years have been very important to understand the

dynamics and interaction of whale species in the ecosystem, as recognized by the IWC SC review of those programs (IWC, 2008; IWC, 2014a).

This scientific endeavor needs to be consistent with existing legal frameworks. The ICRW, in particular, provides a fundamental ground for such research program on whales in the Antarctic. As discussed below, this research plan is developed in accordance with the object and purpose as well as provisions and other legal requirements of the ICRW including paragraph 10 (e) of its Schedule which established the so called commercial whaling moratorium.

The main objectives of this research plan are twofold. The first main objective is "improvement in the precision of biological and ecological information for the application of the Revised Management Procedure (RMP) to the Antarctic minke whales" (see Section 2.1). This is aimed at contributing to the consideration and work of the IWC SC in improving the IWC's RMP that is the single-species management procedure adopted by consensus of its members in 1994 to calculate a catch limit of baleen whales for commercial whaling. The IWC SC continues to work on several issues on how the RMP could be improved and how it could be implemented. The program under this proposed research plan will collect and estimate the parameters required for application of the RMP to the Antarctic minke whale. It will also contribute to the improvement of the RMP itself.

The second main objective is "investigation of the structure and dynamics of the Antarctic marine ecosystem through building ecosystem models" (see Section 2.2). Previous research programs have found that distributions and abundances of Antarctic minke, humpback, and fin whales, which are important parts of the Antarctic marine ecosystem, have been changing appreciably and that there could be competitive relations among those whale species in terms of food (Matsuoka *et al.*, 2011; Matsuoka and Hakamada, 2014; Hakamada *et al.*, 2013; Murase *et al.*, 2014). It can be hypothesized that this phenomenon was caused by increased pressure on Antarctic minke whales from the recovering populations of humpback and fin whales that had been overexploited up to the 1960's, and resultant stagnation/decrease of the Antarctic minke whale populations that had experienced a drastic increase supported by improved food availability as a result of the overexploitation of larger whales. They may also be caused by fluctuations in the abundance of krill populations that are the common food resource for the three species of whales or possibly as a result of changes in the ocean environment associated with climate change. Investigation of the causes and current dynamics is essential for the future conservation and management of the whale resources as well as for the understanding of the Antarctic marine ecosystem.

These objectives come within the research categories identified by the IWC SC in its Annex P (IWC, 2013b) (see Section 2.3).

Achieving a sustainable balance between the maintenance of the unique marine ecosystem and the utilization of its abundant resources is, as is common for all seas and oceans, an important challenge for the Antarctic Ocean. This is also consistent with the objectives of the ICRW stipulated in its preamble; "to provide for the proper conservation of whale stocks and thus make possible the orderly development of the whaling industry".

1.2 The International Court of Justice (ICJ) Judgment

On 31 March 2014, the ICJ issued its Judgment in the case concerning *Whaling in the Antarctic* (Australia v. Japan: New Zealand intervening) (for the Judgment, dissenting opinions, separate opinions and declarations, see http://www.icj-cij.org/docket/files/148/18136.pdf).

Although one needs to read through the entire Judgment in order to understand the context and precise meaning of the Judgment as a whole, salient parts of what was stated in the Judgment are introduced in this section

In its operative paragraph, the Court found that "the special permits granted by Japan in connection with JARPA II do not fall within the provisions of Article VIII, paragraph 1, of the International Convention for the Regulation of Whaling" (paragraph 247 (2) of the Judgment. Hereafter, "paragraph ..." refers to

the paragraph of the ICJ Judgment mentioned above), and decided that "Japan shall revoke any extant authorization, permit or license granted in relation to JARPAII, and refrain from granting any further permits in pursuance of that programme." (paragraph 247 (7)). It should also be noted that the Court found that "Japan has complied with its obligations under paragraph 30 of the Schedule to the International Convention for the Regulation of Whaling with regard to JARPA II." (paragraph 247 (6)).

The Judgment, in its reasoning part, stated that "it is to be expected that Japan will take account of the reasoning and conclusions contained in this Judgment as it evaluates the possibility of granting any future permits under Article VIII, paragraph 1, of the Convention" (paragraph 246). Although this statement was not made in the operative part of the Judgment, Japan takes account of "the reasoning and conclusions contained in this Judgment", key elements of which are as follows.

With respect to the function of Article VIII of the ICRW, the ICJ found that "since Article VIII, paragraph 1, specifies that 'the killing, taking, and treating of whales in accordance with the provisions of this Article shall be exempt from the operation of this Convention', whaling conducted under a special permit which meets the conditions of Article VIII is not subject to the obligations under the Schedule concerning the moratorium on the catching of whales for commercial purposes, the prohibition of commercial whaling in the Southern Ocean Sanctuary and the moratorium relating to factory ships" (paragraph 55).

The Court continued that, concerning the relationship between Article VIII and the object and purpose of the Convention, "the preamble of the ICRW indicates that the Convention pursues the purpose of ensuring the conservation of all species of whales while allowing for their sustainable exploitation" and thus "amendments to the Schedule and recommendations by the IWC may put an emphasis on one or the other objective pursued by the Convention, but cannot alter its object and purpose" (paragraph 56). It then observed that "neither a restrictive nor an expansive interpretation of Article VIII is justified" and noted that "the current Guidelines, Annex P, list three broad categories of objectives. Besides programs aimed at 'improv[ing] the conservation and management of whale stocks', they envisage programs which have as an objective to 'improve the conservation and management of other living marine resources or the ecosystem of which the whale stocks are an integral part' and those directed at 'test[ing] hypotheses not directly related to the management of living marine resources'" (paragraph 58).

Based upon these considerations, the Court considered, with regard to the issuance of special permits, that "Article VIII gives discretion to a State party to the ICRW to reject the request for a special permit or to specify the conditions under which a permit will be granted" but "whether the killing, taking and treating of whales pursuant to a requested special permit is for purposes of scientific research cannot depend simply on that State's perception" (paragraph 61).

This led the Court to establish a standard of review; firstly "whether the programme under which these activities occur involves scientific research" and secondly "if the killing, taking and treating of whales is 'for purposes of' scientific research by examining whether, in the use of lethal methods, the programme's design and implementation are reasonable in relation to achieving its stated objectives" (paragraph 67). For assessing a program against this standard of review, the ICJ stated that "the Court will look to the authorizing State, which has granted special permits, to explain the objective basis for its determination" (paragraph 68).

As for the first limb of the standard of review, the Court observed that "the JARPA II Research Plan describes areas of inquiry that correspond to four research objectives and presents a programme of activities that involves the systematic collection and analysis of data by scientific personnel. The research objectives come within the research categories identified by the Scientific Committee in Annexes Y and P (see paragraph 58 above). Based on the information before it, the Court thus finds that the JARPA II activities involving the lethal sampling of whales can broadly be characterized as "scientific research" (paragraph 127). The Court in this context noted that, concerning the avoidance of adverse effects on stock, "in the particular context of JARPA II [....] Australia does not maintain that meeting the target sample sizes would have an adverse effect on the relevant stocks" (paragraph 85).

The second limb of the standard of review concerns "the programme's design and implementation" and its relevant elements are set forth in the judgment. The Court firstly confirmed its views that "the stated research objectives of a programme are the foundation of a programme's design" while noting that it is not for the Court "to decide whether the design and implementation of a programme are the best possible means of achieving its stated objectives". It then found that "in order to ascertain whether a programme's use of lethal methods is for purposes of scientific research, the Court will consider whether the elements of a programme's design and implementation are reasonable in relation to its stated scientific objectives", suggesting that "such elements may include: decisions regarding the use of lethal methods [Element 1]; the scale of the programme's use of lethal sampling [Element 2]; the methodology used to select sample sizes [Element 3]; a comparison of the target sample sizes and the actual take [Element 4]; the time frame associated with a programme [Element 5]; the programme's scientific output [Element 6]; and the degree to which a programme co-ordinates its activities with related research projects [Element 7]" (paragraph 88, bracketed parts are added for the reference in the present plan). The Court then provided its analysis on these seven elements (for the detailed discussions, see Section 1.3).

In its conclusion of assessments as regards the second limb of the standard of review, the Court firstly found that "the use of lethal sampling per se is not unreasonable in relation to the research objectives of JARPA II". However, with respect to the design of JARPA II, the Court concluded that "the target sample sizes in JARPA II are not reasonable in relation to achieving the programme's objectives" (paragraph 224). Four problems were specifically identified: The first three problems relate to the selection of sample sizes relating to the Elements 2 and 3 above. The Court said: firstly "the evidence does not reveal how those differences between the broad objectives of JARPA and JARPA II lead to the considerable increase in the scale of lethal sampling in the JARPA II Research Plan"; secondly "the sample sizes for fin and humpback whales are too small" and "the programme's design appears to prevent random sampling of fin whales"; and thirdly the process used to determine the sample size for minke whales lacks transparency." The fourth problem relates to the use of lethal methods relating to the Element 1 above. The Court said, "little attention was given to the possibility of using non-lethal research methods more extensively to achieve the JARPA II objectives and [...] funding considerations, rather than strictly scientific criteria, played a role in the programme's design" (paragraph 225). With regard to the implementation of JARPA II, the Court found, in line with the Element 4 mentioned above, that: firstly "no humpback whales have been taken"; secondly "the take of fin whales is only a small fraction of the number that the JARPA II Research Plan prescribes"; and thirdly "the actual take of minke whales has also been far lower than the annual target sample size in all but one season. Despite these gaps between the Research Plan and the programme's implementation, Japan has maintained its reliance on the JARPA II research objectives". The Court also noted, in line with the Elements 5, 6 and 7 above, "other aspects of JARPA II also cast doubt on its characterization as a programme for purposes of scientific research, such as its open-ended time frame, its limited scientific output to date, and the absence of significant cooperation between JARPA II and other related research projects" (paragraph 226). The Court thus concluded that "the special permits granted by Japan for the killing, taking and treating of whales in connection with JARPA II are not "for purpose of scientific research" pursuant to Article VIII, paragraph 1, of the Convention" (paragraph 227).

1.3 Japan's consideration of the reasoning and conclusions in the ICJ Judgment

As mentioned above, the Judgment, in its reasoning part, stated that "it is to be expected that Japan will take account of the reasoning and conclusions contained in this Judgment as it evaluates the possibility of granting any future permits under Article VIII, paragraph 1, of the Convention" (paragraph 246). The Court maintains that, in the use of lethal methods, the programme's design and implementation need to be reasonable in relation to achieving its stated objectives (paragraph 67). The Court thus provides meaningful guidance on the issuance of future special permits by Japan. The Court also states that it will look to the authorizing State to explain the objective basis of its determination to grant special permits

(paragraph 68). In this research plan, which duly takes account of "the reasoning and conclusions contained in [the] judgment", Japan explains the objective basis of its determination to grant special permits.

Japan has in particular taken seriously the Court's finding that the decision to grant special permits under Article VIII, paragraph 1, of the ICRW, "cannot depend simply on that State's perception" (paragraph 61). It has therefore announced its determination to "follow an internationally open and highly transparent process through securing the participation of renowned scientists from Japan and abroad, and through other processes including discussions at the IWC SC's workshop and coordination with other institutions conducting relevant studies" (statement issued by the Minister of Agriculture, Forestry and Fisheries, on April 18, 2014). Japan always welcomes comments from outside that are based upon scientific consideration to which it will give due regard even after the research plan is finalized. The process for development of the original proposal of the research plan, which was placed under the review by the NEWREP-A Review Panel, is outlined in Appendix 1.

The question "whether the programme under which these activities occur involves scientific research" (the first limb of the standard of review adopted by the Court) is examined in more detail in relevant parts in Sections 2, 3 and 6 respectively.

In the following part, it is explained that "in the use of lethal methods, the programme's design and implementation are reasonable in relation to achieving its stated objectives" (the second limb of the standard of review), through the examination of each of the aforementioned seven elements identified by the Court.

Element 1: "decisions regarding the use of lethal methods"

- The Court noted, in respect of "decisions regarding the use of lethal methods", that Japan and Australia "agree that non-lethal methods are not a feasible means to examine internal organs and stomach contents" (paragraph 133) and took into account "the evidence indicating that non-lethal alternatives are not feasible, at least for the collection of certain data" (paragraph 135). However, the Court observed that "the JARPA II Research Plan should have included some analysis of the feasibility of non-lethal methods as a means of reducing the planned scale of lethal sampling in the new programme" (paragraph 137). In the course of its analysis, it paid a close attention to Japan's response to a question from a member of the Court on "what analysis it had conducted of the feasibility of nonlethal methods prior to setting the sample sizes for each year of JARPA II, and what bearing, if any, such analysis had had on the target sample sizes" (paragraph 138). The Court then concluded that "the papers to which Japan directed it reveal little analysis of the feasibility of using non-lethal methods to achieve the JARPA II research objectives. Nor do they point to consideration of the possibility of making more extensive use of non-lethal methods in order to reduce or eliminate the need for lethal sampling, either when JARPA II was proposed or in subsequent years" (paragraph 144).
- Japan continues to uphold its scientific policy with regard to whales that it does not use lethal means more than it considers necessary. Taking account of the Court's above assessment, this research plan explains how the feasibility of non-lethal methods as a means of reducing the planned scale of lethal sampling is carefully reviewed (see Section 3). It has been concluded, in particular, in this plan that there is no non-lethal methods for obtaining age-data required for achieving Main Objective I. This plan also explains a variety of non-lethal methods to be tested, including satellite tagging, biopsy sampling and their associated analytical methodologies, in order to further study their feasibility and practicability (see Section 4). It should be noted here that the proposed plan specifically includes the following:
 - Investigating the feasibility of age-determining methods other than earplug reading (i.e. DNA methylation analysis);

- Investigating the feasibility of biopsy sampling to collect skin and blubber samples necessary for analysis of stock structure and other potential research items of Antarctic minke whales;
- Conducting satellite tagging on Antarctic minke whales to elucidate the location of their breeding grounds, in preparation for the collection of genetic samples from the breeding grounds;
- Investigating the feasibility of data-logger use for the research on feeding behavior of Antarctic minke whales;
- Investigating the feasibility of chemical markers as potential body condition indicator by using biopsy samples (biomarker).

<u>Elements 2 and 3: "the scale of the programme's use of lethal sampling" and "the methodology used to select sample sizes"</u>

- Element 2, namely, "the scale of the programme's use of lethal sampling" and Element 3, namely, "the methodology used to select sample sizes" are both related to the selection of sample sizes. The Court noted that "the evidence relating to the minke whale sample size, like the evidence for the fin and humpback whale sample sizes, provides scant analysis and justification for the underlying decisions that generate the overall sample size. For the Court, this raises further concerns about whether the design of JARPA II is reasonable in relation to achieving its stated objectives" (paragraph 198). It should also be noted, as mentioned above, that Japan and Australia "agree that non-lethal methods are not a feasible means to examine internal organs and stomach contents. The Court therefore considered that the evidence shows that, at least for some of the data sought by JARPA II researchers, non-lethal methods are not feasible." (paragraph 133).
- As noted above, age-data is indispensable for achieving Main Objective I of this research plan (see Section 3). This is because it forms an essential input for detecting changes in biological parameters such as age-at-sexual-maturity (ASM) and recruitment rates that are necessary for this objective. The ASM makes considerable contribution to achieving Main Objective I in the sense that it helps improve the Statistical Catch-at-Age Analysis (SCAA) performance for estimating other key parameters such as MSYR. This leads to the better assessment and management of the Antarctic minke whales. It therefore constitutes the scientific ground for the sample size calculation under this research plan. In the initial stage of developing the research plan, availability, feasibility and practicability of non-lethal research methods for obtaining age-data of the Antarctic minke whales were carefully considered. However, it has been concluded that in order to obtain age-data at the annual scale, the collection of earplugs of whales is indispensable and the sole practicable mean available at the present time. This can only be achieved by lethal sampling (the calculation of sample size to be explained in Section 3.3.1.)
- Under Main Objective II, stomach contents are required for assessing prey consumption that is necessary for investigating the structure and dynamics of the Antarctic marine ecosystem (see Section 2.2). This also requires lethal methods as explicitly found in the ICJ Judgment as mentioned above.
- Multiple approaches have been applied in order to calculate the sample size for this research program including extensive simulations using ecosystem models. As a result, the final sample size was calculated based on age-data necessary for estimating ASM which is one of the main interests under the Main Objective I. However, it has been found that the approaches attempted under the Main Objective II have not provided basis for identifying specific sample sizes (see Section 3.3).
- This proposed sample size is the best possible estimation at the present level of scientific knowledge. As the research progresses and new information becomes available in the coming years, there will be more knowledge that could improve the calculation of a sample size. Under this research plan, revised sample sizes could be proposed, for the

consultation with the IWC SC, at the mid-term review after the first six year period and/or other occasions as it becomes necessary.

Element 4: "a comparison of the target sample sizes and the actual take"

- Concerning "a comparison of the target sample sizes and the actual take", the Court noted that "Japan's continued reliance on the first two JARPA II objectives to justify the target sample sizes, despite the discrepancy between the actual take and those targets, coupled with its statement that JARPA II can obtain meaningful scientific results based on the far more limited actual take, cast further doubt on the characterization of JARPA II as a programme for purposes of scientific research. This evidence suggests that the target sample sizes are larger than are reasonable in relation to achieving JARPA II's stated objectives" (paragraph 212).
- As research activities could be disrupted by both natural and human factors including dangerous sabotage activities by an extreme anti-whaling NGO, this research plan establishes a contingency backup plan in order to secure the scientific value of data for the purpose of achieving the established scientific objectives. This back-up plan responds to the recommendation made at the Expert Workshop to Review the Japanese JARPA II Special Permit Research Programme, held in Tokyo, Japan, February 24-28, 2014 (JARPA II review workshop) (see section 12.1.2 in IWC, 2014a).
- Issues arising from possible disruptions could include: reduced sample size, unsurveyed research areas, missed research periods and sampling bias. The contingency back-up plan addresses three aspects; (i) adjustments of research protocols at the scene of disruption, (ii) adjustment of proposed research plans including research period, sample size, and research areas, and (iii) consideration of analysis methods to compensate the effects of disruptions as much as possible. This ensures pre-planned response based on scientific consideration to such disruptions, addressing concerns expressed in the Judgment.

Element 5: "the time frame associated with a programme"

- The Court observed, in relation to "the time frame associated with a programme", that "with regard to a programme for purposes of scientific research, as Annex P indicates, a "time frame with intermediary targets" would have been more appropriate" (paragraph 216).
- This research plan has set its research period as 12 years. It has also established "intermediary targets", as suggested by the Judgment, specifying concrete target outputs of the first 6 years, together with a system of mid-term review. An assessment of the plan, including on the feasibility of non-lethal methods will be made on a yearly basis. These time frames are determined in view of the prospect of scientific works and practical factors including the natural environment in the Antarctic Ocean, required time for verifications and analyses of collected data and information, the capacity of the research and financial constraints (See Sections 3.1.2 and 6).

Element 6: "the programme's scientific output"

- In the context of "the programme's scientific output", the Court found that "in light of the fact that JARPA II has been going on since 2005 and has involved the killing of about 3,600 minke whales, the scientific output to date appears limited" (paragraph 219).
- This research plan is primarily intended to contribute to the improvement of conservation and management of whale resources in the Antarctic in accordance with the ICRW as shown in its research objectives. It follows that its scientific output will be produced first and primarily for the review and discussion in the IWC SC. At the same time, in line with an observation by the Court, there will be increased efforts to publish its scientific achievements in peer-reviewed journals outside the IWC (see Section 8).
- It should also be noted that scientific data generated from this research will be made available to other scientists both inside and outside the IWC SC in accordance with the IWC Data Availability Agreement (IWC, 2004) and the Institute of Cetacean Research

(ICR)'s protocol for access to samples/data. Data availability arrangement is also specified in this plan (see Section 8) in order to ensure that the research results will be utilized broadly by scientists worldwide.

Element 7: "the degree to which a programme co-ordinates its activities with related research projects"

- Regarding "the degree to which a programme co-ordinates its activities with related research projects", the Court observed that "some further evidence of co-operation between JARPA II and other domestic and international research institutions could have been expected in light of the programme's focus on the Antarctic ecosystem and environmental changes in the region" (paragraph 222).
- Scientists of the ICR, which is expected to conduct the research program under this plan, have already started to strengthen existing collaboration with relevant research institutions that have direct interest in the Antarctic marine ecosystem such as Commission for the Conservation of Antarctic Marine Living Resources (CCAMLR), Japan's National Institute of Polar Research and Japan's National Research Institute of Far Seas Fisheries, among others. It should also be noted that deepening co-operation/co-ordination with these institutions will be sought in planning and implementing the krill research and analysis and utilization of data derived from it, including abundance estimate. For example, scientific outputs of this research plan will be shared with those research institutions and joint research will be proposed to relevant research institutions, including CCAMLR scientists. Section 8 will deal with specific discussions on this element.

As the ICJ found that "Japan has complied with its obligations under paragraph 30 of the Schedule" to the ICRW (paragraph 247(6)), it will continue to abide by this provision. This plan specifies, as required, (a) objectives of the research (see Section 2), (b) number of the whales to be taken (see Section 3.3), (c) opportunities for participation in the research by scientists of other nations (see Section 8) and (d) possible effect on conservation of stock (see Section 5) in the following sections. Sex, size and stock of the whale to be taken are not specified as this program adopts random sampling procedure based upon scientific consideration (see Section 3.2.2). Comments and suggestions offered during the review process of the original proposal of the research plan by the IWC SC and the NEWREP-A Review Panel were taken into account in finalizing this plan. Finally, whales taken under this program will so far as practicable be processed and the proceeds will be dealt with in accordance with the direction issued by the government of Japan in conformity with Article VIII, paragraph II, of the ICRW.

It is important to reiterate that the Court confirmed that the objects and purposes of the ICRW include sustainable exploitation of whale stocks and that special permit whaling under Article VIII, paragraph 1, of the ICRW is legally permissible under certain conditions. Japan has therefore announced that it confirmed its basic policy of pursuing the resumption of commercial whaling, and collecting and analyzing necessary data through special permit whaling for this purpose, in full accordance with legal requirements including the ICRW, its paragraph 10 (e) of the Schedule which establishes the moratorium on commercial whaling, as well as in light of the ICJ Judgment. This plan explains that the program meets expectations of the Court and that Japan is thus ready to conduct its special permit whaling in the Antarctic for purposes of scientific research under Article VIII in full compliance with international law, and gives due consideration to the comments from the IWC SC in accordance with paragraph 30 of the Schedule and Annex P, comments outside the IWC SC, and the recommendations and suggestions by the NEWREP-A Review Panel.

1.4 Overview of JARPA/JARPAII outcomes

JARPA was started in the 1987/88 and 1988/89 austral summer seasons as a two-year feasibility study. The full program started in 1989/90 and was completed in 2004/05. The JARPA II started in the 2005/06

and 2006/07 as a two-year feasibility survey. The full program started in 2007/08 and the first six-year period was completed in 2010/11. The JARPA II was cancelled after the 2013/14 season in compliance with the ICJ Judgment. The objectives of JARPA and JARPA II as well as details of the scientific outputs from the research were summarized in Pastene *et al.* (2014a).

Data and results from the JARPA were reviewed in three workshops: an IWC intersessional working group meeting held in May 1997 (IWC JARPA mid-term review) (IWC, 1998a); a non-IWC JARPA review workshop called by the Government of Japan in January 2005 (Japan JARPA review) (Anon, 2005); and the final IWC intersessional workshop held in December 2006 (IWC JARPA final review) (IWC, 2008).

Data and results of JARPA II obtained in the first period (2005/06-2010/11), being combined with JARPA data, were reviewed under the IWC SC during a specialist workshop held in February 2014 (IWC, 2014a). The workshop welcomed the scientific contribution of JARPA/JARPAII. At the same time it identified those areas where further work was required and provided useful suggestions and recommendations. Several of those suggestions and recommendations are also relevant for some of the analyses proposed in this research plan, and these were taken into consideration to improve the research program proposed.

Below are some key scientific outcomes of JARPA and JARPAII:

JARPA (Pastene et al., 2014a; IWC, 2008)

- Two biological stocks of the Antarctic minke whale (I and P stocks) distribute in the JARPA research area.
- Several biological parameters related to growth were estimated by biological stock.
- The average annual natural mortality rate and the age-specific natural mortality were estimated by biological stock.
- Abundance of the Antarctic minke whale remained broadly stable while those of the humpback and fin whales increased appreciably (and statistically significantly) during the JARPA period.
- Changes in whale species composition and distribution were observed in the JARPA research area.

JARPAII (Pastene *et al.*, 2014a; IWC, 2014a)

- Trends of demographic parameters of Antarctic minke whale in 1940s-1970s were consistent with the pattern expected under the krill surplus hypothesis.
- The changes in the Antarctic marine ecosystem suggested under the JARPA research *e.g.* changes in whale species composition and distribution, and possible biological changes in krill predators such as the Antarctic minke whale, were confirmed.
- The stock structure hypothesis of Antarctic minke whale derived from JARPA research was confirmed and refined. There is a soft boundary between stocks, which changes by year and sex.
- Hypotheses on stock structure of humpback, fin and southern right whales in the feeding grounds, were proposed.
- Progress was made toward the development of two types of ecosystem models: multispecies production model and Ecopath with Ecosim. Additional data and analyses were identified to further progress this work.

2. RESEARCH OBJECTIVES

2.1 Main Objective I

Main Objective I

Improvements in the precision of biological and ecological information for the application of the RMP to the Antarctic minke whales.

In order to highlight the significance of the Main Objective I, this section describes i) adoption of commercial whaling moratorium and introduction of the RMP; ii) the basic idea of RMP catch limits for the Antarctic minke whales; and iii) necessity of improvement in the precision of biological and ecological information for the application of the RMP to the Antarctic minke whales.

2.1.1 Adoption of commercial whaling moratorium and introduction of the RMP

Whale stocks in the Antarctic had been heavily utilized since the beginning of 20th Century until the 1980s. There is no doubt that the large baleen whales such as blue, fin and humpback whales were overhunted, which caused sequential depletion of those whale stocks in the Antarctic (Allen, 1980).

As a temporary measure responding to the situation, "the Commercial Whaling Moratorium" was established by adopting Paragraph 10(e) of the Schedule to the ICRW in 1982. Its text reads: "Notwithstanding the other provisions of paragraph 10, catch limits for the killing for commercial purposes of whales from all stocks for the 1986 coastal and the 1985/86 pelagic seasons and thereafter shall be zero. This provision will be kept under review, based upon the best scientific advice, and by 1990 at the latest the Commission will undertake a comprehensive assessment of the effects of this decision on whale stocks and consider modification of this provision and the establishment of other catch limits (underline added)."

During discussions at the IWC that resulted in the establishment of the Commercial Whaling Moratorium, it was argued that the scientific information for the management of whales was uncertain and therefore scientific knowledge should be accumulated while suspending all commercial whaling. This is specifically reflected in the text of Schedule 10(e) as underlined above.

With this awareness, the RMP was finalized in 1992 by the IWC SC for the purpose of undertaking "a comprehensive assessment" of the effects of the IWC's decision on whale stocks (reflected by the population models underlying the RMP *ISTs*), and of establishing non-zero catch limits, as stipulated in paragraph 10(e) (IWC, 1986, 1987 and 1988).

The RMP is a risk-averse, powerful and important management tool developed by the IWC SC. The full RMP process is composed of three steps: i) *pre-implementation assessment*, ii) implementation process including the *Implementation Simulation Trials (ISTs)*, and iii) application of the Catch Limit Algorithm (CLA) based on the output from steps i) and ii). The implementation process (ii) above takes two years to be completed. Each of the steps requires data identified in the Requirements and Guidelines for Implementations under the Revised Management Procedure (IWC, 2012a).

2.1.2 The RMP and catch limits for the Antarctic minke whales

The basic idea behind the RMP is as follows:

- 1) The RMP is to calculate a catch limit for a target area assumed to consist of a single stock, based on a specified algorithm, the CLA. The RMP has a built-in safety threshold (i.e. zero catch if the population size is estimated below 54% of carrying capacity). The RMP employs a simple surplus production model and the CLA requires only time series of past catches and available abundance estimates (with their associated CVs) with several tuning levels (60-72% of target depletion level of population size after 100 years implementation for a specified trial with the lowest Maximum Sustainable Yield Rate (MSYR) value) to calculate catch limits. The calculation requires regular availability of abundance estimates with reasonable precision. According to the definition of the target area and spatial stock structures, several harvesting scenarios in terms of areas and seasons (i.e. variants) for calculation of catch limits are considered.
- 2) Apart from the simple model above, a wide variety of realistic models ("trials", not necessarily the same as the model used in the CLA but rather complicated because of model uncertainty concerning

the population dynamics itself and population structure) are proposed in order to replicate future population dynamics based on supposed implementation of the CLA above and consideration of uncertainty in dynamics, estimation and so on. Based on this form of comprehensive simulation testing, only variants of harvest rules with acceptable levels of risks are selected for calculating the actual catch limits (IWC, 2012a).

As mentioned above, the RMP *ISTs* require several types of information for the management of whales, which include

- time series of past catches by area
- time series of abundance estimates (and their CVs) by area
- hypotheses of stock structure (for trial structures, definition of target areas, variants of the CLA)
- biological information on natural mortality, production rate, maturity and recruitment relationship etc. (for trial structures)
- stochastic variation of migration, mixing etc. (for trial structures)

Based on the historical and legal background mentioned above and also based on the fact that the IWC SC has been engaged in improving the basis for the *Implementation* of the RMP, the calculation of catch limits of the Antarctic minke whales through the application of the RMP is consistent with the provision of Schedule 10(e) and the work of the IWC SC (Morishita, 2013). The specific idea of the RMP and the flow of the full process of the RMP *Implementation* is described below.

The IWC SC carried out in 1993 a trial application of the RMP to Antarctic minke whales and calculated a range of circumpolar annual catch limits of approximately 2,049 to 4,490 (IWC, 1994). However, this trial calculation can be characterized as being based on excessively conservative assumptions in view of the limited knowledge of the Antarctic minke whale stock structure at that time; for example, the longitudinal width of *Small Areas* (reflecting possible separate stocks) was set as 10 degrees and the lower end of the MSYR (for the mature component of the population) was 1%. In recent years new information became available that could improve the implementation of the RMP on the Antarctic minke whale (see below).

In this context, it should be noted that the most conservative whaling scenarios are selected as a precautionary approach, when there is uncertainty in the biological and ecological information required in the RMP *ISTs* such as stock structure and reproductive parameters (uncertainty is related to the limited amount of data, which in turn implies less precision in the parameters). In other words, further biological and ecological information will contribute to reducing uncertainty, and therefore less conservative whaling scenarios can be used, which usually provide larger catch limits. In this way, additional data to be obtained under this new research program could be used to increase the allowable catch limits of Antarctic minke whales without increasing the depletion risk. This will be in line with one of the IWC's management objectives for commercial whaling under the RMP, which is "making possible the highest possible yield from the stock" (IWC, 1990).

IWC Resolution 1997-5 "INSTRUCTS the Scientific Committee not to consider Southern Hemisphere minke whales in the context of implementation of the RMP unless advised to do so by the Commission" (IWC, 1998b). From this perspective, it should be noted that this research plan does not request the IWC SC to conduct the *implementation* of the RMP on Antarctic minke whales. Furthermore, the general RMP issues of improving the SCAA analysis and investigating means to incorporate ecosystem effects into the RMP have been recognized as high priority issues by the IWC SC (see for example section 13.2 in IWC, 2014e) and this new research program is expected to contribute to the IWC SC discussions.

Since the adoption of the RMP, the RMP itself has served as a good example for recent management procedures developed for other domestic/international fishery management organizations. Such a management scheme with comprehensive simulation testing to produce objective harvest control rules is now well-known as the framework of "Management Strategy Evaluation (MSE)", which makes it clear that the RMP is a pioneer of modern fishery resource management.

2.1.3 Improvements in the precision of biological and ecological information for the application of the RMP to the Antarctic minke whales

The IWC SC has been investigating the population dynamics of the Antarctic minke whales through the SCAA modeling approach. However, the results of SCAA analyses conducted until now have led to the conclusion that issues such as the improvement of the precision of MSYR estimates still require further relevant information and warrants further investigation on the time-varying parameters such as carrying capacity and productivity (IWC, 2014c).

Under this Main Objective I, not only abundance estimates and hypotheses of the stock structure but also certain biological and ecological parameters, including ASM estimated by age and reproductive data, are keys in the specification of trials, and therefore narrowing the reasonable range of their values and hypotheses is an important issue for calculating reasonable catch limits and thus ensuring the sound management of whaling once paragraph 10 (e) of the Schedule is lifted. Using this basic idea, the IWC SC has improved RMP *ISTs* to allow improved selection amongst RMP variants.

Abundance data

Some international and national surveys have been conducted in the Antarctic Ocean for estimating whale population sizes and monitoring their population trends. Particularly, the IDCR and the SOWER surveys under the auspices of the IWC have provided valuable information on the distribution and abundance of large whales at the circumpolar scale. Also, the Japanese surveys in the Antarctic (JARPA/JARPA II) contributed information on finer time-scale trends in abundance because of the availability of abundance estimates virtually every other year. From these surveys, recovery of population sizes for the blue, fin and humpback whales have been confirmed (Branch, 2007; Matsuoka and Hakamada, 2014; Matsuoka *et al.*, 2011). Also, for the Antarctic minke whales, the problem of g(0) attributed to their surfacing/diving behavior was resolved by a new statistical model (OK method, Okamura and Kitakado, 2012), and a set of abundance estimates agreed by the IWC SC was produced (IWC, 2013a). These estimates of g(0) were also used in the JARPA/JARPA II estimates and abundance trends in the study areas reflecting near-stability have been confirmed (Hakamada *et al.*, 2013; Hakamada and Matsuoka, 2014).

Biological and ecological parameters

For the Antarctic minke whales, in addition to the abundance estimates mentioned above, several types of biological data have become available since the commercial whaling era (e.g. Kato, 1982). In particular, the technical procedures for determination of age and maturity status were well-developed and became consistently used over years (Bando et al., 2006; Zenitani et al., 1997). These advances could then give key information for inferring population dynamics of the Antarctic minke whales. The catch-at-age data and knowledge on maturity by age, which usually play a central role in fishery population assessments, are important sources of information for estimating population dynamics of the Antarctic minke whale, including yearly changes in carrying capacity and productivity. The current changes observed in the Antarctic ecosystem warrants continuing research efforts to focus on the changes in the carrying capacity and productivity.

The parameters of age-structured population dynamics models have been estimated using both the abundance estimates and catch-at-age data provided by samples from commercial whaling and the JARPA/JARPAII (Mori and Butterworth, 2006; Punt, 2014a). In the population dynamics model, parameters such as the "natural mortality rate (M)" (whose age-dependence was also determined (Punt, 2014b)) and the "MSYR" are important. It should be noted that the estimate of MSYR is still sensitive to model assumptions and thus can take a wider range of values, including those which are unrealistically high in view of knowledge for other baleen whales. The biological parameters including these which

will be estimated from data collected by this research program will contribute to the trials structure in the RMP *ISTs*.

These biological parameters are estimated using catch-at-age modeling methods. Several of such methods have been already applied to data for Antarctic minke whales to draw inferences regarding trends in abundance for this species. As one of these methods, the SCAA was first presented to the Scientific Committee in 2005 (Punt and Polacheck, 2005). It has been modified over time to more adequately mimic the available data on length, conditional age-at-length and indices of abundance from IDCR/SOWER and JARPA/JARPAII. A summary of the history of the application of the SCAA to Antarctic minke whales was presented by Punt (2014a). The SCAA model is recognized as the "best currently available model for examining stock dynamics for Antarctic minke whales" (IWC, 2014a).

Typical outputs from the SCAA analysis are the historical trends in abundance (total abundance and recruitment), MSYR, natural mortality (M), recruitment rates and growth curves. The 2013 IWC SC meeting considered that some of the conclusions regarding the application of the SCAA to Antarctic minke whales appear to be supported "quite robustly" (IWC, 2014c). This includes the result that relative trends in past abundance are generally estimated consistently (IWC 2014c). The SCAA is designed to detect, among others, changes in recruitment rates and to provide better estimation of these changes, which have important contribution to RMP trial specifications for the species as a time-varying parameter. Antarctic minke whales have exhibited growth rates well in excess of 1% per year (Punt, 2014b). Using such updated parameters, sustainable catch levels are considered to be much greater than outputs from the current RMP would provide. This leads to a reasonable scientific assessment that an improved RMP could be developed for Antarctic minke whales that would allow appreciably greater catches without any risk to the population. This is in line with both the management objectives agreed by the Commission for the RMP and the objectives and purposes of the Convention itself.

There are, however, certain difficulties associated with these important parameters such as MSYR. In fact, although MSYR can be estimated by the SCAA, its output still has considerable level of uncertainty. One of the possible approaches to overcome this challenge is the use of ASM. The ASM is influential to the numbers of mature animals, especially females, and possible future changes in the ASM affect the carrying capacity and the productivity. In addition, the ASM is of great importance for contributing to information on the proportion of matured Antarctic minke whales that is used for SCAA. This could lead to the improvement of the SCAA performance for estimating aforementioned key parameters such as MSYR. The mean ASM can be derived directly from an analysis of the proportion of mature animals at each age in the catch. Usually the age at which 50% of the animals are sexually mature is taken as the average at first sexual maturity. This is identified through direct observation of reproductive organs. There are also indirect methods for estimating ASM. As noted in the relevant literature, "[i]ndirect methods of estimations of mean age at sexual maturation (tm) include estimating tm from mean length at age curves when mean body length at first sexual maturity is known, calculating the age at first ovulation from regression of age on corpus number, and analysis of the earplug transition phase" (Lockyer, 1984a). This research plan will use the former method, namely direct observation of reproductive organs. Samples will be arranged by cohorts (i.e. years in which the whales born). As noted above, the ASM is crucial to define the mature component in the population, which can contribute to its reproduction, and thus has an implication to the recruitment process and will contribute to achieving Main Objective I of the research plan. This data item can also be used as an indicator of prey availability over time, since less availability of prey would cause delayed sexual maturity. In this regard, the ASM is useful for understanding inter-species competition among baleen whales over krill in the Antarctic Ocean and it is also relevant to the Main Objective II of the research plan.

Stock structure

When it comes to the stock structure, Pastene (2006) showed that there are two genetically different stocks (so-called I-stock and P-stock named after "Eastern Indian Ocean stock" and "Western South Pacific Ocean stock", respectively) and these two stocks are mixed in a transition area. This inference was confirmed by integration of various sources of information provided by the JARPA/JARPA II

programs such as different genetic markers, morphometrics and other biological traits. In addition, a recent study using microsatellite loci and morphometric measurements suggests that the spatial distribution of the two stocks has a soft boundary in the main JARPA/JARPAII study area (IWC Management Areas IV and V) and the mixing pattern depends on year and sex. However, the JARPAII review workshop noted that another interpretation of the data is possible instead of variable mixing of two stocks over years, and furthermore the certainty associated with these conclusions becomes weaker for locations outside the main research area (IWC, 2014a). Therefore it is considered that further analyses with the existing data in the current study area and additional data from the outside of the main area are worth conducting. These analyses may provide extensive information to specify valid population structure hypotheses in more detail, and these would also provide a basis for improvement of the SCAA, trial specifications and application of the CLA.

2.1.4 Conclusion

As described above, the RMP is a management procedure which guarantees sustainability with safe levels of catch limits over a period of 100 years (IWC, 2012a). While considerable data and information are already available through past research, more refined information over a longer period is necessary for improvement of the RMP *Implementation* (see Table 1).

Therefore, the Main Objective I aims at providing required data for application of the *ISTs* and improving the assessment process under the RMP *Implementation*. Furthermore, the actual catch limits which may be used for the lifting of the Moratorium, will be derived from the achievement of this research objective. The specific data and samples required in this proposed program for the Main Objective I are amplified in the subsequent section (Section 2.4).

Table 1. Existing information and problems to be addressed relevant to RMP *Implementation* for the Antarctic minke whales.

	Existing values and knowledge	Problems/pending issues	Aspects to potentially be cleared with past data and new data under the proposed research plan
1) Abundance	IDCR/SOWER (OK estimates) JARPA/JARPAII	IDCR/SOWER estimates become subject to the "phase-out rule" · usable for conditioning purpose · usable for CLA in trials · no estimates agreed for use in the actual CLA as yet	Obtain new series of abundance estimates from surveys with IWC oversight with g(0) correction Application of the g(0) estimate for correcting JARPA/JARPAII Repeated calculation of abundance estimates for study areas will be helpful to draw information on inter-annual variation of distribution (better estimate of additional variance and inference of factor affecting the variation) and to improve

			adjustments made for g(0) (Objective I(i))
2) Biological and ecological parameters	M: Age-dependent values were estimated by the SCAA MSYR(1+): 1 ~ 4% (revised range given by meta-analysis) ASM: Time-invariant maturity (age at 50% maturity was assumed to be 8.5)	 How the time-varying parameters such as K and productivity will change in the future is unknown The estimate of MSYR(1+) by the SCAA is sensitive to the model configuration ASM seems to be slightly increasing 	 Improvement of precision of biological/ecological information Further monitoring of the changes in the carrying capacity and productivity Try to narrow the range of MSYR with longer series of catch-at-age data, abundance estimates Detection of changes in ASM over years (Objective I(ii))
3) Stock structure	A hard boundary with a process error is assumed in statistical catch-at- age analysis.	Soft boundaries with annual variation and single stock with isolation by distance constitute another possible assumption	Further investigation with additional data in previously uncovered areas and additional repeated data in the main study areas (Objective I(iii))
4) Specification of RMP/IST	Possible to use existing outputs from Punt's SCAA	 Need to define future changes in carrying capacity No specific species interaction, which likely occur 	Trials structure can be considered with changes in carrying capacity etc. and linkage between recruitment and environmental changes Possible contribution from ecosystem modeling (in Main Objective II) (Objective I(iv))

As shown in Table 1, there are several research items necessary to achieve the Main Objective I. For this reason, this proposed plan also identifies four sub-objectives as follows (see also Appendices 3-7):

Objective I(i): Abundance estimates for Antarctic minke whales taking into account of g(0) and additional variance

Objective I(ii): Improvement of precision of biological and ecological parameters

Objective I(iii): Refinement of stock structure hypotheses of Antarctic minke whale in Areas III-VI for the implementation of the RMP

Objective I(iv): Specification of RMP ISTs for the Antarctic minke whales

Main Objective I Application of RMP for the Antarctic minke whales

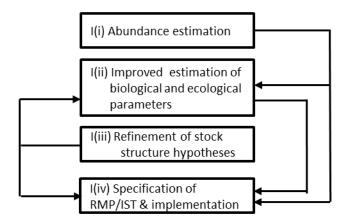


Figure 1. Relationship among the four sub-objectives under the Main Objective I (the directed arrows mean flows of information among them)

2.2 Main Objective II

Main Objective II

The other main research objective is *Investigation of the structure and dynamics of the Antarctic marine ecosystem through building ecosystem models*.

2.2.1 Importance of the investigation of the Antarctic marine ecosystem

JARPA and JARPA II have found, through their lethal and non-lethal research components, the possibility of ongoing and substantial changes in the Antarctic marine ecosystem as reflected by annual shifts in the distribution and migration of the main species of whales (*e.g.* Antarctic minke, humpback, and fin whales) (Murase *et al.*, 2014) and changes in the nutritional condition of minke whales (Konishi *et al.*, 2008; 2014, Konishi and Walløe, 2014). Such findings indicate that whales are crucial species in understanding the changes in the Antarctic marine ecosystem.

One important indicator of shifts in the ecosystem is ASM. In fact, ASM in the Antarctic minke whales changed from 12 years old to 7 years old between 1946 and 1970 and stabilized subsequently until now (Thompson *et al.*, 1999; Zenitani and Kato, 2006). It has been hypothesized that such changes occurred in response to change in nutritional condition, which became better for the Antarctic minke whales after the depletion of large whales such as blue and humpback whales, which fed on the same prey, the Antarctic krill.

Furthermore in view of potential ecosystem functions in the Antarctic marine ecosystem such as climate control and sustainable provisions of useful natural resources, investigation of the structure and dynamics of the Antarctic marine ecosystem through building ecosystem models is thus critically important.

While numerous attempts have been made to build ecosystem models in the Antarctic and other marine areas, it is well recognized that the exercise is not simple and requires substantial investment in terms of time, data, analysis and trials.

2.2.2 Ecosystem modeling

In the Antarctic Ocean, Antarctic krill (*Euphausia superba*) is the main prey species and plays a crucial role in influencing the ecosystem surrounding large baleen whales like blue, fin, humpback and Antarctic minke whales and other krill predators like seals and sea birds. On the other hand, as mentioned earlier, a sequential depletion took place in the 1950s and 1960s amongst the baleen whales in the Antarctic, and the pattern of utilization of krill among the whale species may also have been affected by these occurrences. It is necessary under these circumstances to build models to quantify interactions among predators like whale species for the main prey species, krill, and to examine the dynamics of the Antarctic marine ecosystem. This will enable to conduct the investigation on what has happened in the past and to predict what is going to happen in the Antarctic.

A broad range of approaches have been proposed and attempted for building ecosystem models (e.g. Plagányi, 2007). One of the most comprehensive approaches (or so-called the "whole-of-ecosystem" modelling, e.g. Plagányi et al., 2012) is Ecopath-with-Ecosim (EwE, e.g. Pauly et al., 2000), in which a complicated system is represented by multi-directional linkages among component species with consideration of an initial mass-balancing of the system and its subsequent dynamics. This approach has been widely used for constructing marine ecosystem models, but it also requires various types of information such as the biomass levels, the diet compositions, per capita growth rates and the extent of necessary consumption with consideration of required energy, and these types of information are often influential with regard to the models' results. Furthermore, predicting future population dynamics demands further information and assumptions. Nevertheless, it is still beneficial, recognizing the challenges and limitations, to try to draw a bigger picture of the ecosystem through attempting this approach.

There is another framework of ecosystem modeling. The dynamics of the population size of wildlife species has traditionally been expressed as a form of production model (*e.g.* Butterworth and Rademeyer, 2005; Hilborn, 1990). This contains, of course, the effect of density dependence, which is usually linked with the relative depletion of a population compared to its carrying capacity. The model is then extended to consider prey availability together with competition in utilizing them to inform of the impact of predators on prey species. This can be regarded as a model of intermediate complexity of an ecosystem (*e.g.* Plagányi *et al.*, 2012). Pioneering works were developed by Lotka and Volterra (Lotka, 1910; Volterra, 1931). Each of them derived different sets of equations, which are now called "predator-prey interactions" and "competition for food and space". Since then, the models have been further developed to take into account multiple prey and predator species simultaneously while facing "the curse of dimension", especially in the presence of many species to be considered in, for example, a region of the Antarctic. Although the multiple-predators and multiple-prey system potentially requires a large number of parameters, the Antarctic marine ecosystem is simpler than most of the others, and hence offers a particularly good opportunity to impose limits on the component species and their hierarchy without unduly jeopardizing the realistic consideration of the model.

When considering this simple hierarchical structure, the extended production model might work well without encountering the difficulty of over-parameterization. This proposed program thus re-visits a modeling approach in a preceding study conducted by Mori and Butterworth (2006) and Kitakado *et al.* (2014a). Although the multi-species production model is less complex than the other approaches to ecosystem modeling, it still has many unknown parameters. Some modifications were therefore added into their basic system of equations for multiple predators (some baleen whales and seals) and single prey species (krill) by considering bioenergetic and allometric reasoning (*e.g.* Yodzis and Innes, 1992). The maximum consumption is linked to the body weight of the predators.

This sort of model has the potential to estimate the extent of competition between the baleen whales. For example, since the population sizes of other larger baleen whales such as blue, fin and humpback whales declined through commercial harvesting in the past, there may have been a state of krill surplus in the Antarctic (Laws, 1977; 1985). This surplus could have been taken by the Antarctic minke whales, and since that time this species may have been and may still remain above their pre-exploitation level (Pastene *et al.*, 2014a).

In view of examining multi-species competition among whales, it is important to collect relevant information on humpback and fin whales. This is mainly because it has been observed that they have increasing biomass and are in possible competition with other whale species over space and krills for food in the Antarctic marine ecosystem. Data on distribution and abundance for these species will be obtained by the non-lethal component of the program and this program will also seek to utilize proxies derived from existing scientific knowledge accumulated through JARPA and JARPA II on other species of whales in the Antarctic and the same species in other seas at this stage, pending the result of the midterm review after the first six year-period of the program.

This program therefore attempts to build ecosystem models through analyzing data (data required under the Main Objective II is listed under section 2.4) in the Antarctic Ocean; 1) Modeling spatial interaction among baleen whales, 2) estimation of multi-production models with consideration of the predator-prey system and allometric reasoning.

As the second main objective, this plan proposes the Main Objectives II "Investigation of the structure and dynamics of the Antarctic marine ecosystem through building ecosystem models". Ecosystem modeling was one of the primary objectives in the JARPAII. However, it became evident that the lack of information on krill abundance and its trend caused difficulty in the identification of key parameters in the model. In this new research proposal, the krill survey component is strengthened to obtain that key information. In addition, observations of the body condition of Antarctic minke whales conducted in JARPA and JARPA II are proposed to continue, and improvement of the indicators of body condition and nutrition level through the collection and use of blubber thickness and stomach contents will be attempted so as to detect an effect on the prey consumption by the Antarctic minke whales. It should also be noted that such an effort will further contribute to the better interpretation of ASM data that is an important indicator, as mentioned above, of the Antarctic marine ecosystem. These data are to be integrated for better ecosystem modeling and estimation.

It is important to reiterate that the Main Objective II has also scientific significance regarding the elucidation of the Antarctic marine ecosystem as a whole from mid- and long- term perspective, despite current uncertainties in the models investigated, as recognized among scientists/by IWC SC(IWC, 2014a).

This plan therefore identifies the following sub-objectives to achieve the Main Objective II (see also Appendices 8-12):

Objective II(i): Ecological Research (krill abundance estimation and oceanographic observation)

Objective II(ii):Abundance estimate of some cetacean species as input data for ecosystem modeling

Objective II(iii): Estimation of prey consumption by the Antarctic minke whale and its nutritional condition

Objective II(iv): Ecosystem modeling (Spatial interaction among baleen whales and consideration of predators-prey system and allometric reasoning)As noted by the NEWREP-A Review Panel, the Main Objective II of NEWREP-A "relates to a field in development which makes specifying the overall outcomes in a 12-year program more difficult than for" the Main Objective I (IWC, 2015a pp39). Appendix 13 includes charts showing the structure of the models, basic analyses, data/samples, and the steps required to accomplish the Main Objective II of NEWREP-A. This Appendix is prepared to facilitate understanding of complexity of the structure of the Main Objective II.

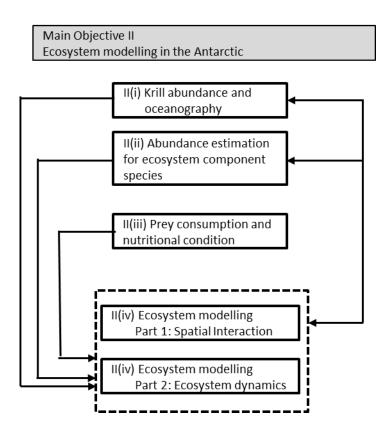


Figure 2. Relationship among the four sub-objectives under the Main Objective II (the directed arrows mean flows of information among them)

2.2.3 Cooperation with CCAMLR

The CCAMLR has been engaged in scientific research and analyses regarding the Antarctic marine ecosystem, and in 2008 an IWC/CCAMLR joint workshop on ecosystem modeling in the Antarctic (the Joint CCAMLR-IWC Workshop to Review Input Data for Antarctic Marine Ecosystem Models) was held. The proponent contributed to the Workshop through providing information on whale abundance, biology and ecology for the baleen and toothed whales in the Antarctic. Furthermore the CCAMLR Working Group on Ecosystem Monitoring and Management (WG-EMM) agreed in its 2014 meeting to further IWC/CCAMLR joint work. Data obtained from this new scientific research program, *e.g.* abundance and trends of key whale species, will contribute to this joint effort.

As noted earlier, within the Antarctic marine ecosystem the krill is a key prey species, supporting different species of baleen whales, pinnipeds, birds and fish. Information on krill biomass trends and krill size structure is fundamental for the development of ecosystem models, independent of the specific purposes the modeling exercise could have. This was recognized by the JARPA II review workshop that recommended, in the context of ecosystem modeling research, 'that future high priority be given to obtaining new estimates to allow an area-based time series of krill biomass estimates' (IWC, 2014a).

The JARPA II review workshop recommended that 'future surveys conducted during JARPA II employ survey design standards, including the spacing between net-tow stations along the acoustic transect lines, similar to those developed and implemented for the CCAMLR 2000, BROKE and BROKE-West surveys' (IWC, 2014a). With this recommendation the workshop recognized the experience accumulated by CCAMLR on the study of krill biology and abundance.

The best way to collect the information on abundance and size structure of krill, in line with the recommendations by the JARPA II review workshop, is through international research collaboration with CCAMLR experts. In this way, data on krill collected by this research can be used for multiple purposes and by multiple organizations, *e.g.* for the purpose of ecosystem modeling under the Japanese research program, and for ecosystem modeling under CCAMLR.

As described in Appendix 8, two kinds of krill surveys are proposed, *i.e.* (i) a krill survey along with a dedicated whale sighting survey, aimed to obtain a relative index of abundance (Sighting survey vessel-based krill survey), and (ii) a dedicated krill survey that follows CCAMLR standard methodology, aimed to obtain an absolute index of abundance (CCAMLR-type dedicated krill survey). In order to facilitate the international participation and collaboration, the latter will be conducted outside of, but coordinated with, this program. For this international survey, Japan is considering to provide the research vessel *Kaiyo-Maru* (2,630GT) owned by the Fisheries Agency of Japan. It is under consideration that the vessel would be available for at least one austral summer season in the first six-year period of the research program.

A detailed survey proposal, which will follow previous survey designs developed by CCAMLR, will be submitted to the CCAMLR Scientific Committee meetings for discussion and endorsement.

The research and analyses under this new research proposal, focusing on whale species, and the research activities on the Antarctic marine living resources under the auspices of CCAMLR should be and indeed are mutually complementary in promoting the understanding of the Antarctic marine ecosystem through data exchange, joint analysis and so on.

In consultation with experts to be involved in the international, independent krill survey mentioned above, regular krill surveys (the former of the aforementioned two proposals) will be carried out as part of this research program, by using echo-sounder instruments and sampling net installed in some of the vessels participating in the dedicated whale sighting surveys.

An outline of the future krill surveys as indicated above was already presented to the WG-EMM held in July 2015. The Working Group agreed that detailed information on the design of sighting survey vessel-based krill—survey would be submitted with data from the survey in the first season to the next meeting of the Sub Group on Acoustic Survey and Analyses Methods (SG-ASAM), Working Group on Statistics, Assessment and Modelling (WG-SAM) and WG-EMM.

2.2.4 Contribution to the future improvement of the RMP

While the RMP was developed as a single species whale management system, there are scientific discussions that an attempt should be made in order to incorporate factors such as interactions with other whale species into the RMP evaluation process and also into the RMP itself. This need is recognized by the IWC SC (IWC, 2014d). This research program aims to support the system to calculate sustainable catch limits, the RMP, under the Main Objective I, in accordance with the ICRW while reflecting the reality in the marine ecosystem (*e.g.* multi-species interactions) more accurately. This is regarded as an important long-term project for the IWC SC (IWC, 2014d). It is also consistent with the general direction in the field of fisheries resources management to introduce the ecosystem approach (*e.g.* Plagányi, 2007).

This new research program also intends to contribute scientifically to the future improvement of the RMP through the investigation of the possibility of building ecosystem models which constitutes an important interaction between the Main Objectives I and II of this research program. More specifically, in the existing RMP *Implementation*, species interactions are not taken into account explicitly. However, some adjustment to reflect this multi-species aspect would contribute to the improvement of operating models and harvest controls. For example, the ecosystem model could give some information on changes in carrying capacity and provide a scenario for running "RMP" simulation to see if the current single species CLA may or may not "work" (*e.g.* may fail to allow harvests levels that are perfectly possible without putting a population at risk) under the changes in estimated carrying capacity. In this regard,

this Main Objective II could contribute to the future improvement of the RMP in terms of species interactions.

2.3 Consistency with Annex P

In the development of these objectives, scientific issues that have been actively discussed at the IWC SC (e.g. Implementations and Implementation Reviews of the RMP, in-depth assessments of Antarctic minke and other whales, investigation of marine ecosystems through ecosystem models) as well as the previous four research objectives of JARPAII have been taken into account (see Appendix 2).

Under the framework of the ICRW, the IWC SC has established its guidelines for reviewing scientific research to be conducted under Article VIII, and the current guideline is Annex P adopted in 2008 and most recently revised in 2013 (IWC, 2013b). As discussed in the ICJ Judgment (paragraph 58), Annex P lists these broad categories of objectives for such research: (1) to "improve the conservation and management of whale stocks"; (2) to "improve the conservation and management of other living marine resources or the ecosystem of which the whale stocks are an integral part"; and (3) to "test hypotheses not directly related to the management of living marine resources".

Both of Main Objectives I and II of this program, namely, (i) *Improvements in the precision of biological* and ecological information for the application of the RMP to the Antarctic minke whales and (ii) *Investigation of the structure and dynamics of the Antarctic marine ecosystem through building* ecosystem models are clearly within the research categories identified by the IWC SC in Annex P mentioned above, as were the cases of JARPAII research objectives (Appendix 2). Each of them has more specific and detailed targets and sub-objectives (for further details, see sections 2.1 and 2.2).

It should be noted that, as mentioned above, the ICJ Judgment noted that "[t]he research objectives [of the JARPAII] come within the research categories identified by the Scientific Committee in Annex Y and P" and "based on the information before it, the Court thus finds that the JARPAII activities involving the lethal sampling of whales can broadly be characterized as "scientific research" (paragraph 127).

2.4 Data and samples to be collected for the Main Objectives I and II

In order to achieve the Main Objectives I and II described above, collection of a variety of data and samples, which provide important information for abundance, reproduction and nutritional conditions of whales, will be conducted under this research program (see Table 2). These data will be collected through an appropriate combination of lethal and non-lethal techniques as explained in Section 3.

Valuable data and samples to be collected should be fully utilized as much as possible, so they could also be used in various scientific analyses for purposes other than achieving the Main Objectives I and II (see Table 2). For example, organ abnormalities have been observed in whales in the Northern Hemisphere (*e.g.* Mikaelian *et al.*, 2003), and this situation cannot be discarded for whales in different ocean basins. Therefore in this new program, internal tissues obtained by the lethal sampling of Antarctic minke whale, the necessity of which will be explained in Section 3, would be examined for monitoring pathology. Visual observation for organ abnormalities would be made on internal organs such as liver, lung and thymus. Immunological examinations would be conducted on blood serum samples.

Table 2. Data and samples to be collected under this research program.

	Data		Sample
Abunda	nce estimate		
*#	Weather data		
*#	Effort data		
*#	Sighting record of whales		
*#	Angle and distance experiments		
*#	Ice edge line		
Environ	mental data	***************************************	
#	Temp. Salin. (XCTD)		
#	Echo sound (krill distribution/abundance)		
	Marine debris (sea surface)		
Antarct	ic minke whale		
*#	Catching date and location	#	Prey species in stomach for feeding ecology
	Photographic record of external character	#	Feces and colon contents for feeding ecology
	Record of internal and external parasites	*#	Testis for reproductive study
*#	Sex and body length	*#	Ovary for corpora counting and reproductive study
*	Body proportion for stock structure		Mammary grand and endometrium for reproductive study
*	Skull measurements (length and breadth) for stock structure	*#	Earplug for age determination
*	Satellite tracking for stock structure	*#	Ocular lens for age determination
#	Body weight for feeding ecology	*#	Baleen plates for age determination
#	Organ weight including fat weight for feeding ecology	*#	Tissue samples for genetic study
#	Diatom film record for feeding ecology	*#	Tissue and organ samples for chemical study
#	Blubber thickness for feeding ecology	#	Tissue and plasma samples for physiological study
#	Stomach content: freshness and weight for feeding ecology		Vertebral epiphyses for physical maturity
#	Diving behaviour for feeding ecology		
*#	Testis weight for reproductive study		
	Mammary grand: lactation status and measurement for reproductive study		
	Fetal number, sex, length and weight for reproductive study		
	Marine debris (stomach)		
	Gross pathological observation and sampling		
Otherl	arge whales		
Other I			

^{*:} Data or samples to be used for Main Objective I; #: Data or samples to be used for Main Objective II (Other items will be used for other research purposes)

3. RESEARCH METHODS

In this section, research methods employed under this proposed program are described step by step in the following order: 1) firstly, research area and period are specified (Section 3.1); 2) secondly, survey methods for whales and other targeted objectives are explained, including Japan's consideration of the use of lethal and non-lethal means (Section 3.2); 3) thirdly, having provisionally concluded that there is no other means than lethal methods to obtain age data that is indispensable for achieving the Main Objective I, the calculation of sample size for Antarctic minke whales, including its logic and result is presented (Section 3.3); 4) lastly, research vessels to be used, and personnel to be involved are specified (Section 3.4). New non-lethal techniques to be employed and tested in this research program are described in Section 4.

3.1 Research area and period

3.1.1 Research area

Determination of the research area and period is a crucial component of the proposed project. The Management Areas from III to VI as defined by the IWC SC, including Areas IIIW and VIE that were not covered under JARPAII but were a part of the recommendation of the JARPA II review workshop in 2014 for further research (IWC, 2014a), are proposed to be the primary research areas for the Main Objective I. This is because of the existing level of scientific knowledge about the Antarctic minke whale stocks in those areas and the resultant high potential for the application of the RMP to them. These four areas are also relevant to Main Objective II and data obtained there will be used for the purpose of building ecosystem models. It should be noted in particular that the Management Areas IV and V provide

useful information in this regard as the recent observed shifts of the species composition of whales in those areas might be related to changes in the Antarctic ecosystem (Pastene *et al.*, 2014a).

3.1.2 Research period

The research period under this program is planned to be 12 years with a midterm review after the first 6 years in order to achieve research objectives. The Main Objectives I and II require intensive researches that last for certain period of time. This is because the data sought under these objectives includes long-term trends of changing rates of various biological and ecological parameters. It is therefore considered that certain scientific achievements could be made after about 12 years of the analytical works that is to be conducted based on data obtained through this program (Section 6 shows a flowchart of surveys and analyses to be conducted in the first and second six-year period). In addition, in designing the time frame of this program, practical considerations need to be taken into account, including the natural environment in the Antarctic Ocean including sea and ice conditions where research can be conducted only during a limited period time, required time for verifications and analyses of collected data and information, the capacity of the research such as the number of available vessels, researchers and personnel, and financial constraints. In view of the prospect of scientific works as well as practical considerations, this plan sets 12 years as the timeframe for its research.

This plan is going to conduct a midterm review after the first six year-period of the program, at the half of the entire research period. The detailed timeline of research activities in this program is presented in the Section 6.

An assessment of the plan, including on the feasibility of non-lethal methods will be made on a yearly basis.

After the 12 year research period, with an additional research period if necessary under a contingency backup plans (see Section 7), this research will be completed and reviewed in accordance with Annex P.

3.2 Research Methods

3.2.1 Consideration of non-lethal means and lethal sampling

Under this research program, it is required to collect various types of data which are essential for estimating different parameters necessary under the Main Objectives I and II. Taking account of what is stated in the ICJ Judgment, the most important issue, with regard to its research methods, to be examined first is whether it is necessary to adopt lethal methods in order to achieve its stated objectives. This plan attempts to carefully consider this aspect, namely the availability, feasibility and practicability of non-lethal methods in collecting required data, at the initial stage of the drafting of this new program (for further details, see Section 4). There are a lot of information required for achieving the Main Objectives I and II, namely abundance estimate, stock structure and other biological and ecological parameters including ASM as demonstrated in Section 2. In obtaining such information, one need to look at what types of data are necessary, and in view of the ICJ Judgment, whether such data is obtainable through non-lethal methods. This plan focuses on one of the data that is least likely to be obtained through non-lethal methods and then critically review whether it is indeed unobtainable by methods other than lethal sampling. After giving serious scientific consideration, it has been concluded that age data at the annual scale can be obtained only through lethal sampling methods and thus lethal methods need to be employed under this program (see, Section 3.2.1.1).

3.2.1.1 Main Objective I

It is considered that this plan should look into the possible use of non-lethal methods in collecting agedata for the purpose of achieving the Main Objective I.

There are mainly two potential methods and one well-developed method to collect age-data for whales: DNA-based methods, the use of enantiomers of aspartic acid in lens and counting of growth layer groups (GLGs) accumulated in the earplug of whales. Each of these methods is assessed below.

Firstly, there have been continued academic efforts in recent years to explore the use of DNA-based methods for age determination in different animal taxa. They include 1) telomere length assays; 2) T cell receptor excision circle (TREC) qPCR for humans; 3) changes in levels of specific mRNA in mosquitoes; 4) assays for specific CpG site methylation levels in humans; and 5) accumulation of specific lipids in whale blubber (see Polanowski *et al.*, 2014). In the case of whales, DNA can be extracted from biopsy skin samples, and there have been various attempts, including in JARPAII, to employ and improve this method (Kanda *et al.*, 2014). This indicates that there is a potential that age can be determined using such non-lethal approaches.

However, it should be noted that there is an insurmountable difficulty at this juncture to use these DNAbased methods. This technique does not provide the chronological age of the individual but rather a physiological age that can be used as a proxy for chronological age (IWC, 2015a pp16). The "physiological age" does not change with an annual trigger, but change with constant biological processes. There is almost always population-wide variability in correlation between "physiological age" estimates and "chronological age" because individuals within a population have different genotypes and experience different life histories (Polanowski et al., 2014). A DNA methylation approach was recently developed and applied to humpback whales (Polanowski et al., 2014), and the IWC SC noted that this approach can be applied to increase the power of the close-kin mark recapture by identifying the intergenerational relationship within parent-offspring pairs (IWC, 2014f). It should be recognized, nevertheless, that the precision of the age determination by this method (i.e. SV=2.991) is not large enough for using age data in population dynamics models like the SCAA. Furthermore the IWC SC noted that *de novo* calibration of methylation pattern is required to develop this method to different species as inter-species methylation patterns and accumulation rates are not consistent (IWC, 2014b). This technique may require calibration not only by species but by population and period. There is as yet no experience on this regard, because it has been applied to a single population of a single species of cetacean, i.e. humpback whales, due to the novelty of the technique (IWC, 2015a pp16). This has led to a conclusion that the DNA-based methods are not practicable at this juncture.

Secondly, there is another method for age determination utilizing enantiomers of aspartic acid in lens of whales. These parameters are measured using high performance liquid chromatography (Yasunaga *et al.*, 2014). New statistical methods, such as the Delta method (George *et al.*, 1999), are applied to estimate the precision of age estimation with the enantiomers. Nevertheless, there is no practical method to obtain lenses of whales without lethal sampling. It should also be noted that the precise measurement methodology is still under development (see Appendix 4).

Lastly, these discussions leave counting GLGs accumulated in the earplug of whales as the only practicable mean at the present time to obtain age data at the annual scale. It was indeed pointed out in the scientific literature that the earplug has proved to be a valid and useful tool for age determination (Lockyer, 1984b), and is the only method providing age data accurate enough for population-level analyses such as the SCAA analyses of Antarctic minke whales (Punt *et al.*, 2014a). This methodology has been used for more than 25 years under JARPA and JARPAII and its scientific outcomes have been positively evaluated in previous IWC SC meetings (IWC, 2014a). When it comes to the earplugs of whales themselves, they are formed in the auditory meatus and there have been no academic literature that proves that there is practical alternative method to collect them other than lethal sampling.

Recent investigations have indicated that the precision of methylation-based recruitment estimates from SCAA are much worse than those obtainable from ear-plug based readings, with the methylation-based results hardly better than those in the absence of any age data at all (Kitakado, 2015). Hence at this juncture, indications are that the non-lethal methylation approach cannot provide ageing information as precise as ear-plug base age readings to perform assessments and consequently management.

In light of these scientific considerations, lethal sampling is indispensable for achieving the research objective of this research plan, namely Main Objective I.

However, this does not establish that non-lethal methods will not be effective for the collection of other necessary parameters as well as meaningful age-data in the future. In order to benefit from their practical contribution and to explore possibility of their future use, this plan will utilize non-lethal methods for collecting other parameters. At the same time, assessment of the feasibility of non-lethal methods will be intently conducted in this program (see Section 4), which would lead to future adjustment of the sample size for lethal sampling as appropriate. As part of the efforts, a feasibility study on DNA methylation approach for age determination will attempt i) a *de novo* calibration of methylation pattern for Antarctic minke whale, and ii) to investigate ways on how to increase precision of the age estimate (see Section 4 for details).

3.2.1.2 Main Objective II

Necessary data for Main Objective II are listed in Table 2, Section 2.4 (non-lethal methods, which will be adopted for the Objective, will be described in detail in Section 4). Among them, information about blubber thickness and stomach contents cannot be obtained through non-lethal methods as the ICJ observed (paragraph 133). However, as described more in detail in section 3.3.1, the calculation of a sample size based on the parameters required for Main Objective II is not feasible at this stage. Therefore, biological data, namely blubber thickness and stomach contents, of Antarctic minke whales obtained through lethal methods under the sample size calculated based on the parameter for Main Objective I, in combination with necessary data taken through non-lethal methods, will be utilized and analyzed for Main Objective II as well.

3.2.2 Sampling survey methods for Antarctic minke whales

Antarctic minke whales will be taken in the area south of 60°S in Management Areas III to VI (for the sample size, see Section 3.3). Survey courses will be established in offshore and ice edge waters of the research area by the line transect method. Two or three sampling and sighting vessels will advance along parallel track-lines 7n. miles apart, at a standard speed of 11.5knots. Basically, each of the sampling and sighting vessels will change the track-line order every day to avoid possible sighting bias by fixed position. Starting point of the day will be set at the position where one of the vessels ended the surveys on the previous day in the most advanced position. Other vessel will move to the starting position of the next day after the end of the daily survey. A maximum of two (2) Antarctic minke whales per school sighted will be taken randomly. Sampled whales will be immediately transported to the research base vessel, where biological measurements and sampling will be carried out in a systematic manner.

Detailed annual plan which includes areas to be surveyed is not included in the present plan because of the security reason (i.e. to avoid violent sabotage activities by anti-whaling groups).

All whales will be taken using explosive grenades to ensure instantaneous death in line with existing norms of whale killing methods. If instantaneous death is not achieved by the primary killing method, a suitable secondary method, such as a large caliber rifle or another grenade will be chosen, depending on the condition of the whale.

3.2.3 Sighting survey methods for whales

Abundance estimation is fundamental information necessary for the conservation and management of the whale stocks concerned. Sighting surveys are to be conducted by the line transect method. Survey protocols follow the Requirements and Guidelines for Conducting Surveys and Analyzing Data within the Revised Management Scheme (IWC, 2012c) so that they are conducted under the oversight of the IWC SC. Sighting protocols are the same as those in IDCR/SOWER (Matsuoka *et al.*, 2003). In general,

the surveys will cover areas south of 60°S. Surveys are to cover one IWC Management Area (one of the Areas III to VI) in a year. Cruise tracks will be designed systematically in accordance with mathematical and scientific calculation in the research area. The sighting survey will be conducted using (1) Closing mode and (2) Passing with Independent Observer mode. Both survey modes follow the protocol endorsed for the IDCR/SOWER surveys. Primary search effort will be conducted only when weather conditions are acceptable (Matsuoka *et al.*, 2003) (see details in Appendices 3 and 9).

This sighting survey method was used in JARPA II and it should be noted that the expert panel to review its results ("JARPAII Review Panel") "welcome[d]" the results from sighting surveys conducted under the JARPA/JARPA II programs, mentioning that the "importance of monitoring trends in abundance for cetacean species is of general conservation and management interest as well as providing information relevant to the JARPA II programme objectives and especially as input to ecosystem models" (IWC, 2014a).

The plan of the dedicated sighting survey for each austral summer season including details of the survey design, will be presented to the corresponding annual IWC SC meetings. The survey plan for the dedicated sighting survey in the 2015/16 austral summer season was presented to the 2015 IWC SC meeting (SC/66a/SP08).

The survey design will take into account the specific research activities planned for a particular Area and austral summer season, and will follow the recommendations provided by the NEWREP-A Review Panel (IWC, 2015a pp14-15), including incorporating detailed track lines. NEWREP-A researchers will work closely with the IWC SC before finalizing the survey approaches and to ensure that future survey plans fully follow the IWC SC guidelines for such survey plans.

The survey design for the first two-three years will include feasibility studies on biopsy sampling of and telemetry for Antarctic minke whales, on which the NEWREP-A Review Panel made specific recommendations (IWC, 2015a pp11-12). It will also include krill and oceanographic surveys (see below and Section 6).

In response to the NEWREP-A Review Panel recommendation regarding the g(0) estimation (IWC, 2015a pp13), identification of duplicate sightings will be also conducted for whale species other than the Antarctic minke whale during NEWREP-A sighting surveys in IO mode. In order to determine whether g(0) is significantly different from one, analyses on duplicate sightings will be conducted for those whale species given that sufficient data will be obtained under the NEWREP-A program.

3.2.4 Other established non-lethal survey methods for whales

Photo-ID and biopsy experiments will continue to be conducted for blue, fin, humpback, southern right and killer whales. These methods will provide useful data for stock structure, mixing and movements. The JARPAII Review Panel welcomed photo-ID photographs of humpback, blue, and southern right whales taken during the JARPA and JARPA II cruises, recognizing "their potential to provide information on stock structure, mixing and movements" (IWC, 2014a). The JARPAII Review Panel also "welcome[d]", in general, results of genetic studies on humpback, fin and southern right whales using biopsy samples collected under the JARPA and JARPA II programs (IWC, 2014a). As to Antarctic minke whales, effectiveness and practicability of these techniques are in dispute due to its characteristics such as their size and speed of their movement that are different from other species (IWC, 2014c).

3.2.5 Survey methods for krill

As noted in Section 2.2, two kinds of krill surveys are planned, one under an international research outside of this research program (CCAMLR-type dedicated krill survey), and the other under this program (Sighting survey-based krill survey). Details are shown in Appendix 8.

The former one will apply a standard survey design similar to those developed and implemented for CCAMLR 2000 (Trathan *et al.*, 2001), BROKE (Nicol *et al.*, 2000) and BROKE-West (Nicol *et al.*, 2010)..

The main objective of the krill surveys is to estimate the abundance of Antarctic krill acoustically (absolute and relative), and to obtain the length frequency distribution and maturity stage of Antarctic krill in the survey area.

The proponent will respond to recommendations by the NEWREP-A Review Panel (IWC, 2015a pp20-23) in the following ways.

- i) Ship(s) and echosounder system(s) will be tested before steaming to the Antarctic to determine the likely effective acoustic sampling range (signal-to-noise by depth characterization) and potential for detecting krill.
- ii) Cross-calibration between both types of krill surveys vessels will be carried out the in waters around Japan, in a near synchronous way, as the first priority. However, the proponent might conduct the calibration in the survey area as well if conditions are met.
- iii) Simultaneous survey by the dedicated krill survey vessel equipped with a frame-type trawl and echosounder and that by, the dedicated whale sighting vessel equipped with an echosounder and the whale sampling vessels will be conducted within a small-scale experimental survey area over a short time period (e.g. one week) so that comparisons among vessels/samples can be made.
- iv) Sufficient time will be allocated for net sampling of krill, by following a survey design similar to those developed and implemented for CCAMLR 2000, BROKE and BROKE-West. Approximately 2.5-3 hauls per day will be a standard for net sampling frequency under the proposed CCAMLR-type dedicated krill survey.

Recommendation i) above will be conducted in the 2015/16 season. The other recommendations will be considered in future surveys. Further details of the krill survey design along the dedicated whale sighting vessel in 2015/16 season have been given in SC/66a/SP08.

3.2.6 Other survey methods

(i) Oceanographic observations

Oceanographic observations will be conducted in parallel with krill surveys (see Appendix 8).

The JARPAII Review Panel recommended continuation of oceanographic observations with improvement in several points, stating that "collection of such simultaneous [oceanographic] data is an important component of being able to relate the physical and biological habitat with whale distribution and density and ultimately to assist in developing an understanding of the role of whales in the ecosystem" (IWC, 2014a). The expendable conductivity-temperature-depth profilers (XCTD) and conductivity-temperature-depth profilers (CTD) will continue to be used.

For the CCAMLR-type dedicated krill survey, careful consideration will be given to the scale and design of oceanographic sampling, considering the previous experience of BROKE and BROKE-West surveys. Sampling at approximately 2-3 sites per day will be a standard for the CTD oceanographic sampling frequency under the proposed CCAMLR-type dedicated krill survey.

Further details of the oceanographic survey to be conducted along the dedicated whale sighting vessel in the 2015/16 season have been given in SC/66a/SP08.

(ii) Marine debris

The JARPAII Review Panel commended the collection of debris data, both in the environment and in the stomachs of the whales that could provide valuable baseline data and stated that such information should regularly be analyzed and presented to the IWC (IWC, 2014a). Observation of marine debris will continue to be conducted during sighting surveys.

3.3 Calculation of sample size for Antarctic minke whales

3.3.1 Calculation

As discussed in Section 3.2.1 above, the key data that requires lethal methods is the age. The age will be measured from counting GLGs accumulated in the earplug. The age data themselves will be then used as main data in SCAA analysis for the estimation of biological parameters including the natural mortality, MSYR and parameters in recruitment relationship and also ecological parameters like the time-varying carrying capacity. In addition, an indicator derived from a combination of age and maturity status, ASM, is of great importance not only for contributing information on the proportion of matured animals in the SCAA but also as an important indicator to know the change in the nutritional condition for whale populations.

The examination for the Antarctic minke whales based on the transition phase in the earplugs has shown that the mean ASM gradually got younger since the late 1940's, when other baleen whales went to depletion, and it then became stable or slightly increasing since 1970 (Bando *et al.*, 2014). This behavior might be attributed to the decreased and then increased food competition and it also reflected to the past increase and recent decline of carrying capacity for the Antarctic minke whales (Punt, 2014b), possibly due to the competition with other species.

The population parameters such as natural mortality, ASM, recruitment rate and carrying capacity can be dynamically changing across years by reflecting the ecosystem interaction among species in the Antarctic Ocean. The ASM is affected by the mid- or long-term nutritional condition in the population. This does neither change rapidly nor fluctuate greatly. In this sense, it might be possible to detect a future change in the ASM if it happens. In addition, the ASM can be observed with relatively less uncertainty to monitor and also has an advantage that it can be observed directly. The ASM is influential to the number of mature animals (especially female), and hence a possible future increase in the ASM affects the productivity and of course population dynamics of the Antarctic minke whales. These factors demonstrate that the ASM makes considerable contribution to achieving Main Objective I in the sense that it helps improve the SCAA performance for estimating other key parameters such as MSYR. This leads to the better assessment and management of the Antarctic minke whales. It therefore constitutes the scientific ground for the sample size calculation under this proposed plan.

Given that the ASM has an accessible feature to set a target sample size, a single indicator, the age-at-50% sexual maturity (ASM50), is used to set an annual sample size in this plan (see Section 2.1.3). For this purpose, a conditioning was conducted to infer some effect sizes using maturity data taken from past JARPA and JARPA II samples. Secondly based on conditioned values for the changes in the ASM50 and consideration of the changes in the mean ASM based on transition phase over a couple of last decades, simulation studies are performed to examine sample sizes to detect possible changes. A necessary level of annual sample size is finally determined by the statistical testing and estimation performance. Specifically, the annual sample size is calculated to assure a reasonable degree of statistical power for detecting the change (i.e. the statistical power is greater than 90% for detecting ASM50's annual increase of 0.1 per year, and is close to 80% for the extent of increase 0.075 per year) with 5% significance level (i.e. the level of accuracy) over the whole research period (i.e. 12 years) and to improve estimation precision of the estimate of the extent of change (for more details, see Appendix 13). The annual sample size for Antarctic minke whales is thus set at 333.

The sample size for lethal method is calculated based on the required amount of both earplugs and reproductive organs for collecting age and maturity data respectively. This is directly related to Main Objective I, which includes to perform the SCAA and to provide better specifications for the *IST*s.

It should be noted that, in the initial stage of developing of this proposed plan, it was examined through simulation studies whether required sample size for Main Objective II can be calculated. For determination of the sample size with a simulation framework, the first thing to do is to derive some conditioned models as in the case above. One of the ecosystem models which will be employed in the current plan is a set of multiple production models for prey (krill) and predators (mainly baleen whales) with some links between them (see Section 2.2). It may be possible to set some conditioned values for the models. However, parameters in the models such as abundance estimate might have their own large uncertainty in addition to the inevitable uncertainty of the models themselves, and therefore it was not feasible to fix a set of reliable simulation scenarios at this stage. In addition, the models assume to use both the time series of abundance estimates for component species and that of a nutrition index for the Antarctic minke whales (e.g. stomach fullness or its proxy value), but the latter index from one krill predator, the Antarctic minke whales, is not so influential in the model simulations. Therefore it is not achievable at this stage to assess its contribution to the estimation of ecosystem models. In other words, it is not possible to evaluate the influence of sample size for Antarctic minke whales on models' performance. For this reason, the sample size determination based on the data items to be used for the Main Objective II is not scientifically feasible at this time. As the program progresses, the calculation of sample size of Antarctic minke whales could be retried and the plan could be accordingly modified.

Even though the calculation of a sample size under the Main Objective II is not feasible at this stage, tissue and other biological samples and data to be obtained from their analysis under the sample size calculated based on the ASM as described above will be fully utilized for achieving the Main Objective II in combination with information from non-lethal research methods and other researches and studies. With such effort, there must be certain progress in achieving the Main Objective II, while it is not possible to foresee the level of expected achievement by using statistical methods at present time. (For the effect of the proposed sample size on the stocks, see Section 5.)

3.3.2 Possible adjustment to the latest available scientific knowledge

This proposed sample size is the best estimation at the present level of scientific knowledge in relation to ASM. However, the sample sizes should be calculated based on the existing knowledge or estimates at the time and it is hard to deny that the assumptions used above could turn out to be inappropriate in light of updated information generated during this research plan. In this regard, the calculated sample sizes should be re-visited at the time of mid-term review (see Sections 3.1 and 6). This research program allows for possible modification of the future sample size by taking advantage of the latest available knowledge.

3.3.3 Necessity of new samples and importance of the long time-series data set

In accordance with the IWC's review process (IWC, 2013b), special permit research proposals shall include an assessment of why: 1) non-lethal methods; 2) methods associated with any ongoing commercial whaling; or 3) analyses of past data; have been considered to be insufficient. An assessment on the first item is already presented in Section 3.2, and for the second item, it is obvious that methods associated with ongoing commercial whaling are not available since there is not such an operation in the Antarctic Ocean. In relation to the third item, as described in Section 2 and the earlier part of Section 3, this research program's objectives require to ascertain ongoing changes occurring in whale resources and Antarctic ecosystem through the collection of relevant data on a yearly basis. This inevitably needs new samples under this program.

The data to be obtained from the new samples, being combined with the past data derived from JARPA and JARPA II as well as from commercial whaling operations, will form an integral part of the long time-series data set, while there is a one-year gap in between (2014/15). Such a data set would be the foundation for the better understanding of the long-term transition of the Antarctic ecosystem including whales and for the conservation and management of whale resources.

The critical importance of the long time-series data set is duly recognized within the framework of the ICRW, and clearly expressed in the convention, specifically Article VIII, paragraph 4, which says "continuous collection and analysis of biological data in connection with the operations of factory ships and land stations are indispensable to sound and constructive management of the whale fisheries" (emphasis added). This idea is not unique to the ICRW. Agenda 21, an action plan with regard to sustainable development, adopted at the United Nations Conference on Environment and Development in 1992, provides that "States, with the support of international organizations, whether sub-regional, regional or global, as appropriate, should cooperate to exchange on a regular basis up-to-date data and information adequate for fisheries management" (paragraph 17.56 (b)). To have long time-series data set in both fisheries-dependent and fisheries-independent by continued accumulation of new data is a common practice in regional fisheries management organizations established under international conventions.

3.4 Research vessels to be used and personnel to be involved

For this research program, one research base vessel, a few sighting and sampling vessels and one or two dedicated sighting vessels will be used. The vessels are equipped with necessary facilities that are suitable for collecting the data and samples required to achieve the research objectives.

The research under this plan will be conducted by the ICR. Scientific experts from the ICR will be engaged in the systematic collection of data and samples on board and laboratorial analyses of them, in cooperation with other relevant researchers.

4. TRIAL, EVALUATION AND DEVELOPMENT OF NEW NON-LETHAL TECHNIQUES

There are established non-lethal research methods, non-lethal methods of which their usefulness is in dispute, and newly emerging non-lethal methods at very preliminary development stages. In this research plan, all those non-lethal methods will be employed and tested as described below.

JARPA and JARPAII employed a series of non-lethal research methods for research items that require non-lethal methods or that can be conducted with non-lethal methods. Those non-lethal methods which will continue to be used in this research program are described in Section 3.2.4.

Taking account of the 31 March 2014 ICJ Judgment, specifically its statement that "the JARPA II Research Plan should have included some analysis of the feasibility of non-lethal methods as a means of reducing the planned scale of lethal sampling in the new programme" (paragraph 137) the new research proposal includes the examination of non-lethal techniques, which could potentially complement or replace lethal techniques used in the context of achieving the research objectives. The proposed lethal sample size in Section 3.3 represents the best estimation at the present time taking account of the available and established non-lethal research methods. However, it should be noted that these methods could be further improved in the future depending on the progress of the feasibility experiments in this Section.

This Section presents a summary of the non-lethal techniques to be considered and tested in this new research program. The aim is to investigate the potential of these techniques for achieving the objectives of the program. In the investigation it is necessary to consider the following four major tests in assessing the feasibility of non-lethal methods; whether the same data sought (*e.g.* age) can be obtained by a non-lethal method; whether it is of sufficient quality for analysis (*i.e.* accuracy); whether the cost to obtain the data is realistic and reasonable; and whether enough data can be obtained in terms of quantity for

statistical analysis. These tests are often inter-related and not exhaustive. Depending on the results of the investigation, as well as the results of the feasibility study of non-lethal methods under JARPN II, this research plan could be revised in relation to the lethal sample size.

It should be noted that the number of samples to be obtained by non-lethal methods is not necessarily the number to be reduced from the lethal sample size. For example, even if the age-data can be obtained from a non-lethal method in the same quality and accuracy as from the lethal method, when the cost of obtaining the data by the non-lethal method is prohibitive, a sample size trade-off cannot be established in a simple manner.

4.1 Age data (Objective I (ii), Appendix 4) (also see Section 3.2.1.1)

In the previous as well as this new research program, age-data used in population dynamics models, are obtained from counting GLGs accumulated in the ear plugs of whales, which requires lethal sampling of whales. However, additional new methods of age determination, both lethal and non-lethal, will be developed and tested under this new research plan.

One of the methods is based on enantiomers of aspartic acid in lens of whales, which are measured using high performance liquid chromatography (Yasunaga *et al.*, 2014). New statistical methods, such as the Delta method (George *et al.*, 1999), are applied to estimate the precision of age estimation with the enantiomers. Lens of whales are obtained by lethal sampling.

In recent years some potential ageing method at a DNA level, have been reported (*e.g.* Koch and Wagner, 2011). If such methods are further developed there is a potential that whale ages can be estimated from analysis of DNA extracted from biopsy samples. In particular a DNA methylation approach was recently applied to humpback whales (Polanowski *et al.*, 2014).

In this new research plan, the DNA methylation approach will be evaluated to see whether or not the method is feasible for Antarctic minke whales (see Objective I(ii)).

Following the recommendations by the NEWREP-A Review Panel (IWC, 2015a pp16), at first, the stock effect on the DNA methylation (DNA-M) technique will be examined using 50 highly readable earplugs each from I-stock animals sampled in Area IVW and P-stock animals sampled in Area VE under JARPA II, ideally from whales sampled in a same year in each Area. In the next step the effect of the period will be examined by using a similar number of samples from different austral summers. DNA methylation changes will be identified at the Antarctic minke whale genes homologous to the humpback whales. The procedure for identification of age-related DNA methylation site and measurement of methylation level will follow previous studies (Maegawa *et al.*, 2010; Bocklandt *et al.*, 2011; Polanowski *et al.*, 2014). The proponent has already contacted a specialized laboratory and a university in Tokyo so that this study can be conducted in collaboration with experts. Preliminary results will be reported at the 2017 IWC SC meeting and final results at the 2018 IWC SC meeting. In this way the proponent will respond to the Review Panel recommendation that this feasibility study be conducted early in the NEWREP-A program (IWC, 2015a pp16). The proponent will also address the issue of inter-laboratory calibration in consultation with the IWC SC.

4.2 Stock structure (Objective I(iii), Appendix 6)

As explained in Objective I(iii), stock structure analyses of Antarctic minke whales are based on both DNA and morphometric data since both data have been informative for studying the stock structure. As recently reported by Kitakado *et al.*, (2014b, c), indeed, morphometric data are as useful as genetic information. Accurate and detailed morphometric measurements used in the analyses require lethal sampling.

However DNA analyses can be carried out using skin samples obtained by biopsy as materials. Some previous studies have reported success in biopsy sampling of Antarctic minke whales, especially from areas near the ice-edge and for large school sizes (Gales *et al.*, 2013). There is however the need to conduct a feasibility study to examine whether or not biopsy sampling is possible for this species in

offshore areas and for small schools because these are cases where approaching whales are more difficult. This new proposed program will include such a feasibility study.

The feasibility study on biopsy sampling in Antarctic minke whale will be undertaken along with the dedicated sighting surveys, at an early stage of the first six-year period of NEWREP-A (Section 6 and SC/66a/SP08). As the program develops, the design of the feasibility study on biopsy sampling will take account of the four specific recommendations by the NEWREP-A Review Panel [1) involve people with expertise in successfully biopsy sampling common minke whales in the North Atlantic; 2) mimic the sampling strategy developed for lethal sampling; 3) record information on time taken, sea state, swell, etc. to enable a plausible measure of effort required to be developed; and 4) consider the amount of tissue and nature of tissue required (for each analysis and in total)], as far as practicable (IWC, 2015a pp11-12). In planning and implementing the feasibility study, the program will make use of experiences obtained by foreign (e.g. colleagues working with North Atlantic minke whale) and Japanese (e.g. colleagues working with North Pacific minke whale) experts. The "ideal" framework of the study suggested by the NEWREP-A Review Panel (e.g. representativeness of the entire area and time period as well as, random sampling techniques) (IWC, 2015a pp11-12) will be considered as a second step after examining the practicability of the sampling technique itself in the initial stage. It should be noted that the value of biopsy samples for the NEWREP-A research objectives should be evaluated in parallel with the results of feasibility studies of analytical approaches that potentially use biopsy samples (e.g. DNA-M, stable isotopes, hormones).

The new research considers the use of satellite tracking as an additional non-lethal approach to contribute to the study of stock structure of the Antarctic minke whale.

The feasibility study on telemetry for Antarctic minke whale will be undertaken along the dedicated sighting surveys, at an early stage of the first six-year period of NEWREP-A. With regard to field methods as well as tag types, the proponent will collaborate and consult with experienced foreign and Japanese colleagues (Section 6 and SC/66a/SP08).

4.3 Prey consumption (Objective II(iii), Appendix 10)

In JARPA and JARPA II, consumption by the Antarctic minke whale has been estimated using theoretical energy requirements and the diurnal changes of stomach contents mass through the lethal sampling of whales (Tamura and Konishi, 2014).

However it is very difficult to obtain robust estimates of consumption by Antarctic minke whales in their feeding area because of year-to-year variation of krill availability and uncertainty in the migration patterns and daily feeding behavior of the Antarctic minke whale. Successful attachment of several logger and transmitters for satellite tracking for the Antarctic minke whale has been reported (Gales *et al.*, 2013), that could potentially reduce the uncertainty in the consumption estimate. Therefore in the new research plan, the proponent will carry out both satellite tracking (migration patterns) and data logger trials (daily feeding behavior) to contribute to the study of feeding ecology and estimation of consumption by the Antarctic minke whale.

The JARPAII Review Panel noted that 'several cetacean research programmes include the collection of faeces from wild population to assess diet and food habit' and that 'the JARPAII programme is in position to assess the efficacy and accuracy of this non-lethal approach'. Furthermore the review workshop recommended that faecal samples (from the colon) be compared with stomach samples for species composition (IWC, 2014a). Japanese scientists have already examined the feasibility of prey identification based on DNA of the colon content, under the whale research program in the North Pacific (JARPN/JARPNII) (Kobayashi, 2010).

It is considered that this feasibility experiment is more relevant to North Pacific whale research because of the high diet diversity of whales in this region. It is already known that in the Antarctic, the Antarctic minke whales feed mainly on a single species, the Antarctic krill. Furthermore this technique does not provide quantitative information of food consumed, which is important for ecosystem model studies.

Therefore there is not much value to attempt this technique for investigating 'species composition' in the Antarctic minke whale diet.

In response to the NEWREP-A Review Panel suggestions and recommendations on the use of stable isotopes (IWC, 2015a pp 24-25), the new research plan also considers the examination for concurrent analysis of krill samples to ensure the correct determination of stable isotope ratios (δ 13C and δ 15N) in studies on feeding ecology. For the feasibility study different whale tissues will be used, which include skin that can be potentially obtained through biopsy sampling.

As a first step in the work to compare the overlap in diet amongst predator species using stable isotopes, prey species (krill) samples will be analysed to ensure the correct determination of stable isotope baselines. This will be done during the first six-year period of NEWREP-A (Section 6). Sampling of krill will be conducted in different geographical strata. At least five samples per sub area (e.g. East-South, East-North and Prydz Bay) will be analysed. Stable isotopes ratios for krill in each sub-area will be analysed each year. Accumulated information on isotope ratios in krill will be applied for the analysis of the overlap in diet amongst fin, humpback and Antarctic minke whales. After the first six years, the baseline data for krill will thus have been obtained. These results will be evaluated at the mid-term review of NEWREP-A (Section 6).

Concurrently, the overlap in diet amongst fin, humpback and Antarctic minke whales will be examined using stable isotope ratios in skin samples collected by lethal method (Antarctic minke whales) and by biopsy sampling (fin and humpback whales) under NEWREP-A program. This analysis will be carried out at an early stage of the NEWREP-A program based on the information on stable isotope baselines of krill in each area (Section 6) accumulated under the program. Results will be reported to the IWC SC meeting whenever available during the first six-year period and evaluated during the mid-term review of NEWREP-A.

Stable isotopes analyses on the edge of baleen plates will also be conducted under the NEWREP-A program to estimate time spent by Antarctic minke whale in feeding grounds. Stable isotopes in baleen plates (n=10-20) of Antarctic minke whales sampled under JARPA II will be analysed early in the research program, as an initial step. The results of the analyses will be reported to the IWC SC in 2018.

Regarding food consumption estimates, efforts will be made to develop more sophisticated calculations by adding some categories such as age, growth and maturity status based on energy requirements measured by changes in blubber mass and lipid contents, as recommended and suggested by the NEWREP-A Review Panel (IWC, 2015a pp24-26). The use of data collected previously is important in building the basic structure of the bioenergetics model but collection of recent data is essential for monitoring and forecasting the dynamics of the Antarctic marine ecosystem. Thus, the proponent will use the data collected previously for body weight-length relationships and maturity status as much as possible; however they also need recent data from NEWREP-A samples to examine the lipid contents and blubber weight relationship, and to monitor and forecast the dynamics of the Antarctic marine ecosystem. Preliminary results of the analyses will be reported to the 2016 IWC SC meeting. It also should be noted that the proponent will conduct consumption calculations based on the stomach contents in parallel.

For both of the calculation methods based on allometry (bioenergetics model) and stomach contents, the proponent will conduct the Monte-Carlo simulation trials which have been recommended by the JARPAII Review Panel and the IWC SC to take into account the actual range of the estimates including the uncertainties (see Tamura and Konishi. 2014). In the calculation based on the stomach contents, the proponent will continue to make efforts to minimize uncertainties in r (the ratio of low/high feeding intake), the length of the feeding season and the extent of night feeding for better estimation by satellite tagging, stable isotope of baleen plates and night-time sampling.

4.4 Chemical markers as potential body condition indicator (Objective II(iii), Appendix 10)

The body condition indicators such as blubber thickness, length of girth and total fat weight that can be obtained by lethal sampling have been used throughout JARPA and JARPA II periods (Konishi *et al.*, 2014, Konishi and Walløe, 2014). These data will continue to be collected as they will contribute to the Main Objective II. In 2014 the IWC SC agreed that the body condition and stomach content trends of Antarctic minke whales were decreasing significantly during the JARPA period (IWC, 2015b pp46).

In response to the suggestion by the NEWREP-A Review Panel (IWC, 2015a pp25), instead of retinol, the proponent plan to analyse the lipid content % of blubber throughout NEWREP-A research. At first, the total lipid content % of blubber from mid lateral position (near the position used in JARPA and JARPAII for the point measurement of blubber thickness), will be analysed since this position has been informative of changes of blubber thickness in relation to feeding season (Konishi *et al.*, 2008). The lipid content % of blubber will be compared to other body condition indicators such as blubber thickness and girth measurements. As an initial feasibility test, the measurements of body condition (blubber thickness, girth, total fat weight and the lipid content % of blubber) obtained from Antarctic minke whales collected under NEWREP-A will be used. At least 50 whales for each sex and sub area will be used for this analysis. (see details in Appendix 10).

4.5 Others

To respond the recommendation by the NEWREP-A Review Panel to investigate potential non-lethal techniques to determine sexual maturity in whales (IWC, 2015a pp17), progesterone analyses will be conducted in blubber tissues of Antarctic minke whales using JARPAII samples. Females with various reproductive status such as ovulating, resting and pregnant with small fetus will be selected, and hormone levels will be compared with histological information for reproductive organs. Analysis of the progesterone level for a total of 50 Antarctic minke whale samples will be assayed using the compact ELISA workstation (Crocodile, Titertek Berthold). Progesterone will be assayed with the Cayman Practice ELISA Kits (Cayman Chemical), according to the manufacturers' protocols. Preliminary results will be reported in the 2017 IWC SC meeting.

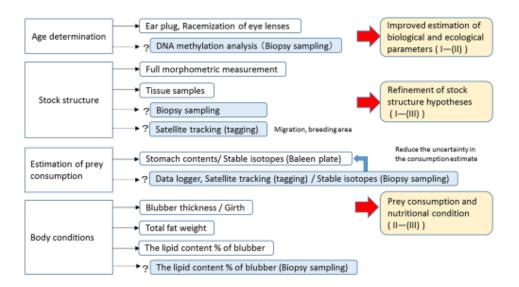


Figure 3. Use and evaluation of non-lethal techniques on Antarctic minke whale in the new research program (White: Lethal methods; Blue: Non-lethal methods that will be evaluated in this research)

5. EFFECT ON THE STOCKS

It is recognized that the RMP developed by the IWC SC is the most appropriate way to calculate catch limits for baleen whales without threatening the stock. At the JARPA II review workshop, it was noted that "the most appropriate way to assess the impact of future Special Permit catches on stocks is within the framework of an RMP-type process" (IWC, 2014a). However, the RMP *Implementation* for Antarctic minke whales has not been conducted for the last 20 years. In the calculation of the catch limits, conducted in 1993 (IWC, 1994), very narrow areas (i.e. 10 degrees longitudinal sectors) were used as *Small Areas* because limited information on the stock structure for the Antarctic minke whales was available at that time. Now that, some updated information is available for the stock structure of the Antarctic minke whales (*e.g.* "two-stock with mixing hypothesis" by Pastene *et al.*, 2014a, b, Kitakado *et al.*, 2014b, c) wider *Small Areas* might be a more appropriate definition to apply the CLA compared to the previous definition. Since the Main Objective I is to provide improved information for the RMP *Implementation*, a comprehensive RMP *Implementation* would be conducted after the completion of the proposed program.

Given this situation, a simple CLA application was made based on some new definitions of *Small Areas* using the most recent agreed abundance estimates (IWC, 2013a) shown in Table 3, the historical catch series in the study area and tuning level of 0.6, which is one of the tuning levels used by the IWC SC to examine performance of candidate management procedures (IWC, 1992). Catch limits obtained by the CLA were compared with the sample size in this proposed research plan (n= 333) in order to assess the effects of sampling on the stocks.

Table 3. Abundance estimates for each half area and for each stocks applied to CLA

Area	Year	P	CV	Stock	Year	P	
IIIW	1987/88	69,267	0.277	I-stock	1988/89	58,545	-
	1994/95	33,375	0.209	Base case	1996/97	90,532	
IIIE	1987/88	11,782	0.440	I-stock	1987/88	116,818	_
	1994/95	34,659	0.237	Sensitivity	1998/99	114,534	
IVW	1988/89	23,668	0.237	P-stock	1987/88	281,047	
	1996/97	29,745	0.477	Base case	2000/01	228,303	L
IVE	1988/89	23,096	0.237	P-stock	1987/88	222,774	(
	1998/99	26,127	0.477	Sensitivity	2000/01	204,301	_
VW	1985/86	105,951	0.159				
	2001/02	43,640	0.139				
VE	1985/86	154,658	0.189				
	2003/04	136,457	0.134				
VIW	1990/91	20,438	0.271				
	1995/96	48,206	0.177				
VIE	1990/91	29,928	0.308				
	2000/01	25,683	0.199				

Based on information from JARPA and JARPAII two stock scenarios were examined: i) a base-case where two stocks occur in the research area, one distributed west of 130°E (I stock) and the other east of that longitude (P stock), and ii) a sensitivity test where two stocks occur in the research area (I and P stocks) which mix to each other in Area VW with a proportion of the I stock of 55% (Pastene and Kanda, 2005). Catches are made to the west of 130°E in the first year (austral summer season 2015/16), and to the east of that longitude in the second year (austral summer season 2016/17). Subsequently future catches are conducted in each of the sectors every two years alternately (therefore figures obtained by the CLA should be compared with sample size n= 333/2). Calculated catch limits by the CLA for the two sectors (stocks) are shown in Table 4 for both base case and sensitivity case. Under both cases the catch limit for I and P stocks are much larger than the sample size in this proposed research plan. Calculations by the CLA were also made for half of the Management Areas (Table 5). Total catch limits for Areas IIIE, IVW and IVE combined (I stock in the base case) is 571 while that for the combined Areas VW, VE and VIW (P stock in the base case) is 3,169. For all cases examined the calculated catch limits were larger than the proposed sample size.

Table 4. Catch limits obtained by the CLA for I and P stocks under the base case (left) and the sensitivity case (right)

Istock	Pstock	Total	Istock	Pstock	Total
739	3,153	3,892	1,260	2,857	4,117

Table 5. Catch limits obtained by the CLA by half Management Area

IIIE	IVW	IVE	Sub-total	VW	VE	VIW	Sub-total	Total
202	186	183	571	614	2,142	413	3,169	3,740

Because CLA provides catch limits that would not threaten the Antarctic minke whale stock for 100 years, which is much longer period than the proposed research period, an alternative approach to evaluate the effect of the catches on the stock was required. For this aim the Hitter-Fitter population dynamics model was used. The same stock scenarios in the calculation of the CLA above, were used (base case and sensitivity case). A constant carrying capacity was assumed. In the Fitter runs, some biological parameters were used: MSY level of 0.6, ASM 7.5 (50%) and 10 (95%). 50% ASM of 7.5 is assumed because 50% ASM for recent cohort ranges 7-8 years in both stock (Bando *et al.*, 2014). It is assumed natural mortality for the I stock for ages <=3, 10-20 and >=40 are 0.077yr-1, 0.048yr-1, and

0.107 respectively and the corresponding values for the P stock are 0.074yr-1, 0.046yr-1, and 0.103 yr-1 respectively and that natural mortality is assumed to vary linearly with age between ages 3 and 10, and between ages 20 and 40 (Punt, 2014c). Using a fixed MSYR (1+) option of the Hitter-Fitter program, runs were conducted for MSYR (1+)=1% and 4% and for agreed abundance estimates and historical catches. Abundance estimates to be fitted to the population model for the base case stock scenario are 58,545 (CV=0.161) in 1988/89 and 90,532 (CV=0.229) for I stock and 281,047 (CV=0.122) in 1987/88 and 228,303 (CV=0.092) in 2000/01 for P stock. Those for the sensitivity scenario are 116,818 (CV=0.113) in 1987/88 and 114,534 (CV=0.183) in 1998/99 for I stock and 222,774 (CV=0.138) in 1987/88 and 204,301 (CV=0.100) for P stock based on the abundance estimates in IWC (2013). The effect on the stocks was evaluated by examining the population trajectory for 1+ component of both stocks. Additionally two sensitivities were considered regarding future catches for the base case scenario: zero catch and double of the proposed annual sample size (n=666). Therefore four scenarios were considered for each stock and each MSYR.

Figure 4 shows the population trajectory for the I and P stocks, respectively, for MSYR (1+)=1% and 4%, under the four scenarios. The trajectory for I stock shows an increasing trend after the 2015/16 for all scenarios examined (Figure 4 left). In the case of the P stock, trajectory remains constant around the pre-exploitation level (Figure 4 right).

In conclusion both approaches used, CLA and Hitter-Fitter, suggest no negative effect on the stocks of the proposed annual sample size of the new research program.

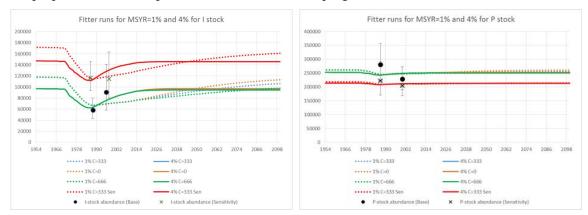


Figure 4. Population trajectory for the I (left) and P (right) stocks for MSYR (1+)= 1% and 4% under four scenarios. Vertical lines indicate 95%CI of abundance estimate.

The effect of catches on the stocks was also evaluated for future catches in Areas IIIW and VIE. For this evaluation and with regard to I and P stocks, the same base and sensitivity cases in the previous calculations were used. The stock structure in Areas IIIW and VIE is unknown. For the effect of the calculations, the most conservative scenario was considered *e.g.* the occurrence in Area IIIW of a different stock (no occurrence of I stock), and the occurrence in Area VIE of a different stock (no occurrence of the P stock). Abundance estimates derived from IDCR/SOWER were employed in the estimation. The estimates in Area IIIW used were 69,267 (CV=0.277) in 1987/88 and 33,375 (CV=0.209) in 1994/95. Those in Area VIE were 29,928 (CV=0.308) in 1990/91 and 25,683 (CV=0.199) in 2000/01. Whales are sampled one every two years in the sectors west (occupied mainly by I stock) and east (occupied mainly by P stock) of 130°E, respectively. In the western sector, catches among sectors are distributed as follow: IIIW: 90; IIIE: 90; IVW: 77 and IVE: 76 (total: 333). In the eastern sector catches are distributed as follow: VW: 106; VE: 76; VIW: 76 and VIE: 75 (total: 333). Figures change according the wide of the longitudinal sector.

Catch limit obtained by the CLA in Area IIIW was 182 which is larger than the level of catch (90/2) in that Area. Catch limits obtained by the CLA in Area VIE was 259 which is larger than the level of catch (75/2) in that Area.

For the Hitter-Fitter analysis the same additional sensitivities described in the previous calculations were used. Figure 5 show the population trajectories for the population in Areas IIIW and VIE. In both cases the trajectories are stable or show an increase on the time.

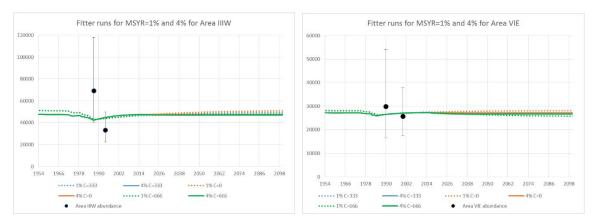


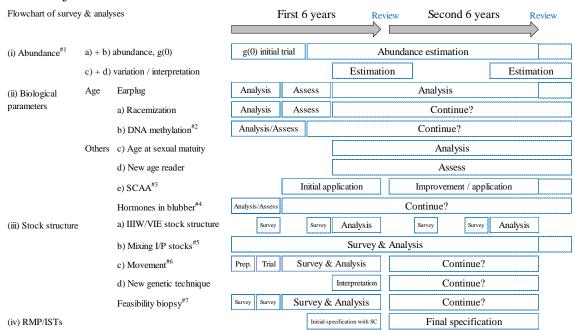
Figure 5. Population trajectory for Areas IIIW (left) and VIE (right) for MSYR (1+)= 1% and 4% under three scenarios. Vertical lines indicate 95% CI of abundance estimate.

Even under the conservative scenario used, the catch of the future research has no negative effects on the stocks in Areas IIIW and VIE.

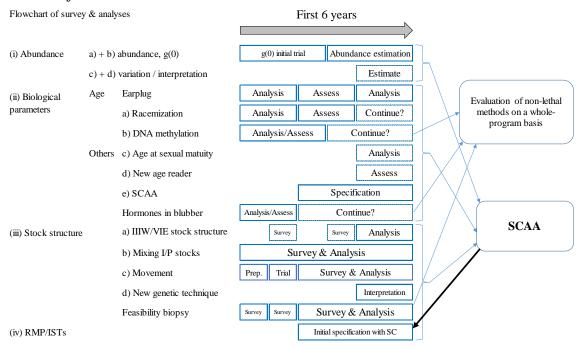
6. TIMELINE OF RESEARCH ACTIVITY

The chart below shows the kinds of research activities and associated timeline under the NEWREP-A's Main Objectives I and II. For both of the Main Objectives, the upper part of the figures shows the timeline for the research activities in the entire 12-year research period. The lower part of the figures shows the timelines for the first six-year period, specifying the outputs and evaluation to be obtained after the first six-year period. Details are explained in the footnotes to the charts.

Main Objective I



Main Objective I



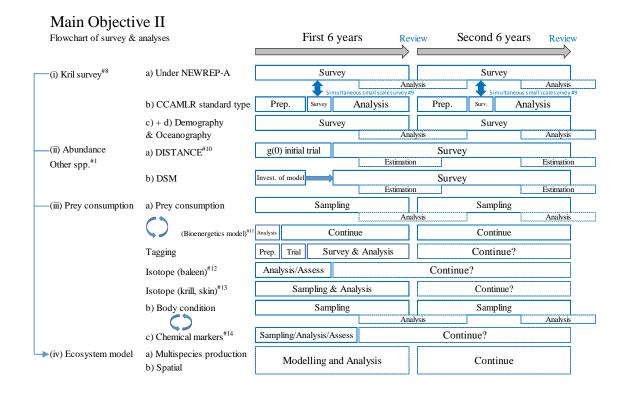
The survey plan for each austral summer season including details of the survey design, will be presented to the annual IWC SC meetings. The survey design will take into account the specific research activities planned for a particular Area and austral summer season, and will follow the seven guidelines or suggestions provided by the Review Panel regarding survey design. The survey design for the first two-three years will include feasibility studies on biopsy sampling and telemetry of Antarctic minke whale. The survey plan for the 2015/16 austral summer season was presented to the 2015 IWC SC meeting as Document SC/66a/SP08.

^{#2} At first, the stock effect on the DNA methylation technique will be examined using 50 highly readable earplugs each from I and P-stocks. In the next step the effect of the period will be examined by using similar number of samples from different austral summers. A specialized laboratory and a university in Tokyo have been contacted so that this study can be conducted in collaboration with experts. Preliminary results will be reported to the 2017 IWC SC meeting and final results at the 2018 IWC SC meeting.

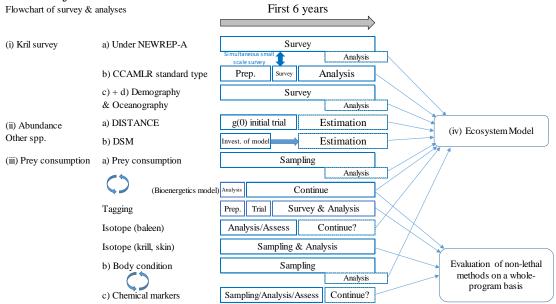
^{#3} The proponent responded preliminarily to several SCAA/RMP-related recommendations from the NEWREP-A Review Panel at the 2015

IWC SC meeting (SC/66a/SP08).

- The proponent will conduct progesterone analyses in blubber tissues of Antarctic minke whales using JARPAII samples. Females with various reproductive status such as ovulating, resting and pregnant with small fetus will be selected, and hormone levels will be compared with histological information of reproductive organ. Analysis of progesterone level for a total of 50 Antarctic minke whale samples will be assayed using the compact ELISA workstation (Crocodile, Titertek Berthold). Progesterone will be assayed with the Cayman Practice ELISA Kits (Cayman Chemical), according to the manufacturers' protocols. Preliminary results will be reported to the 2017 IWC SC meeting.
- #5 The estimation of performance will consider the use of a random effect model for the mixing proportions, by using a statistical principle of "borrowing strength".
- #6 The feasibility study on telemetry in Antarctic minke whale will be undertaken along the dedicated sighting surveys, in the early stage of the first six-year period of NEWREP-A. Detailed study design will be provided in the sighting survey plan to be presented and discussed at the annual IWC SC meetings. With regard field methods as well as tag types, the proponent will collaborate and consult with experienced foreign and Japanese colleagues (see details in the main text).
- #7 The feasibility study on biopsy sampling in Antarctic minke whale will be undertaken along the dedicated sighting surveys, in the early stage of the first six-year period of NEWREP-A. Because this feasibility should be conducted early in the program, it will be conducted in the IWC Area to be covered by the sighting surveys in 2015/16, 2016/17 and 2017/18. Detailed study design will be provided in the sighting survey plan to be presented and discussed at the annual IWC SC meetings (see details in the main text).



Main Objective II



- #8 Since the NEWREP-A review workshop, progress has been made on the installation of equipments (echosounder, Norpac net and CTD) in the research vessel that will be engaged in krill surveys under NEWREP-A. During this work the specifications made by the Review Panel for those instruments are being taken into consideration.
- #9 The proponent will consider to conduct a simultaneous survey by a krill survey vessel equipped with a frame-type trawl and echosounder, the dedicated sighting vessel equipped with an echosounder and the whale sampling vessels within a small-scale experimental survey area over a short time period (e.g. one week) so that comparisons among vessels/samples can be made.
- **10 Identification of duplicate sightings will be also conducted for whale species other than Antarctic minke whale during NEWREP-A sighting surveys in IO mode. To determine whether $g(\theta)$ is significantly different from one, analyses on duplicate sightings will be conducted for those whale species where sufficient data are obtained during the NEWREP-A program. Details will be provided in the sighting survey plans to be presented to the annual meetings of the IWC SC.
- plans to be presented to the annual meetings of the IWC SC.

 #11 The proponent will make effort to build more sophisticated models adding some categories such as age, growth and maturity status based on the energy requirement by change in blubber mass and lipid contents. Previously and newly collected data will be used. Preliminary results of this work will be reported to the 2016 IWC SC meeting (see details in the main text).
- #12 Stable isotopes analyses (δ13C and δ15N) along the edge of baleen plates will be conducted to estimate time duration of Antarctic minke whale in the feeding grounds. Stable isotopes in baleen plates (n=10-20) of Antarctic minke whales sampled under JARPA II will be analysed early in the research program, as an initial step. The results of the analyses will be reported to the IWC SC in 2018.
- As a first step in the work to compare overlap in diet amongst fin and Antarctic minke whale using stable isotope, krill samples will be analyzed to ensure the correct determination of stable isotope baselines. This will be done during the first six-year period of NEWREP-A. Concurrently, the proponent will examine the overlap in diet amongst fin, humpback and Antarctic minke whales using stable isotope ratios in skin samples collected by lethal method (Antarctic minke whales) and by biopsy sampling (fin and humpback whales). This analysis will be carried out early in the NEWREP-A program based on the information accumulated on stable isotope baselines of krill in each area. Results will be presented to the IWC SC meeting as appropriate in the first six-year period and evaluated at the mid-term review of NEWREP-A (see details in the main text).
- **14 At first, the total lipid content % of blubber will be compared to other body condition indicators such as blubber thickness and girth measurements. As an initial feasibility test, the measurements of body conditions (blubber thickness, girth, total fat weight and the total lipid content % of blubber) from Antarctic minke whales collected under NEWREP-A will be used. At least 50 whales for each sex and sub areas will be used or this analysis.

7. BACKUP PLAN FOR CONTINGENCIES

As research activities could be disrupted by both natural and human factors including dangerous sabotage activities by an extreme anti-whaling NGO, this proposed research plan establishes a contingency backup plan in order to respond to the contingency and secure the scientific value of data to be collected by this research for the purpose of achieving the stated scientific objectives.

The backup plan would address three aspects; (i) adjustments of research protocols at the scene of disruption, (ii) adjustment of research plans including research period, sample size, and research areas after the year of disruption, and (iii) consideration of analysis methods to compensate the effects of disruptions. Possible issues arising from a disrupted research plan could be reduced sample size, unsurveyed research areas, sampling bias and missed research periods. They could mean reduced accuracies, reduced randomness, and other increased biases. The considerations of these aspects also

responds to the recommendation made at the Expert Workshop to Review the Japanese JARPA II Special Permit Research Programme, held in Tokyo, Japan, February 24-28, 2014 (see section 12.1.2 in IWC, 2014a).

(i) Adjustments of research protocols at the scene and in the year of disruption

In case of a disruption, including sabotage activities, the head of the research (*e.g.* the cruise leader) will declare to the public that lethal parts of the proposed research will be immediately suspended. Non-lethal research activities will continue as planned. If continuation of non-lethal research activities is also disrupted, the whole research will be suspended and could be relocated to other research areas. These adjustments including resumption of the research in other areas and periods than originally planned will be fully recorded and made available for the analysis required in establishing adjusted research plans and in conducting scientific considerations for compensating the effects of the disruption.

(ii) Adjustment of research plans including research period, sample size, and research areas after the year of disruption

When the number of samples does not reach a target sample size, the reduced number could be carried over to the latter years of the research plan and/or the whole research period could be extended when scientific justification based on the careful analyses of the data collected is established in consultation with the IWC SC. When this happens, effects on the sampled stock(s) will be reassessed.

Missed research periods and unsurveyed areas could also require adjustment proposed research plans. Scientific justification for the need of adjustment research plans regarding period and areas will be provided along with pertinent data and its analysis to the IWC SC. Such an adjustment research plan will be established immediately after the completion of the research of the year of disruption.

A summary information on the progress of the research, on an annual basis, in terms of samples obtained and areas and periods covered will be made public. This summary provides transparency in the implementation of the proposed research and background for adjustment research plans if they become necessary.

(iii) Consideration of analysis methods to compensate the effects of disruptions

Effects of disruptions will be analyzed in terms of reliability of scientific data obtained. This would allow the consideration of analysis methods to compensate the effects of disruptions. One of the potential problems likely caused by the disruptions is spatially and temporally imbalanced sampling outcome. Like in the case of discussion in the abundance estimation, lack of design-unbiasedness may be overcome by introducing the model-based estimation approaches, where finding better spatial and environmental covariates is a crucial part for its success. So far, although such heterogeneity has been observed to a certain degree in the JARPA and JARPAII, there is an achievement to treat such effects through statistical methods such as random- or mixed-effect models (Konishi and Butterworth, 2013).

It is also important for the analysis that the adjustments described above in (i) and (ii) are well-planned and executed. However, as the effects of the disruptions could vary substantially, a case-by-case approach might be unavoidable. A high level of transparency will be necessary for the analytical methods employed to address the effects of the disruptions.

8. OUTPUT OF THE RESEARCH, PARTICIPATION OF FOREIGN SCIENTISTS AND COLLABORATION WITH OTHER RESEARCHES/ORGANIZATIONS

This proposed research program is primarily intended to contribute to the improvement of conservation and management of whale resources in the Antarctic in accordance with the ICRW as shown in its research objectives. It follows that its scientific output will be produced first and primarily for the review and discussion in the IWC SC. At the same time, in line with an observation by the ICJ, there will be increased efforts to publish its scientific achievements in peer-reviewed journals outside the IWC

Participation of foreign scientists in the field surveys will be welcomed, so long as they meet the following qualifications established by the Government of Japan: i) costs for participation, travel expenses to and from the home ports, subsistence on board the research vessels and any other ancillary cost, will be covered by the participant; ii) indemnification and insurance for any casualty or personal injury while on board the research vessels, will be covered by the participant; iii) participation of those who are found to have intentionally sabotaged the implementation of the research in the field shall be terminated.

Research collaboration will be sought with relevant scientists and research institutions (national and international), in consideration of the objectives of the proposed research. In the case of Main Objective I, namely "Improvement in the precision of stock assessment and ecological information for the application of the RMP to the Antarctic minke whales and calculation of its catch limits", and its subobjectives, collaboration will be made with scientists of the IWC SC, which have been heavily involved with this type of research in the past, and with scientists from national research institutions such as the Tokyo University of Marine Science and Technology, National Research Institute of Far Seas Fisheries, National Research Institute of Fisheries Science among others. On Main Objective II, namely "Investigation of the structure and dynamics of the Antarctic marine ecosystem through building ecosystem models", and its sub-objectives, collaboration will be made with national and international research institutions involved in the research of the Antarctic ecosystem. As noted earlier, design and implementation of the krill survey will be carried out in collaboration with CCAMLR scientists. Analysis of data will be carried out in collaboration and consultation with scientists from CCAMLR, Tokyo University of Marine Science and Technology, National Research Institute of Far Seas Fisheries, National Research Institute of Fisheries Science, National Institute of Polar Research in Japan among others.

As in the case of JARPA and JARPAII, data obtained by this new proposed research program will be shared with members of the IWC SC in accordance with the IWC SC Data Availability Agreement (DAA) (IWC, 2004). Data and samples from the research will be available for other accredited scientists and research organizations in accordance with the protocol for access to samples/data from the ICR (http://www.icrwhale.org/pdf/appendix2.pdf).

To facilitate the collaboration and analyses, databases will be created after each survey under this program conducted by the ICR, which will specify the kind of samples and data collected during its research activities. Databases which include data from laboratory work will be created in time for the mid-term review of the research program in line with the Annex P (IWC, 2013b). According to the Annex P the proponent of the research program should submit a preliminary data description document that explains the data two months before the Annual IWC SC meeting prior to the specialist workshop that will review the proposed program. It also specifies that the final data description document and the data themselves will be made available in electronic format one month after the close of the Annual IWC SC Meeting. The mid-term review is expected after the completion of the first six-year period of the proposed research plan.

In addition, efforts will be made to respond the NEWREP-A Review Panel specific recommendations regarding research collaboration: (1) *ad hoc* collaborations on specific issues; (2) the development of a formal protocol for outside scientists to express interest; and (3) the development of a strategy to promote incorporation of external Japanese and/or foreign scientists into the research (IWC, 2015a pp41). Regarding the recommendation (1), NEWREP-A will continue contacting potential partners, and co-operative research will be identified and arranged for specific items in due course, preferably before research activities on such items start. As for the recommendation (2), NEWREP-A will develop and post such a protocol on the website of the Institute of Cetacean Research (ICR) (both in Japanese and English) well in advance of the first research cruise under NEWREP-A. With respect to the recommendation (3), NEWREP-A will: i) continue contacting specific potential collaborators directly; and ii) invite co-operative research widely through the internet (e.g. ICR's website) and at related meetings (including those of the IWC SC) indicating specific research items for collaboration. An "annual progress report" to the IWC SC (see the next item), would also help to promote research collaboration with external scientists/organizations by including description on as the involvement of

external scientists/organizations and specification of their roles in the report.

With regard to collaboration with CCAMLR, the outline of the krill surveys plan were already presented and discussed at the WG-EMM held in July 2015. Following the recommendations by the WG-EMM15, data and details of a plan of the whale dedicated sighting vessel-based krill survey to the next SG-ASAM and WG-SAM in 2016.

9. PROGRAM MANAGEMENT, PERSONNEL AND LOGISTIC RESOURCES

As for the whole program management, the Fisheries Agency of Japan (FAJ) will assume responsibility, including provision of the necessary funding. Regarding personnel resources to be engaged in the program, scientists from the ICR will play the leading role in order to pursue research activities and achieve the research objectives of NEWREP-A in collaboration with scientists from other domestic and/or foreign organizations, such as the Tokyo University of Marine Science and Technology, the National Research Institute of Far Seas Fisheries and the National Research Institute of Fisheries Science. In addition to collaboration with CCAMLR, further collaboration with relevant oversea researchers and research institutions will be sought by ICR.

The proponent intend to provide relevant information concerning program management to the IWC SC every year in an annual progress report for the SC's comments and suggestions to facilitate steady progress of the program in an open and transparent manner. This information will include progress achieved for each sub-objective as well as the involvement of external scientists/organizations and their roles. This annual update and comment process through the IWC SC is expected to improve the management of the program throughout its duration.

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Process for development of this research plan

1. Background --- "scientific research" under the framework of the ICRW and other international conventions/agreements concerning conservation and management of marine living resources

The International Convention for the Regulation of Whaling (ICRW) prescribes that amendments of the Schedule, by adopting regulations with respect to the conservation and utilization of whale resources, shall be based on scientific findings (Article V, paragraph 2 (b)). Therefore, the International Whaling Commission (IWC), established under the ICRW, shall use "scientific findings" as the fundamental basis in adopting conservation and management measures for whale resources concerned, and its decisions depend on research programs with clear objectives of providing such "scientific findings". In this regard, it should be noted that, among the three broad categories of objectives for special permit research proposals under Article VIII of the ICRW, which are identified in the "Annex P" review process adopted by the IWC, the first one is "[to] improve the conservation and management of whale stocks" (IWC 2014).

The principle of "science-based conservation and management" is not unique to the ICRW/IWC, but is common in regional fisheries management organizations (RFMOs) established under international conventions/agreements¹, which are responsible for conservation and management of marine living resources. It is also a common practice that research programs to provide such "scientific findings", are planned and implemented by research institutions (of member governments in most cases) which are directly responsible for research concerning the conservation and management of marine living resources concerned, on their own initiatives, in consultation or collaboration with the competent RFMOs.

2. Nature of this scientific research program

This research plan is developed as a scientific research program of which main objectives, includes possible contribution to improving the conservation and management of whale resources under the framework of the ICRW. In other words, this special permit research program intends to provide "scientific findings" mentioned in Article V, paragraph 2 (b) of the ICRW, with a view primarily to providing the basis for amending the Schedule to the ICRW and improving the Revised Management Procedure used to calculate sustainable catch limits for whale stocks, and thus resuming commercial whaling in a sustainable manner in the Antarctic Ocean in accordance with the ICRW. This proposed research program can be characterized as a program of "scientific research" under the framework of the ICRW, in light of the text of the ICRW and the "Annex P" review process, while it is not intended to be a research program for purely academic purposes.

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¹ *e.g.* International Convention for the Conservation of Atlantic Tunas ("The Commission may, on the basis of scientific evidence, make recommendations designed to maintain the populations of tuna and tuna-like fishes that may be taken in the Convention area at the levels which will permit the maximum sustainable catch" (Article VIII, paragraph 1.(a))); Convention on the Conservation of Antarctic Marine Living Resources ("[the Commission shall] formulate, adopt and revise conservation measures on the basis of the best scientific evidence available, subject to the provision of paragraph 5 of this Article" (Article IX, 1.(f))); and Convention on the Conservation and Management of High Seas Fisheries Resources in the North Pacific Ocean ("[In giving effect to the objective of this Convention, the following actions shall be taken individually or collectively as appropriate:] adopting measures, based on the best scientific information available, to ensure that fisheries resources are maintained at or restored to levels capable of producing maximum sustainable yield, taking into account fishing patterns, the interdependence of stocks and any generally recommended international minimum standards, whether subregional, regional or global" (Article 3, (b)(adopted in February 2012, but yet to enter into force as of October 2014))).

3. The judgment of the International Court of Justice (ICJ) and Japan's response

The judgment delivered by the ICJ on March 31, 2014 in the case concerning *Whaling in the Antarctic* (Australia ν . Japan: New Zealand intervening) states that the decision to grant special permits under Article VIII, paragraph 1 of the ICRW "cannot depend simply on that State's perception" (paragraph 61). Japan has taken this finding of the ICJ into consideration, and with respect to the process for development of this proposed research plan and it is determined to "follow an internationally open and highly transparent process through securing the participation of renowned scientists from Japan and abroad, and through other processes including discussion at the IWC Scientific Committee's workshop and coordination with other institutions conducting relevant studies". This was announced on April 18, 2014 by the then Minister of Agriculture, Forestry and Fisheries in a policy statement concerning the future whale research programs.

Following the Minister's statement, and taking account of the observations described in 1 and 2 above, Japan decided to have first and second drafts of the new research plan reviewed by external scientists with a wide range of expertise to get comments and suggestions before it is submitted to the IWC Scientific Committee (i.e. in addition to the official review process in accordance with the "Annex P"). Specifically, it was planned to go through a "voluntary" review process open to external experts to obtain valuable inputs, including constructive criticisms, for improving the draft research plan through: 1) e-mail correspondence in early September; and 2) a meeting held in Tokyo, Japan in October.

4. Chronology of the actual process for development of this research plan

On August 29, 2014, approximately 160 external experts in the following categories were invited to participate in Japan's "voluntary" review process of the new research plan, by an e-mail sent from Japanese scientists participating in the IWC Scientific Committee:

- ✓ Members of the IWC Scientific Committee (Participants in the 2014 meeting of the Committee and other regular participants in the past);
- ✓ External expert panel members for the past special permit research reviews held by the IWC Scientific Committee;
- ✓ External expert reviewers for the 2004 IWC review of the Southern Ocean Sanctuary;
- ✓ Experts who appeared in the ICJ case;
- ✓ Chair of the Scientific Committee of the Commission for the Conservation of Antarctic Marine Living Resources (SC-CAMLR) and conveners of its related working groups;
- ✓ Chairs of scientific subsidiary bodies (and panel members) of RFMOs which covers the waters adjacent to the Antarctic Ocean (i.e. CCSBT, SPRFMO, SEAFO and ICCAT);
- ✓ Director and ex-Directors of Fisheries Resources Management and Research Division, Food and Agriculture Organization of the United Nations (FAO); and
- ✓ Other experts who have good insights into the biology of marine mammals, the Antarctic marine ecosystem and the conservation or sustainable use of marine living resources.

By early September, ten (10) external experts from nine (9) countries (including one (1) from Japan) had confirmed their interest in reviewing (or in just receiving) the initial draft of the proposed research plan. One (1) external expert confirmed his interest in receiving the draft at the beginning of October. Consequently, eleven (11) experts from ten (10) countries expressed his/her interest in the new research plan at that stage. On the other hand, three (3) experts from two (2) countries expressed their view that it was not appropriate to participate in the review process mentioning the implication of the ICJ Judgment to the special permit whaling under the ICRW etc.

On September 6, 2014, the first draft of the new research plan was sent by e-mail to the ten (10) external experts who had confirmed his/her interest in the new research plan by that time. The first draft was also sent to the other expert in early October by e-mail.

By late September, comments on the first draft had been received from six (6) experts. One (1) expert indicated that he had no comment on the first draft. The received comments were considered by the proponent and taken into account as much as possible in developing the second draft of the research plan.

On October 22, 2014, a meeting to review and discuss the second draft was held in Tokyo, Japan with the participation of three (3) external experts from three (3) countries and one (1) local scientist. On the following day, the second draft was sent out by e-mail to all external experts participating in the process, including three (3) additional experts who had agreed by the middle of October to review the second draft, for their review and comments. In total, fourteen (14) external experts from eleven (11) countries (including three (3) from Japan) received the second draft, and four (4) experts (including two (2) experts who attended the meeting on October 22) provided their comments on it by early November.

The comments provided at the meeting and through the e-mail correspondence were considered by the proponent and taken into account as much as possible in developing the final draft of the research plan.

5. The Transparency of the whole process

The process applied for developing this proposed research plan is intended to be more transparent compared with scientific research programs conducted for marine living resources under the competence of other RFMOs, which involve lethal sampling of such resources. Most of those research programs do not usually go through a review process open to external experts when they are being developed, especially in the initial stages of their development.

Given the nature of scientific discussions under this proposed plan, this process was only open to external experts who have expertise in the specific related fields of science. This is considered as appropriate in order to secure and facilitate objective and science-based discussion. More open opportunities for discussing the plan are secured in the later stage of the whole review process, i.e. after the proposed research plan is submitted to the IWC Scientific Committee, including its review process in accordance with the "Annex P." Japan plans to have this proposed research plan circulated by the IWC Secretariat to all Contracting Governments to the ICRW, for their review and comments in order to improve and then finalize the substance of the proposed plan so that the research program can contribute more effectively to improving the conservation and management of whale resources in the Antarctic.

Reference

International Whaling Commission 2014. Report of the Scientific Committee. Annex P. Process for the Review of Special Permit Proposals and Research Results from Existing and Completed Permits.

Clarification of research objectives in the proposed research plan

The previous JARPAII program had four main research objectives: 1) Monitoring of the Antarctic ecosystem, which involved several research items; 2) Modeling competition among whale species; 3) Elucidation of temporal and spatial changes in stock structure; and 4) Improving the management procedure for Antarctic minke whale. The relationship among these four JARPAII objectives, is shown in the figure below.

OBJECTIVES OF PREVIOUS JARPAII I. Monitoring of the Antarctic ecosystem · Whale abundance trends IV. Improving the management procedure for Antarctic minke (1) (2) (4) · Biological parameters trends Recruitment trend Stomach content weight and body condition trends Prev consumption rate trend · Krill biomas (1) Abundance (2) MSYR (*ISTs*) (3) Redefinition of appropriate management Areas Cetacean habitat (oceanography, marine debris) Contribution for future improvement of RMP II. Modeling competition among whale (4) MSYR (CLA algorithm) (5) Incorporation of effects arising from the inter species relationships among the whale species Constructing a model of competition among whale species III. Elucidation of temporal and Defining new management objectives spatial changes in stock structure including the restoration of the cetacean · Antarctic minke whale · Other large whale species

In JARPAII, several parameters considered 'indicators' of changes in the ecosystem were studied and monitored under the Objective I, and several of those parameters were used as input data for the developing of ecosystem models focusing competition among whale species (Objective II), *e.g.* whale abundance, krill biomass, stomach contents and body condition of whales; and for improving the management of Antarctic minke whale (Objective IV) *e.g.* biological parameters such as the MSYR and natural mortality. Elucidation of the stock structure of this species stands as a separate objective (Objective III), which would contribute to improving the management of the species under the RMP (Objective IV). Results of the modeling work under Objective II, would also be used to incorporate the effect arising from the inter-species relationship among whale species into the RMP (Objective IV). Therefore Objective IV depended on the results obtained under the other three objectives.

The four research objectives of JARPAII were not judged as unreasonable by the ICJ. They remain relevant and have been taken into account in the development of this proposed plan.

The two main research objectives of the new research plan are, 1) Improvements in the precision of biological and ecological information for the application of the RMP to the Antarctic minke whales to calculate catch limits; and 2) Investigation of the structure and dynamics of the Antarctic marine ecosystem through building ecosystem models. Sub-objectives were explained in section 2 of the main text.

The previous JARPAII research objectives and items are incorporated into the two objectives of the new plan through the establishment of sub-objectives.

The two main objectives of the new research plan are directly relevant for the conservation and sustainable use of marine living resources, especially whales, in the Antarctic.

On the first main objective: The RMP is the rigorously-tested mechanism that the IWC SC has developed to allow it to provide advice on safe, risk-averse catch limits for commercial whaling of baleen whales, should the Commission ever decide to reinstate such whaling. The RMP comprises all of the rules and data requirements associated with the provision of advice for particular species in particular geographical areas. The RMP was adopted by the Commission in 1994. Because of the moratorium on commercial whaling the RMP has not been applied, with the exception of operations carried out by Norway and Iceland that have lodged an objection or reservation to the commercial whaling moratorium. However the IWC SC continues to work on several issues on how the RMP could be implemented. Specifically every year, the IWC SC discusses general issues of the RMP as well as *Implementation*-related matters. On the latter, the IWC SC has carried out RMP *Implementations* on western North Pacific minke and Bryde's whales and North Atlantic minke and fin whales, each of which took at least two years and involved a huge amount of scientific endeavor. In summary, the RMP is a powerful and important management tool developed by the IWC SC.

On the second main objective. Ecosystem modeling is important not only to investigate and understand the structure and dynamics of the Antarctic ecosystem but also it can be useful for the objectives of conservation and management. There is a growing disillusion with the predictive capacity of single species assessment methods and the management approaches they support, and this has been the primary driver for the development of ecosystem-based approaches. The principle of multi-species management has been discussed, accepted, adopted, and recommended by many international organizations or fora, including the Food and Agriculture Organization of the United Nations (FAO, 2003) and the 2002 World Summit on Sustainable Development (WSSD, 2002). Recognizing the importance of the ecosystem approach, the IWC SC discussed multi-species management approaches under its Standing Group on Environmental Concerns until 2006. Since 2007 a Working Group on Ecosystem Modeling has been discussing prototype ecosystem models. On the other hand the CCAMLR have been developing ecosystem models to evaluate management strategies for the Antarctic krill fisheries. The importance that both organizations (IWC and CCAMLR) give to ecosystem modeling is reflected in a joint workshop on this topic conducted in 2008, and new joint workshops are being programmed for the near future. The recent workshop to review JARPAII research strongly encouraged further work on ecosystem modeling.

The new research plan is, therefore, based on two objectives that reflect research need amply recognized by international organizations in charge of conservation and management of marine living resources. The new Antarctic research plan integrates the previous JARPAII research objectives and research items into these two main objectives, in a coherent and more sensible way. For example the stock structure of Antarctic minke whale did stand as a separate research objective in JARPAII. In the new Antarctic research plan it became a sub-objective under Objective I which make more sense since the stock structure of Antarctic minke whale is conducted for- and is essential information of- the RMP *Implementation*.

On the other hand this new structure will facilitate the understanding and evaluation by the IWC SC because objectives and sub-objectives are specified, as well the time frame for producing results within them.

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Objective I(i)

Abundance estimates for Antarctic minke whale taking account of g(0) and additional variance estimate

1. Background and objectives

Absolute abundance estimates by direct methods such as sighting surveys and their precision are required for catch limit calculations under the Revised Management Procedure (RMP) (IWC, 2012a). In addition to the requirements, data and methods for analyzing them that are used in the application of the RMP should meet the minimum standards described in Requirements and Guidelines for Conducting Surveys and Analyzing Data within the Revised Management Scheme (IWC, 2012a).

There have been two sources of abundance estimates based on sighting data and the Line Transect Method in the Antarctic, IDCR/SOWER (1978/79-2009/10) (Matsuoka *et al.*, 2003) and JARPA/JARPA II (1987/88-2013/14). Based on the former, Antarctic minke whale abundance estimates that took account of *g*(0) estimates were obtained for CPII (1985/86-1990/91) and CPIII (1991/92-2003/04) (Okamura and Kitakado, 2012). These estimates were agreed by the IWC/SC (IWC, 2013). As for the latter, abundance estimates for Areas IV and V were obtained and the results accepted by the JARPAII review workshop. Dedicated sighting surveys during JARPAII have been subject to oversight since 2010/11 but the survey could not be conducted due to external violent interference by an anti-whaling group. The most recent estimates for Antarctic minke whales in Areas IV and V by JARPA/JARPAII cannot be used for RMP since the phase-out rule would be applied because ten years have passed since the year of the most recent abundance estimate (IWC, 2012a).

In order to attain Objective I(i), four sub-objectives are set: i) estimate g(0) based on sighting data collected during the surveys, ii) estimate abundance during JARPA/JARPAII considering the g(0) estimates, iii) estimate inter-annual variation (additional variance), and iv) interpret and understand time series of abundance estimate for Antarctic minke whales.

2. Methods

2.1 Survey methods

Sighting surveys are planned to be conducted by the Line Transect Method following the survey protocols set out in the Requirements and Guidelines for Conducting Surveys and Analyzing Data within the Revised Management Scheme (IWC, 2012b) so that sighting surveys are conducted under oversight of IWC/SC. Sighting protocols are the same as those used in the IDCR/SOWER surveys (Matsuoka *et al.*, 2003). Survey areas are planned to cover one IWC Management Area (one of Areas III, IV, V and VI) in a year. It is planned to survey in Areas III and VI twice and Areas IV and V three times in twelve years. In other two years, ecological survey and sighting survey in waters north of 60°S will be conducted. In this way, it is planned to produce abundance estimates in these Management Areas every six years which can be used in application of RMP for the Antarctic minke whale. In case that sighting surveys cannot be conducted due to external violent interference, there would be possibility that the survey area will be changed from original plan.

Survey mode

The sighting survey will be conducted using (1) Closing mode (NSC) and (2) Passing with Independent Observer (IO) mode. Both survey modes follow the protocol endorsed for the IWC/SOWER surveys. Passing with IO mode are to be conducted as follows. Two topmen observe from the barrel at all times and a third topman is stationed in the independent observer platform (IOP). The topmen report information to upper bridge observers but no information exchange between the barrel and the IOP (Matsuoka *et al.*, 2003). In closing mode, vessels approaches to detected school to confirm its species and school size and therefore observed school size data are more reliable that those obtained during IO

mode. By adopting both closing and passing with IO mode, the merits of both surveys for abundance estimation of Antarctic minke whales are utilized.

Tracking experiment

In order to obtain further information on dive time of the minke whales, tagging methods for data loggers (TDR) and satellite tagging transmitters will be developed. These tagging methods will be also used for ecological surveys (Objective 2(iii)). Observation experiment could be a candidate method for this purpose. Mean dive-time was a key parameter for estimation of g(0) using the OK model as the case of abundance estimate of the minke whales based on IDCR/SOWER (IWC, 2013). Mean dive-time estimate and its precision could be improved by the further information on dive time of Antarctic minke whales.

2.2 Analytical methods

Abundance estimation

The OK model (Okamura and Kitakado, 2012) is applied to estimate abundance taking g(0) estimate into account using sighting data obtained in closing and IO mode. The OK model can examine the effect of covariates such as school size estimate and weather condition on the g(0) estimate. Detection probability function is modeled using Hazard probability model (Schweder *et al.*, 1997) and inter-cue interval. The OK model will be applied treating inter-cue interval as an input parameter. In case that mean dive time should not be obtained successfully by the tracking experiment, it could be estimated using OK model.

Correction of previous abundance estimates

In OK model, detection probability function can be modeled with covariates such as perpendicular and forward distance, school size and weather conditions, which are available for JARPA and JARPA II data. Applying the model above using data obtained by sighting vessels (SVs) in this survey, g(0) can be estimated that considers the effect of the covariates for each sightings of the Antarctic minke whale. This use of g(0) can improve the abundance estimates during JARPA and JARPA II period from those using the *ad hoc* approach in Hakamada *et al.*, (2013).

Inter-annual variance (Additional variance)

The approach in Kitakado and Okamura (2005; 2008; 2009) will be used to estimate yearly variation in abundance levels due to inter-annual change in distribution of the Antarctic minke whale population same as the case that OK model was applied to estimate the Antarctic minke whales abundance based on IWC-IDCR/SOWER. By considering the inter-annual variance, underestimating the variance of the abundance estimate can be avoided. In order to estimate inter-annual variance with more precision, not only the abundance estimate based on this survey but also the JARPA and JARPAII abundance estimates adjusted by g(0) estimate can be used. Abundance trend can be also estimated by the approach in Kitakado and Okamura (2005; 2008; 2009).

3. Expected outcomes

By following the method proposed above, abundance estimates for the Antarctic minke whale taking g(0) estimate and additional variance into account will be obtained for JARPA, JARPAII and the new research program proposed. These estimates can contribute to interpret and to understand the time series of abundance estimate of the minke whales from the start of JARPA. These abundance estimates will contribute to construct/develop population dynamics model of Antarctic minke whales (e.g. SCAA (Punt et al., in press)) and ecosystem modeling (e.g. Kitakado et al., 2014). These estimates can also be used for application of the RMP to Antarctic minke whales.

4. Others

The plan of the dedicated sighting survey for each austral summer season, including details of the survey design, will be presented to the corresponding annual IWC SC meetings. The survey design will take into account the specific research activities planned for a particular Area and austral summer season, and will follow the recommendations or suggestions provided by the NEWREP-A Review Panel (IWC, 2015a pp.14-15), including incorporating detailed track lines. NEWREP-A researchers will work closely with the IWC SC before finalizing the survey approaches and to ensure that future survey plans fully follow the IWC SC guidelines for such survey plans.

The survey design for the first two-three years will include feasibility studies on biopsy sampling of and telemetry for Antarctic minke whales, on which the NEWREP-A Review Panel made specific recommendations. It will also include krill and oceanographic surveys. See Document SC/66a/SP08 for the plan of the dedicated sighting survey in the 2015/16 season.

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Objective I(ii)

Improvement of precision of biological and ecological parameters

Part 1: Improvement of age data precision

Physical age is one of the life history parameters required for management of large whale stocks (Zenitani and Kato, 2006). Various methods have been applied for age estimation of baleen whales such as counting growth layer groups (GLGs) accumulated in the earplug, decoding oscillation patterns of baleen plate surface, recording the number of corpora, examining ossification of vertebra, and examining accumulation patterns of several advanced glycation end products (Ohsumi, 1967; 1977; Singh *et al.*, 2001). Long-term tracking of an identified individual at the breeding area is a direct method to identify true age of the individual, but this method cannot be applied for Antarctic minke whales because these whales have few distinct morphological characteristics for individual identification (Gabriele, *et al.*, 2010; Ohsumi *et al.*, 2007). During the JARPA and JARPAII programs, the method of counting GLGs was adopted for age estimation of Antarctic minke whales, and the obtained data have been used for statistical catch-at-age (SCAA) analysis, which is a population dynamics model used by the IWC/SC (Punt *et al.*, 2013).

In the JARPA and JARPAII programs, the age of each whale was estimated from counting GLGs at the bisected surface of the earplug under a stereoscopic microscope. The advantages of this method are that it requires no specific apparatus, and the dataset obtained using this method have been accumulated since the commercial whaling period. The latter allows us to conduct long term analysis of population dynamics using the SCAA. However, disadvantages of the method are that it requires extensive training for a reader, it often involves inter-reader variability, and some whales, especially younger individuals, have unreadable earplugs.

Recently, the inter-reader variability has been evaluated under suggestions and guidance from the IWC/SC, and after that evaluation, the estimated ages with this variability was incorporated into the SCAA analysis (IWC, 2012; Kitakado *et al.*, 2013, Punt *et al.*, 2013). Although ages of whales with unreadable earplugs were estimated applying age-length-key to be tentatively incorporated into the SCAA, the SCAA analysis will be definitely improved if the ages of those whales are obtained more precisely using alternative methods.

Under this proposed research program, estimation of biological and ecological parameters will be carried out using the SCAA model (see Appendix 5), which is based on age and abundance data. Age data will be then obtained from counting of GLGs as this is the accepted procedure by the IWC/SC. In addition, new methods for age estimation will be explored to improve precision of the data. Ratio of aspartic acid enantiomers in the lens nucleus, and level of DNA methylation will be examined for evaluating its potential use in age estimation of Antarctic minke whales. These chemical methods have high reproducibility compared to age estimated from earplugs. Furthermore, DNA methylation analysis will allow us to pursue age estimation from a small amount of skin sample collected by biopsy sampling.

1. Improving the age estimation system of Antarctic minke whales based on ratio of aspartic acid enantiomers in the lens nucleus

1.1 Backgrounds and objectives

It is well known that the amino acids, such as aspartic acid, in the protein of eye lens of mammals change from L-form to D-form very slowly (Masters *et al.*, 1977). Some authors have applied the racemization of aspartic acid in lens protein for the estimation of age of baleen whales (Nerini, 1983, George *et al.*, 1999). During the age estimation process, however, these authors did not derive two important parameters from the species in question but extrapolated from other whale species and/or animals. In

our preliminary study on Antarctic minke whales, we were able to determine these parameters from laboratory experiments (Yasunaga *et al.*, 2014) using samples of Antarctic minke whales taken from JARPA II.

The objective of this proposed research is to verify the age estimation equation based on aspartic acid enantiomers in the lenses of Antarctic minke whales. Furthermore, this age estimation system will be improved with other statistical methods and biological data.

1.2 Methods

Enantiomers of aspartic acid in lens of whales are measured using high performance liquid chromatography (Yasunaga *et al.*, 2014). The new statistical methods, such as the Delta method (George *et al.*, 1999), are applied to estimate the precision of age estimation with the enantiomers. In addition, theriogenological data, such as number of corpora for both ovaries, is applied to verify the accuracy of the estimated ages (Rosa *et al.*, 2013).

2. Feasibility study of the age determination method based on DNA methylation

2.1 Backgrounds and objectives

Recent evidence suggests that epigenetic changes are both directing the process of ageing and being caused by it. The best studied class of epigenetic change in vertebrates is the methyl group presence or absence at the C5 position of Cytosine residues that are adjacent to Guanidine residues. For cetacean species, Polanowski *et al.* (2014) applied this method to estimate ages of humpback whales. The authors examined age-induced changes in DNA methylation on humpbacks genes that were identified as homologous regions to human and mouse.

It should be noted that this technique does not provide the chronological age of the individuals but rather a physiological age that can be used as a proxy for chronological age (IWC, 2015a pp16). The physiological age change with constant biological processes. Furthermore this technique may require calibration not only by species but by population and period. There is as yet no experience on this regard, because it has been applied to a single population of a single species of cetacean, i.e. humpback whales, due to the novelty of the technique (IWC, 2015a pp16).

This proposed study will evaluate the feasibility of an age-determining method based on DNA methylation analysis for Antarctic minke whales. This study will examine the correlation between DNA methylation levels and whale ages determined by counting GLGs.

2.2 Methods

Following the recommendations by the NEWREP-A Review Panel, at first, the stock effect on the DNA methylation (DNA-M) technique will be examined using 50 highly readable earplugs each from I-stock animals sampled in Area IVW and P-stock animals sampled in Area VE under JARPA II, ideally from whales sampled in a same year in each Area. In the next step the effect of the period will be examined by using a similar number of samples from different austral summers. DNA methylation changes will be identified at the Antarctic minke whale genes homologous to the humpback whales. The procedure for identification of age-related DNA methylation site and measurement of methylation level will follow previous studies (Maegawa *et al.*, 2010; Bocklandt *et al.*, 2011; Polanowski *et al.*, 2014). The proponent has already contacted a specialized laboratory and a university in Tokyo so that this study can be conducted in collaboration with experts. Preliminary results will be reported at the 2017 IWC SC meeting and final results at the 2018 IWC SC meeting. In this way the proponent will respond to the NEWREP-A Review Panel recommendation that this feasibility study be conducted early in the NEWREP-A program. The proponent will also address the issue of inter-laboratory calibration in consultation with the IWC SC.

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Objective I(ii)

Improvement of precision of biological and ecological parameters

Part 2: Refinement of the SCAA model and estimation of biological parameters

Background and Objectives

Under Main Objective I, drawing information on certain biological and ecological parameters is one of key issues for the specification of trials. Although some parameters like the ASM are directly obtainable in the observation, there are several parameters which can only be derived from the results of stock assessment. The IWC has spent a decade for its discussion (see. Punt, 2014a, b, Punt and Polacheck, 2005, 2006, 2007, 2008, Punt *et al.* 2013), and now the SCAA model is recognized as the "best currently available model for examining stock dynamics for Antarctic minke whales" (IWC, 2014).

In the age-structured population dynamics model, parameters such as the "natural mortality rate (*M*)", the "MSYR", recruitment relationship and time-varying carrying capacity are influential to the results. Among these parameters, *M* is reasonably well estimated including its age-dependence, while the estimate of MSYR is still unrealistically high in view of knowledge for other baleen whales. The estimated changes in the recruitment rate (the number of recruited whales divided by that of mature females) and carrying capacity over years were clear so far, but estimating and monitoring such likely changes in the future are of scientific importance given consideration of various changes in the ecosystem and environment in the Antarctic Ocean. Therefore, the biological parameters including these which will be estimated from data collected by this program will contribute to the trials structure in the RMP *ISTs*. That is why "refinement of the SCAA model and estimation of biological parameters" is one of the research sub-objectives.

Methods

Models used in this sub-objective are mostly similar with the current SCAA models developed in Punt (2014a). The basic assumption is same as that in Punt (2014a).

For each breeding stock, the following basic population dynamics is considered.

 $P_{t,a}^{g,b}$: the number of animals of age a and sex g for breeding stock b at the beginning of year t

 R^b : the number of birth for breeding stock b in year t

$$P_{t,a}^{g,b} = \begin{cases} 0.5R_t^b & \text{for } a = 0\\ (P_{t-1,a-1}^{g,b} - C_{t-1,a-1}^{g,b})S_{a-1}^{g,b} & \text{for } 1 \le a \le A_+ - 1\\ (P_{t-1,A_+}^{g,b} - C_{t-1,A_+}^{g,b})S_{A_+}^{g,b} + (P_{t-1,A_+}^{g,b} - C_{t-1,A_+}^{g,b})S_{A_+}^{g,b} & \text{for } a = A_+ \end{cases}$$

$$R_t^b = f^b \tilde{P}_t^{F,b} \left[1 + A^b \left\{ 1 - \left(\frac{P_{t,1+}^b}{K^b} \right)^z \right\} \right] e^{u_t - \sigma^2/2}, \quad u_t \sim N(0, \sigma^2)$$

$$C_{t,a}^{g,b} = \frac{\gamma_a^{g,b} P_{t,a}^{g,b}}{\sum_{a}^{A} \gamma_a^{g,b} P_{t,a}^{g,b}} C_t^{g,b}$$

 $S_a^b = \exp(-M_a^b)$: annual survival probability of animals of breeding stock b and age a

 γ_a^b : selectivity (depends on period)

 f^b : pregnancy rate

 A^b : resilience

 K_t^b : carrying capacity for 1+ population in year t for breeding stock b

 $\tilde{P}_{t}^{F,b} = \sum_{a=1}^{A} \beta_{a} P_{t,a}^{F,b}$: the number of mature female animals for breeding stock *b* at the beginning of year *t*

$$P_{t,1+}^b = \sum_{a=1}^A (P_{t,a}^{M,b} + P_{t,a}^{F,b})$$
: 1+ population size for breeding stock b

 β_a : maturity rate for animals of age a

The following points will be taken into account in the proposed programs:

- Apply longer time-series of newly available abundance estimates with g(0) correction
- Consider a possible change in the age-at-sexual maturity in the model
- Consider any new age-reader's bias and variance in age-reading error matrices
- Extend the existing SCAA models to incorporate updated assumption of population structures for the Antarctic minke whales (time-varying stock boundary and mixing pattern)

It is expected that the results of SCAA analyses will provide improvement in the precision of key biological and ecological parameters as was the case in the previous programs although the extent of improvement cannot be evaluated given recent time-varying parameters such as carrying capacity and productivity etc.

Of course, outcomes based on the new SCAA analyses will contribute to the specification of the Implementation Simulation Trials.

Table 1. Key points in specification

Parameter/Structure in the model	Assumption
Stock structure (# stocks)	2 (I/P) or additional stocks?
Mixing pattern	Base: Hard boundary as in Punt <i>et al.</i> (2014a) Extension: soft, time-varying, sex, maturity
MSYR ₁₊	Estimated
MSYL ₁₊	0.6 (given)
Mortality rate (M) (Immat, Mat)	Age-specific and time-varying
Initial year and initial depletion	D (=1) at 1930
Carrying capacity K_{1+}	Time-varying
Reproduction	Deviation in the recruitment

Parameter/Structure in the model	Assumption
Growth curve	Time-varying
Selectivity	Time-varying
Additional variance (inter-annual distributional change)	Some numbers assumed
Age-reading errors	Assumed

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Objective I(iii)

Refinement of stock structure hypotheses of Antarctic minke whale in Areas III-VI for the implementation of the RMP

Background

The most recent results of the research on stock structure of Antarctic minke whales were based on JARPA and JARPAII samples, genetics based on mtDNA control region sequencing and microsatellite DNA, and morphometrics based on ten body measurements. Results were presented and discussed at the JARPAII review workshop, and subsequently at the 2014 IWC Scientific Committee (IWC SC) Meeting (Pastene *et al.*, 2014; Kitakado *et al.*, 2014; IWC, *in press a;b*).

The hypothesis accepted by the IWC SC is the occurrence of at least two stocks in the JARPAII research area (35°E-145°W), with the cores of these stocks distributed in the western (Indian or 'I' stock) and eastern (Pacific or 'P' stock) sectors of the research area (Figure 1). There is a soft boundary between these stocks in Areas IVE and VW with the mixing proportion between stocks in these sectors changing by year and sex (Kitakado *et al.*, 2014).

Furthermore the JARPAII review workshop noted that 'management procedure considerations on stock structure focus on developing plausible interpretations of available data, not simply the single 'best' interpretation when examining uncertainty' (IWC, *in press* a). In this spirit, at the JARPAII review workshop, an alternative hypothesis was proposed: a single stock that exhibits one-dimensional isolation by distance along a longitudinal gradient (IWC, *in press* a).

The following sub-objectives were identified, all of them aimed to refine the current stock structure hypotheses. Plausible hypotheses can then be used to develop *Implementation Simulation Trials (ISTs)* during the RMP *Implementation* process, which are aimed to span the uncertainty in different aspects, in this case the uncertainty in stock structure.

Sub-objectives

a) Investigation of the stock structure in Areas IIIW (0-35E) and VIE (120-145W)

The JARPAII review workshop commented that 'Antarctic minke whales are more-or-less continuously distributed around the Antarctic continent and that in contrast the JARPAII research area represents just under half of the circumpolar area'. They further noted that 'the lack of information provided for areas outside the programme's research area presents some inherent difficulties in fully meeting the objective to elucidate spatial and temporal variation in stock structure, even though the information developed under JARPAII is probably sufficient for the purposes of developing trials to evaluate RMP variants within the area of sampling' (IWC, *in press* a).

The examination of the stock structure of Antarctic minke whales in the areas immediately contiguous to the JARPAII area, *e.g.* Areas IIIW and VIE is considered useful to refine the current hypothesis on stock structure. This will allow the investigation of the longitudinal expansion of the I and P stocks, to the west and east, respectively. If abundance information also becomes available from those sectors then the combined new information of stock structure and abundance will contribute to sound *ISTs*, and to the optimization in the use of this whale resource as larger catch levels can be estimated without depleting the resource (in the case that the I stock extends till Area IIIW and the P stock until Area VIE).

In addition, longitudinally-extended data sets would probably facilitate the analyses of the second hypothesis on isolation by distance proposed by the JARPAII review workshop.

Samples and data

Genetic samples and morphometric data for studies on stock structure are available from JARPA and JARPAII for Areas IIIE (35°-70°E) and VIW (170°-145°W). As explained above, under this sub-

objective there is a need for acquiring additional samples and data from Areas IIIW and Areas VIE to refine the stock structure hypotheses. There are some genetic samples available for those sectors from the time of commercial whaling, which could be used for this purpose.

The quality of the historical samples for DNA analysis should be investigated first. Genetic samples from the period of commercial whaling in Areas IIIW and VIE involve different kinds of tissues such as blood and liver. These were collected in the 70's. There is a total of 58 samples from Area IIIW (57 blood and 1 liver) and 175 (liver) from Area VIE. A feasibility study is required to investigate whether these samples are useful for extracting and analyzing DNA.

The geographic distribution of the historical samples are restricted to the southern part of Areas IIIW and VIE, probably concentrated around the ice-edge (Figure 2). Considering the study of Goto *et al.* (1998), which showed that samples collected exclusively near the ice-edge are not informative of the mtDNA heterogeneity, samples from commercial whaling in Areas IIIW and VIE alone could not be sufficient for studies on stock structure, and additional samples are required from the offshore areas in those sectors.

As noted above, the analyses on stock structure have involved genetics and non-genetics (morphometric) methods. Both markers have been informative of the stock structure (Kitakado, *et al.*, 2014). Therefore, it is proposed that the research take planned alternatively in Areas IV-IIIE and V-VIW, be expanded at least in two years to cover sectors IIIW and VIE, respectively. This will allow the collection of other information useful for stock structure in addition to DNA, *e.g.* detailed body measurements.

The feasibility study on biopsy sampling in Antarctic minke whale will be undertaken along with the dedicated sighting surveys, at an early stage of the first six-year period of NEWREP-A (Section 6 and SC/66a/SP08). As the program develops, the design of the feasibility study on biopsy sampling will take account of the four specific recommendations by the NEWREP-A Review Panel [1) involve people with expertise in successfully biopsy sampling common minke whales in the North Atlantic; 2) mimic the sampling strategy developed for lethal sampling; 3) record information on time taken, sea state, swell, etc. to enable a plausible measure of effort required to be developed; and 4) consider the amount of tissue and nature of tissue required (for each analysis and in total)], as far as practicable. In planning and implementing the feasibility study, the program will make use of experiences obtained by foreign (e.g. colleagues working with North Atlantic minke whale) and Japanese (e.g. colleagues working with North Pacific minke whale) experts. The "ideal" framework of the study suggested by the NEWREP-A Review Panel (e.g. representativeness of the entire area and time period as well as, random sampling techniques) will be considered as a second step after examining the practicability of the sampling technique itself in the initial stage. It should be noted that the value of biopsy samples for the NEWREP-A research objectives should be evaluated in parallel with the results of feasibility studies of analytical approaches that potentially use biopsy samples (e.g. DNA-M, stable isotopes, hormones).

Design of the feasibility biopsy experiments in offshore waters will take into consideration the previous experience of the IDCR/SOWER (Ensor *et al.*, 2001; 2004) and JARPN/JARPA (Nishiwaki, 2000) surveys.

Laboratory work and analytical procedure

The routine procedure for mtDNA control region sequencing and genotype with a number of microsatellite loci, will be used. In the case of mtDNA the first 500bp at the 5' end of the control region will be sequenced. In the case of microsatellite DNA a set of 12 loci are used: AC045, AC082, AC087, AC137, CA234, DlrFCB14, EV1, EV104, GT23, GT129, GT195 and GT211. Details of the laboratory procedure for those two markers can be found in Kanda *et al.* (2014a).

Regarding morphometrics, up to 10 body measurements will be used, as in the past studies (*e.g.* Figure 2 in Kitakado *et al.*, 2014).

Analysis will be based on hypothesis testing (e.g. Pastene et al., 2014).

b) Investigation of the spatial and temporal pattern of mixing between the I and P stocks in Areas IVE and VW

As reported above modeling work based on genetic and morphometric data showed an area of mixing between the I and P stocks in Areas IVE and VW, with the area of mixing and mixing proportion changing by year and by sex ('soft boundary').

The concept of a 'soft boundary' is a new one within the RMP *Implementation* process. A long-term data series of spatial and temporal pattern of mixing in Areas IVE and VW would be required to capture the uncertainty in stock structure and mixing during the *ISTs*.

Samples and data

Research take will be conducted alternatively in Areas IV and V. At least in two years of the total research period sampling will be designed to cover both Area IVE and VW.

Morphometric and genetic data obtained from the whales sampled can be examined in conjunction with the relevant data from JARPA (16-year period) and JARPAII (6-year period) to create a long-term data series to examine temporal and spatial pattern of stock mixing in the relevant longitudinal sectors.

Analytical procedure

The method developed by Schweder *et al.* (2011), improved following suggestions from the IWC SC (Kitakado *et al.*, 2014), will be used.

c) Investigation of the pattern of movement of Antarctic minke whales within the feeding grounds and between feeding grounds and putative breeding grounds

Movement within the feeding ground

Kato *et al.* (1993) examined all available marks (2,864 mark release and 110 recoveries of Discovery tag) in the Antarctic. Through their analyses it was revealed that: the average distance of longitudinal movement is about 30 degree, recaptured animals showed no preferential east or west movement in the Antarctic and no significant difference was found between sexes in terms of distance moved. Marked movement of whales was observed through 130E, the boundary between Areas IV and V. Regarding boundaries they noted that the absence of mark crossing at 80E and 160E suggest the possibility of separate feeding stocks. Following an examination of mark distribution and catching effort they concluded that discontinuity at 80E could be more suggestive of a boundary between stocks than at 160E.

These results are broadly consistent with the current hypothesis on stock structure, notably the interchange of animals through 130E, the transition area of the current hypothesis, and the possibility of additional structure west of Area IV. This provides support to the idea of additional sampling in Area IIIW under the first short-term objective above.

Movement between feeding grounds and breeding grounds

There is an assumption that the I and P stocks are related to breeding areas in the eastern Indian Ocean and western South Pacific, respectively, where high sighting density areas of minke whales have been identified in October (Kasamatsu *et al.*, 1995) (Figure 1). However there is no direct evidence of this link, and no genetic samples are currently available for Antarctic minke whales in low latitude areas. The IWC SC has recommended on several occasions that research should be conducted to investigate the location of breeding grounds of this species in the Southern Hemisphere, and the collection of genetic samples (IWC, 2008; *in press* a).

Therefore the migratory destination of Antarctic minke whales of the I and P stock in autumn and winter is another important research topic to address. Are minke whales from the I stock migrating in winter to the high density areas in the eastern Indian Ocean? Are minke whales from the P stock migrating in winter to the high density areas in the western South Pacific Ocean?

Method

Further studies on movement of whales within the feeding grounds and movement of whales between high and low latitudes areas can be investigated through satellite tracking. The latter topic should have priority as there is already some information on movement of whales within the feeding grounds.

Some experiments on satellite tagging were conducted on Antarctic minke whales around the pack-ice (Gales *et al.*, 2013). The following kinds of tags were used by those authors: blubber penetrating satellite tags, Wildlife Computers, SPOT 177N; dorsal fin mounted satellite tags, Wildlife Computers, SPOT 240C; dorsal fin mounted satellite, Wildlife Computers, SPLASH 292A.

As noted above the priority at this stage should be the searching for breeding areas, and for this particular aim, long-term monitoring tags should be considered. In the next step the priority should be changed to obtain genetic samples from the putative breeding areas.

Satellite tagging should be carried out at the end of the feeding season for the I and P stocks in their 'core areas', IVW and VE, respectively. If biopsy sampling is conducted at the same time of the satellite tagging in the feeding grounds, some genetic information for whales migrating into potential breeding grounds will be obtained.

d) Application of new genetic techniques to assist the analyses and interpretation of results in the three sub-objectives above

The IWC SC has welcomed the development of alternative analytical procedure in addition to the standard hypothesis testing approach. For example the paternity analysis presented to the JARPAII review workshop (Kanda *et al.*, 2014b) was welcomed by the Review Panel as such method could contribute not only for the development of genetic-based abundance estimate but in the assistance of the interpretation of stocks structure (IWC, *in press* a).

At the 2014 IWC SC meeting, the development of close-kin mark recapture methods for a number of species, including North Atlantic minke whales and Antarctic blue whales, was welcomed. For example Tiedemann *et al.* (2014) reported a method for finding relatives among North Atlantic common minke whales based on microsatellite data. They investigated the relationship between false discovery rate and detection power. This method could be used for investigating relatives among Antarctic minke whales, and this information will be very valuable for the interpretation of the hypothesis on stock structure.

These techniques will be applied to assist the analyses and interpretations under the three sub-objectives above, and both JARPA/JARPAII and new samples will be used.

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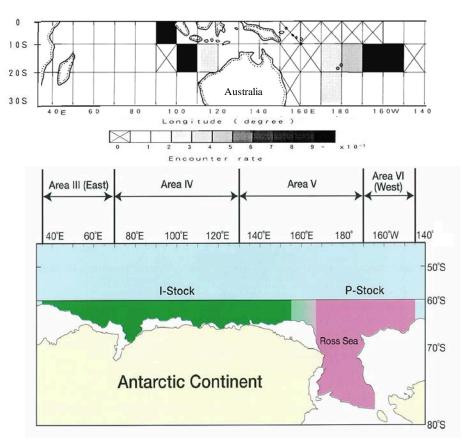


Figure 1. Hypothesis on stock structure of the Antarctic minke whale (lower figure) and sighting density in low latitudes in October (upper figure) (Kasamatsu *et al.*, 1995).

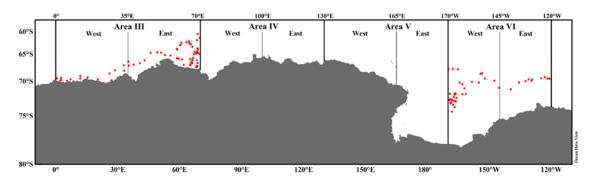


Figure 2. Geographical distribution of genetic samples in Areas III and VI obtained during the period of commercial whaling. There is a total of 58 genetic samples in Area IIIW (0-35E) (57 blood samples obtained in 1973/74; 1 liver sample obtained in 1979/80), and 175 in Area VIE (145-120W) (175 liver samples taken in 1981/82).

Objective I(iv)

Specification of RMP ISTs for the Antarctic minke whales

This appendix briefly outlines the specification of the RMP *Implementation Simulation Trials (ISTs)* planned under the proposed research plan. This corresponds to one of the sub-objectives under Main Objective I. More details of relevant information are provided in the main text of the research plan.

The models assumed in the trials are basically same as those used in the existing Statistical Catch-at Age Analysis (SCAA) (Punt, 2014), and the parameter values estimated and used in the SCAA will be provided as sets of plausible ranges of parameters. Results of new SCAA runs (see Appendix 5) as well from other sub-objectives under Main Objective I (see Appendices 3, 4 and 6), will contribute to the specification. The kind of parameter/structure in the model and potential contribution from the new research is shown in Table 1. Also, some implication or results from Main Objective II (investigation of ecosystem in the Antarctic Ocean), will also be taken into account for specifying the trial structures.

Note that *Small Areas* will be defined based on the updated information on stock structure (see Appendix 6) and possible whaling operations.

Table 1. Template of trial specification

Parameter/Structure in the model	Source of information	
MSYR ₁₊	Existing range/outcomes from this new research plan	
$MSYL_{1+}$	0.6 (given)	
Stock structure	Existing hypotheses/outcomes from this new research plan	
Mixing pattern	Outcomes from this new research plan	
Mortality rate	Outcomes from this new research plan	
Max pregnancy rate	Outcomes from this new research plan	
K_{1+}	Outcomes from this new research plan	
Time varying <i>K</i>	Outcomes from this new research plan	
Additional variance (inter-annual distributional change)	Existing range/outcomes from this new research plan	
Small Areas and Management Variants	Several variants will proposed with consideration of stock stucture hypotheses and possible whaling operations	

Reference

Punt, A.E. 2014. Assessment of Antarctic Minke Whales using Statistical Catch-at-age Analysis. Paper SC/F14/O02 presented to the Expert Workshop to Review the Japanese JARPAII Special Permit Research Programme, February 2014 (unpublished). (99pp).

Objective II(i)

Plan for ecological research (krill abundance estimation and oceanographic observation)

1. Background and objectives

The Southern Ocean can be considered as a wasp-waist ecosystem featuring many species at the bottom and top trophic levels, and few dominant species at the mid-level. Unlike other wasp-waist ecosystems where small pelagic, planktivorous fish such as sardine and anchovy are dominant at the mid-level, a plankton species, the Antarctic krill (*Euphausia superba*), is dominant at that level in the Southern Ocean (Bakun, 2006; Atkinson *et al.*, 2014). The mid-trophic level species are also called forage species which play a pivotal role in marine ecosystem and economics by sustaining many predators and fisheries directly and indirectly (Pikitch *et al.*, 2014). Abundance and demography of Antarctic krill is fundamental information to understand the Southern Ocean ecosystem. In addition, monitoring of oceanographic conditions in the Southern Ocean is important as decadal scale changes in oceanographic properties have been reported around the Antarctic (Jacobs and Giulivi, 2010; Meredith *et al.*, 2014; van Wijk and Rintoul, 2014).

The main objective of this survey is to estimate the abundance of Antarctic krill acoustically in the survey area by applying a standard survey design similar to those developed and implemented for CCAMLR 2000 (Trathan *et al.*, 2001), BROKE (Nicol *et al.*, 2000a) and BROKE-West (Nicol *et al.*, 2010). The estimates will be used as an important input parameter of ecosystem models. Demography of Antarctic krill in the survey area will also be studied to understand its ecology. Spatial interactions among baleen whales and krill will also be investigated using the obtained data. The second objective is to collect oceanographic data to test whether oceanographic conditions in the Southern Ocean are changing.

2 Methods

- 2.1 Research under this program (Sighting survey-based krill survey)
- 2.1.1 Abundance estimation of Antarctic krill by an echosounder

Simple surveys for krill abundance by using echosounder and net-tow are planned with R/V. The type of R/V may be modified based on logistical reasons.

2.1.2 Demography of Antarctic krill

Simple surveys for estimating of demography of Antarctic krill by using sampling net would be planned. The type of sampling net may be modified based on logistical reasons.

2.1.3 Oceanographic observation

The expendable conductivity-temperature-depth profilers (XCTD) will also be used as a supplementary instrument. The XCTD would be deployed on an opportunistic basis. Information on water temperature and salinity is required to estimate sound velocity and absorption coefficient for correction of echosounder data.

- 2.2. Research outside of this program (CCAMLR-type dedicated krill survey)
- 2.2.1 Abundance estimation of Antarctic krill by an echosounder

Area IV overlaps with the CCAMLR management areas 58.4.1 and 58.4.2. Area V overlaps with the CCAMLR management areas 58.4.1, 88.1 and 88.2. The survey area and design which was used in

BROKE and BROKE-West would be proposed when the survey is conducted in Area IV. Oceanographic observations will also be conducted along with the krill survey. Detailed survey plans would be presented to the IWC/SC, CCAMLR-EMM and SC-CAMLR in advance of the surveys to receive their comments. The R/V *Kaiyo Maru* is a desirable candidate research vessel for this survey since it would be capable of conducting a krill survey that fulfills the requirements of the CCAMLR. The vessel would be available for at least one austral summer season in the first six-year period

2.2.2 Net sampling

A frame-type trawl net would be towed at either predetermined or targeted station. Targeted tows would be conducted to identify the species and size compositions of biological backscattering detected by the quantitative echosounder. The purpose of the predetermined tows would be to estimate the abundance and distribution of planktons independently from the echosounder. The nets would be run down to 1,000 m or near the sea bottom at the predetermined stations. Either a rectangular midwater trawl (RMT) with nominal mouth areas of 8 and 1 m2 (1 + 8) and mesh sizes of 4.5 and 0.33 mm (Baker *et al.*, 1973), an Isaacs-Kidd Midwater Trawl (IKMT) or a Matsuda-Oozeki-Hu-Trawl (MOHT) (Oozeki *et al.*, 2004) would be proposed. As the main objective of this exercise is the estimation of Antarctic krill using the echosounder, priority would be given to targeted tows. Length frequency data of Antarctic krill obtained by the net sampling is required to estimate abundance of Antarctic krill by using the echosounder. Species compositions and size structure of net samples would be analyzed.

2.2.3 Demography of Antarctic krill

Length frequency distribution and maturity stage of Antarctic krill will be constructed using samples from stomachs of Antarctic minke whales in JARPA and JARPAII periods in addition to samples (both from stomachs and net haul) obtained in upcoming surveys. Regional and temporal difference of demography will be investigated. Comparison between samples from stomach and net will also be attempted.

Construction of population dynamics models will be considered using the above mentioned demography data. Modeling approaches similar to the past studies (Reid *et al.*, 1999, 2002, 2006, 2010; Murphy and Reid, 2001) will be applied with modifications whenever these are appropriate.

Information on population genetic structure of Antarctic krill is important for an appropriate interpretation of the results of the demographic analyses. Therefore stock structure of Antarctic krill within Areas IV and V will be examined using samples obtained by this research.

2.2.4 Oceanographic observation

The conductivity-temperature-depth profilers/Rosette Multi-bottle Samplers (CTD/RMS) would be used as the primary oceanographic observation instrument. The XCTD would also be used as a supplementary instrument. The CTD would be deployed at predetermined and targeted stations down to 3,500 m or near the sea bottom while water samples would be collected by the RMS for the purpose of studying physical, chemical and biological oceanography. The XCTD would be deployed on an opportunistic basis. Information on water temperature and salinity is required to estimate sound velocity and absorption coefficient for correction of echosounder data.

2.3 Response to recommendations by the NEWREP-A Review Panel

The latter CCAMLR-type dedicated krill survey will apply a standard survey design similar to those developed and implemented for CCAMLR 2000 (Trathan *et al.*, 2001), BROKE (Nicol *et al.*, 2000) and BROKE-West (Nicol *et al.*, 2010).

The proponent will respond to recommendations by the NEWREP-A Review Panel (IWC, 2015a pp20-23) in the following way.

- i) Ship(s) and echosounder system(s) will be tested before steaming to the Antarctic to determine the likely effective acoustic sampling range (signal-to-noise by depth characterization) and potential for detecting krill.
- ii) Cross-calibration between both types of krill surveys vessels will be carried out the in waters around Japan, in a near synchronous way, as the first priority. However, the proponent might conduct the calibration in the survey area as well if conditions are met.
- iii) Simultaneous survey by the dedicated krill survey vessel equipped with a frame-type trawl and echosounder, and that by the dedicated whale sighting vessel equipped with an echosounder and the whale sampling vessels will be conducted within a small-scale experimental survey area over a short time period (e.g. one week) so that comparisons among vessels/samples can be made.
- iv) Sufficient time will be allocated for net sampling of krill, by following a survey design similar to those developed and implemented for CCAMLR 2000, BROKE and BROKE-West. Approximately 2.5-3 hauls per day will be a standard for net sampling frequency under the proposed CCAMLR-type dedicated krill survey.

Recommendation i) above will be conducted in the 2015/16 season. The other recommendations will be considered in future surveys.

3. Distinctive features and expected outcome

The abundance estimates and the size structure of krill would be used as an important input parameter of ecosystem models. Spatial interactions among baleen whales and krill would also be investigated using the obtained data. The estimated abundance would be reported to the IWC/SC and CCAMLR-EMM. The obtained data would contribute to not only management of cetaceans but also management of krill. Only two krill abundance estimates accepted by the CCAMLR were available in Areas IV and V. BROKE was conducted in the area between 80°E and 150°E south of 62°S in 1996 (Nicol *et al.*, 2000a). BROKE-West was conducted in the area between 30°E and 80°E south of 62°S in 2006 (Nicol *et al.*, 2010). The R/V *Kaiyo-maru* conducted a krill survey in the Ross Sea in 2004/2005 although the abundance estimate was not accepted by the CCAMLR (Naganobu *et al.*, 2010). The new krill abundance estimates obtained by this research and other researches outside of the program would enhance our knowledge on krill in the survey area where data are scarce.

Though several demographic studies on Antarctic krill in the survey area have been reported (Nicol *et al.*, 2000b; Pakhomov, 2000; Sala *et al.*, 2002; Taki *et al.*, 2008; Kawaguchi *et al.*, 2010;) the information is scarce in comparison with that of the Antarctic Peninsula region. The obtained data would contribute to understanding the ecology of Antarctic krill in the survey area. This study would potentially provide information on population dynamics of Antarctic krill in the survey area where no population dynamics study on Antarctic krill has been carried out. Stock structure studies at a circumpolar scale (Zane *et al.*, 1998) and sub-regional scales in the Western Antarctic Peninsula region (Batta-Lona *et al.*, 2011) have been carried out. However, no such studies have been carried out in the survey area. The results of combination of this study and other studies outside of the program would potentially provide new information on stock structure of Antarctic krill in the survey area, which is important for interpreting demographic results.

Oceanographic data would provide us information on the current status of oceanography in the survey area. The data would be sent to the appropriate data depository for further analysis integrating data obtained by JARPA, JARPAII and other surveys.

Preliminary results of the analysis would be reported to the IWC/SC and CCAMLR-EMM within 2 years after the completion of each survey. The analysis would be completed within 4 years after the completion of each survey and submitted to peer review journals.

The overall research plan may be modified based on the type of R/V available.

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Objective II(ii)

Abundance estimate of some cetacean species as input data for ecosystem modeling

1. Background and objectives

Abundance estimates for humpback, fin and blue whales have shown significant increase based on JARPA and JARPA II sighting surveys (Matsuoka *et al.*, 2011; Matsuoka and Hakamada, 2014). In other studies, significant increase of the humpback whales (*e.g.* Noad *et al.*, 2011; Hedley *et al.*, 2011; Branch 2011) and blue whales (Branch *et al.*, 2004) was reported. Result of the assessment for breeding stock D (BSD) humpback whales showed that the abundance in 2012 was about 90% of the initial population size (IWC, 2014a). Like the Antarctic minke whale these baleen whales feed on krill. Population dynamics of these baleen whales could affect that of the minke whales and therefore it is necessary to examine abundance estimates and trends for these species. Such abundance estimates and trends will be used for input of ecosystem modeling (Kitakado *et al.*, 2014) especially multi-species-production type models.

Sighting surveys for the Antarctic minke, humpback and blue whales probably cover the peak of their distribution in the austral summer. However, for other species such as fin whales, this is not the case. The Expert Workshop to review JARPA II reiterated that considerable care should be given to the interpretation of abundance and trends of fin whales (IWC, 2014b).

Information on spatial distribution of cetaceans in finer scales can be derived by spatial modeling. The updated requirements and guidelines for conducting surveys and *Implementations* (Hedley and Bravington, 2014) include the guideline for abundance estimation in spatial modeling approach. At the JARPAII review workshop, the existence of competition amongst baleen whales in spatial context was discussed and but no final conclusion on this was reached. Given the important management implications such competition would have, it is important to gather data to assist in resolving this issue. Spatial abundance estimates derived from the spatial modeling approach that follows the guideline agreed by IWC/SC could facilitate such further investigations of cetacean interactions. Such estimates can be used to estimate occupied area indices (*e.g.* Murase *et al.*, 2014)

In order to attain Objective II(ii), two sub-objectives are set: i) produce abundance estimates of some cetacean species as input data for ecosystem models, and ii) produce abundance estimates using the spatial modeling approach based on the updated IWC/SC guidelines, for investigation of cetacean interactions in a spatial context (Objective II(iv)).

2. Methods

2.1 Survey methods

Sighting surveys

Survey protocols follow the 'Requirements and Guidelines for conducting surveys and analyzing data within the Revised Management Scheme' (IWC, 2012). Sighting protocols are the same as those used in IWC-IDCR/SOWER (Matsuoka *et al.*, 2003). Survey areas are planned to cover one IWC Management Area (Areas III, IV, V and VI) in a year. Following the recommendation at the Expert Workshop to review JARPAII (IWC, 2014b), if an initial sighting occurs near to a vessel, effort will be make in identifying the ecotype of killer whale.

Sighting survey in northern area

To examine distribution and abundance of fin whales, a sighting survey will be conducted in one year out of 12 years in the latitudinal band of 55-60°S. It should be recognized that this band is not sufficient to cover all of the distribution in summer as suggested by JSV data (Miyashita *et al.*, 1995), but it is

what can be covered in a single year. The longitudinal area will be decided considering the distribution pattern of the species and logistics.

Acoustic survey

Acoustic survey would be used as an alternative method for situations when sighting survey cannot be conducted (e.g. during night, within pack ice, bad weather, etc.). There were some previous studies using acoustic devices to collect the data for (relative) abundance estimation. For example, combination of towed hydrophone and visual surveys (Barlow and Taylor, 2005), using two towed acoustic devices with visual surveys (Kimura et al., 2014), and setting passive acoustic devices at fixed site (Van Opzeeland et al., 2014). It remains to be examined how often baleen whales make sounds and how correctly the species can be differentiated from the recorded sound. The acoustic survey should be considered as a feasibility survey at this stage.

2.2 Analytical methods

Abundance estimation

The Standard methodology of line transect surveys (e.g. Branch and Butterworth, 2001) will be applied to estimate abundance. For abundance estimation, recommendations made at the Expert Workshop to review JARPA II should be considered; (1) present a more thorough description of methods and assumptions used: (2) highlight the issues related to small sample size and the recommendations made at the JARPA review meeting; (3) identify more clearly the differences between ideal and realised tracklines by year including percentage achieved coverage; (4) explain more fully the treatment of additional variance components in the estimation of rates of increase; (5) include more extensive discussion than at present of the results including interpretation of the results with respect to stock structure and proportion of range covered; and (6) include an updated power analysis of the effects of survey interval and estimation of trend to inform consideration of levels of effort and survey design in the future. (IWC, 2014b). These recommendations will be considered on drafting whale abundance estimation papers. Recommendation (2) will be considered by investigating appropriate stratification of data to estimate abundance, especially in case that sample size of detected whales is too small to estimate abundance for each stratum. Recommendation (6) will be considered taking into account abundance estimates and trends considering recommendations (1)-(5). Candidates for abundance estimates are humpback, fin, blue, southern right, sperm, southern bottlenose and killer whales. Abundance estimate for species such as sperm and southern bottlenose whales may need to be treated as relative because g(0) was estimated less than 1 for these species (Kasamatsu and Joyce, 1995).

In response to the NEWREP-A Review Panel recommendation regarding the g(0) estimation (IWC, 2015a pp13), identification of duplicate sightings will be also conducted for whale species other than the Antarctic minke whale during NEWREP-A sighting surveys in IO mode. In order to determine whether g(0) is significantly different from one, analyses on duplicate sightings will be conducted for those whale species given that sufficient data will be obtained under the NEWREP-A program.

Spatial modeling

Density Surface Modeling (DSM) (Miller *et al.*, 2013) will be used, which is one of the packages in the DISTANCE grogram (Thomas *et al.*, 2010). But there is a room for trying other options (*e.g.* smoother, modeling framework, variance estimation method etc.) to improve abundance estimates. Furthermore, the utility of using environmental covariates in the spatial modeling for improving the abundance estimates will be investigated.

3. Features and expected outcome

Abundance estimate for the species listed above can be used as input data for the ecosystem modeling, especially multi-production type, and for management purposes. Preliminary analysis of spatial

modeling using the sighting data can contribute to discussion on the guideline for abundance estimation using spatial modeling approach by providing the example to check whether the proposed guidelines are appropriate. If acceptable spatial abundance estimates are obtained, they can contribute to the investigation of interactions among cetaceans in a spatial context

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Objective II(iii)

Estimation of prey consumption by Antarctic minke whale and its nutritional condition

1. Background

The krill surplus hypothesis (Laws, 1977) is based on the concept of species interactions between krill predators and was investigated using population dynamics models by Mori and Butterworth (2006). Looking toward the conservation and sustainable use of whale resources in the future, the IWC needs to consider multi-species management approaches in the Antarctic Ocean where the world's largest whale resources exist. Since the Antarctic minke whale plays an important role in the Antarctic ecosystem as a predator with its abundance estimated at over 500,000 individuals (IWC, 2013), estimation and monitoring of prey consumption by the Antarctic minke whale and its nutritional condition will be useful for verification of the Multi-production model (objective II(iv) Part 2).

The Scientific Committee of IWC (SC65b) agreed that a decline in blubber thickness and in fat weight that was statistically significant at the 5% level had occurred in JARPA period. It also agreed that the model presented in SC65b would benefit from extension to determine whether the trends identified showed any indication of change over time. It was noted that, even if not statistically significant at the 5% level, estimates of changes in trends would ultimately be important to take into account in fitting multi-species models. Furthermore Japanese whaling surveys have been conducted for more than 20 years in the Antarctic and have collected body condition data that are a unique example that can be used as described above (IWC, 2014a). Taking into consideration this situation, body condition studies including its monitoring are important for whale studies in the Antarctic.

2. Objectives

2.1 Estimation of prey consumption

The total krill consumption by the Antarctic minke whale was estimated to be approximately 2.7-18.9 % of krill biomass in IWC management Areas IV and V (Tamura and Konishi, 2009), however the precision of the estimate should be increased by accounting for some uncertainties of daily and seasonal prey consumptions (Leaper and Lavigne, 2007; Tamura *et al.*, 2009; IWC, 2014b). Although the daily feeding pattern and the duration of feeding days are important parameters to estimate consumption, little information is available for these. The main objective is, therefore, to increase the precision of estimates of consumption by the Antarctic minke whale by analysis of data from night-time sampling of stomach contents and by the tagging studies.

2.2 Body condition

One of the objectives for examining body condition is to monitor any trend of body condition as an indication of possible ecosystem change and to verify the results of ecosystem models to see whether a selected scenario fits the reality. Besides this monitoring purpose, the proposed research will assess the body condition indicators to examine the whales' biological condition by comparing these to the amount of total fat storage. Additionally other chemical body condition markers will be tested to examine their usability.

3. Research methods

3.1 Improvement of estimation of prey consumption

The prey consumption will be calculated by the time change of stomach content weights and basal metabolism using the Monte-Carlo simulations to increase the precision (Leaper and Lavigne, 2007; IWC, 2014b; Tamura and Konishi, 2014). The plan includes samplings whales during the night in the

feeding area of the Antarctic minke whale with plankton net surveys to examine the of qualitative vertical migration patterns of krill in the same area.

To consider the uncertainties in calculation of consumption, the tagging and satellite tracking that records the position of whales for a long period will be conducted for the Antarctic minke whale to examine the migration patterns and duration of stay in the Antarctic waters. The tagging of sensor data logger (depth and acceleration) will be conducted for the Antarctic minke whale to examine its daily feeding patters.

In theory, daily consumption can be estimated by the number of lunge feeding and krill density and some biological and environmental parameters only if examined simultaneously (IWC 2014a), this estimation is going to be done after some successful bio-logging attachments with simultaneous krill surveys. For this calculation, the mouth volume of the whales is necessary, therefore measurement of the mouth volume of the sampled whales will be conducted on board the mother research vessel.

3.2 Body conditions

The measurements of body condition (blubber thickness, girth, total fat weight) and stomach contents will be conducted in this research program in the same manner as in JARPA and JARPA II for the sampled whales (Konishi *et al.*, 2008, 2014a, 2014b). The blubber, ventral groove and intestinal fat will be weighed respectively. Then, for chemical analyses, blubber and internal fat are sampled from the entire body of the whales and the lipid content % of samples will be analyzed to calculate total lipid contents per individual. The total lipid content will be compared to body condition indicators, such as blubber thickness, girth measurements and chemical marker described below to assess their efficiency. Analyses on body condition will be based on the inner and outer layers of the blubber. As an initial feasibility test, the measurements of body conditions (blubber thickness, girth, total fat weight and some chemical markers) from Antarctic minke whales collected from this new research program will be used. Based on the Texel pollution workshop (IWC, 1999) the sample sizes would need to be at least 50 in each 'cell' where the cell will vary by important variables. At least 50 whales would be required for each sex and area.

3.3 Chemical assessment of body condition indicators

The body condition indicators such as blubber thickness, length of girth and total fat weight that can be obtained by lethal sampling have been used throughout JARPA and JARPA II periods (Konishi *et al.*, 2014, Konishi and Walløe, 2014). These data will continue to be collected as they will contribute to the Main Objective II. In 2014 the IWC SC agreed that the body condition and stomach content trends of Antarctic minke whales were decreasing significantly during the JARPA period (IWC, 2015 pp46).

In response to the suggestion by the NEWREP-A Review Panel, instead of retinol, the proponent plans to analyse the lipid content % of blubber throughout NEWREP-A research. At first, the total lipid content % of blubber from mid lateral position (near the position used in JARPA and JARPAII for the point measurement of blubber thickness), will be analysed since this position has been informative of changes of blubber thickness in relation to feeding season (Konishi *et al.*, 2008). The lipid content % of blubber will be compared to other body condition indicators such as blubber thickness and girth measurements. As an initial feasibility test, the measurements of body condition (blubber thickness, girth, total fat weight and the lipid content % of blubber) obtained from Antarctic minke whales collected under NEWREP-A will be used. At least 50 whales for each sex and sub area will be used for this analysis.

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Objective II(iv)

Ecosystem modeling

Part 1: Spatial interaction among baleen whales

1. Backgrounds and objectives

Spatial distribution of cetaceans have been widely studied using empirical (statistical) models in recent years such as generalized additive models (GAMs) (Beekmans *et al.*, 2010; Friedlaender *et al.*, 2006; Hedley *et al.*, 1999; Murase *et al.*, 2013; Williams *et al.*, 2006), maximum entropy models (MaxEnts) (Ainley *et al.*, 2012; Ballard *et al.*, 2012; Bombosch *et al.*, 2014; Friedlaender *et al.*, 2011) and Getis—Ord G statistics and spatial regression models (Santora and Reiss, 2011). These empirical models are commonly termed as species distribution models (SDMs). However, it was recognized that SDMs could be complex and often difficult to formulate, and development of guidelines and recommendations for modeling steps for cetaceans are now considered in the IWC/SC (IWC, 2014). Furthermore, interaction among cetaceans and krill in spatial context has not been studied extensively in the Antarctic with few exceptions (Friedlaender *et al.*, 2011; Murase *et al.*, 2014).

Though results of SDMS provide insight into the relationship between spatial distribution of cetaceans and their environment, mechanisms which drive the spatial distribution can be derived from them and the necessity of use of mechanistic models are recognized (Palacios *et al.*, 2013). Spatially explicit ecosystem models can be considered as spatial mechanistic models. Several types of spatially explicit ecosystem models have now been developed such as Atlantis (Fulton *et al.*, 2011), Ecospace module of Ecopath with Ecosim (EwE) (Steenbeek *et al.*, 2013), NEMURO and NEMURO.FISH (Kishi *et al.*, 2011) and a spatial ecosystem and populations dynamics model (SEAPODYM) (Lehodey *et al.*, 2008). However, there has been no attempt to construct spatially explicit ecosystem models in the Antarctic specifically mainly targeting cetaceans.

The objectives of this proposed research are (1) constructing appropriate SDMs of cetaceans as well as krill based on the guidelines developed by the IWC/SC, (2) developing appropriate methods to measure interactions among cetaceans and krill in a spatial context using output from SDMs and (3) constructing spatially explicit ecosystem models which mainly focus on cetaceans.

2. Methods

2.1 Species distribution models (SDMs)

A various types of SDMs such as a generalized linear model (GLM), GAM, Ecological Niche Factor Analysis (ENFA), Random Forest (RF), Boosted regression tree (BRT) and MaxEnt will be tested using data obtained by upcoming surveys. Data of the IDCR/SOWER, JARPA and JARPAII will also be used in the analysis. Appropriate methods to formulate and compare results among models will be considered throughout the exercise. The results will also be compared with the results obtained by model based abundance estimation methods which are currently underdeveloped in the IWC/SC (IWC, 2014).

Development of appropriate methods to measure interaction among cetaceans and krill in a spatial context will be considered using the results of SDMs. Both estimated abundance and presence/absence of cetaceans will be considered in the proposed research. The magnitude of interactions among cetaceans and krill will then be measured using the results.

2.2 Spatially explicit ecosystem models

Construction of a spatially explicit ecosystem model using Ecospace module of EwE will be attempted as we have already constructed an initial EwE model in Area IV (Kitakado *et al.*, 2014). The output of

SDMs will be considered as input data of the ecosystem models. Construction of other spatially explicit ecosystem models will be considered once modeling of Ecospace module of EwE is completed. The results of the spatially explicit ecosystem models will be compared with the results of multi-species-production ecosystem models.

Initially, we will start to design EwE-Ecospace models in Area IV and V using available data mainly obtained from the literature. The models will be updated once data from the ecological research are available. Environmental factors such as global warming and ocean acidification which are poetically affect Antarctic marine ecosystem (Constable *et al.*, 2014) will be considered in the updated models as external forces as in the case of Ainsworth *et al.* (2011). Coupling EwE-Ecospace and lower trophic models such as NEMURO (Kearney *et al.*, 2012) will be considered as an ambitious attempt.

3. Distinctive features and expected outcome

The proposed research will contribute the development of guidelines for SDMs and model based abundance estimation method which are actively considered in the IWC/SC. The results of SDMs will contribute to understand temporal changes of spatial distributions of cetaceans in the survey area. SDMs will contribute to development of more advanced spatially explicit ecosystem models as the output of former models will be used as input of latter models. Comparisons between results of spatially explicit ecosystem models multi-species-production ecosystem models will contribute to refine ecosystem models in the Antarctic. Overall results under this research will provide fundamental information on the Antarctic marine ecosystem and contribute to management of cetaceans as well as other living species.

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Objective II(iv)

Ecosystem modelling

Part 2: Investigation of ecosystem dynamics in the Antarctic Ocean

1. Background and objectives

This exercise was intended to refine the earlier approach by Mori and Butterworth (2006) and Kitakado *et al.* (2014), but some twists were added into their basic system of equations for multiple predators (some baleen whales and seals) and single prey species (krill). Although the multi-species production model is simpler than the approach of whole-of-ecosystem modelling, it still has many unknown parameters. To reduce the number of parameters to be estimated, some bioenergetic and allometric reasoning by Yodzis and Innes (1992) were incorporated for the predator species. The maximum consumption is linked to the body weight of the predators.

This sort of model has the potential to estimate the extent of competition between the baleen whales. For example, since the population sizes of other larger baleen whales such as blue, fin and humpback whales declined through commercial harvesting, there may have been a state of krill surplus in the Antarctic. This surplus could be utilized by the Antarctic minke whales, and since that time the minke whales may be still above their pre-exploitation level.

The works on ecosystem modelling is primarily to model competition among whale species and interaction with the krill, but it would also contribute to development of management objective and procedures. In the existing RMP implementation, any species interaction is not explicitly taken into account. However, some multispecies adjustment would contribute to improvement of operating models and harvest controls to make more effective use of this resource. The multi-species production model, which development is undergoing now in the Antarctic Ocean, must be a good tool to precede such process.

For example, the model could give some information on changes in carrying capacity and provide a scenario for running "RMP" simulation to see if the current single species CLA may or may not work under the changes in carrying capacity. This was a reason for our further development of multi-species production models. In this regard, it might be possible to extend our research objective toward work for linking ecosystem modelling with management with consideration on species interaction etc.

In this sub-objective, the following ancillary research items are handled:

- (a) Elucidation of history of the ecosystems in the Antarctic using multi-species production models
- (b) Future projection of ecosystem using multi-species production models
- (c) Contribution to the RMP trial specification using ecosystem models
- (d) Examination of performance of the current RMP under ecosystem interactions

2. Specification of multi-production models: an example

One of basic models to be used for multiple predators (some baleen whales and seals) and a single prey species (krill) is given as follows:

$$P_{t+1,i} = P_{t,i} - M_i P_{t,i} + E_i f_i(B_t) P_{t,i} - \alpha_i P_{t,i}^2 - C_{t,i}, \quad (i = 1, 2, ..., n)$$

$$f_i(B_t) = c_{\max,i} \frac{B_t^{\theta}}{H_i^{\theta} + B_t^{\theta}},$$

$$B_{t+1} = B_t + r B_t \left(1 - \frac{B_t}{K}\right) - \sum_{i=1}^n f_i(B_t) P_{t,i},$$

$$(1)$$

where $P_{t,i}$ and B_t are respectively the population sizes for predators and prey species, and M_i , E_i and α_i are the natural mortality rate, the energy conversion factor and a parameter for the density dependence for predator species i, respectively. The function "f" is a so-called functional response, which is used to link the predator with prey. The parameters, $c_{\max,i}$ and H_i , are respectively a maximum consumption per the i-th predator in the feeding season and the prey biomass when the consumption of the i-th predator drops to a half of $c_{\max,i}$. These become the Type II and Type III functional responses for cases of o_{i} and o0, respectively. The parameters o1 and o2 are the ones used in the usual logistic production model.

As shown above, Mori and Butterworth (2006) assumed a density-dependence within each species, but there may be some other density dependence by other predator species. It may not be a useful way to simply include all other predators' effects in each species dynamics. Instead, one possible way is to modify the available prey abundance by considering a food competition among the predators as follows:

$$P_{t+1,i} = P_{t,i} - M_{i} P_{t,i} + E_{i} c_{\max,i} \frac{(\omega_{t,i} B_{t})^{\theta}}{H_{i}^{\theta} + (\omega_{t,i} B_{t})^{\theta}} P_{t,i} - \alpha_{i} P_{t,i}^{2} - C_{t,i},$$

$$B_{t+1} = B_{t} + r B_{t} \left(1 - \frac{B_{t}}{K} \right) - \sum_{i=1}^{n} c_{\max,i} \frac{(\omega_{t,i} B_{t})^{\theta}}{H_{i}^{\theta} + (\omega_{t,i} B_{t})^{\theta}} P_{t,i},$$

$$\text{where } \omega_{t,i} = \frac{s_{i} P_{t,i} W_{i}^{0.75}}{\sum_{j} s_{j} P_{t,j} W_{j}^{0.75}}.$$

$$(2)$$

Here, s_i is a selectivity of the *i*-th predator to the prey species, and it is assumed to be 1 for all the predators in this application.

This model is further developed to incorporate the species-internal completion (density dependence) into the functional response itself as in the following formula:

$$P_{t+1,i} = P_{t,i} - M_i P_{t,i} + E_i c_{\max,i} \frac{(\omega_{t,i} B_t)^{\theta}}{H_i^{\theta} + (\omega_{t,i} B_t)^{\theta} + \phi_i P_{t,i}} P_{t,i} - C_{t,i}.$$
(3)

This model can be regarded as an extension of Beddington (1975)'s model. The carrying capacity for the krill and the parameters for the density dependence for the predators are solved through an assumption of an equilibrium condition to reduce the number of parameters to be estimated.

Though the multi-species production model is simpler than other ecosystem modelling approaches like whole-of-ecosystem modelling, it still has lots of unknown parameters. To reduce the number of parameters to be estimated, some bioenergetic and allometric reasoning by Yodzis and Innes (1992) was incorporated over predator species. The maximum consumption and the rate of increase are linked with the masses of predators (W) as

$$c_{\max,i} = \beta W_i^{0.75},$$

$$\mu_i = \gamma W_i^{-0.25}.$$
(4)

Then the energy conversion factor was calculated as

$$E_{i} = \frac{\mu_{i}}{c_{\text{max }i}} = \frac{\gamma W_{i}^{-0.25}}{\beta W_{i}^{0.75}} = \delta W_{i}^{-1}.$$
 (5)

Note that the exponent in the rate of increase "-0.25" may not be suitable for marine mammals (Duncan *et al.* 2007). Using a dataset of Duncan *et al.* 2007, the exponent for cetaceans is roughly estimated as -0.0315 and this was used as a proxy value for predators.

The past and newly available data on time series of abundance for all the component species and nutrition indices for the Antarctic minke whales are used for the estimation of the parameters using the maximum likelihood method.

$$\log \hat{P}_{t,s} = \log P_{t,s} + \varepsilon_{t,s}, \quad \varepsilon_{t,s} \sim N(0, c\hat{v}_{t,s}^2).$$

$$\log \left(\text{nutrition index}\right)_t = \log a + \log f_{\text{Minke}}(B_t) + v_t, \quad v_t \sim N(0, \sigma^2)$$

Table 1. Summary of estimated and fixed parameters in the model (EC: equilibrium condition)

	Parameter	Treatment	
	Initial population size in 1900/01 (P_0)	Estimated separately for predators	
Predators	Natural mortality (M)	Fixed at some values	
(5 species)	Energy conversion (E)	Estimated as a common coefficient (δ)	
	Density dependence (α or ϕ)	Solved by EC	
Prey (krill)	Initial population size in 1900/01 (B_0)	Estimated	
	Intrinsic rate of increase (r)	Fixed at 0.5	
	Carrying capacity (K)	Solved by EC	
Functional response	Maximum consumption (c_{max})	A fixed common coefficient (β)	
	Prey biomass at which a half of predator consumption attains (H)	Heffmated cenarately	
	Exponent (θ)	Fixed at 1 or 2	
Nutritional indicators	Constant coefficient (a)	Estimated	
	Model errors variance	Estimated	

3. Some remarks

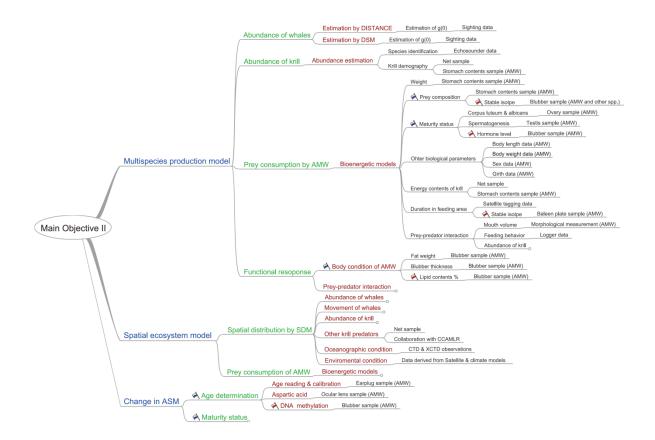
This proposed research program includes the two Main Objectives I and II, but these are not necessarily separated. The Main Objective I intends to contribute scientifically to the future application of the *IST*s for the Antarctic minke whales through the improvement of biological and ecological parameters and update of its stock structure hypotheses. The Main Objective II of this program also includes the relevance to the management through investigation of possibility of building ecosystem models. Those works constitutes an important bridge between the two Main Objectives. More specifically, in the existing RMP Implementation, species interactions are not taken into account explicitly neither in the trials not for the catch limit. However, some adjustment to reflect this multispecies aspect would contribute to improvement of operating models and harvest controls. For example, the ecosystem model could give some information on changes in carrying capacity and provide a scenario for running "RMP" simulation to see if the current single species CLA may or may not "work" (e.g. may fail to allow harvests levels that are perfectly possible without putting a population at risk) under the changes in

estimated carrying capacity. In this regard, this research objective could contribute to the future improvement of the RMP in terms of species interactions.

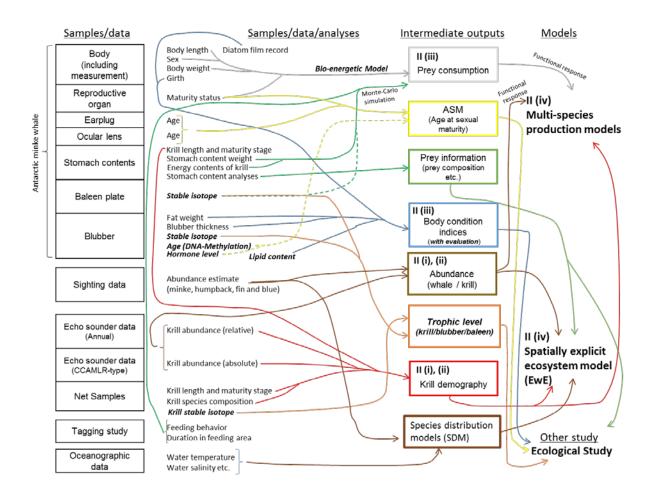
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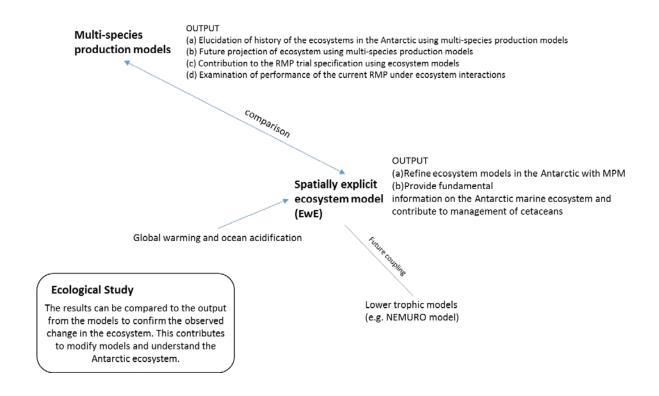
A: Schematic chart showing the nested structure of models, basic analyses and data/samples required to accomplish the Main Objective II of NEWREP-A. Analyses added to NEWREP-A or adjusted based on recommendations by the NEWREP-A Review Panel are red flagged. Analyses requiring methodological development to compare lethal and non-lethal methods are blue flagged. Redundant child nodes (*e.g.* maturity status) are hidden (hidden nodes are indicated as circles). ASM: age at sexual maturity; AMW: Antarctic minke whale; DSM: density surface model; SDM: species distribution model.



B: A schematic flow chart showing the steps from required samples and data to modeling exercises to achieve the Main Objective II in NEWREP-A. Samples and analyses recommended by the NEWREP-A Review Panel have been included (italic and bold letters). Non-lethal methods will be evaluated in the NEWREP-A program. The absence (as yet) of established analyses or sampling is illustrated by dotted lines.



C: Investigation of the structure and dynamics of the Antarctic marine ecosystem through building ecosystem models under the Main Objective II in NEWREP-A.



Sample size calculation based on detectability of changes in the age-at-sexual maturity

1. Introduction

The age-at-sexual maturity (ASM) is an important indicator of ecological and nutritional conditions for whale populations. Examination for the Antarctic minke whales based on the transition phase in the ear plugs has shown that the ASM declined by around 3 years old from 1955 to 1970, when nutrition is likely to have been better than in the past, because of the lowest food competition with the other baleen whales which were highly depleted over that period. However, since then the trend has become stable or slightly increasing (Bando *et al.* 2014). This might be attributed to the increased food competition and reflected to the decline of carrying capacity for the Antarctic minke whales (Pastene *et al.*, 2014, Punt, 2014), possibly due to the recent recovery of other competing species. The ASM has an impact on the number of mature animals (especially females), and therefore possible future increases in the ASM would affect the productivity and population dynamics of the Antarctic minke whales, which in turn has implications for management.

Here, first analyses are conducted to see if the changes in the ASM have taken place over the last two decades using maturity data taken from past JARPA and JARPA II samples. Secondly based on some specified values for the change in ASM, simulation studies are performed to indicate the sample size needed to detect the changes.

2. Conditioning: estimation of ASM50 using past JARPA and JARPAII data

Materials and Methods

The data employed are the maturity status of whales taken during the JARPA and JARPA II programs as determined from examination of the reproductive organs. The cohorts which are considered in this analysis are those from 1980 to 2000 (which are well sampled in the data available). Let Y_{ta} be the observed number of female mature animals aged as a among the total sample size of age a animals in the t-year cohort (say N_{ta}). Y_{ta} is assumed to be distributed as a binomial distribution $Bin(N_{ta}, p_{ta})$, where p_{ta} is the maturity probability. The maturity p_{ta} is further assumed to be a logistic function of age, in which the function's point of symmetry corresponds to the age at 50% sexual maturity (say ASM50). The ASM50 is also considered to be cohort-specific or a function of cohort, where this difference is just a way of handling cohort as a continuous or categorical variable in terms of a generalized linear model.

More specifically, the following three models are applied:

```
Model 1: logit p_{ta} = \alpha + \beta t + \gamma a, (common intercept, cohort as a continuous variable)
Model 2: logit p_{ta} = \alpha + \beta_t + \gamma a, (common intercept, cohort as a categorical variable)
Model 3: logit p_{ta} = \alpha_t + \beta_t + \gamma a, (cohort specific intercept, cohort as a categorical variable)
```

where the intercept α_t controls the range of maturity, β or β_t is a coefficient for cohort t and determines the ASM50, and γ is a coefficient for the continuous value of age a. The estimate of ASM50 is calculated by the usual transformation:

$$ASM 50_t = -\frac{\alpha_t}{\gamma} - \frac{\beta_t}{\gamma},$$

and its standard error is computed by the delta method.

For the model with continuous cohort values, the identification or otherwise of a yearly change in the ASM50 is determined simply by whether or not the value estimated is significantly different from zero. In contrast, for the model with categorical cohort dependence, a further linear regression analysis needs to be conducted using the estimated ASM50 values. In this case, a linear model with inverse-variance weighting is employed.

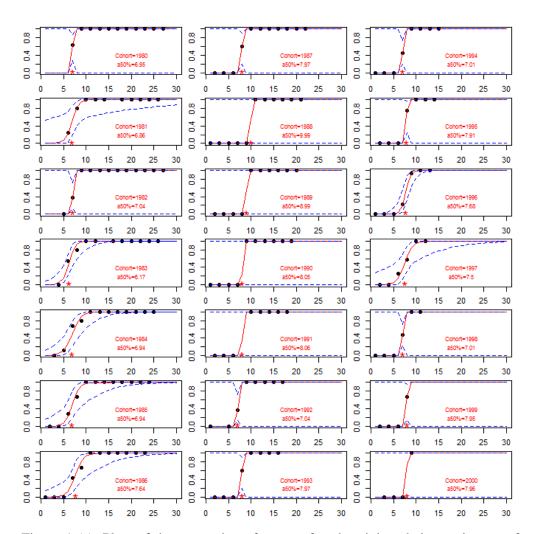


Figure 1-(a). Plots of the proportion of mature female minke whales against age for Area IV. Filled circles are observed mature proportions, and the red (solid) and blue (dotted) lines are estimated proportion mature curves with 95% confidence intervals, respectively, based on Model 3 with year-specific ASM50 and slope.

As shown in Figure 1-(a) and 1-(b), the range of ages which affect the analysis is narrower than the range covered by the plots. In fact, parameter estimates are not changed if data for a shorter range from ages 4 to 13 are used (see Figure 2 for an example).

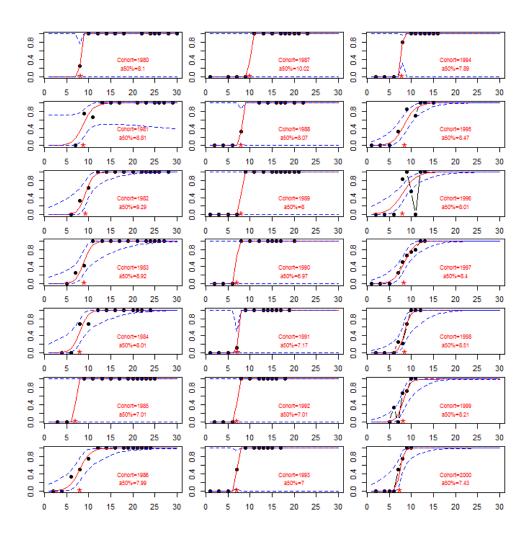


Figure 1-(b). Plots of the proportion of mature female minke whales against age for Area V.

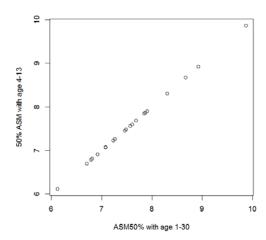


Figure 2. Comparison of ASM50 estimates in Area IV derived from the age range 4-13 against those from the age range 1-30. The model applied is a categorical cohort model with a common intercept for each set of data.

Results

In Area IV,. the estimated year effects were positive and significant for all the cases. The range of recent incrase over 1980-2000 cohorts is from 0.038 (/year) to 0.076 (/year). On the other hand, In Area V, negative year effects were evident. In addition, in each case the SE is roughtly 0.02, and therefore the effect was not significant. In Area V, there is a mixture of the two putative stocks (I- and P- stocks) and the boundary between them may have been changing over time (*e.g.* Kitakado *et al.* 2014). Furthermore, although the maturity status of female whales does depend on the latitude in the Ross Sea, the habitat and ice conditions also change year by year. In this sense, it is not easy to infer the potential effect size from Area V at this stage.

Table 1. Results for the estimation of year effect for the ASM50. The numbers in parentheses are the standard errors. The values of comparable AIC are shown.

Year effect	No. parameters in GLM	AIC in GLM
0.0385 (0.0173)	3	127.34
0.0380 (0.0231) *	22	122.41
0.0440 (0.0178)	3	90.00
` ′	•	104.15
0.0431 (0.0170)	17	104.13
-	42	-
0.0759 (0.0130)*	14	-
Year effect	No. parameters in GLM	AIC in GLM
-0.0344 (0.0206)	3	188.18
-0.0347 (0.0257)*	22	191.18
-0.0331(0.0221)	3	157.71
-0.0331(0.0215)*	17	160.63
_	42	_
	0.0385 (0.0173) 0.0380 (0.0231) * 0.0440 (0.0178) 0.0431 (0.0170)* - 0.0759 (0.0130)* Year effect -0.0344 (0.0206) -0.0347 (0.0257)* -0.0331(0.0221)	0.0385 (0.0173) 3 0.0380 (0.0231) * 22 0.0440 (0.0178) 3 17 - 42 0.0759 (0.0130)* 14 Year effect No. parameters in GLM -0.0344 (0.0257)* 22 -0.0331 (0.0221) 3

3 (partial data) -0.0255(0.0497)*

^{*}Note that the year effect in Model 2 is estimated by the linear model based on the ASM50 estimated by the GLM model. No year effect was estimated for the Model 3 with the full data beceause most of point estimates have large uncertainty. Rather, results with a partial data set with SE less than 1 is meaningful.

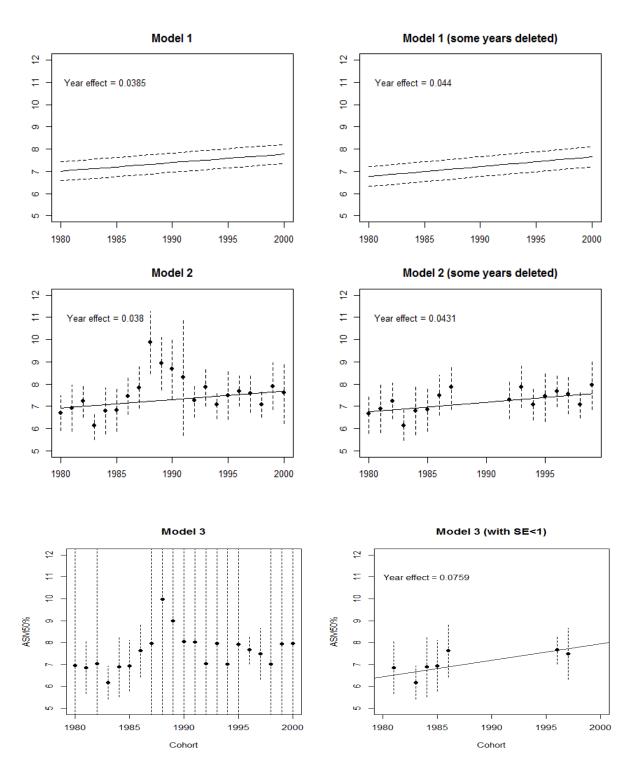


Figure 3. Estimated ASM50 for Area IV for each of the three models. For Models 1 and 2, results based on only cohort years with proportion mature indices greater than 0 and less than 1 are also shown because essentially only those cohorts contribute to the estimate of the year effect. In Model 3, a similar treatment with a criterion of the standard error less than 1 was applied.

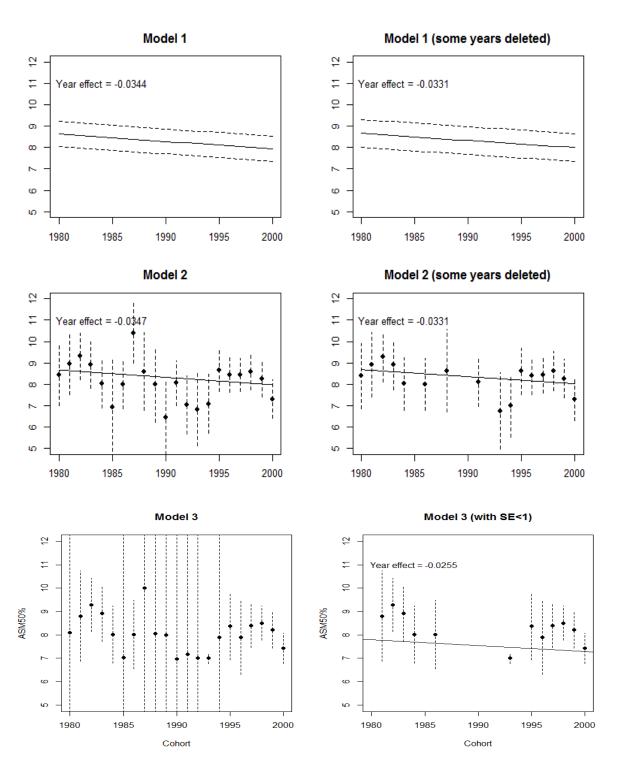


Figure 4. Estimated ASM50 for Area V for two models.

Simulation for investigation of sample size

In the conditioning process for simulation setting, the potential effect size, a yearly change in the ASM50, ranged from 0.038 to 0.076 in Area IV. These values are slightly smaller than those provided by the transition phase analysis. Figure 5 showed time series of the mean age at sexual maturity for I-stock and P-stock female animals (Bando *et al.* 2014). Although it has been suggested that a fringe effect should be considered in the analysis, even after allowing for this coarsely, it is clear that the ASM declined by at least around 3 years from 1955 to 1970, which means that the yearly change was -0.2 per year. The extent of change observed in the past is greater than for recent estimated values, but if such a relatively fast change were to happen again, this would have implications in a management context and would also warrant further investigation of species competition and habitat utilization for prey. However, it may not be plausible that the change starts in the rate of 0.2 per year. Rather it seems to start more slowly. Here, target effect sizes are set as the maximum value of estimates of recent yearly changes (c.a. 0.075 per year) and a half value of the past extent (0.10). As a reference, another effect size, 0.05 (/year) is also considered.

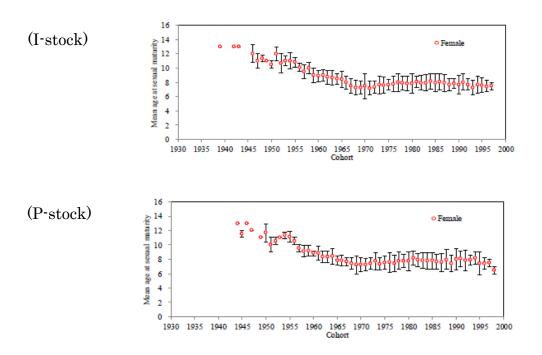


Figure 5. Time series of the mean age at sexual maturity for I- and P- stocks female animals (extracted from Bando *et al.* 2014). Open circles are the point estimates and the error bars are ranges of one sigma (SD).

In the simulation for this sample size investigation, it is assumed that female age composition in each cohort is observed. Given the current sampling plan, in which sampling is conducted 2nd year in each Area, maturity statuses for odd ages are observed in the odd year classes, and those for even ages in the even year classes. It is also assumed that the age composition of sampled whales is stable and same as the average of 1980-2000 cohorts (see Figure 6).

In the simulation, the following scenarios, with sample size for age range 4-13, and the performance measures indicated below are considered.

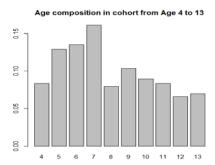


Figure 6. An assumed stable age composition of the year class (calculated from cohorts of 1980-2000)

Simulation scenarios

- 1) The effect size (yearly change in age-at-50%-sexual maturity, ASM50) is assumed to be 0.05, 0.075 or 0.10. ASM50 for the initial year class is 7.5 and it increases by the effect size under consideration for each successive year under consideration.
- 2) Maturity range (MR=75%-25%) is assumed to be constant at 1.5, which is similar with the value estimated
- 3) A total of simulation 1,000 replicas are generated.

Sample size

Female animals of age from 4 to 13 are used as the sample size in the simulation. The total sample size required is a reciprocal of the sampling rate of female whales of age from 4 to 13. The proportion of females with age range 4-13 years old in the yearly total catch varied from 0.10 to 0.19 during JARPA and JARPA II period in area IV (Table 2). The mean value was 0.15. Therefore, the sampling rate of 0.15 was used for calculation of the total sample size.

Table 2. Number and proportion of female Antarctic minke whales with age range 4-13 years old sampled during JARPA and JARPA II surveys in Area IV.

Research year	Number of	Number of females	Proportion of females
	total samples	with age range 4-13	with age range 4-13
1987/88	272	46	0.17
1988/89			
1989/90	326	48	0.15
1990/91			
1991/92	288	43	0.15
1992/93			
1993/94	330	44	0.13
1994/95			
1995/96	330	40	0.12
1996/97			
1997/98	328	33	0.10
1998/99			
1999/2000	330	51	0.15
2000/01			
2001/02	330	52	0.16
2002/03			
2003/04	330	51	0.15
2004/05			
2005/06	573	99	0.17
2006/07			
2007/08	235	35	0.15
2008/09			
2009/10	101	19	0.19
2010/11			
		Mean	0.15

Performance measures

1) RRMSE (relative root mean squared error, a proxy for CV because the ASM50 can take negative values)

$$RRMSE = \frac{1}{\theta} \sqrt{\frac{\sum_{i=1}^{n} (\widehat{\theta_i} - \theta)^2}{n}},$$

where θ is the true parameter (ASM50, here), and $\widehat{\theta}_i$ is the *i*-th estimate among the *n* replications in each sample size.

2) Power of the statistical test for a null hypothesis of no year effect in age-at-sexual maturity

Results of simulation and conclusion

The simulation results are shown in Figure 7 (a)-(c). Of course, for the larger the effect size, the smaller sample size is required to detect it. In the case of the smallest effect size (0.05), detection probability is quite small unless a larger sample size is made. However, such an effect may not be very urgent change to detect within next 12 years' research plan. In contrast, a bigger change has a larger implication if we miss to detect. In case of the case with effect size = 0.10, in which result is shown in Fig 7-(c), a (female) sample size of 50 is a reasonable size in terms of the RRMSE (about 0.2 and less improvement if the sample size is increased) and statistical power (over 90% if 5% significance level is applied). This sample size also guarantees a reasonable statistical power (close to 80% for 5% significance level) and precision for the case of effect size 0.075 as shown in Figure 7(b).

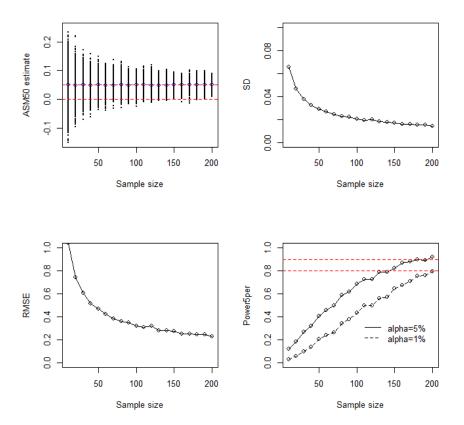


Figure 7-(a). Simulation results for the effect size 0.05 (/year). In the top left panel, 1000 estimates of ASM50 are plotted against the sample size of 4-13 female animals. Blue points are the mean of estimates in each sample size. In the top right and bottom left panels, the standard deviations and relative root mean squared relative errors are drawn to correspond to the sample size under consideration. The bottom right graph shows the statistical power for two significance levels (5% and 1%). The dashed red lines are 80% and 90% power levels.

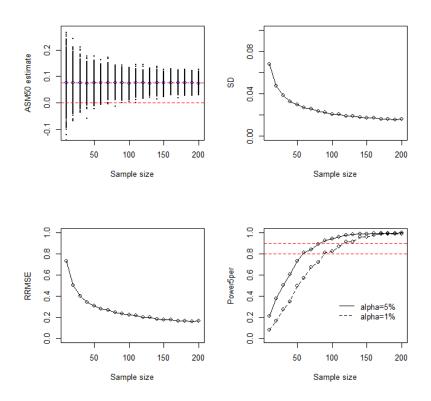


Figure 7-(b). Simulation results for the effect size 0.075 (/year).

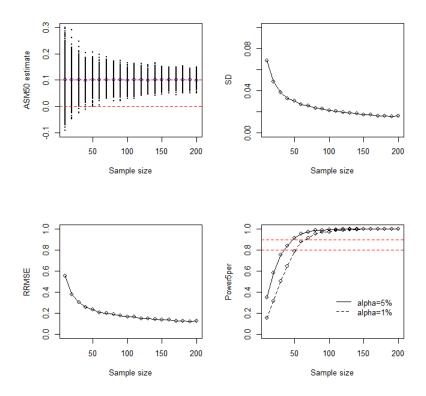


Figure 7-(c). Simulation results for the effect size 0.10 (/year).

It should be noted that no ageing errors were considered in the conditioning and simulation analyses. As studies on the age-determination errors have been conducted (e.g. Kitakado et al. 2013), it might be possible to take this into account in the sample size investigation. The age-reading error matrix is estimated as conditional probabilities for the expected observed ages given a true age. However, to account for this error in this study, an inverse transformation consisting of a conditional distribution for true ages given an observed age is performed. This transformation causes large uncertainty because the true age distribution is unknown in practice. The current age-reader has been in charge of age-reading since JARPAII, and the same person will continue to provide age readings in this program, so that consistent age-reading performance will be maintained.

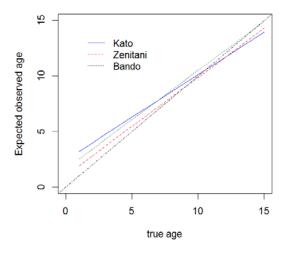


Figure 8. Age-reading errors estimated in Kitakado et al. (2013)

Conclusion

Based on the simulation above, an annual sample size of 50 female animals of age from 4 to 13 is suggested as appropriate. Therefore, taking account of the average sampling rate of those animals (i.e. 0.15), the total sample take required is calculated as 50/0.15=333.

The calculation is based on the estimated effect size from the direct observation of maturity in Area IV as well as the past knowledge based on the longer time series from the transition phase for both the stocks. While the analysis of the past ASM data has shown that its changes could be varied by stock in terms of the magnitude and direction, it could be assumed that it is biologically not plausible for different stocks of the Antarctic minke whale to have radically different responses to external stimulus such as environmental fluctuations and food availability, especially when those stocks have diverged only recently as in the case of the Antarctic minke whale stocks. Therefore it is not unreasonable to hypothesize that the sample size calculated from I-stock can be used as a proxy to P-stock or any other stock that could occur in Areas IIIW and VIE at this stage. The result of the calculation itself remains applicable to other Areas covered by this research program to detect whether a substantial change in the ASM50 has occurred. It should be noted that, however, the change in the ASM must essentially be pursued stock-wise. This proposed research program assumes that the samples will alternately be taken from areas IV (or IV+III) and V (or V+VI), and the annual sample size may be adjusted based on the acquired/updated knowledge on the stock structure after the first 6 years of this research for investigating a better allocation of sample sizes.

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