Stock structure of North Atlantic common minke whale (*Balaenoptera acutorostrata*): a multidisciplinary review of the Icelandic Research Program results

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ABSTRACT

In response to the report of the Independent Expert Panel following the Icelandic Research Program (IRP) review, we present here an integrated paper incorporating information from genetics, morphometry, telemetry, biological parameters, stable isotopes, fatty acids and pollutants (recommendation 12.1.2 (1) of the report of the expert workshop (SC/65A/Rep3)).
The integrated results emphasized the fact that the genetic markers employed did not detect any significant genetic structure among the IWC stock boundaries while other biological information suggested that stock structure might exist on a local scale (north vs. south Iceland), on a large geographical scale (feeding grounds) and that, some subtle structure might exist on breeding grounds.

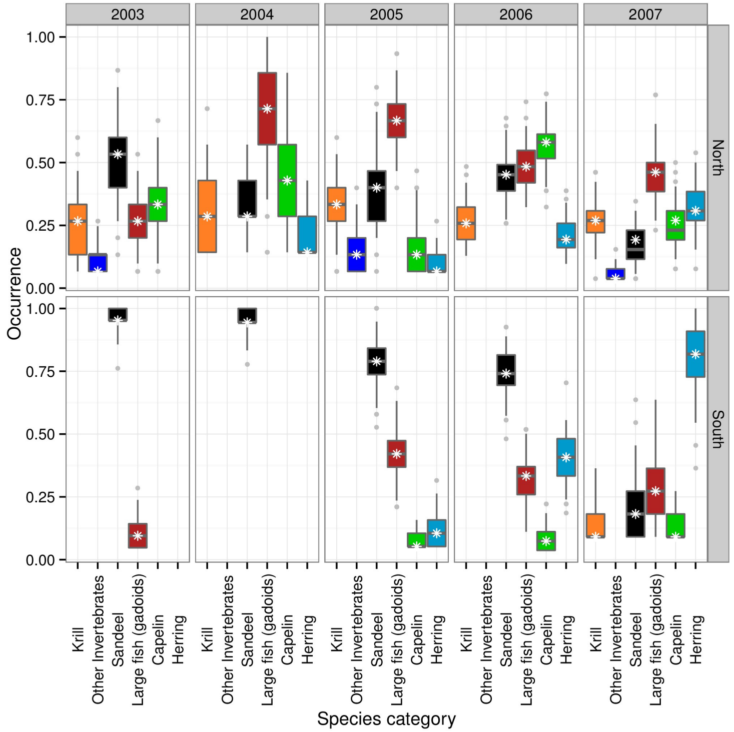
INTRODUCTION

In response to the report of the Independent Expert Panel following the Icelandic Research Program (IRP) review, we present here an integrated paper incorporating information from genetics, morphometry, telemetry, biological parameters, stable isotopes, fatty acids and pollutants (recommendation 12.1.2 (1) of the report of the expert workshop (SC/65A/Rep3): Produce a fully integrated paper incorporating the information from genetics, morphometry, telemetry, biological parameters, stable isotopes, fatty acids and pollutants). This multidisciplinary approach is based on the Annex D of the Report of the Expert Workshop to Review the IRP, presenting the summary of potential indicators of structure developed by proponents during the workshop.
The results of the different techniques potentially giving useful information for stock structure were considered regarding three potential indication, stock structure at local feeding grounds (Icelandic waters), at large scale feeding grounds (North Atlantic), and at breeding grounds.

FEEDING ECOLOGY AND ENERGETICS

The main feeding ecology study (SC/F13/SP2_Rev) showed that animals captured off the North and South of Iceland had a significantly different diet composition, indicative of a very strong local feeding structure. The diet of animals caught in the southern part of Iceland was mainly composed of sandeel while animals caught in the North mainly had more diverse diet composition. In addition, despite the changes observed in the diet of animals during the studied period, the difference of diet between animals caught off the south and north of Iceland still differed markedly (see Figure 1). This geographic difference in diet is consistent with the difference in the marine environment between north and south Icelandic coastal waters (SC/F13/SP2_Rev).

a)



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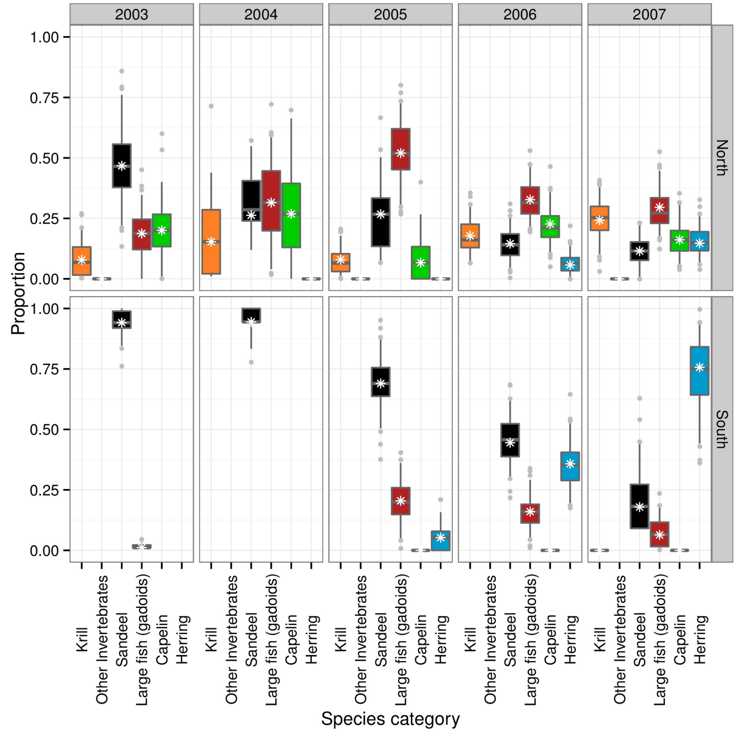

Figure 1 [Figure 3a and3b of SC/F13/SP2_Rev]. Boxplots of spatial and temporal variation in diet composition of minke whales expressed as **a)** frequency of occurrence (FO) and **b)** reconstructed weight (RW). The point estimate is indicated by a white star while the solid gray lines through the boxes represent the bootstrapped medians. The box indicates the inter-quartile range and the whiskers 95% confidence intervals. Any further outlying data points are indicated as points. See SC/F13/SP2 for information on the calculations of the indices and distribution of animals studied.

Information retrieved from the energetics study (SC/F13/SP10_Rev) tend to support the feeding ecology one as animals captured from the South of Iceland were fatter than animals captured from the North. It should be noted that the differences between north and south Icelandic waters can be confounded by the spatial and temporal segregation of minke whales by sex within the study area (SC/F13/SP14).

GENETICS

In the last decades, genetic markers have frequently been used in an attempt to resolve uncertainties related to stock structure within and/or between management regions in Cetaceans (Andersen *et al*., 2003; Jorde *et al*., 2007; Kanda *et al*., 2007), with or without success. During the IRP, two main papers were presented on Stock structure using genetic markers (a suite of 16 microsatellite loci and mtDNA), the paper of Anderwald *et al.* (2011) (also called SC/F13/SP16 in the review workshop, and herein) and SC/F13/SP17. However, the results of these two studies are not directly comparable as they performed analyses based on a different set of microsatellite loci and a different set of individuals collected during the IRP. The total number of individuals used during these two studies differs also quite a lot. SC/F13/SP17 used 535 and 565 individuals respectively for microsatellite loci and mtDNA respectively, i.e. approximately double numbers than in SC/F13/SP16.
In essence the objectives of the two studies also differ slightly. SC/F13/SP17 aimed at studying the spatio-temporal genetic structure at feeding grounds while SC/F13/SP16 intended to study the geographical structure and infer the potential mixing of minke whale at different feeding grounds. The analytical approach developed in both studies is therefore quite different, with the use of different software and, therefore simulation methods.

Despite the different number of samples and microsatellite loci used, and the somehow different objectives, both studies can nevertheless be interpreted in a similar way, although the conclusions of each study differed.

At microsatellite loci, SC/F13/SP17 failed to show any significant differences among samples collected at different geographical regions, and the level of differentiation among samples was not significant and too low (F_{ST} of 0.0003; 95% CI: -0.0005-0.0011) to apply any Bayesian cluster analyses. SC/F13/SP16 confirmed the lack of significant differentiation among samples from different geographical regions as only F_{ST}'s among North Atlantic and Japan exhibited significant values. However, SC/F13/SP16 performed further analyses detecting two possible cryptic subpopulations roaming in the North Atlantic supported by analyses subsequent to an initial Bayesian cluster analysis and suggested that further analyses should be performed to assess the accuracy of the finding.

At mtDNA both studies revealed a lack of genetic structure in the North Atlantic although SC/F13/SP17 found two haplotypes groups but there was a lack of concordance between geographic and phylogenetic position of mtDNA haplotypes, i.e. the distribution of the two haplotypes groups did not correspond to any IWC stocks boundaries. The results obtained with mtDNA during SC/F13/SP17 can either reflect a residual ancestral polymorphism or a "recent" isolation of two populations at breeding sites, which roam through large parts of the North Atlantic Ocean during the feeding migration as suggested by Palsbøll (1989) and Bakke *et al*. (1996) results. SC/F13/SP16 found that for some regional sample sets mtDNA differentiation was concordant with the proposed two putative populations based on nuclear DNA markers.

To conclude, genetic studies during the IRP did not reveal any clear genetic pattern and are calling for additional studies using other more appropriate genetic markers, such as Single Nucleotide Polymorpshims (SNPs). So far, the genetic studies revealed that the IWC stock boundaries are not genetically distinguishable. Although firm conclusion can not be drawn from such results, they might not necessarily indicate that the North Atlantic minke whale is composed of only one population, but merely that the genetic markers used were not sufficient to confirm possible cryptic genetic structure. Recent relatedness analyses based on microsatellite loci (SC/F13/SP20_Rev) suggested that relatedness pairwise comparisons were significant between individuals occurring at different feeding areas, spanning from few years to few generation and suggesting that within/among generation, gene flow among areas separated by the current IWC stock boundaries is quite high, therefore confirming the lack of genetic differentiation.

MORPHOMETRY

Although morphometric measurements have been suggested to be useful for stock discrimination (Cadrin, 2000), very few studies have been conducted on Cetaceans so far (Hakamada and Bando, 2009; Christensen *et al*., 2010; Nicolosi and Loy, 2010). During our study (SC/F13/SP19), multivariate statistical analyses of 17 morphometric characters were performed in order to evaluate potential heterogeneity between predefined common minke whales stock unit areas in the North Atlantic (West Greenlandic, East Greenlandic, and Icelandic waters, North Seas and Norwegian and Barents Sea), including data from Christensen *et al*. (1990). Considering sexes separately (analyses conducted separately for males and females), the multivariate analysis showed that morphometric measurements were significantly different between all samples collected at different geographical regions, but the overlap of the data was so pronounced that individual assignment to their specific geographical region of origin was impossible. The overlap of the different measures was also too large between North and South of Iceland to find any significant differences.

TELEMETRY

Due to the development of promising tracking methods, migration studies have been flourishing in recent decades, with particular emphasis on multiple year patterns (Egevang *et al*., 2010; Klaassen *et al*., 2011; Matthews *et al*., 2011; Olifiers *et al*., 2011; Vardanis *et al*., 2011). In this context, data from telemetry observation allow the investigation of migration patterns of marine mammals and can be relevant for stock structure.

During the experiment set-up in Icelandic waters from 2001-2010, we were able to track the movement of six whales of which three moved out of Icelandic waters during autumn. Among the tagged individuals, a total of 5 tagged animals in the North of Iceland exhibited high site fidelity during all the period, therefore suggesting that migration among feeding areas and IWC stock boundaries rarely occurred (Figure 2 [Figure 2 of SC/F13/SP18]).

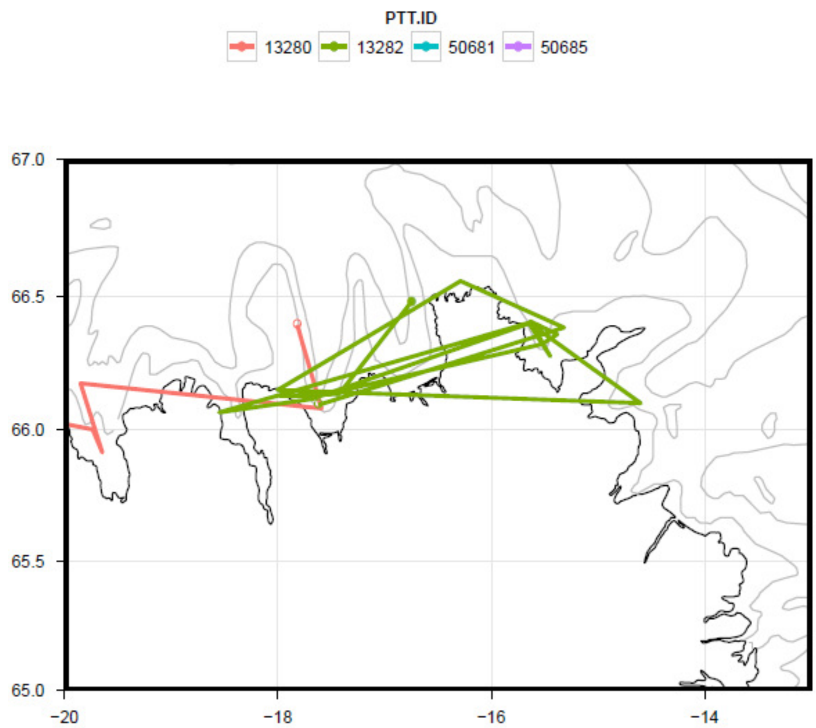

Figure 2 [Figure 1 of SC/F13/SP18]. Example of satellite tracking of two minke whales tagged (#50683 and #50686) in the North of Iceland in August 2001. Animals for which signals (2 out of 4) were recovered were constantly visiting fjords in the North of Iceland with no apparent offshore activities.

Although more tagging should be performed to firmly draw conclusion about migration pattern among feeding areas and IWC stock boundaries, the present results tend to suggest that migration rarely occurred among IWC stock boundaries.

Migration pattern could also be investigated during migration from Iceland to potential breeding grounds for a total of 3 animals (see Figure 3). The tracks of these three minke whales that departed from Icelandic waters provide the first indications of the migration route and possible winter destination of minke whales summering in Icelandic waters. All three followed an offshore route in the middle of the Atlantic heading south. Contact was lost early with two of the whales but they were all heading in the same direction. The third whale, where contact was maintained past the Azores, continued south to a position in the mid Atlantic at about 28^{o}N. This represents the longest tracking record for a minke whale worldwide both in terms of distance travelled (3,700 km) and time (100 days).

Although, more tagging should be performed to draw any firm conclusion about the route of migration of minke whales to their potential breeding areas, tagging data tend to reveal that routes of migration stayed within the Central North Atlantic area for the three-long-migration information we obtained. However, it is interesting to note that individuals 50683 and 3960, and individual 50686, also staying in the same are clearly took different routes East and West of the area. Further tagging might, indeed, based on these results indicate that several breeding grounds exist, and although we can not conclude at present, telemetry seems to be a promising method despite the current absence of relevant information related to stock structure due to the low number of retrieved data.

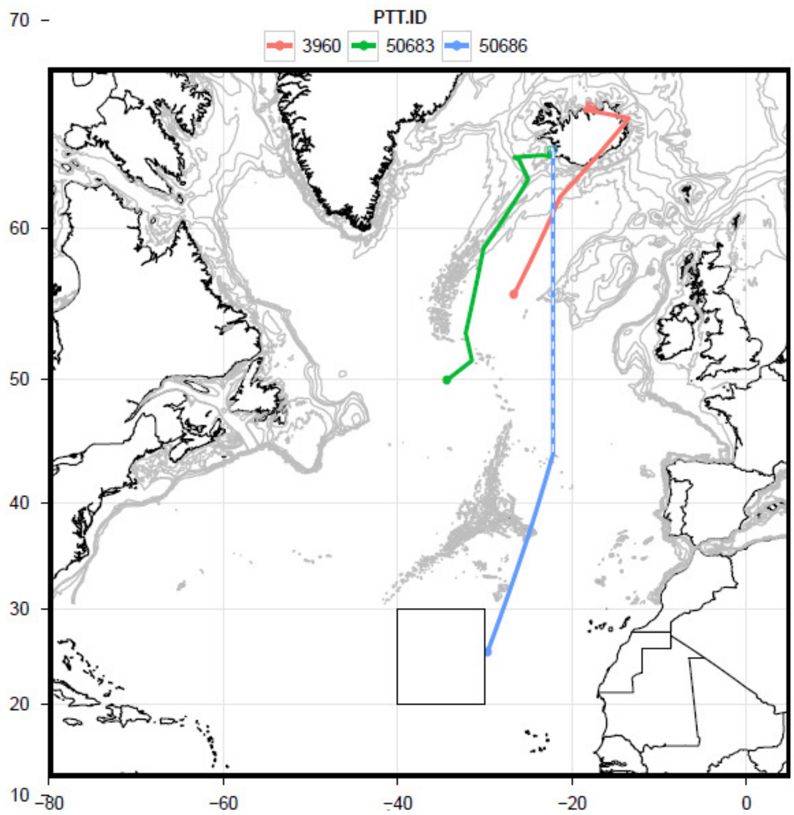

Figure 3 [Figure 2 of SC/F13/SP18]. Satellite tracking of two minke whales tagged (#50683 and #50686) in Faxafloí bay from 27th August – 23rd of September effort and of one minke whale tagged (#3960) in the North of Iceland. All three animals turned southwards after having spent some time along the continental shelf of Iceland.

STABLE ISOTOPES

Stable isotopes have recently been suggested as potential indicators of ecological separation of minke whale as they reflect possible differences in foraging behaviour among geographical areas (Born *et al*., 2003). Comparison of the overall levels of stable carbon and nitrogen isotope ratios in muscle samples analysed during the IRP, revealed differences between the Icelandic minke whale and minke whale from other geographical regions in the North Atlantic (as well as from the North Pacific). This supports segregation of these animals of the North Atlantic in the weeks or months prior to the sampling (Figure 4), *i.e.* potentially at breeding ground, and/or during the migration route and/or before capture at feeding grounds. However, Hobson *et al*. (2004) argued that the turnover rate of muscle tissue of minke whales was about a month, which might indicate that the isotope values observed in Icelandic minke whale in the later part of the summer reflect feeding in Icelandic waters. In addition, the values for the Antarctic minke whales (*B. bonaerensis*) differed markedly from the values of minke whales from both the North Atlantic and the North Pacific, where the Antarctic minke whales were much more depleted in δ^{13}C (δ^{13}C being -24.6±0.3 ‰ (95% CI)) and higher in δ^{15}N (14.6±0.5 ‰ (95 % CI) than their North Atlantic and North Pacific counterparts (Endo *et al*., 2012).

In addition, analyses of relationship of isotope ratios in various minke whale tissues to a set of explanatory variables using generalised linear mixed model gave indications of difference in carbon isotope ratios between the North and South regions of Iceland. This suggests low migration rates between these areas during the feeding season in Icelandic waters.

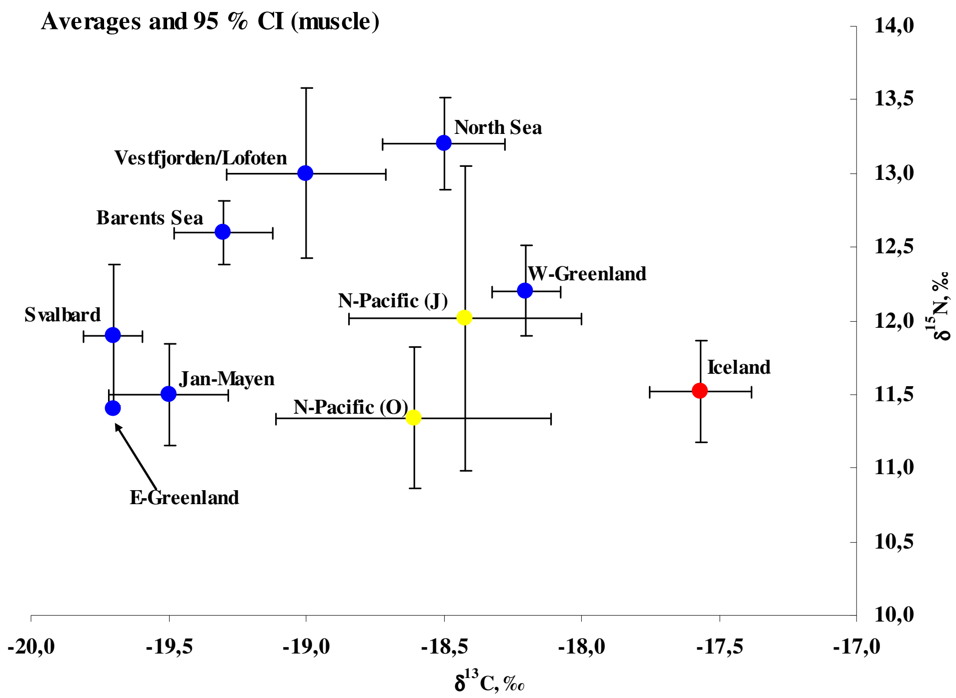

Figure 4. Comparisons of the ratio $\partial^{13}C$ and $\partial^{15}N$ levels in minke whale muscle in the North Atlantic and North Pacific. Values for North Atlantic regions were retrieved from Born *et al.* (2003) while values for the North Pacific were obtained from Endo *et al.* (2012)

FATTY ACIDS (FA)

FA technique is commonly used to provide information on the diet (Kharlamenko *et al.*, 2001; Dahl *et al.*, 2003; Pétursdóttir *et al.*, 2012). Such investigations provide an integration of prey consumed over periods ranging from weeks to months (Fry, 1988; Dalsgaard *et al.*, 2003). Even though FA signature of predator and its prey resembles each other they are very seldom identical as both diet and endogenous synthesis influences the FA profiles of the depot fat.

Recently new analytical tool i.e. fatty acid (FA) profiles of depot fat, has been used to assist with analyzing population structure of marine mammals (i.e. Grahl-Nielsen *et al.*, 1993; Iverson *et al.*, 1997 and Møller *et al.*, 2003). Møller *et al.* (2003) suggested that FA signatures in the depot fat (i.e. blubber) of minke whales could be used for studying population structure. The FA profiles in outer and inner blubber of minke whales from the North Atlantic (Møller *et al.*, 2003) and genetic study performed on the same samples (Andersen *et al.*, 2002) were in agreement. These samples were collected during a single whaling season, from 6 May to 31 October in 1998 in the North Atlantic region: West Greenland (WG), East Greenland (CG), Jan Mayen (CM), Svalbard (ES), Barents Sea (EB), Vestfjorden/Lofoten (EC) and the North Sea (EN) but the neighboring Canadian and Icelandic waters were not included in this study as whales were not harvested there at that time. Multivariate analyses on the fatty acid profiles proposed the following three regions: Greenland (WG, CG), the Northeast Atlantic (CM, ES, EB, EC) and the North Sea (EN). Further work on population studies of minke whales in the North Atlantic should however include minke whales from the Icelandic and Canadian waters as well. Tshe recent study about FA compositions of minke whales in Icelandic waters (SC/F13/SP4_Rev) could possibly contribute to such a study. Admittedly, further studies on the affect of both environmental and internal factors, i.e. metabolism of the blubber, and the turnover rates of the FAs, that affect the FA composition of the depot fat, are necessary to develop this technique in population studies.

POLLUTANTS

Trace elements and organic micropollutants have been used to characterize stocks of minke whales (*e.g.* Kunito *et al.*, 2002; Hobbs *et al.*, 2003; Born *et al.*, 2003; Born *et al.*, 2007). During the IRP, a series of both organic pollutants and trace elements were analysed in several tissues of the Icelandic minke whale stock (SC/F13/SP21; SC/F13/SP22_Rev; SC/F13/SP23_Rev; SC/F13/SP24; SC/F13/SP25; SC/F13/SP26). Furthermore, the radioactive Cs-137 was also analysed in muscle tissue (SC/F13/SP30).

Trace elements

As regards the trace elements, no significant difference was found between feeding areas north and south of Iceland in any of the 11 trace elements analysed in skin, livers, kidneys, and muscle tissues (As, Se, Hg, Cd, Pb, Cu, Cr, Ni, Mn, Fe and Zn) (log-transformed tissue levels linearly regressed on length, sex, and area). Gonads were also analysed for cadmium but the levels of cadmium did not show difference between the two feeding areas off Iceland. The hepatic and renal levels of cadmium were higher in the Icelandic stock than other stocks in the Northeast Atlantic (Figure 5). There seems to be a decreasing gradient of hepatic cadmium levels from Southeast Greenlandic and Icelandic minke whales towards lower levels to the east and south, lowest levels occurring in the North Sea. Renal levels correlate with hepatic levels and therefore similar picture is obtained for renal and hepatic concentrations of minke whales of the Northeast Atlantic (SC/F13/SP23_Rev). In addition, hepatic cadmium levels in Antarctic minke whales (Kunito *et al.*, 2002) are much higher than those in the North Atlantic (SC/F13/SP23_Rev). In spite of higher hepatic and renal cadmium levels, the muscular cadmium levels are low in the Icelandic stock, similar to the lowest levels in the North Sea (Figure 6). Therefore, the Icelandic stock differs from other stocks in cadmium levels alone. As a further differentiation between these stocks of the North Atlantic, the mercury levels of the Icelandic minke whales (muscle, liver, and kidneys) are lower in the Icelandic stock than those in the minke whales from Southeast Greenland, the stock most similar to the Icelandic minke whale stock in hepatic and renal cadmium levels (Figure 7). Mercury levels in skin, kidneys, livers, and muscle correlate in the Icelandic minke whales.

On the basis of Hg, Se and Cd in various tissues of North Atlantic minke whales, Born *et al.* (2003) found that for female whales, significant differences in at least one long-term diagnostic element occurred between several areas where the following population substructure was inferred: (a) West Greenland, (b) a central group represented by whales from Jan Mayen, (c) a northeastern stock including the Barents Sea, Svalbard and coastal Norway, and (d) the North Sea. In this work of Born *et al.* (2003), males appeared to fall into similar groupings as females but due to smaller sample sizes of males, fewer significant differences occurred between areas.

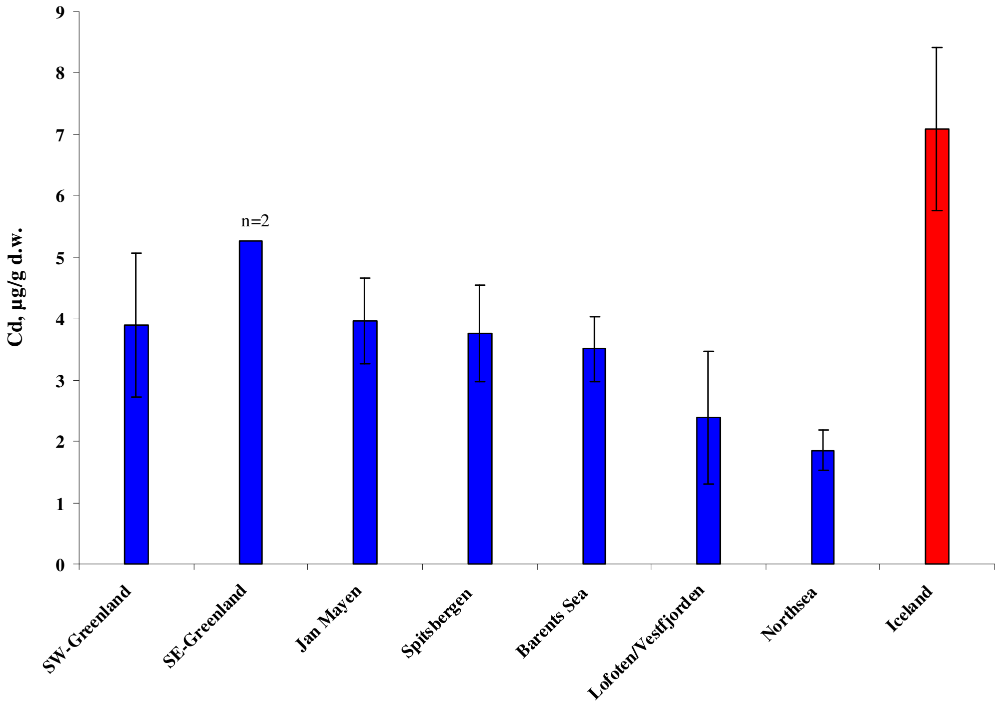

Figure 5. Cadmium levels in livers of minke whales from the NE-Atlantic on a dry weight basis. The bars on each column represent the 95% confidence intervals. The blue column data derive from Born *et al.* (2003).

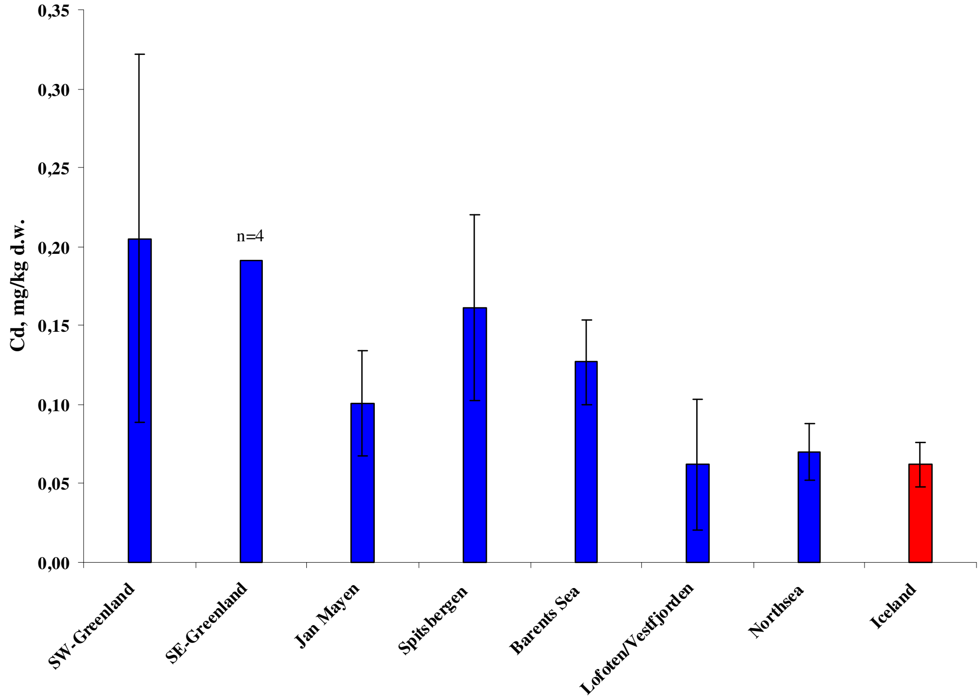

Figure 6. Cadmium levels in muscle tissue of minke whales from the North Atlantic on a dry weight basis. The bars on each column represent the 95% confidence intervals. Data of blue columns derive from Born *et al.* (2003).

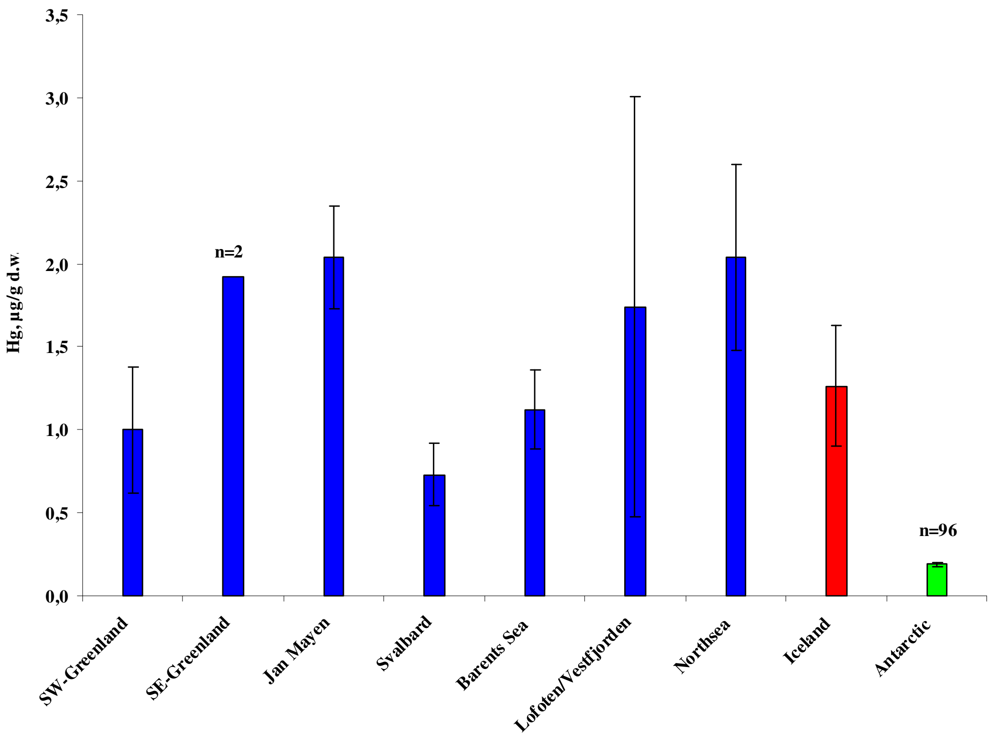

Figure 7. Mercury in livers of minke whales from the North Atlantic and the Antarctic on a dry weight basis. The bars on each column represent the 95% confidence intervals. Blue column data derive from Born *et al.* (2003) while the data for the Antarctic derive from Honda *et al.* (1987).

Organic contaminants

As concerns the legacy POPs (PCBs (#28, 31, 52, 101, 105, 118, 138, 153, 156, 170 and 180), DDTs (p,p'-DDE, p,p'-DDD, p,p'-DDT, and o,p'-DDT), HCHs (α-, β-, and γ-HCH), HCB, Chlordanes (trans-nonachlor, α-chlordane, γ-chlordane, and oxychlordane), toxaphenes (26, 50, and 62), and dieldrin) and PBDEs (47, 99 and 100), no significant difference was found between the foraging areas north and south of Iceland where log-transformed contaminant data (blubber, liver and muscle on lipid weight) were regressed on length, sex, and area (SC/F13/SP22_Rev). The most striking difference between the Icelandic stock and other stocks in the North Atlantic is revealed in the concentrations of toxaphenes in their tissues (Figure 8). Generally, it may be concluded from studies on levels of organic contaminants in blubber of minke whales worldwide, that the pattern of the legacy POPs and the three PBDEs in the blubber of Icelandic minke whales is different than that found in other minke whale stocks of the North Atlantic and very different than that of the North Pacific and Antarctic (SC/F13/SP22_Rev).

The main methoxylated PBDEs (6-MeO-PBDE47 and 2′-MeO-BDE68), most likely of natural origin, were of noticeably lower levels in blubber of minke whales off Norway than the levels in minke whales sampled off Southwest Greenland and West Iceland (SC/F13/SP21). No other minke whale stocks of the North Atlantic were studied with respect to MeO-PBDEs but these results suggest that these compounds should be explored further in other stocks as well.

Studies of Hobbs *et al.* (2003) on minke whale stocks of six regions of the North Atlantic (not Iceland or Canada) showed that the general similarity in mean levels of ΣPCBs, ΣDDT and ΣCHL, as well as mean principal components analysis scores, among minke whales sampled at Jan Mayen, Svalbard, Vestfjorden/Lofoten, the North Sea, and the Barents Sea suggests that the whales are quite mobile and may feed in multiple areas within the northeastern Atlantic. However, the differences in concentrations of ΣPCBs and ΣDDT suggest that West and Southeast Greenland minke whales of low loadings may represent one group of whales, which are distinct from both the Jan Mayen minke whales and those from other IWC defined stocks in northern European waters, especially the North Sea.

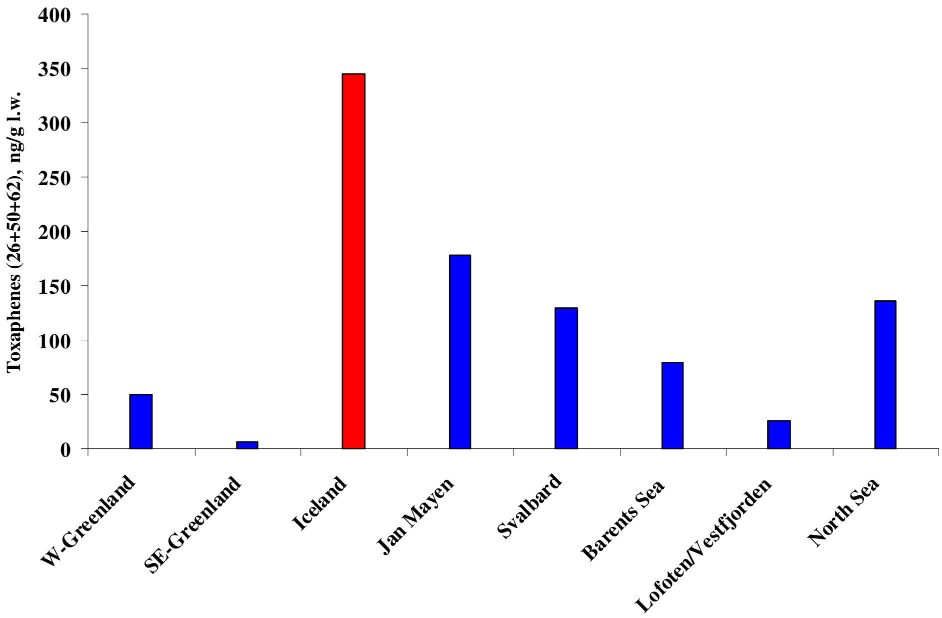

Figure 8. Toxaphenes (sum of 26, 50, and 62), mean values on a lipid weight basis, in blubber of minke whales from different areas. The blue column data derive from Gouteux *et al.* (2008).

Studies of Born *et al.* (2007) using Canonical Discriminant Analysis (CDA) on mercury and cadmium in muscle, liver, and kidney, and eight organochlorines (Ocs) and four unsaturated fatty acids in blubber of minke whales of the six regions of the North Atlantic of Hobbs *et al.* (2003) above indicated that a multi-elemental approach based on long-term deposited compounds (with different ecological and physiological pathways) can be used for identification of subpopulations of marine mammals. Based on this approach, four subpopulations of North

Atlantic minke whales were identified: 1) a West Greenland group, 2) a Central Atlantic group of minke whales from Jan Mayen, 3) a Northeast Atlantic group (Svalbard, Barents Sea, and Northwestern Norway), and 4) a North Sea group.

Caesium-137

Levels of caesium-137 have been analysed in muscle tissue of minke whales from 7 regions of the North Atlantic but not including Iceland and Canada (Born *et al.* 2002). Levels of Cs-137 in muscle of Icelandic minke whales from 2003-2004 (SC/F13/SP30) are significantly lower than the levels found in muscle of most minke whale stocks from the North Atlantic 1998 (Figure 9). However, the levels found in minke whales from Svalbard are not significantly different from the levels of the Icelandic minke whales.

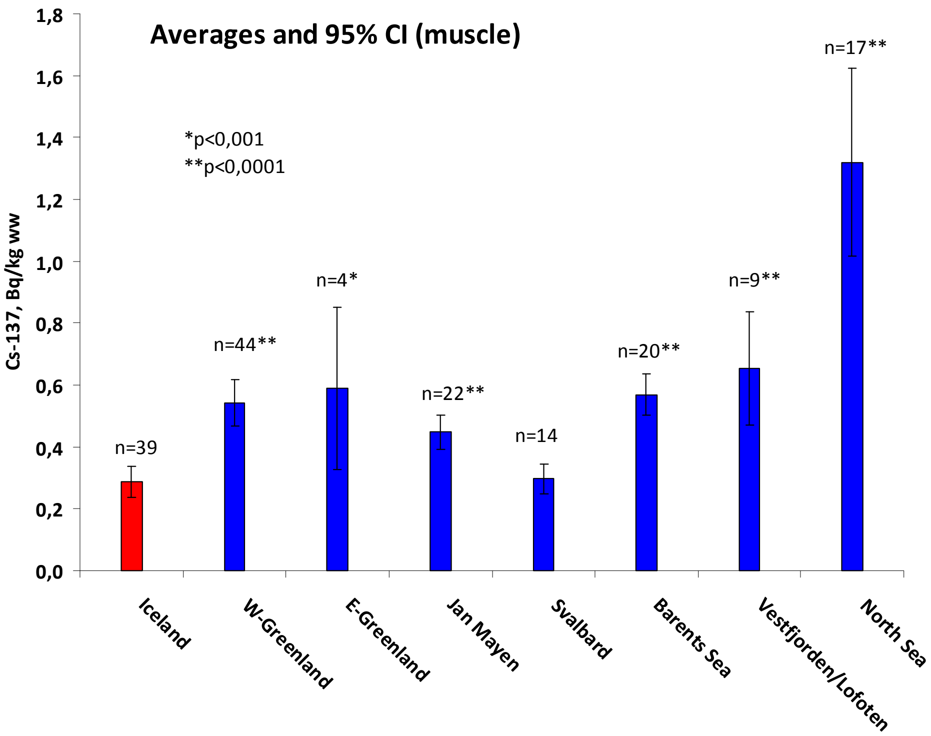

Figure 9. Cs-137, mean values with 95% CI, in muscle tissue of minke whales from different areas. The blue column data derive from Born *et al.* (2002).

BIOLOGICAL PARAMETERS

Variation of biological parameters among different geographical areas has often been used as a mean to define stock structure. During the IRP, several biological parameters indicated significant differences among minke whale caught in Icelandic waters and whale caught in other geographical areas such as in Norwegian waters.

For example, SC/F13/SP13, which presented a report on blood testosterone and progesterone concentrations of the North Atlantic minke whale (*Balaenoptera acutorostrata*) during the feeding season in Icelandic waters, suggested that the rise in the serum testosterone (T) measurements in the Icelandic sample happens during the days 215–243 of the year while the rise in the Norwegian measurements appears during days 180–220 of the year, i.e. earlier. Such a difference between individuals of these different IWC stock boundaries' populations indicates that the reproductive cycle of the individuals feeding in these two distinct geographical areas is different. Therefore, it might indicate that whales caught in Iceland and Norway represent two distinct populations breeding at a single breeding ground not overlapping in time, or, alternatively, that they represent two distinct populations reproducing at two different geographical breeding grounds. Although the conclusion cannot really be drawn concerning the breeding grounds until further analyses are done, the rise in serum testosterone clearly indicated a stock structure between Iceland and Norway. This stock structure was supported by the body mass which also differed between minke whale caught in Iceland and minke whale caught in Norway. Icelandic minke whale generally exhibited lower weights than Norwegian minke whale for similar length (SC/F13/SP11).

Finally, sex segregation was also observed at the local scale i.e. between North and South of Iceland (SC/F13/SP14), and was consistent to what has been observed in other areas. Thus, off W-Greenland (Laidre *et al*., 2009) and Norway (Tore Haug pers. comm.) the females ratios increase with latitude as in this study.

PARASITES

Parasites data have been recently suggested as potential indicators of stock structure in several species (Oliva and Gonzalez, 2004; Timi *et al*., 2005; Williams and Lester, 2006) but have rarely been used in Cetaceans (but see Dailey and Vogelbein, 1991; Balbuena and Raga, 1994). A review on the use of parasite as indicators of social-structure and stock identity of marine mammals suggested that parasites might indeed provide valuable insight into the behavioural features of marine mammals, but that stock structure indication are rather limited due to the lack of control over sampling procedure and the paucity of information about the biology of the parasites on their host (Balbuena *et al*., 1995).

During the IRP, the Anisakids (nematodes) complex was studied (SC/F13/SP28). However, although the abundance and prevalence of Anisakids might represent a promising method related to stock structure, the data retrieved during our study could not reveal any significant variation in these estimates in Icelandic minke whale (see SC/F13/SP28), mostly due to the lack of comparative data in minke whale from other areas of the North Atlantic, and the lack of information about the biology of the parasite. However, one clear conclusion was that the parasite indices (abundance and prevalence) were not significantly different at a local scale, i.e. no significant distinction could be done between minke whales from the North and the South of Iceland.

CONCLUSION

Although one of the most common approaches to discriminate stocks is the use of genetic markers, several multi-approach studies have emphasized the importance and the need of multidisciplinary investigation (Cadrin *et al*., 2010; Higgins *et al*., 2010). During the IRP, the genetic markers employed did not detect any significant genetic structure among the IWC stock boundaries while other biological information suggested that stock structure might exist and that, in addition, some subtle structure might exist on breeding grounds. The absence of genetic structure does not necessarily mean that the North Atlantic common minke whale is composed of only one population, but merely that the genetic markers used were not sufficient to confirm possible cryptic genetic structure. In this case, biological information such as reproductive status, pollutants levels, parasites fauna, morphometry and telemetry might give a better insight into migration pattern and potential isolation of populations within and between IWC stock boundaries. Unfortunately, a global conclusion on stock structure, based on the data retrieved from the IRP, is limited by the external availability of data, i.e. data on parasite, fatty acids, or other techniques, were not available from other geographical regions/IWC stock boundaries. Nevertheless, some of the retrieved information might be relevant for future development of research.

Evidence of structure at a local feeding ground (Iceland):

Several of the studies conducted during the IRP suggested a medium to large structuring at the local scale, i.e. mainly between minke whales caught in the north and south of Iceland. One of the largest evidence came from the primary objective of the research proposal, the feeding ecology (SC/F13/SP2_Rev). The diet composition of whales caught in the north of Iceland was more diverse than the ones caught in the South (mainly sandeel and herring, SC/F13/SP2_Rev). Despite the observed temporal changes in the diet composition from 2003 to 2007 a clear difference between these two areas still persisted throughout the period. The energy storage study (SC/F13/SP10) also reflected this observation and the differences related to the changes in diet composition at a local scale. Stable isotopes also showed significant differences between whales caught in the North and South of Iceland (SC/F13/SP3), but might reflect feeding ecology in the past month instead of current diet composition (when the whales were caught). It might therefore supports the structuring at local scale and also indicate different route of migration of these two groups.

Finally, for the strong evidence of local structuring, sex segregation was also observed between north and south of Iceland. This segregation may be connected to a potential difference in feeding strategies of males and females or even by reproductive classes. The data collected in this feasibility study (n=190) are however insufficient to make firm conclusions on that point.

Two other methods showed differences at local feeding ground but were rated as low indication of structure (1) either due to statistical reasons or due to a small number of observation. Indeed, morphometric measurements (SC/F13/SP19) showed differences between whales caught north and south of Iceland, but the overlap of the measurements was so high that it led to non-significant differences and to the impossibility of assigning individuals to their region of origin. Likewise, telemetry (SC/F13/SP18) indicated that two individuals tagged in the north of Iceland exhibited site fidelity during the period of investigation (late summer/autumn). Although satellite tracking has great potential to be informative on local movements and stock structure, the method has proven more difficult to apply to common minke whales than other baleen whales (SC/F13/SP18). Thus, out of around 20 tags used, informative data on local movements were retrieved from only five individuals. However, each successful tag can provide large amount of useful information and the MRI will thus continue its effort in this field.

Evidence of structure at feeding grounds (large geographical scale):

Several of the studies conducted during the IRP also suggested a medium to large structuring at feeding grounds (large geographical scale), i.e. between IWC stock boundaries.

One of the strongest evidence came from the pollutants levels, especially from cadmium, mercury, toxaphenes and Cs-137 (SC/F13/SP30).

The levels of stable isotopes (SC/F13/SP3) also suggested some structure at feeding grounds (large scale), especially when examining the ratio $\partial^{13}C$ and $\partial^{15}N$ levels in muscles. Most of the geographical regions exhibited significant differences in this ratio.

Biological parameters such as the rise in the serum testosterone (SC/F13/SP13) and the variation in body mass (SC/F13/SP11) also suggested significant differences among Icelandic and Norwegian minke whale. These results might indicate that animals living in these two areas display different reproductive cycle and might belong to different reproductive groups using the same breeding ground with temporal variation or using different breeding grounds.

Although satellite trackings have been limited in scope thus far (SC/F13/SP18), it is noteworthy that during the total combined tracking period of 370 days (8 individuals combined), no whale was observed crossing the IWC Schedule boundaries for the Central North Atlantic stock.

Finally, two other methods suggested structure at large feeding grounds but were rated as low indication of structure, i.e. the morphometry (SC/F13/SP19) and the genetic (SC/F13/SP16). The morphometric measurements revealed significant differences among minke whales from different geographical areas, but due to a large overlap of the measurements, assignment of individuals to their group of origin was impossible. One microsatellite loci study also suggested some cryptic genetic structure among feeding areas in the North Atlantic but also recommended further analyses to clearly picture the observed pattern.

Evidence of structure at breeding grounds:

Very few analyses performed during the IRP allow for conclusion about potential structure at breeding ground. However, information such as:

1) the difference in the rise in the serum testosterone between the minke whales from Iceland and Norway (SC/F13/SP13),
2) the levels of pollutants such as cadmium, mercury, toxaphenes, and Cs-137 (SC/F13/SP22_Rev, SC/F13/SP23_Rev, SC/F13/SP30), which might at least partly also reflect isolation on breeding ground,
3) the differences in stable isotopes which might reflect the diet composition on breeding ground (SC/F13/SP3),
4) the weak indication of cryptic structure reported with microsatellite (SC/F13/SP16) and mtDNA (SC/F13/SP17),

might give valuable information for future research regarding investigation on potential structure at breeding grounds.

In addition, the satellite tracking experiments have already provided the first indication of a potential winter breeding ground in the deep waters off West Africa, but obviously further trackings are needed to verify the migration routes and winter destination(s) (SC/F13/SP18).

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Annex 1[1]

Summary of potential indicators of structure developed by proponents

The Table considers structure at two levels, i.e. indication of structure at breeding grounds and of structure in the IWC Schedule stock boundaries for the Central North Atlantic stock (feeding ground). In the latter, the table considers local structure (north vs south Iceland) and at larger scale (North Atlantic). Explanatory comments are indicated to clarify the observed results.

Methods	Breeding	Feeding Local	Feeding Large	Comments
Feeding ecology				
Diet – SP2_Rev	na	3	na	
Energetics				
Energy storage – SP10_Rev	na	2	na	Differences related to change in diet composition north vs south.
Genetics				
Microsatellite – SP16	1	na	1	Possible indication of two breeding populations, distribution of these two groups across the North Atlantic shows differences in IWC stock boundaries areas.
Microsatellite – SP17	0	0	0	No genetic pattern observed (different loci than SP16).
mtDNA – SP16	0	na	0	
mtDNA – SP17	1	0	0	Two groups of haplotype detected with no geographical partitioning.
Relatedness – SP20_Rev	0	0	0	High rate of related individuals across the North Atlantic. Age and geographical location information are crucial.
Morphometry – SP19	na	1	1	But non-significant pattern due to too large variance in estimates
Telemetry – SP18	0[§]	1[†]	*	5[†] individuals tagged in the North exhibited site fidelity during all the period. [§] All individuals migrating south stayed within the Central North Atlantic area (370 days of tracking for 8 whales).
Stable Isotopes – SP3	*	2	*	Reflects feeding in the past.
Fatty acids – SP4_Rev	*	0	*	Reflects feeding in the last 1-6 months.
Pollutants – SP21, SP22_Rev, SP23_Rev, SP25, SP30				
Cadmium	2	0	3	Differences in concentration in Iceland and other areas.
Mercury	2	0	2	Differences in concentration in Iceland and other areas.
Toxaphene	2	0	3	Differences in concentration in Iceland and other areas.
Polychlorinated naphthalenes	2	0	3	Differences in concentration in Iceland and other areas.
PBDEs-BDE47	2	0	3	Differences in concentration in Iceland and other areas.
Cs-137	2	0	3	Differences in concentration in Iceland and other areas.
Biological parameters				
Reproductive seasonality – SP13	1	0	2	Differences between the rise in the serum testosterone measurements in Norway and Iceland.
Variation in body mass – SP11	na	0	2	Differences between Norway and Iceland.
Sex segregation – SP14	0	2	0	Sex segregation in Iceland is similar to what is known in other areas.
Parasites (Anisakids) – SP28	na	0	na*	

na: information not available. * Promising methods despite the current absence of relevant information related to stock structure. “0” denotes the absence of detected structure, “1” low indication of structure, “2” medium indication of structure, “3” strong indication of structure.

[1] This is a revised version of the Annex D of the Independent Expert Panel report (SC/65A/Rep3).