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Maternal and infant body composition in relation to fish and meat intake during pregnancy

- A randomized longitudinal dietary intervention study

Degree Project in Medicine

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Table of content

1. List of abbreviations

ADP; air displacement plethysmography

BMI; body mass index

FFQ; food frequency questionnaire

FFM; fat-free mass

FM; fat mass

FM %; fat mass percent

GDM; gestational diabetes mellitus

GWG; gestational weight gain

IOM; Institute Of Medicine

LGA; large for gestational age

NNR; Nordic Nutrition Recommendations

NW; normal weight

OB; obese

PONCH- study; Pregnancy Obesity Nutrition and Child Health study

2. Abstract

Degree project, Programme in Medicine, *Maternal and infant body composition in relation to fish and meat intake during pregnancy*, Agnes Dickèr, 2019, Institution of Neuroscience and Physiology, Gothenburg, Sweden

Introduction: Many women start their pregnancy at a high BMI and gain more weight than recommended during pregnancy, thus increasing the risk for severe pregnancy complications and children born large for gestational age. Polyunsaturated omega-3 fatty acids found in fish might have an effect on reducing obesity. Few studies have investigated the relation between fish intake and maternal and infant body composition.

Aims: To determine whether the PONCH (Pregnancy Obesity Nutrition and Child Health) dietary intervention study had an impact on maternal dietary intake and to investigate body composition in mother and infant in relation to energy, meat and fish intake during pregnancy.

Method: Normal weight (NW) and obese (OB) pregnant women were randomized to either a control or intervention group, the latter receiving dietary guidance. Study visits took place in each trimester with measurements of body composition by air displacement plethysmography (ADP) and collection of food frequency questionnaires. Infant anthropometry and body composition were measured (by ADP) at 1 and 12 weeks after birth.

Results: NW and OB intervention groups reported an increased intake of high-fat fish between first and second trimester (increase of 118 g/week for NW, $p<0.001$, and 150 g/week for OB, $p=0.015$). OB intervention had a lower frequency of excessive gestational weight gain compared with OB control (23% versus 61%, $p = 0.046$). OB women with gestational weigh gain under or within recommendations reported higher intake of high-fat fish (198 g/week) than OB women with weight gain above recommendations (131 g/week) ($p=0.049$). Negative correlations were found between maternal high-fat fish intake and infant weight, length and fat free mass.

Conclusion: This study shows that dietary guidance can help women increase their fish intake during pregnancy. Obese women with high fish intake during pregnancy more often gain weight within IOM recommendations. Moreover, intake of high fat fish during gestation is correlated with smaller children at birth.

Key words: Fish intake, pregnancy, infant, body composition

3. Introduction

Obesity is rapidly increasing worldwide with prevalence not only expanding in the adult population but also affecting children (1, 2). According to the WHO, the global prevalence of obesity has nearly tripled during the last 40 years (3). Body mass index (BMI) presents an index frequently used for classifying weight status and comparing the prevalence of obesity, both within and between populations (1). Obesity is defined as BMI above 30 kg/m^2 and is a major cause of impaired health. The abnormal accumulation of fat causes increased risk for numerous serious diseases, such as cardiovascular disease, diabetes, osteoarthritis and other musculoskeletal disorders (1, 3, 4). Furthermore, there is growing evidence that obesity, through secretion of bioactive substances and activation of inflammatory signalling creates a pro-tumorigenic environment and plays a central role in carcinogenesis (5). Facing this increasing social, health care and economic burden, there is a general consensus that prevention and treatment strategies should be targeted not only toward adults, but also youths (6).

Human gestation features numerous physiological changes, including processes leading to weight gain. Due to accelerated maternal and fetal tissue synthesis and adjustments in basal metabolism there is a need of increased energy intake (7, 8). Many women, however, gain more weight than is generally recommended by the medical profession, resulting in an increased maternal fat accumulation which can impact the long-term metabolic health of both mother and child (8, 9). Moreover, due to the growing obesity epidemic, more women of childbearing age start their pregnancy with a high BMI (10). In 2017, 26% of the women were classified as overweight and 15% as obese when registered at Swedish antenatal health clinics. The Swedish national board of Health and Welfare witness a distinct rise in the average maternal BMI since 1992 when documentation of this data from medical records was first initiated (11, 12). In 2009, the American Institute of Medicine (IOM) presented new recommendations on weight gain in pregnant women according to corresponding BMI category before conception. In contrast to previous guidelines, this version presents a specific and rather narrow range regarding gestational weight gain (GWG) for obese women. The recommended range of total weight gain during gestation is; 11.5-16 kg for normal weight women and 5-9 kg for obese women (13). Although obese pregnant women generally show a smaller GWG, the majority still gain more than the recommended weight for their BMI class (14, 15).

An excessive GWG or maternal obesity is known to increase the risk for pregnancy complications such as gestational diabetes (GDM), pre-eclampsia, pregnancy induced hypertension, preterm delivery and the need for emergency caesarean section (16-18). Long term effects include post-partum weight retention and overweight in mother (15). In addition, fetal consequences include large for gestational age (LGA; birth weight above the 90th percentile) and therefore increased risk of shoulder dystocia (19), perineal lacerations grade III-IV (12), fetal macrosomia (birth-weight >4500 g), congenital birth defects and longer hospital stay (17, 18, 20). While these findings emphasize immediate implications during pregnancy and delivery, several studies have also demonstrated consequences of maternal obesity and GDM when examining long-term effects in children. Infants exposed to intrauterine metabolic alterations associated with maternal obesity or GDM, and/or have high birth weight appear to have an increased risk of developing metabolic syndrome and obesity in childhood (21-26).

According to the Swedish National Food Agency, a prescribed selection of foods that follows the general dietary recommendations in the Nordic Nutrition Recommendations, NNR, provides satisfactory conditions for good health, both in the pregnant mother and fetus (27). Numerous nutrients are of great importance for fetal development, such as folate, proteins and essential fatty acids (8, 27). Meat is a source of protein but also micronutrients such as iron

(27) of which requirements gradually rise as pregnancy proceeds (28). Numerous studies on obese individuals have presented results indicating that high protein diets promote weight loss, weight maintenance and beneficial changes in body composition (29, 30) in terms of a reduction in fat mass whilst preserving fat free mass (31-33). Previous studies of maternal macronutrient intake in relation to infant body composition suggest that protein intake is positively correlated with infant birth weight (34, 35) but the effect on FM and FFM in infant is yet relatively unexplored.

Polyunsaturated omega-3 fatty acids such as eicosapentaenoic acid (n-3 EPA) and docosahexaenoic acid (n-3 DHA) can be found in different types of fish, especially high-fat fish e.g. salmon, herring or mackerel. Pregnant women are recommended 2-3 servings of fish per week during gestation in order to reach a daily intake of 0,1-0,3 g DHA (36). This nutrient has a significant role in fetal neurodevelopment (37) but previous studies also indicates positive effects of omega-3 intake on reducing obesity (38, 39). Considerable evidence from previous rodent studies suggest that supplementation of long-chained omega-3 polyunsaturated fatty acids (n-3 PUFA) might have beneficial impact on body composition, attenuating weight gain and reducing body fat accumulation. Although studies on the effect of supplementation in humans is more scarce, a dietary interventional study on obese women from 2015 showed that when combined with calorie restriction, additional supplementation of n-3 PUFA appeared to enhance the effect of weight loss and reduce the severity of metabolic syndrome (40). Observational studies on obese adults and children have shown that low plasma n-3 PUFA levels is negatively correlated with BMI and unfavourable body measurements (41-43). Moreover, a cohort study conducted on mother-child pairs showed that a higher concentration of n-3 PUFA in the umbilical cord plasma was associated with lower adiposity in offspring. In the same study, higher maternal fish intake was negatively correlated with obesity in children at three years of age (44).

It is suggested that prenatal programming is influenced by epigenetic regulation in utero. These modifications that alter gene expression result in a varied function in fetal tissues and organs, thus changing basic conditions for further development and long-term metabolic health (45, 46). Dietary fatty acids (FA) have been identified as nutrients associated with epigenetic modifications in early metabolic programming in animal models, regulating feeding behaviour, altering liver tissue function and adipose cell proliferation (47, 48). Although underlying mechanisms are not fully resolved, this emerging evidence indicates that maternal health status is a major determinant of offspring and childhood health, and that focus on prevention and lifestyle implementations such as dietary guidance should play a substantial part in maternal health services.

The Pregnancy Obesity Nutrition and Child Health study (PONCH) is an ongoing randomized intervention study investigating the metabolic health in normal weight and obese pregnant women and their offspring. By dietary intervention the aim is to optimize pregnancy outcome in terms of weight gain and complications related to maternal obesity and GDM. Maternal and infant body composition is measured by air displacement plethysmography (49) in order to assess the changes that take place during pregnancy and to see if this may have an impact on both mother and offspring's metabolic health. This study succeeds previous work presented by the PONCH study. In a former publication, Bosaeus et al described that serum EPA and DHA as a biomarker for fatty acid intake were positively correlated with fish intake during early pregnancy in normal weight women (50). However, there is a lack of randomised control trials investigating whether fish and meat intake influence maternal and infant body composition in terms of fat mass (FM), fat mass percent (FM%) and fat free mass (FFM).

4. Aim

The aim of this study was to determine whether the PONCH dietary intervention had an impact on maternal dietary intake in normal weight and obese women, as well as to investigate body composition and anthropometry in mother and infant in relation to energy, meat and fish intake during pregnancy.

4.1 Specific objectives

Firstly, did the women, who were randomized to the intervention group, modify their intake of energy, meat and fish? Also, how did the amount of fish and meat consumed during pregnancy correlate with maternal body composition? Finally, how did the amount of fish and meat consumed by the mother during pregnancy correlate with infant body composition?

5. Methods

5.1 Study population

During a period from April 2009 to August 2018, a total of 132 normal weight (NW: BMI 18,5-24,9 kg/m^2) and 45 obese (OB: BMI > 30 kg/m^2) pregnant women were recruited for the Pregnancy Obesity and Child Health study (PONCH), an ongoing prospective randomized dietary intervention study taking place at Sahlgrenska University Hospital, Gothenburg, Sweden. Information about the study was handed out at six various maternity care centres within the Gothenburg area and through advertisement on websites for pregnant women and public billboards. All participants lived in the region of Västra Götaland at the time of recruitment. Inclusion criteria in this study was age 20-45 years. Women with self-reported diabetes, giving birth to twins, with reported vegetarianism or veganism, use of tobacco or neuroleptic drugs were excluded.

At enrollment, the women were categorized as OB or NW after which they were randomized to either a control group or an intervention group based on age, parity and BMI. Randomization was carried out by a computer-controlled program. As presented in figure 1, the women were henceforth followed throughout pregnancy. Both groups attended regular study visits that took place in trimester one (visit between gestation week 8-12), trimester two (visit between gestation week 24-26) and trimester three (visit between gestation week 35-37). At the last visit before estimated time of birth, the participating women were asked if they would admit their child into the PONCH-study. Met with approval, these children were divided into groups with reference to corresponding group (NW or OB) of the mother (Figure 2).

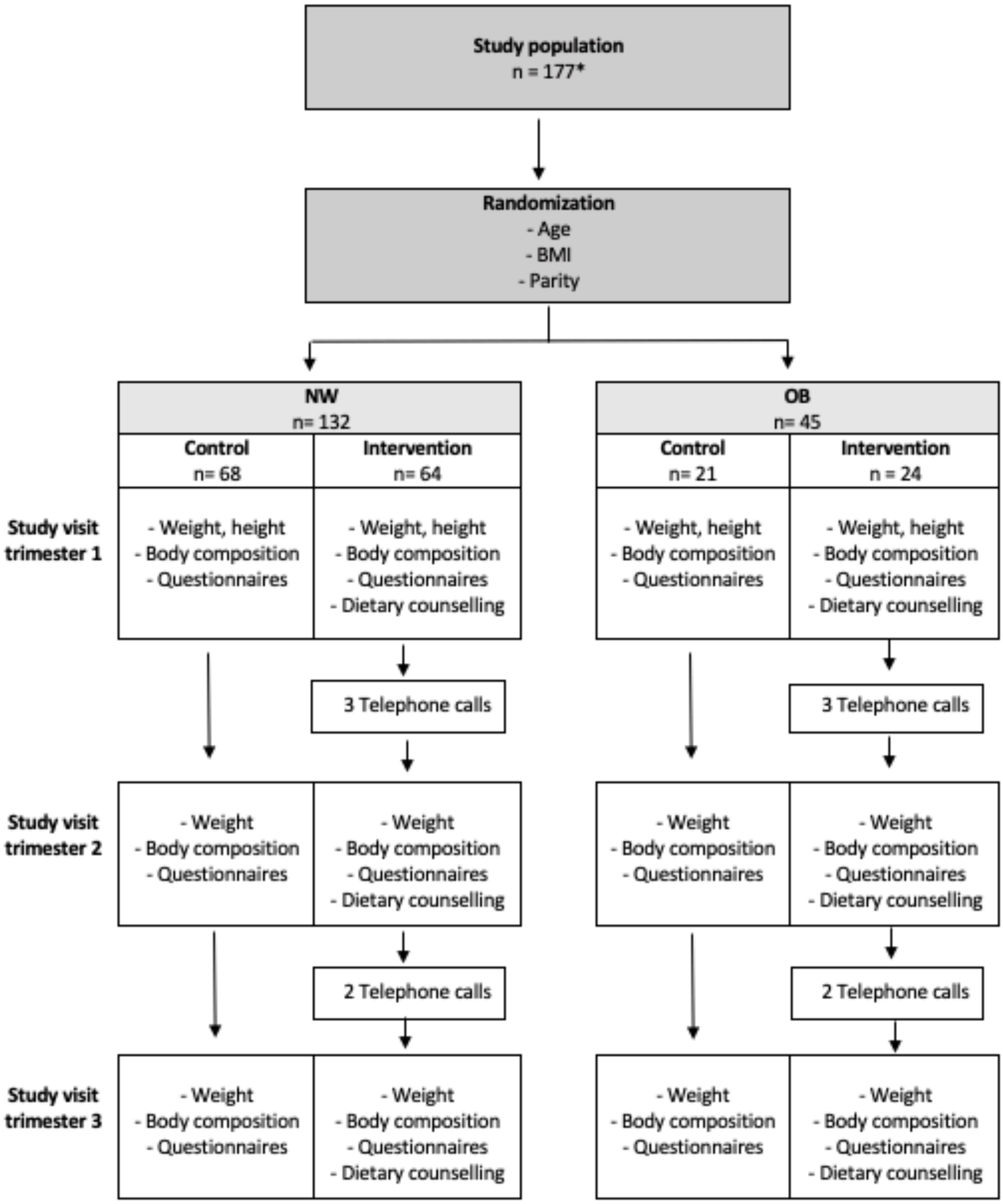

Figure 1: Flow chart of maternal study protocol

Flow chart visualizing recruitment and number of normal weight (NW) and obese (OB) women randomized to either a control or intervention-group and henceforth followed through pregnancy with regular study visits.

* Material for this study consist of data from women in the PONCH - study who participated in all three trimesters. n = number of participants.

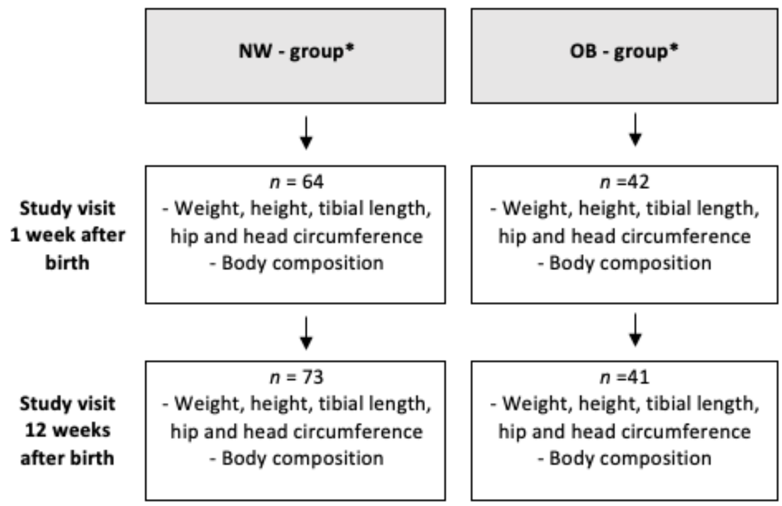

Figure 2: Infant study protocol

* Infants were divided into groups with reference to corresponding group of the mother, NW = normal weight or OB=obese, whom at the point of trimester three agreed to enroll their child.

n = number of subjects attending study visits.

5.2 Maternal study visits

Maternal study visits took place in the morning after overnight fasting and included anthropometric measurements of weight and body composition, completion of food frequency questionnaires and blood sampling. At the first visit, the height of each participant was measured to the nearest 0.5 cm, and BMI was calculated.

Body composition was determined using the BOD POD (software version 5.4.0; Cosmed, Rome, Italy) which is a validated method (51-53). With the subject dressed in underwear and bathing cap, measurements of body volume were performed twice. In case of inconsistency between the values, the software asked for a third measurement. Based on air-displacement plethysmography, ADP, the software calculates body density (kg/L) (49). Due to increased hydration of FFM during gestation, adjusted values of FFM and FM were calculated according to previous studies (54, 55) as follows:

In trimester 1, FM and FFM were calculated using the equation (FM_{ADPvR1}) published by van Raaij et al.(54) where BW_1 is body weight (kg) and Db_1 is body density (kg/L) in first trimester:

$$FM_{ADPvR1} \text{ (kg)} = BW_1/100 * (496.4/Db_1 - 451.6)$$

$$FFM_{ADPvR1} \text{ (kg)} = BW_1 - FM_{ADPvR1.}$$

Accordingly, FM and FFM in trimester 2 were calculated using the equation (FM_{ADPvR2}) developed by the estimation of FFM density in mid pregnancy set at 1.095 kg/L published by van Raaij et al. (54). BW_2 is body weight (kg) and Db_2 is body density (kg/L) in second trimester:

$$FM_{ADPvR2} \text{ (kg)} = BW_2 * (5.0538/Db_2 - 4.6154)$$

$$FFM_{ADPvR2} \text{ (kg)} = BW_2 - FM_{ADPvR2}$$

FM and FFM in trimester 3 were acquired using the equation (FM_{ADPvR3}) presented by Hopkinson et al.(55), who estimated FFM density in late pregnancy to 1.089 kg/L. BW_3 is body weight (kg) and Db_3 is body density (kg/L) in trimester 3:

$$FM_{ADPvR3} \text{ (kg)} = BW_3 * (5.19/Db_3 - 4.76)$$

$$FFM_{ADPvR3} \text{ (kg)} = BW_3 - FM_{ADPvR3}$$

A validated self-administered food frequency questionnaire (FFQ) comprising the participant's dietary habits 3 months prior to the study visit was used to calculate energy intake (56). In addition, questionnaires regarding fish and meat intake were also collected at each study visit, identifying frequency and type of fish or meat respectively (Appendix 1). This questionnaire, developed at the Institute of Neuroscience and Physiology at Sahlgrenska

Academy, was evaluated in 2015 where a study performed by Bosaeus et al observed that the concentration of PUFA in serum positively correlated with reported fish intake (50). Following the Norwegian Health Authorities, it is assumed that one serving of fish is equivalent to 150 g and one serving of meat is equivalent to 175 g (36). Hereby the frequency of intake could be converted into grams. Subjects were also questioned whether they used any dietary supplementation containing fish oil or ω-3 fatty acids.

Women allocated to the intervention group received additional dietary counselling by an authorized dietician, emphasizing the recommendations for pregnant women established by the Swedish National Food Agency and presented in the NNR 2004 (57). In detail, the subjects of the NW intervention group were given directions to A) consume at least three servings of fish per week (to avoid pollutants, appropriate types of fish to consume were presented) B) reduce intake of sugar, preferably reaching a level below 10 % of daily energy intake, C) consume at least 500 g of fruits and vegetables daily and D) increase daily energy intake according to trimester (350 kcal in second trimester and 500 kcal in third trimester). Furthermore, advise on suitable snacks, food frequency and fibre intake were given if needed. For OB women, the participants in the intervention group were given identical directions as NW women, with the exception of energy intake. The OB women were handed a dietary plan with a 20% energy restriction. This was derived from estimated basal metabolic rate, calculated with Harris Benedict's equation (58).

Regular telephone calls in between study visits aimed to increase adherence to the recommendations received. Between first and second visit, the subjects were contacted three times, and between second and third visit they were contacted twice.

5.3 Infant anthropometric measurements

For infant outcomes, all women that enrolled their child into the study and attended a trimester 3 visit were included. Infants were divided into groups with reference to corresponding group of the mother, NW = normal weight or OB=obese (n= 73 for NW, n = 42 for OB, figure 2). Infant weight and length measurements at time of birth were collected from medical records. Further measurements in terms of anthropometry and body composition were made at 1 week (4-10 days) and 12 weeks (80-90 days) after birth (figure 2). Lower leg length, head circumference, length, waist and hip circumference to the nearest 0.5 centimetre was measured at both visits. Weight, percentage of body fat, FM and FFM was obtained by the use of PEA POD (software version 3.3.0; COSMED, Italy), which in accordance with the BOD POD system generates values using ADP validated for measuring body composition of infants (from birth up to 6 months) (59-61).

5.4 Data collection

With the exception of some FFQs as well as surveys regarding fish and meat intake, data prior to the current study was already collected and accessible. A number of mothers and new-borns in OB group were recruited later than NW and additional study visits took place before completion of data base. Further data specific for the FFQ were entered into a computer-accessed program for calculating nutritional values whereupon it was transferred into excel and the statistical software SPSS, enabling data analysis.

Considering the short amount of time between delivery and the first infant visit (1-week post-partum), some visits scheduled for infants were missed owing to incomplete maternal recovery. Moreover, with a number of participants preferring to complete questionnaires at home, some food-frequency surveys were unfortunately left uncompleted. Numerous FFQ's were reported to have been sent to the department although some were not received.

5.5 Statistical methods

For statistical analyses, SPSS version 25 (IBM SPSS Statistics, Armonk, NY: IBM Corp) was used. Continuous variables are presented by their mean and standard deviation (Mean (SD)) unless otherwise stated. In addition to visual analysis of histograms, Shapiro-Wilk test was used to test for normality. To test for differences between groups, Student's independent t-test was applied for normally distributed data and Mann-Whitney U test for non-normally distributed data. Pearson's chi-squared test was used to examine relationships between categorical values, e.g education or parity when analysing background characteristics. To assert changes within a group, Wilcoxon signed-rank test was performed.

Correlations between dietary intake and maternal and infant body composition were tested by Spearman's rank correlation coefficient (for non-normally distributed data) and Pearson correlation coefficient (for normally distributed data). Using multiple linear regression, the maternal data were adjusted for pregestational BMI, age and education. Outcomes regarding infant body composition were adjusted for pregestational BMI, maternal age and infant sex. Initially, pairwise relations between dependent and predictor variables were examined with linear regression. Significance below 0.25 was adjusted for. Predictor variables were controlled for collinearity using Pearson's correlation coefficient, where no correlations above 0.3 were found which enabled further calculations in a multiple linear regression model.

GWG was determined by calculating the difference in weight between trimester one and trimester three. GWG were then in accordance with IOM guidelines categorized as "under", "within" or "above" recommendations, between which different characteristics could be examined. Kruskal Wallis-test was used when comparing mean values between all three subgroups, and Mann-Whitney U was used when comparing means between two subgroups.

Unless noted otherwise, the analyses were performed on data gathered from women participating in all study visits and follow up between trimester one and trimester three, n=177 (figure 1). Considering the relatively low number of infants enrolled in the study, women registered later than trimester one or women with missed visits during the study were also included together with participants' child in the analyses of correlations. All tests performed were two-tailed and throughout analysis, significance was defined as $P < 0,05$.

6. Ethics

This project was covered by the ethical approval of the PONCH study (Dnr 402-08).
All women received oral and written information regarding the study. Written consent from all participants was collected prior to enrolment. Each participant was anonymized by the use of codes.

7. Results

7.1 Maternal background characteristics

Table 1 displays maternal background characteristics. Reflecting the difference in OB and NW group classification, pregestational BMI was significantly higher in OB group than NW. A higher education level was found in NW compared to OB. Higher education was associated with lower pre-gestational BMI for all women combined ($r = -0.226$. $p < 0.001$). For age, height and parity, there were no significant differences between OB and NW groups. Between the subcategorized intervention and control groups, no significant differences were found for background variables (table 1).

Table 1: Maternal background characteristics

	NW						OB						
	Intervention		Control			Total	Intervention		Control			Total	P [b]
	n	Mean (SD)	*n*	Mean (SD)	P [a]	Mean (SD)	*n*	Mean (SD)	*n*	Mean (SD)	P [a]	Mean (SD)	
Age (years)*	64	30.6 (3.6)	68	31.2 (3.6)	0.344	30.8 (3.8)	24	31.5 (3.2)	21	30 (4.2)	0.174	30.8 (3.8)	0.916
Pregestational weight (kg)	64	62.2 (6.4)	67	61.4 (6.4)	0.489	61.8 (6.5)	23	100.9 (18.0)	21	99.0 (11.9)	0.691	100.0 (15.2)	**<0.001**
Height (m)	64	1.68 (0.06)	68	1.68 (0.06)	0.505	1.68 (0.1)	24	1.70 (0.07)	21	1.68 (0.05)	0.542	1.69 (0.07)	0.423
Pregestational BMI	64	21.9 (1.5)	67	21.8 (1.6)	0.772	21.8 (1.5)	23	35.0 (4.5)	21	34.9 (4.3)	0.796 [c]	35.0 (4.4)	**<0.001** [c]
Parity 0/1/2/3 % [d]	64	50/36/14/0	68	60/35/4/0	0.134	55/36/9/0	23	50/42/4/4	21	48/43/10/0	0.712	49/42/7/2	0.280
Education level: 3 years of high school / < 3 years of university / > 3 years of university (%) [d]	62	11/11/77	68	10/19/71	0.466	11/15/74	23	30/22/48	21	38/14/48	0.769	34/18/48	**0.001**
Weight 1st trimester (kg)	64	62.9 (6.2)	68	62.3 (6.4)	0.552	62.6 (6.3)	22	100.9 (16.6)	18	97.7 (11.9)	0.86 [c]	99.5 (14.6)	**<0.001** [c]
Fat mass 1st trimester (kg)	63	16.8 (3.4)	68	16.7 (4.9)	0.791	16.8 (4.1)	22	48.0 (11.3)	18	45.2 (10.3)	0.543 [c]	46.7 (10.8)	**<0.001** [c]
Fat mass percent 1st trimester (%)	63	26.7 (4.4)	68	26.5 (6.3)	0.497	26.6 (5.4)	22	47.2 (4.8)	18	45.9 (5.5)	0.427	46.6 (5.1)	**<0.001**
Fat free mass 1st trimester (kg)	63	46.1 (5.0)	68	45.5 (4.6)	0.821	45.8 (4.8)	22	53.0 (7.6)	18	52.5 (4.7)	0.645 [c]	52.7 (6.4)	**<0.001** [c]

NW = normal weight, OB=obese.
* At time of trimester one.
[a] P-values calculated with Student's independent sample t-test between intervention and control
[b] P-values calculated between NW and OB group with Student's independent sample t-test.
[c] P-values were calculated by Mann–Whitney U test.
[d] Categorical values were analyzed by Chi squared-test.

7.2 Reported dietary intake

7.2.1 NW group

As shown in table 2, there were no significant differences between NW intervention and control group in reported fish, meat or energy intake in absolute amount in the first and consecutive trimesters. However, the NW intervention group significantly increased their intake of high fat fish and total fish intake between trimester 1-2 and 1-3, which was not seen in the NW control group (table 3 and visualized in figure 3). These changes in fish intake for the NW intervention group were also significantly higher when compared with the NW control group (table 3). NW control group reported a significantly higher use of n-3 PUFA supplementation in trimester 3 than NW intervention (table 2). NW control reported a significantly increased meat intake between trimester 1-3 and this was also significantly higher when compared with NW intervention (table 3).

7.2.2. OB group

No significant differences between intervention and control group were found when examining reported fish, meat or energy intake in the first and consecutive trimesters (table 2). The OB intervention group reported an increased intake of total fish between trimester 1 and trimester 2. OB control reported an increased intake of high-fat fish between trimester 2-3, significant when analysed within the group (table 3). When comparing the changes between control and intervention, they showed no significant differences (table 3). OB intervention group reported no significant reduction in energy intake between trimester 1 and trimester 3. The self-reported (and not significant) decrease of 159.1 kcal/day would account for a 7% energy restriction with reference to energy intake at baseline in trimester 1.

7.2.3. NW vs OB

Whereas reported intake of total amount of fish and high fat fish in trimester 1 was somewhat higher in NW, OB reported a higher meat intake. However, in both cases these differences were not significant (table 2). Reported total fish intake in trimester 3 was significantly higher in NW compared to OB. During the same period OB reported significantly lower energy intake (table 2). Remaining data showed no significant differences (table 2).

Table 2: Reported dietary fish, meat and energy intake

	NW						OB						
Trimester	**Intervention**		**Control**			**Total**	**Intervention**		**Control**			**Total**	**P [c]**
	n	Mean (SD)	*n*	Mean (SD)	P [a]	Mean (SD)	*n*	Mean (SD)	*n*	Mean (SD)	P [a]	Mean (SD)	
1st													
High-fat fish (g/week)	59	147.5 (133.9)	63	168.5 (136.4)	0.304	158.3 (135.0)	18	156.3 (87.5)	15	117.5 (105.0)	0.094	138.6 (96.3)	0.846
Total fish (g/week)	62	359.3 (217.3)	66	431.3 (273.0)	0.082	396.4 (249.3)	22	327.3 (201.2)	17	352.9 (339.3)	0.873	338.5 (266.5)	0.114
Meat (g/week)	62	1120.6 (574.1)	66	962.5 (529.5)	0.108 [b]	1039.1 (555.0)	22	1141.5 (589.4)	17	1286.8 (627.8)	0.463 [b]	1204.8 (602.7)	0.115
Energy (kcal/day)	60	2144.0 (556.6)	66	2329.5 (590.1)	0.076	2241.2 (579.6)	21	2269.0 (539.8)	17	2125.9 (794.1)	0.191	2205.0 (664.3)	0.655
2nd													
High-fat fish (g/week)	62	189.3 (164.0)	66	149.4 (111.5)	0.146	168.8 (140.3)	23	164.7 (99.7)	18	108.3 (80.2)	0.056	139.6 (96.3)	0.367
Total fish (g/week)	63	475.0 (243.6)	68	447.8 (248.9)	0.487	460.9 (245.8)	21	450.0 (231.2)	19	355.3 (270.2)	0.240 [b]	405.0 (251.8)	0.260
Meat (g/week)	63	1090.3 (519.0)	66	1092.4 (609.4)	0.791	1091.4 (564.9)	21	1083.3 (585.3)	19	1211.2 (640.5)	0.513 [b]	1144.1 (607.6)	0.707
Energy (kcal/day)	63	2238.3 (504.3)	67	2338.2 (644.2)	0.498	2289.8 (580.6)	21	2126.0 (596.6)	18	2178.2 (738.3)	0.808 [b]	2150.1 (657.2)	0.162
3rd													
High-fat fish (g/week)	63	207.7 (155.5)	65	169.6 (140.8)	0.051	188.4 (148.8)	20	178.1 (98.0)	18	131.3 (104.7)	0.096	155.9 (102.6)	0.364
Total fish (g/week)	62	477.8 (187.3)	67	429.4 (266.5)	0.142	452.7 (232.2)	22	395.5 (230.5)	20	337.5 (239.0)	0.429 [b]	367.9 (233.6)	**0.037**
Meat (g/week)	61	1095.9 (473.8)	67	1164.1 (605.1)	0.482 [b]	1131.6 (545.4)	22	1101.7 (480.5)	20	1321.3 (583.8)	0.189 [b]	1206.3 (537.1)	0.398
Energy (kcal/day)	62	2275.8 (595.4)	67	2376.4 (577.4)	0.272	2328.0 (586.0)	22	2096.8 (492.8)	20	2175.6 (601.4)	0.644 [b]	2134.3 (541.9)	**0.042**
Reported use of n-3 PUFA supplementation (%) [d]	64	7.8	68	22.1	**0.023**	15.2	23	26.1	21	9.5	0.155	18.2	0.634

NW = normal weight, OB=obese.
[a] P-values between control and intervention group were calculated with Mann–Whitney U test.
[b] P-values between control and intervention group were calculated with Student's independent sample t-test.
[c] P-values between NW and OB group were calculated with Mann–Whitney U test.

Table 3: Dietary changes of fish, meat and energy intake calculated between follow-up visits

	NW							OB						
Trimester	**Intervention**			**Control**			**P [b]**	**Intervention**			**Control**			**P [b]**
	n	Mean (SD)	P [a]	*n*	Mean (SD)	P [a]		*n*	Mean (SD)	P [a]	*n*	Mean (SD)	P [a]	
Trimester 1 - 2														
High-fat fish (g/week)	59	45.8 (146.4)	**0.022**	63	- 14.9 (108.5)	0.463	**0.006**	18	39.6 (104.9)	0.075	15	- 7.5 (63.7)	0.268	0.128
Total fish (g/week)	61	118.0 (178.3)	**<0.001**	66	23.3 (189.0)	0.215	**0.004**	20	150 (250.5)	**0.015**	15	- 20.0 (277.0)	0.73	0.092
Meat (g/week)	61	- 49.1 (394.3)	0.463	64	119.2 (478.5)	0.051	0.082	20	- 65.6 (684.8)	0.868	15	- 140.0 (754.7)	0.474	0.763 [c]
Energy (kcal/day)	59	112.4 (446.4)	0.219	65	44.7 (608.9)	0.626	0.577	20	-155.2 (475.8)	0.247	15	48.0 (910.7)	0.46	0.398 [c]
Trimester 1 - 3														
High-fat fish (g/week)	59	57.8 (143.6)	**0.003**	63	3.6 (133.8)	0.670	**0.031**	18	33.3 (131.1)	0.298	15	15 (44.4)	0.197	0.621
Total fish (g/week)	60	111.3 (159.5)	**<0.001**	65	1.3 (232.4)	0.896	**<0.001**	21	71.4 (206.0)	0.132	16	- 46.9 (326.2)	0.723	0.528
Meat (g/week)	59	- 47.3 (454.8)	0.792	65	200.3(428.8)	**0.001**	**0.010**	21	- 4.7 (525.5)	0.736	16	43.8 (736.3)	0.783	0.914
Energy (kcal/day)	58	128.2 (433.6)	**0.037**	65	69.0 (634.4)	0.251	0.746	20	- 159.1 (581.9)	0.370	16	- 2.9 (970.7)	0.642	0.553 [c]
Trimester 2 - 3														
High-fat fish (g/week)	62	16.9 (157.2)	0.382	65	18.5 (97.9)	0.241	0.661	20	- 5.6 (75.3)	0.406	18	22.9 (48.4)	**0.025**	0.142
Total fish (g/week)	61	0.0 (172.1)	0.660	67	- 20.6 (187.2)	0.478	0.437	21	- 57.1 (178.9)	0.184	19	- 19.7 (141.1)	0.609	0.446
Meat (g/week)	60	- 15.8 (394.1)	0.831	65	58.4 (376.8)	0.078	0.232	21	20.8 (513.1)	0.255	19	138.2 (850.0)	0.346	0.890
Energy (kcal/day)	61	34.2 (524.0)	0.534	66	28.4 (534.7)	0.648	0.891	21	- 11.8 (565.5)	0.614	18	- 43.3 (802.3)	0.744	0.612

NW = normal weight, OB=obese.
[a] P-values for paired means within intervention or control group calculated with Wilcoxon signed-rank test.
[b] P-values between control and intervention group were calculated with Mann–Whitney U test.
[c] P-values between control and intervention group were calculated with Student's independent sample t-test.

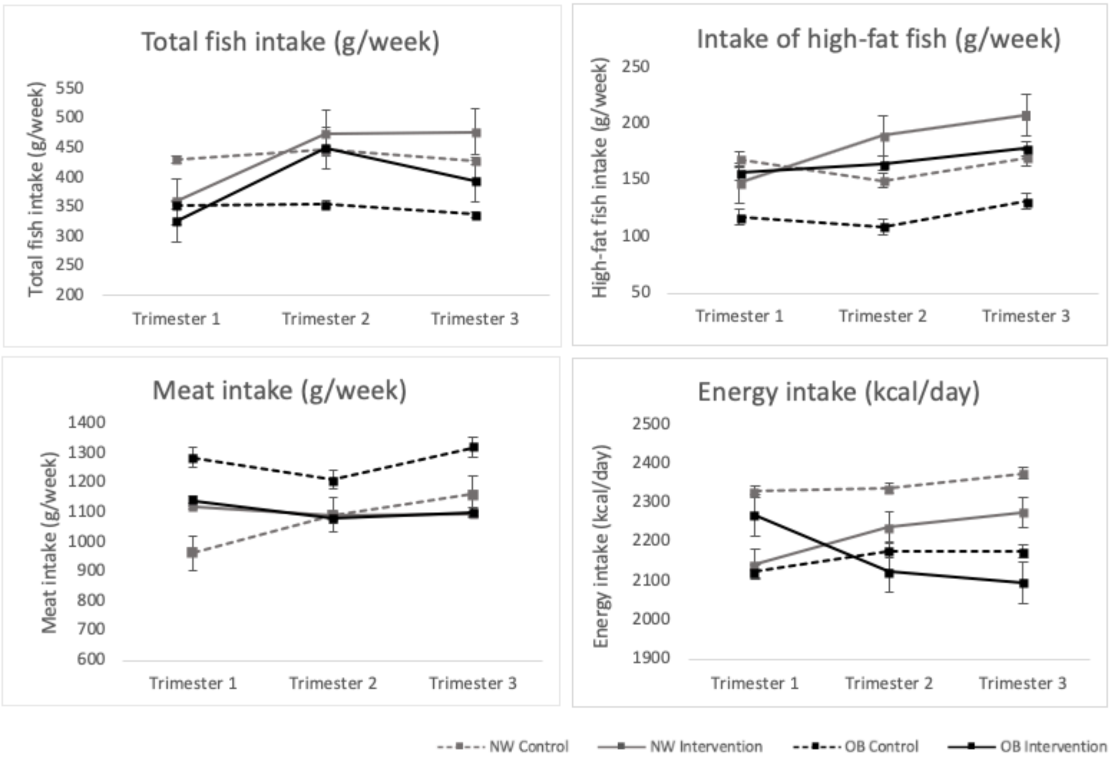

Figure 3: Reported fish, high-fat fish, meat and energy intake at study visit trimester one, two and three.
Points show mean (± SE). For p-values regarding dietary intake at follow-up, see table 2.
For p-values regarding change in dietary intake within and between each group, se table 3.

7.3 Maternal body composition

As shown in table 4, all parameters of maternal body composition differed significantly between NW and OB in all trimesters. No differences were found between subcategorized intervention and control groups.

Mean GWG and changes in FM and FM% were greater for NW than OB, the latter reducing mean fat mass with 2.1% (table 4). Though not significant, there was a tendency to a smaller GWG, FFM and FM-gain in the OB intervention group compared with OB control group (table 4).

Table 4: Body composition changes between trimester 1 and trimester 3

	NW							OB							
	Intervention		Control		P [b]	Total		Intervention		Control		P [b]	Total		P [a]
	n	Mean (SD)	*n*	Mean (SD)	0.478	Mean (SD)		*n*	Mean (SD)	*n*	Mean (SD)	0.15	Mean (SD)		
Trimester 1 – 3															
Gestational weight gain (kg)	64	11.5 (3.4)	68	11.5 (2.8)	0.521	11.5 (3.1)		22	6.9 (3.5)	18	8.9 (5.3)	0.157	7.8 (4.4)		**< 0.001**
Change in fat free mass (kg)	61	6.9 (2.5)	67	7.0 (2.0)	0.899	6.9 (2.3)		22	5.9 (2.1)	18	7.0 (3.1)	0.514	6.4 (2.6)		0.283
Change in fat mass (kg)	61	4.7 (2.8)	67	4.5 (2.9)	0.933	4.6 (2.8)		22	1.0 (3.3)	18	1.9 (4.4)	0.744	1.4 (3.8)		**< 0.001**
Change in fat mass percent (%)	61	2.1 (3.1)	67	1.9 (3.2)	0.478	2.0 (3.2)		22	- 2.1 (2.3)	18	-2.1 (2.5)	0.15	-2.1 (2.4)		**< 0.001**
GWG according to IOM recommendations Under/Within/Above (%) [c]	66	61/30/9	70	47/46/7	0.181	54/38/8		22	27/50/23	18	17/22/61	**0.046**	23/37/40		

NW = normal weight, OB=obese.
[a] P-values calculated with Mann-Whitney U test, analyzed between NW and OB group.
[b] P-values between control and intervention group calculated by Mann–Whitney U test.
[c] Categorical values were analyzed by Chi squared-test.

7.4 PONCH results compared with IOM recommendations

When examining GWG according to the guidelines stated by the IOM, NW control and intervention groups exceeded recommended GWG to an extent of 7 % and 9 %, respectively, but with no significant difference between control and intervention. For the OB groups there was however a significant difference between intervention and control. Of participants in OB control group, 61% exceeded recommendations (GWG > 9 kg), whereas for OB intervention group, the corresponding proportion was 23% (p=0.046) (table 4 and figure 4). OB women with GWG within or under IOM recommendations reported significantly higher intake of high-fat fish (198 g/week) than OB women with GWG above recommendations (131 g/week) (p =0.049).

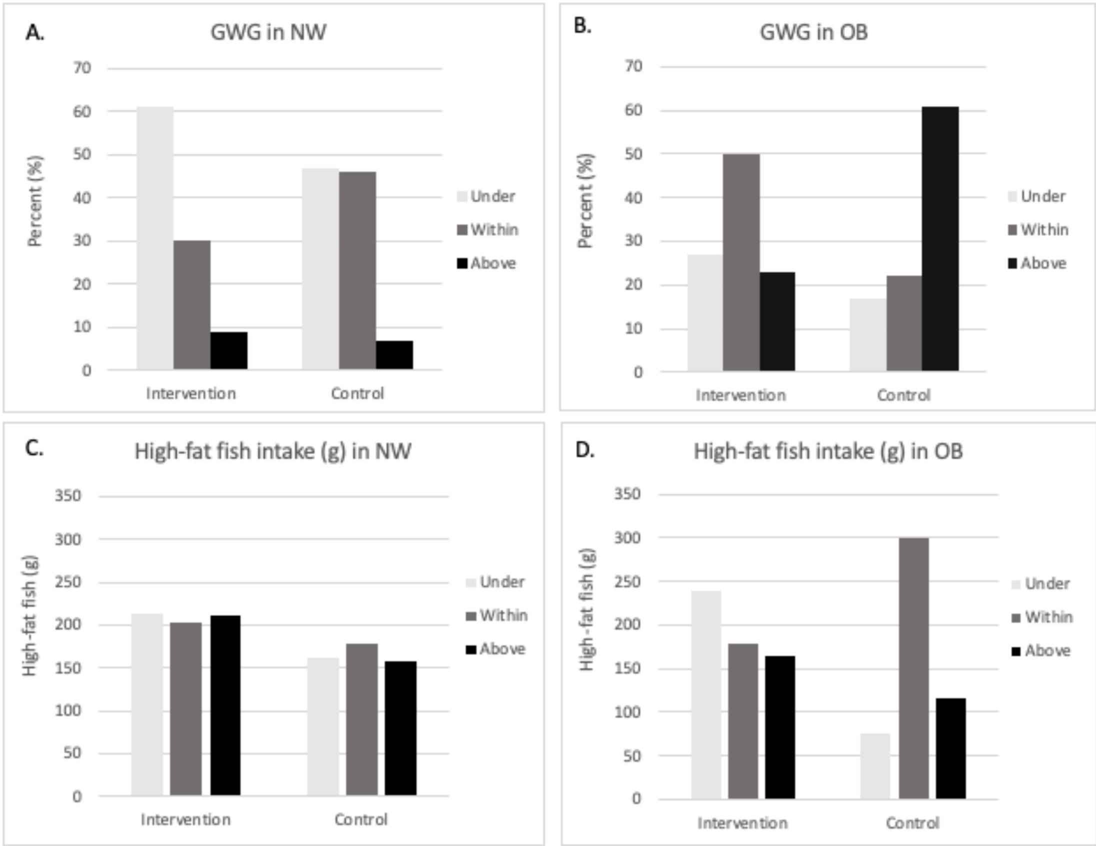

Figure 4: A) and B) percentage of women under, within or above IOM recommendations of GWG in NW and OB women. C) and D) intake of high-fat fish in each corresponding group of NW and OB women under, within or above IOM recommendations of GWG.
NW = normal weight. OB = obese. GWG = gestational weight gain.
Recommended range of total GWG in NW women: 11.5-16 kg
Recommended range of total GWG in OB women: 5-9 kg.

7.5 Infant characteristics and anthropometry measurement

Regarding gender, an even distribution could be seen in children born to NW as well as OB mothers (table 5).

Table 5: Gender distribution of infants participating in study visit 1 week and 12 weeks after birth

	NW [1]					OB [1]				
Gender [2]	Female		Male			Female		Male		
	n	% of NW	*n*	% of NW	P	*n*	% of OB	*n*	% of OB	P
1 week after birth	33	51.6	31	48.4	0.223	18	40.9	26	59.1	0.812
12 weeks after birth	34	47.2	38	52.8	0.221	15	37.5	25	62.5	0.070

[1] Children were divided into groups with reference to corresponding group of the mother NW = normal weight, OB=obese.
Women recruited later than trimester one, or with lower frequency of study visits were not excluded.
[2] Gender distribution calculated with Chi Squared-test.

Differences in infant anthropometry measurements were identified between children born to NW and OB mothers (table 6). Mean length was significantly greater in OB infants than NW infants 1 and 12 weeks after birth. Difference in weight was significant and measured to approximately 200 g at birth as well as 1 week thereafter. Although difference in weight could be seen 12 weeks after birth, this was not statistically significant. FM % was 2% higher in OB infants 1 week after birth (table 6).

Table 6: Anthropometry at birth and body composition at 1 and 12 weeks after birth in infants born to normal weight and obese mothers

	NW [1]		OB [1]		P [a]
	n	Mean (SD)	*n*	Mean (SD)	
At birth					
Length (cm)	99	50.4 (2.17)	58	51.6 (2.05)	0.527 [b]
Weight (g)	98	3609.9 (488.8)	59	3867.5 (514.0)	**0.002**
BMI (kg/m^2)	98	14.1 (1.2)	58	14.5 (1.3)	0.102
1 week after birth					
Length (cm)	64	51.3 (2.04)	44	52.7 (2.09)	**< 0.001**
Weight (g)	64	3685.6 (471.7)	44	3877.4 (497.9)	**0.045**
BMI (kg/m^2)	64	13.9 (1.1)	44	13.9 (1.2)	0.884
Waist circumference (cm)	51	35.2 (2.6)	40	35 (2.3)	0.650
Hip circumference (cm)	45	31.4 (2.0)	35	31.1 (2.2)	0.601
Lower leg length (cm)	42	13.2 (0.7)	38	13.3 (0.7)	0.777
Fat mass (kg)	63	0.5 (0.2)	44	0.6 (0.2)	**0.010**
Fat mass percent (%)	63	13.7 (4.1)	44	15.7 (3.8)	**0.012**
Fat free mass (kg)	63	3.2 (0.3)	44	3.3 (0.4)	0.196
12 weeks after birth					
Length (cm)	73	61.0 (2.4)	42	62.1 (2.1)	**0.019**
Weight (g)	73	6142.2 (806.2)	42	6441.0 (825.4)	0.060
BMI (kg/m^2)	73	16.5 (1.7)	42	4.7 (0.5)	0.523
Waist circumference (cm)	67	41.8 (3.1)	39	41.4 (2.9)	0.602
Hip circumference (cm)	56	39.4 (3.1)	33	39.8 (3.0)	0.580
Lower leg length (cm)	52	15.9 (0.8)	36	16.0 (0.9)	0.576
Fat mass (kg)	69	1.5 (0.5)	40	1.6 (0.4)	0.599 [b]
Fat mass percent (%)	69	24.3 (5.8)	40	25.5 (3.9)	0.308 [b]
Fat free mass (kg)	69	4.58 (1.7)	40	4.74 (1.6)	0.129

[1] Children were divided into groups with reference to corresponding group of the mother
NW = normal weight, OB=obese. Women recruited later than trimester one, or with lower frequency of study visits were not excluded.
[a] P-values between NW and OB group calculated with Student's independent sample t-test.
[b] P-values between NW and OB group were calculated with Mann–Whitney U test

7.6 Correlations between dietary patterns and anthropometric measurements in mother and infant

Correlations between reported dietary fish, meat and energy intake in trimester three and maternal body composition are displayed in table 7. With NW and OB grouped together, significant negative correlations were found between total fish intake and weight, FM and FM%. Reported meat intake was positively correlated with FM and FM % in OB group. After adjusting for pre-gestational BMI, age, and education, these correlations regarding fish and meat intake did not maintain significance. Moreover, in NW group, energy intake showed a significant positive correlation with weight, FM and FM%. FM and FM%. These correlations were still significant after adjustments of potential confounders (table 7).

Table 7: Spearman's rank correlations between total fish intake (grams/week), high-fat fish intake (grams/week), meat intake (grams/week), energy intake (kcal/day) in trimester three and maternal body composition in trimester three [a]

	Weight (kg)			Fat mass(kg)			Fat mass percent (%)			Fat free mass (kg)		
Trimester 3	*n*	R	P	*n*	R	P	*n*	R	P	*n*	R	P
Total fish intake (g/week)												
NW	133	- 0.03	0.774	129	-0.12	0.234	129	- 0.11	0.204	129	0.06	0.526
OB	59	- 0.03	0.834	59	0.03	0.844	59	0.12	0.365	59	- 0.17	0.206
TOTAL	192	-0.18*	**0.012**	188	-0.21*	**0.003**	188	-0.21*	**0.004**	188	-0.13	0.077
Intake of high-fat fish (g/week)												
NW	131	- 0.13	0.156	128	- 0.13	0.152	128	- 0.10	0.244	128	- 0.05	0.579
OB	51	- 0.10	0.509	51	-0.04	0.804	51	0.06	0.670	51	-0.20	0.151
TOTAL	182	- 0.16 *	**0.034**	179	-0.16*	**0.035**	179	- 0.14	0.063	179	- 0.13	0.096
Meat intake (g/week)												
NW	132	0.06	0.510	129	0.12	0.182	129	0.12	0.185	129	-0.05	0.575
OB	59	0.25	0.061	59	0.27*	**0.037**	59	0.28*	**0.030**	59	-0.03	0.840
TOTAL	191	0.10	0.171	188	0.14	0.054	188	0.14*	**0.049**	188	0.02	0.837
Energy intake (kcal/day)												
NW	133	0.20*	**0.022**	129	0.22*	**0.011**	129	0.18*	**0.048**	129	0.09	0.319
OB	59	0.06	0.635	59	0.13	0.327	59	0.14	0.284	59	-0.03	0.821
TOTAL	192	0.04	0.610	188	0.05	0.503	188	0.03	0.686	188	0.02	0.822

[a] Correlations calculated with data gathered from all women attending a trimester three visit and choosing to enroll their child.
Women recruited later than trimester one, or with lower frequency of study visits were not excluded.
P-values were calculated using Spearman's rank correlation.
* $P < 0.05$ Correlation is significant at the 0.05 level.

Furthermore, correlations between dietary intake in mother and body composition in infant were examined (table 8). Maternal meat intake showed no correlations with infant body composition. Negative significant correlations were found between maternal fish intake and several body composition measurements in infants. Both length, weight at birth and anthropometric measurements at follow up correlated negatively with fish intake for all infants combined. Additionally, strong significant negative correlations were seen in NW and OB separately, especially between total fish intake and NW infant body composition 12 weeks after birth, but also between high fat fish intake and weight 12 weeks after birth in OB group (table 8). When adjusting for pregestational BMI, maternal age and sex in infant, all correlations between high-fat fish intake and anthropometry measurements at birth and 1 week after birth maintained their significance and equivalent rank. Total fish intake showed no significant correlations after adjustments.

Table 8: Spearman's rank correlations between maternal dietary fish, meat and energy intake in trimester three and body composition in infant at 1 and 12 weeks after birth.

Infant body composition	Maternal dietary intake in trimester three					
	Total fish intake (g/week)			High-fat fish intake (g/week)		
	NW	OB	TOTAL	NW	OB	TOTAL
At birth						
Length (cm)	-0.16 (0.146)	-0.213 (0.142)	-0.25* **(0.003)**	-0.26* **(0.011)**	-0.17 (0.257)	-0.27* **(0.001)**
Weight (g)	-0.11 (0.298)	-0.112 (0.44)	-0.18* **(0.04)**	-0.25* **(0.015)**	-0.08 (0.601)	-0.24* **(0.005)**
BMI (kg/m^2)	-0.04 (0.711)	0.027 (0.856)	-0.02 (0.864)	-0.10 (0.320)	-0.01 (0.935)	-0.10 (0.24)
1 week after birth						
Length (cm)	-0.13 (0.315)	-0.332* **(0.032)**	-0.31* **(0.001)**	-0.27* **(0.03)**	-0.44* **(0.006)**	-0.38* **(<0.001)**
Weight (g)	-0.19 (0.129)	-0.256 (0.102)	-0.26* **(0.007)**	-0.21 (0.104)	-0.24 (0.152)	-0.26* **(0.009)**
BMI (kg/m^2)	0.19 (0.134)	-0.055 (0.727)	-0.09 (0.357)	-0.07 (0.589)	-0.03 (0.883)	-0.05 (0.629)
Fat mass (kg)	-0.13 (0.319)	-0.21 (0.183)	-0.26* **(0.008)**	0.02 (0.907)	-0.19 (0.257)	-0.12 (0.232)
Fat mass percent (%)	-0.07 (0.571)	-0.163 (0.302)	-0.19 (0.053)	0.11 (0.394)	-0.19 (0.26)	-0.04 (0.663)
Fat free mass (kg)	-0.16 (0.214)	-0.188 (0.232)	-0.21* **(0.037)**	-0.30* **(0.018)**	-0.24 (0.147)	-0.30* **(0.002)**
12 weeks after birth						
Length (cm)	-0.13 (0.293)	-0.312 (0.056)	-0.23* **(0.015)**	0.22 (0.061)	-0.24 (0.174)	-0.15 (0.12)
Weight (g)	-0.36* **(0.002)**	-0.226 (0.173)	-0.33* **(<0.001)**	-0.07 (0.575)	-0.38* **(0.025)**	-0.23* **(0.018)**
BMI (kg/m^2)	-0.444* **(<0.001)**	-0.072 (0.667)	-0.310* **(0.001)**	-0.19 (0.124)	-0.234 (0.183)	-0.177 (0.071)
Fat mass (kg)	-0.353* **(0.003)**	-0.182 (0.288)	-0.305* **(0.002)**	-0.03 (0.98)	-0.111 (0.538)	-0.075 (0.456)
Fat mass percent (%)	-0.288* **(0.016)**	-0.073 (0.672)	-0.225* **(0.021)**	-0.216 (0.132)	0.09 (0.619)	0.036 (0.723)
Fat free mass (kg)	-0.251* **(0.037)**	-0.207 (0.255)	-0.260* **(0.007)**	-0.038 (0.762)	-0.373* **(0.033)**	-0.263* **(0.008)**

Correlations presented as R(P) unless noted otherwise. *
P < 0.05 Correlation is significant at the 0.05 level.

8. Discussion

This study aimed to examine body composition in mother and infant in relation to reported fish and meat intake. A total of 132 normal weight and 45 obese pregnant women were followed throughout gestation as they participated in regular study visits with measurements of body composition and collection of FFQ. Women randomized to intervention received dietary guidance. Enrolled infants were measured at 1 and 12 weeks after birth.

8.1 Main findings

8.1.1. Reported dietary intake and changes in maternal body composition

Mean total fish intake in each trimester corresponded with the amount recommended during pregnancy according to The Norwegian Health Authorities, set to 2-3 portions of fish per week assuming one portion is equivalent to 150 g (36). Both OB and NW intervention group reported an increased consumption of high fat fish particularly between first and second trimester, NW also significantly increasing total fish intake throughout the entire intervention period. NW control reported a significantly increased meat intake during the follow up period. Although no significant decrease was seen in intervention group, the tendency could be explained by the corresponding increase in fish intake. These results suggest that the PONCH dietary intervention had an effect on fish intake.

OB women showed a smaller GWG than NW women. Similar trends have previously been reported in several studies (14, 62, 63). Although 40% of OB women gained more weight than recommended during pregnancy according to IOM guidelines, mean GWG for both NW and OB women was within the range of weight gain recommended for corresponding BMI class (13). Participants in OB intervention group were in addition to increasing fish intake also instructed to lower energy intake by 20% in reference to their BMR at baseline. Subjects were unable to fulfil this implementation reporting mean energy intake in trimester 3 at 7%

below baseline. This did not result in a significantly lower GWG than in OB control. Nevertheless, in comparison to OB control, there was a significantly lower percentage of OB intervention that gained weight above recommendations, which indicates a successful intervention.

We found a higher intake of high-fat fish in OB women with GWG within or under recommendations, when analyzed against OB women with excess weight gain. In a former Norwegian cohort study, Hillesund et al presented evidence that high adherence to a diet consisting of whole grains, fish and large quantities of berries, fruits and vegetables optimized gestational weight gain with reference to IOM guidelines (64). Considering fish to be part of a beneficial diet, this intake might represent an overall healthy consumption of foods rather than affecting the outcome in body composition per se. With the knowledge that an excess GWG in especially overweight and obese mother increase the risk of adverse outcomes such as gestational hypertension, emergency caesarian section and neonates needing intensive care (65-67), it is of great importance to direct preventive strategies towards these individuals. Further investigations of this sample could be targeted at analyzing the quality and distribution of nutrient in more detail and compare GWG with adherence to NNR, with the aim to optimize guidelines further.

No significant correlations were found between fish as well as meat intake and maternal body composition after adjusting for potential confounders. However, several correlations pointed toward a negative tendency between fish intake and weight, fat mass and fat mass percent with NW and OB grouped together. Regarding meat intake, Bosaeus et al reported positive correlations between reported meat intake and fat free mass in first trimester (50). We found no such correlation in this study, perhaps due to another sample of participants involving both NW and OB women. In addition, we adjusted correlations for pre-gestational BMI, maternal age and education.

In accordance with research presented by Holowko et al (68) and a report from The Swedish national board of Health and Welfare published in 2015 (12), we found that lower education is associated with higher pregestational BMI.

8.1.2. Infant anthropometric measurements

In this study, we found that high maternal fat fish intake was inversely correlated with infant length and weight at birth and at one week of age, even after adjusting for potential confounders. Before adjusting for pre-gestational BMI, infant sex and maternal age, significant negative correlations between total fish intake and infant fat mass were seen 1 and 12 weeks after birth. This was not only seen with NW and OB analyzed together, but also in NW group 12 weeks after birth. This result is of particular interest since the group of NW women constitutes a highly homogenous sample of women that does not differ in many other aspects. Results from previous studies investigating the effect of maternal dietary fish intake on infant body composition are inconclusive. On the one hand, some studies present a positive correlation between fish intake and birth size in terms of weight and head circumference (69). On the other hand, some studies demonstrate the opposite relationship with an inverse association between fish intake and birth size as well as body fat in childhood (70-72). With the use of PUFA supplementation, Bergmann et al concluded that increased DHA intake during pregnancy might result in lower BMI in infant (73). Since the participants in NW control group reported a significantly higher use of n-3 PUFA supplementation in trimester 3 than NW intervention, we might have difficulty distinguishing the true effect of fish intake on maternal and infant body composition in this study. As PUFA supplementation might represent a potential confounder in this context, this ought to be considered in future studies involving this data.

On the one hand, we found no correlations between maternal meat intake and infant body composition. Previous studies have suggested that high protein intake is associated with higher birth weights (34, 35). On the other hand, these studies did not isolate meat intake as analyzed in this study. Further objectives for this material could take this into consideration, seeing that the need for protein also can be met from dairy products and non-animal sources of protein.

Infants born to obese mothers had higher weight, length and fat mass. This has been shown for the PONCH study prior to this project (Andersson et al) (74). Starling et al performed an observational study in 2015 featuring corresponding results of pregestational BMI in direct relation to neonatal adiposity (75). With several studies indicating that that maternal obesity may increase the risk of overweight, obesity and metabolic syndrome in childhood (21, 26) along with research suggesting that the epigenetic regulations taking place in utero may affect the child's metabolic profile in a long-term perspective, (44, 71, 76) it would be motivated to continue studying the effect of maternal fish intake in children as they grow older.

8.2 Methodological considerations

Strengths of the PONCH study include 1) the study design as a randomized controlled trial 2) that participants in the intervention groups acquired individualized information 3) that follow up and measurements took place at one location and in contact with the same researcher and dietician 4) that the use of BOD POD and PEA POD is golden standard for measuring body composition in pregnant women and infants respectively.

Due to the aim of studying the dietary intervention starting in trimester one and continuing throughout pregnancy, analyses were performed on women participating in all study visits. This reduced the number of subjects involved in analyses, an aspect undermining the statistical power, but motivated according to the specific objectives. Drop out was partly due to short time between delivery and first infant visit. Some food-frequency surveys were left uncompleted, but answering frequency was consistently high at approximately 95% throughout the follow up period.

With self-assessed FFQ, there might be a limited ability to draw accurate conclusions about the effect of dietary intake, due to subjects under- or overestimating quantity of food. Some studies suggest that report bias may occur when the nutrient in question is referred to as socially desirable e.g. fish (77). Nevertheless, a strength in this study was the longitudinal follow up where the subject served as their own control. Findings from several studies investigating the validation of FFQ in groups consisting of obese, normal weight or pregnant participants suggest that FFQ has good validity when comparing with other methods as well as biomarkers (78-80). Each participant reported the frequency of which their meals contained either fish or meat in a separate questionnaire. By estimating one portion to 150 g and 175 g respectively, our calculations might have overestimated the quantity consumed. A possible improvement in this matter would be to ask the participants to define their portion size with reference to the amount stated in the questionnaire. In short, isolating the effect of certain nutrients tends to be challenging. To gain more knowledge, further research within this field could also investigate the possible correlations between serum fatty acid profile as a biomarker for fish intake and body composition in mother and offspring.

Higher education was associated with lower body weight. A large body of evidence also suggest correlations of higher education with a healthier nutrient intake in general, (81-83) also including a higher fish intake (84, 85). With this in mind, correlation analyses were adjusted for education even though the result of analyses indicated only modest associations between education and fish intake in this study. In addition, one must consider selection bias, since women with a more pronounced interest in the field of health and nutrition are more inclined to participate in this type of study.

8.3 Conclusions and implications

In conclusion, this study shows that dietary guidance can help women increase their fish intake during pregnancy. Obese women with a high fish intake during pregnancy more often gain weight within IOM recommendations. Moreover, our results suggest that a large intake of high-fat fish during pregnancy is correlated with smaller children. This information could contribute to the research aiming to optimize recommendations for women in risk of pregnancy complications and increased post-partum weight retention, as well as long term health of the children.

9. Populärvetenskaplig sammanfattning

”Den gravida kvinnans och nyföddas kroppssammansättning i förhållande till fisk- och köttintag under graviditet”

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Om den gravida kvinnan inleder sin graviditet med ett Body Mass Index (BMI) motsvarande graden för fetma (BMI över 30 kg/m^2) eller har en allt för kraftig viktökning under graviditet så ökar risken för graviditetsdiabetes, allt för stor fostertillväxt, komplikationer vid förlossning samt risken för långsiktig påverkan hos barnet i form av ofördelaktiga hälsoparametrar och fetma. Med den växande fetmaepidemin är det således av största vikt att hitta preventiva strategier för att i möjligaste mån minska riskerna under graviditet. Fisk med sitt innehåll av fleromättade fettsyror är ett födoämne som den gravida kvinnan rekommenderas äta två till tre gånger per vecka. Fettsyrorna har, förutom sin avgörande roll i utvecklingen av fostrets nervsystem, i upprepade studier indikerat en reducerande effekt på fetma, både hos vuxna som barn.

Följande studie syftade till att studera den gravida kvinnans fiskintag och den möjliga effekten på kroppssammansättning hos både henne och det nyfödda barnet. Från år 2009 till 2018 rekryterades 177 kvinnor till PONCH-studien, ett forskningsprojekt med ett övergripande ändamål att studera möjliga angreppspunkter för att främja kvinnohälsa under graviditet. Kvinnorna som antingen klassificerades som normalviktiga eller obesa (BMI> 30 kg/m^2) delades in i ytterligare två subgrupper, en interventionsgrupp och en grupp som svarade som kontroll. Vid totalt tre tillfällen kom kvinnorna på mottagningsbesök där man med hjälp av ett tillförlitligt instrument mätte kvinnans kroppssammansättning i form av vikt, mängden fettmassa, kroppsfett i procent samt fettfri massa. Samtliga fyllde i heltäckande frågeformulär rörande kostvanor. Till interventionen tillkom kostrådgivning med en legitimerad dietist, som mellan besöken kontaktade kvinnorna för uppföljning av kostråden. Mot slutet av graviditeten blev kvinnorna tillfrågade ifall de kunde tänka sig att registrera sitt barn i studien. Vid godkännande fick barnen komma på motsvarande mätning av kroppssammansättning en respektive tolv veckor efter födsel.

Utifrån kvinnornas rapporterade intag kunde vi se att båda interventionsgrupperna ökade sitt intag av fisk under de första månaderna av graviditet. Dock kunde man inte utläsa statistiskt säkerställda samband mellan den gravida kvinnans kostintag och kroppssammansättning i sen graviditet. Emellertid kunde vi urskilja trender, där ett ökat fiskintag visade samband med en lägre vikt, fettmassa och kroppsfett hos modern. Hos det nyfödda barnet kunde man se ett samband mellan ett högre intag av fet fisk hos mamman och lägre vikt samt kortare längd hos barnen vid födsel och en vecka därefter. Tidigare studier har påvisat liknande resultat där ett högt fiskintag visat sig ha en association med lägre vikt och mindre kroppsfett hos den nyfödda.

Gällande kostinterventioner föreligger alltid ett problem att isolera effekten av enskilda näringsämnen. Eftersom fisk är föda associerat med en generellt mer hälsosam kosthållning kan den sammansatta effekten av bättre kost leda till mer fördelaktiga parametrar för hälsan. Biomarkörer i blod som representerar intag av fleromättade fettsyror kan vara nästa led i hur man kan studera effekten av fiskintag på kroppssammansättning.

Sammanfattningsvis visade denna studien att strukturerad rådgivning kan bidra till ökat fiskintag under graviditet. Våra resultat indikerar att obesa kvinnor som äter mer fisk tenderar att i större grad hamna inom ramen för rekommenderad viktuppgång under sin graviditet. Barn som föds till kvinnor med högre konsumtion av fet fisk uppvisar dessutom en lägre vikt och längd kring födsel. Ovanstående kan innebära en reducerad risk för befarade komplikationer hos den gravida kvinnan med ett högt BMI. Således kan dessa resultat bidra till den forskning som syftar till att utforma individuella råd för kvinnor som befinner sig i risk för graviditetskomplikationer.

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12. Appendix

Appendix 1: Questionnaire concerning weekly fish- and meat intake

Namn: **Personnummer:** **Datum:** **Trimester:** **ID-nummer :**

1. **Hur många lagade måltider per vecka äter du:**

Fisk och skaldjur: ……………………………………………………gånger/vecka

Kött: ……………………………………………………………………gånger/vecka

2. **Hur ofta äter du något av nedan nämnda livsmedel? Sätt ett kryss i den ruta som stämmer bäst in på ditt fisk- och köttintag de senaste 3 månaderna. Om du äter något mindre än 1 gång i månaden sätt ett kryss på aldrig.**

Frekvens	Aldrig	1 gång per månad	2 gånger per månad	1 gång per vecka	2 gånger per vecka	3 gånger per vecka	4-6 gånger per vecka	1 gång eller mer per dag
Vit fisk*								
Lax, makrill, öring								
Fiskbullar/-pudding/-kakor								
Fiskpinnar								
Kräftor/kräftsjärtar, räkor, krabba								
Rödspätta								
Sill/ strömming								
Inlagd sill								
Tonfisk i vatten/olja								
Skaldjursallad/räksallad och liknande								
Annat:								
Fiskmåltider som äts utanför hemmet								
Fiskmåltider som äts i hemmet								
Fisk till frukost								
Fisk till lunch								
Fisk till middag								
Annat								

*Till exempel hoki, kolja, sej, torsk, Alaska Pollock, pangasius, eller annan fisk med vitt kött.

Vänligen vänd! →

Frekvens	Aldrig	1 gång per månad	2 gånger per månad	1 gång per vecka	2 gånger per vecka	3 gånger per vecka	4-6 gånger per vecka	1 gång eller mer per dag
Nötkött (inte färsrätter)								
Fläskkött (inte färsrätter)								
Lammkött (inte färsrätter)								
Kyckling/kalkon (inte färsrätter)								
Quornprodukter								
Sojaprodukter								
Linser/bönor								
Korvprodukter								
Inälvsmat								
Färsrätter**								
Leverpastej som pålägg								
Skinka som pålägg								
Korv/salami som pålägg								
Annat:								
Köttmåltider som äts utanför hemmet								
Köttmåltider som äts i hemmet								
Kött till frukost								
Kött till lunch								
Kött till middag								
Annat:								

**Köttbullar, köttfärssås etc.