CHAPTER 7

General discussion
In this chapter, the main findings of this thesis are discussed and put into a broader perspective, followed by methodological considerations and recommendations for future research and public health practice.

**REFLECTION ON THE RESULTS**

*The effects of a preconception lifestyle intervention on women’s lifestyle and health*

The preconception lifestyle intervention was successful in decreasing the intake of high caloric snacks and beverages and in increasing physical activity. However, changes in lifestyle were modest. For instance, women in the intervention group decreased their intake of sugary drinks with half a glass per day, decreased their savoury snack intake with almost two handful per week, decreased their sweet snack intake with almost two portions per week and were more physically active for 25 minutes per day compared to women in the control group (chapter 2).

We hypothesised that the wish to conceive would be a strong motivator for women to change their dietary intake and physical activity\(^1\)\(^-\)\(^4\) and we therefore expected to find larger intervention effects on lifestyle then we observed. The fact that we only found modest changes in lifestyle could be explained by several factors. Women were advised by the intervention coaches to avoid large caloric reductions because they were actively trying to conceive. Furthermore, because our study population consisted of infertile women who tried to become pregnant\(^5\), the stress associated with infertility may have interfered with women’s ability to change lifestyle.\(^6\)\(^-\)\(^8\) Personal communications with women participating in the LIFEstyle study indeed confirmed that concerns and stress due to their infertility made it hard to focus on a healthy lifestyle. The Lancet series about preconception health, published in 2018, showed a model of preconception action phases suggesting that active support for a healthy preconception lifestyle, e.g. one-to-one counselling or a group intervention, should be provided from the moment a couple intends to become pregnant.\(^9\) Our study suggests that, among obese infertile women, active support to change lifestyle after one year of trying to conceive might not be the ideal period for lifestyle changes. Instead, we might have achieved larger lifestyle changes if we would have targeted these women earlier or already before they had an active wish to conceive, which might also have positively affected their fertility.\(^10\)\(^,\)\(^11\)

Regardless of timing, changing lifestyle and maintaining a healthy lifestyle in the long term are notoriously difficult: Lifestyle interventions often show a typical pattern of successful lifestyle and weight change in the short term, followed by diminished
adherence to the intervention in the long term, and often disappointing long term health outcomes. Many participants even regain half of their lost weight within a year after following a lifestyle program and return to their baseline weight within 3-5 years.\textsuperscript{12} We observed a similar pattern in our data: stronger intervention effects of our preconception lifestyle intervention on dietary intake and physical activity in the short term (3 months after randomisation), after which intervention effects decreased (12 months after randomisation; chapter 2). However, we found indications of sustained effects over time since women allocated to the intervention group reported a lower energy intake at 5.5 years after randomisation compared to women in the control group (chapter 3). It should be noted that this lower energy intake was not reflected in differences in BMI between the two groups. Nevertheless, women in the intervention group who were successful in losing weight during the intervention period (lost ≥5% of their original body weight or reached a BMI<29kg/m\textsuperscript{2}) did have a lower weight at follow-up. Hence, being successful in changing lifestyle in response to the intervention predicts beneficial effects on lifestyle and weight in the long term. These results might also suggest that, in order to achieve sustainable long term results after a preconception lifestyle intervention, additional support is needed for women who are not able to lose weight successfully in the short term.

As discussed, changes in dietary intake and physical activity over time were modest and most women were still obese at follow-up (chapter 4). Nevertheless, these small changes were associated with improved cardiometabolic health in the short term\textsuperscript{13}, as well as with a lower BMI in the long term among women who successfully lost weight during the intervention (chapter 3). We therefore investigated the individual variation in lifestyle change and weight loss in relation to cardiometabolic health 3-8 years later. Indeed, we observed trends between increases in fruit intake and moderate to vigorous physical activity during the first six months after randomisation with higher fat free mass 3-8 years later, and between weight loss during the first six months after randomisation with higher HDL-cholesterol 3-8 years later (chapter 4). However, after correction for current lifestyle, we did not observe statistically significant associations between changes in lifestyle over the first six months of the LIFEstyle study and cardiometabolic health 3-8 years later. This indicates that maintaining a healthy lifestyle, e.g. by prolonged lifestyle coaching\textsuperscript{12}, is important to improve cardiometabolic health in the long term.
CHAPTER 7

Women’s lifestyle around the period of pregnancy and offspring’s cardiovascular health

The preconception lifestyle intervention lowered the intake of high caloric snacks and beverages during the first year of the LIFEstyle study, but did not show an effect on vegetable and fruit intake (chapter 2). We speculate that this lack of intervention effect on the intake of vegetables and fruit might be because of the focus on eating less calories to achieve weight loss. Nevertheless, higher preconception intake of vegetables and fruit were associated with more favourable offspring cardiovascular health (chapter 5). We hypothesise that these associations are driven by a higher maternal fibre intake, which is associated with lower maternal body weight and body fat, and thereby might favourably program offspring susceptibility to high blood pressure later in life. Furthermore, these higher intakes of vegetables and fruits probably represent a more healthy preconception dietary pattern and thereby positively influence offspring cardiovascular health. These results suggest that preconception lifestyle interventions in obese women should not only focus on discouraging unhealthy behaviours to lose weight, but also encourage healthy behaviours to optimize offspring health. Currently, the Netherlands Nutrition Centre focusses a lot on foods that should be avoided during pregnancy (e.g. raw milk cheeses, raw fish and shellfish, limited intake of caffeine and liquorice, fennel or anise tea). More attention should be given to the general guidelines for a healthy diet with additional requirements on how to eat healthy when pregnant.

Reviewing the available literature suggests that there was evidence for an association between higher maternal carbohydrate intake during pregnancy with higher offspring blood pressure, and a negative association between maternal protein intake with offspring carotid intima media thickness (chapter 6). These results indicate that not only maternal lifestyle before conception, but also maternal lifestyle during pregnancy is important for the future health of next generations.

METHODOLOGICAL CONSIDERATIONS

The following paragraphs discuss the methodological considerations from a broader perspective, which should be taken into account when interpreting our findings and putting them into a wider context.

Follow-up rate

In this thesis, we used data collected within one year after randomisation into the LIFEstyle trial and during our follow-up study at 3-8 years after randomisation (WOMB
The participation rate of our follow-up study was low\textsuperscript{16}, with follow-up rates of 38.3\% for the questionnaires (chapter 3) and 21.1\% for the physical examinations in women (chapter 4), and of 16.7\% for the physical examinations in children (chapter 5). This attrition reduces statistical power.\textsuperscript{16} In our study of the long term effects of the preconception lifestyle intervention on dietary intake and physical activity we had sufficient power to detect statistically significant differences between the intervention and control group (chapter 3). However, the statistical power of our study was limited when studying the association between changes in preconception lifestyle and women’s cardiometabolic health (chapter 4) and offspring’s cardiovascular health (chapter 5). Therefore, our results should be replicated in larger preconception studies before robust conclusions can be drawn.

The role of selection bias in follow-up studies

Another important consequence of the attrition is the potential introduction of selection bias, which can be defined as a systematic difference between participants included in the study and the non-participants. This limits the generalisability of our results. In this paragraph we focus on the generalisability of our results to the total LIFEstyle study population.

By comparing the participants to the non-participants insight can be gained in potential selection bias, which specifically impacts the interpretation of our results in chapter 3 and chapter 4. Women who were successful in losing weight during the intervention were more likely to participate in our follow-up study. Our conclusion that a preconception lifestyle intervention has sustainable long term effects therefore only applies to the women included in our follow-up study (chapter 3). We cannot conclude with certainty that also the women allocated to the intervention group that declined participation in our follow-up study had a lower energy intake at follow-up.

In our study relating changes in dietary intake and physical activity during the intervention to women’s cardiometabolic health 3-8 years later (chapter 4), the non-participants on average changed their dietary intake, physical activity and body weight to a larger extent than the participants, which might explain why we only observed trends instead of statistically significant associations.
Bias in self-reported and objectively measured data

To assess the amount of physical activity in our study, we used two measurement instruments: 1) The Short QUestionnaire to ASsess Health-enhancing physical activity (SQUASH) during the LIFEstyle trial, and 2) Actigraph accelerometers during follow-up (Table 7.1). The use of accelerometers to examine physical activity is a strength of our study, since accelerometers are considered a more reliable, valid and objective measurement compared to self-reported questionnaires as the SQUASH. However, accelerometers only capture highly dynamic activities. Activities like cycling and swimming are not (well) captured, but self-reported cycling and swimming activities did not differ between women allocated to the intervention and control group (chapter 3). Although considered a more objective measurement, when wearing an accelerometer participants might change their physical activity pattern compared to their normal physical activity due to the awareness of being studied, also known as the Hawthorne effect. This does, however, not influence our conclusions that a preconception lifestyle intervention has no long term effect on physical activity (chapter 3). Filling out the SQUASH might be influenced by social desirability bias, with a larger extent in the intervention group compared to the control group, because women in the intervention group were actively motivated and educated on a healthier lifestyle. This social desirability bias might explain part of the differences in reported physical activity between the intervention and control group (chapter 2), but since the changes in physical activity were reflected in long term cardiometabolic health (chapter 4) we consider it unlikely that the intervention effect can completely be attributed to social desirability bias. Additionally, people tend to overestimate their physical activity filling out the SQUASH. Indeed, if we plot the moderate to vigorous physical activity (MVPA) data measured by accelerometers against the self-reported MVPA data from the SQUASH we see that participants overestimated their time in MVPA filling out the SQUASH.

To examine dietary intake we used two different food frequency questionnaires (FFQ): 1) a 33-item FFQ during the LIFEstyle trial and 2) a validated semi-quantitative 173-item FFQ during follow-up (Table 7.1). The reason to include two different FFQs in our study is that the 33-item FFQ is limited to specific food groups the intervention focussed on, while the 173-item FFQ collects information on the complete dietary pattern. Accurately measuring habitual dietary intake is challenging: there is no easy to use alternative for self-reported data, which is highly subject to biases. Among other biases, FFQs are prone to recall bias, misreporting of dietary intake which is often affected by personal characteristics (e.g. obese people tend to underestimate their energy intake).
and social desirability bias. These measurement errors might explain part of the differences observed between the intervention and control group (chapter 2 and chapter 3). However, since we observed that the preconception lifestyle intervention led to a lower BMI in the long term in women who were successful in losing weight during the trial (chapter 3), and that changes in diet, although not statistically significant, pointed towards an improved cardiometabolic health (chapter 4), we consider it unlikely that the observed effects of a preconception lifestyle intervention on dietary intake can be fully explained by these biases. Furthermore, because of these measurement errors, the associations between lifestyle and health can be stronger or weaker than observed (chapter 4 and chapter 5).

**Confounding factors**

When studying associations between exposures and outcomes confounding factors are important to take into account. Confounding is the “dependence of counterfactual outcomes and exposure, possibly conditional on covariates”. The absence of confounding is an important requirement when defining relationships as causal. In general, we assume that randomisation minimizes the effects of confounding by equal distribution of (un) measured factors. However, an important condition is that randomisation is successful, i.e. there are no differences in baseline characteristics between the randomised groups and, when performing follow-up measurements, the balanced distribution of possible confounding factors remains the same over time. In chapter 2, describing the short term effects of the preconception lifestyle intervention on dietary intake and physical activity, we showed that the randomisation in our study population was successful as baseline characteristics did not differ between the intervention and control group. In chapter 3, describing the long term effects of the preconception lifestyle intervention on dietary intake and physical activity, women in the intervention group had a shorter duration of infertility compared to the control group. Women with a longer duration of infertility might be more motivated to change their lifestyle and we therefore corrected, among others, for this particular covariate. It might however be that also unmeasured confounders differed between the intervention and control group. This could have strengthened or weakened the associations.

When studying the association between preconception lifestyle with women’s (chapter 4) and offspring’s health (chapter 5), we no longer used the randomised controlled trial design. We instead used a cohort design, not distinguishing between women or offspring of mothers allocated to the intervention or control group (Table 7.1). We therefore needed
to control for confounding factors in our statistical analyses, using multivariable models. If a study population is large enough, multivariable models can handle a large number of confounders. However, our study population at follow-up was small in both studies (N ranged from 50-78 in chapter 4 and from 32-46 in chapter 5). We were therefore only able to add a limited number of confounders into our models and to correct for the strongest confounding factors. This means there might be confounding left in the associations between preconception lifestyle and women’s as well as offspring’s health, which could have strengthened or weakened the associations.

**External validity**

The external validity or generalisability of our results to other populations than our study population, might be limited because of different reasons. First, the population included in our studies is very specific, namely obese infertile women. Women who get pregnant more easily might face other facilitators and barriers to change their preconception lifestyle. Also, the general Dutch population is more healthy (i.e. a lower BMI on average and a lower percentage of smokers), higher educated and is more ethnically diverse than the participants in our study. Since these are all characteristics that affect lifestyle behaviours, our results may not be generalizable to the general Dutch population. In addition, it might be that the effects of lifestyle interventions on cardiometabolic health differ for Caucasian versus non-Caucasian persons.
Table 7.1. Overview of data used in this thesis to answer the research questions.

<table>
<thead>
<tr>
<th>Chapter 2</th>
<th>Chapter 3</th>
<th>Chapter 4</th>
<th>Chapter 5</th>
<th>Chapter 6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Study</td>
<td>LIFEstyle study</td>
<td>WOMB project</td>
<td>LIFEstyle study (exposure)</td>
<td>LIFEstyle study (exposure)</td>
</tr>
<tr>
<td>Study population</td>
<td>All women randomised into the LIFEstyle study</td>
<td>All women participating in stage I of the WOMB project</td>
<td>All women participating in stage II of the WOMB project</td>
<td>All children participating in stage II of the WOMB project with maternal data on dietary intake and physical activity during the preconception period</td>
</tr>
<tr>
<td>Study design</td>
<td>RCT</td>
<td>RCT</td>
<td>Cohort</td>
<td>Cohort</td>
</tr>
<tr>
<td>Total N</td>
<td>511-535</td>
<td>155-179</td>
<td>50-78</td>
<td>32-46</td>
</tr>
<tr>
<td>Exposure</td>
<td>Randomisation group</td>
<td>Randomisation group; Successful weight loss, unsuccessful weight loss, control group</td>
<td>Change in dietary intake (vegetable, fruit, sugary drinks, savoury and sweet snacks), physical activity (total MVPA) and weight during the first six months of the LIFEstyle study</td>
<td>Last measured maternal dietary intake (vegetable, fruit, sugary drinks, savoury and sweet snacks), mean physical activity (total MVPA) during the LIFEstyle study before conception</td>
</tr>
<tr>
<td>Outcome</td>
<td>Dietary intake (33-item FFQ): • Vegetable • Fruit • Sugary drink • Savoury snack • Sweet snack</td>
<td>Dietary intake (173-item FFQ): • Energy intake • Macronutrient intake • DHD15-index score</td>
<td>Women’s: • Body Mass Index • SBP and DBP • Fat mass and fat free mass • PWV • Glucose • Insulin • HOMA-IR</td>
<td>Offspring’s: • Body Mass Index • Waist:height ratio • SBP and DBP • Fat mass and fat free mass • PWV</td>
</tr>
<tr>
<td></td>
<td>Physical activity (SQUASH): • Total MVPA • Leisure time MVPA</td>
<td>Physical activity (accelerometers): • Total PA • Total MVPA</td>
<td>Offspring’s:</td>
<td></td>
</tr>
</tbody>
</table>

RCT = randomised controlled trial; MVPA = moderate to vigorous physical activity; DHD15-index score = Dutch Healthy Diet 2015 index score; PA = physical activity; SBP = systolic blood pressure; DBP = diastolic blood pressure; PWV = pulse wave velocity; HOMA-IR = Homeostatic model assessment (HOMA) or β-cell function and insulin resistance (IR); LDL = low-density lipoprotein; HDL = high-density lipoprotein.
RECOMMENDATIONS FOR FUTURE RESEARCH

Future research about optimizing preconception lifestyle is essential, because the unhealthy lifestyle and rising obesity among women of reproductive age affect health in even more than two generations.  

*Intervention period: start earlier and stop later*

We expected that the most important motivation for obese infertile women to change their lifestyle preconception would be that obesity reduces the chances of becoming pregnant. However, the struggle with infertility and the accompanying stress might have made lifestyle changes more difficult. Additionally, personal communications with study participants have shown that many women were unaware of the link between obesity and infertility. We therefore recommend to target obese women for lifestyle intervention in future research already before they develop an active wish to conceive. However, when intervening before women have an active wish to conceive one cannot use the period around pregnancy as “teachable moment” to achieve a (more) healthy lifestyle. Another triggering event that can make lifestyle change more easy is a medical trigger. For example, a one year lifestyle intervention among obese adolescent girls diagnosed with polycystic ovarian syndrome (PCOS) showed a BMI reduction of -3.9 kg/m², improved cardiovascular risk factors and decreased intima media thickness among girls who lost weight during the intervention. Future studies should examine if early intervening in dietary intake and physical activity, for example using medical triggering events like diagnosis of PCOS, would result in larger changes in lifestyle with the final aim to start pregnancy with a (more) healthy body weight. Additionally, the offspring examined in the WOMB project already had a high BMI and blood pressure at age 3-6 years. We therefore recommend to study if prolonged lifestyle coaching after birth would be more beneficial for the health of offspring of obese infertile mothers.

*Involve the partner*

Lifestyle behaviours are complex to change since an individual’s lifestyle is influenced by many factors. An important factor directly influencing an individual’s lifestyle is her family and home environment. Our preconception lifestyle intervention specifically focussed on women of reproductive age. The intervention did not directly involve the partner of the women, although social support is an important factor to achieve beneficial lifestyle change. For example, studies among pregnant women showed that family is important for social support when a woman changes or aims to maintain a healthy diet and aims to be or become physically active during pregnancy.
Moreover, a previously conducted preconception lifestyle intervention study (93.8% of the couples were subfertile and women’s median BMI was 25.3 kg/m\(^2\) [range: 18.4-42.4 kg/m\(^2\)]) also included the partners for dietary and lifestyle counselling. This study showed, among others, that the percentage of women consuming the recommended fruit intake increased with 15% in a three month time window.\(^{52}\) Within the LIFEstyle study, the percentage of women included in the intervention group consuming the recommended fruit intake increased with almost 8% in a three month time window. Comparing those studies should be done with caution because of different counselling techniques and frequencies of counselling, but it is interesting that actively involving the partner in less frequent preconception counselling showed larger effects on fruit intake compared to our more intensive preconception lifestyle intervention. To achieve larger and more sustainable preconception lifestyle change, we recommend to actively involve the partner in future preconception lifestyle interventions.

**Assessment of maternal lifestyle: dietary patterns over time, micronutrients and physical activity**

Dietary intake consists of a combination of different nutrients and foods. It is our dietary pattern that is associated with our health.\(^{53}\) It is therefore important that future research incorporates the assessment of the complete dietary pattern, instead of focussing on specific foods, food groups or nutrients. Furthermore, we disregarded micronutrient intake because of our measurement instruments. However, maternal nutrient status, which is closely linked to micronutrient intake, seems to have an important role in early life programming of the offspring.\(^{54-56}\) For example, high maternal folate and low vitamin B12 status are associated with higher adiposity rates and insulin resistance in the offspring.\(^{57}\) It is therefore important that future studies take into account micronutrient intake as well as maternal nutrient status.

Additionally, future preconception studies should not stop lifestyle assessment after conception. A systematic review showed that during pregnancy, women increase their fruit and vegetable consumption, and decrease their intake of fried and fast food, coffee and tea compared to their preconception intake according to a systematic review.\(^{58}\) Furthermore, the amount of physical activity changes during pregnancy: some studies show a decrease in physical activity,\(^{59,60}\) while other studies show an increase up until mid-gestation after which physical activity decreases.\(^{61}\) Studying lifestyle during the preconception phase as well as during pregnancy, including changes over time from the preconception phase to the different trimesters of pregnancy, provides more
insight in the programming effect of preconception lifestyle versus lifestyle during pregnancy. Additionally, the combined effect of preconception lifestyle and lifestyle during pregnancy on offspring health can be studied.

The programming role of maternal physical activity is studied less extensively compared to the programming role of maternal dietary intake (chapter 6), while increasing maternal physical activity during pregnancy might be an early intervention to improve cardiovascular health of the offspring.\textsuperscript{62,63} Moreover, rising obesity rates in women of reproductive age\textsuperscript{64–66} give an indication that dietary intake is unhealthy and physical activity is insufficient. Animal studies suggest a positive effect of restoring the energy balance by increasing physical activity to compensate for increased energy intake\textsuperscript{67}, which underlines the importance for future studies to examine preconception dietary intake and physical activity combined.

Assessment of health outcomes: need for a Delphi study
Our systematic review (chapter 6) showed that variations in assessment of dietary intake, physical activity and offspring health hampered meta-analyses, and therefore are a serious barrier to scientific progress regarding early life programming.\textsuperscript{68} To be able to draw more robust conclusions, harmonized exposure assessments should be used and offspring health outcomes should be measured consistently, at the same offspring age.\textsuperscript{68,69} A Delphi study\textsuperscript{70}, in which consensus is obtained about core exposure and outcome sets, would reduce waste of research resources and speed up scientific progress.

IMPLICATIONS FOR PUBLIC HEALTH PRACTICE
The effects of our preconception lifestyle intervention were too small to define concrete implications for public health practices. However, our results did show the potential of intervening during the preconception period.

Over the last decade, attention in public health practice increased regarding the importance of a healthy lifestyle around the period of pregnancy. Currently, there are multiple initiatives to improve preconception health aiming to optimize lifestyle and health of multiple generations. In the Netherlands, the minister of Health, Welfare and Sport endorses the importance of the first thousand days (from conception to second birthday) for later life health, and recently launched the national program “Kansrijke Start” aiming to give all children the best start in life.\textsuperscript{71} Besides this nationwide program,
there are multiple municipalities in the Netherlands using health promotion programs starting before or during pregnancy. Additionally, online health promotion programs are available for interested future mothers/couples who want to improve lifestyle before and during pregnancy.

Initiatives to improve lifestyle and health around the period of pregnancy are important for society, since an unfavourable start in life is, for example, associated with higher hospitalisation rates, all-cause mortality and unfavourable employment outcomes. These associations underline the importance of promoting a healthy preconception lifestyle, and thereby body weight, from a societal and economic perspective.

CONCLUSIONS
Our preconception lifestyle intervention showed modest changes in dietary intake and physical activity in obese infertile women, with indications for favourable health effects of preconception lifestyle changes in two generations. Intervening in dietary intake and physical activity of obese infertile women before their active wish to conceive would possibly lead to larger changes in lifestyle. Furthermore, in this way they would start pregnancy with a (more) healthy body weight, which is beneficial for the long term cardiometabolic health of women as well as their offspring. Additionally, we suggest that lifestyle interventions should go further than the preconception period alone. Both partners should be motivated for and informed about a healthy lifestyle for the benefit of their own health and that of their offspring.
REFERENCES


