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Fatty acids in the blubber of minke whales (*Balaenoptera acutorostrata*) -stratification and relation to diets

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Preface

First of all I want to thank my advisers Tore Haug, Sonnich Meier and Ulf Lindstrøm, and the Institute of Marine Research (IMR) which have given me the opportunity to do this thesis. The advisers have given me help with all practical issues, and have carefully read drafts and given me advice through the entire writing process.

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Lars Aage Gade-Sørensen

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Abstract

The fatty acid (FA) composition of 37 minke whales (*Balaenoptera acutorostrata*) from 4 different areas, Spitsbergen, Bjørnøya, Vesterålen and the North Sea was determined. Stratification between inner, middle and outer blubber was present. Most saturated fatty acids (SFAs) and monounsaturated fatty acids (MUFAs) with 20 and 22 carbon atoms had a higher relative amount in the inner blubber while short-chain MUFAs with 14, 16, 17 and 18 carbon atoms had a higher amount in the outer blubber. Whales from the different areas had a different fatty acid composition in inner blubber layer, but a more similar FA composition in outer blubber layer. The inner blubber layer were found different from all potential prey species, even though prey species from expected and observed diets were found with most similar fatty acid composition as the inner blubber layer. Fatty Acid Trophic Markers (FATMs) indicating the copepod *Calanus sp*. based food-webs, were found in high relative numbers with all whale samples from all areas, FATMS indicating diatoms were found with Spitsbergen/Bjørnøya samples, and FATMs indicating dinoflagellates were found with samples from Vesterålen and the North Sea. FA profiles have the potential in bio monitoring minke whale diet, but more study of influence of metabolism in FA incorporation of blubber are needed.

List of abbreviations used in the thesis:

FA = fatty acid

FATM = Fatty Acid Trophic Marker

FAME = Fatty Acid Methyl Esters

IWC = International whaling commission

MUFA = monounsaturated fatty acids

PCA = Principal Component Analysis

PC1 = first principal component

PC2 =second principal component

PL = phospholipids

PUFA = polyunsaturated fatty acids

QFASA = quantitatively fatty acid signature analysis

RSD = residual standard deviation

SFA = saturated fatty acids

SI = Stratification index

SIMCA = Soft Independent Modeling of Class Analogies

TAG = triacylglycerol's

WE = wax esters

1.0. Introduction

1.1. Minke whale

The minke whale (*Balaenoptera acutorostrata*) is the smallest and probably the most abundant baleen whale in the North Atlantic, and is one of the most conspicuous high-trophic-level predators in this area. The population is subdivided into four stocks: the East Canadian stock, the West Greenland stock, the Central Atlantic stock and the Northeast Atlantic stock, by the International Whaling Commission (IWC). The population has been commercially exploited in Norway from the 1930s to present, with a stop in the years 1987 – 1992.

All stocks undertake seasonal migration between low latitude breeding areas, and temperate and polar regions where they exploit the biological production (Jonsgård 1951). The whales are segregated by sex and size during the northward migration, with adult females and juveniles inhabiting more coastal areas, and adult males which tend to stay in more open waters (Jonsgård 1951).

Evidence from commercial and scientific catching operations (Folkow et al 2000; Haug et al 1995A; 1996; 1997; 2002; Jonsgård 1982; Lydersen et al 1991; Olsen & Holst 2001; Windsland et al 2007) has revealed that the minke whale in the northeastern Atlantic has a flexible feeding pattern and is feeding on a wide variety of prey species, consisting of several species of zooplankton and fish. They prefer feeding on energy rich fish species such as herring (*Clupea harengus*) and capelin (*Mallotus villosus),* but gadoid species (*Gadidae*), sandeels (*Ammodytes sp.)*, krill (*Thysanoessa sp* and *Meganyctiphanes sp.)* and copepods (*Calanus sp*.) are also part of their diet. The whale is able to adapt to local prey densities, because prey abundance varies with different areas and time of the season.

It is important to understand the interactions between the minke whale and its prey, for management of a sustainable minke whale stock and the economically important fish species, which the whale preys upon.

1.2. Minke whale blubber

The blubber of the minke whale serves several fundamental roles in the life of the aquatic animal, and is a subcutaneous lipid-rich layer, surrounding the entire body. Blubber is a highly modified form of adipose tissue, which consists of adipocytes contained in a supportive matrix of collagen and elastin fibers (see e.g. Koopman 2006; Pond 1998; Pabst et al 1999). It contributes to buoyancy and streamlining necessary for diving and swimming, and functions as an isolation layer, important for maintenance of thermal balance (see e.g. Strandberg et al 2008; Ryg et al 1988; Koopman et al 2002). The blubber is also the main storage site for lipids in the form of triacylglycerol (TAG), and serves as the main energy source of the animal in periods of fasting.

Several investigations of marine mammal blubber have documented that the FAs in the blubber is stratified through the blubber column. This was shown already in 1932 by Heyerdahl, who found

difference in iodine value through the blubber column, and that outer blubber had higher and less varying fat content than inner blubber (refereed in Olsen & Grahl-Nielsen 2003). Ackman et al (1965) showed that there is a difference in the FA composition of the inner and outer layers of fin whales (*Balaenoptera physalus*). The stratification of minke whale blubber is documented by Fehn (1996) and Grahl-Nielsen & Olsen (2003), and is also found in four species of seals (Fredheim et al 1995), ringed seal (*Phoca hispida;* Käkelä and Hyvärinen 1996), harbor porpoise (*Phocoena phocoena;* Koopman et al 1996*),* harp seals (*Phagophilus groenlandicus*; Grahl-Nielsen et al 2011) and in southern elephant seals (*Mirounga leonine*; Best et al 2003)

The outer layer is primarily structural and thermoregulatory, and consists of more short-chain MUFAs and less SFAs, than the inner layer (Strandberg et al 2008). The inner layer is metabolically active, and the fatty acid composition is strongly affected by recent or ongoing lipid mobilization or deposition. The middle layer is a storage site that contracts and expands with food availability and consumption. The reason for this stratification is likely differential metabolism through the blubber.

1.3. Blubber analysis

FAs have been widely used in ecology to reveal food web relationships, and have been suggested as a tool to reveal the diet of predators. The advantage of FAs is the potential to provide information on dietary intake and food constituents of the predator over a longer period of time, than more traditional gut content analysis, which only gives as snap-shoot of the last meal of the predator. Blubber samples have also the potential to be obtained from biopsy without killing the animal, and can be used both for DNA analysis and biochemical analysis.

Budge et al (2006) has reviewed the use of FA in trophic ecology, and has suggested that there is three ways in which FA can be used to study forage ecology and food webs. The first is a qualitative approach were changes in FA distribution of the predator alone is examined, in order to reveal spatial and temporal variations in diets, both among and within individuals or populations. The second use of FA is also qualitatively, but relies on the use of individual biomarkers, that can be, specific FAs which can be traced to a single origin or prey species, so called FATMs. It can also be ratios between specific FAs or unusual levels of certain FA in the prey, which tell about their dominance or unimportance in the diet. The third approach in which fatty acids are used, are Quantitative Fatty Acid Signature Analysis (QFASA), which quantitatively estimate the diet of predators from FA signatures of predator and prey (Iverson et al 2004). This method uses a statistical model to compute the most likely combination of prey FA signatures that comes closest to matching that observed in the predator. The metabolism of FA is accounted for by mathematically weighting individual FA, by use of estimated calibration coefficients.

Blubber analysis is widely used by researchers studying marine mammal ecology. Lund (1934) used the degree of unsaturation of whale oil, the iodine value, to distinguish between different stocks

of fin whales (*Balaenoptera physalus*), and Lund (1936) used the oil of humpback whales (*Megaptera novaeanglia*), to distinguish between a stock from the Indian Ocean and a stock from the South Atlantic Ocean (refereed in Olsen & Grahl-Nielsen 2003). Grahl-Nielsen et al (1993) did stock identification of individual harp seals, by use of FA composition in the jawbone of the seal, and Käkelä et al (1993) could distinguish between a fresh-water population of ringed seals (*Phoca hispida saimensis*) and a Baltic population (*Phoca hispida botnica*), by use of blubber FAs. Falk-Petersen et al (2009) used FATMs to trace harp seals to diatom based food chains and hooded seals to dinoflagellate and the prymnesiophyte *Phaeocystis phouchetii*-based food chains.

The degree to which FAs are able to tell about the diet of predators is highly disputed, and the biggest constrains with use of FAs are that no single FA can be assigned uniquely to any one species, and that FAs are not metabolically stable, depending on the condition and metabolic strategy of the consumer.

Crucial in understanding of how FAs can be used to inform about diet, is the way FAs that enters the body of a predator are metabolized (reviewed in Budge et al 2006). FAs enters the gut of a predator, such as a marine mammal or a fish, in the form of phospholipids (PL), triacylglycerol (TAG) and wax esters (WE), which are emulsified by bile salts and hydrolyzed by pancreatic lipases into free FAs. The free FAs are further absorbed into intestinal epithelial cells where they are re-esterified into PL or TAG, before they are transported in the blood to tissues. TAG that enters the tissue is again hydrolyzed to free FAs, which is either metabolized for energy, or re-esterified into TAG for storage in adipose tissue or into PL in the cell membranes. Some FAs are transported into storage in the body in a selective and direct way, while others are exposed for modification through *de novo* biosynthesis. This *de novo* biosynthesis is mainly limited to SFA and MUFA, in birds and mammals, and is inhibited during both fasting and consumption of high fat diets.

Another complicating factor for the use of FAs to inform about diet is the fact that blubber is controlled by the DNA of the organism. Genetic differences between individuals, populations and species could therefore be visible in the biochemical composition of the blubber, masking the effect by the diet. The lipid composition could also be affected by seasonal factors in the life history of the predator, and of biotic and abiotic factors such as food abundance, water temperature and salinity. The lipid composition of fish has shown to differ with season, for instance with sexual maturation and spawning in capelin (Henderson et al 1984).

1.4. The three different ecosystems

FA composition of minke whales has previously been shown to differ between the Norwegian Sea and the North Sea, in a study by Olsen & Grahl-Nielsen (2003). Møller et al (2003) have studied FA signatures of minke whales from the North Atlantic, and have suggested a “three-geographic region model” of minke whales, where the regions are: Greenland, the Northeast Atlantic and the North Sea.

In order to investigate the FA distribution of minke whales in different areas, and the potential influence by prey, blubber samples from three different areas were selected, together with prey samples from the same areas. The areas selected in this study are Spitsbergen/Bjørnøya, Vesterålen and the North Sea. A short description of these three areas and the main minke whale prey in these areas follows:

1.4.1 Spitsbergen/Bjørnøya

This ecosystem belongs to the east and northernmost parts of the Barents Sea ecosystem, which is a large and highly productive shelf sea. The west coast of Svalbard is dominated by the warm West Spitsbergen Current with Atlantic water, and the cold South Cape Current with arctic water (Ingvaldsen & Loeng 2009). The Bjørnøya area lays at the polar front and is influenced by both warm Atlantic water and cold arctic water (Ingvaldsen & Loeng 2009). The minke whale diet in this area is dominated by capelin and krill (Folkow et al 2000; Haug et al 1996; 2002). Other potential prey species in this area, found in stomach analysis, is cod (*Gadus morhua*), polar cod (*Boreogadus saida*) and haddock (*Melanogrammus aeglefinus*) (Windsland et al 2007).

1.4.2 Vesterålen

This ecosystem belongs to the Norwegian Sea, and is dominated by the Norwegian coastal current which carries relatively fresh water along the Norwegian coast, and the outer laying Norwegian Atlantic Current with saltier water masses (Ingvaldsen & Loeng 2009). This area is the main spawning site for the North Eastern Atlantic cod stock, and the southern bound for the spawning of capelin (Gjøsæter 2009). A stomach content analysis by Lydersen et al (1991) found herring to dominate as prey in the area, while prey species like cod, saithe (*Pollahcius virens*) and haddock, were found in lesser degree. Other prey species found to be available for minke whale in this area is blue whiting (*Micromesistius poutassou*) (Windsland 2007), sand eel and krill (Haug et al 1996).

1.4.3 The North Sea

The North Sea is very different from the Barents Sea and the Norwegian Sea, because it is a closed shallow ecosystem, heavily influenced by human activity. It opens to the Atlantic but receives low-salinity water from the Baltic trough the Kattegat and Skagerrak, as well as from rivers (Ducrotoy et al 2000). Sand ell is found to be the most important diet of the minke whale in this area, while mackerel (*Scomber scombrus),* herring and haddock were found in lesser degree in stomach samples (Olsen & Holst 2001). Saithe and blue whiting are also thought to be potential prey species here, since they are known as minke whale prey (Haug et al 1996; Windsland et al 2007) and are abundant in the area (Huse 2011; Høines 2011).

1.5. Objective of the investigation

The knowledge about diet of minke whales is based on stomach content analyses. We want to investigate if FA analyses can be used to monitor the diet of minke whales. FA analyses are not as comprehensive as stomach content analyses in regard to sampling, and they provide information about diet from a much longer time span, potentially from weeks to months. Stomach content does only tell about the last meal of the whale.

In order to investigate the potential of FA analyses as a tool to monitor minke whale diet, it was decided to address the four questions:

1. Is the blubber layer of the minke whale stratified, with respect to FA composition?
2. Is the minke whale blubber characterized by the ecosystem were the whale is caught?
3. Is the minke whale blubber characterized by the stomach content of the whale?
4. Is there a relationship between minke whale blubber and potential prey species?

2.0 Materials and methods

2.1 The procedure for minke whale sampling

The sampling of blubber profiles and inspection of stomach contents from minke whale was done during the Norwegian commercial minke whale catch, onboard three whaling vessels: m/s Kato, m/s Reinebuen and m/s Reinefangst. Three large scale areas were sampled: the North Sea 2009, Vesterålen June 2010 and Spitsbergen/Bjørnøya May 2010.

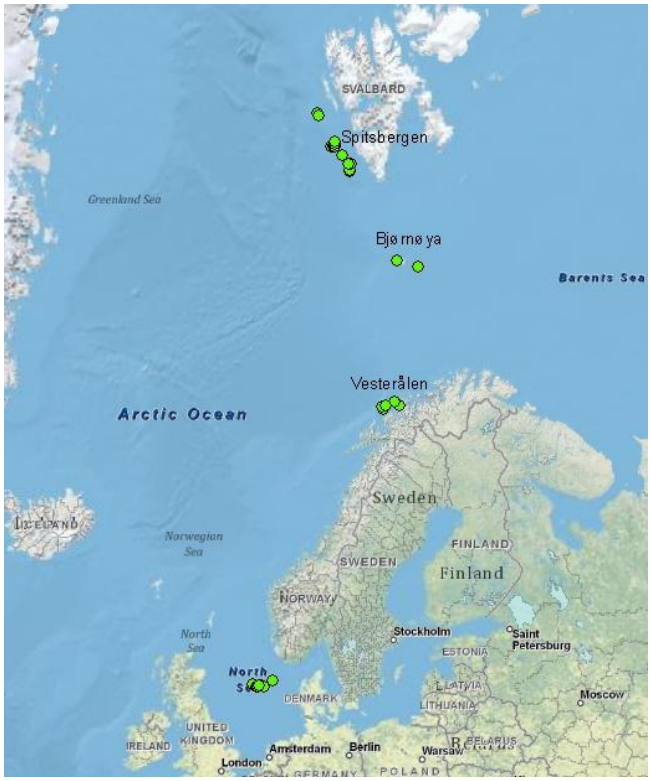


Figure 1: Map showing catch positions for the 37 sampled whales (green circles) caught during the whaling in the areas Spitsbergen, Bjørnøya, Vesterålen (2010) and the North Sea (2009).

An ordinary fishing vessel equipped with a harpoon, a crow`s nest and a "running grind" between the wheel house and the harpoon was used for the whaling operation. Lookout for whales was held in the crow`s nest, and when a whale was spotted the animal was approached. Some curious whales, called "seekers", were approaching the boat themselves. With the whale close to the ship, all men were at deck, either in the crow's nest or at the top of the wheel house, holding lookout in every direction to see where the whale was breaking the surface. The whale was chased until it finally was in front of the boat and the harpoon. A shoot with a spear armed with a high-explosive shell, and attached with a rope to the boat, was then fired against the animal. The rifleman was aiming at the white spot on the flipper of the whale, so that the abdominal cavity and the spinal cord were hit. The high-explosive shell would in 80 % of all shots kill the animal instantaneously (Knudsen & Øen 2003). If the whale was not killed instantaneously, the rifleman would immediately shoot it with a large caliber rifle. After the animal

was dead it was hauled aboard, and secured by a chain in the tail, before it was dragged up on the deck of the ship. During flensing the stomach and intestines were removed and a fast inspection into the fore-stomach was conducted, in order to assess the dominating prey species.

A blubber sample of 5 x 5 cm was taken from the back just behind the blowhole (Figure 1). The whole blubber column from skin to muscle was sampled, and packed in aluminum foil and plastic bags before it was frozen at – 20°C. Total length of the whale was measured in a straight line from the tip of the upper jaw to the apex of the tail fluke notch. The sex of the whale was also identified.

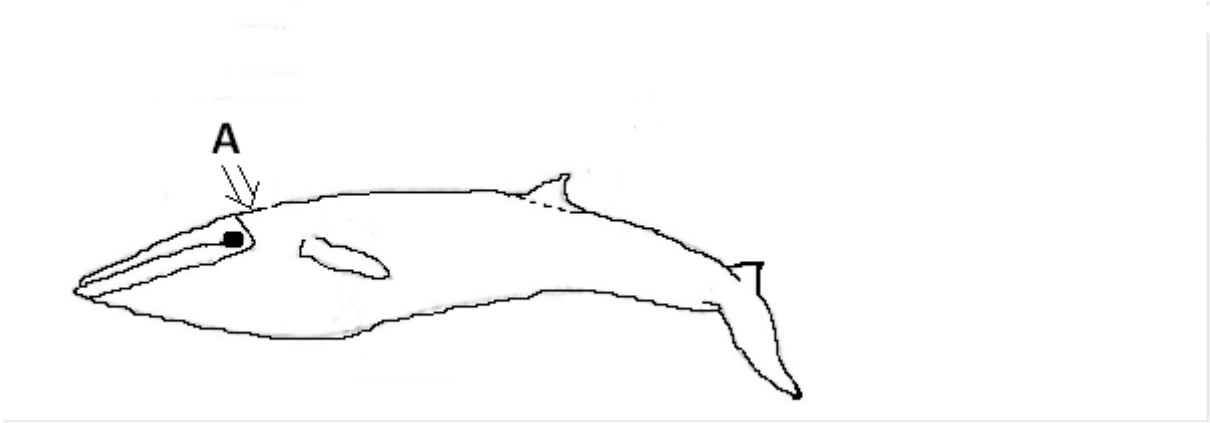


Figure 2: A blubber sample of 5x5 cm was taken from location A on the picture.

2.2 Fish samples

In the analyses of potential prey animals, a combination of historic data and recently sampled species were used. The prey species were selected from relevant literature on previous stomach content analyses in the relevant areas by Folkow (2000), Haug et al (1995a,b; 1996; 2002), Jonsgård (1982) Lydersen et al (1991), Olsen & Holst (2001) and Windsland (2007).

Most of the fish data were taken from a previous FA study of harp seal blubber and prey by Grahl-Nielsen et al (2011). These prey species, *Meganyctiphanes sp*., *Thysanoessa sp*., capelin (*Mallotus villosus*), cod (*Gadus morhua*), haddock (*Melanogrammus aeglefinus*), and polar cod (*Boreogadus saida*) were collected in the northwestern part of Barents Sea in May-June 2006, i.e., comparable in area and season with the northern samples of minke whale from 2010. FA data from herring (*Clupea harengus*) in the central Norwegian Sea were obtained from Kurita et al (2003).
Blue whiting (*Micromesistius poutassou*), mackerel (*Scomber scombrus*), saithe (*Pollachius virens*) and lesser sand eel (*Ammodytes tobianus*) were sampled in the North Sea summer 2010 and analyzed at the Chemistry lab at the Institute of Marine Research in Bergen, Norway, simultaneously with the whale blubber analyses.

All the data from Grahl-Nielsen et al (2011) comes from homogenates of whole fish, and the same is the case for sand eel. For mackerel and herring, the analyses were done on fillet, which is

expected to be a good estimate of the FA profile of the whole fish. For blue whiting and saithe, both liver and fillet samples were analyzed, and the FA profile of the "whole fish" were calculated from assumed tissue contribution to the total fatty acids profile using the formula (1):

(1). Fatty acids profile of FA = F-FA x F-C + L-FA x L-C.

F-FA: The fatty acids % of FA in fillet

L-FA: The fatty acids % of FA in liver

F-C: Contribution of FA in the fillet to the whole fish FA content.

L-C: Contribution of FA in the fillet to the whole fish FA content.

In both blue whiting and saithe, the fatty acids liver did dominate the contribution to the "whole fish FA content" and contributed with 92.6 ± 1.8 % and 90.4 ± 1.4 % of the total amount of FA, respectively.

2.3 Chemical analysis

The FA analyses of the minke whale blubber were conducted at the Chemistry lab at the Institute of Marine Research in Bergen, Norway. Analysis of blubber from whales, and fillet and liver from fish, followed a one-step extraction/methylation method for determination FA in marine tissue (Meier et al 2006).

Collection of subsamples was done while the blubber was still frozen: first, 1 cm of the outer part of the blubber from skin to muscle was removed; to get rid of oxidized blubber. Small subsamples weighing 20-50 mg were subsequently taken from the blubber core at depths described in section 2.5 and 2.6.

Similar small samples were taken of the prey; sand eel (homogenate of whole fish), mackerel and herring (fillet), blue whiting and saithe (fillet and liver).

All subsamples were transferred to thick-walled glass tubes with Teflon-lined screw caps. Beforehand 100µl of a chloroform solution with an accurately determined concentration of the 19:0 Fatty Acid Methyl Ester (FAME) (nonadecanoid acid), had been added to the tubes, and the chloroform was evaporated. This left an accurately determined concentration of around 1.000 mg 19:0 FAME as an internal standard in each tube. For blue whiting and saithe fillet and lesser sand eel homogenate an intern standard with less concentration, 0.101 mg was used.

Methanolysis-reagent, 1ml, anhydrous methanol with toluene, in the proportion methanol:toluene (4:1 v/v), containing hydrogen 2 M hydrogenchloride (HCl) was then added to the tubes. The tubes were left in an oven at 100°C for 2 hours, were the methanolysis occurred.

Half of the methanol was evaporated with nitrogen-gas. Then 1 ml distilled water (H_2O) was added, together with 2 ml hexane. The contents were mixed by a whirl-mixer, followed by

centrifugation at 2000 rpm for 3 min. The upper layer of hexane was transferred to thick-walled glass tubes by Pasteur pipette. The extraction procedure was repeated with 2 ml hexane.

The extracted hexane was diluted or concentrated to obtain a suitable chromatographic response. One microliter was injected split less into a Hewlet-Packard P5890 AM gas chromatograph, equipped with a flame ionization detector and an autosampler. The column was 25m x 0.25 mm fused silica carbowax coated with polyethylene-glycol (CP-WAX 52CB Chrompack) of 0.2 µm thickness, and helium was used as the mobile phase at 1 ml/min constant column flow (20 psi). The injection temperature was set to 270°C. The oven was programmed as follows: 90°C for 0 min, 30°C/min to 165°C, then 2.5°C/min to 225°C, where the temperature was held for 20 min. The temperature of the flame ionization detector was set at 300°C.

Blanks of hexane and blanks of the metanolysis-reagent containing the same amount of the intern standard 19:0 FAME as the samples were also run. Every tenth sample run was a FAME standard, containing 20 FAMEs (GLC-463 Nu-Chek-Prep, Elysian, minn, USA). The detector output was converted, recorded, stored and treated in the software EzChrom.

Forty two well-defined peaks in the chromatogram were selected, and identified by comparisons with a chromatogram of the standard mixture (Nuchek) and retention index maps and mass spectral libraries (http://www.chrombox.org/index.html) from previous analysis of marine FAMEs. (Forty one fatty acids were selected in the validation, section 2.5)

2.4 Statistics

Every FA was corrected by empirical response factors relative to the FA 18:0 in 20 FAMEs present in known proportions in the standard Nuchek mixture. Response factors for those FA not present among the 20 FAMEs in the Nuchek standard, were estimated by comparisons with the FA that resembled them most in terms of chain length and number of double bonds. The data were normalized by expressing each FA as percentage of the sum of all 42 FAs.

To obtain the combined information from all FAs simultaneously, FAs were treated with multivariate statistics based on principal component analysis (PCA), by the software SIRIUS version 8.0. (Kvalheim & Karstang 1987). The relative values (i.e. percent of the sum) were scaled by dividing each value by the mean of the values of all samples for that particular FA, with the intention to level out the quantitative difference among the FAs, leaving them all to vary around one. The samples were then positioned in a 41-dimensional space, were new coordinates (principal components), through the centroid of the samples, in the directions of the largest and second largest variance among the samples were computed (40-dimensional space in the validation, section 2.5. The FA 18:1(n-11) was not included in PCA, se discussion.) The systematic relationship among the samples could then be described in two dimensions, PC1 and PC2, instead of the original 41 dimensions, with negligible loss

of information. The samples (objects) were projected onto the PC1 – PC2 plane to give scoreplots, and the FAs (variables) were projected onto the PC1 – PC2 plane to give loading plots.

A stratification index was calculated to illustrate the degree of difference between the inner and outer blubber layer for all the 42 fatty acids. This index (SI) was calculated by subtracting the percentage in inner layer (F_i) from the percentage in outer layer (F_0), and dividing the difference by the mean of the percentages in the inner and outer layer.

$$SI = \frac{(F_0 - F_i)}{(F_0 + F_i)/2}$$

SIMCA (Soft Independent Modeling of Class Analogies; Wold 1976, 1978; Ugland and Massart 1996), available in the SIRIUS software package, were used to determine the distance of prey samples to the inner blubber. A PC-model of the inner blubber was first generated. Cross validation were used to check if the different components were significant, and the distances (RSD-values) to the different prey samples were calculated.

2.5 Method Validation

A validation of the entire lab procedure was first conducted, and the results from this exercise were used to select subsample spots in the blubber column. One blubber column (Sample RB1) was used to take subsamples which included 7 different depths of the blubber. Five replicates were taken 2mm from the skin (O), then 3 replicates from each of 5 different depths of the middle layer (MA – ME), before 5 replicates were taken 5 mm from the muscle (I) (Figure 3). All subsample replicates were analyzed according to the chemical methods described in section 2.3, and data were corrected by empirical response factors, normalized and treated with PCA as described in section 2.4.

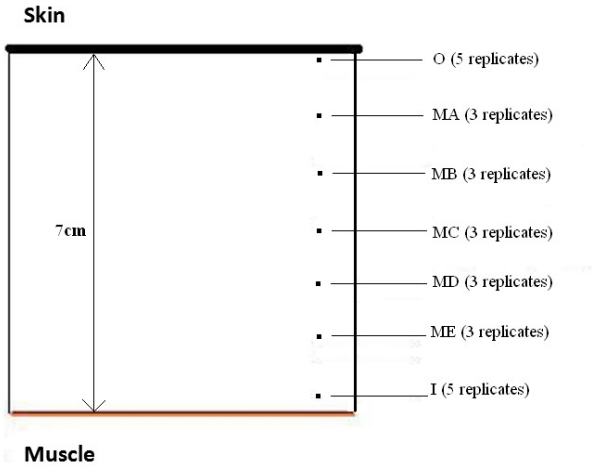
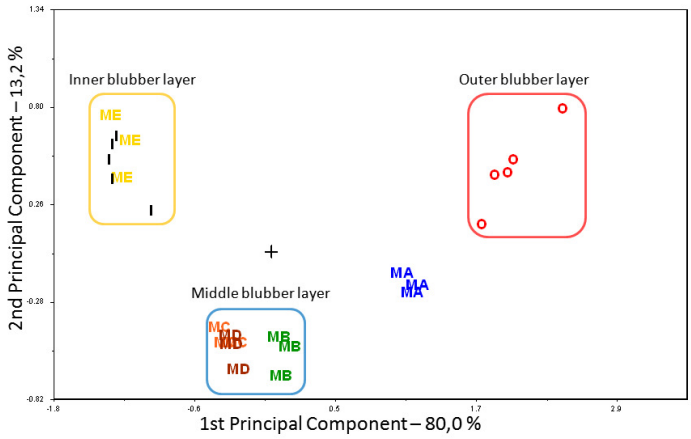


Figure 3. Location in the blubber of subsamples used in the validation analyses. Five replicates were taken 2mm from the skin (O), then 3 replicates from each of 5 different depths (MA – ME), before 5 replicates were taken 5 mm from the muscle (I).

Forty one FAs were identified in the validation (Table 1). The average percentage sum of SFAs varied from 17.9 % to 22.7 % for the subsample locations. The average percentage sum of MUFAs varied between 58.4 % and 64.7 %. The amounts of polyunsaturated fatty acids (PUFAs) were close to 20 %, at all depths, except in the inner layers, ME and I, which had around 15 %.

A PCA was run with the normalized data from the validation of the lab procedure, as given in Table 1. The two principal components PC1 and PC2 were together able to explain 93.2 % of the variance (Figure 4A). In this analysis the five samples from the outer blubber layer grouped together to the right in the plot. Samples from the depth MB, MC and MD grouped in the middle, while samples from depth MA grouped between the middle group and the outer blubber layer group. Samples from depth ME grouped together with the samples from the inner blubber layer.

A



B

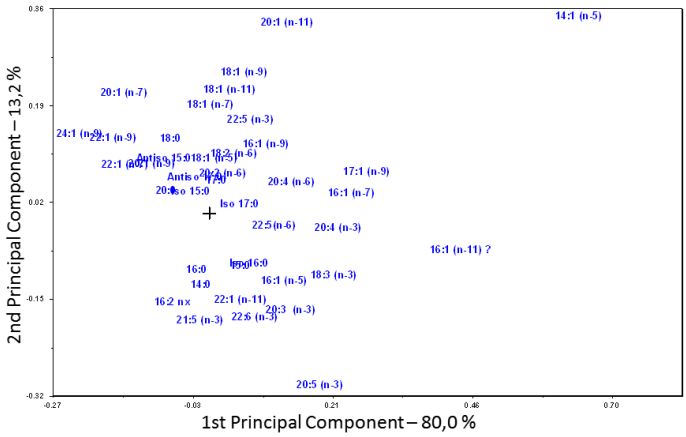


Figure 4A: Scoreplott of a PCA run on the analyzed samples of the validation. O = outer blubber layer, MA = middle layer location A, MB = middle layer location B, MC = middle layer location C, MD = middle layer location D, ME= middle layer location E and I = inner blubber layer.

Figure 5B: Loadingplott from the PCA run on the analyzed samples of the validation.

The results from the validation (Figure 4A) show a clear difference between the samples from the outer, middle and inner blubber layer in fatty acid composition. It was therefore decided to use three subsamples (outer, middle and inner blubber layer) in the analysis of the whale samples from 2010 (Figure 5). In the 2009 samples, outer blubber layer was not analyzed.

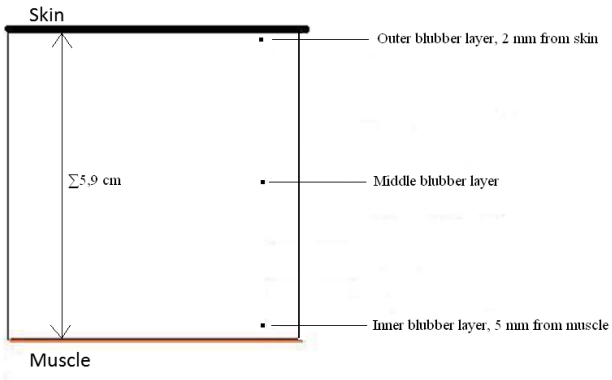


Figure 5: Subsamples of blubber used in the analysis: one sample 2mm from the skin (O = outer), one approximately in the middle (M= middle) and one 5mm from the muscle (I= inner). Average blubber thickness for all blubber samples is also shown (5.9 ± 1.7 cm).

3.0. Results

3.1. Whale samples and stomach contents

In total 37 minke whales were caught during the sampling in 2009 and 2010 (Table 2). Nine whales were cought in the North Sea in 2009 and all had sand eel in their stomacks. Twenty three minke whales were caugth in the area Spitsbergen/Bjørnøya in 2010. Three of the whales had stomach content dominated by codfish, mainly of the species haddock, while 20 had stomach content dominated by krill and capelin. The stomach content of one whale were unidentified. Five whales were caught at the coast of Vesterålen in 2010. Two had sand eel in their stomachs, while 3 had herring.

3.2. Stratification of the blubber layer – difference between inner, middle and outer blubber

The samples from the North Sea 2009 is not included in the analysis of blubber stratification, because samples from outer blubber layer lacks from this area. The FA analysis of the samples from Spitsbergen/Bjørnøya and Vesterålen 2010 displayed several differences in FA compostion between inner, middle and outer blubber layer.

The individual whale samples were varying a lot with regard to average quantified amount of FA among the three different blubber layers (Figur 6). No consistent pattern in fat content among the three different layers were found, but 22 of 37 individuals had most fat in the middle layer. Note that outer blubber samples is missing for the 2009 samples, together with to inner blubber samples from 2010, which were found to be outliers.

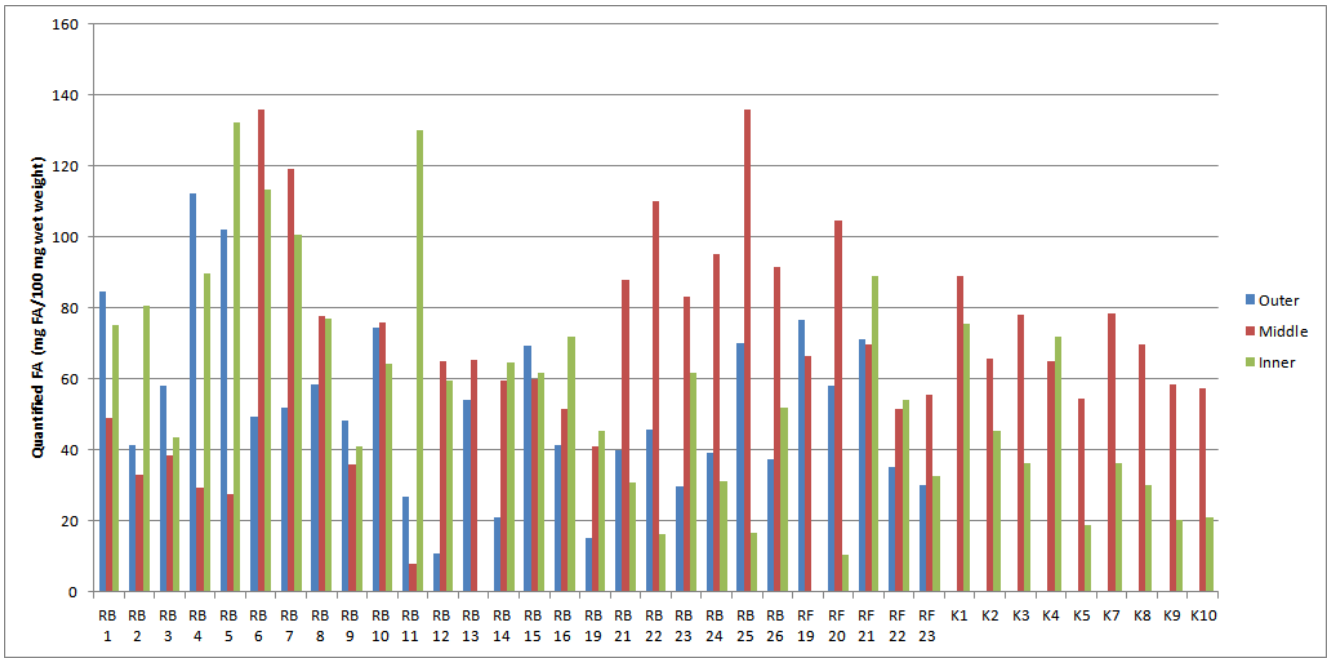


Figure 6: Average quantified FA (mg FA/ 100 mg wet weight) for the three different blubber layers from the individual whale samples from 2010 and 2009 (Table 2). Note that outer blubber samples for the 2009 samples are missing, together with inner blubber samples for the individuals RB 13 and RF 19.

The MUFAs was the group of FA with the highest average percent in all three layers, with a sum varying from 60 to 65 % (Table 3). A sum of 18 to 22 % SFAs and 18 to 19 % PUFAs were found in the three layers.

The relatative abundance of the individual FA varies a lot (Figure 7 and Table 3). The SFAs with the highest abundances are 14:0 and 16:0. The MUFAs with highest abundance are 16:1(n-7), 18:1(n-9), 18:1(n-7) 20:1(n-9) and 22:1(n-11). 18:1(n-9) has the highest abundance of all the 42 FAs. The PUFAs with highest abundance are 18:2(n-6), 18:4(n-3), 20:5(n-3) and 22:6(n-3). The MUFAs with 14, 16 and 18 carbon atoms had a higher percentage in the outer layer, except 18:1(n-11) which had the highest percentage share in the inner layer. The inner blubber had the highest share of MUFA with 20 carbon atoms, except 20:1(n-11) which has highest share in the outer blubber, and 22:1(n-11) which has it highest share in the middle layer. The FA 18:1(n-11) was found in all blubber layers (Table 3), but was almost not present in the FA composition of the prey (Table 4).

Outer blubber layer
Middle blubber layer
Inner blubber layer
Relative fatty acid composition %
0
5
10
15
20
25
14:OO
i-15:O
ai-15:O
15:O
i-16:O
16:O
i-17:O
ai-17:O
17:O
18:O
20:O
14:1n5
16:1n11
16:1n9
16:1n7
16:1n5
17:1n9
18:1n11
18:1n9
18:1n7
18:1n5
20:1n11
20:1n9
20:1n7
22:1n11
22:1n9
22:1n7
24:1n9
16:2nx
16:4n1
18:2n6
20:2n6
20:4n6
22:5n6
18:3n3
18:4n3
20:3n3
20:4n3
20:5n3
21:5n3
22:5n3
22:6n3

Figure 7: Relative fatty-acid compostitions of outer blubber layer (blue), middle blubber layer (red) and inner blubber layer (green) shown for all 42 FAs. Error bars are the standard deviations of the means of the relative percentage values. All whales from 2010 are included in the analyses.

The relative amount for almost all FAs, 28 of the 42 total, were either increasing or decreasing towards the skin or towards the muscle (Table 3). Some FAs were found with highest or lowest relative amount in the middle layer. The FAs: 18:1(n-11), 22:1(n-11), 16:4(n-1), 20:2(n-6), 18:4(n-3), 20:5(n-3) and 22:6(n-3) was found with highest relative amount in the middle layer, while the FAs: 17:0, 16:1(n-9), 18:1(n-9), 18:1(n-7), 18:1(n-5), 20:1(n-11) and 22:5(n-3) was found with lowest relative amount in the middle layer.

The relative stratification index for all FAs (Figure 8) shows a clear pattern in FA distribution between the inner and outer blubber layers. The inner layer has higher levels of SFAs, and MUFAs with 20 or more carbon atoms. The outer layer has high levels of MUFAs with 14, 16 and 18 carbon atoms. For the PUFAs it varies which fatty acid which got the highest level in the inner and outer layers.

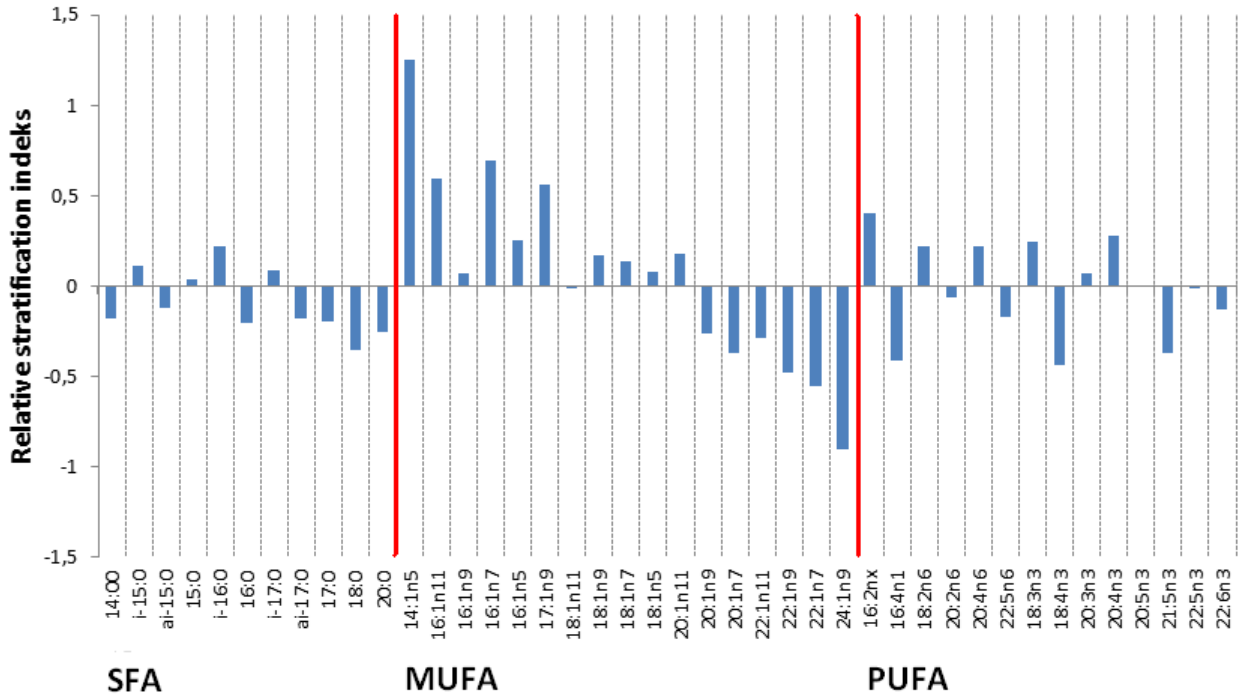


Figure 8: Stratification index for all 42 blubber fatty acids obtained by subtracting the percentage in inner layer from the percentage in outer layer, as given in Table 3, and dividing the difference by the mean of the percentages in the inner and outer layers. Values higher than zero indicate higher level of the acid in the outer than in the inner layer, and values below zero indicate higher level of the acid in the inner layer than in the outer.

A PCA with all blubber samples from outer, middle and inner layers for the 2010 samples, was conducted to examine the stratification of the blubber (Figure 9A, with corresponding loading plot;

Figure 9B). The two components PC1 and PC2 explained 52.7 % of the variance. Samples from the outer blubber layer gathered in the right side of the plot, while samples from the inner and middle layer gathered spread to the left in the plot (Figure 9A). A small overlap between the samples is found in the middle of the plot. Figure 9B, the loading plot of Figure 9A, shows that the FAs 14:1(n-5), 16:1(n-7), 16:1(n-11) and 17:1(n-9) explains the position of the outer blubber layer samples to the right in the plot, while 16:4(n-1), 18:4(n-3), 22:1(n-7), 22:1(n-9), 22:1(n-11) and 24:1(n-9) explains the position of the middle and inner blubber layer to the left in the plot.

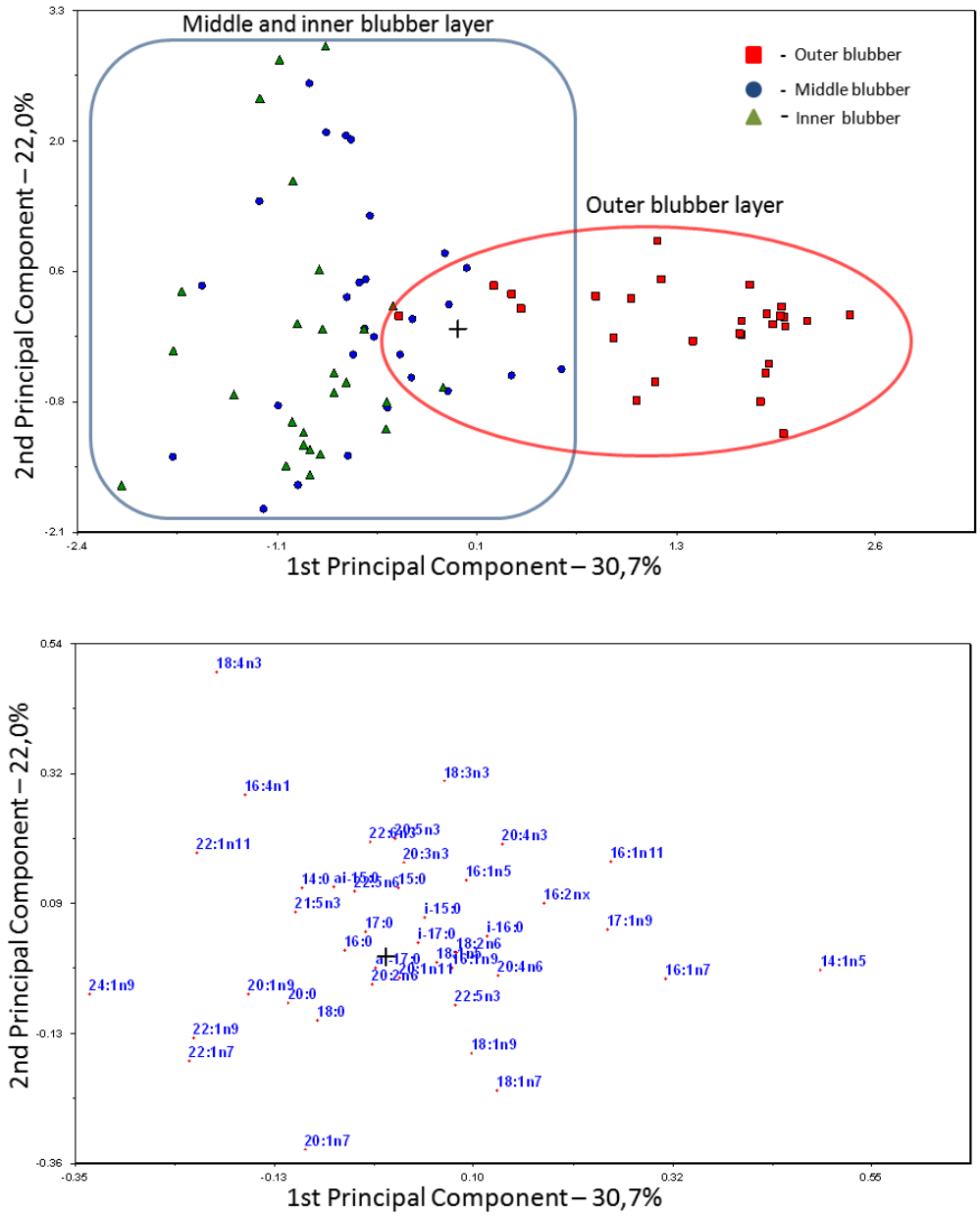


Figure 9

A: Score plot of samples from inner, middle and outer blubber layers from the 2010 samples.

B: Loading plot of samples from inner, middle and outer blubber layers from the 2010 samples.

3.3. Variation in inner blubber composition among different ecosystems

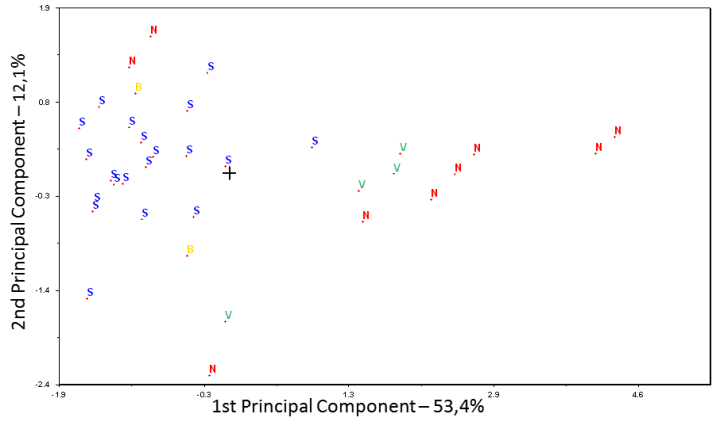
3.3.1. Inner blubber

Inner blubber samples from 2010 (areas Spitsbergen/Bjørnøya and Vesterålen), and 2009 (area the North Sea), where run in a PCA (Figure 10ABC). The two components PC1 and PC2 explained 65,5 % of the variance. The samples were first marked by their catch locality (Table 2 and Figure 10A). Samples from the North Sea are situated to the right in the plot, Vesterålen a bit to the left and Spitsbergen/Bjørnøya farthest to the left. Two samples from the North Sea were also situated high up to the left in the plot together with the Spitsbergen/Bjørnøya samples.

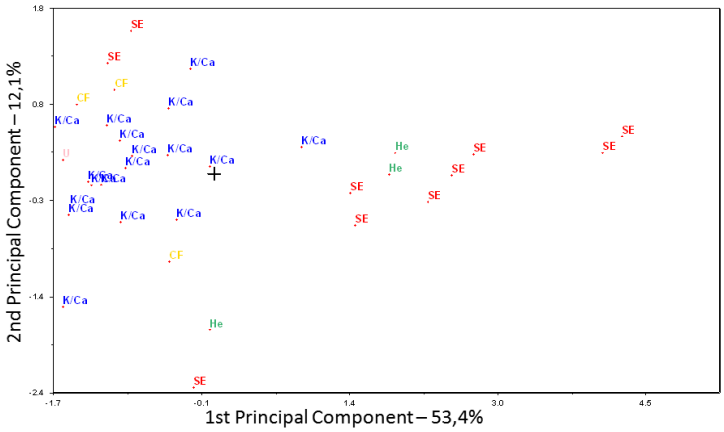
Second, all whale samples were marked by their main stomach content (instead of catch locality), based on observation done during whaling (Table 2 and Figure 10B). The North Sea whales had sand eel stomachs; the Spitsbergen samples had krill capelin stomachs except for one cod fish stomach and one unknown stomach, the Bjørnøya samples had cod fish stomachs and the Vesterålen had three herring stomachs and one sand eel stomach.

The loading plot (Figure 10C) shows that the FAs responsible for the position of the North Sea samples to the right in the plot are 16:4(n-1), 18:4(n-3) and 18:3(n-3). The position of four North Sea samples and three Vesterålen samples in the middle of the plot is due to the FAs 16:1(n-5), 16:1(n-11), 16:2nx, 20:4(n-3), 20:5(n-3), 21:5(n-3), 22:5(n-6) and 22:6(n-3). The position of the Spitsbergen samples farthest to the left on the plot is mainly due to the fatty acids 18:1(n-7), 18:1(n-9) and 20:1(n-7).

A



B



C

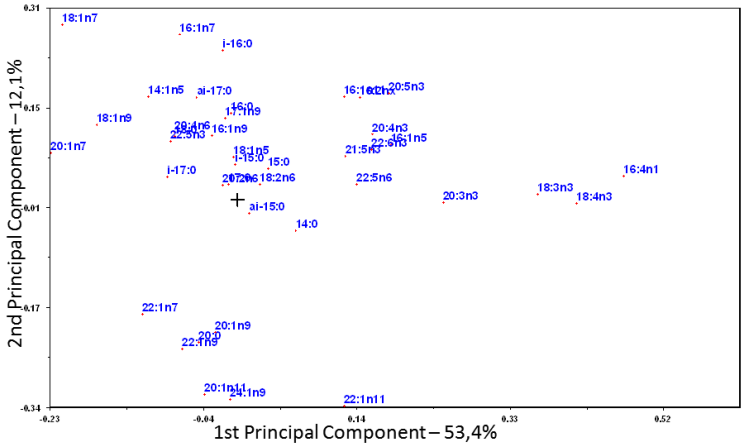


Figure 10

A: Score plot of a PCA with all inner blubber whale samples from the three different areas, marked by their catch locality; S = Spitsbergen (blue), B = Bjørnøya (yellow), V = Vesterålen (green) and N = the North Sea (red).

B: Score plot of a PCA with all inner blubber whale samples from the three different areas, marked by their main stomach content; K/Ca = krill-capelin stomach (blue), CF = cod fish stomach (yellow), He = herring stomach (green), SE = sand eel stomach =(red) and U = unknown stomach (violet).

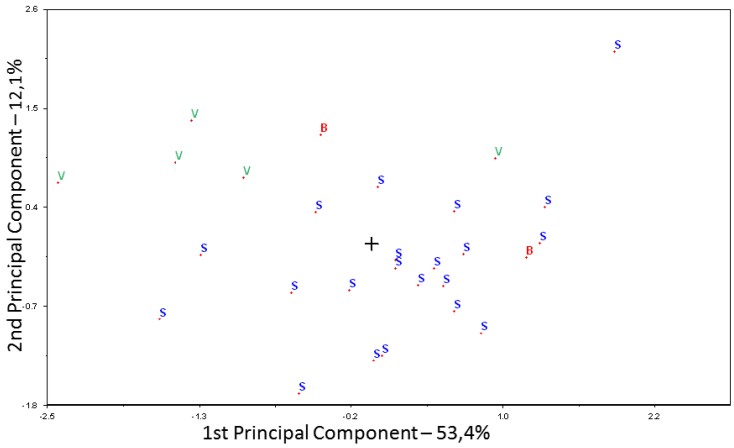
C: Loading plot of the PCA with inner blubber samples from the three different areas Spitsbergen/Bjørnøya, Vesterålen and the North Sea.

3.3.2 Outer blubber

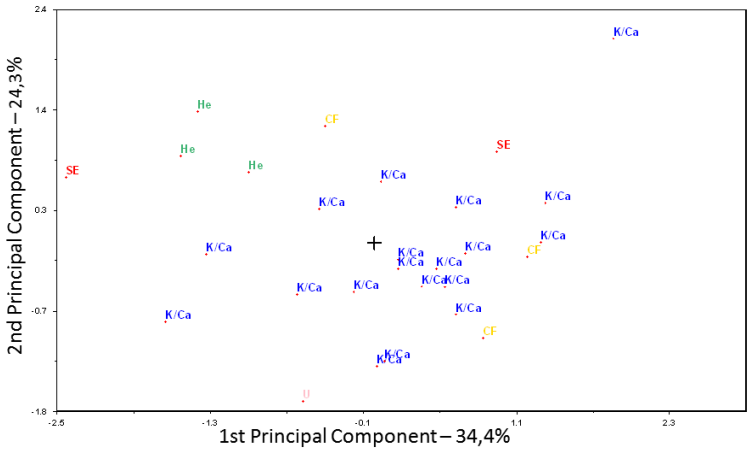
Outer blubber samples from 2010 (areas Spitsbergen/Bjørnøya and Vesterålen), were run in a PCA (Figure 11ABC). The two components PC1 and PC2 explained 58.7 % of the variance. The samples were first marked by their catch locality (Table 2 and Figure 11A). No clear distribution pattern was found, but samples from Spitsbergen/Bjørnøya grouped together in the lower middle part of the plot, while four of the five Vesterålen samples gathered together at he left side of the plot.

Second, all whale samples from the outer blubber layer were also marked by their main stomach content (instead of catch locality) as observed during whaling (Table 2 and Figure 11B). No clear distribution pattern were found here either, but most of the krill-capelin whales gathered together in the middle of the plot and the three herring whales gathered together at the left side of the plot. The sand eel whales and the codfish whales were placed spread in the plot.

A



B



C

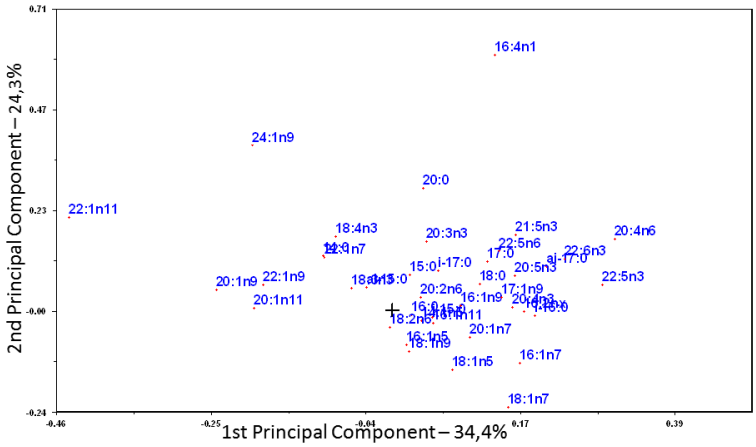


Figure 11

A: Score plot of a PCA with all outer blubber whale samples from the 2010 samples, marked by their catch locality; S = Spitsbergen (blue), B = Bjørnøya (red) and V = Vesterålen (green).

B : Score plot of a PCA with all outer blubber whale samples from the 2010 samples, marked by their stomach content K/Ca = krill-capelin stomach (blue), CF = cod fish stomach (yellow), He = herring stomach (green), SE = sand eel stomach =(red) and U = unknown stomach (violet).

C: Loading plot of the PCA run with outer blubber samples from the two areas Spitsbergen/Bjørnøya and Vesterålen.

3.4. Inner blubber layer in relation to possible prey species

In order to investigate the relationship between the FA compositions of the inner blubber layer with the FA composition of various potential prey species several PCA were conducted. First, all whales and prey from all areas were analyzed jointly. Subsequently, in order to investigate the different catch locations, the whale samples were divided into three ecosystems; Spitsbergen/Bjørnøya, Vesterålen and the North Sea. PCA analyses with whale samples from the three areas together with the potential prey species representative for that ecosystem were run. Fewer prey species in each PCA analysis would reduce the amount of noise in the analyses.

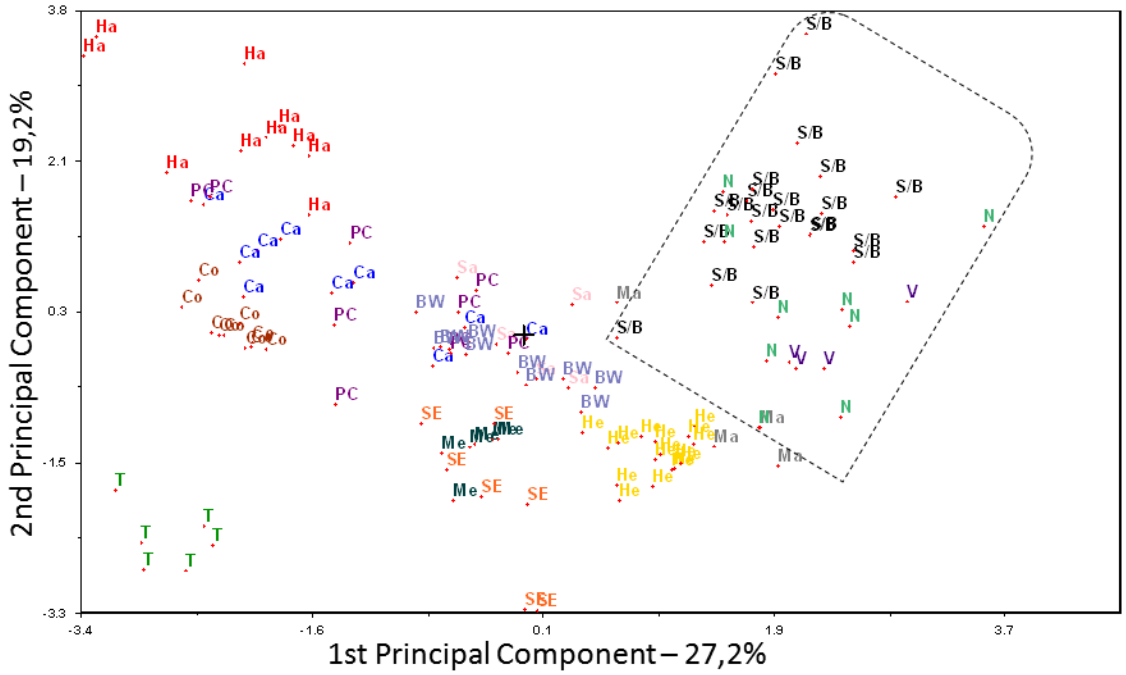
3.4.1 All areas pooled

One PCA with all inner blubber layer whale samples and all potential prey species included, were run (Figure 12A). The two components PC1 and PC2 explained 46,4 % of the variance. All whale samples were positioned to the right in the plot. Mackerel, herring, blue whiting, sand eel, saithe, and *Meganyctephanes* sp. were positioned in the middle, and haddock, polar cod, cod, capelin and *Thysanoessa* sp. were positioned to the left in the plot. The prey species lying closest to the whale samples were mackerel, herring, blue whiting and saith.

The loading plot (Figure 12B) shows that the fatty acids responsible for the position of the whale samples to the right in the plot are; 14:0, ai-15:0, 20:0, 14:1(n-5), 20:1(n-9), 20:1(n-11), 22:1(n-9), 22:1(n-11) 18:3(n-3), 20:4(n-3) and 22:5(n-3). The level of these fatty acids in the whale blubber is therefore high compared with the prey species.

The results of the SIMCA analysis (Figure 13) based on a model of all inner blubber whale samples and compared with all prey samples, shows that the prey samples with average smallest relative standard deviation (RSD) to the inner blubber model, are those laying closest at the scoreplot (Figure 12A). In one end, mackerel and herring got the smallest average RSD, while Thysanoessa sp. and haddock at the other end, got the highest RSD values (Figure 13).

A



B

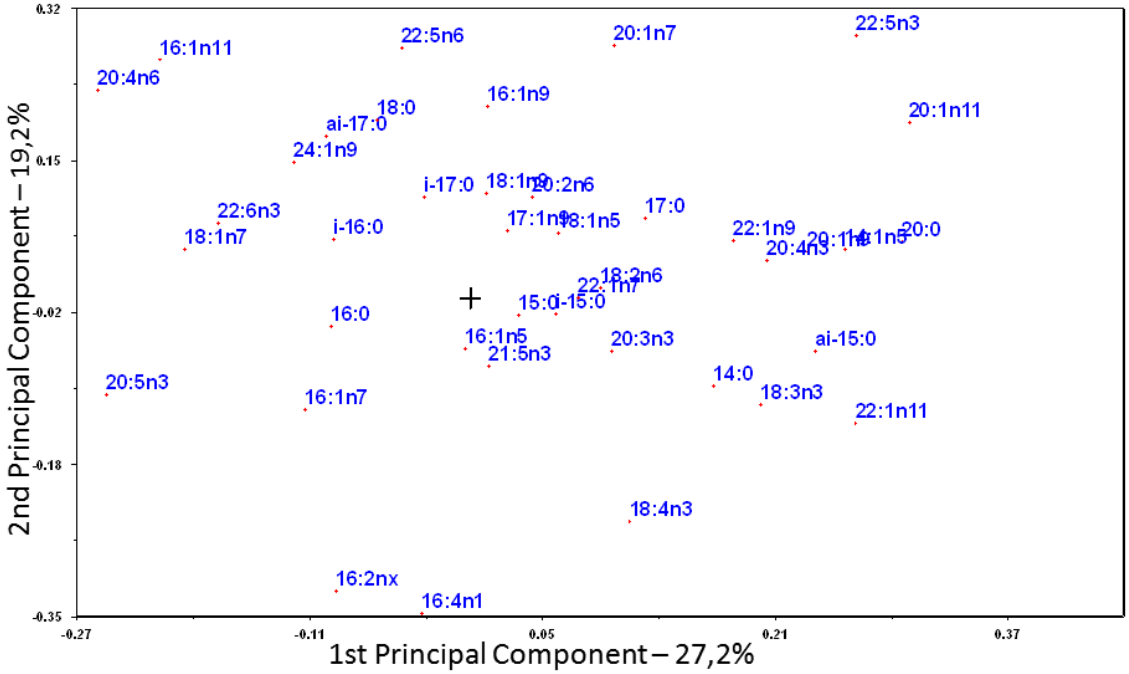


Figure 12

A: Score plot from a PCA with all inner blubber layer whale samples and all samples from potential minke whale prey species included. The whales are marked: S/B = Spitsbergen/Bjørnøya (black), V = Vesterålen (indigo), N = the North Sea (Sea-green). The prey species are marked: Ha = haddock (red), Ca = capelin (blue), Sa= saithe (pink), Ma= mackerel (gray), Me = krill of the species *Meganyctephanes* sp (dark olive green), Co = cod (brown), PC = polar cod (violet), T = krill of the species *Thysanoessa* sp (green), SE = sand eel (orange), BW = blue whiting (blue-gray) and He = herring (gold).

B: Loading plot of the PCA on all inner whale samples and all prey samples.

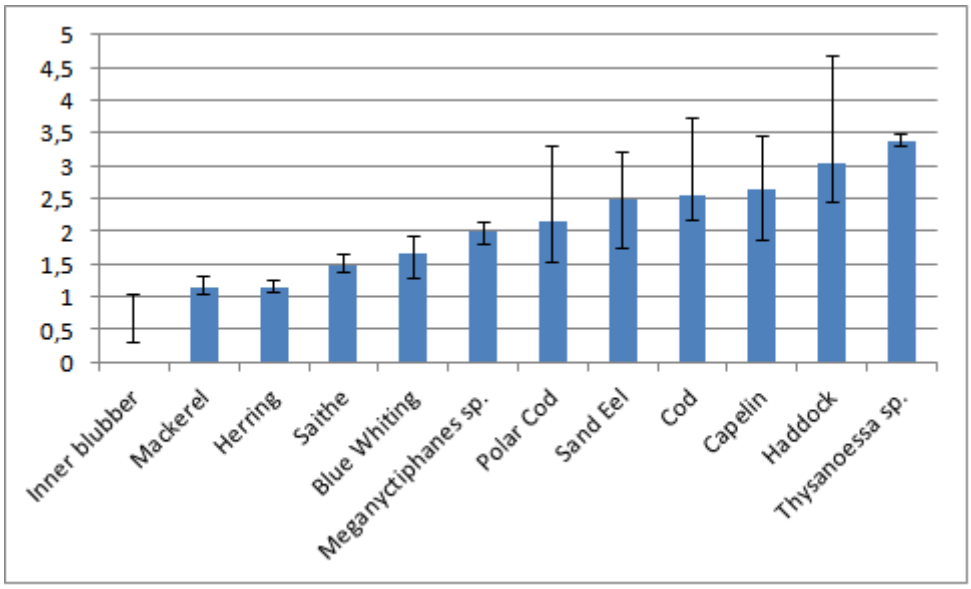


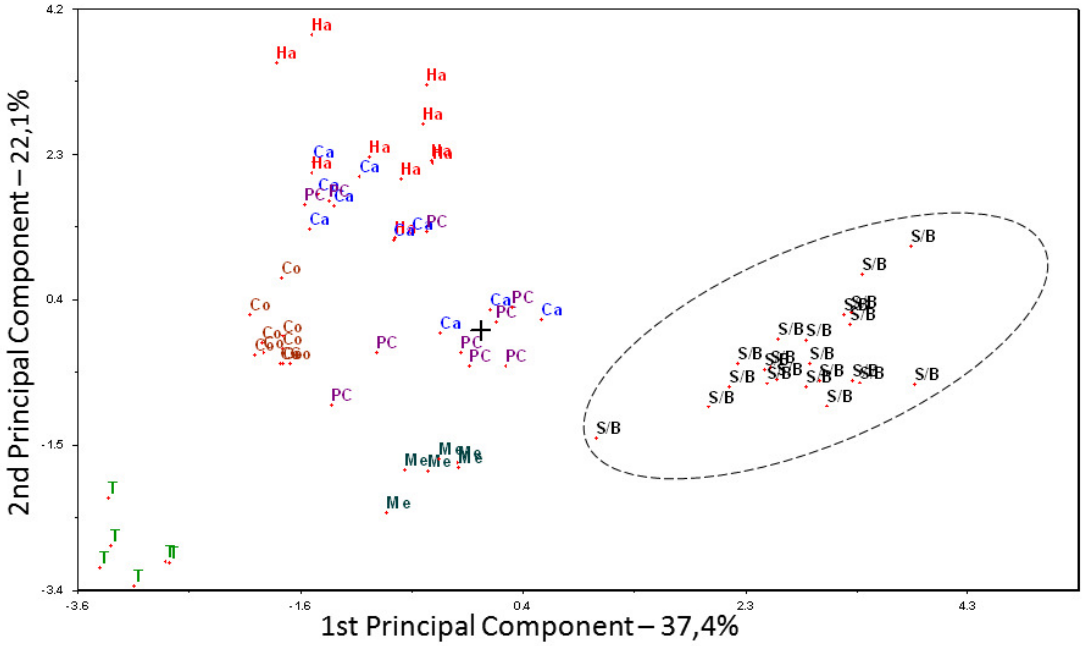
Figure 13: Relative standard deviation of the average values of all blubber and all the prey samples, as given in Table 4, from a model of the inner blubber samples based on 41 FAs. The model has 4 significant components (as tested by cross validation), covering 75.8 % of the total variance among the samples. The lowest and highest RSD of individual samples for each blubber/prey are indicated as error bars.

3.3.2. Spitsbergen/Bjørnøya

A PCA with all inner blubber samples from the 2010 Spitsbergen-Bjørnøya whales, and the potential prey species haddock, capelin, cod, polar cod, *Meganyctiphanes* spp. and *Thysanoessa* spp. was run (Figure 14A). The two components PC1 and PC2 explained 60.6 % of the variance. The whale samples were positioned at the right side of the plot. Haddock, capelin, cod polar cod and *Meganyctephanes* sp. were positioned in the middle a bit to the left, while *Thysanoessa* sp. were placed in the lower left corner. The prey species laying closest to the whale samples were capelin, polar cod and *Meganyctephanes* sp, which indicate that these species has the most similar fatty acid composition as the inner blubber. The variables responsible for the positioned of the whale samples to the right in the plot were the fatty acids: ai-15:0, 20:0, 14:1(n-5), 20:1(n-7), 20:1(n-9), 20:1(n-11), 22:1(n-11), 22:4(n-3) and 22:5(n-3) (Figure 14B).

The results of the SIMCA analysis (Figure 15) based on a model of the inner blubber from the Spitsbergen/Bjørnøya whale samples, shows that the prey samples with average smallest RSD to the inner blubber model, are those laying closest at the scoreplot (Figure 14A). Polar cod got the lowest average RSD while Thysanoessa sp. got the highest average RSD (Figure 15). Combined information from (Figure 14A and Figure 15) shows that the prey species polar cod, cod, *Meganyctiphanes* sp. and capelin got a FA distribution closest to that of the inner blubber.

A



B

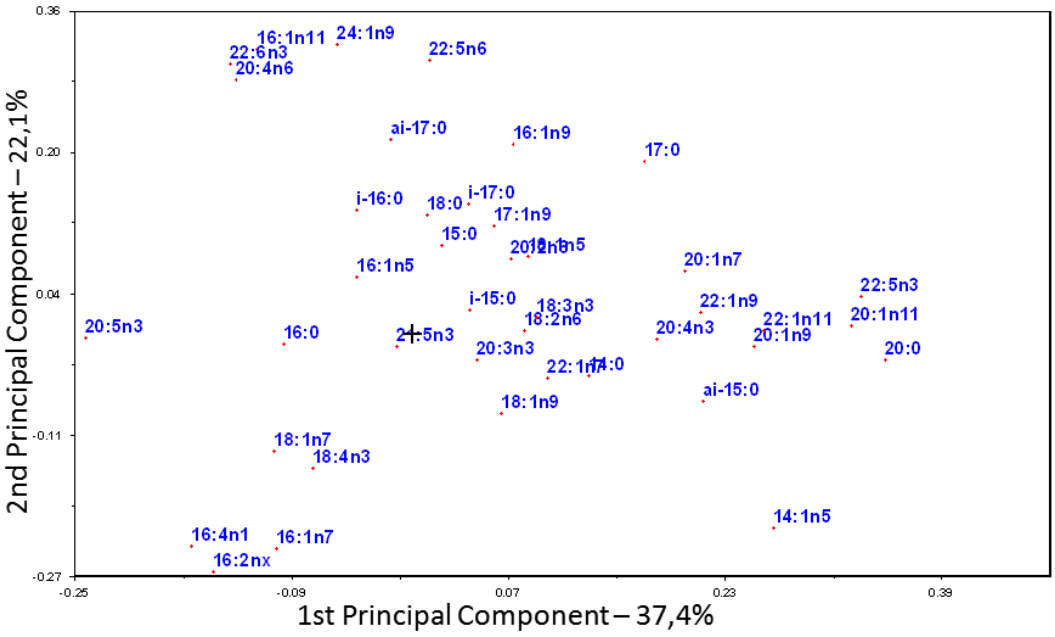


Figure 14

A: Score plot of a PCA on the inner blubber samples of whales from the Spitsbergen/Bjørnøya area. S/B= Spitsbergen/Bjørnøya, Ha = haddock (red), Ca = capelin (blue), Me = krill of the species *Meganyctephanes* sp. (dark olive green), Co = cod (brown), PC = polar cod (violet) and T = krill of the species *Thysanoessa sp*. (green).

B: Loading plot of a PCA on all inner whale samples from the area Spitsbergen/Bjørnøya together with the prey species haddock, capelin, *Meganyctephanes* sp, cod, polar cod and *Thysanoessa* spp.

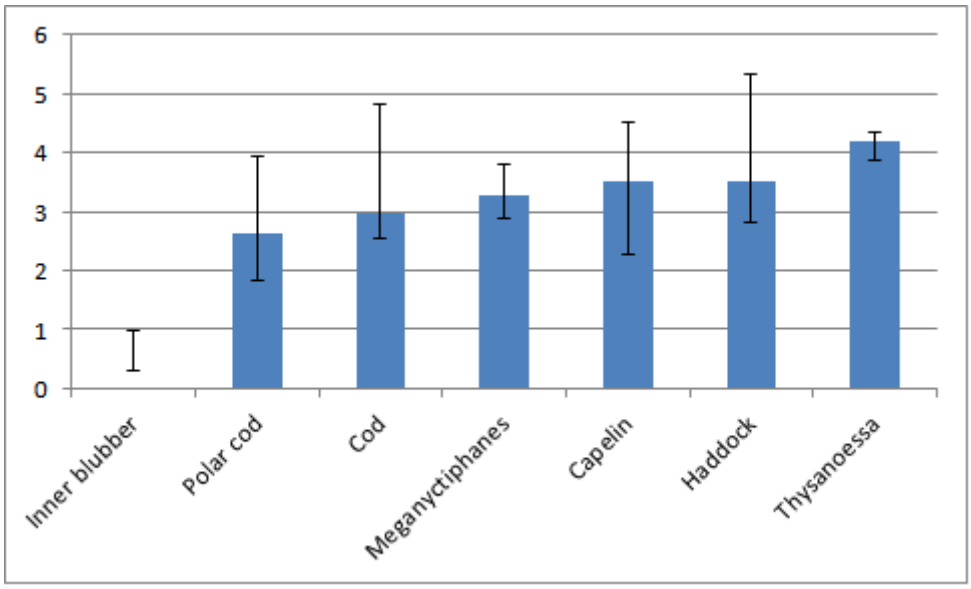


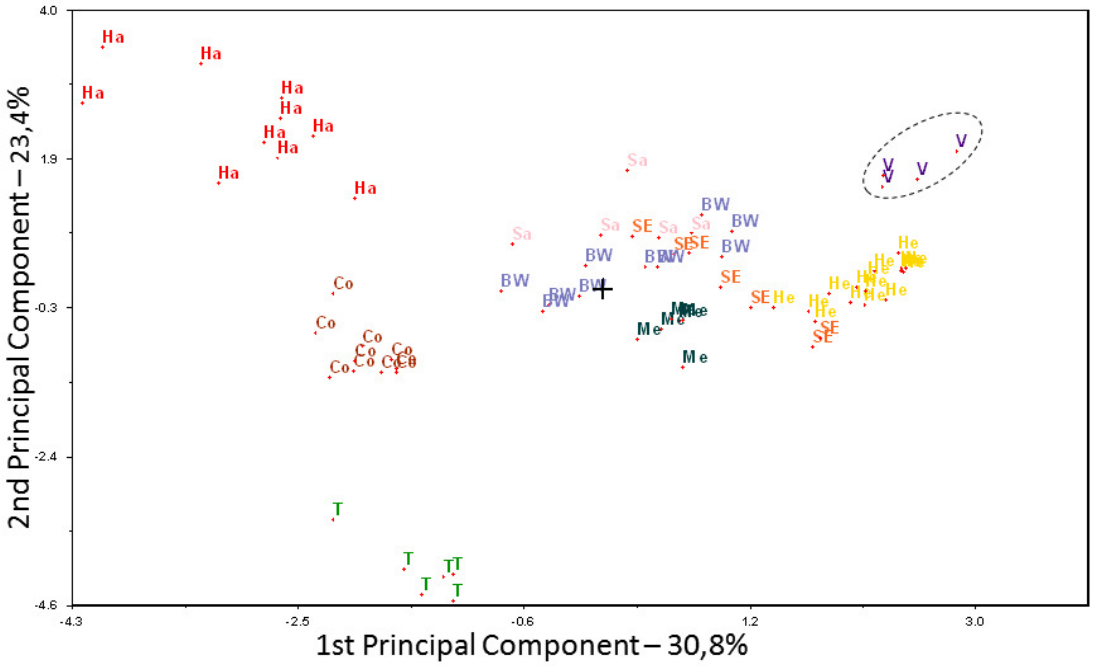
Figure15: Relative standard deviation of the average values of the Spitsbergen/Bjørnøya blubber and potential prey samples, from Table 4, from a model of the inner blubber samples based on 41 FAs. The model has 4 significant components (as tested by cross validation), covering 76.8 % of the total variance among the samples. The lowest and highest RSD of individual samples for each blubber/prey are indicated as error bars.

3.4.2. Vesterålen

A PCA with all inner blubber samples from the 2010 Vesterålen whales and the prey species; haddock, cod, *Thysanoessa* sp., saithe, blue-whiting, sand eel, *Meganyctephanes* sp. and herring was run (Figure 16A). The two components PC1 and PC2 were together able to explain 54.2 % of the variance. The whale samples grouped together in the upper right corner with the prey species herring lying right under. Saithe, sand eel, blue whiting and *Meganyctephanes* sp. were poisoned in the middle of the plot, while haddock, cod and Thysanoessa spp. were placed to the left. This plot indicates that herring, sand eel, blue whiting, saithe and *Meganyctephanes* spp. are the most likely prey of whales from this area. The variables responsible for the position of the whale samples were ai-15:0, 20:0, 20:1(n-9), 20:1(n-11), 22:1(n-9), 22:1(n-11) and 18:3(n-3) (Figure 16B).

The results of the SIMCA analysis (Figure 17) based on a model of the inner blubber from the Vesterålen whale samples, shows that the prey samples with average smallest RSD to the inner blubber model, are those laying closest at the scoreplot (Figure 16A). Herring got the lowest average RSD while Thysanoessa sp. got the highest average RSD (Figure 17). The prey species with the most similar FA composition to the inner blubber are herring, lesser sand eel, saithe, blue whiting and Meganyctiphanes sp. (Figure 16A and Figure 17).

A



B

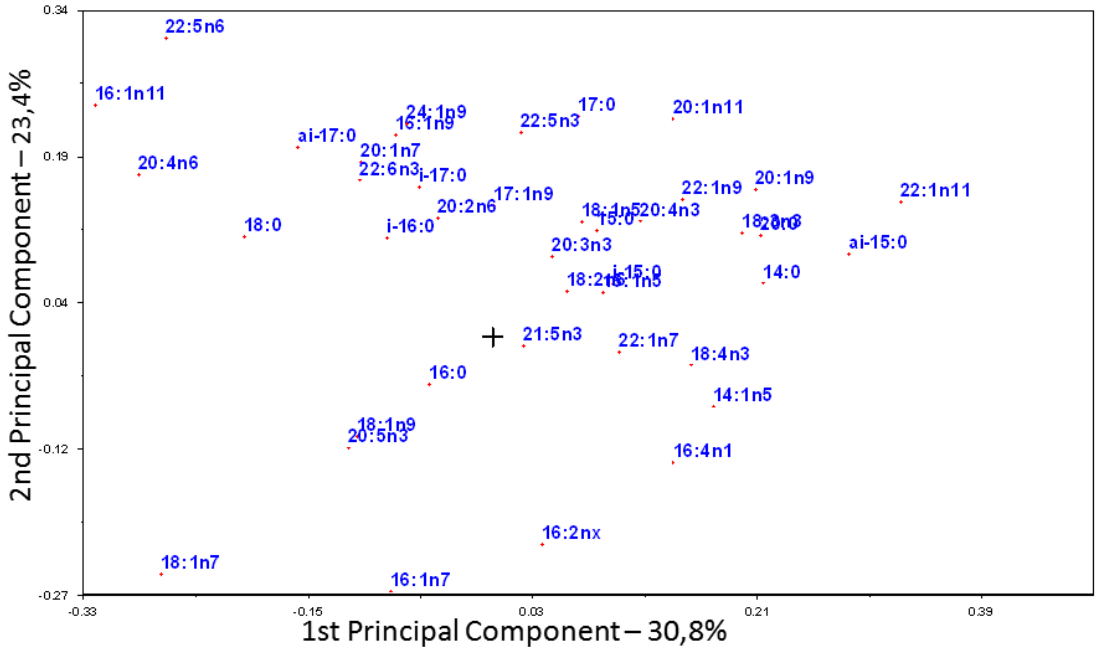


Figure 16

A: Score plot from a PCA with all inner blubber layer whale samples from the area Vesterålen together with samples from potential prey species. V = Vesterålen (indigo), Ha = haddock (red), Sa= saithe (pink), Me = krill of the species *Meganyctephanes* sp. (dark olive green), Co = cod (brown), T = krill of the species *Thysanoessa* sp. (green), SE = sand eel (orange), BW = blue whiting (blue-gray) and He = herring (gold).

B: Loading plot from a PCA with all inner blubber layer whale samples from the area Vesterålen together with the potential prey species; haddock, saithe, *Meganyctephanes* sp, cod, *Thysanoessa* sp, sand eel, blue whiting and herring.

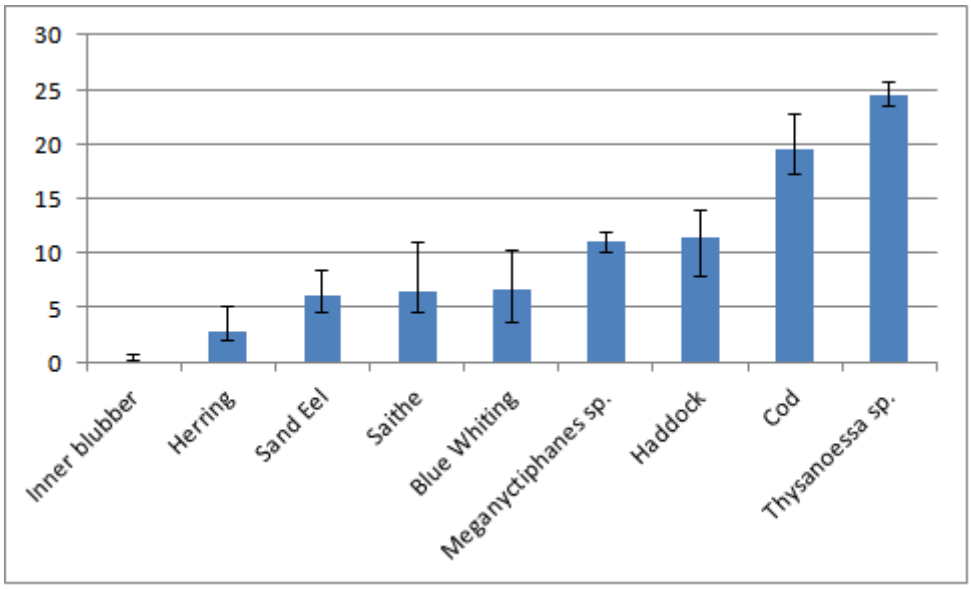


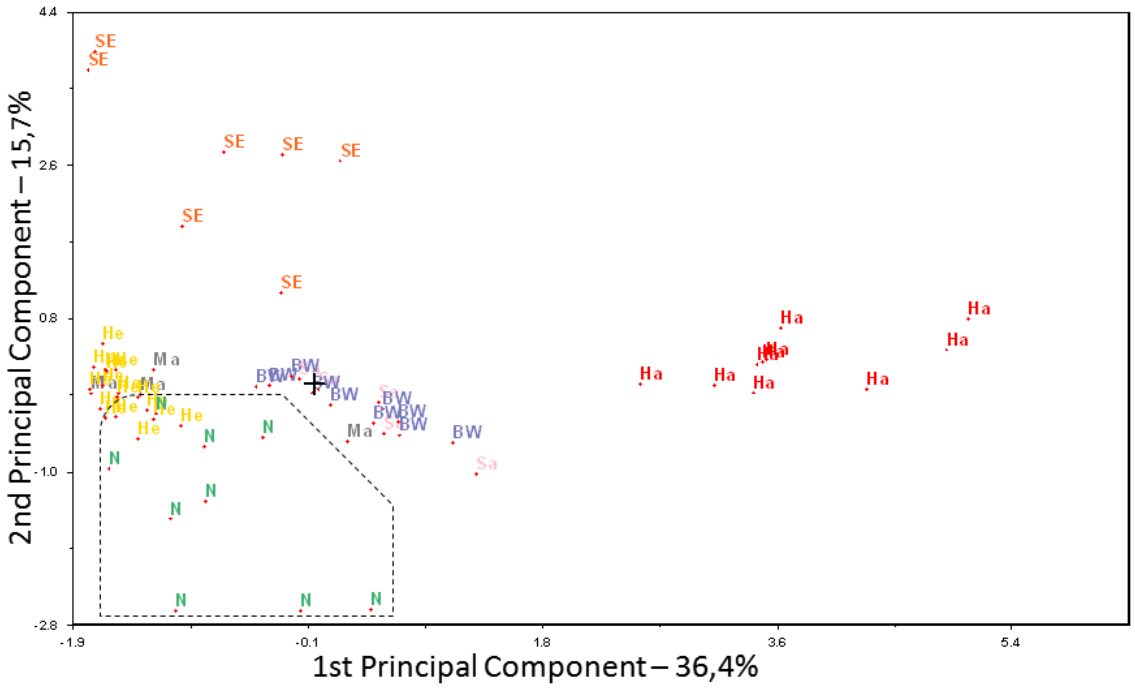
Figure17: Relative standard deviation of the average values of the Vesterålen blubber and potential prey samples, from Table 4, from a model of the inner blubber samples based on 41 FAs. The model has 1 significant component (as tested by cross validation), covering 79.5 % of the total variance among the samples. The lowest and highest RSD of individual samples for each blubber/prey are indicated as error bars.

3.4.3. The North Sea

All inner blubber layer samples from the 2009 North Sea whales were included in a PCA together with the potential prey species; haddock, sand eel, blue whiting, saithe, mackerel, and herring (Figure 18A). The two components PC1 and PC2 were together able to explain 52.1 % of the variance. The whale samples were positioned in the lower left corner of the plot, with herring and mackerel lying close above. Blue whiting and saithe were also lying close to the whale samples, a bit to the left. Sand eel were lying a bit above, and haddock were lying to the left in plot. This plot suggest that herring, mackerel, blue whiting and saith is the most likely prey of the whales from this area. The fatty acids responsible for the position of whale samples were 14:0, ai-15:0, 20:0, 14:1(n-5), 20:1(n-9), 20:1(n-11), 22:1(n-11), 18:3(n-3), 18:4(n-3) and 20:4(n-3) (Figure 18B).

The results of the SIMCA analysis (Figure 19) based on a model of the inner blubber from the North Sea whale samples, shows that the prey samples with average smallest RSD to the inner blubber model, are those laying closest at the scoreplot (Figure 18A). Mackerel and herring got the lowest average RSD while haddock got the highest average RSD (Figure 19). The prey species with the most similar FA composition to the inner blubber are mackerel, herring, blue whiting and saith (Figure 18A and Figure 19).

A



B

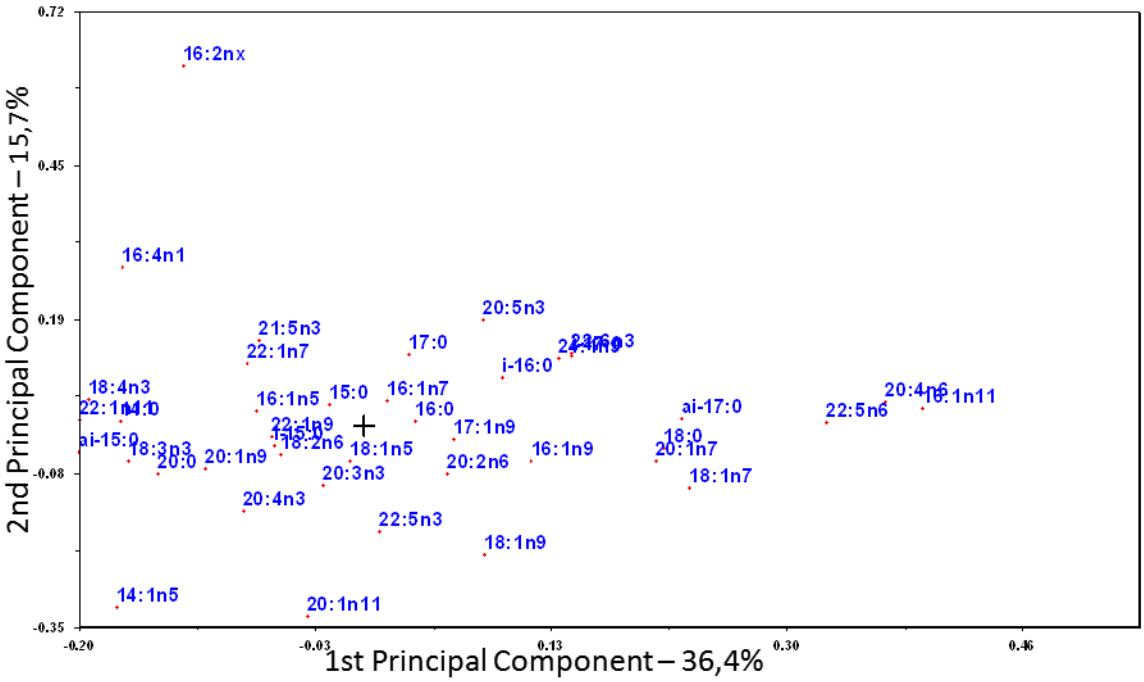


Figure 18

A: Score plot from a PCA with inner blubber layer whale samples from the area the North Sea together with samples from potential prey species. N = the North Sea (Sea-green), Ha = haddock (red), Sa= saithe (pink), Ma= mackerel (gray), Co = cod (brown), SE = sand eel (orange), BW = blue whiting (blue-gray) and He = herring (gold).

B: Loading plot from a PCA with inner blubber layer whale samples from the area the North Sea together with the potential prey species; haddock, saithe, mackerel, cod, sand eel, blue whiting and herring.

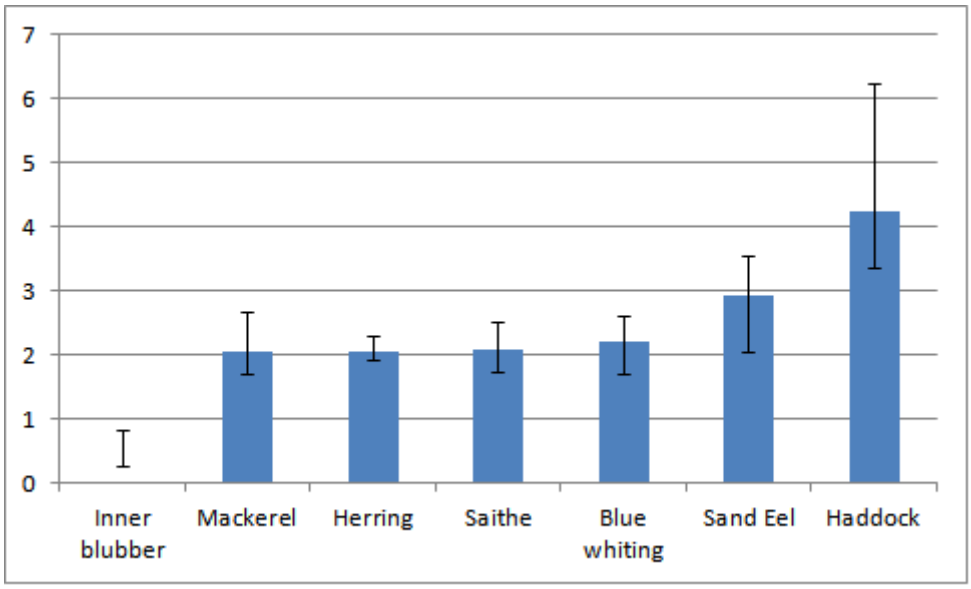


Figure 19: Relative standard deviation of the average values of the North Sea blubber and all potential prey samples, from Table 4, from a model of the inner blubber samples based on 41 FAs. The model has 2 components, the first is significant while the second has a CsvSD at 1.00 (as tested by cross validation). The two components are covering 77.2 % of the total variance among the samples. The lowest and highest RSD of individual samples for each blubber/prey are indicated as error bars.

4.0. Discussion

4.1. Blubber stratification

Most reported blubber FA analyses include only subsamples from outer and inner blubber layer, while only a few include subsamples from the middle layer (Lund 1936; Ackman et al 1965; Fredheim et al 1995 and Olsen & Grahl-Nielsen 2003 Strandberg et al 2008). In those cases, the middle layer had a FA composition which was intermediate between the inner and outer layer, and a continuous change in FA composition from the inner to the outer layer was therefore suggested. In the present investigation the relative amount of almost all FAs were either increasing or decreasing towards the skin or towards the muscle (Table 3), while some FAs were found with highest or lowest relative amount in the middle layer. This was also found by Olsen & Grahl-Nielsen (2003) and Strandberg et al (2008).

Stratification in FA was found with higher amounts of SFA and MUFA with 20 carbon atoms in the inner layer, and highest amounts of short-chain MUFA with 14, 16, 17 and 18 carbon atoms in the outer layer (Table 2, Figure 8;9). Similar stratification pattern is found in previous FA investigations in minke whale (Fehn 1996; Olsen & Grahl-Nielsen 2003; Møller et al 2003) and in other marine mammals (Fredheim et al 1995; Käkelä and Hyvärinen 1996; Koopman et al 1996; Best el at 2003; Grahl-Nielsen et al 2005;) and is thought to be a characteristic for marine mammal blubber (Strandberg et al 2008).

The stratification was also evident from the PCA-analysis of the 2010 samples from inner, middle and outer blubber layer (Figure 9A). The samples from the outer layer grouped jointly, and at some distance from the inner and middle layer samples. In particular it was the FAs 14:1(n-5), 16:1(n-7), 16:1(n-11) and 17:1(n-9) that explained the position of the outer blubber layer samples, while 16:4(n-1), 18:4(n-3), 22:1(n-7), 22:1(n-9), 22:1(n-11) and 24:1(n-9) explained the position of the middle and inner blubber layer (Figure 9B). These FAs was also the same found with highest concentration in the respective layers (Table 3 and Figure 8).

The observed stratification pattern in FA distribution seems to be similar in all species of marine mammals (Strandberg et al 2008), indicating that the composition of the blubber is determined by the same mechanisms in all species, and are connected with the function of the blubber. The presence of short chain MUFAs in the outer layer is probably a result of selective deposition or biosynthesis in the blubber, aimed to make the outer blubber appropriate for insulation and body streamlining (see e.g. Strandberg et al 2008; Ryg et al 1988; Koopman et al 2002). Unsaturated FAs and FAs with low chain lengths have low melting points. A low melting point is advantageous in maintaining membrane fluidity, and is also reducing the potential for heat loss across the body surface (Stryer 1988 refereed in Best et al 2003). The stratification between inner and outer blubber, with higher numbers of MUFAs with low chain lengths in the outer layer could therefore be explained by the thermal capacity for these FAs in the outer layer.

The underlying biochemical mechanisms involved in the maintenance of the outer layer it not clear, but slow mobilization or efficient *de novo* synthesis of specific TAG is suggested (Strandberg et al 2008). The enzyme Δ-9 desaturase are induced and activated by low tissue temperature, and introduces a double bond into SFA to make low-chain MUFAs (Tiku et al 1996; Kouba et al 1999). Strandberg et al (2008) calculated the Δ9-desaturation index (Δ-9-DI), the ratio between potentially endogenous created MUFA and the corresponding SFA, in the blubber of the ringed seal. They found that the Δ-9-DI values were highest in the outermost layers of blubber and decreased towards the inner blubber. The vertical profile curve Δ-9-DI dropped steeply in the outer section of the blubber and then ceased at the end of the outer layer. The enzyme Δ-9 desaturase are one mechanism making the stratification by creation of low-chain MUFA in the outer layer. The mechanisms involved in stratification need more study.

The inner layer serves mainly as a short term energy reserve, and is the primary site for lipid mobilization (Koopman et al 2002). The FAs from the most recent diet is first deposited in this blubber layer. The middle layer is a tissue for energy storage, and is not influenced heavily by environmental temperatures or recent metabolic activity (Strandberg et al 2008). Large variation in blubber thickness was found in this study (Table 2), which can be related to the metabolic state of the whale. Seasonal variation in blubber thickness is found (Næss et al 1998). At periods of intensive feeding the middle layer is expanding due to lipid deposition, and at periods of starvation the layer is shrinking.

4.2. Variation in blubber composition - in relation to catch position and observed stomach content

In the PCA with all inner blubber whale samples (Figure 10A), the samples from the North Sea and the samples from Vesterålen grouped relatively close, while the samples from Spitsbergen and Bjørnøya grouped at some distance from the two former. The Vesterålen whales and the North Sea whales seem therefore to be more similar to each other in FA composition, as compared with the Spitsbergen and Bjørnøya samples. The PCA-plot marked with the stomach content of the whales (Figure 10B) shows that whales with herring and sand eel in the stomach group close and at some distance from krill/capelin and codfish whales. This coincide with expected diets of the whales in the different areas from previous stomach content analysis, were sand eel is found dominant in North Sea whales (Olsen & Holst 2001), herring in Vesterålen whales (Lydersen et al 1991) and krill/capelin and codfish in Spitsbergen/Bjørnøya whales (Haug et al 1996; 2002; Folkow et al 2000; Windsland et al 2007).

Exception is two North Sea whales which group with the Spitsbergen and Bjørnøya whales (Figure 10A). The reason for this is not obvious, but one possible explanation could be that the two whales had arrived relatively recent in the North Sea. e.g., they had been foraging in more northern areas before they were shot at the southern location.

Differences in FA profiles between whales from different areas, as found in this study, has also been found between samples from the Norwegian Sea and the North Sea, by Olsen & Grahl-Nielsen (2003). Møller et al (2003) suggested a "three-geographic region model" of minke whales based on FA signatures, were the regions were Greenland, the Northeast Atlantic and the North Sea.

The current PCA-plots (Figure 10ABC) imply that there is a relationship between FA composition of inner blubber samples, catch position and observed stomach content. The important question is whether this area difference can be related to difference in diet. The integration of prey into blubber is shown to take periods from weeks to months (Bowen 2000; Arim & Naya 2003), and in harp seals it has been suggested that the appearance of new dietary FAs are evident within 1-2 weeks after a switch in diet (Kirsch et al 2000; Iverson et al 2004). The prey species present in the whale stomach when it is shot, is therefore thought to have no influence on the FA composition, other than indicating the most likely diet of the whale. If the whale stays in an area, and forage there for a longer period (weeks to months), it is reasonable to assume that the blubber FA composition of the whale can be "colored" by the diet in the area.

Møller et al (2003) explains the observed difference in FA composition between minke whales from the different regions of the North Atlantic by differences in diet, short-term diet differences in the inner blubber and long-term average diet differences in the outer blubber. Borobia et al (1995) have supposed that interspecific differences in FA composition of the outer blubber of fin whales and humpback whales is caused by difference in long term diet, but they also suggest that differences in metabolism and/or genetics, in addition to differences in diet, may explain the species differences.
The PCA-plot with outer blubber samples from 2010 (Figure 11A) shows no clear distribution pattern, but most samples from Spitsbergen and Bjørnøya grouped together, and four of the five Vesterålen samples grouped together. The marking of the samples by their stomach content (Figure 11B), does not give any clearer distribution pattern.

Obviously, the pattern among the whale samples due to catch locality and stomach content are easier visible in the inner layer samples than in the outer layer samples. This indicates that the variation in FA composition of whales from the same area is larger in inner blubber layer than in outer blubber layer, were the FA profiles are more similar. A potential diet influence in blubber is more likely to be present in inner blubber layer, than in outer blubber layer. The outer blubber of almost all marine mammals is thought to have the same FA pattern, and diet have little or no influence on this layer (Strandberg et al 2008). The inner blubber differs more between whales due to different diet, and/or genetics, as earlier described.

4.3. Inner blubber layer in relation to possible prey species

4.3.1. All areas pooled

In the PCA-analysis run on all whale samples and all prey samples (Figure 12A), the whales grouped jointly. Mackerel, herring, blue whiting, sand eel, saithe and *Meganyctiphanes* sp. was the prey species positioned closest to the whale samples, and was therefore thought to be the most likely prey species. This is supported by the results of the SIMCA analysis (Figure 13) which shows that the prey species with smallest RSD-values are those which are grouped closest to the whales at the scoreplot.

The corresponding loading plot (Figure 12B) shows that the fatty acids responsible for the grouping of whale samples are; 14:0, ai-15:0, 20:0, 14:1(n-5), 20:1(n-9), 20:1(n-11), 22:1(n-9), 22:1(n-11) 18:3(n-3), 20:4(n-3) and 22:5(n-3). The level of these fatty acids in the whale blubber is therefore high compared with the prey species. Long chain MUFAs, 20:1 and 22:1 are FATMs which indicate carnivores eating calaniode copepods (Dalsgaard et al 2003). The FAs 20:1(n-9), 20:1(n-11), 22:1(n-9) and 22:1(n-11) indicate that the whales are linked to calanus based food chains. 22:5(n-3) are also a tracer of diatoms (Dalsgaard et al 2003;Falk-Petersen et al 2004), indicating diatoms as important phytoplankton in the bottom of the food chain.

The FA 18:1(n-11) was found in all blubber layers (Table 3), but was almost not present in the FA composition of the prey (Table 4). This FA is a result of peroxisomal β-oxidation of 22:1(n-11) and 20:1(n-11), while FA metabolism usually happens with mitochondrial β-oxidation, and the entire FA is transferred into energy (Cooper 2006; Käkelä et al 2009). This means that this FA is not related to diet, other than indirectly because it originates from the long chain MUFAs. This FA is therefore left out of the PCA-analyses.

4.3.2. Spitsbergen/Bjørnøya

The prey species grouping closest to the whale samples from Spitsbergen/Bjørnøya in the PCA-plot (Figure 14A) and the prey species with the lowest RSD-values (Figure16) compared with the inner whale blubber, were capelin, polar cod, cod and krill (*Meganyctiphanes sp*). Those species seems, therefore to have the most similar FA composition to the inner blubber layer of the whales from the area. These prey species were previously found to be the primary diet of the whales from stomach content analysis in the area (Folkow et al 2000; Haug et al 1996; 2002; Windsland et al 2007), and they are in accordance with observation of stomach content in the whales.

Among the variables responsible for the positioning of the whale samples, are the calanus FATMs: 20:1(n-7), 20:1(n-9), 20:1(n-11) and 22:1(n-11), and the diatom FATM 22:5(n-3) (Figure 14B). 22:5(n-3) explains the position of the Spitsbergen/Bjørnøya whales in several PCA-plots (Figure 10A, Figure 12, Figure14) but not the position of whales from Vesterålen (Figure 16) and the North Sea (Figure 18), and might therefore be a trait for the Spitsbergen/Bjørnøya whales. Diatoms are

thought to be the major phytoplankton in spring blooms in the Barents Sea (Sakshaug et al 2009). It is therefore reasonable to find FATMs of diatoms in the FA profiles of the whales from the northern most waters. The spring bloom happens earlier in more southern waters, and the food webs here are probably dominated by other microalgae, for instance dinoflagellates.

4.3.3. Vesterålen

The prey species grouping closest to the Vesterålen whales in both the PCA-plot (Figure 16A) and the species which had the smallest RSD-values (Figure 17) compared with inner whale blubber are herring, blue whiting, saithe and krill (*Meganyctiphanes sp*). These prey species were also observed to be the diet of the whales from previous stomach content analysis in the area (Lydersen et al 1991, Windsland et al 2007).

Among the variables responsible for the position of the whale samples in this area are the calanus FATMs: 20:1(n-9), 20:1(n-11), 22:1(n-9), 22:1(n-11) found in close vicinity to the whale samples (Figure 16B). C18 PUFAs are regarded as FATMs for dinoflagellates (Dalsgaard et a 2003; Falk-Petersen 2004), and 18:3(n-3) are found among the variables positioning the whale samples. Dinoflagellates are thought to dominate the phytoplankton community in the summer, when the water is getting stratified and nutrient limited (Dalsgaard et al 2003).

4.3.4. The North Sea

The prey species grouping closest to the Vesterålen whales in the PCA-plot (Figure 18A) and the species which had the smallest RSD-values (Figure 19) compared with inner whale blubber are herring, mackerel, blue whiting and saithe. These prey species were observed to be the diet of the whales from previous stomach content analysis in the area (Olsen & Holst 2001).

Among the fatty acids responsible for the position of whale samples were the Calanus FATMs 20:1(n-9), 20:1(n-11), and 22:1(n-11), and the dinoflaggelate FATMs 18:3(n-3) and 18:4(n-3) (Figure 18B). These two FA are also responsible for the positioning of the whale samples in the PCA plot of all inner blubber samples. A link between the whales and dinoflagellates, an important primary producer, is therefore present in whale FA profiles from this area.

4.4. Is it possible to predict the minke whale diet from fatty acid analysis?

The FA composition of the potential prey species was all different from that of the inner blubber, but the prey species which in previous stomach content analyses had been observed to be the main prey for the whales at the different areas, were the ones laying closet to the whale samples at the PCA-plots, and the ones having the smallest RSD values. This indicates that there is a similarity in FA composition of expected prey species and the FA composition of the inner blubber of whales, and that it could be possible to tell the most likely diet of a whale from its FA composition, qualitatively.

One method in monitoring whale diet could be to determine the FA profile of small blubber samples of the inner blubber of a whale and compare it with a database of potential prey FA profiles, by the use of PCA and SIMCA. A match between the whale blubber and the most likely prey species could be found, by a defined probability value or a significance level. Different prey species has to be caught at different time during the season and at different locations, in order to compare the whales with the most representative prey as possible. The most optimal would be to compare the whales with prey caught in the same area one to two months before the whale, since it takes time before new FAs are evident in the blubber (Kirsch et al 2000; Iverson et al 2004).

The FA composition of the inner whale blubber is not an average of the potential prey FAs, since the samples are not placed in the middle of the prey in the PCA-plots (Figure 12A;14A;16A;18A), but are always placed aside the prey samples. This indicates that the minke whale FA composition always differ from its prey, and there is a general accepted view that the blubber does never match the diet entirely (Iverson et al 2004; Tucker et al 2009a,b; Grahl-Nielsen et al 2011). The FAs in the inner blubber layer are derived from the recent diet, but the FA composition of this tissue is different from the FA in the source of the lipids. This strongly indicates that the FAs are either metabolized or physiologically selected before incorporation into the blubber. This modification is likely caused by the chemical properties of the various FA to serve different functions in the blubber. The difference in FA composition between the inner blubber and prey is caused by different enzymes acting on FAs through digestion, transport and deposition in blubber. Grahl-Nielsen et al (2011) suggest that the metabolism will convert dietary FA composition to a given blubber composition regardless of the FA composition of the prey.

Other studies on diet and blubber samples have also shown that the FA composition of prey and predator differ, for instance Kirsch et al (2000) who found the blubber of harp seals different from FA composition in herring after a one year feeding experiment. The inner blubber of harbor seals from Spitsbergen had a FA composition that was significantly different from that of 18 prey species (Andersen 2001). Grahl-Nielsen et al (2011) tested the results of determined FA profiles in harp seal with that of 4 other investigations, based on seals from all three harp seal stocks. By use of SIMCA they tested their inner blubber model, with that of 55 inner blubber samples from all investigations together. The distances (measured as RSDs) showed a remarkable similarity between harp seals from the 3 different stocks, sampled over a period of more than 10 years. Since both spatial and temporal changes in consumed prey must have taken place, a genetic effect on the FA composition must exist, to make them so similar.

Wether the diet of a predator can be quantatively estimated by the QFASA method or not, is intensively debated. One part suggest that lipids are transferred into storage in blubber in an easy and direct way, and that mathematically estimated calibration coefficients can account for metabolism of FAs in the blubber (Iverson et al 2004; Thiemann 2009). Characteristic FA signatures from prey such

as marine invertebrates and fish are thought to be deposited in the predator in a predictable way. To estimate the diet composition by predators should be possible by comparing the FA signatures of all potential prey to a fat sample of the predator, using a statistical model. The diet is here thought to be the main determinant of blubber FA composition. The opponent part suggests that the composition of FA in blubber is determined mainly by metabolism, and that diet is not related to FA composition (Olsen & Grahl-Nielsen 2003;Grahl-Nielsen et al 2004;2009;2011). The knowledge about how dietary FAs affect the FA composition of the blubber is also found not sufficient enough, to draw conclusions about the diet of predators.

To quantitatively estimate the diet of minke by the QFASA method as suggested by Iverson et al (2004), would be impossible, because the method is based on calibration coefficients which compensate for metabolism in the blubber, and they are calculated by feeding experiments on captured animals. To capture minke whales and other large cetaceans, in order to examine the effect of each potential prey species on blubber composition, would be impossible, because of their big size.

Specific FA-ratios between FAs, might overrule the metabolism in the blubber, according to the FATM concept (Dalsgaard et al 2003). FATMs can be identified to specific taxa such as diatoms and dinoflagellates, and they can provide insight into energy transfer from one trophic level to the next. Falk-Petersen et al (2009) could find that FAs in harp seals originate from diatom dominated food chains, whereas hooded seals originate from dinoflagellate and the prymnesiophyte *Phaeocystis phouchetii*-based food chains, by the use of FATMs.

The long chained MUFAs 20:1 and 22:1 are Calanus FATMs (Dalsgaard et al 2003), and all inner whale samples analyzed contained high numbers of these FA (Figure 12B;14B;17B;18B). A link between minke whales and calanus based food chains is therefore present. C16 PUFAs are FATMS indicating diatoms, while C18 PUFAs are FATMS indicating dinoflagellates (Dalsgaard et al 2003; Falk-Petersen et al 2004). A link between diatoms and Spitsbergen/Bjørnøya whales (Figure 14B), and dinoflagellates and theVesterålen and the North Sea whales (Figure 18B) are present. According to the FATM concept, would it therefore be possible to trace the minke whales back to the origin of the food webs, the primary and secondary consumers, by use of FA profiles from analyzed blubber samples.

4.5. Method evaluation – potential sources of error

One of the biggest constrains in this study is that the prey species are not simultaneous in space and time. The optimal solution would be to capture prey 1-2 months before the whales and in the same area. Unfortunately, this was not possible in this master-project. FA composition of fish is thought to vary with distribution and time of the season (see e.g. Dalsgaard et al 2003). An example is herring were FA composition changes during sexual maturation and spawning (Henderson et al 1984). Some prey samples used in the analyses were caught in a different area than the whales, for instance haddock which was caught in the Barents Sea, and used in analyses of North Sea whales, and so on. The

difference between the inner blubber samples and the prey might be larger, than the “real” differences, because the FA composition is much different than that of the original prey consumed by the whales.

Outer blubber samples from 2009 are missing. Therefore it was not possible to compare exactly the North Sea whales with the Spitsbergen/Bjørnøya whales and the Vesterålen whales. The PCA plots of inner blubber samples and outer blubber samples is not based on the same amount of objects, so the comparison between them, will not tell the whole true picture about inner and outer blubber layer samples.

5.0. Conclusion

Stratification between inner, middle and outer blubber layer is present, in order to meet the different functional roles of the blubber.

Use of FA profiles in bio monitoring the diet of minke whales seems like an applicable method, since a relationship between inner blubber and the prey species expected to be the minke whale prey exist. A difference in FA profiles between minke whales caught in different areas, is present in the inner blubber layer, but not in the outer blubber layer. The knowledge of how metabolism affect the incorporation of FAs into the blubber is not good enough, and it is therefore difficult to draw any strong conclusions on FA profiles from whale blubber and potential prey.

However, the use of FATMs seems promising, and it’s reasonable that some FAs might overrule the metabolism in the blubber. The minke whales in the study can be traced back to the origin of the food web. All whales were traced to calanus based food webs, Spitsbergen/Bjørnøya whales were traced to diatoms, and Vesterålen and North Sea whales was traced to dinoflagelates.

6.0. References

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7.0. Appendix

Table 1. Average percentage share all fatty acids from the validation. Sample RB1 used (O = outer, MA-ME middle, I = Inner, se figure 3

	O	MA	MB	MC
14:0	5,8 ± 0,2	6,3 ± 0,1	7,0 ± 0,0	7,0 ± 0,1
Iso 15:0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,3 ± 0,0
Antiso 15:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
15:0	0,4 ± 0,0	0,4 ± 0,0	0,4 ± 0,0	0,4 ± 0,0
Iso 16:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:0	8,8 ± 0,3	9,4 ± 0,1	9,9 ± 0,1	10,7 ± 0,1
Iso 17:0	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
Antiso 17:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
17:0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
18:0	1,8 ± 0,0	1,7 ± 0,0	1,7 ± 0,0	2,0 ± 0,1
20:0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
∑SFA	**17,9 ± 0,5**	**19,0 ± 0,2**	**20,1 ± 0,1**	**21,2 ± 0,1**
14:1 (n-5)	0,8 ± 0,1	0,6 ± 0,0	0,3 ± 0,0	0,2 ± 0,0
16:1 (n-11) ?	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:1 (n-9)	0,3 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
16:1 (n-7)	11,0 ± 0,6	9,4 ± 0,1	7,5 ± 0,1	6,9 ± 0,2
16:1 (n-5)	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
17:1 (n-9)	0,4 ± 0,0	0,4 ± 0,0	0,3 ± 0,0	0,2 ± 0,0
18:1 (n-11)	1,3 ± 0,2	1,0 ± 0,1	1,1 ± 0,1	1,1 ± 0,2
18:1 (n-9)	16,4 ± 0,8	13,9 ± 0,3	11,4 ± 0,2	12,2 ± 0,2
18:1 (n-7)	2,6 ± 0,1	2,2 ± 0,0	2,1 ± 0,0	2,4 ± 0,0
18:1 (n-5)	0,4 ± 0,0	0,4 ± 0,0	0,4 ± 0,0	0,4 ± 0,0
20:1 (n-11)	2,4 ± 0,5	1,4 ± 0,3	1,5 ± 0,3	1,6 ± 0,1
20:1 (n-9)	12,2 ± 0,6	14,2 ± 0,2	15,9 ± 0,2	16,3 ± 0,1
20:1 (n-7)	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
22:1 (n-11)	12,8 ± 1,0	16,1 ± 0,2	16,8 ± 0,2	14,5 ± 0,1
22:1 (n-9)	0,7 ± 0,0	0,9 ± 0,0	1,1 ± 0,0	1,2 ± 0,0
22:1 (n-7)	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
24:1 (n-9)	0,3 ± 0,0	0,4 ± 0,0	0,6 ± 0,0	0,7 ± 0,0
∑MUFA	**62,5 ± 0,5**	**62,1 ± 1,3**	**60,2 ± 1,3**	**58,7 ± 1,1**
16:2 nx	0,0 ± 0,0	0,0 ± 0,0	0,0 ± 0,0	0,0 ± 0,0
18:2 (n-6)	1,8 ± 0,1	1,6 ± 0,0	1,5 ± 0,0	1,6 ± 0,0
20:2 (n-6)	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
20:4 (n-6)	0,4 ± 0,0	0,4 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
22:5(n-6)	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,1 ± 0,0
18:3 (n-3)	1,2 ± 0,0	1,1 ± 0,0	0,9 ± 0,0	0,9 ± 0,0
18:4 (n-3)	1,6 ± 0,1	2,0 ± 0,0	2,4 ± 0,0	2,5 ± 0,0
20:3 (n-3)	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,1 ± 0,0
20:4 (n-3)	1,4 ± 0,0	1,2 ± 0,0	1,0 ± 0,0	1,0 ± 0,0
20:5 (n-3)	4,2 ± 0,1	4,2 ± 0,1	4,5 ± 0,1	4,1 ± 0,1
21:5 (n-3)	4,2 ± 0,0	4,2 ± 0,1	4,5 ± 0,1	4,1 ± 0,1
22:5 (n-3)	2,4 ± 0,1	2,1 ± 0,1	1,8 ± 0,0	2,0 ± 0,1
22:6 (n-3)	5,6 ± 0,1	5,5 ± 0,1	6,1 ± 0,1	6,7 ± 0,0
∑PUFA	**19,6 ± 0,1**	**18,9 ± 0,3**	**19,7 ± 0,2**	**20,0 ± 0,1**

	MD	ME	I
14:0	7,4 ± 0,1	6,3 ± 0,2	6,4 ± 0,2
Iso 15:0	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
Antiso 15:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
15:0	0,4 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
Iso 16:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
0,00	11,6 ± 0,3	9,8 ± 0,4	9,9 ± 0,6
Iso 17:0	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
Antiso 17:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
17:0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
18:0	2,1 ± 0,0	2,5 ± 0,1	2,2 ± 0,3
20:0	0,2 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
∑SFA	**22,7 ± 0,4**	**20,1 ± 0,6**	**20,2 ± 0,8**
14:1 (n-5)	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:1 (n-11)	0,1 ± 0,0	0,0 ± 0,0	0,1 ± 0,0
16:1 (n-9)	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
16:1 (n-7)	7,3 ± 0,1	5,7 ± 0,2	5,8 ± 0,3
16:1 (n-5)	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
17:1 (n-9)	0,3 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
18:1 (n-11)	1,0 ± 0,1	1,3 ± 0,2	1,2 ± 0,1
18:1 (n-9)	12,8 ± 0,3	15,5 ± 0,1	15,6 ± 0,4
18:1 (n-7)	2,5 ± 0,1	2,9 ± 0,0	2,9 ± 0,0
18:1 (n-5)	0,4 ± 0,0	0,5 ± 0,0	0,5 ± 0,0
20:1 (n-11)	1,4 ± 0,2	1,9 ± 0,3	1,6 ± 0,3
20:1 (n-9)	16,0 ± 0,1	20,1 ± 0,4	20,0 ± 0,7
20:1 (n-7)	0,3 ± 0,0	0,5 ± 0,0	0,5 ± 0,0
22:1 (n-11)	13,7 ± 0,1	12,8 ± 0,2	13,0 ± 0,4
22:1 (n-9)	1,1 ± 0,0	1,5 ± 0,0	1,4 ± 0,2
22:1 (n-7)	0,2 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
24:1 (n-9)	0,6 ± 0,0	0,9 ± 0,0	0,9 ± 0,1
∑MUFA	**58,4 ± 1,1**	**64,7 ± 1,5**	**64,6 ± 2,5**
16:2 nx	0,0 ± 0,0	0,0 ± 0,0	0,0 ± 0,0
18:2 (n-6)	1,7 ± 0,0	1,8 ± 0,0	1,8 ± 0,0
20:2 (n-6)	0,2 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
20:4 (n-6)	0,3 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
22:5(n-6)	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
18:3 (n-3)	0,9 ± 0,0	0,6 ± 0,0	0,6 ± 0,0
18:4 (n-3)	2,4 ± 0,1	1,7 ± 0,0	1,7 ± 0,1
20:3 (n-3)	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
20:4 (n-3)	1,0 ± 0,0	0,7 ± 0,0	0,8 ± 0,0
20:5 (n-3)	3,8 ± 0,1	2,3 ± 0,2	2,3 ± 0,2
21:5 (n-3)	3,8 ± 0,1	2,3 ± 0,2	2,3 ± 0,2
22:5 (n-3)	1,9 ± 0,1	2,2 ± 0,1	2,2 ± 0,1
22:6 (n-3)	5,8 ± 0,5	4,6 ± 0,4	4,7 ± 0,2
∑PUFA	**18,9 ± 0,8**	**15,1 ± 0,6**	**15,3 ± 0,3**

Table 2. Summary over sample information.

Boatname	Whale nr	Sample	BT*	Lati	GrMi	Long	GrMi	Sex	L (cm)*	Stomack content
Reinebuen	1	RB 1	7	N	7431	E	1817	Female	740	Codfish (haddock), 3/4 full, numerous nematodes
Reinebuen	2	RB 2	4	N	7420	E	2035	Female	565	Whitefish, haddock? Quite digested. Almost full, no nematod
Reinebuen	3	RB 3	5,5	N	7651	E	1323	Female	812	Grey matter, unidentified, 1/3 full
Reinebuen	4	RB 4	6	N	7650	E	1327	Female	826	Well digested krill + some capelin, 1/2 full
Reinebuen	5	RB 5	7	N	7653	E	1321	Female	710	Little digested small capelin, 90% full
Reinebuen	6	RB 6	6,5	N	7653	E	1321	Female	707	Capelin (80%). Parathemisto (20%) full
Reinebuen	7	RB 7	3,5	N	7714	E	1233	Female	715	Well digested capelin, 1/3 full
Reinebuen	8	RB 8	6	N	7725	E	1125	Female	725	Capelin (90%), haddock (20 %), nematodes, 1/4 full
Reinebuen	9	RB 9	7,5	N	7725	E	1124	Female	810	Capelin (30%), krill (70%), middle digested, 1/5 full
Reinebuen	10	RB 10	8,5	N	7725	E	1143	Female	805	Well digested capelin, 1,3 full
Reinebuen	11	RB 11	7	N	7728	E	1140	Female	845	Krill/capelin, 50/50
Reinebuen	12	RB 12	6,5	N	7727	E	1141	Female	750	Some digested capelin/krill
Reinebuen	13	RB 13	5	N	7725	E	1140	Female	812	Pure capelin stomach, little digested, 1/2 full
Reinebuen	14	RB 14	6,5	N	7728	E	1148	Female	760	Digested capelin, some nematodes, 1/2 full
Reinebuen	15	RB 15	7,5	N	7728	E	1148	Female	852	Middle digested capelin, some nematodes, 1/2 full
Reinebuen	16	RB 16	8	N	7726	E	1150	Female	840	Well digested capelin + some whitefish, 3/4 full
Reinebuen	19	RB 19	10	N	7730	E	1146	Female	797	Well digested capelin, 1/2 full
Reinebuen	21	RB 21	4,5	N	7810	E	955	Female	666	Middle digested big haddock, 1/2 full
Reinebuen	22	RB 22	6,5	N	7808	E	1008	Female	731	Well digested capelin/krill, 50/50, 1/4 full
Reinebuen	23	RB 23	5,5	N	7732	E	1149	Female	760	Capelin, little digested, 1/4 full
Reinebuen	24	RB 24	6	N	7732	E	1146	Female	755	Capelin, some digested, 3/4 full
Reinebuen	25	RB 25	4,5	N	7702	E	1327	Female	643	Krill/capelin, (60/40), well digested, 1/3 full
Reinebuen	26	RB 26	4,5	N	7701	E	1310	Female	697	Well digested capelin (100%), 1/3 full
Reinefangst	19	RF 19	4,6	N	6951	E	1833	Male	680	Lesser sand eel, little stomach content
Reinefangst	20	RF20	4,6	N	6958	E	1806	Male	810	Not observed, but whale shot in the same area as whale 19
Reinefangst	21	RF 21	3,5	N	6942	E	1651	Male	765	Herring
Reinefangst	22	RF 22	3,5	N	6947	E	1649	Male	820	Herring
Reinefangst	23	RF 23	4,5	N	6952	E	1711	Male	800	Herring
Kato	1	K1	No data	N	5647	E	0400	Female	510	Sand eel
Kato	2	K2	No data	N	5652	E	0350	Female	830	Sand eel
Kato	3	K3	No data	N	5645	E	0417	Male	520	Sand eel
Kato	4	K4	No data	N	5645	E	0334	Female	670	Sand eel
Kato	5	K5	No data	N	5646	E	0337	Male	810	Sand eel
Kato	7	K7	No data	N	5649	E	0343	Female	720	Sand eel
Kato	8	K8	No data	N	5648	E	0347	Male	710	Sand eel
Kato	9	K9	No data	N	5705	E	0513	Female	880	Sand eel
Kato	10	K10_2T	No data	N	5706	E	0511	Male	790	Sand eel

*BT = blubber thickness *L = Length (body)

Table 3. Average percentage share all fatty acids outer, middle and inner blubber layer.

	Outer			Middle			Inner		
14:00	4,9	±	0,9	5,8	±	1,0	5,9	±	1,2
i-15:0	0,2	±	0,1	0,2	±	0,0	0,2	±	0,0
ai-15:0	0,1	±	0,0	0,1	±	0,0	0,1	±	0,0
15:0	0,4	±	0,0	0,4	±	0,1	0,3	±	0,1
i-16:0	0,1	±	0,0	0,1	±	0,0	0,1	±	0,0
16:0	9,2	±	0,9	10,8	±	1,3	11,4	±	1,5
i-17:0	0,4	±	0,1	0,3	±	0,1	0,3	±	0,1
ai-17:0	0,1	±	0,0	0,1	±	0,0	0,1	±	0,0
17:0	0,2	±	0,0	0,2	±	0,1	0,3	±	0,1
18:0	1,9	±	0,3	2,2	±	0,5	2,8	±	0,6
20:0	0,2	±	0,1	0,2	±	0,0	0,2	±	0,1
∑SFA	**17,5**	±	**1,5**	**20,4**	±	**1,5**	**21,6**	±	**1,8**
14:1n5	0,8	±	0,1	0,3	±	0,1	0,2	±	0,0
16:1n11	0,1	±	0,0	0,1	±	0,0	0,1	±	0,0
16:1n9	0,3	±	0,0	0,2	±	0,1	0,3	±	0,1
16:1n7	12,6	±	3,0	7,5	±	2,0	6,1	±	1,7
16:1n5	0,3	±	0,0	0,2	±	0,0	0,2	±	0,1
17:1n9	0,5	±	0,1	0,3	±	0,1	0,3	±	0,1
18:1n11	1,8	±	0,6	1,9	±	0,9	1,8	±	0,6
18:1n9	21,5	±	3,0	17,2	±	4,5	18,2	±	4,4
18:1n7	4,3	±	1,3	3,7	±	1,5	3,7	±	1,3
18:1n5	0,5	±	0,1	0,4	±	0,1	0,5	±	0,1
20:1n11	2,2	±	0,6	1,7	±	0,6	1,8	±	0,6
20:1n9	10,6	±	2,9	13,3	±	3,0	13,9	±	3,5
20:1n7	0,4	±	0,1	0,5	±	0,2	0,5	±	0,2
22:1n11	7,7	±	4,1	11,2	±	3,8	10,3	±	3,5
22:1n9	0,7	±	0,2	1,1	±	0,3	1,2	±	0,3
22:1n7	0,1	±	0,0	0,2	±	0,1	0,2	±	0,1
24:1n9	0,3	±	0,1	0,6	±	0,1	0,7	±	0,2
∑MUFA	**64,6**	±	**1,1**	**60,6**	±	**5,1**	**60,1**	±	**5,5**
16:2nx	0,4	±	0,1	0,3	±	0,1	0,3	±	0,1
16:4n1	0,1	±	0,1	0,1	±	0,1	0,1	±	0,1
18:2n6	1,8	±	0,2	1,5	±	0,2	1,4	±	0,2
20:2n6	0,3	±	0,0	0,3	±	0,0	0,3	±	0,1
20:4n6	0,4	±	0,1	0,3	±	0,1	0,3	±	0,1
22:5n6	0,1	±	0,0	0,1	±	0,1	0,2	±	0,1
18:3n3	1,0	±	0,1	0,8	±	0,4	0,8	±	0,4
18:4n3	1,2	±	0,3	2,1	±	1,1	1,8	±	1,2
20:3n3	0,1	±	0,0	0,1	±	0,0	0,1	±	0,0
20:4n3	1,2	±	0,3	1,0	±	0,3	0,9	±	0,3
20:5n3	3,4	±	1,3	3,9	±	1,4	3,4	±	1,2
21:5n3	0,2	±	0,1	0,3	±	0,1	0,3	±	0,1
22:5n3	2,6	±	0,8	2,1	±	0,6	2,6	±	0,7
22:6n3	5,0	±	1,8	6,0	±	2,0	5,7	±	1,8
∑PUFA	**17,8**	±	**3,5**	**19,0**	±	**4,6**	**18,4**	±	**4,2**

Table 4. Average percentage share all fatty acids, all prey species and all whale samples. RB = Reinebuen, RF = Reinefangst, K= Kato, O =outer, M =middle and I = inner.

	Haddock	Capelin	Saith	Mackerel
14:00	2,8 ± 0,6	4,8 ± 1,7	4,8 ± 0,7	7,4 ± 1,1
i-15:0	0,2 ± 0,1	0,3 ± 0,1	0,2 ± 0,0	0,3 ± 0,0
ai-15:0	0,0 ± 0,0	0,1 ± 0,0	0,0 ± 0,0	0,1 ± 0,0
15:0	0,4 ± 0,1	0,5 ± 0,1	0,4 ± 0,0	0,5 ± 0,0
i-16:0	0,1 ± 0,1	0,1 ± 0,0	0,0 ± 0,0	0,1 ± 0,0
16:0	15,0 ± 1,7	15,5 ± 1,4	14,6 ± 0,8	13,1 ± 1,8
i-17:0	0,5 ± 0,1	0,5 ± 0,1	0,4 ± 0,0	0,4 ± 0,1
ai-17:0	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
17:0	0,3 ± 0,0	0,2 ± 0,0	0,3 ± 0,0	0,4 ± 0,1
18:0	3,9 ± 0,5	2,1 ± 0,5	2,9 ± 0,3	2,2 ± 0,9
20:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,3 ± 0,0
∑SFA	**23,4 ± 1,8**	**24,3 ± 0,4**	**23,9 ± 0,6**	**24,7 ± 1,6**
14:1n5	0,0 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:1n11	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,0 ± 0,0
16:1n9	0,3 ± 0,1	0,3 ± 0,1	0,2 ± 0,1	0,2 ± 0,0
16:1n7	5,6 ± 1,5	5,1 ± 1,1	4,5 ± 0,5	3,5 ± 0,3
16:1n5	0,2 ± 0,0	0,4 ± 0,1	0,2 ± 0,0	0,3 ± 0,1
17:1n9	0,3 ± 0,1	0,3 ± 0,1	0,4 ± 0,1	0,3 ± 0,1
18:1n11	0,1 ± 0,1	0,0 ± 0,0	1,1 ± 0,8	0,3 ± 0,1
18:1n9	11,6 ± 0,9	9,3 ± 1,2	13,9 ± 5,4	8,3 ± 5,6
18:1n7	4,2 ± 0,7	2,8 ± 0,4	3,0 ± 0,9	1,6 ± 0,9
18:1n5	0,4 ± 0,1	0,6 ± 0,1	0,4 ± 0,1	0,4 ± 0,1
20:1n11	0,8 ± 0,2	0,4 ± 0,1	1,4 ± 0,2	0,7 ± 0,1
20:1n9	6,8 ± 2,6	7,1 ± 3,1	7,9 ± 1,5	7,7 ± 1,6
20:1n7	0,5 ± 0,1	0,3 ± 0,1	0,2 ± 0,1	0,2 ± 0,1
22:1n11	4,7 ± 2,9	5,9 ± 2,5	9,2 ± 2,1	14,3 ± 4,4
22:1n9	0,8 ± 0,2	0,6 ± 0,2	0,6 ± 0,1	0,9 ± 0,1
22:1n7	0,1 ± 0,1	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
24:1n9	1,6 ± 0,4	2,1 ± 0,6	0,7 ± 0,1	0,9 ± 0,1
∑MUFA	**38,4 ± 6,6**	**35,4 ± 6,6**	**44,0 ± 3,2**	**39,8 ± 0,6**
16:2nx	0,3 ± 0,1	0,3 ± 0,1	0,4 ± 0,1	0,3 ± 0,1
16:4n1	0,2 ± 0,1	0,4 ± 0,2	0,4 ± 0,2	0,4 ± 0,2
18:2n6	1,0 ± 0,2	1,3 ± 0,2	1,4 ± 0,2	1,7 ± 0,2
20:2n6	0,3 ± 0,1	0,2 ± 0,1	0,3 ± 0,0	0,3 ± 0,0
20:4n6	1,2 ± 0,3	0,7 ± 0,2	0,4 ± 0,1	0,4 ± 0,1
22:5n6	0,4 ± 0,1	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
18:3n3	0,4 ± 0,1	0,5 ± 0,1	1,2 ± 0,3	1,6 ± 0,3
18:4n3	1,1 ± 0,4	1,4 ± 0,3	3,1 ± 1,0	6,1 ± 1,6
20:3n3	0,1 ± 0,0	0,1 ± 0,0	0,2 ± 0,0	0,3 ± 0,0
20:4n3	0,4 ± 0,1	0,5 ± 0,1	0,8 ± 0,2	1,1 ± 0,1
20:5n3	12,0 ± 1,1	11,9 ± 2,2	8,2 ± 0,6	7,5 ± 0,8
21:5n3	0,3 ± 0,1	0,3 ± 0,0	0,4 ± 0,1	0,5 ± 0,1
22:5n3	1,0 ± 0,2	1,0 ± 0,2	1,1 ± 0,3	1,2 ± 0,5
22:6n3	19,5 ± 4,7	21,7 ± 4,7	14,0 ± 1,6	14,0 ± 0,7
∑PUFA	**38,2 ± 5,2**	**40,3 ± 6,7**	**32,1 ± 3,3**	**35,5 ± 2,0**

	Meganyctephanes	Cod	Polarcod	Thysanoessa
14:00	5,2 ± 0,2	3,1 ± 0,4	3,4 ± 0,9	2,0 ± 0,1
i-15:0	0,3 ± 0,0	0,2 ± 0,0	0,2 ± 0,1	0,1 ± 0,0
ai-15:0	0,1 ± 0,0	0,0 ± 0,0	0,0 ± 0,0	0,0 ± 0,0
15:0	0,5 ± 0,0	0,3 ± 0,0	0,3 ± 0,0	0,1 ± 0,0
i-16:0	0,1 ± 0,0	0,1 ± 0,1	0,1 ± 0,0	0,0 ± 0,0
16:0	15,0 ± 0,6	18,7 ± 0,8	14,3 ± 2,6	19,3 ± 0,3
i-17:0	0,4 ± 0,0	0,2 ± 0,0	0,3 ± 0,1	0,1 ± 0,0
ai-17:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,0 ± 0,0
17:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,0 ± 0,0
18:0	1,3 ± 0,1	3,6 ± 0,3	2,4 ± 0,6	1,6 ± 0,1
20:0	0,1 ± 0,0	0,0 ± 0,0	0,0 ± 0,0	0,0 ± 0,0
∑SFA	**23,2 ± 0,8**	**26,4 ± 1,0**	**21,4 ± 2,4**	**23,4 ± 0,4**
14:1n5	0,1 ± 0,0	0,1 ± 0,0	0,0 ± 0,0	0,1 ± 0,1
16:1n11	0,0 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,1
16:1n9	0,1 ± 0,0	0,2 ± 0,0	0,3 ± 0,1	0,0 ± 0,0
16:1n7	12,9 ± 0,6	7,6 ± 0,7	8,4 ± 2,1	17,8 ± 0,5
16:1n5	0,3 ± 0,0	0,2 ± 0,0	0,4 ± 0,1	0,1 ± 0,0
17:1n9	0,3 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,1 ± 0,0
18:1n11	0,0 ± 0,0	0,0 ± 0,0	0,0 ± 0,0	0,6 ± 1,0
18:1n9	10,2 ± 0,3	16,0 ± 0,8	10,8 ± 2,1	19,3 ± 1,0
18:1n7	4,5 ± 0,2	7,5 ± 0,5	3,9 ± 1,2	8,9 ± 0,3
18:1n5	0,5 ± 0,0	0,3 ± 0,0	0,5 ± 0,1	0,1 ± 0,0
20:1n11	0,5 ± 0,0	0,4 ± 0,1	0,9 ± 0,3	0,1 ± 0,1
20:1n9	10,5 ± 1,1	2,9 ± 0,6	11,5 ± 4,2	0,3 ± 0,3
20:1n7	0,6 ± 0,0	0,2 ± 0,0	0,3 ± 0,1	0,0 ± 0,0
22:1n11	6,9 ± 0,4	1,3 ± 0,2	6,3 ± 2,9	0,2 ± 0,0
22:1n9	1,0 ± 0,1	0,3 ± 0,0	1,1 ± 0,3	0,1 ± 0,1
22:1n7	0,2 ± 0,0	0,1 ± 0,0	0,3 ± 0,1	0,2 ± 0,0
24:1n9	0,6 ± 0,1	0,8 ± 0,2	1,4 ± 0,5	0,1 ± 0,1
∑MUFA	**49,3 ± 1,3**	**38,4 ± 1,7**	**46,5 ± 6,8**	**48,1 ± 1,7**
16:2nx	0,7 ± 0,0	0,4 ± 0,0	0,5 ± 0,2	1,0 ± 0,0
16:4n1	1,0 ± 0,1	0,2 ± 0,0	0,3 ± 0,2	0,5 ± 0,1
18:2n6	1,0 ± 0,0	1,1 ± 0,1	1,1 ± 0,2	0,8 ± 0,0
20:2n6	0,3 ± 0,1	0,3 ± 0,0	0,3 ± 0,1	0,1 ± 0,0
20:4n6	0,5 ± 0,0	0,8 ± 0,1	0,7 ± 0,3	0,3 ± 0,0
22:5n6	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,1	0,1 ± 0,1
18:3n3	0,3 ± 0,0	0,6 ± 0,1	0,5 ± 0,1	0,2 ± 0,1
18:4n3	2,0 ± 0,1	2,5 ± 0,3	1,3 ± 0,5	2,6 ± 0,1
20:3n3	0,3 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,0 ± 0,0
20:4n3	0,5 ± 0,0	0,4 ± 0,1	0,5 ± 0,1	0,2 ± 0,0
20:5n3	11,9 ± 0,8	14,3 ± 0,6	11,0 ± 2,0	18,1 ± 1,3
21:5n3	0,4 ± 0,0	0,5 ± 0,1	0,3 ± 0,0	0,3 ± 0,0
22:5n3	0,5 ± 0,0	0,6 ± 0,1	1,0 ± 0,3	0,1 ± 0,1
22:6n3	8,1 ± 0,6	13,4 ± 1,6	14,5 ± 4,0	4,1 ± 0,4
∑PUFA	**27,6 ± 1,6**	**35,2 ± 1,7**	**32,2 ± 5,3**	**28,5 ± 1,8**

	Sand eel	Blue whiting	Herring	RBO
14:00	5,9 ± 1,1	3,8 ± 0,9	7,7 ± 0,6	4,6 ± 0,6
i-15:0	0,2 ± 0,1	0,1 ± 0,0	0,3 ± 0,0	0,2 ± 0,1
ai-15:0	0,1 ± 0,0	0,0 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
15:0	0,4 ± 0,1	0,3 ± 0,0	0,5 ± 0,0	0,3 ± 0,0
i-16:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:0	13,9 ± 1,8	18,9 ± 2,1	13,9 ± 1,6	9,2 ± 0,9
i-17:0	0,5 ± 0,2	0,4 ± 0,0	0,2 ± 0,0	0,4 ± 0,1
ai-17:0	0,1 ± 0,0	0,1 ± 0,0	0,0 ± 0,0	0,1 ± 0,0
17:0	0,4 ± 0,1	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,1
18:0	2,2 ± 0,5	2,9 ± 0,5	1,1 ± 0,1	1,9 ± 0,3
20:0	0,1 ± 0,0	0,2 ± 0,0	0,1 ± 0,0	0,2 ± 0,1
∑SFA	**23,9 ± 1,8**	**27,0 ± 2,1**	**24,2 ± 1,6**	**17,3 ± 1,4**
14:1n5	0,0 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,8 ± 0,1
16:1n11	0,1 ± 0,0	0,1 ± 0,0	0,0 ± 0,0	0,1 ± 0,0
16:1n9	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,3 ± 0,0
16:1n7	7,1 ± 1,8	4,9 ± 0,8	4,8 ± 0,4	13,6 ± 2,2
16:1n5	0,3 ± 0,1	0,2 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
17:1n9	0,3 ± 0,1	0,4 ± 0,0	0,2 ± 0,0	0,5 ± 0,1
18:1n11	0,3 ± 0,1	0,4 ± 0,1	0,3 ± 0,1	1,8 ± 0,5
18:1n9	5,0 ± 1,1	14,9 ± 6,0	9,3 ± 2,3	21,9 ± 3,1
18:1n7	1,6 ± 0,3	2,8 ± 1,2	1,5 ± 0,4	4,7 ± 1,1
18:1n5	0,4 ± 0,1	0,3 ± 0,0	0,4 ± 0,1	0,5 ± 0,1
20:1n11	0,4 ± 0,1	1,1 ± 0,3	0,8 ± 0,1	2,1 ± 0,6
20:1n9	8,2 ± 4,1	6,6 ± 1,6	11,0 ± 1,8	9,9 ± 2,5
20:1n7	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,1	0,4 ± 0,0
22:1n11	12,3 ± 4,5	11,8 ± 2,9	17,1 ± 2,6	6,6 ± 2,8
22:1n9	0,8 ± 0,2	0,6 ± 0,1	1,0 ± 0,3	0,7 ± 0,1
22:1n7	0,2 ± 0,0	0,1 ± 0,0	0,2 ± 0,1	0,1 ± 0,0
24:1n9	1,1 ± 0,2	0,8 ± 0,1	0,9 ± 0,1	0,2 ± 0,1
∑MUFA	**38,5 ± 7,4**	**45,2 ± 3,3**	**48,4 ± 2,9**	**64,5 ± 4,2**
16:2nx	1,5 ± 0,7	0,3 ± 0,1	0,3 ± 0,1	0,4 ± 0,1
16:4n1	0,8 ± 0,2	0,4 ± 0,1	0,5 ± 0,2	0,1 ± 0,1
18:2n6	1,6 ± 0,6	1,3 ± 0,2	1,2 ± 0,2	1,8 ± 0,1
20:2n6	0,2 ± 0,1	0,3 ± 0,0	0,2 ± 0,0	0,3 ± 0,0
20:4n6	0,3 ± 0,1	0,5 ± 0,1	0,3 ± 0,1	0,4 ± 0,2
22:5n6	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
18:3n3	1,0 ± 0,2	0,9 ± 0,3	1,1 ± 0,2	1,0 ± 0,1
18:4n3	3,7 ± 0,5	2,7 ± 0,8	3,8 ± 0,7	1,1 ± 0,3
20:3n3	0,1 ± 0,0	0,2 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
20:4n3	0,6 ± 0,1	0,7 ± 0,1	0,5 ± 0,0	1,3 ± 0,3
20:5n3	11,6 ± 2,0	7,1 ± 0,4	8,5 ± 0,9	3,5 ± 1,3
21:5n3	0,6 ± 0,1	0,3 ± 0,1	0,3 ± 0,0	0,2 ± 0,1
22:5n3	0,9 ± 0,1	0,7 ± 0,1	0,6 ± 0,1	2,7 ± 0,7
22:6n3	14,7 ± 4,3	12,2 ± 0,9	9,9 ± 1,0	5,1 ± 1,8
∑PUFA	**37,7 ± 6,2**	**27,7 ± 2,6**	**27,3 ± 2,1**	**18,2 ± 3,4**

	RBM	RBI	RFO	RFM
14:00	5,5 ± 0,8	5,5 ± 0,9	6,0 ± 1,1	7,2 ± 1,0
i-15:0	0,2 ± 0,0	0,2 ± 0,1	0,3 ± 0,0	0,2 ± 0,0
ai-15:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
15:0	0,3 ± 0,1	0,3 ± 0,0	0,4 ± 0,0	0,4 ± 0,0
i-16:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:0	10,9 ± 1,4	11,3 ± 1,6	9,1 ± 0,9	10,3 ± 0,6
i-17:0	0,3 ± 0,1	0,3 ± 0,1	0,4 ± 0,0	0,4 ± 0,1
ai-17:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
17:0	0,2 ± 0,1	0,3 ± 0,1	0,2 ± 0,0	0,2 ± 0,0
18:0	2,3 ± 0,5	2,9 ± 0,6	1,8 ± 0,4	1,8 ± 0,2
20:0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
∑SFA	**20,3 ± 1,6**	**21,3 ± 1,8**	**18,6 ± 1,6**	**20,9 ± 0,5**
14:1n5	0,3 ± 0,1	0,2 ± 0,0	0,6 ± 0,1	0,2 ± 0,0
16:1n11	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:1n9	0,2 ± 0,1	0,3 ± 0,1	0,3 ± 0,0	0,2 ± 0,0
16:1n7	8,0 ± 1,8	6,5 ± 1,6	8,3 ± 2,3	5,3 ± 0,8
16:1n5	0,2 ± 0,0	0,2 ± 0,1	0,2 ± 0,0	0,3 ± 0,0
17:1n9	0,3 ± 0,1	0,3 ± 0,1	0,4 ± 0,1	0,3 ± 0,1
18:1n11	1,7 ± 0,5	1,8 ± 0,5	1,9 ± 1,0	2,6 ± 2,0
18:1n9	18,7 ± 3,4	19,5 ± 3,3	19,6 ± 1,6	10,4 ± 1,4
18:1n7	4,2 ± 1,1	4,2 ± 0,9	2,6 ± 0,9	1,5 ± 0,2
18:1n5	0,5 ± 0,1	0,5 ± 0,0	0,4 ± 0,2	0,4 ± 0,1
20:1n11	1,6 ± 0,5	1,8 ± 0,5	2,5 ± 0,6	2,0 ± 0,7
20:1n9	13,4 ± 3,2	14,2 ± 3,6	13,9 ± 2,6	13,2 ± 1,8
20:1n7	0,5 ± 0,2	0,6 ± 0,2	0,3 ± 0,1	0,2 ± 0,1
22:1n11	10,1 ± 3,1	9,3 ± 2,5	12,7 ± 5,5	16,6 ± 1,6
22:1n9	1,2 ± 0,3	1,2 ± 0,3	0,9 ± 0,1	1,0 ± 0,1
22:1n7	0,2 ± 0,1	0,2 ± 0,1	0,2 ± 0,0	0,2 ± 0,0
24:1n9	0,6 ± 0,1	0,7 ± 0,1	0,4 ± 0,1	0,6 ± 0,1
∑MUFA	**61,8 ± 4,1**	**61,4 ± 4,1**	**65,2 ± 4,0**	**54,8 ± 5,5**
16:2nx	0,3 ± 0,1	0,3 ± 0,1	0,3 ± 0,1	0,3 ± 0,1
16:4n1	0,1 ± 0,1	0,1 ± 0,1	0,1 ± 0,0	0,1 ± 0,1
18:2n6	1,5 ± 0,2	1,5 ± 0,2	1,7 ± 0,2	1,4 ± 0,1
20:2n6	0,3 ± 0,0	0,3 ± 0,1	0,3 ± 0,0	0,2 ± 0,0
20:4n6	0,3 ± 0,1	0,3 ± 0,1	0,3 ± 0,1	0,2 ± 0,1
22:5n6	0,1 ± 0,0	0,2 ± 0,1	0,1 ± 0,0	0,2 ± 0,1
18:3n3	0,7 ± 0,2	0,6 ± 0,2	1,1 ± 0,2	1,2 ± 0,7
18:4n3	1,7 ± 0,6	1,4 ± 0,6	1,5 ± 0,1	3,9 ± 0,9
20:3n3	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
20:4n3	0,9 ± 0,2	0,9 ± 0,2	1,0 ± 0,2	1,4 ± 0,4
20:5n3	3,6 ± 1,2	3,1 ± 0,9	3,0 ± 1,1	5,2 ± 1,5
21:5n3	0,3 ± 0,1	0,3 ± 0,1	0,2 ± 0,0	0,3 ± 0,1
22:5n3	2,2 ± 0,5	2,8 ± 0,5	1,9 ± 0,9	1,7 ± 0,5
22:6n3	5,7 ± 1,7	5,3 ± 1,4	4,5 ± 1,8	7,8 ± 2,5
∑PUFA	**17,9 ± 3,5**	**17,3 ± 3,0**	**16,1 ± 3,9**	**24,2 ± 5,8**

	RFI	KM	KI
14:00	7,8 ± 0,3	6,1 ± 0,8	5,8 ± 0,5
i-15:0	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,1
ai-15:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
15:0	0,4 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
i-16:0	0,0 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:0	11,8 ± 0,8	10,8 ± 2,0	10,5 ± 1,8
i-17:0	0,3 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
ai-17:0	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
17:0	0,3 ± 0,1	0,2 ± 0,0	0,2 ± 0,0
18:0	2,0 ± 0,2	2,6 ± 0,6	2,4 ± 0,4
20:0	0,2 ± 0,1	0,2 ± 0,1	0,2 ± 0,1
∑SFA	**23,3 ± 1,0**	**20,9 ± 2,0**	**20,2 ± 1,8**
14:1n5	0,1 ± 0,0	0,1 ± 0,1	0,2 ± 0,1
16:1n11	0,1 ± 0,0	0,1 ± 0,0	0,1 ± 0,0
16:1n9	0,2 ± 0,0	0,2 ± 0,0	0,2 ± 0,0
16:1n7	4,3 ± 0,8	5,5 ± 2,3	6,6 ± 2,6
16:1n5	0,3 ± 0,1	0,3 ± 0,1	0,3 ± 0,1
17:1n9	0,3 ± 0,0	0,2 ± 0,0	0,3 ± 0,0
18:1n11	2,0 ± 1,2	1,4 ± 0,5	1,4 ± 0,3
18:1n9	10,8 ± 1,8	12,5 ± 5,6	13,8 ± 3,7
18:1n7	1,4 ± 0,1	2,4 ± 1,6	2,8 ± 1,4
18:1n5	0,4 ± 0,1	0,5 ± 0,1	0,5 ± 0,1
20:1n11	2,1 ± 0,9	2,1 ± 0,9	2,0 ± 0,4
20:1n9	12,3 ± 3,0	13,9 ± 2,2	14,0 ± 2,3
20:1n7	0,2 ± 0,0	0,3 ± 0,1	0,4 ± 0,1
22:1n11	16,2 ± 1,5	13,1 ± 5,5	13,4 ± 5,6
22:1n9	0,9 ± 0,2	1,0 ± 0,2	1,0 ± 0,2
22:1n7	0,2 ± 0,1	0,2 ± 0,0	0,2 ± 0,0
24:1n9	0,6 ± 0,2	0,6 ± 0,3	0,6 ± 0,3
∑MUFA	**52,4 ± 6,3**	**54,5 ± 7,4**	**57,8 ± 3,0**
16:2nx	0,3 ± 0,0	0,4 ± 0,1	0,4 ± 0,1
16:4n1	0,2 ± 0,1	0,4 ± 0,2	0,3 ± 0,1
18:2n6	1,3 ± 0,1	1,8 ± 0,2	1,8 ± 0,1
20:2n6	0,2 ± 0,0	0,3 ± 0,0	0,3 ± 0,0
20:4n6	0,2 ± 0,0	0,3 ± 0,1	0,3 ± 0,1
22:5n6	0,2 ± 0,1	0,2 ± 0,1	0,2 ± 0,0
18:3n3	1,6 ± 0,3	1,6 ± 0,7	1,3 ± 0,4
18:4n3	4,1 ± 1,1	3,6 ± 2,1	2,7 ± 1,1
20:3n3	0,1 ± 0,0	0,2 ± 0,1	0,2 ± 0,1
20:4n3	1,3 ± 0,3	1,3 ± 0,3	1,2 ± 0,1
20:5n3	5,1 ± 1,3	4,3 ± 1,7	3,9 ± 0,7
21:5n3	0,3 ± 0,1	0,4 ± 0,1	0,4 ± 0,1
22:5n3	1,5 ± 0,1	2,3 ± 0,4	2,3 ± 0,4
22:6n3	7,9 ± 2,5	7,5 ± 2,4	6,7 ± 1,1
∑PUFA	**24,3 ± 5,5**	**24,6 ± 7,3**	**22,0 ± 3,0**