Maternal nutrition and newborn health outcome

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Maternal nutrition and newborn health outcome

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(met een samenvatting in het Nederlands)

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door

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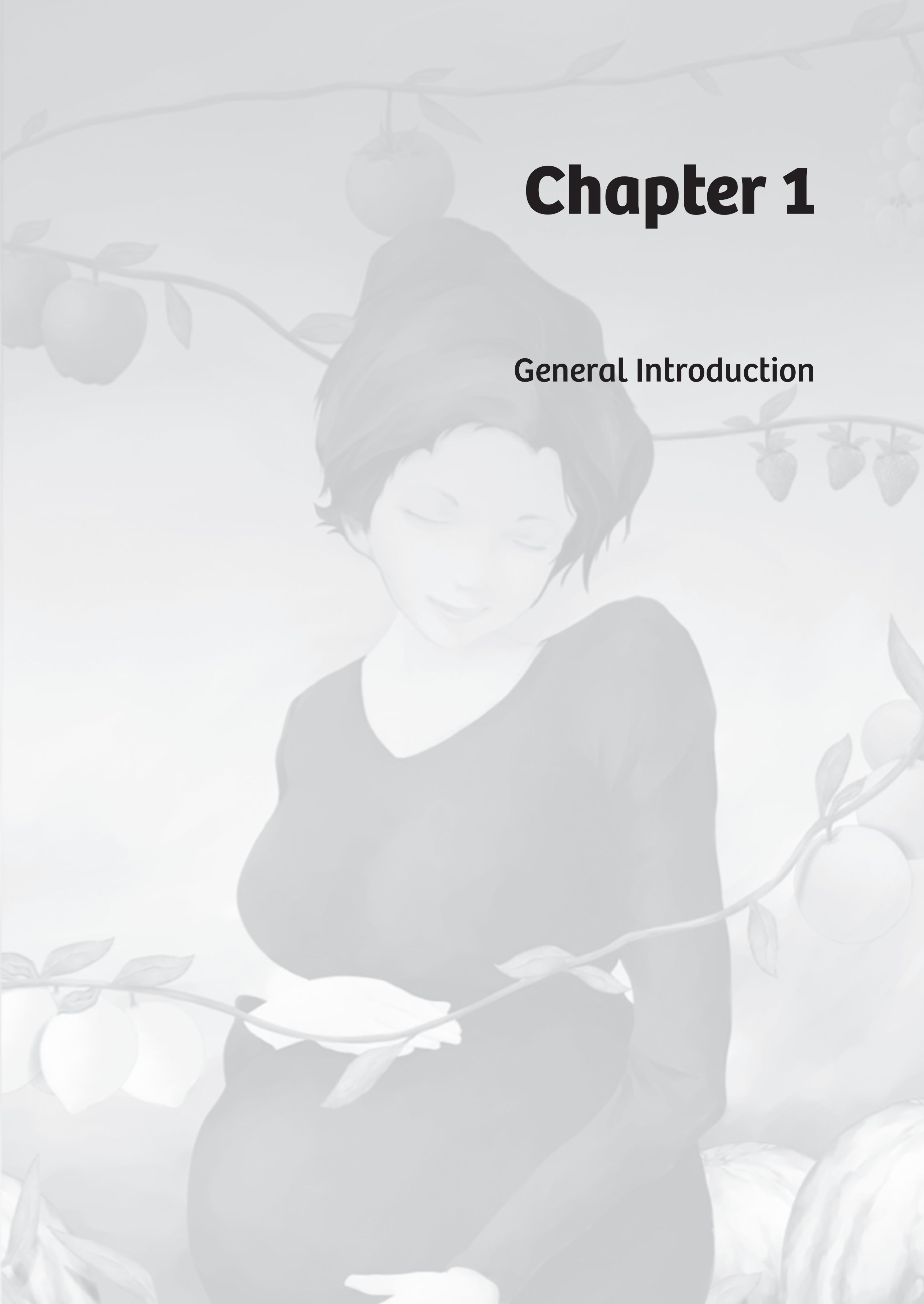

Chapter 1

General Introduction

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Background and Objectives

Nutrition in early life, during the period from conception until early childhood, influences health outcomes, both in the short- and long-term. Suboptimal nutrition in-utero caused by maternal under-nutrition, for instance, could hamper fetal growth and development, and subsequently lead to adverse pregnancy outcomes, such as low birth weight and prematurity.[1,2] Furthermore, the emergence of the hypothesis of Developmental Origin of Adult Diseases (DOHad) by Barker and colleagues,[3] has drawn much attention to the possible long-term effects of impaired nutrition in early life. Studies in this field suggest that poor nutrition during early life may have detrimental effects on the programming of body systems, which in the long term could manifest as diseases such as obesity, diabetes, metabolic syndrome and cardiovascular events.[4,5]

Despite increasing evidence in this area, knowledge gaps still remain. This thesis is aimed to provide further evidence on the effect of particular nutritional circumstances in early life, mainly during the period of preconception and gestation, on the pregnancy and birth outcomes. We examined the effects of over nutrition as represented by women's pre-pregnancy body mass index (BMI), as well as under-nutrition exposures such as in the case of hyperemesis gravidarum. We also investigated if transient lifestyle changes related to Ramadan fasting during pregnancy, which might cause a nutritional disturbance, had any impact on mothers' and babies' health. Furthermore, a protocol of a randomized trial on breastfeeding, which investigates the health effects of breastfeeding, is also described in this thesis. Our ultimate objective is that the evidence that we provide serves as a basis for preventive action as well as promotes further investigation into the best nutritional intervention measures, both in the context of prevention of adverse birth outcomes and in the prevention of later chronic diseases in adulthood.

Methods

The studies described in this thesis were based on the following cohorts or trial:

1. A cohort of 130 pregnant Muslim women who attended antenatal care in midwifery practices or hospitals in Amsterdam and Zaanstad, The Netherlands between 13 September and 31 December 2010 and were pregnant during the month of Ramadan in the respective year. These women were asked about their adherence to Ramadan fasting and being followed up for their pregnancy and newborn outcomes.
2. A cohort of Indonesian pregnant women who visited antenatal care in a maternal and child health primary care referral center, Budi Kemuliaan Hospital and its branch in Jakarta, Indonesia. As many as 2,252 women were recruited from July 2012 until October 2014 and were being followed up for their pregnancy and newborn outcomes. Within this cohort, we investigated if women's pre-pregnancy BMI, hyperemesis gravidarum, or Ramadan fasting affects various obstetric and birth outcomes. In addition, within this cohort, we also identified several factors that determine women's decision to observe Ramadan fasting during pregnancy.
3. Eleven annual birth cohorts from the Perinatal Registry Netherlands (Perined), a nationwide registry which documents all maternal, obstetric, postpartum, and neonatal information. All babies who were born from 1 January 2000 through 31 December 2010 were included, comprising 1,987,124 babies from various ethnical backgrounds. Our analyses were restricted to Dutch and Mediterranean singleton babies with known gestation length, which respectively concerned 1,481,435 and 139,322 babies. Within these babies, we studied the effect of in-utero Ramadan exposure on various birth outcomes.
4. A randomized breastfeeding optimization trial which is currently ongoing in Jakarta, Indonesia. This trial aims to provide some evidence about the effect of breastfeeding optimization program on breastfeeding rate and the effect of breastfeeding on children's growth and development as well as vascular and cardiac characteristics.

Outline

This thesis consists of three parts. The first part focuses on Ramadan (fasting) exposure as a model for transient nutritional disturbance during pregnancy that may influence obstetric and birth outcomes. We begin this part by describing factors that influenced women's decision to fast during pregnancy. In the next two chapters, we show the effect of Ramadan fasting on obstetric and birth outcomes of Indonesian women and Dutch women from Moroccan and Turkish origins. We end this part by describing the effect of Ramadan overlap with pregnancy on various birth outcomes in Mediterranean babies who were born in The Netherlands.

The second part focuses on the possible role of maternal nutritional circumstances early in pregnancy in determining obstetric and birth outcomes. Women's pre-pregnancy BMI and a diagnosis of hyperemesis gravidarum were examined as they may influence the risk of pregnancy complications, neonatal outcomes, as well as placental measures.

Lastly, in the third part, we describe the protocol of an ongoing randomized trial on breastfeeding promotion in the setting of developing country with low breastfeeding rate.

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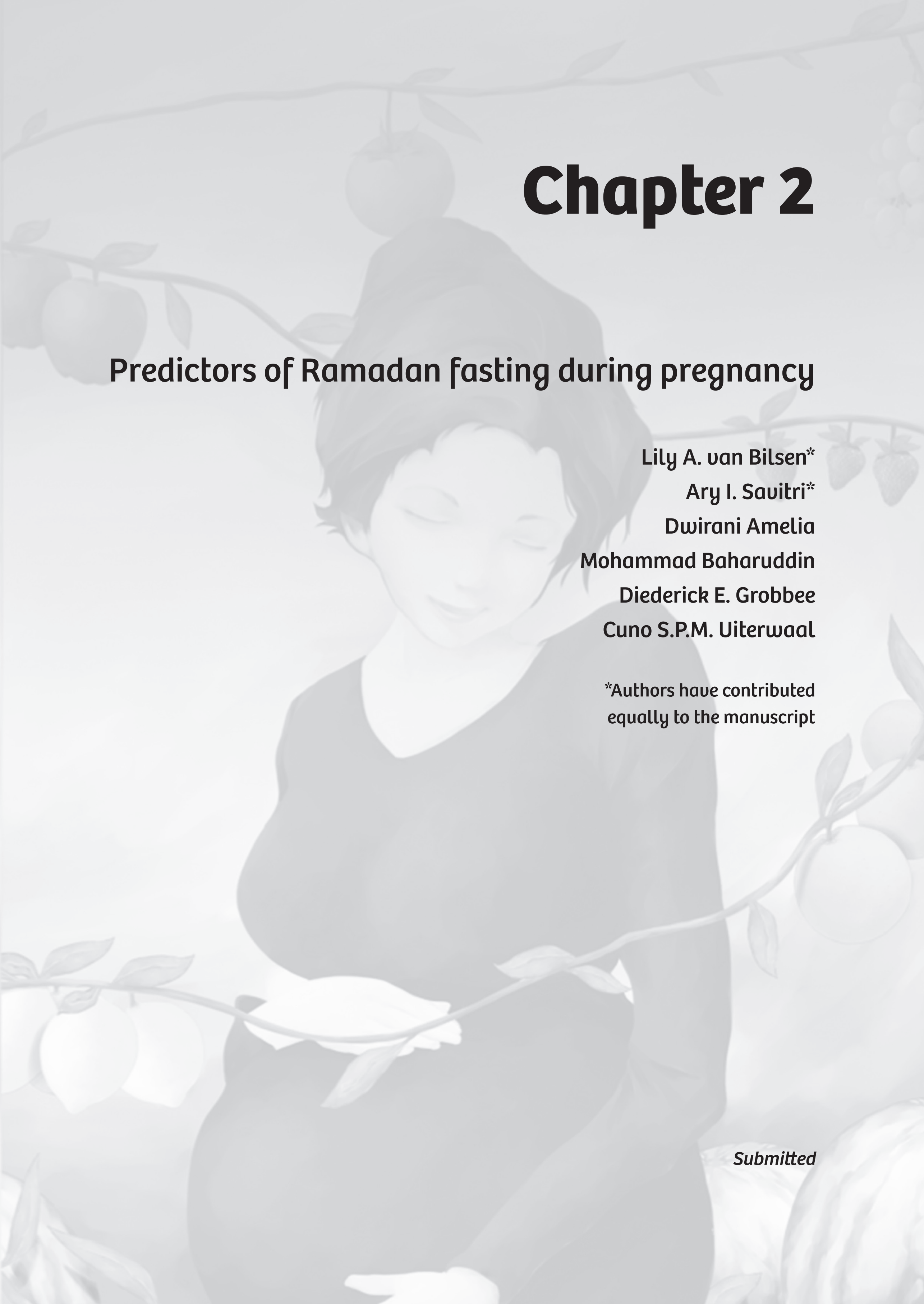
PART 1

Ramadan in pregnancy



Chapter 2

Predictors of Ramadan fasting during pregnancy

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Submitted

2

Abstract

Background

While it is unclear whether there are health effects of Ramadan fasting during pregnancy, it is crucial to investigate predictors and motivational factors involved in women's decision to observe the fast during pregnancy.

Methods

This cross-sectional study was conducted in 187 women within an established prospective cohort of the Budi Kemuliaan Hospital in Jakarta, Indonesia. The motivations and predictors were measured using a questionnaire.

Results

Adherence to Ramadan fasting was associated with earlier gestational age during Ramadan (odds ratio [OR] 0.96; 95% CI 0.92, 1.00; p = 0.06) and with higher pre-pregnancy BMI (OR 1.16; 95% CI 1.01, 1.33; p = 0.03). Non-participation was associated with opposition from husbands (OR 0.27; 95% CI 0.09, 0.73; p = 0.01) and with women's expressed fear of adverse fasting effects on their own or the baby's health (OR 0.47; 95% CI 0.22, 1.01; p = 0.05 and OR 0.43; 95% CI 0.21, 0.89; p = 0.01, respectively), although the latter was attenuated in multivariable analysis. Neither age, income, education, employment, parity, experience of morning sickness nor fasting during pregnancy outside of Ramadan determined fasting during pregnancy.

Conclusions

Earlier gestational age during Ramadan, higher pre-pregnancy BMI, and husband's opinion strongly influence women's adherence to Ramadan fasting during pregnancy. Fear of adverse health effects of Ramadan fasting is common in both fasting and non-fasting pregnant women, and should be appropriately addressed by care providers during counseling.

Introduction

Ramadan is a month according to the Islamic calendar when adult Muslims perform daylight intermittent fasting. Among the approximately 1.6 billion Muslims worldwide,[1] we estimate that there are some 300-400 million Muslim women of childbearing age. Although obligatory for all healthy adults and adolescents, Islamic law exempts pregnant and breastfeeding women from fasting. As Ramadan lasts for one month in a Lunar calendar, Ramadan fasting will overlap with pregnancy in every three of four births accounting for roughly 1 billion Muslims alive today who were in utero during Ramadan.[2] Despite the exemption, most pregnant women still fast.

There could be health effects of fasting on mothers and fetus, although no consensus was established on the matter. Several studies showed that prenatal exposure to Ramadan fasting results in lower birth weight, increased risk of hyperemesis gravidarum, urinary tract infections, and reduction in fetal breathing movements.[2-5] Long-term implications in offspring, possibly through fetal programming, were also reported. [6] On the contrary, many studies showed no effect of fasting on intrauterine growth, birth weight, and birth-time indices.[7-11]

Against the background of a lack of consistent health information about Ramadan fasting and the high proportion of women that adhere to fasting, it is important to know the motivations for or against fasting during pregnancy. There is a gap of knowledge concerning motivations for pregnant women's decision to fast during Ramadan. Most research focuses on attitudes and knowledge on the exemption law.[12] For women insisting to fast, it is crucial that knowledge is assembled to understand their motivations. This will prove useful for doctors and health workers, religious advisors, and women's peers, to adequately counsel Muslim pregnant women with regards to Ramadan fasting. In the present study of pregnant women, we evaluate factors that influence women's adherence to Ramadan fasting.

Methods

Study population

This cross sectional analysis was conducted within an established prospective cohort of pregnant women in a private hospital specialized in mother-child health services, Budi Kemuliaan Hospital in Jakarta, Indonesia. Pregnant women were recruited to the cohort during their regular first visits for antenatal care (ANC). All pregnant women attending visits were invited to join the study and if agreed, signed written informed consent. Participants were examined and interviewed by midwives according to standard clinical care and were followed up until they gave birth. This study was ethically approved by the Institutional Review Board of Budi Kemuliaan Hospital. Women, who paid an ANC visit before the month of Ramadan (10 July 2013 until 7 August 2013), were also asked to join this study. After the midwives gave an explanation, women who agreed gave their informed consent.

Data collection

Data on women's fasting adherence were collected using a daily self-administered questionnaire. During the visit prior to Ramadan (from 1 month to 1 week before the start of Ramadan), women were provided with a questionnaire. They were asked to fill in the questionnaire everyday during the month of Ramadan, indicating whether they fasted per day. After Ramadan, the women were asked to return the questionnaire to the midwives.

Furthermore, at their visit during the month of Ramadan, these women were also interviewed about factors that influenced their decision to fast or not to fast. Women were given several alternative answers for their motivations, which they ticked if they agreed with the statement. If their motivations were not included in the questionnaire, women were allowed to provide their own answer.

Women who indicated that they had fasted for one day or more were classified as having adhered to Ramadan fasting during pregnancy, while women who did not fast at all during pregnancy were classified as having not adhered to Ramadan fasting.

Maternal age was calculated as the difference between her date of birth and the date of her first ANC visit. Gestational age was calculated in days by subtracting the first day of last menstrual period (LMP) from the last day of Ramadan. Education was categorized as low education (elementary school or senior high school), medium education (senior high school), and high education (university or above).

Data analysis

Women's clinical and demographic data were tabulated by fasting adherence groups for descriptive purposes. All clinical, demographic, and motivation-related variables were considered as predictors of fasting adherence. Univariable logistic regression was first performed to evaluate the unadjusted relation between each predictor and fasting adherence. Furthermore, a multivariable logistic regression was performed to investigate the relation between each predictor and fasting adherence, adjusted for all other predictors. Results are expressed as odds ratios with 95% CIs and corresponding *p* values. Statistical significance were considered to be a 2-sided p value < 0.05. All analyses were done with SPSS version 22.0 for Mac (SPSS Inc., Chicago, IL).

Results

Of the 187 pregnant women included in the study, 149 (80%) fasted during Ramadan and 38 did not fast at all. **Table 1** shows the demographic characteristics of the study population separately for fasting and non-fasting women. All women were married, with a mean age of 29.1 years. Furthermore, most women completed senior high school in both groups. In addition, approximately half of the women were not employed and had a household income of less than 2.5 million IDR. In both groups, the proportion of multigravidas and primigravidas was similar. With regards to the stage of pregnancy, few women were in their first trimester during Ramadan. Moreover, the mean gestational age in the fasted group was lower than in the non-fasted group. The mean pre-pregnancy BMI was higher in the fasted group than in the non-fasted group.

Table 1. Baseline characteristics of the study population

Variable	Ramadan fasting		p-value
	Yes n = 149	No n = 38	
Age (SD)	29.1 (5.7)	30.0 (5.6)	0.88[a]
Gestational age in days (SD)	177.0 (70.7)	206.2 (68.7)	0.02[a]
Income in IDR (%)[#]			0.66[b]
< 2.5 million	70 (47.0)	22 (57.9)	
2.5 – 5 million	58 (38.9)	11 (28.9)	
> 5 million	12 (8.1)	3 (7.9)	
Refused to answer	9 (6.0)	2 (5.3)	
Education* (%)			0.92[b]
Low	17 (11.5)	4 (10.5)	
Medium	92 (62.2)	25 (65.8)	
High	39 (26.4)	9 (23.7)	
Employment (%)			0.77[b]
Not working	86 (57.7)	20 (52.6)	
Non-formal job	8 (5.4)	3 (7.9)	
Formal job	55 (36.9)	15 (39.5)	
Gravida (%)			0.99[b]
Primigravida	62 (42.4)	16 (42.1)	
Multigravida	85 (57.8)	22 (57.9)	
Trimester (%)			0.18[b]
First	22 (14.8)	3 (7.9)	
Second	61 (40.9)	12 (31.6)	
Third	66 (44.3)	23 (60.5)	
BMI Pre-pregnancy (SD)	22.8 (4.0)	21.6 (3.2)	0.11[a]

Abbreviations: SD, standard deviation; IDR, Indonesian Rupiah; BMI, body mass index.
Values are means with standard deviations in case of continuous variables and numbers with percentages in case of frequencies.
[#]Estimated family income.
*Low education, completed elementary and junior high school; medium, completed senior high school; High, completed university or above.
[a]Independent samples t-test
[b]Chi-square test

In **Table 2**, the results were found to be statistically significant when cross-tabulated with certain motivations and predictors. All of the women who did not partake in Ramadan fasting were aware of their pregnancy during that time. The non-fasted group was proportionately more concerned that fasting during Ramadan will affect the baby's health and their own health in comparison to the fasting group. For the women who did not fast, more than half of their husbands held the opinion that women should not fast. In the fasted group, 28.9% of the husbands had that point of view. In contrast, the percentage of women whose husband thought they should fast was higher in the fasted group (14.8%) compared to the non-fasted group (10.5%). Morning sickness was as common in the fasted group as in the non-fasted group.

Table 2. Women's motivation for Ramadan fasting

Variable	Ramadan fasting		p-value[a]
	Yes n = 149	No n = 38	
Aware of pregnancy during Ramadan (%)	134 (89.9)	38 (100)	0.04
Experienced morning sickness (%)	53 (35.6)	15 (39.5)	0.66
Afraid fasting during pregnancy may affect fetal health (%)	63 (42.3)	24 (63.2)	0.02
Afraid fasting during pregnancy may affect own health (%)	32 (21.5)	14 (36.8)	0.05
Husband's opinion regarding fasting during pregnancy (%)			0.02
Women should not fast	43 (28.9)	21 (55.3)	
Women should fast	22 (14.8)	4 (10.5)	
No opinion	81 (54.4)	13 (34.2)	
Unknown	3 (2.0)	0 (0.0)	
Fasted outside of Ramadan during pregnancy (%)	19 (12.8)	1 (2.6)	0.07

Values are numbers with percentages from the frequency variables.
[a]Chi-square test

As shown by **Table 3**, several factors such as age, income, education, employment, gravida, experience of morning sickness, and fasting during pregnancy outside of Ramadan had little or no influence on whether a woman partakes in Ramadan fasting during pregnancy, neither in univariable nor in multivariable analysis. Lower gestational age was related to a greater likelihood of participating in Ramadan. Additionally, a higher pre-pregnancy BMI predicted fasting in multivariable analysis, although borderline statistically significant. Women who expressed fear that fasting during pregnancy may affect their own health or their baby's health had a lower chance of partaking in Ramadan, however, this relationship was no longer statistically significant in multivariable analysis. When the husband believed a woman should not fast during pregnancy, according to the multivariable regression, the probability that she partook in fasting decreased by more than 60%. Furthermore, in **Tables 4 and 5**, 42.1% of the women in the non-fasted group listed their husband's opinion as a motivator to not fast. In contrast, only 11.5% of the fasted group participated in Ramadan because their husband told them to do so.

Table 3. Predictors of Ramadan fasting during pregnancy

Variable	Univariable		Multivariable	
	OR (95% CI)	p-value	OR (95% CI)	p-value
Age	1.01 (0.94–1.07)	0.88	0.95 (0.85–1.06)	0.33
Gestational age in days	0.96 (0.92–1.00)	0.03	0.95 (0.89–1.00)	0.06
Income in IDR[#]				
< 2.5 million	Reference		Reference	
> 2.5 million	1.57 (0.74–3.32)	0.24	2.06 (0.76–5.59)	0.16
Refused to answer	1.41 (0.28–7.04)	0.67	4.16 (0.33–52.86)	0.27
Education*				
Low	Reference		Reference	
Medium	0.87 (0.27–2.81)	0.81	0.27 (0.05–1.57)	0.15
High	1.02 (0.28–3.77)	0.98	0.23 (0.03–1.81)	0.16
Employment				
Not working	Reference		Reference	
Non-formal job	0.62 (0.15–2.55)	0.51	1.23 (0.15–10.08)	0.85
Formal job	0.85 (0.40–1.81)	0.68	0.92 (0.32–2.68)	0.89
Gravida				
Primigravida	Reference		Reference	
Multigravida	1.00 (0.49–2.07)	0.99	0.58 (0.18–1.91)	0.37
BMI Pre-pregnancy	1.09 (0.98–1.21)	0.11	1.16 (1.01–1.33)	0.03
Aware of pregnancy during Ramadan	[a]		[a]	
Experienced morning sickness	0.85 (0.41–1.76)	0.66	0.82 (0.30–2.25)	0.70
Afraid fasting during pregnancy may affect baby's health	0.43 (0.21–0.89)	0.02	0.47 (0.15–1.48)	0.20
Afraid fasting during pregnancy may affect own health	0.47 (0.22–1.01)	0.05	0.62 (0.22–1.74)	0.37
Husband's opinion regarding fasting during pregnancy				
No opinion / Unknown	Reference		Reference	
Women should not fast	0.32 (0.15–0.69)	<0.01	0.27 (0.09–0.73)	0.01
Women should fast	0.85 (0.25–2.87)	0.80	0.78 (0.18–3.31)	0.74
Also fasted outside of Ramadan (during pregnancy)	5.41 (0.70–41.75)	0.11	[a]	

Abbreviations: OR, odds ratio; CI, confidence interval; IDR, Indonesian Rupiah; BMI, body mass index. All results are (univariable or multivariable) logistic regression coefficients with 95% confidence intervals.
[#]Estimated family income. *Low education, completed elementary and junior high school; medium, completed senior high school; High, completed university or above.
[a]Can not be analyzed because there were too few cases in the fasted or non-fasted women

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The fasted and non-fasted group answered separate questions regarding their motivations to fast or not to fast. These additional motivations are described in **Tables 4 and 5**. The majority of the fasted group indicated that they believed they were exempted from fasting but wanted to try it anyway. In addition, approximately half of the women who participated in Ramadan fasting had the perception that fasting during pregnancy is not harmful. Among the women who did not fast during pregnancy, perceived difficulty was the most frequent motivator. None of these women indicated that they did not fast because of the exemption granted to pregnant women. Few women acknowledged that their decisions were influenced by their friends and family or by the fear that fasting would affect their own health.

Table 4. Motivations to perform Ramadan fasting among the fasted pregnant women

Variable	Frequency n = 149
According to Islam, pregnant women are obliged to fast (%)	39 (26.2)
According to Islam, I do not have to fast but I want to anyway (%)	91 (61.5)
Ramadan fasting during pregnancy is not harmful (%)	77 (52.0)
Ramadan fasting during pregnancy is healthy (%)	73 (49.3)
Because my husband told me to fast (%)	17 (11.5)
Because I do not want to have to make up the fasting later (%)	23 (15.5)
Because I want to share spiritual and social experiences with my family (%)	72 (48.6)
Because I used to fast during Ramadan (%)	84 (56.8)
Because I did not know I was pregnant during Ramadan (%)	16 (10.8)
Because I also did Ramadan fasting on my previous pregnancy(ies) and there is no problem at all (%)	27 (18.2)
Because my midwife and doctor told me that Ramadan fasting during pregnancy is not harmful (%)	25 (16.9)

Values are numbers with percentages from the frequency variables.

Table 5. Motivations not to perform Ramadan fasting among the non-fasted pregnant women

Variable	Frequency n = 38
Fasting is too difficult to be done during pregnancy (%)	32 (84.2)
I am not used to fasting (also when I was not pregnant) (%)	0 (0.0)
Because I am afraid that it will affect my baby's health (%)	16 (42.1)
Because I am afraid that it will affect my own health (%)	6 (15.8)
Because my husband told me not to fast (%)	16 (42.1)
Because my family and friends suggest me not to fast (%)	2 (5.3)
Because my midwife and doctor told me not to fast (%)	12 (31.6)
Because according to Islam, pregnant women do not have to fast (%)	0 (0.0)
Because I have other illness or pregnancy complications (%)	5 (13.2)

Values are numbers with percentages from the frequency variables.

Discussion

Overview results from the study

The findings in this study among pregnant Indonesian women suggest that adherence to Ramadan fasting was associated with earlier gestational age during Ramadan and higher pre-pregnancy BMI. Non-participation to Ramadan fasting was associated with opposition from husbands and with women's expressed fear of adverse health effects on own or fetal health.

Strengths & Limitations

This study is among the first to investigate motivational predictors of pregnant women's adherence to Ramadan fasting. The study population, which comprised of Indonesian women living in an urban area (Jakarta), was diverse in terms of ethnic group and socioeconomic background. Women's characteristics were based on medical records while motivational factors of Ramadan fasting were collected through individual interview to assure confidentiality and women's freedom in giving response to each question. The interviews were performed during Ramadan, when the women were still pregnant to avoid possible recall bias. Data on the number of fasted days were collected after Ramadan thereby allowing for gathering of the accurate number of days fasted by the women. However, in the analysis, we classified women who fasted for at least a day during Ramadan as part of the fasted group. We are aware that the effect estimate of association between predictors and Ramadan fasting may vary slightly by this classification. We ran the logistic regression with 10 days of fasting as the cutoff and the same results were found.

To our knowledge, studies investigating motivational predictors of Ramadan are scarce. Currently, studies had focused on women's knowledge of the exemption in Islamic law and their perceived harm of fasting during pregnancy, mainly physiological factors that influence women's decision to fast.[12, 13] Social, religious and spiritual context are key in discussing the medical implication of fasting in pregnant women. The inquiry regarding motivational factors during pregnancy is thus a strength of this study. Many studies do not take into account the different support networks of the family such as the husband, family and friends.

Interpretation of the results

In the present study, 80% of pregnant women fasted at least one day during Ramadan. The proportion of women who fasted a full month, almost a full month (21-28 days), half a month (11-20 days) and a few days (1-10 days) were 14%, 16%, 19%, and 30% respectively. Similarly, in a study of Singaporean women, 87% of pregnant women fasted at least one day during Ramadan. [13] In another study, 88% of Pakistani women fasted during their pregnancies. [12] In contrast to our study, the majority of the fasted groups fasted for a whole month in both of these studies (33% and 42.5% respectively).[12 13]

Our finding showed that women in earlier gestational age during Ramadan were more likely to fast. This is supported by several other studies which reported higher adherence to Ramadan fasting among women who were in the first trimester, as compared to those in the second or third trimester.[9, 14] Although another study [15] found a similar adherence of Ramadan in women from various trimester of pregnancy. The higher rate of fasting adherence in women in earlier gestational age could partly be explained by the fact that some women were not aware of their pregnancy when they performed Ramadan fasting or that they found out about their pregnancy later during Ramadan. Furthermore, perhaps women who were still in the early stage of pregnancy perceived the burden as less severe or are less fearful of harmful health affects than women in a later stage of their pregnancy. Morning sickness, which is common in these women, could also provide some ease to perform fasting, since they do not wish to eat anyway. However, severe morning sickness may discourage women from fasting.

In addition, our study shows that women who fasted had on average a higher pre-pregnancy BMI. This suggests that women with different BMI perceived the burden of fasting differently. In accordance to our finding, an Iranian study showed that the fasted group had a significantly higher BMI than the non-fasted group.[16] The authors suggest that women with a higher BMI may perceive the burden or pregnancy as less severe. For this reason, they may feel healthier and thus are more likely to fast than women with a lower BMI who may be more concerned about their health. In contrast to our study, there were no significant differences found between different fasted groups in terms of BMI at the beginning of pregnancy in another Iranian study.[7]

Women with higher pre-pregnancy BMI might also consider Ramadan fasting as a means for losing weight or to control their gestational weight gain. It is important to note that weight loss through/via Ramadan fasting could put a woman's unborn child at risk. Maternal weight loss during pregnancy was associated with a lower birth weight and reduced fat and lean mass in neonates.[17] In overweight and obese women, weight loss during pregnancy was associated with some improved maternal and neonatal outcomes although this effect was lessened by increased odds of SGA (small for gestational age) status and preterm delivery.[18]

Ramadan fasting has been reported to decrease women's weight,[19] although on the contrary, a study in Saudi families showed a reported weight gain after Ramadan.[20] Excess weight gain, especially in the first trimester, increases gestational diabetes mellitus (GDM) risk among women in the study. This needs more attention from health care providers as this may represent a modifiable risk factor for GDM.[21]

It is also important to recognize the role of gender in Muslim society. Our study suggests that the husband's opinion influences a woman's decision to fast. Women were more likely to not observe the fast if their husbands were opposed to it. In the fasted group, most husbands had no opinion, whereas in the non-fasted group, most husbands thought women should not fast. In one study, the majority of the husbands and family units were supportive and only 7% of spouses were against their wives fasting during pregnancy.[13] The opposition could be explained by their concern for the women's or the child's health.

Our study provides evidence of considerable fear that fasting is harmful for fetal health and mother's own health, both in the fasted and non-fasted women, although more common in the non-fasted women. This fear can be manifested as (psychological) stress in the pregnant women, which could affect pregnancy outcomes. High stress and anxiety levels in pregnant women are associated with an increased risk for spontaneous abortion, preterm labor and growth retardations in offspring.[22] Furthermore, stress during pregnancy may influence the fetal programming of brain function resulting in grave implications for the offspring. One study found that pregnancy-specific anxiety is a predictor of lower mental and motor development in infants.[23]

In our study, women's parity was not associated with women's fasting adherence. In another study, however, there was a significant relationship reported where multiparous women were reported to fast more than the nulliparous.[24] Similar to our study, employment also did not differ between the Ramadan fasted group and non-fasted group in Lebanese women.[10] Furthermore, maternal education did not differ between Iranian women who fasted and those who had not.[16]

In the absence of a consensus on health outcomes of Ramadan fasting during pregnancy, health care personnel face the difficult task of accurately counseling this group of women. The clinical and medical implications of fasting in pregnant women are complex, entrenched by the social, religious, and spiritual context in which health beliefs and practices are affected. Understanding the health beliefs, motivations, and perceptions regarding women's choice to fast, provides valuable insight for health care providers to appropriately counsel these women and address their respective medical, social, and religious needs.

Conclusion

In conclusion, earlier gestational age during Ramadan, higher pre-pregnancy BMI, and husband's opinion strongly influences women's adherence to Ramadan fasting during pregnancy. Fear of adverse health effects of Ramadan fasting is common in both fasting and non-fasting pregnant women.

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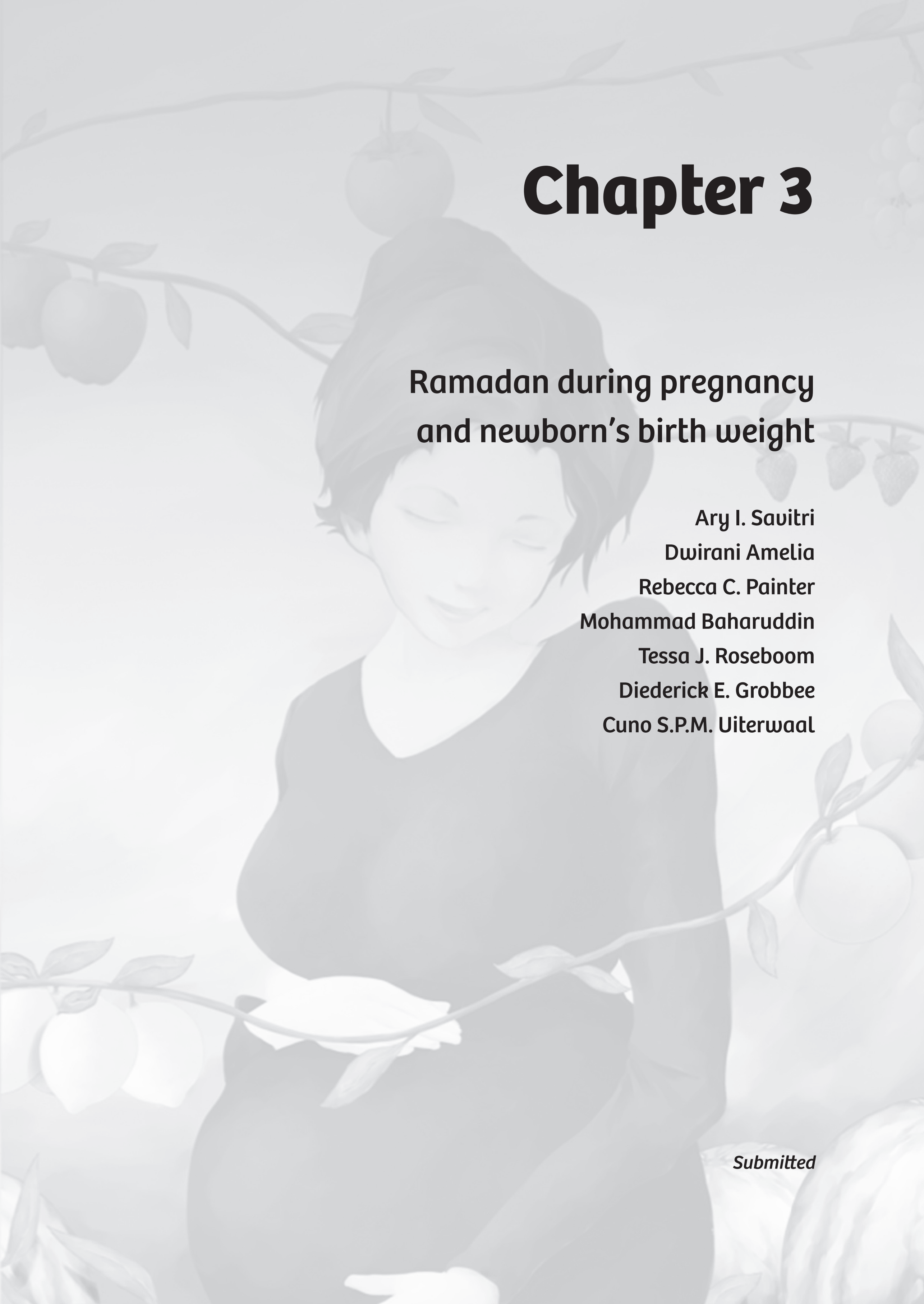

Chapter 3

Ramadan during pregnancy and newborn's birth weight

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Abstract

Background

Women's lifestyle and nutrition during pregnancy are associated with their own short and long-term health and that of their babies. Adherence to Ramadan fasting by pregnant women might affect the health of the woman and her baby. We evaluated the association between Ramadan exposure and fasting in pregnancy on the women's risk of gestational hypertension and newborns' size at birth.

Methods

We used data from 1,066 women from an on-going prospective cohort in Budi Kemuliaan Hospital, Jakarta, Indonesia, who gave informed consent at their first antenatal care visit and were followed until delivery. Based on their last menstrual period and date of delivery, women were classified as having been exposed to Ramadan in pregnancy or not. Women who came to the hospital in Ramadan in 2013 were interviewed about their fasting behavior. Dietary assessments using 24-hour dietary recall were also performed during Ramadan and one month later. Data on outcomes were retrieved from the hospital records or based on telephone interview if women had given birth elsewhere.

Results

Women who fasted had significantly lower intake of total energy, macronutrients, and water as compared to the non-fasting. Women's intakes were also lower during Ramadan (both in the fasting and non-fasting group) as compared to one month later. No difference was found with respect to women's risk of gestational hypertension. Newborn's birth weight was generally higher with Ramadan exposure during pregnancy as compared to without exposure. However, analysis within the Ramadan exposed women, showed no effect of women's fasting status on newborn's birth weight (10.18 grams, 95%CI: -167.74, 188.11, p= 0.91).

Conclusions

Life style changes that occur with Ramadan (fasting) exposure during pregnancy are associated with lower nutrition intake, however, women's risk of gestational hypertension and offspring's birth weight does not seem to be affected. Further studies focusing on other pregnancy outcomes and the offspring's long-term health are needed.

3

Introduction

Adverse fetal environment could give serious consequences on the offspring's health outcomes. Exposures such as sub-optimal maternal diet, smoking, and stress could inhibit fetal growth and development, which further cause lower birth weight and other poor birth outcomes.[1–3] Studies on the long term impact of fetal growth restriction, as represented by lighter weight at birth, showed a greater risk of later chronic diseases, including coronary heart disease, stroke, type 2 diabetes, hypertension, and other cognitive and emotional problems.[4–8]

During the month of Ramadan, various degrees of behavioral changes occur among pregnant Muslim women. Ramadan is the ninth month of the Islamic calendar. During Ramadan, daytime fasting is obligated to every adult Muslim. From the sunrise until sunset, activities such as eating, drinking, smoking, and sexual activities are prohibited.

Meal frequencies are usually reduced to only two times a day; one large meal when breaking the fast in the evening and another smaller meal during "sahoor" at dawn. Many seasonal meals which consist of sugary and fatty foods are commonly served during this month.[9–13] During this month, physical activities and sleeping pattern are also affected as people tend to be more active at nights as compared to other months. Because Ramadan is calculated based on the Lunar calendar, it shifts forward by approximately 11 days each year.[11–14]

Three of every four pregnant Muslim women are exposed to Ramadan at any stage of their pregnancies.[15] According to Islamic rule, pregnant women, together with breastfeeding mothers, ill people, and old people are exempted from the obligation to fast during Ramadan. They are permitted to postpone their fasting until after delivery. However, many women choose to fast because they do not want to have to make up fasting alone later. The proportion of fasting pregnant Muslim women varies globally, ranging from 50-70% (in Iran, The Netherlands)[16–18] to 70-90% (in England, Singapore, US, Gambia, and Yemen).[15,19–21]

Despite of its widespread adherence, evidence on the health effects of Ramadan fasting during pregnancy (both maternal and fetal) is still limited. Dietary intake and weight gain of fasting pregnant women were reported to be less than in the non-fasting,[22,23] Several metabolic alterations associated with fasting have also been reported, which include reduction in serum glucose and insulin levels[24] and elevation in serum triglyceride, cortisol, and leptin concentrations.[25,26] Several studies showed no association between Ramadan exposure during pregnancies and birth weight or risk for low birth weight.[16–18,22,26–31] On the contrary, other studies reported a lower birth weight with Ramadan exposure.[32,33] Many of these studies did not differentiate

Ramadan exposure (as overlap between pregnancy and the month of Ramadan) from actual fasting exposure, which potentially leads to dilution of effects.

Women's nutrition has been shown to determine the risk for developing gestational hypertension;[34] women with highest caloric, carbohydrates, and sodium intake had the highest risk while women with highest intake of vegetable had the lowest risk.[35–37] Studies on Dutch famine also showed that women with caloric rations during pregnancy had lower mean of systolic blood pressure near delivery.[38] In conditions where nutritional disturbance occur, such as in hyperemesis gravidarum, women's risk for preeclampsia increased.[39,40] However, there is still limited evidence on the effect of Ramadan exposure on the occurrence of gestational hypertension.

In the present study, we used data from an on-going cohort of Indonesian pregnant women to evaluate the effect of Ramadan exposure during pregnancy and maternal Ramadan fasting on women's risk of gestational hypertension and newborns' birth weight. Maternal dietary intakes were evaluated during and after Ramadan, both in the fasting and non-fasting women in an attempt to seek for possible explanatory factors.

Methods

Study population

The present study was conducted within a prospective cohort of pregnant women in Budi Kemuliaan Hospital, Jakarta, Indonesia, a private municipal hospital that specialized in maternal and child health care, training (midwives), education, and research. This cohort was built in an attempt to develop a prediction rule for gestational hypertension (including pre-eclampsia and eclampsia) in Indonesian women.

Recruitment of participants took place from July 2012 until October 2014. All pregnant women who were paying antenatal care visits at the hospital were asked to participate and, if so, provided written informed consent. Participants were examined and interviewed by the midwives according to the standard clinical care, and then followed on the subsequent antenatal care visits until delivery.

Ramadan and fasting exposure

The present study coincided with three Ramadan months, which occurred on 21 July 2012– 18 August 2012, 10 July 2013 – 7 August 2013, and 29 June – 27 July 2014. Pregnancies were classified as exposed to Ramadan if the women's last menstrual period coincided with Ramadan or if Ramadan started after the women's last menstrual period and before delivery. All pregnancies that overlapped with Ramadan in any number of days were all classified as exposed.

Women who came to the hospital during the Ramadan month in 2013 were interviewed about their adherence to Ramadan fasting during their pregnancy. They were also asked about factors that influenced their decision either to adhere to Ramadan fasting or not. In addition, women's dietary intakes were assessed by a nutrition officer using a single 24-hour dietary recall. This assessment was done irrespective of their fasting status and was repeated one month later. Twenty-four hour dietary recall is a retrospective method of dietary assessment in which every individual is interviewed about his or her food and beverage consumption during the previous day or 24 hours. A computerized data analysis system (Nutrisurvey, 2007, Indonesian version) was used to convert food intake into nutrient intake based on the portion sizes, preparation of recipes, and food tables. The system is a translation from a German nutrition software (EBISpro) which has been adapted to local food tables.

Outcome measurement

The occurrence of gestational hypertension and newborn's birth weight were investigated in relation to both Ramadan exposure and fasting exposure. Data on the occurrence of hypertensive disorder and birth weight were retrieved from the medical records or retrieved by phone call to the women if they had given birth elsewhere. Gestational hypertension was defined as systolic blood pressure ≥ 140 mmHg and/or diastolic blood pressure ≥ 90 mmHg at two or more occasions, develops for the first time after 20 weeks of gestation.[41] Among women who were exposed to Ramadan in pregnancy, this outcome was limited to cases that were diagnosed after Ramadan had started. We were not informed about the presence or absence of gestational diabetes mellitus (GDM) in our cohort, due to an absence of screening for GDM in our cohort.

Data analysis

Maternal and babies' characteristics were tabulated by Ramadan exposure and Ramadan fasting exposure separately, for both descriptive purposes and for initial evaluation for possible confounding. We analyzed the effect of Ramadan (fasting) exposure on the outcomes using two approaches. First, we compared women who were exposed to Ramadan during pregnancy (either with fasting adherence or not) to the unexposed. Secondly, within women who were exposed to Ramadan during pregnancy, we compared the outcomes of fasting women as compared to the non-fasting.

We used multivariable linear regression for outcome of birth weight and multivariable logistic regression for outcome of gestational hypertension. Uni-variable analyses were first performed and followed by multivariable analyses for adjustment for confounders. Potential confounders were secondhand smoking exposure, family income, maternal education, pre-pregnancy BMI, gestational duration, and nulliparity. Results are expressed as linear regression coefficients with 95% CIs or OR from the logistic regression coefficients and corresponding *p* values. Statistical significance were considered to be a 2-sided p value < 0.05. All analyses were done with SPSS version 21.0 for Windows (SPSS Inc, Chicago, IL).

Results

The initial number of pregnant women in the total cohort was 1,777. One hundred twenty women withdrew their consent or experienced miscarriage (drop out), 113 women could not be contacted for their pregnancy outcomes (loss to follow up), and 334 women were still pregnant during this analysis. After excluding the women with missing data on last menstrual period and/or date of delivery ($n = 70$), miscoded gestational durations ($n = 48$), and twin pregnancies ($n = 26$), the cohort included 1,066 women for analysis. The flowchart of study population is shown in **Figure 1**.

The baseline characteristics of women based on their exposure to Ramadan during their pregnancy and their fasting status is shown in **Table 1**. The majority of the women were exposed to Ramadan during their pregnancy (85.0%). Women who were exposed to Ramadan had higher pre-pregnancy BMI and less weight gain during pregnancy as compared to the unexposed, although not statistically significant. Gestational duration was slightly longer in the exposed group and preterm birth was less common. The exposed and unexposed women were similar with respect to age, education, family income, secondhand smoking exposure, nulliparity, and babies' sex.

There were 171 women who came to the hospital during Ramadan in 2013, to whom interview about Ramadan fasting adherence was conducted. As compared to other Ramadan exposed women who did not come to the hospital, these women were in later stage of their gestation (26.0 weeks vs 20.3 weeks [$p<0.01$]). There were 135 (78.9%) women who fasted to some extent during Ramadan and 36 (21.1%) women did not fast in any day. Among women who fasted, the median number of days fasted was 14.00 days. Fasting women had a slightly higher prepregnancy BMI, higher family income, and were less likely to deliver preterm babies, although not statistically significantly. Gestational weight gain were significantly less in the fasting women than in the non-fasting. The two groups were not different with respect to age, education, secondhand smoking exposure, nulliparity, gestational durations, and babies' sex.

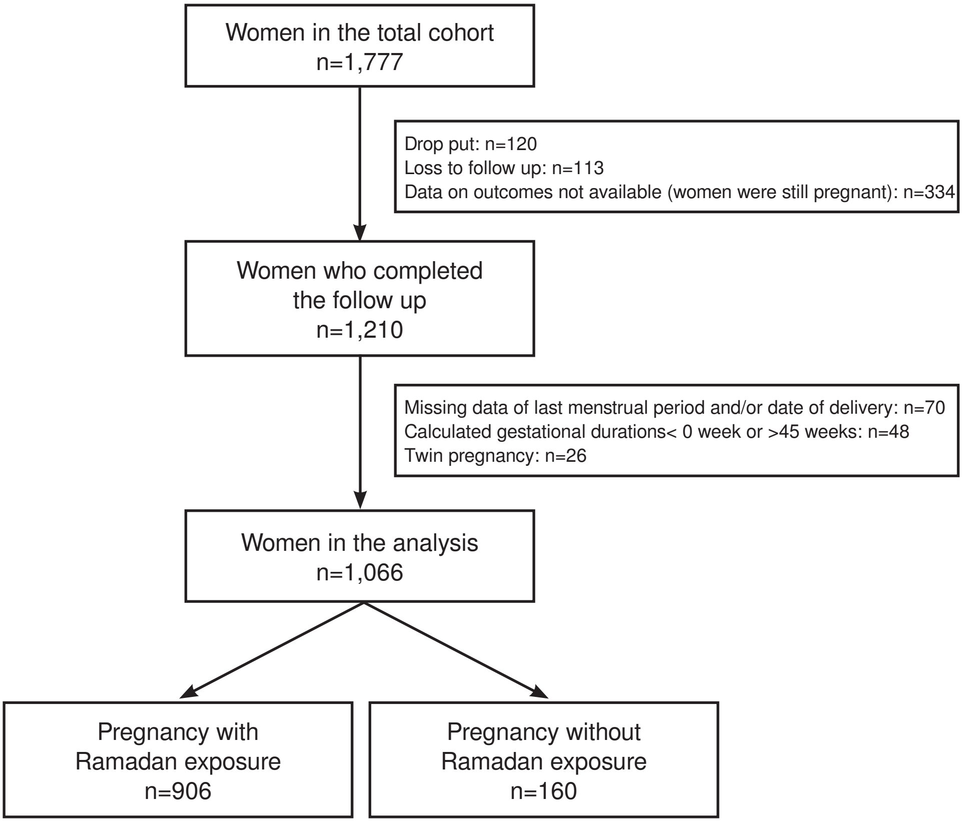

Figure 1. Overview of the study population

During Ramadan in 2013, we performed dietary assessment among 96 women out of 171 women who came to the hospital for antenatal care visits. There were 42 women who were fasting and 54 women who were not fasting at the day of the interview. The median of gestational age was not different between the fasting and non-fasting group (33 weeks vs 30 weeks, p=0.46). **Table 2** shows the comparison of intake between these two groups. Women who were fasting had significantly lower percentage intakes of energy from protein. They also, in general, had lower intakes of carbohydrates, protein, total energy, water, vitamin A, sodium and potassium, although none were statistically significant. There was no difference in consumption of fat, dietary fiber, PUFA, cholesterol, carotene, vitamin E, vitamin B1, vitamin B2, vitamin B6, folic acid, vitamin C, calcium, magnesium, phosphorus, iron, zinc, as well as percentage of energy from fat and carbohydrates as compared to women who were fasting that day.

3

Table 1. Baseline characteristics of women based on Ramadan exposure and their fasting status

	Ramadan exposure			Ramadan fasting exposure		
	Yes	No	p-value	Fasting	Non-fasting	p-value
Number	906 (85.0)	160 (15.0)		135 (78.9)	36 (21.1)	
Maternal age	28.9 ± 0.20	29.1 ± 0.44	0.82[a]	29.2 ± 0.52	29.1 ± 0.96	0.91[a]
Maternal pre-pregnancy BMI#	22.2 (16.6-27.8)	21.6 (15.7-27.5)	0.07[b]	22.9 (17.04-28.76)	21.3 (17.98-24.62)	0.09[b]
Maternal weight gain (in kg)	12.5 ± 0.33	13.7 ± 0.74	0.13[a]	11.7 ± 0.82	16.9 ±1.37	0.002[c]
Maternal education			0.39[c]			0.92[d]
Elementary or Junior High School	170 (18.8)	24 (15.0)		13 (9.7)	4 (11.1)	
Senior high school	560 (62.0)	100 (62.5)		85 (63.4)	23 (63.9)	
University	173 (19.2)	36 (22.5)		36 (26.9)	9 (25.0)	
Family income			0.89[c]			0.08[d]
Less than 1 million IDR	85 (9.6)	13 (8.2)		5 (3.8)	6 (17.1)	
1-2.5 million IDR	412 (46.4)	77 (48.7)		56 (42.1)	15 (42.9)	
2.5-5 million IDR	279 (31.5)	50 (31.6)		54 (40.6)	10 (28.6)	
More than 5 million IDR	54 (6.1)	7 (4.4)		10 (7.5)	3 (8.6)	
Secondhand smoking exposure			0.47[c]			0.86[d]
Yes, everyday	142 (46.7)	34 (54.8)		18 (42.9)	4 (57.1)	
Yes, sometimes	34 (11.2)	5 (8.1)		5 (11.9)	0 (0.0)	
No	128 (42.1)	23 (37.1)		19 (45.2)	3 (42.9)	
Nulliparity	291 (42.9)	59 (45.0)	0.65[c]	59 (44.4)	17 (47.2)	0.76[c]
Gestational duration (days)#	275.0 (260.0-290.0)	271.0 (254.5-287.5)	0.001[b]	276.0 (262.3-289.8)	274.0 (258.0-290.0)	0.31[b]
Preterm birth*	57 (6.4)	18 (11.3)	0.03[c]	0 (0.0)	3 (8.3)	0.09[d]
Baby's sex (boy)	457 (50.8)	90 (56.3)	0.20[c]	67 (50.0)	19 (52.8)	0.77[c]

Values are means with standard errors in case of continuous variables and percentages in case of frequencies. In case of skewed data (#), medians with interquartile range were presented.#Not normally distributed, *Preterm birth is defined as the birth of baby at less than 37 weeks of gestational age.
[a] Independent sample T-test , [b] Mann-Whitney Test, [c] Chi square test, [d] Fisher's exact test

Table 2. Maternal dietary intake in one Ramadan day based on their fasting status at the day of interview

	Fasting (n = 42)		Non-fasting (n= 54)		p-value
	Mean value	**Std. Error Mean**	**Mean value**	**Std. Error Mean**	
Total energy (Kcal)	1646.1	60.5	1831.7	80.8	0.07
Water (g)	1870.2	92.5	1940.1	114.3	0.64
Protein (g)[#]	50.9	25.7	63.6	39.13	0.07
Fat (g)[#]	55.5	48.3	58.1	48.0	0.53
Carbohydrates (g)	215.2	9.3	232.8	9.9	0.21
Dietary fiber (g)[#]	10.4	7.2	9.8	5.7	0.62
PUFA (g)[#]	11.2	15.6	8.9	15.0	0.55
Cholesterol (mg)[#]	227.5	266.3	217.4	264.5	0.30
Vit A (mg)[#]	1076.2	1244.2	1377.0	2093.9	0.16
Carotene (mg)[#]	0.0	245.4	0.0	398.2	0.47
Vit E (mg)[#]	0.0	3.6	0.0	2.4	0.94
Vit B1 (mg)[#]	0.6	0.6	0.7	0.6	0.81
Vit B2 (mg)[#]	0.9	0.6	1.1	0.92	0.63
Vit B6 (mg)[#]	1.2	0.9	1.2	1.0	0.66
Folic acid (ug)[#]	0.0	339.4	0.8	260.8	0.67
Vit C (mg)[#]	63.6	87.5	76.0	114.6	0.60
Sodium (mg)[#]	384.2	627.8	632.3	911.0	0.14
Potassium (mg)	1793.2	142.6	1906.9	104.3	0.51
Calcium (mg)[#]	588.6	507.8	571.4	569.9	0.96
Magnesium (mg)	255.6	17.4	264.5	15.5	0.70
Phosphorus (mg)[#]	843.2	472.4	937.7	865.7	0.45
Iron (mg)[#]	10.4	13.1	11.5	12.4	0.98
Zinc (mg)	7.2	0.4	8.3	0.5	0.14
Protein (% TE)	12.9	0.6	15.0	0.4	< 0.01
Fat (% TE)	29.3	2.5	29.8	1.9	0.86
Carbohydrates (% TE)	52.2	1.3	51.6	1.1	0.73

PUFA, Poly-unsaturated fatty acid; % TE, percentage of total energy
Results are based on estimation using 24-hour nutrition recall.
[#]Not normally distributed
In case of skewed data ([#]), medians with interquartile range were presented instead of means and standard errors.
p-values are based on Student's t-test or Mann-Whitney U-test in case of skewed data.

In **Table 3**, we compared dietary intake of the fasting women during Ramadan with one month after Ramadan. There were 15 women who could be re-interviewed for this purpose. During Ramadan, fasting women consumed significantly less water, fat, and vitamin A, and had lower percentage intakes of energy from carbohydrates. Water and fat intakes were significantly lower than one month later by on average 570 and 16 grams while carbohydrate intake was approximately 50 grams less. Intake of total energy, carbohydrates, vitamin C, sodium, calcium, and phosphorus, as well as percentage of energy from protein, and fat was not significantly different.

Table 3. Maternal dietary intake in one Ramadan day as compared to one month after Ramadan within fasting mothers

	During Ramadan (n=15)		After Ramadan (n=15)		p-value
	Mean value	**Std. Error Mean**	**Mean value**	**Std. Error Mean**	
Total energy (Kcal)	1527.3	103.2	1744.1	165.3	0.22
Water (g)	1686.5	110.9	2255.3	152.6	< 0.01
Protein (g)	52.5	5.0	54.8	5.0	0.69
Fat (g)#	42.0	63.3	58.1	34.2	0.02
Carbohydrates (g)	196.3	16.4	246.9	26.0	0.07
Dietary fiber (g)	10.4	0.9	10.1	1.2	0.80
PUFA (g)#	11.6	12.0	7.7	8.1	0.41
Cholesterol (mg)	234.7	45.3	228.8	40.5	0.93
Vit A (mg)#	1049.8	1224.3	2011.5	1581.7	0.02
Carotene (mg)#	0.0	356.3	0.0	0.0	0.06
Vitamin E (mg)#	0.0	2.4	0.0	0.3	0.51
Vitamin B1 (mg)#	0.6	0.4	0.5	0.4	0.87
Vitamin B2 (mg)	0.8	0.10	0.8	0.14	0.82
Vitamin B6 (mg)	1.2	0.14	1.2	0.16	0.97
Folic acid (ug)#	0.0	210.0	0.0	6.5	0.95
Vitamin C (mg)	62.4	12.0	78.7	16.2	0.40
Sodium (mg)#	383.9	644.9	285.9	906.8	0.78
Potassium (mg)	1583.4	153.7	1589.0	191.1	0.98
Calcium (mg)	485.9	71.7	378.9	71.5	0.26
Magnesium (mg)	240.1	25.6	222.6	25.2	0.59
Phosphorus (mg)#	835.2	448.4	646.5	321.4	0.39
Iron (mg)#	8.2	12.4	8.0	4.4	0.23
Zinc (mg)	6.3	0.6	6.5	0.6	0.81
Protein (% TE)	13.9	1.1	13.0	0.7	0.49
Fat (% TE)#	29.8	38.1	29.5	7.9	0.09
Carbohydrates (% TE)	50.8	2.3	56.4	1.5	0.01

PUFA, Poly-unsaturated fatty acid; % TE, percentage of total energy
Results are based on estimation using 24-hour nutrition recall.
#Not normally distributed
In case of skewed data (#), medians with interquartile range were presented instead of means and standard errors.
p-values are based on paired sample t-test or Wilcoxon signed ranks test in case of skewed data.

In **Table 4**, we compared dietary intake of the non-fasting women in one day of Ramadan with one month later. There were 27 women who were accessible for this analysis. As compared to one month later, these women consumed significantly less water and had lower percentage intakes of energy from protein. Intakes of all other nutrients were similar during Ramadan as compared to one month later.

Table 4. Maternal dietary intake in one Ramadan day as compared to one month later in women who were not fasting

	During Ramadan (n=27)		After Ramadan (n=27)		p-value
	Mean value	Std. Error Mean	Mean value	Std. Error Mean	
Total energy (Kcal)	1888.3	132.7	1986.9	112.4	0.56
Water (g)	1965.7	179.1	2462.8	90.6	< 0.01
Protein (g)#	68.7	56.7	58.9	26.6	0.26
Fat (g)	62.2	7.8	69.2	4.7	0.44
Carbohydrates (g)	245.6	16.5	281.7	18.2	0.10
Dietary fiber (g)#	9.0	7.7	9.8	8.0	0.48
PUFA (g)#	9.6	16.6	11.0	10.2	0.98
Cholesterol (mg)#	234.4	274.3	247.6	203.8	0.55
Vitamin A (mg)#	1743.9	2573.6	2442.4	1541.6	0.52
Carotene (mg)#	0.0	8.0	0.0	0.0	< 0.01
Vitamin E (mg)#	0.5	3.6	0.1	3.1	0.79
Vitamin B1 (mg)#	0.8	0.8	0.9	0.7	0.87
Vitamin B2 (mg)#	1.2	1.2	1.1	0.6	0.51
Vitamin B6 (mg)#	1.6	1.1	1.7	1.2	0.80
Folic acid (ug)#	2.8	351.2	4.0	350.0	0.82
Vitamin C (mg)#	58.7	118.3	73.7	83.5	0.60
Sodium (mg)#	560.4	580.0	533.8	672.7	0.85
Potassium (mg)	1978.2	165.3	1839.3	164.3	0.45
Calcium (mg)#	530.6	659.7	620.6	753.1	0.88
Magnesium (mg)	282.7	25.3	279.2	20.0	0.91
Phosphorus (mg)#	1151.9	893.0	958.9	498.5	0.14
Iron (mg)#	14.3	12.7	10.4	10.3	0.06
Zinc (mg)#	9.7	8.2	7.8	4.7	0.34
Protein (% TE)	15.4	0.6	13.0	0.6	0.01
Fat (% TE)#	28.2	11.9	30.7	8.9	0.41
Carbohydrates (% TE)	52.8	1.8	56.2	1.5	0.14

PUFA, Poly-unsaturated fatty acid; % TE, percentage of total energy
Results are based on estimation using 24-hour nutrition recall.
#Not normally distributed
In case of skewed data (#), medians with interquartile range were presented instead of means and standard errors.
p-values are based on paired sample t-test or Wilcoxon signed ranks test in case of skewed data.

Table 5a shows the associations between Ramadan exposure and Ramadan fasting during pregnancy with the occurrence of gestational hypertension. Pregnancies were categorized into three exposure groups; unexposed (n = 155), Ramadan fasting exposed (n = 135), or Ramadan exposed but unexposed to fasting (n = 36). The unexposed pregnancies consisted of all pregnancies in the total cohort that did not overlap with Ramadan and were taken as the reference group. Women who were exposed to Ramadan fasting had no different risk for gestational hypertension as compared to the reference group, neither in the crude nor in the adjusted model. Women who were exposed to Ramadan but unexposed to fasting also had similar risk for hypertensive disorders during pregnancies as compared to the reference group.

Furthermore, within women who were exposed to Ramadan during pregnancy, the risk of gestational hypertension was compared between the fasting and non-fasting women (**Table 5b**). The risk of gestational hypertension was similar in both groups, both in the crude and adjusted model. We summarized the effect estimates presented in **Table 5a** and **Table 5b** in **Figure 2**.

Table 5a. Associations between Ramadan exposure and maternal fasting during pregnancy with the occurrence of gestational hypertension

	N	**Gestational hypertension**		**OR (95% CI)**		
		Yes (*N*)	**No (*N*)**	**Crude**	**Model 1**	**Model 2**
Unexposed	155	33	122	Reference	Reference	Reference
Ramadan fasting exposed	135	21	114	1.47 (0.80, 2.69)	1.38 (0.75, 2.53)	1.43 (0.74, 2.75)
Ramadan exposed but unexposed to fasting	36	8	28	0.95 (0.84, 1.99)	0.95 (0.39, 2.30)	1.00 (0.38, 2.61)

OR, odds ratio
Results are logistic regression coefficients for newborns birth weight with exposure categories.
Model 1 is adjusted for smoking exposure, family income categories, and maternal education categories.
Model 2 is adjusted for as model 1 and for pre-pregnancy BMI and nulliparity.

Table 5b. Associations between maternal Ramadan fasting during pregnancy with the occurrence of gestational hypertension

	N	**Gestational hypertension**		**OR (95% CI)**		
		Yes (*N*)	**No (*N*)**	**Crude**	**Model 1**	**Model 2**
Ramadan exposed but unexposed to fasting	36	8	28	Reference	Reference	Reference
Ramadan fasting exposed	135	21	114	0.64 (0.26, 1.61)	0.80 (0.29, 2.18)	0.96 (0.33, 2.79)

OR, odds ratio
Results are logistic regression coefficients for newborns birth weight with exposure categories.
Model 1 is adjusted for smoking exposure, family income categories, and maternal education categories.
Model 2 is adjusted for as model 1 and for pre-pregnancy BMI and nulliparity.

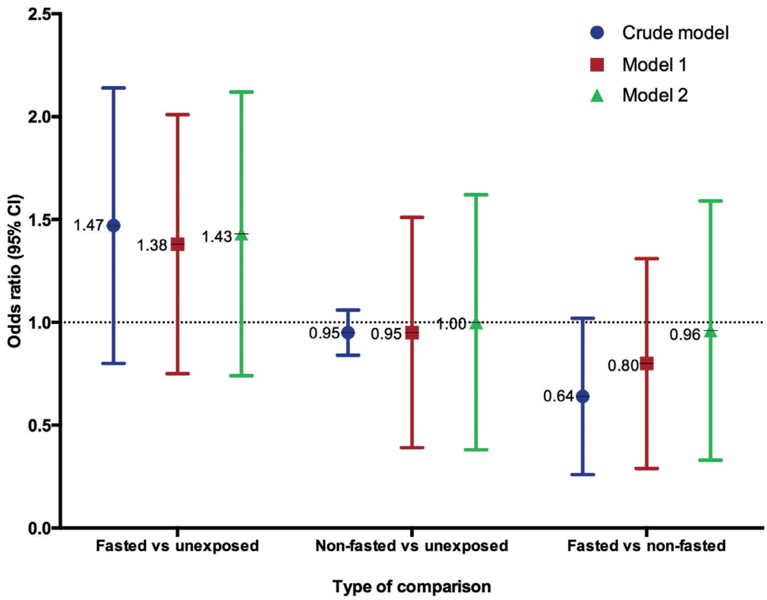

Figure 2. Associations between Ramadan (fasting) during pregnancy and gestational hypertension
CI, confidence interval.
Results are logistic regression coefficients for newborn's birth weight
Model 1: adjusted for smoking exposure, family income, and maternal education
Model 2: adjusted for Model 1 and for pre-pregnancy BMI, gestational duration and parity

In **Table 6a**, the association is shown between Ramadan exposure and Ramadan fasting during pregnancy with newborn's birth weight. As compared to the reference group, babies from women who were exposed to Ramadan fasting during their pregnancies were 205 grams heavier in the crude analysis. The association changed into 202 grams heavier after adjustment for smoking exposure, family income, and maternal education (model 1), and finally attenuated to 128 grams heavier in the fully adjusted model (model 2). Babies whose mothers were exposed to Ramadan but unexposed to fasting during their pregnancies were 178 grams heavier in the crude analysis and after adjustment for potential confounders in model 1 and model 2, these babies were consecutively 178 and 198 grams heavier than the reference group.

Within women who were exposed to Ramadan during their pregnancy, comparison of their newborn's birth weight was made according to the fasting status. (**Table 6b**) In the crude analysis, birth weight of newborns of the fasted women was similar to the birth weight of newborns of women who did not fast. This remained unaltered after adjustment for potential confounders in model 1 and model 2. We summarized the effect estimates presented in **Table 6a** and **Table 6b** in **Figure 3**.

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Table 6a. Associations between Ramadan exposure and maternal fasting during pregnancy with newborn's birth weight

	N	Regression coefficients (95% CI)		
		Crude	Model 1	Model 2
Unexposed	155	Reference	Reference	Reference
Ramadan fasting exposed	135	205.1 (90.1, 320.2)#	201.6 (86.4, 316.9)#	127.8 (8.3, 247.2)*
Ramadan exposed but unexposed to fasting	36	178.5 (-1.4, 358.3)	177.7 (-2.2, 357.6)	197.7 (22.1, 373.3)*

CI, confidence interval
Results are linear regression coefficients for newborns birth weight with exposure categories.
Model 1 is adjusted for smoking exposure, family income categories, and maternal education categories.
Model 2 is adjusted for as model 1 and for pre-pregnancy BMI, gestational duration in days, and nulliparity.
*$p < 0.05$, #$p < 0.01$.

Table 6b. Associations between maternal Ramadan fasting during pregnancy and newborn's birth weight

	N	Regression coefficients (95% CI)		
		Crude	Model 1	Model 2
Ramadan exposed but unexposed to fasting	36	Reference	Reference	Reference
Ramadan fasting exposed	135	26.7 (-132.8, 186.2)	25.2 (-137.9, 188.3)	10.2 (-167.7, 188.1)

CI, confidence interval
Results are linear regression coefficients for newborns birth weight with exposure categories.
Model 1 is adjusted for smoking exposure, family income categories, and maternal education categories.
Model 2 is adjusted for as model 1 and for pre-pregnancy BMI, gestational duration in days, and nulliparity.

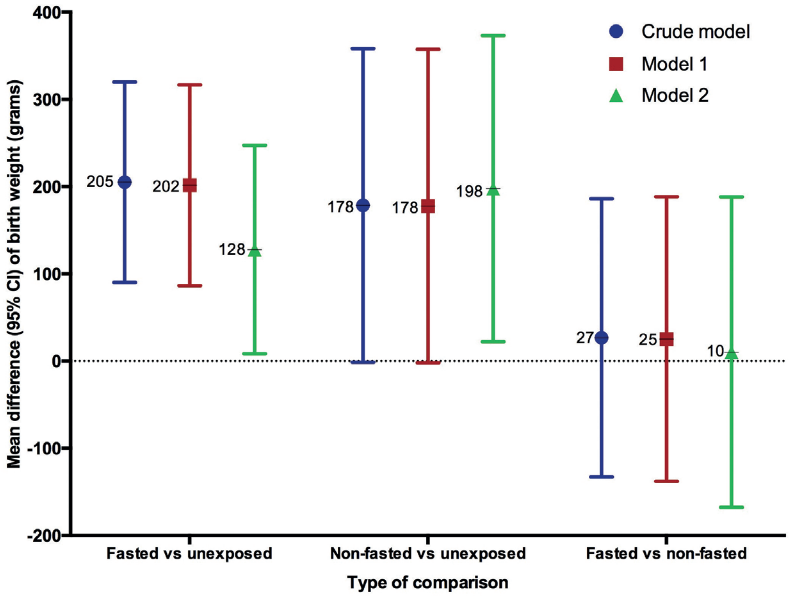

Figure 3. Mean difference of newborn's birth weight according to maternal Ramadan (fasting) exposure
CI, confidence interval.
Results are logistic regression coefficients for newborn's birth weight
Model 1: adjusted for smoking exposure, family income, and maternal education
Model 2: adjusted for Model 1 and for pre-pregnancy BMI, gestational duration and parity

Discussion

Approximately 85% of all women in our study had Ramadan exposure in any time during their pregnancies and about 80% of them fasted to some extent during the month. In the present study, we showed that women who fasted had significantly lower intake of total energy, macronutrients, and water as compared to women who did not fast. Women's intake were also less during Ramadan as compared to one month later, both in the fasted and non-fasted group. No association was found between Ramadan exposure (and maternal fasting) during the gestational period and the occurrence of gestational hypertension. Ramadan exposure is associated with higher birth weight, irrespective of maternal fasting status, as shown with consecutively 130 grams and 200 grams higher birth weight in babies from the fasting and non-fasting women as compared to non-Ramadan exposed. However, in the analysis within the Ramadan exposed pregnancies, birth weights of babies of the fasting women appeared similar to the babies of non-fasting women.

To the best of our knowledge, we are the first to study the effect of Ramadan exposure on birth weight and gestational hypertension by looking both on the effects of the Ramadan exposure and maternal fasting behavior while taking women with no Ramadan exposure as the reference. Furthermore, we are also among the first to evaluate women's dietary intake during and after Ramadan, both in the fasting and non-fasting group.

There were some limitations of the present study. Firstly, interviews about women's fasting status were limited only to women who came to the hospital during Ramadan in year 2013 and these women were in a later stage of their gestation as compared to women who were not accessible for interviews. Furthermore, within women that we interviewed about their fasting behavior, women with available data on nutritional intake were at later gestational age (28.9 vs 20.9 weeks, $p<0.01$) and had lower pre-pregnancy BMI (22.2 vs 23.2 kg/m^2, $p=0.05$) as compared to women without nutritional data. The difference in the gestational age could be caused by the fact that women at later gestational age had more visits to the hospital, while we can only speculate about the difference in BMI. The use of 24-hour dietary recall as nutritional assessment may give some disadvantages since it relies on subject's memory, cooperation, and communication ability. Finally, a relatively small number of subjects is a limitation of our study.

Our dietary assessments showed that women who were fasting during Ramadan in general had lower intake of energy and macronutrients (mainly carbohydrates and protein,) as well as water, vitamin A, sodium, and potassium as compared to the non-fasting women. Fasting women also had lower percentage of energy from protein as compared to the non-fasting women. The differences in women's intakes were unlikely caused by the difference in their gestational age since women in both groups had similar gestational age at the interview. This result is in accordance with findings from previous studies on dietary intake of women during Ramadan which found significantly less energy intake among women who fasted as compared to those who did not,[22] and less energy intake during Ramadan as compared to outside Ramadan.[42] In contrast, our result is different from a study conducted in Saudi families in whom weight gains were reported after Ramadan due to higher consumption of carbohydrates and fat rich food during Ramadan.[9]

Ramadan also appeared to influence dietary intakes of women both in the fasting and non-fasting group. It was shown by lower intakes of total energy, carbohydrates, fat, and water during Ramadan as compared to intakes of the same women one month later, both in the fasting and non-fasting women. More substantial changes of the intake during Ramadan as compared to one month later, however, were evident among fasting women. We suggest that in the non-fasting women, dietary habits were also influenced by their fasting families; as seen by the reduction of the frequency and/or the portion size of women's meal. Furthermore, using Indonesian Estimated Average Requirement (EAR) for pregnant women, we found that women's mean intakes of protein and fat consistently exceeded the recommendation both in the fasting and non-fasting women during and after Ramadan. However, carbohydrate intakes were generally lower than recommended. The EARs for protein, fat, and carbohydrate are consecutively 43-52 g, 40-42 g, and 270-285 g.[43,44]

In the present study, we found similar risks for gestational hypertension in mothers who experienced Ramadan during their pregnancy as compared to those who did not. This finding is in accordance with several studies, which reported similar mean blood pressure among healthy (non-pregnant) men and women during and post-Ramadan.[45–47]

A higher average birth weight of newborns with Ramadan exposure during their gestation was found. This is in contrast with previous studies which reported that Ramadan exposure was not associated with newborns' birth weight.[27,48] However, within women who were exposed to Ramadan, their newborn's birth weight were similar regardless of the maternal fasting status. This is in line with several earlier studies that reported similar birth weight in babies from women who had fasted during Ramadan as compared to those who had not.[16–18,26,29,30]. On the other hand, this finding is different from our previous study conducted among pregnant women in Amsterdam (The Netherlands) from Moroccan and Turkish background.[32] We suggest that the difference in the finding can be attributed to the contrast in cultural and dietary habits as compared to Indonesian population.

Several possible confounders were included in our model as those variables were found to be associated both with the exposure and with the outcome. Secondhand smoking exposure, lower family income, and lower maternal education were slightly more common among the unexposed or the non-fasting women. A slightly higher pre-pregnancy BMI among women who were unexposed to Ramadan as compared to the exposed was most likely due to chance. Within women who were exposed to Ramadan, fasting women had a slightly higher pre-pregnancy BMI and significantly less gestational weight gain. Several maternal characteristics have been known to predict maternal fasting during Ramadan, such as pre-pregnancy BMI, parity, maternal age, and socio-economic status.[17,32,49,50]

The associations between Ramadan exposure and higher newborn's birth weight, particularly among the non-fasting women, could partially be explained by the disparity of average maternal weight gain during pregnancy. Women who were exposed to Ramadan and did not fast had the highest weight gain during pregnancy (16.9 kg ± 1.37), as compared to women who fasted (11.7 kg ± 0.82) and women who were not exposed to Ramadan (13.7 kg ± 0.74). This finding is consistent with several studies[16,22,23,51] which reported significantly lower gain in women who fasted during pregnancy as compared to women who did not. Women who were exposed to Ramadan and did not fast might have had the highest fat or sugar intake, through the consumption of seasonal meals.

We could not exclude the possibility that the finding of a higher birth weight with Ramadan exposure was due to the fact that babies with longer gestational length (and therefore higher birth weight) had higher chance to be exposed to Ramadan; hence reverse causation. Women who were exposed to Ramadan during pregnancy had on average 4 days longer gestation duration than those who were not exposed (**Table 1**). Although we had adjusted for gestational length in our main analyses, the problem might still remain.

Ramadan is a brief period of exposure that is characterized by various degrees of life style changes, which include intermittent fasting, dietary modification (including the type, frequency, and size of meals), changes in physical activity level and sleeping pattern. Since Muslims are not allowed to smoke during daylight, a significant reduction of (secondhand) smoke exposure is also expected to occur during Ramadan.[52,53] Thus, it may provide a temporarily healthier environment for mothers and fetus. Furthermore, Ramadan is followed afterwards by several days of *Eid* celebration. During this celebration, people gather with family and friends and usually have meals that have high fat and sugar contents. We suspect that *Eid* celebration, which is less likely to be experienced by women who did not have Ramadan in their pregnancy, may also contribute to the weight gain of exposed women and the higher birth weight of the baby.

Conclusion

In conclusion, life style changes that occur with Ramadan (fasting) exposure during pregnancy are associated with lower nutrition intake, however, women's risk of gestational hypertension and offspring's birth weight does not seem to be affected. Further studies focusing on other pregnancy outcomes and the offspring's long-term health are needed.

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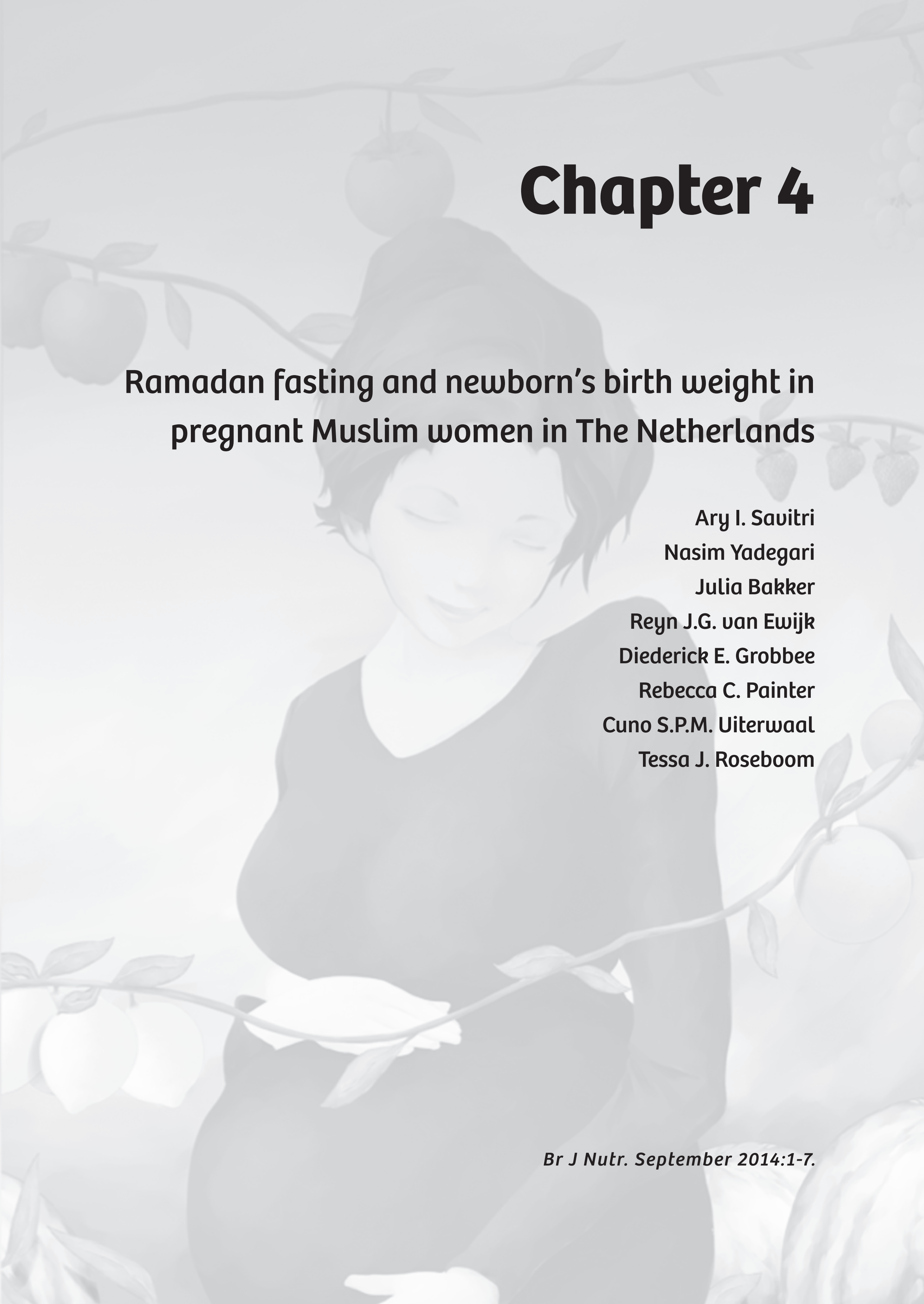

Chapter 4

Ramadan fasting and newborn's birth weight in pregnant Muslim women in The Netherlands

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Br J Nutr. September 2014:1-7.

Abstract

Background

Many Muslim women worldwide are pregnant during Ramadan and adhere to Ramadan fasting during pregnancy. We determined whether maternal adherence to Ramadan fasting during pregnancy has an impact on the birth weight of the newborn, and whether the effects differed according to trimester in which Ramadan fasting took place.

Methods

A prospective cohort study was conducted in 130 pregnant Muslim women who attended antenatal care in Amsterdam and Zaanstad, The Netherlands. Data on Ramadan fasting adherence during pregnancy and demographics were self-reported by pregnant women, and the outcome of the newborn was retrieved from medical records after delivery.

Results

Half of all the women adhered to Ramadan fasting. With strict adherence to Ramadan fasting in pregnancy, newborns' birth weight tended to be lower than with non-fasting, although this was not statistically significant (-198 g, 95% CI: -447 to 51, p=0.12). Children of mothers who fasted in the first trimester of pregnancy were lighter at birth than those whose mothers had not fasted (-272 g, 95% CI: -547 to 3, p=0.05). There were no differences in birth weight between children whose mothers had or had not fasted if Ramadan fasting had taken place later in pregnancy.

Conclusions

Our findings suggest that Ramadan fasting during early pregnancy may reduce birth weight of the newborn. These findings call for further confirmation in larger studies which should also investigate potential implications for perinatal and long-term morbidity and mortality.

Introduction

Maternal nutrition and lifestyle during pregnancy have important effects on the health and wellbeing of the offspring.[1] Poor maternal nutrition and stress during pregnancy are associated with decreased birth weight among newborns.[2-4] Low birth weight may have substantial effects on the morbidity and mortality of the newborn, both in the short and long term. Studies have shown consistent relations between low birth weight and an increased risk of chronic diseases later in life, such as hypertension, coronary heart disease, obesity, and diabetes mellitus.[5-7] Moreover, the effects on the offspring caused by altered maternal nutrition during pregnancy have been demonstrated to vary according to its timing during pregnancy.[5]

Lifestyle changes as characterized by intermittent fasting and altered eating and sleeping patterns may take place among pregnant Muslim women if Ramadan coincides with their pregnancy. Ramadan is an annual period of daytime fasting that lasts for a month. During Ramadan, Muslims refrain from food and fluid from dawn to sunset. Smoking and sexual activities are prohibited when fasting. Physical activities during the days are usually reduced, while activities during evenings and nights are increased. All healthy adult Muslims are obliged to fast. Pregnant women and breastfeeding mothers are exempted from this religious obligation if they are worried about their own health or that of their baby. However, many women still fast in order to share spiritual and social experiences with family.[8] Estimates of the proportion of pregnant Muslim women that fast for at least a day during Ramadan range from 50 - 70 % (Iran)[9-11] to 70 - 90 % (England, Singapore, US, Gambia, and Yemen),[12-14] but are unknown among many migrant communities in continental Europe.

Throughout Ramadan, modest under-nutrition occurs during the days and continues with a relatively well-nourished state during the nights. Women's body weight and body mass index (BMI; weight/height^2) are reduced during this period.[12,15,16] The degree of lifestyle changes during Ramadan could be different across countries. In many Muslim predominant countries, government often reduces the number of working hours during Ramadan and grants several days of public holiday. Some seasonal meals that comprise of foods high in fat, sugar, and salt are often served at the end of fasting. Fasting duration also varies for Muslims living in the northern and southern hemisphere, as longer summer days mean a longer fasting.

In spite of the widespread adherence to Ramadan fasting, it is still unclear whether this mode of fasting during pregnancy is related to health outcomes of the offspring. A number of studies report that maternal fasting during Ramadan has no significant effect on neonatal birth weight and fetal development.[9,10,17,18] However, most of these prospective studies were small and did not take into account potential confounders.

Two large studies conducted in Birmingham[1] and Unizah, Saudi Arabia[2] also reported no effect of Ramadan exposure on birth weight. Yet, effect estimates found in both studies may be subjected to bias since there seems to have been incomplete adjustment for confounders. Inaccuracy of exposure classification may also have been an issue in Birmingham study where distinction between Muslim and Non-Muslim Asian was only based on the first three letters of surnames in the birth data set. A large cohort study among 50,000 mothers of Arab descent in the USA demonstrated a significantly lower birth weight among Muslims who had been in utero during Ramadan. The small difference in birth weight found in this study is likely an underestimate since it also included non-Muslim women and lacked information on the degree to which women had actually fasted.[21] Furthermore, a long-term effect of Ramadan was found from a large population study in Indonesia, in which exposure to Ramadan during pregnancy led to smaller and thinner adult body size, poorer general health and increased prevalence of symptoms indicative of coronary heart problems and type 2 diabetes.[8,13]

Ramadan is clearly demarcated from the rest of the months, and allows us to study the impact of a short episode of metabolic disturbance during pregnancy on neonatal outcome. In the present study, we investigated the relation between maternal Ramadan fasting adherence, as well as its timing during pregnancy and the birth weight of newborns in a Muslim population living in a Western country.

Methods

Study design and population

A cohort study was conducted in the regions of Amsterdam and Zaanstad, The Netherlands, during Ramadan 2010. The study population comprised Muslim women who were pregnant during the month of Ramadan in 2010 (11 August until 10 September 2010), and who signed informed consent. Women with gestational age less than 4 weeks at the beginning of Ramadan and women who gave birth before the end of Ramadan were excluded from the study. In 2010, Ramadan took place during summer with approximately 13 to 15 fasting hours each day. At the start of prenatal care, women were classified into two categories based on their risk of developing complications during pregnancy. All midwifery practices, providing care for low risk pregnancies (n=34) and hospitals, providing care for high risk pregnancies (n=7) in Amsterdam and Zaanstad were asked to participate. 18 midwifery practices, 3 community hospitals, and 1 university hospital participated.

Maternal education was used to estimate socio-economic status of the mother. Women were classified into high educational level attained (finished higher vocational, college, or tertiary education), low educational level attained (finished primary school, lower vocational or secondary school), or unknown. Maternal smoking during pregnancy and during the Ramadan was classified into three categories; non-smoking, infrequent smoking, and daily smoking. Ethnicity was classified into three groups; Turkish, Moroccan, and other ethnicity.

Data collection

Data on Ramadan fasting exposure during pregnancy were collected from 13 September 2010 until 31 December 2010 by midwifery students who interviewed the women using questionnaires. In addition, demographic data were collected.

Women were asked to provide written informed consent for retrieval of their medical records after delivery. They were also asked whether they would agree to be approached in the future for long term follow-up. There were 232 eligible women who had been approached to join the study; 133 of them provided their informed consent and birth-weight data of 130 newborns were accessible from medical records. The flowchart of data collection is shown in **Figure 1**. Medical records of women were requested from the relevant obstetrician/gynaecologist to collect birth outcome.

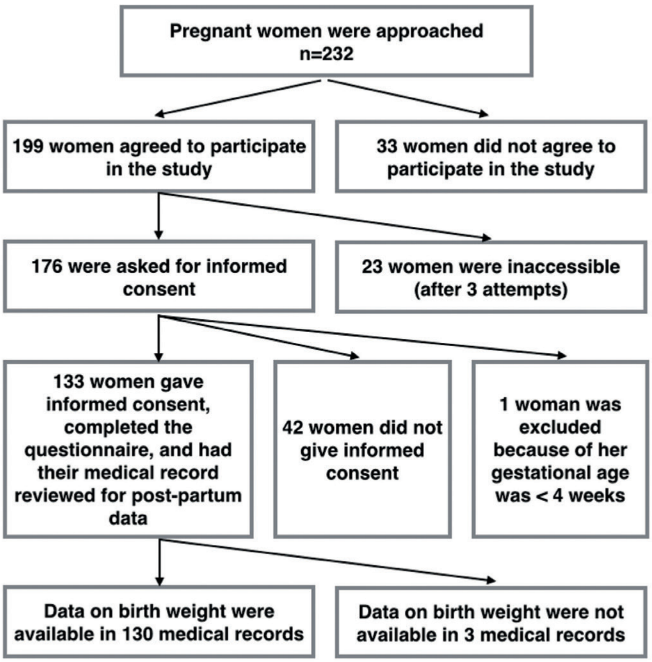

Figure 1. Flowchart of data collection

Data analysis

A multi-variable linear regression analysis was performed to determine the relation between women's adherence to Ramadan fasting during pregnancy (as independent variable) and the birth weight of the newborn (as dependent variable). Women's adherence to Ramadan fasting was classified into indicator categories; 'fasted half a month or less', or 'fasted more than half a month', with 'did not fast' (if women did not fast at any time) as the reference. To determine whether timing of fasting during pregnancy was important, we compared the trimesters in which Ramadan fasting occurred during pregnancy. Women who fasted (both fasted half a month or less and fasted more than half a month) were classified into the categories: fasted in first trimester (T1), fasted in second trimester (T2), and fasted in third trimester (T3). To evaluate if the association between adherence to maternal Ramadan fasting and newborn birth weight depended on the pregnancy trimester in which Ramadan occurred, we use the likelihood ratio test to compare models without and with interaction terms for fasting adherence and trimester.

Maternal smoking, ethnicity, first pregnancy, risk (e.g., care) classification of pregnancy, and maternal education were *a priori* considered as possible confounders.

Results are expressed as linear regression coefficients with 95% confidence intervals and corresponding *P* values. Statistical significance was considered reached at a 2-sided *P* value less than 0.05. Analyses were performed using SPSS version 20.0 for Windows (IBM Corp, Armonk, NY) and R open source software using *lm* function.

Results

General characteristics

The general characteristics of women with regard to their fasting adherence are shown in **Table 1**. Of all the study population, 70 women (53.8%) fasted to some extent, of whom 21 women (16.2%) fasted half a month or less and 49 women (37.7%) fasted more than half a month during Ramadan. The three fasting adherence groups were similar in terms of age and having been born outside The Netherlands. Women who fasted half a month or less and women who fasted more than half a month were of similar parity, educational level attained, mean gestational age at the start of Ramadan, and pregnancy risk status as compared to non-fasting women. Smoking was more common in women who did not fast and fasted half a month or less compared to those who fasted more than half a month. Moroccan women fasted more often than Turkish women. No difference was found with respect to gestational duration and gender of the babies.

Table 1. Baseline characteristics of women by fasting adherence

	Ramadan fasting adherence			p-value
	Did not fast	**Fasted half a month or less**	**Fasted more than half a month**	
Number (%)	60 (46.2)	21 (16.2)	49 (37.7)	
Mother's age	29.3 (4.2)	28.9 (4.9)	28.9 (4.7)	0.93[1]
Nulliparity (%)	31.7	23.8	24.5	0.69[2]
Ethnicity (%)				< 0.01[3]
Turkish	58.3	33.3	8.2	
Moroccan	25.0	52.4	87.8	
Others	16.7	14.3	4.1	
Born outside the Netherlands (%)	36.7	38.1	38.8	0.97[2]
Socio-economic status (%)				0.61[3]
High educational level attained	33.3	28.6	24.5	
Low educational level attained	65.0	71.4	69.4	
Unknown	1.7	0.0	6.1	
Smoking in Ramadan (%)				0.01[3]
No	85.0	85.7	100	
Infrequent	1.7	4.8	0.0	
Every day	13.3	9.5	0.0	
Smoking outside Ramadan/pregnancy (%)	21.7	14.3	2.0	< 0.01[3]
Gestational age at Ramadan (in weeks)	18.8 (8.6)	17.9 (10.1)	16.6 (8.8)	0.44[1]
Trimester at Ramadan (%)				0.36[3]
I	30.0	38.1	40.8	
II	48.3	28.6	42.9	
III	21.7	33.3	16.3	
Pregnancy risk classification (%)				0.95[3]
First line	93.3	100	93.9	
Second line	5.0	0.0	4.1	
Third line	1.7	0.0	2.0	
Gestational duration (days)	277.4 (8.8)	277.4 (9.8)	278.5 (17.4)	0.90[1]
Gender of the baby (% of female)	46.7	45.0	46.9	1.00[2]

Values are means (SD), unless otherwise indicated.
[1] ANOVA, [2] Chi-Square test, [3] Fisher's exact test.

Adherence to fasting and birth weight of the newborns

Table 2 shows associations between Ramadan fasting adherence and the birth weight of the newborn, both crude and adjusted. There were associations between Ramadan fasting and birth weight, although none were statistically significant.

Newborns of mothers who fasted half a month or less and fasted more than half a month tended to be 163.1 g heavier and 64.6 g lighter than with non-fasting. These associations respectively changed to 143.7 g and -132.4 g with adjustment for smoking and ethnicity, to 112.1 g and -169.6 g with further adjustment for nulliparity, and to 95.1 g and -198.1 g with further adjustment for pregnancy risk classification and socioeconomic factors.

Table 2. Relation between maternal adherence to Ramadan fasting in pregnancy with birth weight of newborns

	Birth weight (grams)	
	Linear regression coefficient (95% CI)	**p-value**
Model 1		
Did not fast	Reference	
Fasted half a month or less	163.1 (-109.7, 435.9)	0.24
Fasted more than half a month	-64.6 (-271.7, 142.6)	0.54
Model 2		
Did not fast	Reference	
Fasted half a month or less	143.7 (-133.9, 421.3)	0.31
Fasted more than half a month	-132.4 (-381.8, 117.0)	0.29
Model 3		
Did not fast	Reference	
Fasted half a month or less	112.1 (-160.6, 384.7)	0.42
Fasted more than half a month	-169.6 (-415.2, 76.1)	0.17
Model 4		
Did not fast	Reference	
Fasted half a month or less	95.1 (-182.0, 372.2)	0.49
Fasted more than half a month	-198.1 (-447.4, 51.2)	0.12

Linear regression coefficients indicate the difference in birth weight in grams with the reference category.
Model 1 was a crude model.
Model 2 was adjusted for mother's smoking status during Ramadan and pregnancy and mother's ethnicity.
Model 3 was adjusted as for model 2 and for nulliparity.
Model 4 was adjusted as for model 3 and for risk classification of pregnancy at the start of prenatal care and for socioeconomic factors (mother's education)

Timing of fasting during pregnancy and birth weight

A comparison between the trimesters in which Ramadan occurred during pregnancy with respect to the birth weight of newborns is shown in **Table 3**. The newborns of women who fasted in their first trimester were on average 171 grams lighter (95% CI: -415 to 74.5, p=0.17) compared to the non-fasting (reference group). Adjustment for maternal smoking status and ethnicity, parity, pregnancy risk classification, and socio-economic factors, resulted in a lower birth weight of 272 in newborns of women who fasted in their first trimester (95% CI: -547 to 3, p = 0.05). There were no associations found with respect to birth weight in newborns of women who fasted in the second and third trimester of their pregnancies.

Table 4 shows the mean birth weights of newborns of women with respect to their adherence to fasting and trimester of pregnancy at the start of Ramadan. Newborns of women who fasted more than half a month in the first trimester had the lowest birth weight, although we could not demonstrate statistical interaction suggesting opposing effects of adherence to Ramadan fasting per trimester of exposure (p=0.43).

Table 3. Relation between Ramadan fasting in pregnancy with birth weight of newborns

	Birth weight (grams)	
	Linear regression coefficient (95% CI)	***P* value**
Model 1		
Did not fast (n=60)	Reference	
Fasted in T1 (n=28)	-170.5 (-415.4, 74.5)	0.17
Fasted in T2 (n=27)	141.3 (-106.8, 389.3)	0.26
Fasted in T3 (n=15)	81.4 (-227.6, 390.4)	0.60
Model 2		
Did not fast	Reference	
Fasted in T1	-208.7 (-483.9, 66.6)	0.14
Fasted in T2	97.3 (-165.3, 359.9)	0.47
Fasted in T3	69.0 (-266.1, 404.1)	0.68
Model 3		
Did not fast	Reference	
Fasted in T1	-242.6 (-513.2, 28.0)	0.07
Fasted in T2	52.0 (-207.3, 311.3)	0.69
Fasted in T3	58.8 (-269.0, 386.7)	0.72
Model 4		
Did not fast	Reference	
Fasted in T1	-272.1 (-546.9, 2.8)	0.05
Fasted in T2	28.3 (-234.9, 291.6)	0.83
Fasted in T3	46.8 (-289.3, 382.9)	0.78

Linear regression coefficients indicate the difference in birth weight in grams with the reference category.
Model 1 is a crude model
Model 2 is adjusted for mother's smoking status during Ramadan and pregnancy and mother's ethnicity.
Model 3 was adjusted as for model 2 and for nuliparity.
Model 4 was adjusted as for model 3 and for risk classification of pregnancy at the start of prenatal care and for socioeconomic factors (mother's education).

Table 4. Newborns mean birth weight according to Ramadan fasting adherence and pregnancy trimester

	Mean Birth Weight (in grams)		
	Did not fast	**Fasted half a month or less**	**Fasted more than half a month**
Trimester 1	3421	3439	3226
Trimester 2	3494	3862	3524
Trimester 3	3428	3621	3467

P value for interaction = 0.43

Discussion

In the present study, we provide direct evidence that maternal Ramadan fasting during early pregnancy in an urban Dutch Muslim population may be associated with some reduction in the birth weight of newborns, although it is of borderline statistical significance. The effect may be particularly marked among women who fasted more than half a month in the first trimester of pregnancy, and was robust after correction for potential confounders.

Our findings are in line with an earlier large study, which demonstrated a small reduction in birth weight among babies who were exposed to Ramadan during pregnancy, although that study lacked information on maternal fasting.[12] These findings are also consistent with previous studies, which showed lower birth weight of newborns of women who fasted, particularly if it lasted more than 20 days.[9,23] Our findings are at odds with small studies carried out in predominantly Muslim countries, which found similar birth weight among the babies of women who had fasted during Ramadan and of those who had not.[10,11,24] Although these studies did directly assess maternal fasting, confounders (such as other health related lifestyle, parity and socioeconomic status) were not addressed in the analyses, where we believe that confounding is a major issue in this type of research question. Studies that took ultrasound-based fetal growth indices as the outcome also did not show significant differences between offspring of fasting and non-fasting women.[25-27] Another explanation for the discrepancy with our current findings may be the fact that the number of hours of fasting during Ramadan in our population was up to 25% shorter due to the geographical latitude.

The effect of fasting during pregnancy on the birth weight of the newborns may be mediated through several mechanisms. Fasting can produce 'accelerated starvation'[28] during pregnancy, with more rapid and marked metabolic and endocrine changes (lower blood glucose level, higher cortisol level, lower lipid level) than in non-pregnant women.[26,29,30] Hyperemesis gravidarum may also be more prevalent or severe among fasting women, adding further to metabolic alterations. A study in Israel showed a significant increase in the hospital admission of pregnant Muslim women in their first trimester during Ramadan due to hyperemesis gravidarum.[31]

The relatively small sample size is a limitation of our study. A relatively low response rate of 56% may have induced selection bias, although we find it difficult to perceive how the association could be different among participants and non-participants. We are aware that women's nutritional status could confound our findings, since it may relate to both birth weight of the newborns and women's fasting adherence. A significantly higher BMI was reported in mothers who fasted than in mothers who did not fast,[10] while other studies showed no differences in BMI.[23,30] We could not correct for BMI due to the absence of data regarding women's anthropometric status. Data on placental measurements were not accessible in the present study, which prohibits further explanatory analysis. However, we believe that placental measurement was not a confounder in the association between Ramadan fasting and newborn's birth weight. In the present study, we assumed that the fasting and non-fasting women experienced an equivalent level of lifestyle changes, which may include changes in sleeping pattern and duration, nutritional intake, and physical activities. However, we cannot exclude the possibility that lifestyle changes are dependent of women's fasting adherence and may then have different consequences on pregnancy outcome.

To the best of our knowledge, we are the first to study Ramadan fasting during pregnancy in Muslim women who mostly originated from the Mediterranean and Eurasian region, while living in a Western country. These women could have experienced different degrees of lifestyle changes during Ramadan as compared to women living in other countries, thus the present results may be specific to the present study population. Ramadan fasting in pregnancy was reported by 54% of the women in our study. This suggests a relatively moderate attitude towards adherence to Ramadan fasting in pregnancy compared to in other parts of the world, especially in Middle-eastern countries where some 70 to 90 percent of pregnant women adhere to fasting.[11,12] The proportions of fasting mothers during pregnancy differed across ethnicities in our study (78.9% in Moroccan, 21.7% in Turkish, and 40% in other ethnicities). Proportions were also different between trimesters of the pregnancy and slightly higher in the first trimester. This finding was similar to a previous study,[11] and might be due to the fact that during the first trimester of pregnancy the women were still unaware about their pregnancy or because the burden of pregnancy is lower.[12] Our present findings indicate that Ramadan fasting in the first trimester may lead to a decrease in birth weight, highlighting the need for further study.

In the present study, birth weight among newborns of women who fasted during the first trimester was 272 grams lower than newborns of non-fasting women. Fasting in the second and third trimester was not associated with birth weight. This corresponds with a previous finding that newborns whose mothers fasted in their first trimester had a 1.5 fold risk for low birth weight than newborns of non-fasting mothers.[9] in contrast, this finding is different from a study in Saudi Arabia, which showed similar birth weights in newborns who were exposed to Ramadan in the first, second, or third trimester of pregnancy compared to those who were unexposed.[32] However, in that study, actual adherence of fasting was not recorded and confounders may be an issue since effect estimates were only adjusted for gender, time through the study, and gestational age.

Possibly, placental and fetal growth trajectories are both irreversibly affected by maternal endocrine and metabolic alterations caused by Ramadan fasting. A study showed that Ramadan exposure in the second and third, but not in the first trimester of pregnancy, is associated with smaller placenta.[32] Placenta responds to maternal limited ability to deliver nutrients to them by slowing their growth and increasing their efficiency, so that fetal growth is sustained. Since similar changes were not found in the placenta of newborns who were exposed to Ramadan in the first trimester of pregnancy, the authors speculated that this placental adaptation appears later in the pregnancy, when the maternal blood supply is established. The fact that in our study birth weights were lowest among children of women who fasted in early pregnancy may indicate a maladaptive placental response towards dietary changes during Ramadan. Fasting early in pregnancy, during critical period of organogenesis, may also imply metabolic alterations in other organ systems, as has been demonstrated in animal models of undernutrition.[33]

Conclusion

Our findings suggest that Ramadan fasting during early pregnancy may reduce the birth weight of newborns. It is still unknown to what extent this reduction relates to perinatal morbidity and mortality. Further studies with larger sample size are needed to provide more robust evidence.

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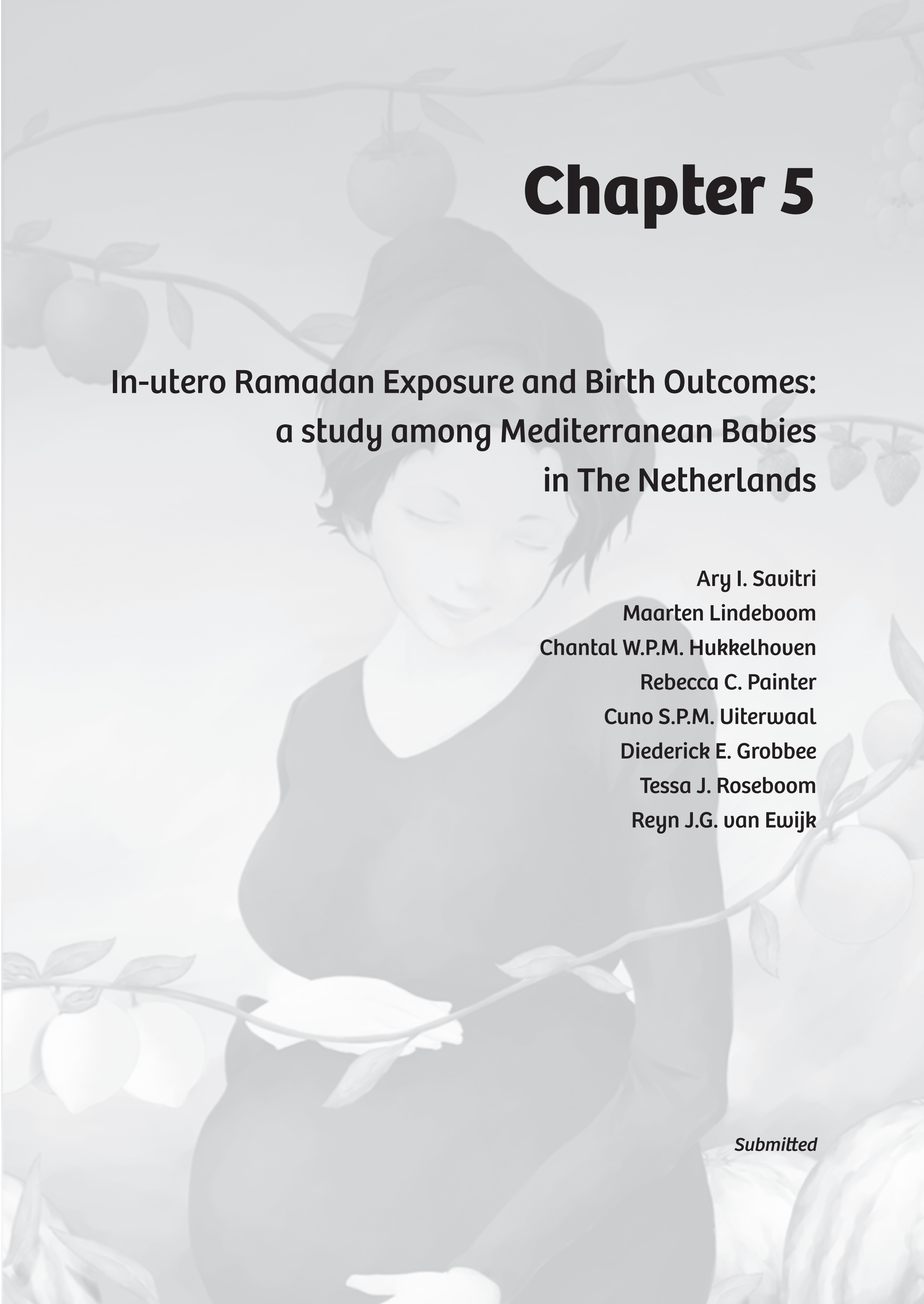

Chapter 5

In-utero Ramadan Exposure and Birth Outcomes: a study among Mediterranean Babies in The Netherlands

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Abstract

Background

In the month of Ramadan many Muslims fast between sunrise and sunset. Ramadan overlaps with the great majority of pregnancies and although pregnant women are exempted, many prefer to fast. Ramadan is associated with major but transient behavioral changes, including intermittent fasting as well as changes in physical activity and sleeping pattern, which may affect the well-being of the fetus. Previous research suggests negative long-term health effects to prenatally exposed children.

Methods

We investigated whether in-utero Ramadan exposure, as a proxy for Ramadan fasting and other lifestyle changes, has an impact on various pregnancy outcomes (birth weight, baby's sex, Apgar score, congenital anomaly, prematurity, and perinatal death). We used all births (2000 until 2010) available from the Perinatal Registry of the Netherlands (Perined), which includes maternal, obstetric, postpartum, and neonatal information of all deliveries from 22 weeks gestation onwards in the Netherlands (n = 1,987,124). Main analyses concerned 139,322 babies from women of Mediterranean origin (as a proxy for Muslims), 7.0% of the total births registered. Ramadan exposure was defined as overlap between gestation and Ramadan in the corresponding year. Depending on the type of outcome we used (multiple) linear or logistic regression, with adjustments for possible confounders.

Results

In the analysis within Mediterranean babies, Ramadan exposure during pregnancy was associated with 12.0 g (95% CI 4.4, 19.6) higher birth weight, lower odds for small for gestational age (SGA) (adjusted Odds Ratio (aOR) 0.92; 0.86, 0.97) and higher odds for severe congenital anomaly (aOR 1.18; 1.01, 1.37), which was most pronounced with third trimester exposure (aOR 1.15; 1.01, 1.30). No associations were found with regards to all other outcomes. Based on the difference in difference analysis, coincidence with Ramadan had no effect on all outcomes.

Conclusions

Exposure to Ramadan in pregnancy does not seem related to health outcomes in babies.

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Introduction

In-utero environment plays an important role in determining health outcomes of the offspring, both in the short and long term. Maternal dietary restriction and stress during pregnancy have been known to provide an adverse intra-uterine environment, which consequently hampers fetal growth and development.[1–3] Maternal under-nutrition not only increases the risks for lower birth weight and poorer birth outcomes, but also raises the risk for developing chronic diseases later in life. Studies have demonstrated an increased risk of coronary heart disease and stroke, obesity, type 2 diabetes, hypertension, as well as cognitive and emotional dysfunction due to poor in-utero environment.[4–6]

Nutrition and life style changes do occur among pregnant Muslim women who practice fasting during Ramadan. Ramadan is an annual period of daytime fasting that lasts for a month. Eating, drinking, smoking, and sexual activity are prohibited from dawn to sunset. Ramadan is considered both as a moment for spiritual activities and self-restraint,[7] and also as a religious festival.[8,9] It involves various degrees of lifestyle changes; including changes in nutritional intake, sleeping pattern and physical activity levels.[9–12] Meal frequency is usually reduced to only two times a day. A large meal is taken when breaking the fast at sunset and a smaller meal is taken at "sahoor" before the sun rises. Seasonal meals that consist of sugary and high fat foods are often served during this month.[8,13] Physical activities during the days are usually reduced, and people tend to be more active at nights. Since Ramadan is based on the Islamic Lunar calendar, it shifts forward by approximately 11 days each year and has occured during each period within the seasonal year over a 33-year cycle.[11,14] Fasting in the northern hemisphere is most difficult to perform during summer due to the long days and hot temperatures.

The great majority of pregnant Muslim women have pregnancies that overlap with Ramadan, and although they are permitted to postpone their fasting,[7] many choose to fast. The proportion of pregnant Muslim women who fast for at least a day during Ramadan range from 50 – 70 % (in Iran, The Netherlands)[15-18] to 70 – 90 % (in England, Singapore, US, Gambia, and Yemen).[19-22] Although fasting during Ramadan in pregnancy is a common phenomenon, there is still little evidence regarding possible health effects. The dietary intake and weight gain of fasting pregnant women are lower than those among pregnant women that do not fast in pregnancy.[23,24] Fasting in pregnancy has been associated with several metabolic alterations including reduction in serum glucose, insulin, lactate, and carnitine levels,[25] as well as elevation in serum triglyceride, cortisol, and leptin concentrations.[24–27] A large study from the USA found a slightly lower birth weight and a lower fraction of male births.[19] But a study with over 1,000,000 babies born to Muslims in Germany found no such effects.[28] Several

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smaller studies also showed no adverse effects of Ramadan during pregnancy with regard to birth weight and risk of low birth weight,[15-17,23,27,29-33] , nor with respect to amniotic fluid volume,[25,34], uterine arterial blood flow,[35] Apgar score,[36] and gestational length.[27] But other studies did find lower birth weights,[18,37] reduced fetal breathing movements,[25,38] lower placental growth,[39] decreased amniotic fluid volume.[40,41] A few large studies investigated the long-term implications for the offspring and found increased rates of disabilities,[19] poorer performance in school,[42] effects on adult body size,[43] and increases in symptoms indicative of type 2 diabetes and coronary heart problems among the elderly.[44] A lesser degree of life style change, although not absent, are expected to occur among women that postpone their fasting. Sudden behavioral changes of the family, which include change in dietary pattern, physical activity, and sleeping pattern might also affect the women and their unborns. Much of the previous literature does not distinguish between effects due to fasting and effects due to other lifestyle changes.

Many of the studies on this topic were small and were done at one point in time, which allows limited correction for the confounding effects of seasonality. In the present study, we used data from a nationwide registry to evaluate birth outcomes of all babies born to Mediterranean mothers in The Netherlands who were exposed to Ramadan during their prenatal period, irrespective of lifestyle changes. In The Netherlands, Mediterranean mainly comprises people of Moroccan and Turkish descent, most of whom are Muslims. People from other origins who were most likely were not Muslim, such as Spanish or Italian, could still be classified into this ethnic group. However, due to the very small number, we could disregard them in the analysis.

Methods

Study population

We analyzed data from the Perinatal Registry of the Netherlands (Perined), a nationwide registry that includes maternal, obstetric, postpartum, and neonatal information of all deliveries in the Netherlands from January 2000 through December 2010. Midwives and doctors who provided antenatal care and assistance during childbirth filled out a form, based on data they had collected from history taking, clinical and laboratory examination, as well diagnosis and intervention. Perined includes all births (live births and stillbirths) that occur after 16 weeks of gestational age, and it contains approximately 96% of all births in the Netherlands.[45]

In the Perined database, a variable is dedicated to describe mother's ethnicity, which could be Dutch, Mediterranean, other European ethnicities, Creole, Indian, Asian, or other. We restricted the analyses to Mediterranean and Dutch babies by excluding babies from all other ethnicities. All babies (live births and stillbirths) with a gestational age of 22^{+0} weeks onwards were included in the analyses, except for outcomes of Apgar score < 7 and prematurity where only live births were analyzed. To obtain information on socioeconomic status (income) at the postal code level, the Perined data were complemented with data from Statistics Netherlands.

Ramadan exposure

Ramadan exposure was based on real overlap between Ramadan and gestation. Gestation begins at the estimated date of conception, calculated as 14 days after the start of women's last menstrual period, and ends at the babies' date of birth. Mediterranean origin was used as a proxy for being Muslim, since in the Netherlands it predominantly comprises two ethnicities with a large share of Muslims, mainly from Turkish and Moroccan ethnicity. The proportion of Muslims in Turkish and Moroccan descendants in The Netherlands is 87% and 94%, respectively.[46]

Pregnancies were classified as overlapping with Ramadan if the estimated conception date coincided with Ramadan or if Ramadan started after the estimated date of conception and prior to the date of birth. All pregnancies that overlapped with Ramadan in any number of days were classified as Ramadan exposed. Babies who were conceived within 7 days after Ramadan were controlled for in the analyses as the possibility cannot be excluded that their health was still affected by Ramadan. Further Ramadan exposure classification using trimesters were made according to the trimester that most of the Ramadan period coincided with. Babies were classified as being exposed to Ramadan in the first trimester if they were conceived during Ramadan or if Ramadan started between the 1st and 88th day of pregnancy. They were classified as being exposed in the second and third trimester if Ramadan started between 89th to 177th day and at 178th day or later of the pregnancy, respectively. Since Ramadan lasts for 29 or 30 days each year, a trimester in which at least 16 days of Ramadan fell into was considered as the trimester of exposure. In some cases where there were equal numbers of days in two trimesters that overlapped with Ramadan, the exposure was classified to occur in the earlier trimester.

For outcomes for which it was not possible to control for gestational length in regressions (such as gestational duration itself and prematurity), exposure to Ramadan during pregnancy was defined as overlap between pregnancy and Ramadan based on the calculated date of conception with the assumption that all pregnancies lasted for 266 days since conception, which is the average length of gestation in human.

Perined does allow for accurate measurement of individual exposure to the Ramadan period but it does not include data on Ramadan fasting behavior or other lifestyle changes. Therefore, in our main analyses, exposure to Ramadan was used as an approximation for Ramadan fasting and other lifestyle changes.

Outcome measurement

Several birth outcomes were investigated in relation to in-utero Ramadan exposure. Used outcome measures of fetal growth were birth weight, occurrence of low birth weight (LBW; birth weight lower than 2,500 grams), and small for gestational age (SGA; birth weight below the 10^{th} percentile for gestational age). Outcome measure of neonatal health was the occurrence of an Apgar score lower than 7. Further outcome measures were: sex ratio of the babies (the share of male babies as compared to female babies), any congenital anomalies, mild congenital anomalies, severe congenital anomalies, gestational duration, prematurity (born at less than 37^{+0} weeks of gestation), and perinatal mortality (occurrence of stillbirth or death during delivery or within 7 days after birth). Mild congenital anomalies include disorders of the eyes, ears, skin defect, palatoschisis or other organs which are not life-threatening. Severe congenital anomalies consist of serious or potentially lethal disorders such as anencephaly, microcephaly, disorders of the heart, and disorders of the respiratory tract. Gestational duration was defined as the time since women's last menstrual period until the date of delivery. Babies were classified as being premature if they were born at less than 37 weeks of gestational age.

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Data analysis

Maternal characteristics, pregnancy complications, and babies' characteristics were tabulated by Ramadan exposure for descriptive purposes and for first evaluation of possible confounding, both within Mediterranean and Dutch pregnancies separately. Independent samples t-tests, Chi-square tests, and Mann-Whitney tests were used to test for differences, wherever appropriate.

We used linear regression to analyze continuous dependent variables and logistic regression to analyze dichotomous dependent variables. Univariable analyses were first performed and followed by multivariable analyses for adjustment for confounders. Potential confounders were a-priori set to include month of conception (seasonal effects), gestational length, gestational length squared, gestational length cubed, maternal age, maternal age squared, baby's sex, average income based on postal code (socio-economic status), parity, year of delivery, maternal smoking, and conceived within 7 days after Ramadan. We considered gestational length as the most important confounder since it determined the fetus' chance for being exposed to Ramadan, while it is also correlated with most of the outcomes. Adjustment for gestational length, however, was not necessary when we analyzed SGA, since this variable is already corrected for gestational age. Moreover, adjustment for gestational length was inappropriate in the analyses where the dependent variable was gestational duration itself, or where the dependent variable was a function of gestational length (the variable prematurity). For outcome of sex ratio, variable of baby's sex was excluded from the adjusted model.

First, analyses were done within Mediterranean babies by comparing birth outcomes of babies who were exposed to Ramadan during gestation with babies who were not exposed. However, this "intent to treat" (ITT) design may have limitations in separating seasonal effects from the true Ramadan effects. Seasonality has been known to relate with various pregnancy outcomes, particularly on birth weight and preterm birth.[47,48] Since Ramadan shifts forward by approximately 11 days each year, it has occured during each period of the seasonal year over 33-year cycle.[11,14] In the case where there were data covering 33 years of birth cohorts, a standard seasonal controls approach could remove possible confounding effects of seasonality. However, in the present study, analyses were based on 11 birth cohorts (year 2000 to 2010), suggesting potentially strong seasonal effects. Therefore, we used a "difference in difference" strategy to overcome this problem by looking at Ramadan effects on Mediterranean babies and further subtracted any effects for Dutch babies caused by possible seasonal effects. We developed a model that included the variables "Mediterranean", "Ramadan exposure", all potential confounders, the interaction terms between the variable "Mediterranean" and all potential confounders, and the interaction term between "Mediterranean" and "Ramadan exposure". The effect estimates of Ramadan came from the coefficients of the latter interaction term ("Mediterranean" * " Ramadan exposure"). This strategy was similar to the one performed in two previous studies.[28,42]

Results are expressed as linear regression coefficients or odds ratio (OR) with 95 % confidence intervals and P values. All analyses were done using Stata/SE 11. 1.

Results

The initial number of registered babies was 1,987,124. Non-Mediterranean and non-Dutch babies (n=224,505), twins or triplets (n=68,810), babies with uncertain length of gestation and babies who had miscoded gestational lengths (n=73,052) were excluded leaving 1,620 757 babies for analysis. Mediterranean babies comprised 139,322 (8.6% of the total study population). The flow chart of subject selection is shown in **Figure 1**.

The baseline characteristics are shown in **Table 1**. As compared to the unexposed, Mediterranean mothers who had been exposed to Ramadan were slightly younger, had lower average income based on postal code, and had on average 2 days longer gestational length. The difference in gestational age is “by construction”, as shorter pregnancies have a lower probability of overlapping with a Ramadan. In addition, spontaneous and instrumental vaginal delivery was more common in women who were exposed. No difference was found with regard to parity and pregnancy complications (hyperemesis gravidarum and hypertension). Within Dutch mothers, those who were pregnant during a Ramadan were slightly younger, had higher average incomes, were less likely to be primiparous, and had on average 2 days longer gestational length. Spontaneous and instrumental vaginal deliveries were also more common among them. No difference was found in smoking and pregnancy complications. Characteristics of Mediterranean and Dutch pregnancies were also compared in **Appendix 1**.

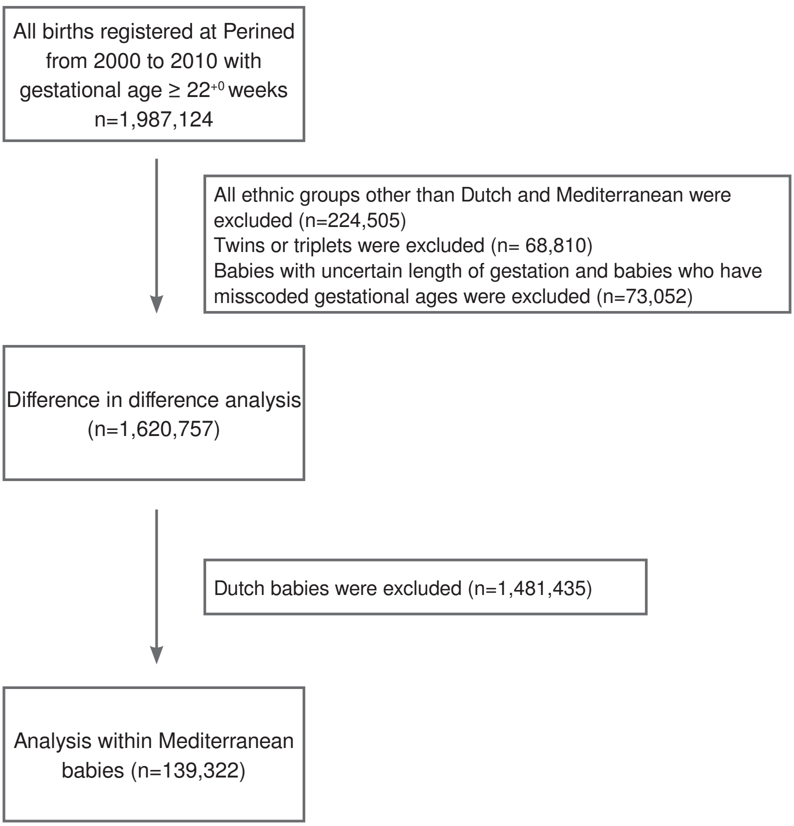

Figure 1. Flow chart of subject selection

Table 1. Baseline characteristics of study population based on Ramadan exposure

Characteristics	Mediterranean (n=139,322)		p-value	Dutch (n=1,481,435)		p-value
	Exposed	Unexposed		Exposed	Unexposed	
Number (%)	116,010 (83.3)	23,312 (16.7)		1,219,606 (82.3)	261,829 (17.7)	
Maternal age	28.7 (5.4)	28.9 (5.5)	< 0.01[1]	30.8 (4.6)	30.7 (4.6)	0.01[1]
Maternal smoking (%)	215 (0.2)	51 (0.3)	0.062[2]	4,553 (0.3)	854 (0.3)	0.79[2]
Average income in euro based on postcode*	1751.8 (317.5)	1756.7 (317.5)	0.009[3]	1949.5 (364.5)	1946.8 (365.2)	< 0.01[3]
Primiparous (%)	41,927 (36.1)	8,500 (36.5)	0.352[2]	576,147 (47.2)	126,747 (48.4)	< 0.01[2]
Gestational length in days*	266 (13)	264 (15)	< 0.01[3]	266 (13)	264 (15)	< 0.01[3]
Pregnancy complications (%)						
Hyperemesis gravidarum	167 (0.1)	23 (0.1)	0.09[2]	925 (0.1)	213 (0.1)	0.36[2]
Hypertension	1,966 (1.7)	380 (1.6)	0.48[2]	44,324 (3.6)	9,560 (3.6)	0.67[2]
Highest diastolic pressure			< 0.01[2]			< 0.01[2]
<90 mmHg	106,878 (93.3)	21,474 (93.2)		1,047,590 (87.0)	224,569 (86.9)	
90-110 mmHg	6,634 (5.8)	1,295 (5.6)		138,862 (11.5)	29,108 (11.3)	
≥ 110 mmHg	1,062 (0.9)	278 (1.2)		18,271 (1.5)	4,737 (1.8)	
Type of delivery (%)			< 0.01[2]			< 0.01[2]
Spontaneous	93,444 (80.7)	18,612 (80.0)		903,486 (74.2)	192,027 (73.5)	
Vacuum, forceps and instrumental vaginal delivery	8,817 (7.6)	1,681 (7.2)		137,363 (11.3)	28,470 (10.9)	
Primary cesarean section	5,516 (4.8)	1,387 (6.0)		77,905 (6.4)	20,269 (7.8)	
Secondary cesarean section	8,003 (6.9)	1,577 (6.8)		98,010 (8.0)	20,400 (7.8)	

Values are means with standard deviations for continuous variables and percentages for frequencies. In case of skewed data (*), median with inter-quartile range were presented.

Exposed group comprised of babies with in-utero Ramadan exposure at any time of gestation, while unexposed babies consisted of babies without in-utero Ramadan exposure.

[1] Two sample t-test, [2] Chi-Square test, [3] Mann-Whitney test

Table 2 shows the adjusted associations between in-utero Ramadan exposure with birth outcomes based on the analyses within Mediterranean babies. The crude associations are presented in **Appendix 2**. Ramadan overlapped with 116 010 (83.3%) gestations of all Mediterranean babies. Consecutively 28.7%, 25.7%, and 28.9% of Mediterranean babies were exposed to Ramadan in the first, second, and third trimester. In-utero Ramadan exposure was associated with 112 grams higher birth weight, but with adjustment for confounders (month of conception [seasonal effects], gestational length, gestational length squared, gestational length cubed, maternal age, maternal age squared, baby's sex, average income based on postal code [socio-economic status], parity, year of delivery, maternal smoking, and conceived within 7 days after Ramadan), this attenuated to 12 grams. Especially adjusting for gestational length is likely to reduce bias in the estimates, as Ramadans by construction more often occur during longer pregnancies. Analyses by trimester showed similar findings, and after adjustment for confounders, only exposure mainly in the third trimester showed slightly higher birth weights. A 49% lower risk for low birth weight (LBW) with Ramadan exposure was completely explained by confounders. We found similar patterns for exposure in each trimester. Risk for small for gestational age (SGA) was 8% lower with Ramadan exposure after adjustment for confounders. Risk for Apgar score lower than 7 did not differ in exposed babies as compared to unexposed babies after adjustment for confounders, both in total and trimester-specific analyses. With Ramadan exposure, the risk for any congenital anomaly was not higher than among the unexposed, although a trimester-specific analysis showed a higher risk with second trimester exposure. Risk for mild congenital anomalies did not differ with regards to exposure group. However, the risk for severe congenital anomalies was higher after Ramadan exposure, mainly with third trimester exposure. Babies' sex ratio was not different in the exposed and unexposed babies. Gestational duration and risk for prematurity appeared to be similar. Likewise, the risk of perinatal death was also similar in the exposed as compared to the unexposed group.

Table 2. Adjusted associations between in-utero Ramadan exposure and birth outcomes of Mediterranean babies, based on analysis within Mediterranean

Outcomes	Timing of exposure			
	In any trimester	**First trimester**	**Second trimester**	**Third trimester**
Birth weight in grams	12.0 (4.5, 19.6)‡	5.7 (-2.2, 13.7)	5.2 (-3.3, 13.7)	9.3 (3.0, 15.7)‡
Low birth weight	0.97 (0.86, 1.08)	0.99 (0.87, 1.12)	0.96 (0.84, 1.09)	0.96 (0.87, 1.06)
Small for Gestational Age (SGA)	0.92 (0.87, 0.97)‡	0.94 (0.89, 1.00)	0.96 (0.90, 1.03)	0.97 (0.93, 1.02)
Apgar score < 7*	1.04 (0.89, 1.22)	1.04 (0.88, 1.23)	0.91 (0.76, 1.09)	0.99 (0.86, 1.13)
Any congenital anomaly	1.10 (0.97, 1.24)	1.07 (0.94, 1.22)	1.16 (1.01, 1.33)†	1.09 (0.98, 1.21)
Severe congenital anomaly	1.18 (1.01, 1.37)†	1.05 (0.89, 1.23)	1.12 (0.94, 1.33)	1.15 (1.01, 1.30)†
Mild congenital anomaly	0.97 (0.79, 1.18)	1.11 (0.90, 1.38)	1.22 (0.97, 1.52)	0.97 (0.82, 1.16)
Sex ratio	1.00 (0.96, 1.03)	1.03 (0.99, 1.07)	1.02 (0.98, 1.07)	1.00 (0.97, 1.03)
Sex ratio*	0.99 (0.96, 1.03)	1.03 (0.99, 1.07)	1.02 (0.98, 1.07)	1.00 (0.97, 1.03)
Gestational duration in days	-0.1 (-0.4, 0.2)	-0.3 (-0.6, -0.01)†	-.0.3 (-0.7, 0.06)	0.2 (-0.1, 0.5)
Prematurity*	1.05 (0.96, 1.15)	1.09 (0.99, 1.20)	1.13 (1.01, 1.26)†	1.00 (0.90, 1.11)
Perinatal mortality	0.85 (0.71, 1.03)	0.85 (0.68, 1.06)	0.90 (0.72, 1.14)	0.97 (0.82, 1.15)

CI, confidence interval; OR, odds ratio

Exposure was measured based on actual overlap between pregnancy and Ramadan; babies who were not exposed to Ramadan nor conceived within 7 days after Ramadan were taken as reference. For analysis on outcomes of gestational duration and prematurity, exposure was determined based on overlap between pregnancy and Ramadan based on the calculated date of conception and the assumption that all pregnancies lasted for 266 days since conception.

Results are expressed as linear regression coefficients (95% CI) in continuous outcomes or OR (95% CI) from logistic regression in case of dichotomous outcomes.

Adjustments are made for month of conception (seasonal effects), gestational age, gestational age squared, gestational age cubed, maternal age, maternal age squared, baby's sex, socio-economic status (averaged income based on postal code), parity, year of delivery, maternal smoking, and conceived within 7 days after Ramadan. For outcome of sex ratio, result was not adjusted for baby's sex. For outcome of small for gestational age (SGA), gestational duration, and prematurity, results were not adjusted for gestational age, gestational age squared, and gestational age cubed. *Only live births were analyzed, † $p < 0.05$, ‡ $p < 0.01$

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In **Table 3**, we present the adjusted associations between Ramadan exposure and birth outomes based on difference in difference analysis. The crude associations are presented in **Appendix 3**. In both the crude and adjusted analyses, no effects of Ramadan exposure were found with respect to all birth outcomes investigated when we aggregate exposure across all semesters. And the analyses by trimester only show a higher rate of severe congenital anomalies among those exposed in the third trimester.

Table 3. Adjusted associations between in-utero Ramadan exposure and birth outcomes of Mediterranean babies, based on difference in difference analysis

Outcomes	Timing of exposure			
	In any trimester	First trimester	Second trimester	Third trimester
Birth weight in grams	8.0 (-0.003, 16.1)	7.0 (-1.4, 15.5)	2.0 (-7.0, 11.0)	5.7 (-1.0, 12.4)
Low birth weight	0.99 (0.88, 1.11)	0.98 (0.86, 1.11)	0.99 (0.87, 1.14)	0.98 (0.89, 1.09)
Small for Gestational Age (SGA)	0.96 (0.90, 1.01)	0.94 (0.88, 1.00)	0.99 (0.93, 1.06)	1.00 (0.96, 1.06)
Apgar score < 7*	1.07 (0.91, 1.27)	1.04 (0.87, 1.24)	0.95 (0.79, 1.15)	1.00 (0.87, 1.16)
Any congenital anomaly	1.10 (0.96, 1.25)	1.05 (0.91, 1.21)	1.11 (0.96, 1.28)	1.10 (0.98, 1.22)
Severe congenital anomaly	1.17 (1.00, 1.37)†	1.03 (0.87, 1.22)	1.08 (0.90, 1.29)	1.15 (1.00, 1.31)†
Mild congenital anomaly	0.97 (0.79, 1.20)	1.08 (0.86, 1.35)	1.17 (0.92, 1.49)	0.99 (0.83, 1.19)
Sex ratio	1.00 (0.97, 1.04)	1.03 (0.99, 1.07)	1.02 (0.98, 1.07)	1.01 (0.98, 1.04)
Sex ratio*	1.00 (0.97, 1.04)	1.03 (0.99, 1.07)	1.02 (0.98, 1.07)	1.01 (0.98, 1.04)
Gestational duration in days	0.01 (-0.3, 0.3)	0.04 (-0.3, 0.4)	0.07 (-0.3, 0.4)	-0.01 (-0.3, 0.3)
Prematurity*	1.04 (0.95, 1.14)	1.03 (0.93, 1.14)	1.04 (0.93, 1.17)	1.04 (0.94, 1.16)
Perinatal mortality	0.81 (0.66, 0.99)	0.82 (0.65, 1.04)	0.89 (0.70, 1.14)	0.94 (0.79, 1.13)

CI, confidence interval; OR, odds ratio

Exposure was measured based on actual overlap between pregnancy and Ramadan; babies who were not exposed to Ramadan nor conceived within 7 days after Ramadan were taken as reference. For analysis on outcomes of gestational duration and prematurity, exposure was determined based on overlap between pregnancy and Ramadan based on the calculated date of conception and the assumption that all pregnancies lasted for 266 days since conception.

Results are expressed as linear regression coefficients (95% CI) in continuous outcomes or OR (95% CI) from logistic regression in case of dichotomous outcomes.

Adjustments are made for month of conception (seasonal effects), gestational age, gestational age squared, gestational age cubed, maternal age, maternal age squared, baby's sex, socio-economic status (averaged income based on postal code), parity, year of delivery, maternal smoking, and conceived within 7 days after Ramadan. For outcome of sex ratio, result was not adjusted for baby's sex. For outcome of small for gestational age (SGA), gestational duration, and prematurity, results were not adjusted for gestational age, gestational age squared, and gestational age cubed. All covariates are interacted with an indicator variable for "Mediterranean". *Only live births were analyzed, † $p < 0.05$, ‡ $p < 0.01$

Discussion

About 83% of all Mediterranean babies in our study population were exposed to Ramadan during pregnancy. We found that in-utero exposure to Ramadan was associated with a slightly higher birth weight, lower risk for small for gestational age (SGA), and higher risk for severe congenital anomaly as compared to Mediterranean pregnancies that were not exposed to Ramadan in utero. No association was found between in-utero Ramadan exposure with risk of low birth weight (LBW), risk of having Apgar score lower than 7, risk of any congenital anomaly, risk of mild congenital anomaly, sex ratio, gestational duration, risk of prematurity, risk of perinatal mortality (stillbirth, death during delivery, or within 7 days after birth). Using a “difference in difference” strategy, no associations between Ramadan exposure and outcomes was found, except for an increased occurrence of severe congenital anomalies among those exposed in the third trimester.

To our knowledge, this study is among the first to investigate the association between Ramadan exposure during pregnancy with birth outcomes using data from a national registry. A large number of babies were included in the registry, which enabled us to analyze with substantial statistical power. Data on confounders were also available in the dataset, allowing for adequate adjustment. There are also some limitations. Data about religion was not available, thus Mediterranean ethnicity could only be used as a proxy for being Muslim. This approach may have led to misclassification and a slight bias toward finding no effects. Mediterranean babies in our data accounted for 7.0% of the total, while Muslims are estimated to constitute 6% of all population in The Netherlands.[42] Furthermore, in-utero Ramadan exposure was measured based on overlap between pregnancy and Ramadan, without information on actual maternal fasting.

A slightly higher birth weight and lower risk for SGA with Ramadan exposure, which we found in the analysis within Mediterranean babies, are relatively small and do not have clinical significance. Similarly, no effects were found in the difference in difference analysis. This finding is in line with a recently published study about Ramadan effects on Muslim infants born in Germany.[28] Using an ITT approach that is similar to ours, the study reported no effects of Ramadan on birth weight. The difference with our study would be that Muslim population in Germany are mainly dominated by people from Turkish origin, whereas in The Netherlands the majority of Muslims are from Moroccan and Turkish origin (31% and 38%, respectively).[49] Given the relatively lower proportion of observance of Ramadan fasting during pregnancy among Turkish women as compared to the Moroccans (42% vs 75%)[18] the results from the latter study were more likely to underestimate the effect of Ramadan observance.

Our result of no effects of in-utero Ramadan exposure on birth weight were also supported by previous studies conducted in predominantly Muslim countries,[15-17,27,30-32] which showed similar birth weight of babies from fasting and non-fasting women. Most of these studies did directly measure maternal fasting during Ramadan, although potential confounders were rarely addressed in the analyses. Our finding of a small increase in birth weight with Ramadan exposure matches some large studies conducted in Birmingham,[29] Unizah (Saudi Arabia),[50] and Michigan (USA).[37] The latter study investigated the association between in-utero Ramadan exposure and birth outcomes using birth cohorts of 50 000 babies with Arab origins, thus it had a similar approach to ours. In that study, although in-utero exposure to Ramadan was found associated with lower birth weight, the effect was relatively small and limited to exposure during the peak period of daylight hours if Ramadan fell in the first month of gestation. Our present finding of limited effects on birth weight from Ramadan exposure does not exclude such effects of Ramadan fasting, as suggested in our previous small study among pregnant Muslim women in Amsterdam, the Netherlands, which showed that maternal Ramadan fasting, mainly during early pregnancy, may be associated with some reduction of birth weight.[18] In that previous study, although maternal fasting behavior was measured, the study was done at a specific calendar period. Therefore the result could be due to seasonal effects on birth weight. In our present study eleven birth cohorts were analyzed and we were able to control for seasonality.

In the present study, in-utero Ramadan exposure was found to increase babies' risk for severe congenital anomaly by 18% in the adjusted analysis. The effect estimates were particularly more evident with exposure in the third trimester. Consequently, that increase in severe congenital anomalies can hardly be explained by this exposure, since virtually all congenital anomalies have underlying mechanisms that originate during embryogenesis in early pregnancy. Therefore, the association found between Ramadan exposure and severe congenital anomaly is more likely artifactual. Our finding of no association between Ramadan exposure and risk of mild congenital anomaly also supports this conclusion.

Only one small previous study had investigated the relation between in-utero Ramadan exposure and Apgar score. Our finding of no association between exposure and risk for Apgar score lower than 7 was in accordance with this previous study.[51] We also analyzed the effects of Ramadan exposure on Apgar sore as a continuous scale and did not find any relation (data not shown).

We analyzed the associations between Ramadan exposure and sex ratio using two approaches. First, analyses were done within all babies registered in the dataset. Since our dataset only registered babies with gestational length of 22 weeks or more, by analyzing all babies, we could identify if any distortion in sex ratio was due to miscarriage. When analyses were restricted to live births, on the other hand, we could show if it was caused by fetal death that occurred later. Our study showed that in-utero Ramadan exposure did not influence sex ratio, both in the analysis in all babies or in live births. This is similar with the finding of a previous study in the population of Muslim infants born in Germany,[28] but different from other studies which found Ramadan exposure, mainly early in pregnancy, skewed the sex ratio in favor of female babies.[19,52]

In-utero Ramadan exposure was not associated with gestational duration nor with the risk of prematurity. This was in accordance with previous studies which compared outcomes of babies from mothers who fasted during pregnancies with those who did not.[27,51,53,54] In contrast, our finding was different from a large study in Michigan (USA) which reported a reduced gestational duration with Ramadan exposure.[37]

Ramadan exposure also did not seem to affect the risk of perinatal death (stillbirth, death during delivery, or within 7 days after birth). This was again in contrast to findings from the Michigan study which reported a 16 percent decline in live births, mainly among male fetuses, who were exposed to Ramadan around the month of conception.

The differences from our results may be explained by the design and sample of the study. The Michigan study investigated live births among babies with gestation between 37 and 42 weeks while in our study all babies with gestational length longer than 22 weeks were included. Furthermore, the Muslims in the Michigan study were mainly of Arab ancestry and may have had different fasting behaviors and lifestyle changes than the Muslims in our sample. Our study was also different from the Michigan study with respect to the months of the calendar year during which the cohorts were analyzed. Our study mainly analyzed Ramadan exposure during the colder months and shorter days, starting with Ramadan exposure in December 2000 and moving forward by 11 days each year to September 2010 (See **Appendix 4**). Contrarily in the Michigan study, Ramadan occurred in April 1989, moving forward by 11 days each year to September 2006. Since the risk for perinatal mortality likely varied across calendar years due to seasonality, the estimated effect of Ramadan on perinatal mortality can also be affected. Furthermore, climate variations between Michigan and The Netherlands may also have effect on the degree of exposure. Michigan has a humid continental climate that is characterized by a large seasonal temperature differences, while the moderate maritime climate in The Netherlands results in lower seasonal temperature differences.

During Ramadan, Muslims observe life style changes, which include nutritional disturbances, modification in sleeping pattern, and decreased level on physical activity. Although Ramadan fasting in pregnancy has been linked to a significant deficit in women's total calorie consumption,[38] foods that contain high level of sugar, fat and salt are often served during the nights. This may contribute to the finding of slightly higher birth weight among Ramadan-exposed babies. Exposure to Eid al-Fitr (Sugar Feast), which follows after Ramadan ends, may also to some extent, explain the finding. Furthermore, since smoking is prohibited during fasting, Ramadan exposure may lead to a reduction of smoking exposure, mainly secondhand smoking. In this study, Ramadan mostly fell during the colder months, providing relatively shorter duration of fasting and a cold ambient temperature, which substantially reduced the burden of fasting. The mean duration of fasting hours during Ramadan months in our study was 12 hours and 19 minutes (inter-quartile range [IQR]= 4 hours and 48 minutes).

In this study, analyses had to be restricted to the effect of Ramadan occurrence during pregnancy without actual measurements on fasting behavior. In The Netherlands, slightly half of all pregnant Muslims fast during Ramadan, but some of those only fast for a few days of this month.[18] Our results therefore under estimate potential effects of Ramadan fasting. The implicit assumption in our analyses is that all Muslim mothers who were pregnant during a Ramadan were affected by it. To the extent that this is not the case, this approach underestimates the real effect of Ramadan during pregnancy. Our estimates should therefore be regarded as conservative (lower-bound) estimates.[43] An important advantage of this approach is that it avoids bias due to systematic differences between Muslims who do versus do not chose to observe Ramadan during pregnancy.[19] More studies in this field are needed, particularly with further analysis on the behavioral changes during Ramadan that may interact with fasting. Future studies may also explore, rather than short-term outcomes, possible long-term effects of in-utero Ramadan exposure on later health outcomes as well as intergenerational effects. The few existing studies on longer-term consequences indicate that these may be larger than short-term effects and may include disabilities, poorer cognitive performance, coronary heart problems and type 2 diabetes.[19,42,44]

Conclusion

In conclusion, in-utero Ramadan exposure, as a proxy for Ramadan fasting, was associated with statistically significant but clinically irrelevant higher birth weight and lower risk for SGA, while other birth outcomes were not influenced. A slightly higher risk for severe congenital anomaly found with exposure was apparently an artifactual finding.

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Appendix 1. Baseline characteristics of Mediterranean babies as compared to Dutch babies

Characteristics	Mediterranean (n=139,322)	Dutch (n=1,481,435)	p-value
Maternal age	28.7 (5.4)	30.7 (4.6)	< 0.01[1]
Maternal smoking (%)	266 (0.2)	5,407 (0.4)	< 0.01[2]
Average income in euro based on postcode*	1754.2 (317.5)	1947.7 (364.5)	< 0.01[3]
Primiparous (%)	50,427 (36.2)	702,894 (47.4)	< 0.01[2]
Gestational length in days*	266 (13)	266 (14)	< 0.01[3]
Pregnancy complications (%)			
Hyperemesis gravidarum	190 (0.1)	1,138 (0.1)	< 0.01[2]
Hypertension	2,346 (1.7)	53,884 (3.6)	< 0.01[2]
Highest diastolic pressure			< 0.01[2]
<90 mmHg	128,352 (93.3)	1,272,159 (86.9)	
90-110 mmHg	7,929 (5.8)	167,970 (11.5)	
≥ 110 mmHg	1,340 (1.0)	23,008 (1.6)	
Type of delivery (%)			< 0.01[2]
Spontaneous	112,056 (80.6)	1,095,513 (74.1)	
Vacuum, forceps and instrumental vaginal delivery	10,498 (7.6)	165,833 (11.2)	
Primary cesarean section	6,903 (5.0)	98,174 (6.6)	
Secondary cesarean section	9,580 (6.9)	118,410 (8.0)	

Values are means with standard deviations for continuous variables and percentages for frequencies. In case of skewed data (*), median with inter-quartile range were presented.
Exposed group comprised of babies with in-utero Ramadan exposure at any time of gestation, while unexposed babies consisted of babies without in-utero Ramadan exposure.
[1] Two sample t-test, [2] Chi-Square test, [3] Mann-Whitney test

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Appendix 2. Crude associations between in-utero Ramadan exposure and birth outcomes of Mediterranean babies, based on analysis within Mediterranean

Outcomes	Timing of exposure			
	In any trimester	First trimester	Second trimester	Third trimester
Birth weight in grams	112.0 (103.4, 120.6)‡	37.9 (29.9, 45.8)‡	59.7 (50.6, 68.9)‡	100.1 (92.3, 107.8)‡
Low birth weight	0.51 (0.48, 0.54)‡	0.78 (0.73, 0.83)‡	0.67 (0.62, 0.72)‡	0.49 (0.46, 0.52)‡
Small for Gestational Age (SGA)	0.97 (0.93, 1.02)	0.97 (0.93, 1.01)	1.00 (0.96, 1.06)	0.98 (0.94, 1.03)
Apgar score < 7*	0.72 (0.64, 0.82)‡	0.94 (0.83, 1.06)	0.78 (0.68, 0.90)‡	0.72 (0.64, 0.81)‡
Any congenital anomaly	0.93 (0.84, 1.03)	1.00 (0.90, 1.10)	1.03 (0.92, 1.15)	0.94 (0.86, 1.04)
Severe congenital anomaly	0.92 (0.81, 1.04)	0.94 (0.83, 1.06)	0.98 (0.86, 1.13)	0.95 (0.84, 1.07)
Mild congenital anomaly	0.92 (0.78, 1.09)	1.08 (0.92, 1.26)	1.07 (0.90, 1.29)	0.91 (0.78, 1.06)
Sex ratio	0.98 (0.96, 1.01)	1.02 (0.99, 1.05)	1.01 (0.98, 1.04)	0.99 (0.96, 1.02)
Sex ratio*	0.98 (0.95, 1.01)	1.02 (0.99, 1.05)	1.01 (0.98, 1.04)	0.99 (0.96, 1.02)
Gestational duration in days	0.07 (-0.2, 0.3)	-0.03 (-0.3, 0.2)	0.01 (-0.3, 0.3)	0.2 (-0.05, 0.5)
Prematurity*	1.04 (0.96, 1.13)	1.06 (0.97, 1.15)	1.08 (0.99, 1.17)	1.00 (0.92, 1.09)
Perinatal mortality	0.38 (0.34, 0.43) ‡	0.67 (0.59, 0.76)‡	0.51 (0.44, 0.58)‡	0.35 (0.31, 0.40)‡

CI, confidence interval; OR, odds ratio

Exposure was measured based on actual overlap between pregnancy and Ramadan; babies who were not exposed to Ramadan nor conceived within 7 days after Ramadan were taken as reference. For analysis on outcomes of gestational duration and prematurity, exposure was determined based on overlap between pregnancy and Ramadan based on the calculated date of conception and the assumption that all pregnancies lasted for 266 days since conception.

Results are expressed as linear regression coefficients (95% CI) in continuous outcomes or OR (95% CI) from logistic regression in case of dichotomous outcomes.

*Only live births were analyzed

† $p < 0.05$, ‡ $p < 0.01$

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Appendix 3. Crude associations between in-utero Ramadan exposure and birth outcomes of Mediterranean babies, based on difference in difference analysis

Outcomes	Timing of exposure			
	In any trimester	**First trimester**	**Second trimester**	**Third trimester**
Birth weight in grams	-0.6 (-9.9, 8.6)	1.0 (-7.5, 9.5)	2.3 (-7.5, 12.2)	7.8 (-0.5, 16.1)
Low birth weight	0.97 (0.91, 1.03)	0.97 (0.91, 1.04)	0.95 (0.88, 1.02)	0.93 (0.87, 1.00)†
Small for Gestational Age (SGA)	0.99 (0.95, 1.04)	0.98 (0.93, 1.02)	1.03 (0.97, 1.08)	1.00 (0.95, 1.04)
Apgar score < 7*	1.02 (0.90, 1.16)	1.04 (0.92, 1.18)	0.96 (0.83, 1.11)	0.98 (0.87, 1.11)
Any congenital anomaly	1.10 (0.99, 1.23)	1.04 (0.94, 1.15)	1.10 (0.98, 1.23)	1.11 (1.00, 1.23)
Severe congenital anomaly	1.14 (1.00, 1.30)	0.98 (0.86, 1.11)	1.08 (0.93, 1.24)	1.18 (1.04, 1.33)‡
Mild congenital anomaly	1.01 (0.85, 1.21)	1.12 (0.95, 1.33)	1.10 (0.91, 1.32)	1.00 (0.85, 1.17)
Sex ratio	1.00 (0.97, 1.03)	1.02 (1.00, 1.05)	1.01 (0.98, 1.04)	1.01 (0.98, 1.04)
Sex ratio*	1.00 (0.97, 1.03)	1.02 (1.00, 1.05)	1.01 (0.98, 1.04)	1.01 (0.98, 1.03s)
Gestational duration in days	0.3 (0.02, 0.5)†	0.1 (-0.2, 0.4)	0.4 (0.1, 0.7)†	0.3 (0.1, 0.6)
Prematurity*	1.01 (0.93, 1.10)	1.04 (0.96, 1.14)	1.01 (0.93, 1.11)	0.98 (0.90, 1.08)
Perinatal mortality	0.95 (0.85, 1.07)	0.90 (0.79, 1.03)	0.91 (0.79, 1.05)	0.99 (0.86, 1.14)

CI, confidence interval; OR, odds ratio

Exposure was measured based on actual overlapped between pregnancy and Ramadan; babies who were not exposed to Ramadan nor conceived within 7 days after Ramadan were taken as reference. For analysis on outcomes of gestational duration and prematurity, exposure was determined based on overlap between pregnancy and Ramadan based on the calculated date of conception and the assumption that all pregnancies lasted for 266 days since conception.

Results are expressed as linear regression coefficients (95% CI) in continuous outcomes or OR (95% CI) from logistic regression in case of dichotomous outcomes.

*Only live births were analyzed

† $p < 0.05$, ‡ $p < 0.01$

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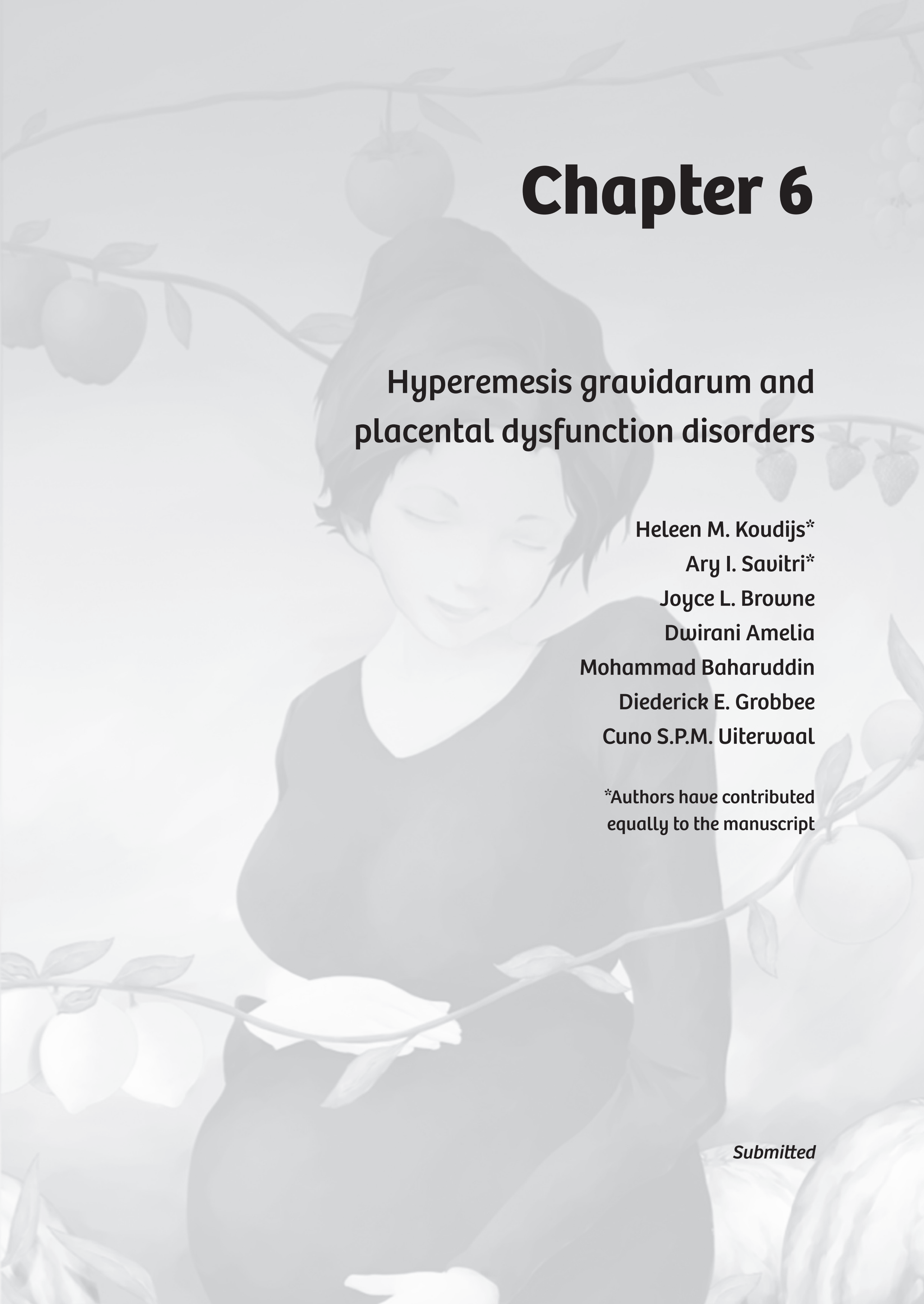
Appendix 4. Gregorian calendar dates of observance of Ramadan in our study[55]

Gregorian calendar dates	Islamic year
27 November 2000 – 26 December 2000	1421
16 November 2001 – 15 December 2001	1422
6 November 2002 – 4 December 2002	1423
26 October 2003 – 24 November 2003	1424
15 October 2004 – 13 November 2004	1425
4 October 2005 – 2 November 2005	1426
24 September 2006 – 22 October 2006	1427
13 September 2007 – 12 October 2007	1428
1 September 2008 – 30 September 2008	1429
22 August 2009 – 19 September 2009	1430
11 August 2010 – 9 September 2010	1430

PART 2

Maternal nutrition status early in pregnancy



Chapter 6

Hyperemesis gravidarum and placental dysfunction disorders

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Submitted

Abstract

Objectives

To study whether hyperemesis gravidarum (HG) is associated with placental dysfunction disorders and neonatal outcome.

Methods

A prospective cohort study was conducted in a maternal and child health primary care referral center, Budi Kemuliaan Hospital and its branch, in Jakarta, Indonesia. 2,252 pregnant women visiting the hospital for regular antenatal care visits from July 2012 until October 2014 were included at their first clinic visit. For women without, with mild and with severe hyperemesis, placental dysfunction disorders (gestational hypertension, preeclampsia, stillbirth, miscarriage), neonatal outcomes (birth weight, small for gestational age [SGA], low birth weight [LBW], Apgar score at 5 minutes, gestational age at delivery) and placental outcomes (placental weight and placental-weight-to-birth-weight ratio [PW/BW ratio]) were studied.

Results

Compared to newborns of women without HG, newborns of women with severe hyperemesis had 172 g lower birth weight in adjusted analysis (95%CI -333.26; -10.18; p=0.04). There were no statistically significant effects on placental dysfunction disorders or other neonatal outcome measures.

Conclusions

Hyperemesis gravidarum seems not to induce placental dysfunction related disorders but it does, if severe, lead to lower birth weight.

Introduction

Nausea and vomiting are common and usually benign symptoms of primarily the first trimester of pregnancy. The onset of nausea correlates with the level of human chorionic gonadotropin (hCG), which typically raises within four weeks after the last menstrual period, peaking at approximately nine weeks of gestation.[1] Sixty percent of nausea cases resolve by the end of the first trimester and 91% by twenty weeks of gestation.[1] Hyperemesis gravidarum is at the severe end of the nausea spectrum and according to The International Statistical Classification of Disease and Related Health Problems (ICD-9), is defined as 'persistent and excessive vomiting starting before the end of the 22nd week of gestation'. Hyperemesis gravidarum is clinically classified as mild or severe, depending on associated metabolic disturbances such as carbohydrate depletion, dehydration and electrolyte imbalance. Its incidence is estimated at 0.3% to 1.5% of all live births but is unevenly distributed on a global level.[2–6] Asian women, for instance are more likely to suffer from hyperemesis than Caucasian women.[3,5] Hyperemesis greatly affects maternal well-being and quality of life[7,8] and is among the most common reasons for pregnancy-associated hospitalization.[6,9,10]

Relatively little is known about the etiology of hyperemesis.[6] Causal roles of sex hormones, thyroid hormones, *H. pylori* infections and paternal genes have been suggested, although consensus has not been reached.[6,11] The level of hCG is associated with the occurrence and severity of this disease, and more common and severe among women with higher levels of hCG (e.g. occurring in multiple- or molar pregnancy).[12,13] While benign nausea and vomiting in early pregnancy are closely related to temporarily increased hCG levels, it has been argued that in women with hyperemesis, the persistently high hCG level dysregulates normal stimulation of trophoblast migration, which consequently alters placentation.[14–16] Ultimately, abnormal placentation could lead to placental dysfunction that clinically manifests as gestational hypertension, pre-eclampsia, as well as miscarriage, stillbirth and intra-uterine growth restriction (IUGR).[15,17–19] In particular, elevated hCG plasma levels in the second trimester are associated with development of these conditions.[13,20] Thus, hyperemesis gravidarum, occurring in first and early second trimester, could be an early pregnancy indicator of a process that results in symptomatic placental dysfunction later.

There is limited evidence about consequences of hyperemesis on maternal and offspring's health. Two large cohort studies in Scandinavian countries showed that hyperemesis was associated with higher risk of preeclampsia, lower birth weight and shorter gestational duration.[15,21] This was supported by several studies which suggested higher risks of low birth weight (LBW), small for gestational age (SGA), and preterm birth if mothers experienced hyperemesis.[22–24] However, another large study[25] and several smaller studies,[26,27] did not show such associations. Women who experience severe hyperemesis have a significantly reduced maternal caloric 'intake' and lose additional nutrients and electrolytes.[28] This state resembles fasting and often involves ketonuria, which is frequently tested by clinicians in women suspected of having hyperemesis.[29,30] Previous studies showed that placental efficiency changes in women exposed to famine. Increased placental weight in women who were pregnant during the Dutch Hungerwinter suggests that compensatory growth of the placenta can occur in situations where nutritional resources are lacking.[31,32] The same compensation might occur in women who experience severe hyperemesis gravidarum, however evidence is lacking.

Both hyperemesis and placental dysfunction constitute substantial maternal and neonatal health threats, particularly in the low and middle income countries where health care resources are limited.[33] Therefore, further exploration of a relation between hyperemesis and such disorders is warranted. We here investigate the relation between hyperemesis and placental dysfunction disorders (gestational hypertension, pre-eclampsia, miscarriage, and stillbirth), and neonatal outcomes, including birth weight, small for gestational age (SGA), Apgar score and gestational age at delivery.

Methods

Study population

We used a prospective cohort of 2,252 pregnant women in the private mother-child health Budi Kemuliaan Hospital and its branch (Budi Kemuliaan Petojo) in Jakarta, Indonesia. Pregnant women were recruited during first regular visits for antenatal care (ANC) between July 2012 and October 2014. All women who attended clinic visits were invited and asked to sign a written informed consent. Participants were examined and interviewed by midwives according to standard clinical care and followed up until delivery. This study was ethically approved by the Institutional Review Board of Budi Kemuliaan Hospital.

After enrolment, information regarding personal affairs, medical status and clinical information was obtained through interviews by midwives at ANC visits. This included socio-economic background of women and partners, women's medical history (including previous surgery, medication), current pregnancy (last menstrual period (LMP), pre-pregnancy weight), obstetrical history (parity, previous morbidity during pregnancy, previous mode(s) of delivery), and family history of disease. Clinical information at each ANC visit included weight of the mother, blood pressure, temperature, occurrence of hyperemesis gravidarum, and presence of proteinuria.

Hyperemesis gravidarum exposure measurement

Hyperemesis gravidarum was diagnosed by midwives during routine ANC visits. Details about duration of complaints, weight loss, metabolic disturbances and associated hospitalization were recorded. For analysis, women were classified as those without, with mild or with severe hyperemesis gravidarum (women with > 5 % weight loss compared to pre-pregnancy weight). Only women with hyperemesis diagnosed in the first or second trimester were included.[34]

Outcome measurements

Hypertensive disorders of pregnancy were classified by International Society for the study of Hypertension in Pregnancy (ISSHP) definitions.[35] Gestational hypertension was defined as systolic blood pressure of 140 mmHg or more and/or diastolic blood pressure of 90 mmHg or more at two occasions in a patient with no hypertension prior to 20 weeks of gestation. In women with gestational hypertension, proteinuria was diagnosed using a urine dipstick. Pre-eclampsia was diagnosed if women with gestational hypertension had proteinuria[35] or if there were also one or more convulsions (eclampsia).[36] Due to the low incidence of eclampsia, we analyzed eclampsia patients combined with the preeclampsia group. Chronic hypertension was defined as blood pressure exceeding 140/90 mm Hg before pregnancy or before 20 weeks gestation,[37] and only when found elevated at ≥ 2 occasions. Women who were diagnosed with gestational hypertension or preeclampsia received treatment according to the standard hospital protocols.

Abortion or miscarriage was defined as fetal loss before 23 weeks, and stillbirth as intrauterine fetal demise after 23 weeks.[38,39] Small for Gestational Age (SGA) was defined as birth weight at a particular gestational age was below the 10th percentile of United States National Reference for Fetal Growth.[40] Low Birth Weight (LBW) was a birth weight below 2500 grams.[41] Apgar scores were measured at 1, 5, and 10 minutes after birth.[42] Reported outcome for Apgar score was the score 5 minutes postpartum. Prematurity was defined as birth before 37 weeks of gestation. Gestational age at delivery was calculated in days by subtracting the first day of last menstrual period (LMP) from the date of admission to delivery room/operation theatre. Confirmation of gestational age using ultrasound scan was not done since only limited women had access to this examination. Nine women with gestational duration > 46 weeks (probably due to inaccuracy of gestational age calculation), were excluded.

After birth, placental dimensions (weight, length, width) and birth weight were measured using a standardized method by midwives. The placenta-to-birth weight ratio was calculated.[28,31] After discharge from hospital, active follow-up was terminated. Analyses on neonatal outcomes were based on singleton live birth pregnancies, and therefore multiple pregnancies, stillbirths and miscarriages were excluded from analysis.

Confounding variables

Analyses of the associations between hyperemesis and placental dysfunction disorders were adjusted for socio-economic status (family income), second-hand smoking exposure, maternal age at delivery, primigravidity and pre-pregnancy body mass index (BMI).

Data analysis

Baseline analysis was stratified for women who were diagnosed with hyperemesis (severe/mild/no hyperemesis), and group differences were evaluated with Chi-square, one-way-ANOVA or Kruskal-Wallis tests where appropriate. For skewed data, we reported median and interquartile range (IQR). Main results were expressed as crude and adjusted linear regression coefficients and odds ratios from logistic regression, with corresponding 95% confidence intervals and p-values in tables. All statistical analyses were run using IBM SPSS (version 22 for Mac).

Results

Study population

Of 2,252 participants, 400 were diagnosed with hyperemesis gravidarum (18.9%). Of the diagnosed, 94 experienced weight loss varying from 1 to 13 kg. There were 1,833 women without, 354 with mild and 46 women with severe hyperemesis (weight loss > 5%). Subject selection is shown in **Figure 1**. The mean age of participants was 28.3 years, 27% were primigravida.

Table 1 shows the baseline characteristics. Family income categories differed statistically significantly across exposure groups. Women with hyperemesis attended the first ANC visit earlier in gestation than women without hyperemesis. Compared to women with no or mild hyperemesis those with severe hyperemesis more often had lower education, higher pre-pregnancy BMI, non-smoking partners, multiparous history, vaginal deliveries and female babies, but none were statistically significant. There were no differences in age, partner’s education, and history of hypertension, diabetes, hypertensive disorders or miscarriages.

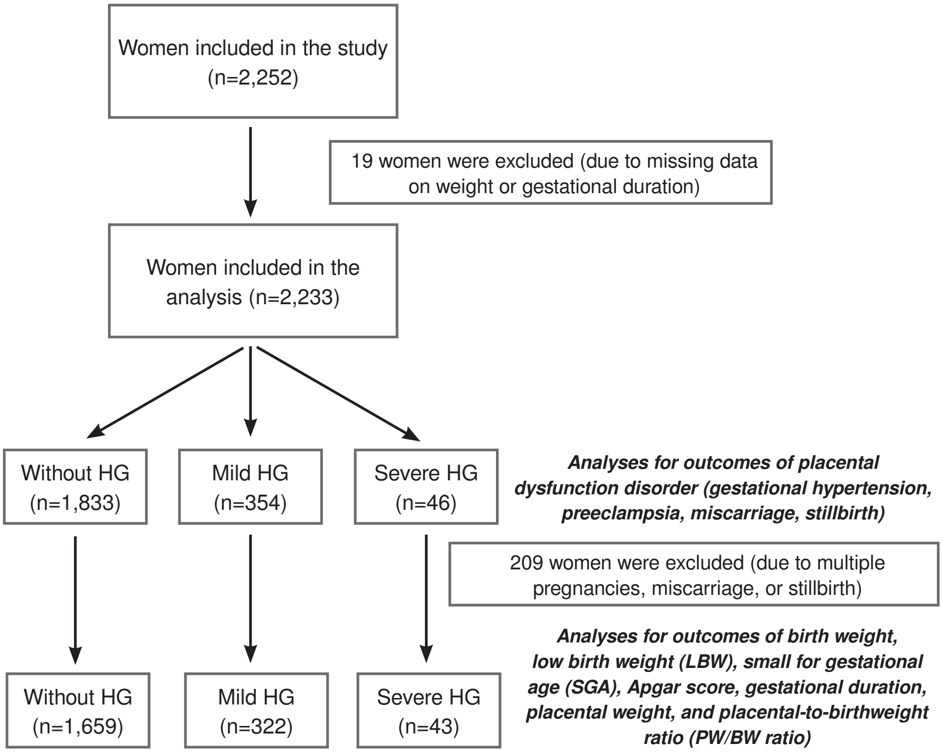

Figure 1. Flowchart of subject selection

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HG exposure and outcomes

Tables 2a, 2b and 2c show associations between hyperemesis and placental dysfunction disorders, neonatal outcomes and placental measures. Mild and severe hyperemesis were not associated with placental dysfunction disorders. After adjustment, babies of women with severe hyperemesis were on average 172 grams lighter at birth than unexposed babies. No associations were found for SGA and LBW offspring, or other adverse neonatal outcomes. Women with mild hyperemesis had slightly lighter placentas while women with severe hyperemesis had heavier placentas, although none were statistically significant. Likewise, placental-weight-to-birth weight ratios (PW/BW ratio) were lower with mild hyperemesis and higher with severe hyperemesis, although not statistically significant.

Additional analyses regarding the effect of absolute and relative maternal weight change during the early part of pregnancy were also conducted within women with hyperemesis gravidarum. However, no associations were found between early pregnancy weight change with later placental dysfunction disorders and neonatal or placental outcomes.

Table 1. Baseline characteristics of pregnant women participating in our cohort by HG exposure status

		HG exposure			p-value
		No HG (n=1,833)	**Mild HG (n=354)**	**Severe HG (n=46)**	
Demographic characteristics	Age (years), median (IQR)	28.09 (8.7)	28.13 (7.4)	28.36 (9.1)	0.62
	Women's education[1]				0.15
	Low	413 (22.6)	73 (20.6)	10 (22.2)	
	Intermediate	1123 (61.4)	208 (58.8)	31 (68.9)	
	High	292 (16.0)	73 (20.6)	4 (8.9)	
	Partner's education[1]				0.23
	Low	323 (17.7)	46 (13.0)	9 (20.0)	
	Intermediate	1199 (65.6)	241 (68.1)	30 (66.7)	
	High	305 (16.7)	67 (18.9)	6 (13.3)	
	Family income[2]				0.01
	< 72 USD	197 (10.8)	26 (7.3)	5 (10.9)	
	72 – 180 USD	799 (43.6)	169 (47.7)	18 (39.1)	
	180 – 360 USD	529 (28.9)	126 (35.6)	16 (34.8)	
	> 360 USD	124 (6.8)	17 (4.8)	3 (6.5)	
	Refused to answer	182 (9.9)	16 (4.5)	4 (8.7)	
Health characteristics	Pre-pregnancy BMI, median (IQR)	22.0 (5.5)	22.2 (5.5)	23.2 (6.6)	0.59
	Chronic Hypertension	27 (1.5)	5 (1.4)	0	0.71
	History of Diabetes	11 (0.6)	2 (0.6)	0	0.87
	Partner's smoking status				0.12
	Smokes daily	394 (48.6)	70 (50.7)	8 (36.4)	
	Smokes occasionally	113 (13.9)	12 (8.7)	1 (4.5)	
	Doesn't smoke	304 (37.5)	56 (40.6)	13 (59.1)	
Obstetric characteristics	Primigravida	480 (38.0)	112 (41.5)	12 (34.3)	0.50
	Gestational age (weeks) at first visit, median (IQR)	16 (20)	9 (4.50)	10 (5.0)	<0.01
	Reported complications in previous pregnancy				
	No	1070 (84.6)	233 (86.0)	29 (82.9)	0.80
	IUGR/ SGA	3 (0.2)	0	0	0.70
	Hypertensive disorder in pregnancy	44 (2.4)	11 (3.1)	1 (2.2)	0.74
	Miscarriage	136 (7.4)	28 (7.9)	2 (4.3)	0.69
	Female baby	651 (46)	133 (46.5)	22 (59.5)	0.27
	Multiple pregnancy	27 (1.9)	1 (0.4)	0	0.12
	Mode of delivery				0.08
	Vaginal	871 (61.3)	168 (58.9)	24 (64.9)	
	Instrumental	33 (3.1)	11 (3.9)	4 (10.8)	
	Caesarean section	506 (35.6)	106 (37.2)	9 (24.3)	

Abbreviations: HG Hyperemesis gravidarum; USD United States Dollar; IUGR intra uterine growth restriction; SGA small for gestational age; IQR inter-quartile range.

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Results are median (inter-quartile range) or numbers (percentage); For continuous outcome variables Kruskall-Wallis test was used, for categorical variables, Pearson's chi-square test. 1 *Low education*= no education, elementary school, senior high school; *Intermediate education*= senior high school or above; *High education*= university; 2 Mean monthly total family income, estimated by the patient

Table 2a. Hyperemesis gravidarum severity groups and placental dysfunction disorders

	n (%)	Crude model		Adjusted model[1]	
		OR (95% CI)	p-value	OR (95% CI)	p-value
Gestational hypertension					
No HG	242 (17.7)	Reference		Reference	
Mild HG	39 (14.5)	0.79 (0.55; 1.13)	0.20	0.87 (0.58; 1.31)	0.51
Severe HG	4 (10.8)	0.56 (0.20; 1.60)	0.28	0.64 (0.22; 1.87)	0.41
Preeclampsia					
No HG	77 (6.0)	Reference		Reference	
Mild HG	15 (5.8)	0.97 (0.55; 1.72)	0.92	0.91 (0.47; 1.76)	0.78
Severe HG	2 (5.4)	0.90 (0.21; 3.81)	0.89	0.99 (0.23; 4.34)	0.99
Miscarriage					
No HG	136 (7.4)	Reference		Reference	
Mild HG	28 (7.9)	1.07 (0.70; 1.64)	0.75	1.17 (0.60; 2.70)	0.64
Severe HG	2 (4.3)	0.57 (0.13; 2.37)	0.44	-	
Stillbirth					
No HG	12 (0.8)	Reference		Reference	
Mild HG	3 (1.0)	1.24 (0.35; 4.43)	0.74	1.41 (0.37; 5.40)	0.62
Severe HG	1 (2.7)	3.26 (0.41; 25.72)	0.26	4.18 (0.50; 35.10)	0.19

Abbreviations: HG, hyperemesis gravidarum; OR, odds ratio. 1. Adjusted model: adjusted for socio-economic status (as reflected by income), smoking status, gravidity, maternal age and pre-pregnancy BMI.

Table 2b. Hyperemesis gravidarum severity groups and neonatal outcome

	Values	Crude model		Adjusted model[1]	
		OR (95% CI)	p-value	OR (95% CI)	p-value
Birthweight (g)[3,4]					
No HG	3100 (476.6)	Reference		Reference	
Mild HG	3116 (464.4)	16.36 (-44.82; 77.54)	0.60	-4.17 (-68.36; 60.02)	0.90
Severe HG	3046 (485.7)	-53.86 (-211.15; 103.43)	0.50	-171.72 (-333.26; -10.18)	0.04
SGA[3]					
No HG	212 (16.6)	Reference		Reference	
Mild HG	54 (19.8)	1.24 (0.89; 1.73)	0.20	1.29 (0.87; 1.91)	0.20
Severe HG	6 (17.6)	1.08 (0.44; 2.64)	0.87	1.74 (0.68; 4.44)	0.25
LBW[3]					
No HG	101 (7.4)	Reference		Reference	
Mild HG	21 (7.5)	1.03 (0.63; 1.67)	0.92	1.36 (0.74; 2.51)	0.32
Severe HG	2 (5.6)	0.74 (0.18; 3.13)	0.68	1.44 (0.33; 6.33)	0.63

Gestational age at delivery (in days)[5]					
No HG	274 (17)	Reference		Reference	
Mild HG	275 (13)	0.27 (-2.22; 2.75)	0.83	-0.03 (-2.60; 2.54)	0.98
Severe HG	276 (19)	0.48 (-5.99; 6.96)	0.88	-2.20 (-8.81; 4.42)	0.52
Apgar score[3,5]					
No HG	9 (0)	Reference		Reference	
Mild HG	9 (0)	-0.06 (-0.17; 0.04)	0.24	-0.08 (-0.19; 0.03)	0.15
Severe HG	9 (0)	-0.06 (-0.35; 0.23)	0.67	-0.09 (-0.39; 0.21)	0.56

Abbreviations: CI, confidence interval; HG hyperemesis gravidarum; g, grams; LBW low birthweight, SGA small for gestational age. 1 Adjusted model: adjusted for socio-economic status (as reflected by income), smoking status, gravidity, maternal age and pre-pregnancy BMI. 2 Results are expressed as linear regression coefficients (95% CI) in continuous outcomes or OR (95% CI) from logistic regression in case of dichotomous outcomes. 3 Multiple pregnancies, stillbirths and miscarriages were excluded from the analyses. 4. Values are means with standard deviations in case of continuous variables or numbers with percentage in case of frequencies. 5. Values are median with inter-quartile range.

Table 2c. Hyperemesis gravidarum severity groups and placental measures

	Values	Crude model		Adjusted model[1]	
		OR (95% CI)	**p-value**	**OR (95% CI)**	**p-value**
Placental weight (g)[3]					
No HG	536 (145.2)	Reference		Reference	
Mild HG	530 (102.3)	-5.98 (-42.92; 30.95)	0.75	-6.05 (-41.25; 29.15)	0.74
Severe HG	547 (98.4)	10.73 (-126.91; 148.38)	0.88	47.76 (-97.62; 193.14)	0.52
PW/BW ratio[3]					
No HG	0.18 (0.05)	Reference		Reference	
Mild HG	0.18 (0.05)	0.001 (-0.01; 0.01)	0.89	0.002 (-0.01; 0.02)	0.72
Severe HG	0.19 (0.05)	0.02 (-0.03; 0.06)	0.54	0.04 (-0.01; 0.09)	0.15

Abbreviations: CI, confidence interval; HG hyperemesis gravidarum; PW/BW ratio, placenta-to-birthweight ratio; g grams.
1 Adjusted model: adjusted for socio-economic status (as reflected by income), smoking status, gravidity, maternal age and pre-pregnancy BMI.
2 Results are expressed as linear regression coefficients (95% CI).
3 Multiple pregnancies, stillbirths and miscarriages were excluded from the analyses.

Discussion

This study shows that severe hyperemesis gravidarum is associated with a significant decrease in birth weight, but no associations between hyperemesis gravidarum and gestational hypertension, preeclampsia or other placental dysfunction disorders were observed. No links were found between hyperemesis gravidarum and neonatal outcomes, such as SGA and LBW.

Strengths of this study comprise the prospective design and the number of included women. This allowed for assessing both first and second trimester effects of hyperemesis gravidarum on neonatal outcomes and particularly birth weight, to give an impression of effects on the placenta, and evaluate early pregnancy weight change as a possible mediator of the effects. However, given the low incidence of placental dysfunction disorders, our study may have been too small for definite inference. Of participants, 18.8% had some form of hyperemesis, which is very high, but our estimates of severe hyperemesis do fit previous reports. Notably, women were recruited from a referral institute for mother and child health, and indeed some with hyperemesis were referred earlier in pregnancy. We do believe that specific referrals for hyperemesis, particularly those in early pregnancy, were for that complaint only, and not for some associated expectation of higher risk for adverse maternal or neonatal outcome, such that selection bias is unlikely. Missing information increased with longer follow-up, but was largely due to women who were temporarily provided antenatal care services and then referred back to primary care, a routine that we consider unrelated to the association of interest. Our self-report questionnaire information may contain measurement error, but as patients were unaware of the study aim, such error was likely random.

Our results do not show that severe hyperemesis gravidarum increases the risk of placental dysfunction disorders. A link between hyperemesis gravidarum and placental dysfunction disorders was first suggested in a 1991 case-control study reporting a 1.6 times higher risk for preeclampsia in women with severe vomiting.[43] Consistent with our findings, previous studies also showed no increased risks of placenta dysfunction disorders with exposure to hyperemesis.[44,45] The largest cohort to date with 1,155,033 pregnancies, of which 13,287 were complicated by hyperemesis leading to hospital admission, showed a slightly increased risk for preeclampsia, and a higher risk for preterm preeclampsia when hyperemesis occurred in the second trimester.[15] It has been suggested that differences in outcome in hyperemetic pregnancies are explained by maternal characteristics such as (gestational) hypertension, (gestational) diabetes, and primiparity.[46] A general problem in comparing findings on relations between hyperemesis and placental dysfunction disorders is the lack of a widely accepted definition of 'severe' hyperemesis. Working criteria range from hyperemesis requiring hospitalization or vomiting with associated metabolic disturbances to classifications purely based on caregiver's diagnosis. In this study, the presence of (maternal) weight loss of more than 5% (compared to the weight prior to diagnosis) was used as criterion to classify severity of hyperemesis.

We showed a (adjusted) 172 g lower birth weight in offspring of women with severe hyperemesis, which was not explained by gestational duration. This was in accordance with several previous reports.[22,46-49] In this study, we did not find any association between hyperemesis and SGA or LBW, although these appeared more common in women with severe hyperemesis. Even though exposed babies were born smaller, it appeared that their chances of passing the threshold of the 10th percentile for the gestational age (SGA) or 2500 g for birth weight (LBW) were equal to the non-exposed. This agrees with several previous reports, including the largest study on neonatal outcome to date.[21,50,27] Differences in exposure definition and classification of its severity may contribute to the variation of findings.

Previous research on the association between hyperemesis and placental measures is limited. In the present study, we detected no effects on placental weight and placenta-weight-to-birth weight ratio (PW/BW ratio). Women with severe hyperemesis, however, had heavier placentas and higher of PW/BW ratio, although not statistically significant. Heavier placentas and higher PW/BW ratio were previously reported, but only for female offspring.[28] Studies on famine and placental weight and PW/BW ratio suggest that with low caloric intake, the placenta compensatorily grows, probably to maintain adequate fetal nutrition.[31,32] This might also occur with hyperemesis and, therefore, further research on the placenta is warranted; especially since there is evidence that compensatory growth of the placenta is associated with cardiovascular problems in later life.[28,51]

Further research requires clear HG definition and severity criteria. Large cohort studies will be needed to estimate relations between hyperemesis and placental dimensions, and rare outcomes, such as eclampsia and stillbirth. Conducting follow up studies of women with severe hyperemesis gravidarum will give a better insight in possible long term effects of hyperemesis on the health of children born out of hyperemetic pregnancies.

Conclusion

Hyperemesis gravidarum is an invalidating disease in early pregnancy, associated with hospitalizations, use of medication and a poorer quality of life. Our findings indicate no relevant impact of hyperemesis gravidarum on placental dysfunction disorders, although if severe, it could lead to a lower birth weight. A close monitoring and intervention against poor pregnancy weight gain in women with severe hyperemesis remains vital for offspring's health.

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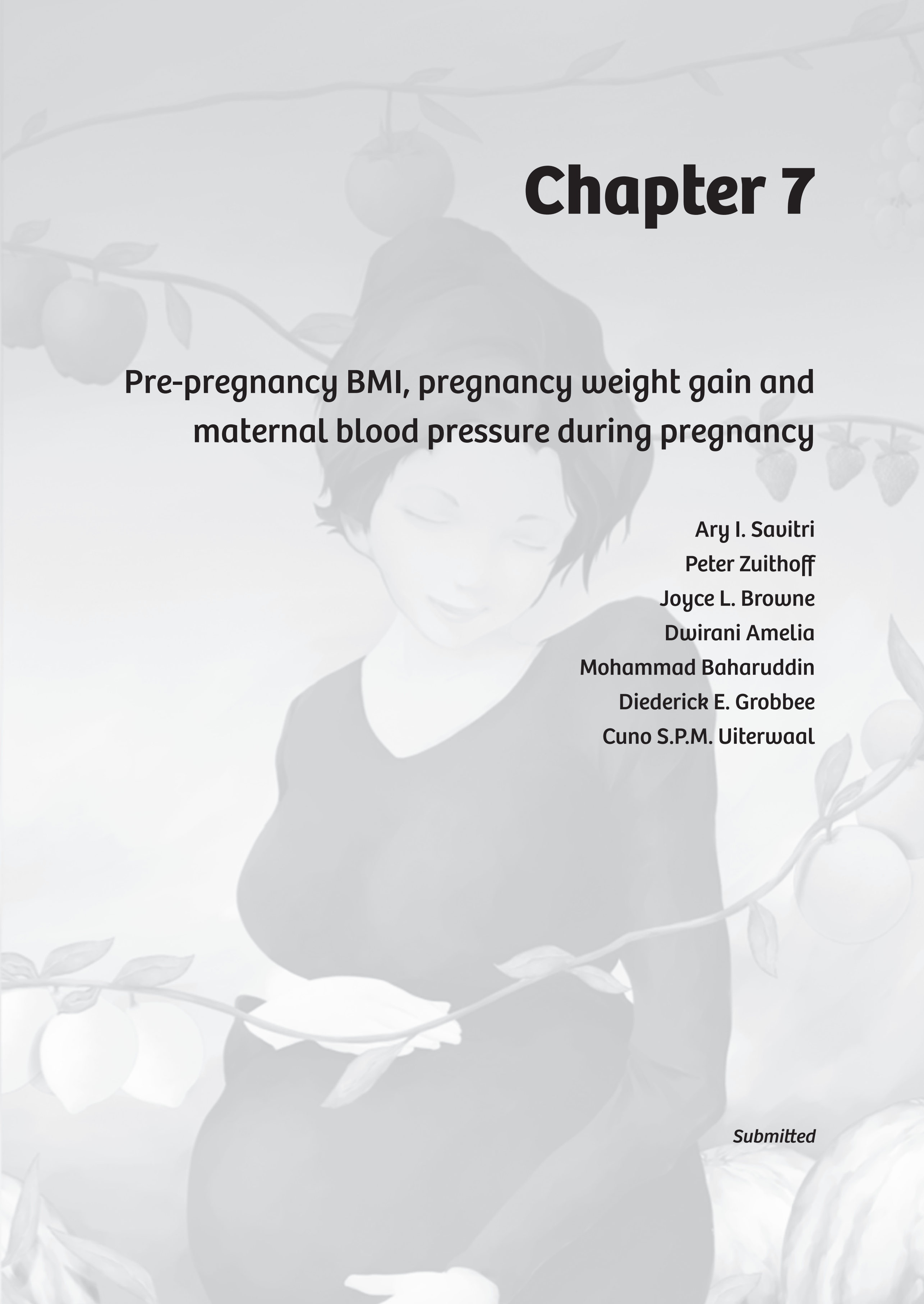

Chapter 7

Pre-pregnancy BMI, pregnancy weight gain and maternal blood pressure during pregnancy

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Submitted

Abstract

Objectives

To evaluate if pre-pregnancy BMI determines blood pressure throughout pregnancy and to explore the role of pregnancy weight gain in this association. In addition, the effects of pre-pregnancy BMI and pregnancy weight gain on the occurrence of gestational hypertension and preeclampsia were investigated.

Methods

A prospective cohort study was conducted in a maternal and child health primary care referral center, Budi Kemuliaan Hospital and its branch, in Jakarta, Indonesia. There were 2,252 pregnant women visiting the hospital for regular antenatal care visits from July 2012 until April 2015. Pre-pregnancy BMI (kg/m^2) was based on self-reported pre-pregnancy weight and measured height at first visit. Pregnancy weight gain was calculated as weight at the day of delivery and compared to the pre-pregnancy weight. Systolic blood pressure (SBP) and diastolic blood pressure (DBP) were measured during pregnancy at every visit. Linear mixed models were used to analyze this relation with repeated blood pressure measures as outcome and pre-pregnancy BMI as predictor. When looking at gestational hypertension and preeclampsia as outcomes, (multiple) logistic regression was used in the analysis.

Results

Independent of pre-pregnancy BMI, SBP and DBP increased by 0.99 mmHg/month and 0.46 mmHg/month, respectively. Higher pre-pregnancy BMI was associated with higher pregnancy SBP (0.25 $mmHg/kg/m^2$; 95% CI 0.17 to 0.34; $p<0.01$) and DBP (0.18 $mmHg/kg/m^2$; 0.13 to 0.24; $p<0.01$) in adjusted analysis. Every 1 kg/m^2 higher pre-pregnancy BMI was associated with 6 and 9 percent higher odds for gestational hypertension (adjusted odds ratio [aOR] 1.06; 95% CI 1.03 to 1.09; $p<0.01$) and preeclampsia (aOR 1.09; 1.04, 1.14; $p<0.01$). Accounting for pregnancy weight gain did not attenuate these associations.

Conclusions

Pre-pregnancy BMI determines the level, not the change, of blood pressure in pregnancy and is linked to higher odds for gestational hypertension and preeclampsia, independent of gestational weight gain.

Introduction

Over the last decades, the prevalence of overweight and obesity among women in reproductive age has increased tremendously both in the developed and developing countries.[1] High body mass index (BMI), together with high systolic blood pressure, have been known as the leading risk factors for mortality and disability-adjusted life-years (DALY) in women worldwide,[2] mainly through manifestation of cardiovascular diseases and metabolic disorders.[3] Being overweight or obese also puts women at a higher risk for developing various pregnancy complications, including gestational hypertension and preeclampsia.[4–10] As compared to normal weight women, overweight and obese women had 3 to 5 times increased risk for preeclampsia.[3]

Higher baseline BMI recorded at the first antenatal visit has also consistently been linked to higher risk of hypertensive disorders of pregnancy.[11,12] Several studies have reported that pre-pregnancy BMI influences systolic and diastolic blood pressure (SBP and DBP) levels during pregnancy.[5,13–15] However, studies that reported the effect of pre-pregnancy BMI on the rate of change in SBP and DBP are limited. Furthermore, the role of weight gain on the relation between pre-pregnancy BMI and blood pressure during pregnancy is also less consistent. Several studies reported that excessive pregnancy weight gain was associated with higher risk of gestational hypertension and preeclampsia[5,15,16], but some other studies did not find such associations.[17,18] It is also unclear whether the relation between obesity and hypertensive disorders of pregnancy was mediated by excessive weight gain, as overweight or obese women were shown more likely to experience excessive weight gain as compared to women with normal BMI.[19]

In the present study, we investigated the effect of pre-pregnancy BMI, as a proxy for maternal adiposity, on several blood pressure measures throughout pregnancy and on the occurrence of gestational hypertension and preeclampsia. Furthermore, we explored the influence of gestational weight gain in these associations.

Methods

Study population

This study was based on a prospective cohort of 2,252 pregnant women in the private mother-child health Budi Kemuliaan Hospital and its branch (Budi Kemuliaan Petojo) in Jakarta, Indonesia. Women were recruited at first visits for antenatal care (ANC) between July 2012 and October 2014 and followed up until delivery, the last in April 2015. All women who attended ANC visits were invited and asked to sign written informed consent. This study was ethically approved by the Institutional Review Board of Budi Kemuliaan Hospital.

Midwives obtained interview information regarding women's demography including age, socio-economic background, education and employment status. Women's education was categorized as low if the women had finished elementary or junior high school, medium if they finished senior high school, or high if they attained education from university. Women's employment status was classified as not working, working in a formal job, or working in a non-formal job (trader, nanny, cleaning service, etc). Women's clinical data included medical history, information on current pregnancy (last menstrual period (LMP), pre-pregnancy weight, and weight at every ANC visit), women's obstetrical history (parity, previous complications, previous mode(s) of delivery), and family history of disease.

Anthropometric measures

Pre-pregnancy weight was self-reported at the first ANC and height was measured using standardized statometers. Pre-pregnancy BMI was then calculated as pre-pregnancy weight in kilograms divided by the square of height in meters. Women's weight was measured at each ANC visit using standard scales.

Outcome measurements

Women's systolic blood pressure (SBP) and diastolic blood pressure (DBP) were routinely measured by midwives at every ANC visit. SBP and DBP were measured once per visit in seated position, after a minimum 30 minutes rest, using a mercury sphygmomanometer. Gestational hypertension and preeclampsia were classified according to the definitions from the International Society for the study of Hypertension in Pregnancy (ISSHP).[20] Gestational hypertension was defined as systolic blood pressure of 140 mmHg or more and/or diastolic blood pressure of 90 mmHg or more at two occasions in a patient with no hypertension prior to 20 weeks of gestation. Preeclampsia is defined as gestational hypertension with proteinuria.

Confounding variables and intermediate variables

Women's age, parity, secondhand smoking exposure, and monthly family income (proxy for socio-economic status) were a priori considered as possible confounders. Age was calculated as the difference between the dates of birth and of women's first ANC visit. Parity was classified as nulli- or multiparous. Data on secondhand smoking exposure was used instead of active smoking because few of these pregnant women smoke. Monthly family income was asked to the women as an estimate range. Gestational age at delivery was derived from the date of last menstrual period and the date of delivery.

As a possible intermediate factor, pregnancy weight gain was calculated as weight at the day of delivery subtracted by pre-pregnancy weight in kilograms.

Data analysis

The baseline characteristics were described by pre-pregnancy BMI tertiles and group differences were tested using Chi-square, one-way-ANOVA or Kruskal-Wallis tests where appropriate. Skewed data were reported as median and interquartile range (IQR). Associations between pre-pregnancy BMI and blood pressures were analyzed using (un-) adjusted linear mixed models for repeated measures, with corresponding 95% confidence interval and p values.

Furthermore, to investigate whether pregnancy weight gain acted as an intermediate factor in this association, we developed an explanatory model which included variables on pregnancy weight gain. Analyses about the effects of pre-pregnancy BMI on women's risk of developing gestational hypertension and preeclampsia were performed using (multiple) logistic regression.

The pattern of blood pressure changes during pregnancy was plotted for each pre-pregnancy BMI tertile by fitting (adjusted) linear mixed models with SBP and DBP as the outcomes, separately. The estimated marginal means of SBP and DBP at 13, 16, 19, 22, 25, 28, 31, 34, 37, and 39 weeks of gestation were used to describe the blood pressure trajectory during pregnancy. The baseline gestational age was set at 13 weeks because this was the median gestational age of the first measurement at the antenatal care visit. Thirty-nine weeks was set as the last BP measurement since this was the median gestational age at delivery. The relation between blood pressure and gestational age (time) was assumed to be linear over the course of pregnancy, as our main goal in this analysis was the mutual ranking of women's blood pressure. In addition, the pattern of weight increase during pregnancy was also presented separately for women in each pre-pregnancy BMI tertile using the same approach. All statistical analyses were performed using IBM SPSS (version 21 for Windows).

Results

Figure 1 provides the study flowchart. Due to missing data on pre-pregnancy BMI, the analysis concerned 2,031 women. Baseline characteristics of women according to tertiles of pre-pregnancy BMI are presented in **Table 1**. As compared to the highest BMI tertile, women in the first and second tertile were younger, more likely nulliparous, and had higher total pregnancy weight gain. Fewer women in the lowest tertile had attained high education. More women in the highest tertile women did not work or worked in a non-formal sector. Monthly family income slightly differed with pre-pregnancy BMI. No difference was found with respect to women's smoking status, secondhand smoking exposure, and gestational age at the first visit and at delivery.

Table 1. Baseline characteristics of women, by tertiles of pre-pregnancy BMI

Characteristics	Tertiles of pre-pregnancy BMI			p-value
	Low	Middle	High	
Number (%)	675 (33.2)	684 (33.7)	672 (33.1)	
BMI (kg/m²), median (inter-quartile range)	18.6 (2.4)	22.1 (1.8)	26.7 (3.9)	<0.01[1]
Age (years), mean (SD)	26.6 (5.6)	28.6 (5.6)	30.5 (5.7)	<0.01[2]
Women's education				0.03[3]
Low education (%)	170 (25.2)	144 (21.1)	132 (19.7)	
Middle education (%)	413 (61.3)	415 (60.8)	415 (61.9)	
High education (%)	91 (13.5)	124 (18.2)	123 (18.4)	
Women's employment status				0.01[3]
Not working (%)	385 (57.2)	398 (58.6)	428 (63.9)	
Non-formal job (%)	44 (6.5)	52 (7.7)	57 (8.5)	
Formal job (%)	244 (36.3)	229 (33.7)	185 (27.6)	
Monthly family income				0.01[3]
Less than 1 million IDR (%)	68 (10.1)	63 (9.2)	68 (10.1)	
1-2.5 million IDR (%)	320 (47.4)	308 (45.0)	291 (43.3)	
2.5-5 million IDR (%)	204 (30.2)	216 (31.6)	203 (30.2)	
More than 5 million IDR (%)	29 (4.3)	59 (8.6)	45 (6.7)	
Refused to answer (%)	54 (8.0)	38 (5.6)	65 (9.7)	
Nulliparity (%)	251 (55.8)	182 (36.6)	132 (25.7)	<0.01[3]
Women's smoking status				0.46[3]
Yes, sometimes (%)	3 (1.1)	2 (0.7)	3 (1.0)	
Yes, everyday (%)	4 (1.5)	1 (0.3)	1 (0.3)	
Partner's smoking status				0.62[3]
Yes, sometimes (%)	42 (15.3)	40 (14.0)	35 (11.8)	
Yes, everyday (%)	125 (45.6)	136 (47.7)	153 (51.5)	
Total pregnancy weight gain (kg), mean (SD)	14.3 (7.0)	12.8 (6.5)	10.2 (7.1)	<0.01[2]
Gestational age at first visit (weeks), median (inter-quartile range)	13.7 (17.0)	12.1 (17.3)	12.7 (16.8)	0.27[1]
Gestational age at delivery (weeks), median (inter-quartile range)	39.1 (2.3)	39.3 (2.1)	39.3 (2.3)	0.10[1]

BMI, body mass index; SD,standard deviation. [1] Kruskal-Wallis test, [2] ANOVA,[3] Chi square test

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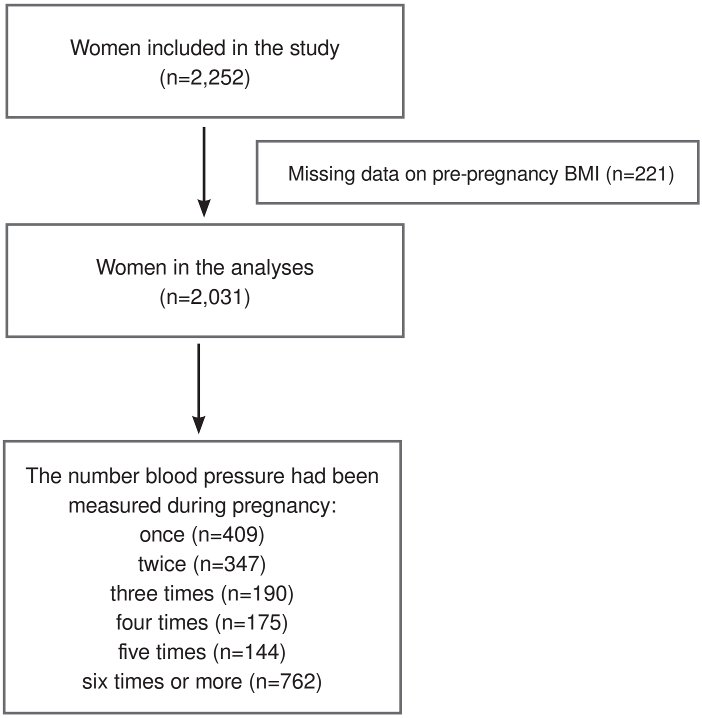

Figure 1. Flowchart of study participants

Table 2 shows that higher pre-pregnancy BMI was associated with a significantly higher systolic and diastolic blood pressure, both crude and confounding adjusted. The explanatory model shows that further accounting for total pregnancy weight gain did not change associations. Adjustments for other measures of weight gain were also performed, by adding a variable of weight gain measured at each antenatal care visit or a variable of weight gain-for gestational age into the adjusted model. However, the use of these measures did not change the findings compared to the presented measurement for pregnancy weight gain (data not shown).

In **Supplement 1**, we present the effect estimates if twin pregnancies were excluded from the analysis (n=26). There were no significant differences found as compared to the analysis with twin pregnancies included.

There were 264 women (13.0 %) in our study who were diagnosed with gestational hypertension and 81 women (4.0 %) with preeclampsia. In **Table 3**, the associations between pre-pregnancy BMI and occurrence of gestational hypertension and preeclampsia were presented. Higher pre-pregnancy BMI were associated with higher odds for gestational hypertension and preeclampsia, both in the crude and adjusted model. Adjustment for total pregnancy weight gain in the explanatory model did not attenuate the associations.

Table 2. Effect of pre-pregnancy BMI on systolic and diastolic blood pressure during pregnancy

		Coefficient	95% CI	p-value
Systolic blood pressure	Crude model	0.32	0.24, 0.40	<0.01
	Adjusted model[a]	0.25	0.17, 0.34	<0.01
	Explanatory model[b]	0.41	0.15, 0.66	<0.01
Diastolic blood pressure	Crude model	0.22	0.16, 0.27	<0.01
	Adjusted model[a]	0.18	0.13, 0.24	<0.01
	Explanatory model[b]	0.27	0.08, 0.45	<0.01

BMI, body mass index; CI, confidence interval. Results are expressed as mixed model coefficients of systolic and diastolic blood pressure in mmHg for every 1 kg/m^2 increase in pre-pregnancy BMI.
[a]Adjusted for maternal age, parity, secondhand smoking exposure, and socio-economic status (monthly family income)
[b]As [a] and for total pregnancy weight **gain** (difference between maternal weight measured at the day of delivery from the pre-pregnancy weight)

Table 3. Effect of pre-pregnancy BMI on the risk for developing gestational hypertension and preeclampsia

		OR	95% CI	p-value
Gestational hypertension	Crude model	1.07	1.04, 1.10	<0.01
	Adjusted model[a]	1.06	1.03, 1.09	<0.01
	Explanatory model[b]	1.10	1.05, 1.14	<0.01
Preeclampsia	Crude model	1.09	1.05, 1.14	<0.01
	Adjusted model[a]	1.09	1.04, 1.14	<0.01
	Explanatory model[b]	1.19	1.11, 1.27	<0.01

BMI, body mass index; CI, confidence interval; OR, odds ratio. Results are expressed as OR (95% CI) for every 1 kg/m^2 increase in pre-pregnancy BMI from (multiple) logistic regression.
[a]Adjusted for maternal age, parity, secondhand smoking exposure, and socio-economic status (monthly family income)
[b]As [a] and for total pregnancy weight **gain** (difference between maternal weight measured at the day of delivery from the pre-pregnancy weight)

Throughout pregnancy, SBP was estimated to increase by 0.99 mmHg/month while DBP increased by 0.46 mmHg/month in all women, independent of their pre-pregnancy BMI tertiles and other possible confounders. **Figure 2** shows that mean blood pressures were consistently higher with higher pre-pregnancy BMI, although the pace of increase did not seem to be affected. The lines that depict the pattern of change in SBP and DBP during pregnancy appeared to be parallel in women from different pre-pregnancy BMI tertiles. In addition, no interaction was found between pre-pregnancy BMI and gestational age (p = 0.91 in the confounder adjusted model).

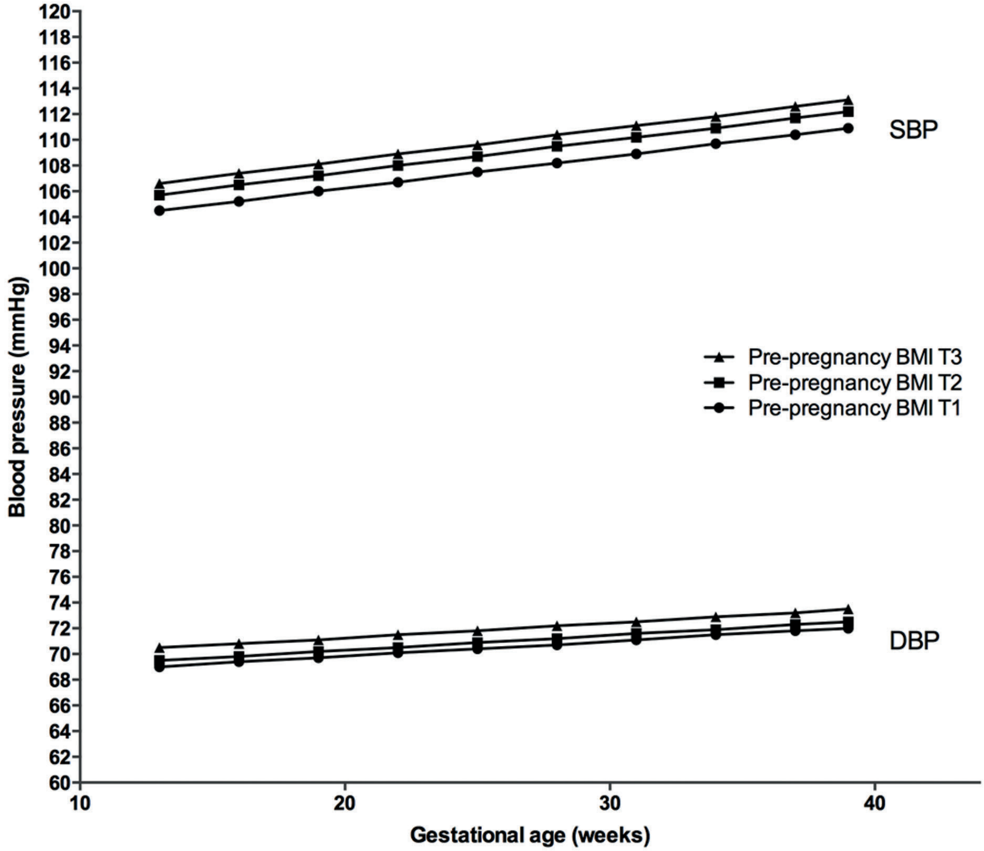

Figure 2. Effect of prepregnancy BMI tertiles on systolic and diastolic blood pressure during pregnancy
DBP, diastolic pressure; SBP, systolic blood pressure; T1, first tertile (low); T2, second tertile (middle), T3, third tertile (high)
Estimated marginal means of systolic and diastolic blood pressure measured during pregnancy were plotted, adjusted for maternal age, parity, secondhand smoking exposure, and socio-economic status (monthly family income)

Figure 3 shows the pattern of weight gain in pregnancy by pre-pregnancy BMI tertile. The level of pregnancy weight gain differed according to women's pre-pregnancy BMI tertiles. The mean pregnancy weight gain of women in the first, second, and third tertile pre-pregnancy BMI were 14.3 kg, 12.8 kg, and 10.2 kg consecutively.

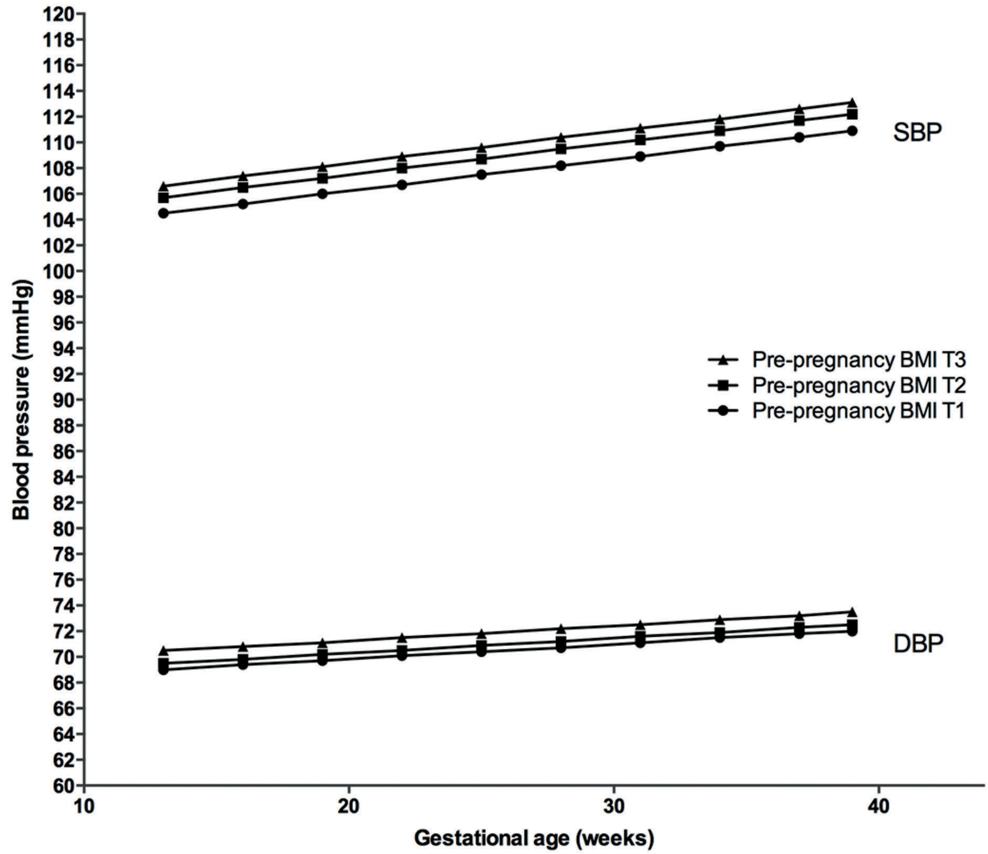

Figure 3. Increase in maternal weight during pregnancy according to pre-pregnancy BMI tertiles
T1, first tertile (low); T2, second tertile (middle); T3, third tertile (high)
Estimated marginal means of maternal weight measured during pregnancy were plotted, adjusted for maternal age, parity, secondhand smoking exposure, and socio-economic status (monthly family income)

Discussion

This study shows that pre-pregnancy BMI determines the level of SBP and DBP in pregnancy, but not the pace of blood pressure increase. Pre-pregnancy BMI was also positively associated with the risk of gestational hypertension and preeclampsia. The effect was independent of gestational weight gain.

The strength of this study is its prospective design. Furthermore, our study differed from other studies largely conducted in the Western countries where poor health behaviors, such as smoking, alcohol consumption and drug abuse, were more common among obese/overweight women.[21,22] The proportion of women who have these habits is very low in Indonesia,[23,24] as shown by the data from the women involved in this study. This enabled us to evaluate the effect of pre-pregnancy BMI, as a measure of adiposity, in isolation from such factors.

Pre-pregnancy BMI was calculated from the pre-pregnancy weight that was based on interview. We are aware that it could have limited accuracy. The main analysis was based on the assumption of linearity of the association between increase of blood pressures during pregnancy with gestational age. Therefore, although the graph could accurately describe the relative effect, the rankings, of pre-pregnancy BMI tertiles on SBP and DBP, it may not fully reflect the actual absolute trajectory of blood pressure changes during pregnancy. Previous studies reported that blood pressure decreases from early to mid-pregnancy and then increases until late pregnancy.[13,18,25] However, our BP measurement began at around 13 weeks of gestation, when SBP and DBP are about to approach their lowest level. Due to missing data, analyses were limited to women with complete data on the determinant, pre-pregnancy BMI, and outcomes, SBP and DBP, but both were fairly complete.

Classification of pre-pregnancy BMI was based on the tertile, rather than using the WHO cut-off criteria. Using this data driven approach, a relatively comparable number of women were classified in each group, ensuring sufficient power for statistical testing. Furthermore, there has been uncertainty about appropriate BMI cut-off for Asian population. Several studies recommend a lower cut-off for overweight and obesity in Asian population.[26]

Our study showed that pre-pregnancy BMI influences the level of SBP and DBP during pregnancy. This effect was present consistently over the course of pregnancy. It was in accordance with the reports from previous studies.[5,13,14,27] Almost similar to our findings, a study which explored the relation between blood pressure during pregnancy with gestational age and pre-pregnancy BMI as continuous scale measurements, also found that SBP and DBP level generally increased with higher pre-pregnancy BMI at any gestational age.[15]

In the present study, SBP and DBP were increased by 0.25 mmHg and 0.18 mmHg per BMI unit, respectively. These findings were similar to those in a previous study[27] which reported an increased mean arterial pressure (MAP) of 0.21 mmHg per BMI unit, but were smaller compared to another study[5] which reported the mean differences of blood pressure in the first, second, and third trimester of 1.03 mmHg, 0.98 mmHg, and 0.89 mmHg for SBP per BMI unit and 0.83 mmHg, 0.81 mmHg, and 0.74 mmHg for DBP per BMI unit. These discrepancies could be attributed to differences in variables included in the adjustment model, where in the latter study pregnancy weight gain was taken into account in the adjusted model but not in our analysis. Furthermore, women's BMI in that study may be higher since there were more overweight and obese women (27.6 %) than in our study (24.8 %). Lastly, differences in women's ethnicity could also contributed to these differences. To the best of our knowledge, we are among the first to study this in Asian women.

Hypertensive disorders of pregnancy (HDP) are a spectrum of diseases that encompass chronic hypertension, gestational hypertension, preeclampsia, and chronic hypertension with superimposed preeclampsia.[20,28] Our findings were also in accordance with several studies which demonstrated the relation between higher pre-pregnancy BMI and increased risk of gestational hypertension and preeclampsia.[10,16,29–33] These were also in line with studies that took obesity or BMI recorded at the first ANC as the determinant.[3,4,6,8,11,12,34–41]

In the present study, we found that pre-pregnancy BMI did not materially affect the pace of increase in blood pressure during pregnancy. An interaction term between BMI tertiles and gestational age that was introduced into the model showed non-significant p-values, suggesting that the relation BMI and blood pressures did not depend on gestational age. The increases in SBP and DBP were running in parallel independent of women's pre-pregnancy BMI. A previous study[14] also supports our finding as it showed relatively unchanged mean differences of SBP and DBP between various pre-pregnancy BMI groups in all trimesters. In contrast, another study showed that the pace of increase in BP did vary across BMI group and gestational age, with a ceiling effect which attenuated the increase of BP in higher BMI groups, particularly in later stages of pregnancy.[15]

As compared to women with normal range pre-pregnancy BMI, women who were overweight or obese before pregnancy had higher risk for excessive weight gain during pregnancy.[19] This excessive weight gain, further, could synergistically amplify the risk of preeclampsia in obese women.[31] Our finding in unselected pregnant women shows that pregnancy weight gain did not mediate the effect of pre-pregnancy BMI on blood pressure level during pregnancy. Our overall finding is in accordance with a study in Sweden showing correlation between DBP during pregnancy with women's baseline BMI, but not with their pregnancy weight gain.[18] Another study also showed that excessive weight gain did not increase the risk of preeclampsia,[17] although it raised the risk of transient hypertension. Many other studies, however, reported a positive relation between excessive weight gain and higher risk of hypertensive disorders of pregnancy that was independent of pre-pregnancy BMI.[5,16,29–33,42] Nevertheless, it is important to note that these studies used excessive gestational weight gain as dichotomous determinant, while in our study pregnancy weight gain was evaluated as continuous scale measurements.

The pathophysiologic mechanism to explain the association between pre-pregnancy BMI and blood pressure levels during pregnancy remains unclear. It has been speculated that adiposity causes a state of increased inflammation, hyperleptinaemia, hyperinsulinemia, and insulin resistance which further leads to disturbances in autonomic function such as sympathetic nervous activation. These mechanisms appeared to be similar in pregnancy and outside of pregnancy as has been extensively studied among preeclamptic pregnant women[43–45] as well as in obese and hypertensive women.[15,46–48] Obesity may also contribute in the dysregulation of leptin function, which consequently cause hypertension, and alternatively hyperleptinemia itself could have a direct effect on the increased level of inflammation and raised in blood pressure.[44] In contrast, several studies in non-pregnant populations suggested that the development of hypertension may precede the weight gain through pregnancy.[49,50] Sympathetic hyperactivity in hypertensive patients might down-regulate beta-adrenergic receptors, which caused a decrease in thermogenic response and therefore increased their propensity to gain weight.

Notably, an adaptive mechanism which involves maternal systemic inflammatory response through insulin resistance occurs in all pregnancies, with preeclamptic pregnancies being in the extreme end of the continuum.[51] This is supported by a study which showed that women's pre-pregnancy BMI strongly influenced women's mean arterial pressure throughout pregnancy in normotensive and chronic hypertensive women alike.[27]

A history of hypertensive pregnancy is known to be associated with higher incidence of high blood pressure, ischemic heart disease, and stroke later in life and occurrence at younger age.[10,52–54] It was argued that the positive association between gestational hypertension and cardiovascular diseases in later life was caused by their common pathology.[55] Obesity and chronic medical conditions such as diabetes or hypertension which are among the most recognized risk factors of preeclampsia,[9] also predispose women to later cardiovascular diseases. This could imply that the associations are due largely to pre-pregnancy risk factors, rather than to factors that operate exclusively in pregnancy.[56] Pregnancy could be seen as metabolic and vascular "stress test" that could unmask women's preexisting subclinical risks, which may manifest in later life as the effects of ageing and long-term exposure to other classical risk factors emerge.[52,55] In addition, excessive pregnancy weight gain is also associated with long-term maternal abdominal adiposity, which in turn may increase women's risks of cardiovascular and metabolic disease in later life.[57]

Our study showed that women's SBP and DBP increased with higher pre-pregnancy BMI, suggesting a stable linear relation of adiposity and blood pressures during pregnancy. This resembles the situations occurring outside of pregnancy, as shown by studies in adults[58,59] and in children and adolescents[60] in which adiposity parameters such as BMI were found linearly related with the increase in SBP and DBP. We speculate that the difference in blood pressure levels were actually present between women with different BMI before the pregnancy started, persisted during pregnancy, and continuously so in the postpartum period. However, we are unable to confirm this hypothesis fully since we do not have data on women's blood pressure prior to pregnancy and post-partum. In contrast, a small study in Brazil showed that although women with excessive weight had higher SBP and DBP than the normal weight women throughout pregnancy, their blood pressures at postpartum period were comparable.[13]

Conclusion

In conclusion, pre-pregnancy BMI, as a measure of adiposity, determined women's blood pressure level during pregnancy but not the pace of its increase. Higher pre-pregnancy BMI was also associated with higher risk for developing gestational hypertension and preeclampsia. Pregnancy weight gain does not appear to mediate these effects. Obese and overweight women in reproductive age should be encouraged to practice healthy life style which includes weight reduction prior to pregnancy. This intervention could protect them against various pregnancy complications and improve their long-term cardiovascular risks. Women who enter pregnancy with high BMI should be monitored more closely for development of hypertensive disorders during pregnancy.

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Supplement 1. Effect of prepregnancy BMI on systolic and diastolic blood pressure during pregnancy (twin pregnancies were excluded)

		Coefficient	95% CI	p-value
Systolic blood pressure	Crude model	0.32	0.24, 0.40	<0.01
	Adjusted model[a]	0.25	0.17, 0.34	<0.01
	Explanatory model[b]	0.34	0.22, 0.46	<0.01
Diastolic blood pressure	Crude model	0.22	0.17, 0.28	<0.01
	Adjusted model[a]	0.19	0.13, 0.24	<0.01
	Explanatory model[b]	0.20	0.12, 0.28	<0.01

BMI, body mass index; CI, confidence interval
Results are expressed as mixed model coefficients of systolic and diastolic blood pressure in mmHg for every 1 kg/m^2 increase in pre-pregnancy BMI.
[a]Adjusted for maternal age, parity, secondhand smoking exposure, and socio-economic status (monthly family income)
[b]As [a] and for total pregnancy weight **gain** (difference between maternal weight measured at the day of delivery from the prepregnancy weight)

PART 3

Breastfeeding

Chapter 8

Breastfeeding Attitude and Volume Optimization (BRAVO) study: a randomized breastfeeding optimization trial

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Submitted

Abstract

Background

A growing body of evidence shows the short-term benefits of breastfeeding, which include protection against infections, allergies and lung diseases. However, evidence on the long-term benefits of breastfeeding is scarce and often conflicting. The BReastfeeding Attitude and Volume Optimization (BRAVO) trial (clinicaltrials.gov NCT01566812) is designed to study the effect of breastfeeding on early signs of later chronic diseases, particularly life time cardiovascular, respiratory, and metabolic risks. In addition, the effectiveness of breastfeeding empowerment in promoting breastfeeding will also be evaluated.

Methods/Design

This study is an ongoing randomized trial in Jakarta, Indonesia that started in July 2012. Pregnant women are screened for their breastfeeding plan in the third trimester and those with low intention to breastfeed are randomly allocated to either receiving an add-on breastfeeding optimization program or usual care. Primary outcomes include breastfeeding rate, lung function, blood pressure, during the first year of life and vascular/cardiac characteristics, which will be measured at the age of 4 to 5 years. We also measured child growth and infection/illness episodes, while cognitive test is planned to be conducted at the age of 5 years.

Discussion

To date, 784 women (80%) have been randomized of 1,000 planned, with satisfactory completeness of follow up (90.1%). Included mothers were of lower socioeconomic status and more often have blue-collar jobs, similar to what was observed in the pilot study.

Conclusions

BRAVO can be successfully conducted in a low resource setting like Indonesia to provide solutions for low exclusive breastfeeding proportions and evidence about the short and long term health effects of breastfeeding, particularly on later cardiovascular and metabolic risks.

Introduction

World Health Organization (WHO) aims to have newborns exclusively breastfed for six months.[1] However, it is estimated that globally only 38% of infants are exclusively breastfed, and every year 800,000 deaths among children under five years of age have been attributed to suboptimal breastfeeding.[2] These acute consequences are urgent, but there has been concern for decades that suboptimal breastfeeding may have lifelong health implications as well.

Breastfeeding is suggested to protect against infections, allergies, lung disease, but also overweight and obesity,[3,4] diabetes mellitus,[5] cardiovascular risk factors,[6,7] and cardiovascular diseases.[8,9] Such suggestions draw heavily on non-experimental research in which, especially with breastfeeding, confounding bias is a major issue. Human breastfeeding experimentation in a fully explanatory way is impossible with firm public and care preferences and strong adherence to WHO recommendations, and given a lack of equipoise for at least short-term benefits.[10,11] However, numerous pragmatic randomized experiments had been done, primarily aimed to prolong breastfeeding duration, using a range of pre- and/or postnatal interventions with different randomized designs, and in a large variety of settings and populations.[12-34] By far the largest and probably most informative is a cluster randomized trial (Promotion of Breastfeeding Intervention Trial/PROBIT) in over 17,000 women in Belarus, showing that empowerment raised breastfeeding rates and decreased infant gastro-intestinal infection and atopic eczema rates, thus clearly underpinning short-term benefits.[35] However, later (non-) randomized analyses on that trial did not show protection against obesity and high blood pressure,[36-38] allergies and asthma,[39] and behavior problems.[40]

Indonesia is a low- to middle income country with a population of 250 million and 4 to 4.5 million births per year. Indonesia lags behind WHO recommendations with only 15 to 40% exclusive breastfeeding.[41-43] Indonesia experiences the epidemiological transition and it is among the regions identified to face a strong rise in non-communicable diseases, including atherosclerotic disease and a significant prevalence of chronic obstructive pulmonary disease.[44, 45] There is much potential for improvement of breastfeeding practices as breastfeeding empowerment programs have been shown effective in various south-east (SE) Asian countries, although not Indonesia.[46-48] Modifiable predictors of non-exclusive breastfeeding in SE-Asian countries have been identified and include the absence of prenatal breastfeeding planning, working by mothers, and non-involvement of fathers in the breastfeeding planning.[49] Generally, breastfeeding promotion programs are probably more successful in achieving exclusive breastfeeding in lower income countries like Indonesia than in economically more affluent societies.[50] Nevertheless, due to the specific local mix of cultures, religions, ethnic backgrounds, and limited resources, the effectiveness of empowerment programs that worked elsewhere, even in SE-Asia, cannot be taken for granted in Indonesia.

A new randomized breastfeeding trial is currently being conducted in Jakarta, Indonesia. The BReastfeeding Attitude and Volume Optimization (BRAVO) trial (clinicaltrials.gov NCT01566812) aims to individually randomize 1,000 women in later pregnancy, when decision-making about breastfeeding is imminent. In a pilot study, the stepped screening for eligibility procedure was tested in 207 pregnant women, of whom 56% intended to breastfeed less than 2 months and 38% were willing to be randomized.[51] Like virtually all reported breastfeeding empowerment trials, BRAVO questions whether breastfeeding can be optimized towards WHO recommendations. However, in BRAVO the principal aim is to study breastfeeding effects on early life signs of later life cardiovascular and -metabolic risk, and respiratory risk which have become particularly pressing in Indonesia.[42, 52] This aim has led us to choose for individual rather than commonly used cluster randomization, for intensive individual pre- and postnatal empowerment intervention for maximal breastfeeding contrast, and to choose a trial size and measurements specifically aimed at detection of cardiovascular, -respiratory, and -metabolic effects in early childhood. BRAVO aims to measure breastfeeding effects on the developing cardiovascular organ system itself.

To date, BRAVO has included and randomized 780 pregnant women in their third trimester. The BRAVO design is here presented, as well as a cross-sectional description of both screened and enrolled populations.

Methods

Study design and setting

BRAVO is a pragmatic parallel group randomized trial that started in July 2012. Before birth, eligible women are randomly allocated to either receive a program aimed at add-on breastfeeding optimization during pregnancy and the first 6 months after delivery or to receive primary care as usual. After randomization, a first cardiovascular assessment will be made when the children are between 4 and 5 years of age. The general design scheme is in **Figure 1**.

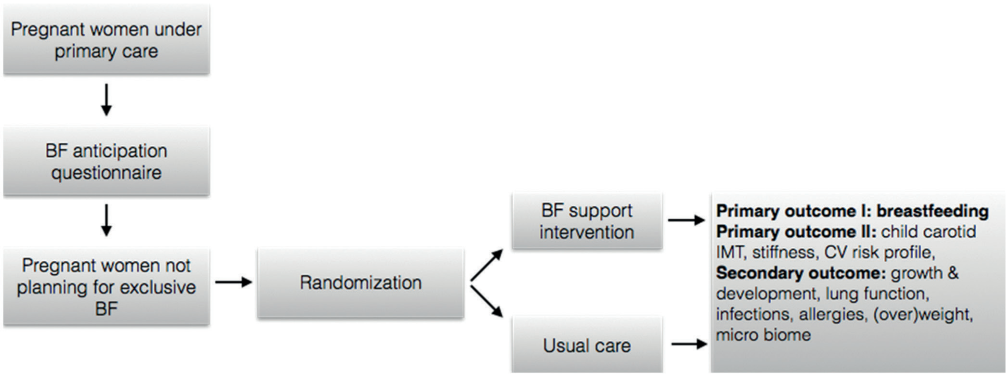

Figure 1. General screening and randomization scheme of BRAVO
BF=breastfeeding, CV=cardiovascular, IMT=intima media thickness

Women under primary or referred care are screened for eligibility. Women are recruited from Budi Kemuliaan Hospital, a private specialized referral center for maternal and child care in the municipality of Jakarta (Director MB), which has a delivery rate of 7000-8000 newborns/year. It performs service delivery, training (midwives), education, and research. The hospital has 10 staff with Good Clinical Practice certificates. Attending women are partly mobile but many live in the vicinity and have the hospital as first choice. Budi Kemuliaan features a home visit program by midwives in training. Restriction to women with Budi Kemuliaan as first choice ensures some 3,000/year for inclusion. Women are also recruited from local primary care centers, including Senen and Jatinegara districts primary care centers. This study was ethically approved by the Institutional Review Board of Faculty of Medicine University of Indonesia/Cipto Mangunkusumo General Hospital (Reference number: 913/UN2.F1/ETIK/X/2015).

Stepped eligibility screening, recruitment, and informed consent

BRAVO has a stepped screening of eligibility procedure of which the details, including inclusion criteria, are given in **Table 1**. Criteria for eligibility include pregnant women under primary or referred health care, who intend to breastfeed their newborn shortly or not at all.

Table 1. BRAVO eligibility screening steps during gestation

Step	Purpose	
1	Plans breastfeeding	Gestation week 28-36: midwives provide a short questionnaire to measure plans for breastfeeding duration ≤ 2 months 2 - ≤ 4 months 4 - ≤ 6 months > 6 months. Women indicating ≤ 2 months are eligible.
2	Stability residence	From the group of women that were selected under Step 1, a further selection is made of those living in the vicinity of the hospital.
3	Inclusion criteria	1. Residing in vicinity of participating hospital or primary care centers 2. Telephone communication possible with mother, concerning present pregnancy 3. Uncomplicated (no morbidity requiring hospitalization or intensive care, e.g. eclampsia) 4. No major fetal congenital disease 5. No known HIV in mother
4	Passed steps 1-3	Formal invitation of women to participate in BRAVO
Women who, after randomization, have stillbirth, premature birth (birth weight < 2500 g, gestational age < 37 weeks), congenital heart disease, multiple congenital anomaly, are not excluded and will be part of the intention to treat analysis.		

Midwives or primary care workers in charge of routine pregnancy care do the recruitment during visits. Eligible women are first informed about BRAVO, then formally invited to participate. Women are given 1 week to decide and are asked to inform their midwives or primary care physicians about their decision at the next visit or by telephone calls if they have then not decided yet. Women who decide to participate are asked to sign a BRAVO informed consent form.

Pilot study

Prior to BRAVO, a pilot study was done, assessing the feasibility of the procedures related with screening, invitation, informed consent, and randomization (see Introduction). The pilot was focused on active roles of midwives and primary care physicians, both from a practical and theoretical viewpoint. Women were subjected to procedures for antenatal and postnatal breastfeeding support as planned (see Interventions). Experiences of women and caretakers were registered and used for fine-tuning of the empowerment intervention. As caretakers are responsible for both BRAVO conduct and future empowerment implementation, their needs and comments were maximally accommodated.

Interventions

1. Care as usual

Care as usual means that there is no interference with breastfeeding propagation practice in current Indonesian care, which in general follows WHO recommendations for successful breastfeeding. The implementation of these recommendations varies across hospitals and primary care centers according to resource availability. Generally, education about breastfeeding is given individually by attending midwives or physician during antenatal visits.

2. Breastfeeding empowerment

Randomized trials in Southeast Asia have shown empowerment to effectively raise exclusive breastfeeding rates.[47] For BRAVO these programs were adapted in both language and culture to the Indonesian setting. Clearly, optimal empowerment comes from both ante and postnatal intervention.[47] The BRAVO program consists of antenatal, perinatal, and postnatal intervention, and special supports for working mothers, including provision of breast pump equipment when necessary and active support at workplaces. A lactation manager coordinates breast pump provision and gives written advocacy to employers to provide lactation room and working hours that allow mothers expressing their breast milk.

During later pregnancy, women in the intervention group are subjected to one private and one group counseling session. To avoid intervention contamination, the antenatal visit in the empowerment intervention is handled by a particular group of midwives or primary care physicians who underwent a standardized breastfeeding counseling training during BRAVO preparation, while the care as usual group have routine antenatal visits to regular caretakers. At private counseling, conducted within 28-37 weeks of gestational age, women and families are enabled to visit a lactation counselor for 30 minutes, mainly to build motivation for breastfeeding. A set of leaflets addressed specifically to mothers, husbands or grandmothers is given at the end of the session. At group counseling, women are subjected to a commercially available video (http://injoyvideos.com/) based on a 16-minute educational video entitled “14 steps to better breastfeeding” (Injoy Videos, Boulder, CO). The video is voiced over in Indonesian language and demonstrates correct positioning, latch on, and breast care. Closest family members (husband and/or women’s mother) are also invited to watch the video. Group counseling also allows mothers to share opinions with peers and mutual encouragement.

Empowerment intervention continues around delivery. Apart from early initiation and lactation support as part of routine care in Budi Kemuliaan Hospital and primary care centers, the intervention arm receives one extra home visit from appointed midwives/breastfeeding counselors to establish breastfeeding, address problems, and check preparations at home.

As part of postnatal intervention, women in the intervention group are visited at home by caretakers within the first postnatal week, both with usual care and experimental lactation support. A second lactation support visit takes place within week 1-2 after delivery. Thereafter, midwives and primary care workers have telephone contact once in two weeks in the first month and once a month thereafter for lactation support until 6 months after delivery. A centrally managed regular short message service (SMS) to encourage breastfeeding practice is sent weekly. A breastfeeding hotline or call center managed by midwives is set up to anticipate urgent questions or problems of mothers. Mothers in the intervention group who return to work before 6 months after delivery, get additional counseling on preparing, storing, and feeding their babies with expressed breast milk. Also, letters are written to formally advocate mothers' employer to facilitate breastfeeding continuation by providing secluded space, working hours, and environments that allow for expressing breast milk. Breast milk pump rental is arranged at request.

Trial retention

To promote trial retention, all mothers, irrespective of intervention arm, get reimbursement for their travel expenses to the primary health care or research hospital and infants who are not covered by government health insurance obtain free basic immunization. Investigators also set up a stepwise standard procedure for mothers who do not turn up for the routine health check, which includes at least two telephone reminders and home visits if mothers were not able to come for the outcome measurement.

Baseline trial measurements

Table 2 shows the baseline parental and child characteristics that are measured in BRAVO.

Endpoints

BRAVO deals with studying every child's first year, but beyond, it intends to follow children until at least their 5th year of life for carotid artery wall measurements, pulmonary function, anthropometry, and disease history. A general scheme of measurements and timing throughout BRAVO follow-up is presented in **Table 3**. Outcome assessors and data analysts are being blinded about the participants' assignment to interventions.

Primary outcome I: Monthly questionnaire/telephone: feeding patterns (breast/formula feeding habits)

Primary outcome II: Vascular wall characteristics at 4 years old (thickness of carotid aortic intima-media, distensibility, and elastic modulus) (see **Table 4**)

Secondary Outcomes: Blood pressure, lung function/respiratory tract disease, infection, allergy, obesity, growth and development, cardiac function, breast milk analysis (especially fat content), and gut/airway microbiome.

For subjects who discontinue or deviate from study protocol, but still live in the Jakarta area, home visits are conducted to collect data for breastfeeding habits, history of illness in the past 1 month, and growth data taken from the infant health card.

Table 2. Baseline parental and child's birth characteristics

Baseline parental characteristics	**Unit**	**Source**
Postal code & contact address		Questionnaire
Age	Years	Questionnaire
Marital status	Give response classes?	Questionnaire
Pre-pregnancy weight	Kg	Questionnaire
Number of (and age of) other children	N, years	Questionnaire
Substance use (smoking, alcohol, etc)	Yes/no Number/day?	Questionnaire
Socio-economic status (highest education of father/ mother)	Give response classes?	Questionnaire
Occupational status (mother/father)	Give response classes?	Questionnaire
Nutrition during gestation.		Questionnaire
Family history of chronic disease ((chronic) lung disease, diabetes mellitus, cardiovascular disease).		Questionnaire
Maternal and paternal disease history. ((chronic) lung disease, diabetes mellitus, cardiovascular disease)		Questionnaire
Children's birth characteristics	**Unit**	**Source**
Gestational age in weeks	Weeks	Midwife registry
Birth weight in grams	Grams	Midwife registry
Birth length in centimeters	Centimeters	Midwife registry
1- and 5-minute Apgar score	Values	Midwife registry
Specifics of gestation: (pre-)eclampsia, diabetes mellitus.		Midwife registry
Specifics of delivery: caesarian section, breech position, and vacuum/forceps extraction.		Midwife registry
Specifics of neonatal morbidity: Neonatal Intensive Care Unit (NICU)/special nursery care requirements, including sepsis, respiratory distress, and hyperbilirubinemia		Midwife registry

Table 3. Schedule of enrolment, interventions, and assessments

	STUDY PERIOD															
	Enrolment	Allocation	Post-allocation													Close-out
TIMEPOINT**	Gestation week		Birth	Month												
	24-36	32-36	*1*	*2*	*3*	*4*	*5*	*6*	*7*	*8*	*9*	*10*	*11*	*12*	*…*	*48-60*
ENROLMENT:																
Eligibility screen	X															
Informed consent	X															
Allocation		X														
INTERVENTIONS:																
Antenatal intervention		X														
Postnatal home visit			X													
Biweekly postnatal contacts			X													
Postnatal visits			X	X		X		X			X			X		
Telephone calls			X	X	X	X	X	X	X	X	X	X	X	X		
SMS			X	X	X	X	X	X								
ASSESSMENTS:																
Maternal ad family data		X														
Lung function SOT			X					X								
Lung spirometry														X		X
Feces sampling			X	X		X		X			X			X		
Breastmilk sampling			X	X		X		X			X			X		
Diary upper/lower RTD, allergy, infections			X	X	X	X	X	X	X	X	X	X	X	X	X	
Blood pressure measurement			X	X		X		X			X			X		X
Anthropometry			X	X		X		X			X			X		X
Vascular US carotid artery test																X
Pulse wave velocity/ stiffness																X
Echocardiography cardiac function																X
IGF-1, IGFBP-3, leptin levels, fibrinogen*														X		X

SMS=short message service, SOT=single occlusion technique, RTD=respiratory tract disease, US=ultrasound, IGF-1=insulin-like growth factor 1, IGFBP-3=insulin-like growth factor binding globulin 3

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Table 4. Cardiovascular endpoint measurements

	Methods	Arterial segment	Timing
Vascular ultrasound	Common carotid artery		48 to 60 months
Arterial stiffness: pulse wave velocity, augmentation index	Arteriography		48 to 60 months
Blood pressure	Oscillometric device	Brachial artery	2,4,6,9.12 months, 48 to 60 months
Cardiac function: cardiac output, LV systolic/diastolic function, RV diastolic function	Echocardiography	-	48 to 60 months

LV=left ventricle, RV=right ventricle

Data management, randomization, statistical power, and data analysis

All trial data are gathered through web-based data entry sheets (www.bravo-trial.com) by research nurses/assistants. Data-management is the responsibility of the Department of Clinical Epidemiology and Evidence Based Medicine (CEEBM) at Cipto Mangunkusumo Hospital in Jakarta, Indonesia. All investigators belong to the BRAVO study group have access to the final trial database through the data manager (SRS). Local principal investigators (NSI, WI) are also granted a direct access to the trial database for monitoring purpose. Public availability of clinical trial data is a pending debate,[53] and awaiting the outcome of that, any BRAVO data usage is decided upon consensus of the BRAVO study steering committee (NSI, CU, SS, WI, DEG). Randomization is centrally performed at that center using computer based random lists for simple parallel groups. All participating women are given individual study numbers and their personal information is collected, shared, and maintained in order to protect confidentiality before, during, and after the trial.

BRAVO is restricted to pregnant women who, from their own free will, plan to give breastfeeding for no longer than two months, if at all. It was estimated that 40% of all pregnant women meet that criterion. Thus, for all 4 eligible women, 10 pregnant women have to be screened, assuming that the number women lost by exclusion criteria is negligible. The effect of the empowerment program on the increase of exclusive breastfeeding was the first required estimate. From a Southeast Asian trial that used no restrictions on women's short breastfeeding planning, exclusive breastfeeding proportions in the experimental group were expected to be at least 1.5 times higher than in the care as usual group. This relative risk was reported throughout the first 6 neonatal months, but decreased from 83% to 53% at 14 days post partum and ultimately to 20% in the experimental group, and to 0% in the usual care group at 6 months. In 5-year old healthy children, breastfeeding vs formula feeding was suggested to have an effect on carotid artery wall intima-media thickness of 20 µm, at an average CIMT of 386 µm (SD 37). Translating to BRAVO, comparison of 54 women in an exclusive breastfeeding group and 54 women in an exclusive bottle feeding group would allow detection of a 20 µm group difference in aortic IMT with 80% power and alpha = 0.05. Obviously, BRAVO does not yield randomized groups of exclusive breastfeeding and exclusive bottle feeding groups. Due to contemporary uncertainties about effects of empowerment on breastfeeding and ultimately on the cardiovascular system, it was assumed that at least a 10 times bigger group was to be randomized. BRAVO therefore aimed to randomize 1000 eligible pregnant women, requiring screening of approximately 2500 pregnant women. A trial of 1000 women can detect small differences in breastfeeding practices between intervention groups. It was estimated that 500 women randomly allocated to each group allowed for statistical detection of a nominal difference of almost 10% in exclusive breastfeeding proportions, with 90% power and a statistical significance level of 0.05, (PASS 2008). From a public health point of view, a difference of 10% or higher was considered meaningful.

With respect to data analysis, both descriptive and baseline prognostically important characteristics will be tabulated against intervention strategy. The chance of successful exclusive breastfeeding for 6 months (yes/no) will be compared between the empowerment and care as usual group by calculating the relative risk with 95% confidence intervals. As an absolute risk estimate to support public health policy making, the number needed to treat (by breastfeeding empowerment) and 95% confidence intervals will be calculated. The duration of breastfeeding will be studied as a function of the interventions by using linear regression with (transformed) duration as the dependent variable and a group indicator as the independent variable. Should there be post-randomization differences that would hamper proper interpretation, regression modeling will be used to adjust all findings for baseline prognosis (propensity score or inverse probability weighting techniques).

The effects of intervention on cardiovascular measurements will be studied using linear regression techniques with cardiovascular measurements as dependent variables and a group indicator variable as the independent variable. All regression models will also be used for adjusting for baseline non-comparability when necessary. All analyses will be performed according to the intention to treat principle, such that all randomized women will be analyzed by their randomized allocation. Possibly biased missingness of 5- year outcome data will be assessed using additional tables of baseline data comparing the available and missing, and apply the above described analytical techniques for dealing with prognostic group differences.[54] Missing data will be analytically addressed using multiple imputation regression techniques.

Results of screening, enrolment, and preliminary follow-up

Table 5 shows the characteristics of women who were screened for BRAVO. The age of included and excluded women was similar. Included women were more often of low socio-economic status. Although there was no statistically significant difference in the proportion of working mothers between excluded and included women, included women more often had blue-collar employment. So far, completeness of follow-up is 90.1%.

Table 5. Characteristics of screened women (n=1,234)

	Excluded N = 707	Included N = 527	p-value
Age	31.3 (5.7)	30.3 (5.9)	0.30[1]
Socioeconomic status			<0.001[2]
Low	78 (12.7)	117 (22.9)	
Middle	432 (70.5)	358(70.1)	
High	103 (16.8)	36 (7)	
Number of pregnancy			0.70[2]
1	233 (37.8)	212 (41.2)	
2	218 (35.3)	173 (33.7)	
3	112 (18.2)	87 (16.9)	
>3	54 (8.8)	42 (8.2)	
Working mother	213 (34.6)	152 (29.7)	0.08[2]
Blue-collar job	49 (23.9)	55 (37.7)	0.006[2]
Previous breastfeeding	310 (43.8)	248 (47.1)	0.14[2]

Values are means with standard deviations for continuous variables and percentages for frequencies. In case of skewed data (*), median with inter-quartile range were presented.
[1] Independent samples t-test, [2] Chi-Square test

Discussion

To our awareness, BRAVO is the first randomized breastfeeding empowerment study that is specifically focused on estimating early life cardiovascular and -metabolic consequences of breastfeeding. Currently at almost 80% of the intended enrolment, the first part of BRAVO has been shown feasible and highly successful.

A strong feature of BRAVO is the number of individually randomized pregnant women, to our knowledge one of the larger of its kind in a low resource setting. The principal aim of BRAVO to study effects of breastfeeding on early life development of cardiovascular function and structure sets it apart from other trials that were primarily designed to measure effects of empowerment on breastfeeding proportions.

A major challenge in trials is to achieve accurate and complete follow-up. This is particularly challenging in BRAVO as it deals with young families in a highly mobile culture. In BRAVO, follow-up, certainly for the first year of life, is designed to follow regular care (vaccination scheme) and is proactive including home visits. So far, follow-up completeness is satisfactory although final results will have to be awaited. BRAVO like all other breastfeeding trials is pragmatic, so its ultimate success depends on the contrast in breastfeeding between intervention arms that can be achieved, and that will still be a function of distributions of breastfeeding duration in both arms and importantly, on distributions of bottle and other infant feeding in both arms. Obviously, BRAVO registers breastfeeding duration in both arms as well as introduction of other feeding habits. The fact that BRAVO just like other trials can only study cardiovascular effects as a function of overall breastfeeding contrast is a limitation that could not and cannot be circumvented by design. On the other hand, the pragmatic character of BRAVO will allow for easier implementation of beneficial results, if found. In principle, a threat to the BRAVO design is that women that are randomly allocated to care as usual will, knowing about the study objectives, change their breastfeeding plans, thus leaving a smaller contrast between experimental intervention arms. An alternative would have been pre-randomization (first randomize, then obtain informed consent), such as Zelen's design.[55] However, since BRAVO focuses on women who already during pregnancy plan to provide for a maximum period of two months breastfeeding, pre-randomization with its associated medical ethical implications, was not considered necessary. In BRAVO, individual women are randomized, rather than clusters of women within, for instance, primary care practices. Monitoring of breastfeeding motivation does take place on an individual basis, and is administered by study nurses. The effects of close monitoring are expected to not easily go from one pregnant woman to another. Although there is in principle a risk of intervention contamination, BRAVO uses individual, rather than cluster randomization for a couple of main reasons. First, the full intervention, both pre-, peri-, and postnatal, is well outside the realm of usual care as it requires high levels of organization (group counseling, home visits). Although cluster randomization was considered, only limited numbers of clusters were feasible in the BRAVO setting, which would have substantially increased the chance of baseline prognostic non-comparability, a much bigger threat to the overall purposes than contamination. A cluster randomized design would have required larger numbers of pregnant women, in whose offspring highly specialized cardiovascular and respiratory measurements were considered unfeasible. Finally, a cluster randomized design bears intrinsic prior uncertainties, such as ultimately achieved (differences in) cluster sizes, and unknown within-cluster correlations. Although such variation can be dealt with in analysis, the BRAVO design with individual randomization was considered a priori a more robust approach, specifically concerning its overall aim, the study of cardiovascular and –metabolic consequences of breastfeeding.

Obviously, breastfeeding randomization with its claimed beneficial health outcomes can only be considered within certain ethical boundaries. In principle, BRAVO does not interfere with current practice and personal maternal choices regarding their children's nutrition, it applies a breastfeeding intervention that is meant to improve on breastfeeding practices. As alluded to above, there are many examples of such randomized breastfeeding empowerment studies, including in (south-east) Asian countries.[48] The ethical justification of such trials has been uncertainty, equipoise, about the effectiveness of empowerment programs on breastfeeding rates. As in the Indonesian setting that effectiveness is currently unknown, it is uncertain whether Indonesian women allocated to care as usual are withheld better care, it is simply unknown if the alternative of empowerment is better. Therefore, as there is equipoise, randomization is considered justified. Moreover, women in BRAVO are selected such that they, by their own choice, plan to provide breastfeeding for a maximum of two months, if at all. If BRAVO were not conducted, all pregnant women would receive no empowerment other than currently available under care as usual.

The first results of BRAVO show that some 40% of screened women participated in BRAVO, which is slightly above the expected number. The enrolled and randomized women were more often of low socio-economic status and more often had blue-collar employment, since they were more likely to have no plans for extended breastfeeding. This was in line with findings from our BRAVO pilot study.[51] and previous studies which showed inverse relationship between breastfeeding rate and socio-economic status.[56-58] The decline rate of participant did not differ according to women's socio-economic status.

We believe that BRAVO is important for a number of reasons. First, since it is performed in Indonesia and strongly embedded in Indonesian maternal and child care, its results with regard to possibilities for breastfeeding optimization will be readily implementable locally. Second, it is well recognized that suggestions about beneficial effects of breastfeeding on later life cardiovascular and –metabolic risk, require stronger supportive evidence.[59] We believe that the BRAVO design presented here can provide such evidence.

Conclusion

We conclude that a complex randomized trial like BRAVO can be successfully conducted in a low resource setting like Indonesia, where solutions for low exclusive breastfeeding proportions and evidence about both short and long-term benefits are urgently needed.

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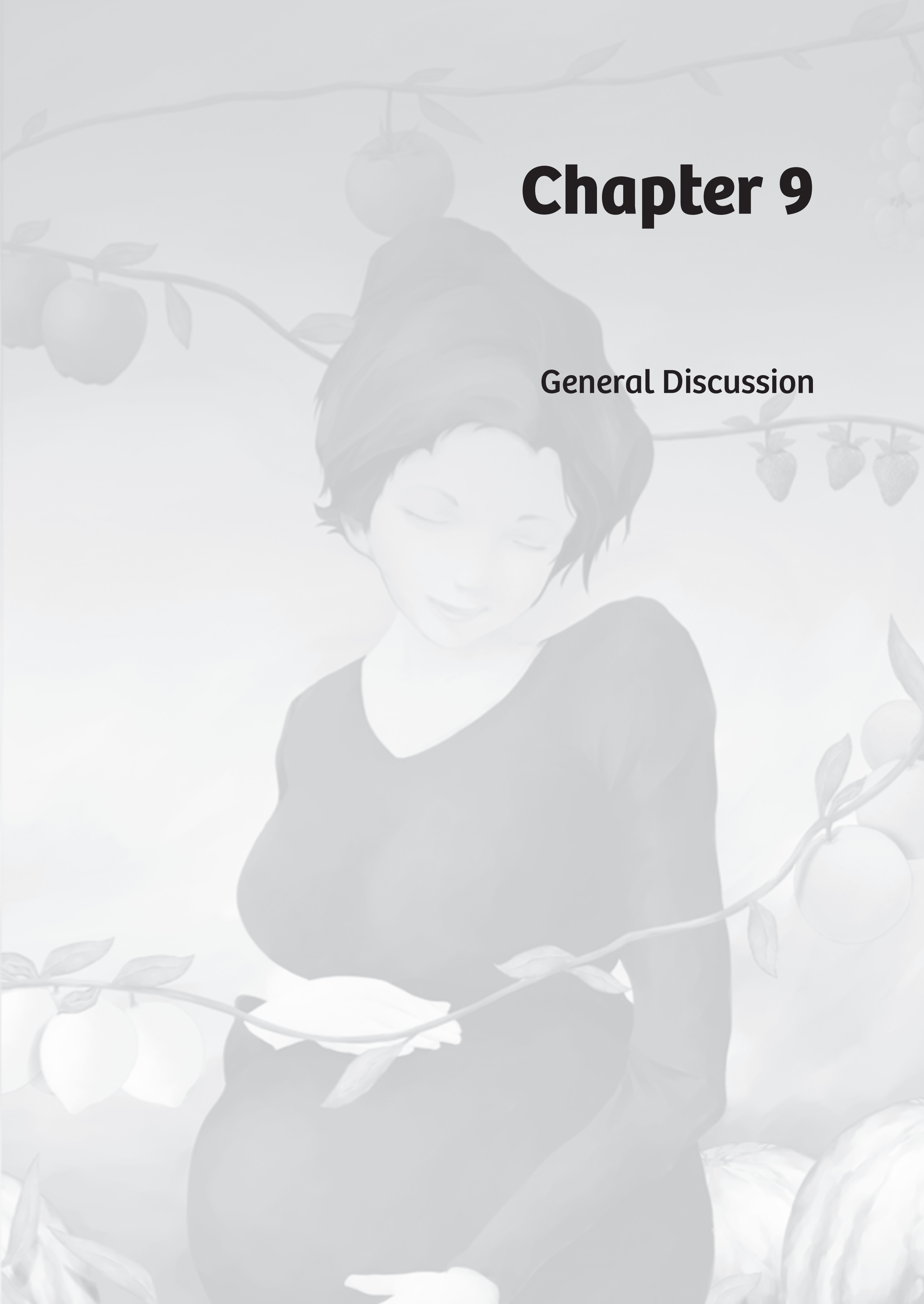
Chapter 9

General Discussion



Alongside genetic factors, environmental factors, particularly those that occur early in life, have been recognized to influence health, both in the short and long term. These environmental factors range from nutritional adequacy, infectious diseases, smoking or pollutants exposure, to psychosocial environmental stressors. Studies have been consistently shown that the aforementioned exposures during prenatal period are associated with a lower birth weight,[1–3] one of the most used parameters of hampered intra-uterine growth and adverse newborn's short and long-term prognosis. The effect of nutrition, in particular, has been studied extensively in relation to various maternal and child health outcomes. This encompasses studies about chronic nutritional deficiency, such as insufficient intake of macro- and micronutrients, to studies about the effect of transient nutritional disturbance. Due to its universal exposure, nutrition in early life appears to be among the most important and potentially modifiable environmental determinants of health.

Early life nutrition refers to women's nutritional circumstances prior to conception and during pregnancy, as well as offspring nutritional conditions during infancy and early childhood. These stages are interrelated and together co-shape future health. The period between conception and early childhood is of critical importance with regard to the growth and development of organs. Any perturbation occurring during this period could lead to serious and sometimes irreversible health consequences.

Studies have been done to investigate the long-term effects of nutritional disturbance around conception and the gestational period. Work that has been quite seminal is that of Barker and colleagues which has led to the Developmental Origins of Health and Disease (DOHaD) hypothesis. Showing correlations between the variation in mortality rates due to coronary heart disease across England and Wales and death rates of newborn, they suggested that events early in life could explain this apparent paradox.[4] Since most newborn deaths at the time were attributed to low birth weight, this finding led to the hypothesis that under-nutrition during the gestational period could permanently change the body's structure, function, and metabolism, leading to later occurrence of coronary heart disease. This first study was later confirmed by many similar studies around the world, including studies on the Dutch hunger winter, an acute short period of extreme famine, showing relationships between poor maternal nutrition during pregnancy with higher risk and earlier onset of coronary heart disease,[5,6] hypertension, obesity, metabolic syndrome, breast cancer, and psychiatric disorders in the offspring.[7,8] Studies in this field also looked further into the effect of undernutrition during the post-natal period and found similar results.[9,10]

Despite the mounting evidence in support of this hypothesis criticism also arose, mainly concerning the causal mechanism behind the associations. Studies in this field suggest the role of developmental plasticity, a critical period when a system is plastic and sensitive to the environment, and harmful events like fetal undernutrition may be followed by loss of plasticity and fixed loss of functional capacity. This phenomenon is suggested to facilitate the evolving of quite varied phenotypes from a single ‘genotype’ in response to the environmental conditions *in utero.*[11] Maternal undernutrition, for instance, could send cues to the growing fetus to develop a ‘thrifty phenotype’.[12] This phenotype enables the fetus to compensate undernutrition by reducing the rate of growth (intrauterine growth restriction), which would benefit short-term survival but lead to adverse health consequences later in life. At birth, babies who were born to malnourished mothers show a quite different complexion as compared to those from well-nourished mothers. Besides being significantly smaller and lighter, their body composition is also different such that they have less protein-rich soft tissues (skeletal muscle and abdominal viscera) but relatively preserved subcutaneous fat.[13] This phenotype seems to persist throughout the postnatal period (childhood and adulthood) as shown by a higher tendency towards central obesity and insulin resistance found in adulthood, particularly if an improved nutrition is encountered. Recent studies suggest that fetal undernutrition may adversely affect epigenetic control mechanisms, including DNA methylation, histone modification, and non-coding microRNA processes, which consequently cause alteration in the organ structure and function.[14,15] Most of these studies, however, are using experimental or animal models, and cannot be easily transferred to human situations. Epigenetic studies in human, including studies in people who were prenatally exposed to Dutch famine[16,17] are increasing. But the findings vary, depending on many other factors, such as the time of exposure and subjects’ sex.

Maternal over nutrition, as represented by obesity during pregnancy, has also been recognized to possibly impact offspring’s health. All-cause mortality has been found increased in adult offspring of obese mothers, as compared to those from normal weight mothers, and that was mainly due to higher rates of cardiovascular events.[18] Intrauterine programming seems to play an important role in the development of this chronic disease, although the observed relations are not easily disentangled from possible shared genes and familial environment, and from postnatal lifestyles that influence risk. Increased nutrient supply during pregnancy, accompanied with a state of compensatory fetal hyperinsulinemia has been proposed to permanently alter offspring’s appetite control, neuroendocrine systems, and energy metabolism, leading to higher risk of adiposity and other cardiovascular risk factors later in life.[19]

Short-Term Health Impact of Nutrition in Early Life

Apart from the serious later health consequences, short-term health impacts of suboptimal nutrition early in life should not be overlooked. This thesis aimed to provide insights on the associations between nutritional circumstances early in life, mainly during preconception and gestation period, with various pregnancy outcomes. Determinants included maternal nutrition status prior to pregnancy (as estimated by pre-pregnancy body mass index [BMI]) and the occurrence of transient nutritional changes during pregnancy (as modeled by hyperemesis gravidarum and Ramadan fasting). For outcomes, we focused on the newborn and maternal health parameters such as birthweight, small for gestational age (SGA), Apgar score, as well as the occurrence of gestational hypertension, miscarriage, and stillbirth.

Both under- and over nutrition during pregnancy seem to have adverse effects on maternal and newborn health. Maternal undernutrition and micronutrient deficiency contribute to the increased risk of maternal death by exacerbating complications including hemorrhage, obstructed labor, pregnancy-induced hypertension, and puerperal infection.[20,21] Maternal obesity and excessive weight gain, similarly, lead to a higher rate of preeclampsia, gestational diabetes, miscarriage, and Cesarean sections.[22–24]

Higher pre-pregnancy BMI has been shown associated with higher systolic and diastolic blood pressures consistently during the whole course of pregnancy (this thesis). A gradual increase of blood pressures occurs during pregnancy, beginning from the mid-pregnancy and reaching its peak around the end of pregnancy. As overweight or obese women enter pregnancy with a higher blood pressure, they are more likely to surpass the cut-off of high blood pressure, and, therefore, to develop complications such as gestational hypertension and preeclampsia.

A transient episode of nutritional disturbance during pregnancy has also been thought to affect maternal and newborn outcome. Hyperemesis gravidarum, for instance, could lead to dehydration and inadequate intake of food, consequently causing suboptimal pregnancy weight gain or even weight loss. We showed that hyperemesis gravidarum, particularly the severe type, leads to a significantly lower birth weight of babies (this thesis).

Another model of nutritional disturbance during pregnancy is the phenomenon of Ramadan fasting among pregnant Muslim women. Ramadan, which is characterized by intermittent (daytime) fasting, offers an opportunity to study the impact of temporary nutritional circumstances on various obstetric and newborn outcomes. Using data from the Mediterranean babies in the Dutch national registry, we showed that in-utero Ramadan exposure at large (not necessarily fasting) does generally not affect maternal or newborn health (this thesis). This was supported by findings from another study in Indonesia in which actual fasting during Ramadan was recorded (this thesis). Current studies on this topic generally support our findings, although our study in a different setting, involving women of Moroccan and Turkish descent who live in The Netherlands, suggested lower birth weights with first trimester Ramadan fasting. This discrepancy could be attributed to the variations in dietary or eating habit that are related with cultural background, as well as fasting duration and geographical location (humidity and temperature).

The mechanism by which maternal malnutrition, both by way of undernutrition and over nutrition, could affect fetal well-being is still under investigation. Studies on animal models suggested that maternal undernutrition causes increases in the plasma corticosterone levels, which in turn alters the process of placentation and placental ability in transferring nutrients.[25] In the case of over nutrition, a state of increased oxidative stress in the placenta was found to impair the establishment of sufficient blood supply which could consequently lead to placenta insufficiency.[26]

Maternal nutritional status also appears to determine offspring health during infancy and childhood. Undernourished women are more likely to give birth to smaller and premature babies,[27] that are at greater risks of neonatal death from infection and asphyxia. Maternal undernutrition may also explain a high prevalence of wasting (low BMI for age) and stunting (short height for age) among their children.[28]

During the postnatal period, one of the most important nutritional determinants is the type of feeding (breastfeeding or bottle-feeding). Breastfeeding has been reported to have various health benefits, such as reduced risk of all-cause- and pneumonia-related mortality in babies under 6 months old.[29,30] Breastfeeding has also been reported to protect against immune-mediated disorders,[31] obesity,[32] and have positive effects on child's cognitive function.[33] Most of these finding, however, are based on non-experimental studies and could therefore be subject to bias. In this thesis, we describe a study protocol of a randomized breastfeeding trial in the setting of a developing country, where there is typically a suboptimal breastfeeding rate. This study will not only provide evidence on the effectiveness of breastfeeding optimization program but also on the effect of breastfeeding on growth and development as well as on vascular wall characteristics, which are important markers for future cardiovascular outcomes (this Thesis).

Consideration for Early Life Nutritional Intervention

Ultimately, evidence from the etiologic studies on nutrition in early life should have some bearing on implementable prevention measures. Nutritional intervention in early life is particularly important in the context of the large burden of maternal and child health problems. The growing evidence about its later health consequences further adds to its importance. Etiologic studies like in this thesis may provide indications for prevention, but we are well aware that findings are not directly translatable to practice. For example, public health workers and clinicians do not meet patients with adjusted measurements, but with actual measurements. Nevertheless, realizing that more research is needed, it is important to consider what future implications for prevention could be.

In principle, there are two types of strategy of prevention; the 'high-risk' approach, which aims to protect the susceptible individuals, and the population approach, which seeks to control the causes of risk on a population level.[34] Both approaches have their own advantages and drawbacks. A 'high-risk' approach is favorable in terms of appropriateness to the individual resulting in a high motivation of subjects. However, this approach has disadvantages, mainly regarding its limited potential to impact the population at large. The population approach, in contrast, is more powerful as it has the potential to shift the distribution of risk factors in the population, including that of high-risk individuals. The disadvantage of this approach is the relatively small benefit of prevention for most individuals, and, therefore, lack of motivation of subjects.

In the context of early life nutrition, intervention needs to target both approaches. However, prevention measures using a population strategy should be prioritized in many situations. This strategy is more feasible and probably more cost-effective, particularly in the setting of limited resources in the developing countries. Intervention early in life is well applicable since most pregnant mothers and young children are usually under general care (via antenatal visits and immunization programs). This time window provides an excellent opportunity for education and health promotion programs. Moreover, intervention done early in life could also bring more sustainable impacts. Dietary modification that occurred in early childhood, for instance, is more likely to be maintained throughout one's life.[35] From the biological perspective, it is also reasonable to start preventive measures as early as possible, before irreversible damage has been done. Every stage of early life has its own critical period in terms of the pathological mechanisms of diseases. Nutritional problems occurring in one stage of early life could cause permanent consequences that cannot be ameliorated with nutrition improvement in later stages.

Given the interrelatedness between maternal and child nutrition status, we also need to reinforce the mother-child dyad,[36] in which mother and baby are seen as a team or a system. Maternal nutrition improvement has been shown to impact both the mother and the babies. For instance, in a setting where undernutrition is common, administration of additional daily snacks and micronutrient supplementation to women in the preconception and gestation period resulted in reduced pregnancy complications and increased newborns' birthweight.[37,38] Similarly, this was shown for education about maternal dietary intake during pregnancy.[39]

The finding that high pre-pregnancy BMI may lead to higher blood pressure in pregnancy (this thesis), for instance, calls for timely preventive efforts. All women of childbearing age need to be advised to maintain a healthy weight. For overweight or obese women, nutritional intervention that involves weight loss seems to be most relevant. This intervention is best done before the pregnancy begins as weight loss during pregnancy could potentially hamper fetal growth and development. It was shown that gestational weight loss did reduce the risk of preeclampsia, but the risks of preterm and small for gestational age (SGA) births were significantly increased.[40]

Achieving weight loss or weight gain control in overweight or obese women will not be easy. Modification of dietary intake and physical activity is challenging as it attempts to alter an already established lifestyle. Similar to many other interventions which require lifestyle modification, such as smoking cessation, weight loss programs are often perceived as burdening and consequently lead to subject's lack of motivation. Furthermore, more studies are needed to provide evidence about the effectiveness of weight loss intervention done prior to pregnancy in preventing the occurrence of adverse pregnancy outcomes. In addition, the best strategy for weight loss in childbearing age women can only be discovered through further studies. Studies like ours could may help to convince women of child bearing age that pre-pregnancy overweight, just like for instance (secondhand) smoke exposure, puts their future pregnancies and offspring health at risk.

Our finding of the effect of severe hyperemesis gravidarum on newborn's birthweight (this thesis), may call for an increased awareness among care providers who treat women with this condition. Although hyperemesis gravidarum is common, it is important for the doctors or midwives to be able to identify early signs which potentially lead to adverse pregnancy outcomes. Improvement in the monitoring of gestational weight gain followed with nutritional intervention if necessary may prevent the babies from being born with low birth weight.

Likewise, our finding that life-style changes that go with Ramadan fasting do not materially affects mothers' and babies' health (this thesis), can probably be translated into direct advice towards pregnant women who face the challenge of having to choose between fasting or not. One approach could be to disseminate this finding through a popular media outlet that women have general access to. At the end of this chapter, we enclose a short opinion article specially written for the general audience in Indonesia, the largest Muslim-majority country in the world. Furthermore, knowledge about factors that determine women's adherence to Ramadan fasting (this thesis) may help doctors or midwives to provide counseling that is customized to the women's situation. Nevertheless, more evidence is still needed on this topic, which includes the possible impact of Ramadan fasting for women with complicated pregnancies (for instance gestational diabetes or undernutrition). The influence of cultural background on women's strict adherence to Ramadan fasting may also need to be investigated further. This also holds for its possible long-term effects on the offspring.

With regard to prevention of various chronic diseases in later life, nutritional intervention during early life seems warranted. However, its implementation is rather complicated due to the fact that the evidence to support an early life strategy is largely of an etiologic nature (see above) and yet incomplete.[41] This is in stark contrast with our knowledge of prevention measures and implementation in adulthood. Taking the example of cardiovascular disease, the evidence about its risk factors for adults (such as age, gender, metabolic syndrome, smoking, hypertension and high cholesterol levels) has been well established. Prevention efforts which address these (combined) risk factors have been proven to significantly reduce the occurrence of diseases, as shown with antihypertensive medication, and lipid lowering.[42] Currently, there is virtually no evidence regarding the effect of modifying early life nutrition on the actual subsequent occurrence of chronic diseases.[41] Most of the interventions done during early life are focusing on the short-term outcomes such as growth and development or infection rate, but very rarely address its impact on health outcomes later in life. Obviously, the long duration of follow-up and logistic matters are the most dominant factors that hamper this kind of studies. There are medium term examples using proxy-outcomes, such as a randomized trial of sodium restriction during infancy, which showed a positive association between intervention and lower blood pressures in adolescence.[43] Nevertheless, as alluded to above, there is substantial evidence on particular health indicators such as birthweight and later life chronic disease. While studies like ours focus on direct maternal and neonatal outcomes, such findings can in our view be extrapolated to adult chronic disease risk, particularly in the absence of long term experimental research. Therefore, we consider nutritional optimization efforts early in life as an essential strategy for preventing chronic diseases.

Conclusion

Nutrition in early life plays an important role in determining future health outcomes, both in the short- and the long-term. Our findings show immediate ways to promote maternal and child health and support further investigations into the best preventive strategies.

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Supplement. Opinion article to be submitted to an Indonesian newspaper and its English translation

Berpuasa saat hamil: amankah?

Bulan Ramadan yang dinanti semakin mendekat, kaum Muslimin menyambut dengan suka cita. Tak terkecuali para muslimah yang kini sedang mengandung. Namun, banyak di antara mereka yang ragu apakah akan berpuasa atau tidak. Dalam Al Quran dijelaskan bahwa ibu hamil, bersama dengan ibu menyusui, termasuk dalam golongan yang mendapat keringanan untuk tidak berpuasa pada bulan Ramadan.[1] Mereka diwajibkan untuk mengganti puasa yang ditinggalkan dan/atau membayar fidyah. Walaupun demikian, cukup banyak pula ibu hamil yang tetap melaksanakan ibadah puasa. Di Indonesia, sekitar 80% ibu hamil berpuasa pada bulan Ramadan setidaknya selama satu hari.

Lalu, mengapa sebagian ibu hamil memilih untuk berpuasa sedangkan yang lain tidak? Ada beberapa penjelasan yang mungkin bisa membantu kita memahami hal ini. Pertama, terdapat interpretasi yang beragam mengenai kewajiban berpuasa pada ibu hamil. Selain itu, lingkungan sosial juga memiliki peranan. Ibu hamil sering kali merasa bersalah jika ia tidak berpuasa saat semua anggota keluarganya berpuasa. Apalagi mengingat ia akan berpuasa seorang diri saat mengganti puasanya nanti.

Annisa, seorang ibu dari dua anak di Kalimantan Timur, misalnya, memutuskan untuk berpuasa pada kedua kehamilannya yang bertepatan dengan bulan Ramadan. “Saya tahu sebenarnya saya boleh untuk tidak berpuasa, tapi saya tidak bisa membayangkan nantinya harus membayar hutang puasa sendirian.” Lain halnya dengan Mila, seorang wanita Indonesia yang kini bermukim di Belanda. Keputusan untuk berpuasa atau tidak pada saat hamil sangat tergantung pada musim di saat Ramadan jatuh. “Saya tidak berpuasa ketika saya hamil, karena waktu itu Ramadan bertepatan dengan pertengahan musim panas sehingga siangnya sangat panjang. Saya khawatir beratnya puasa dapat berpengaruh buruk bagi janin yang saya kandung. Seandainya saja Ramadan jatuh pada musim dingin, saya pasti berpuasa.”

Selain itu, pendapat suami tampaknya juga memiliki peran penting dalam keputusan ibu hamil untuk melaksanakan puasa Ramadan. Ibu hamil cenderung untuk tidak berpuasa jika suaminya menganjurkan demikian. Faktor lain yang tidak terduga, namun ternyata juga mempengaruhi adalah indeks massa tubuh (IMT) ibu hamil dan usia kandungan. Ibu hamil yang memiliki IMT lebih tinggi (lebih gemuk) dan ibu dengan usia kehamilan yang lebih muda memiliki kecenderungan lebih tinggi untuk berpuasa. Hal ini mungkin disebabkan karena mereka merasa lebih kuat untuk menjalani puasa.

Rasa cemas tentang kemungkinan pengaruh buruk puasa terhadap kehamilan ternyata cukup banyak dikeluhkan, baik oleh ibu hamil yang tidak berpuasa, maupun oleh mereka yang memutuskan untuk berpuasa. Hal ini tentu sangat tidak diharapkan karena kecemasan yang berlebihan justru dapat berpengaruh buruk bagi kesehatan janin. Kecemasan ini disebabkan oleh kurangnya pengetahuan ibu dan informasi yang tersedia.

Cukup banyak penelitian yang telah dilakukan mengenai dampak puasa Ramadan selama kehamilan terhadap kesehatan ibu dan janin. Puasa Ramadan diketahui berhubungan dengan menurunnya masukan nutrisi[2,3] dan terjadinya perubahan keseimbangan biokimia dalam darah ibu.[4] Namun demikian, perubahan-perubahan ini bersifat sementara dan akan segera pulih begitu ibu berbuka puasa. Selain itu, pertumbuhan janin berdasarkan pemeriksaan ultrasonografi (USG) juga nampak tidak terpengaruh.

Penelitian kami terhadap lebih dari 130,000 bayi baru lahir di Belanda dari keturunan Turki dan Maroko menunjukkan bahwa bayi yang terpapar Ramadan selama kehamilan tidak berbeda dari bayi yang tidak terpapar dalam berbagai parameter kesehatan, yakni berat lahir, skor Apgar, cacat bawaan, kelahiran prematur, dan kematian perinatal. Hal ini juga didukung oleh temuan dari penelitian-penelitian lainnya.[5-8] Namun demikian, beberapa penelitian lain melaporkan berat lahir yang lebih rendah jika ibu berpuasa, terutama jika puasa dilakukan pada trimester pertama.[9,10] Sementara itu, penelitian tentang efek puasa Ramadan saat hamil terhadap kesehatan ibu menunjukkan bahwa risiko hipertensi dalam kehamilan dan persalinan melalui bedah Caesar sama saja antara ibu yang berpuasa dan yang tidak. [2,11]

Penelitian tentang kemungkinan efek jangka panjang dari puasa Ramadan saat hamil juga telah banyak dilakukan, namun hasilnya tidak selalu sejalan. Beberapa penelitian melaporkan bahwa anak dari ibu yang berpuasa saat hamil memiliki kemampuan akademik yang lebih rendah di sekolah dan kondisi kesehatan yang lebih buruk saat dewasa.[12,13] Namun, penelitian yang lain tidak mendukung temuan tersebut.[11]

Perbedaan pada hasil penelitian yang dipaparkan di atas dapat disebabkan oleh berbagai hal, di antaranya karena variasi pada kondisi medis ibu, keadaan cuaca dan iklim (suhu udara dan kelembapan), durasi puasa, dan kebiasaan diet dan makan yang sangat berkaitan erat dengan kultur.

Secara umum, puasa Ramadan saat hamil tidak berdampak buruk bagi ibu maupun janinnya. Namun, puasa Ramadan dapat berpengaruh buruk terhadap kehamilan jika dilakukan oleh ibu yang status nutrisinya kurang (berat badan di bawah normal) atau memiliki kondisi medis lain. Dengan mempertimbangkan hasil dari berbagai penelitian yang telah ada, ibu hamil yang sehat aman untuk berpuasa saat bulan Ramadan. Dukungan dari anggota keluarga lain, terutama suami, sangatlah penting agar ibu dapat dapat menjalani ibadah puasanya dengan baik.

Ramadan fasting in pregnancy: is it safe?

Ramadan month is approaching and many pregnant Muslim women may wonder whether they should fast or not. According to The Quran, pregnant women are exempted from the obligation of Ramadan fasting.[1] Regardless of this religious pardon, many of them insist to fast. In Indonesia, about 80% Muslim women adhere to Ramadan fasting in pregnancy.

So, why do some Muslim women fast and others don't? There are some possible explanations that could help answer this question. First, Muslim women have a varied interpretation about fasting exemption. Social influence also plays a role in this decision-making process. Women often feel guilty and left alone when they skip the fasting while all other family members and their social network fast. This feeling may get stronger by the obligation to make up the fasting later on. Annisa a mother of two from Bontang, East-Borneo, fasted during her pregnancies that coincided with Ramadan. "I know it was okay for me not to fast at the time, but I could not imagine to have to make up all the fasting alone later." In places where the duration of fasting depends on the season coinciding with Ramadan, the reasoning may be a bit different. Mila, an Indonesian woman who resides in The Netherlands said "I did not fast when I was pregnant because it fell in the height of summer. Such a long duration of fasting could have a bad influence on my baby. If only it fell in winter, I would definitely have fasted". Husbands' opinion also seems to be important, as women are more likely to skip the fast if their husbands have an opposing view about it. There may also be other, perhaps somewhat unexpected, factors involved. Women with higher prepregnancy body mass index (BMI), a measure of overweight, and those who are at earlier gestational age are more likely to fast, possibly because they perceive that they can bear the burden of fasting.

Despite the many factors involved in the fasting decision and the reasoning behind it, fear of possible health effects appear to be as common among the fasting as in the non-fasting women. The fear seems rooted in women's lack of understanding of the issue and could be disadvantageous as the anxiety itself may affect baby's health.

Many studies have been conducted in this field. Ramadan fasting was linked with lower dietary intake.[2,3] Changes in women's blood biochemical balance occurred during the fasting.[4] These changes, however, were quickly reversible once the women broke their fast and did not seem to affect fetal growth. Our research done in more than 130,000 newborns from Turkish and Morrocan background suggests that Ramadan fasting had no effect on various newborn health parameters including birth weight, Apgar score, prematurity, and stillbirth. This was supported by many other studies.[5–8] Yet, some other studies reported a lower birth weight with fasting.[9,10] Maternal health also did not seem to be affected by Ramadan fasting, as shown by similar risks of Cesarean section and gestational hypertension.[2,11]

Researchers have also tried to find out if there are long-term effects of Ramadan fasting in pregnancy, but findings do not always agree. Some studies reported that it is associated with children's poorer academic performance at school and lower health status in adulthood.[12,13] However, another study did not confirm this finding.[11]

The disparity in the results of studies can be attributed to many factors such as differences in pre-existing medical conditions, weather or climate conditions (temperature and humidity), the length of fasting hour, as well as dietary practices and eating habits that are closely related with culture.

In general, Ramadan fasting during pregnancy does not seem to give any adverse impact, neither on the mothers nor on the babies. However, Ramadan fasting may adversely affect pregnancies if the women are underweight, malnourished or have pre-existing medical conditions. Taking into account what the research has told us so far, there is no big reason for healthy pregnant women to skip the fast. Support from family members, especially husband, is very crucial in giving the assurance the women need.

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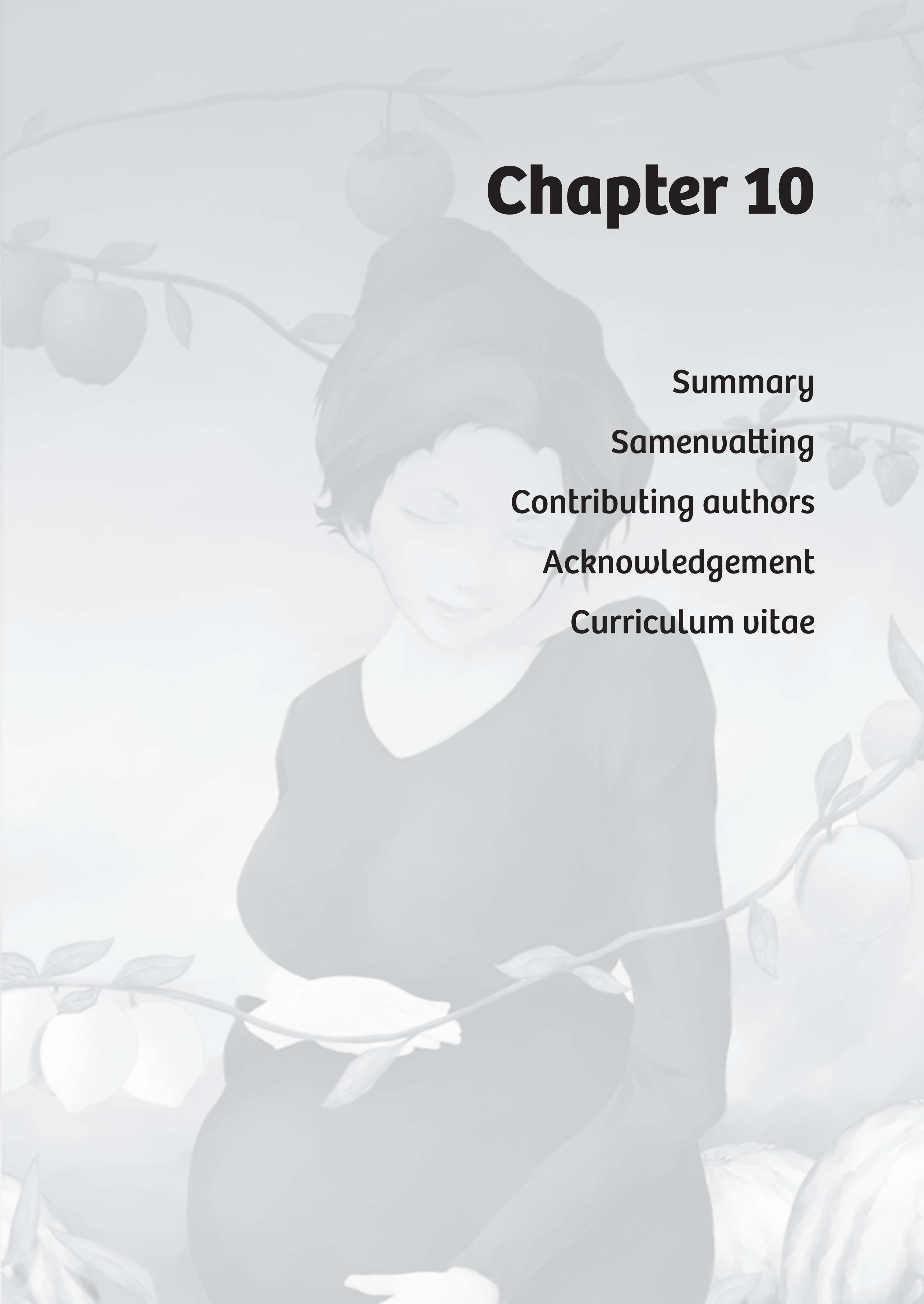

Chapter 10

Summary

Samenvatting

Contributing authors

Acknowledgement

Curriculum vitae

Summary

Early life nutrition is one of the most substantial environmental factors that shapes future health. This extends from the women's nutritional status prior to conception and during pregnancy to the offspring's nutritional conditions during infancy and early childhood. During this critical period, various body organs are undergone growth and development at incredible rates and programming of body systems take place. This thesis basically provides evidence on the associations between nutritional circumstances early in life, particularly during preconception and gestation period, with newborn health outcomes. We outlined this thesis into three parts. The first part (chapters 2, 3, 4, and 5) mainly focuses on *in utero* Ramadan exposure as nutritional determinants that may influence newborn health outcome. The second part (chapters 6 and 7) provides evidence about the effect of maternal nutrition status early in pregnancy on the development of pregnancy complications and newborn outcome. In the third part of this thesis (chapter 8), we described a protocol of a randomized trial on breastfeeding which is aimed to investigate the effect of breastfeeding on health outcomes during the first years of life.

Part 1 begins with a description of various factors that predict women's adherence to Ramadan fasting during pregnancy (**Chapter 2**). Although the health effects of Ramadan fasting during pregnancy are still unclear, it is important to identify various factors behind women's adherence to Ramadan fasting. The results of our cross sectional study among Indonesian women showed that demographic factors such as age, income, education, or employment do not influence fasting adherence, but higher pre-pregnancy body mass index (BMI), earlier gestational age, opposition from husband and fear of adverse fasting effects on their own or the baby's health do. These factors need to be addressed properly during counseling.

In **Chapter 3**, we present the association between Ramadan exposure and fasting during pregnancy with newborn's birth weight and the risk of gestational hypertension. In our Indonesian cohort of pregnant women, we found that birth weight was generally higher with Ramadan exposure as compared to without exposure. Analyses among women with Ramadan exposure revealed that although women who fasted had significantly lower nutrient intake in Ramadan, no different in the newborn's birth weight nor in the risk of gestational hypertension was found. These findings suggest that a transient life style modification that occurs with Ramadan does not seem to affect pregnancy outcomes investigated.

In **Chapter 4**, we show that among women of Moroccan and Turkish background living in The Netherlands, Ramadan fasting during the first trimester of pregnancy was associated with lower birth weight. No difference of birth weight was found if

the fasting took place later in pregnancy. This suggests that the effect of Ramadan fasting on newborn's birth weight may depend on the timing exposure. Furthermore, differences in finding as compared to the study in Indonesian women indicate that the effects may (partially) be influenced by culture or dietary habits as well as the length of fasting duration.

Chapter 5 addresses the association between in-utero Ramadan exposure with various birth outcomes. We analyzed 1,987,124 newborns from 11 birth cohorts of the Perinatal Registry of the Netherlands (Perined), 139,322 of these were classified as Muslims. No clinically relevant effect of Ramadan exposure was found, including on birth weight, Apgar score, congenital anomalies, sex ratio, gestational duration and perinatal mortality.

In Part 2, we focus on the effects of maternal nutrition status early in pregnancy on pregnancy outcomes. We present the associations between hyperemesis gravidarum, as a model of nutritional disturbance, on the development of placental dysfunction disorders in **Chapter 6**. Using the data from our prospective cohort study in Jakarta, Indonesia, we found that women who experienced severe hyperemesis had significantly lower birth weight newborns. Hyperemesis did not affect the development of placental dysfunction disorders (gestational hypertension, preeclampsia, stillbirth, and miscarriage), other newborn health measures (SGA, low birth weight, Apgar score, and gestation length), nor the placental dimension (placental weight and placental-weight-to-birth-weight ratio).

In **Chapter 7**, we elaborate the effect of women's prepregnancy body mass index (BMI) on their blood pressure during pregnancy. Using the data from our prospective cohort study in Jakarta, Indonesia, we showed that pre-pregnancy BMI determined women's systolic and diastolic blood pressure level during pregnancy. Higher pre-pregnancy BMI was also associated with higher risk of gestational hypertension and preeclampsia. These associations imply that pre-pregnancy (cardiovascular) risk factors strongly influence women's blood pressure. This finding also suggests the importance of prevention of pregnancy complications which should be done prior to pregnancy.

Part 3 (**Chapter 8**), we described a study protocol of a randomized trial on breastfeeding optimization (BRAVO) that is on going in Jakarta, Indonesia. This study is aimed to provide evidence about the short and long term health effects of breastfeeding, mainly on the children's cardiovascular and metabolic risks. Pregnant women with low intention to breastfeed are randomly allocated into either usual care or receiving an add-on breastfeeding optimization program which includes antenatal, perinatal, postnatal intervention, and special support for working mothers. Primary outcomes include breastfeeding rate, lung function, and blood pressure during the first year of life and vascular/cardiac characteristics which will be measured when the children are between 4 to 5 years old. So far, BRAVO has been successfully conducted with satisfactory completeness of follow up.

Finally in **Chapter 9** (general discussion), we discuss our findings in light of implementable intervention measures, both in the context of maternal and child health and in the prevention of later health consequences. Findings from the etiologic studies described in this thesis about maternal nutrition, including Ramadan (fasting) exposure in pregnancy, hyperemesis gravidarum, pre-pregnancy BMI, and breastfeeding, although may not directly translatable to practice, could provide indications for prevention and directions future research in this area.

Samenvatting

Voeding in het vroege leven is een van de meest belangrijke omgevingsfactoren die een rol spelen in de toekomstige gezondheid. Die invloed strekt zich uit van de voedingstoestand van de vrouw voor de conceptie en gedurende de zwangerschap tot de voedingsomstandigheden van het jonge kind.

In die kritische periode maken lichaamsorganen en -systemen een enorme groei en ontwikkeling door. Dit proefschrift beschrijft de samenhang tussen voedingstoestand in het vroege leven, vooral gedurende de preconceptie en zwangerschap, en gezondheidsuitkomsten van de pasgeborene. Dit proefschrift kent drie delen. Het eerste deel (**Hoofdstukken 2, 3, 4 en 5**) richten zich met name op in utero blootstelling aan Ramadan als voedingsdeterminant die de gezondheid van de pasgeborene zou kunnen beïnvloeden. Het tweede deel (**Hoofdstukken 6 en 7**) beschrijft de effecten van maternale voedingstoestand in de vroege zwangerschap op de ontwikkeling van zwangerschapscomplicaties bij de moeder en de gezondheid van de pasgeborene. In het derde deel van dit proefschrift (**Hoofdstuk 8**), wordt een protocol beschreven van een gerandomiseerde trial om het effect van borstvoeding op gezondheidsuitkomsten van het kind gedurende de eerste jaren van het leven na te gaan.

Deel 1 begint met een beschrijving van verschillende factoren die voorspellen of een zwangere vrouw zal gaan vasten gedurende de Ramadan (**Hoofdstuk 2**). Hoewel de gezondheidseffecten van Ramadan vasten tijdens de zwangerschap nog onduidelijk zijn, is het van belang om zulke factoren te identificeren. De resultaten van dit cross-sectionele onderzoek bij Indonesische vrouwen laten zien dat demografische factoren zoals leeftijd, inkomen, opleidingsniveau of werk, niet bepalend zijn. Wel bepalend voor het vasten in Ramadan bleken de body mass index (BMI) van de vrouw voor de zwangerschap, kortere zwangerschapsduur, mening van de echtgenoot en angst voor nadelige gevolgen van vasten op de gezondheid van de baby. Deze factoren verdienen goede aandacht bij het adviseren van vrouwen.

In **Hoofdstuk 3** wordt het verband getoond tussen blootstelling aan Ramadan en vasten tijdens de zwangerschap en het geboortegewicht van de pasgeborene en het risico op zwangerschapshypertensie. In ons Indonesische cohort van zwangeren vonden we dat het geboortegewicht over het algemeen hoger was bij blootstelling aan Ramadan dan zonder die blootstelling. Analyse van aan Ramadan blootgestelde vrouwen liet zien dat hoewel vrouwen die hadden gevast ook significant lagere intake van nutriënten hadden, het geboortegewicht van hun kind en het risico op zwangerschapshypertensie niet verschilde. Deze bevindingen suggereren dat een voorbijgaande aanpassing van leefstijl met Ramadan niet leidt tot veranderingen in zwangerschapsuitkomsten zoals onderzocht.

In **Hoofdstuk 4** laten we zien dat in Nederland wonende vrouwen van Marokkaanse en Turkse achtergrond, Ramadan vasten in het eerste trimester van de zwangerschap gerelateerd was aan een lager geboortegewicht. Dit suggereert dat het effect van Ramadan vasten op het geboortegewicht van de pasgeborene afhangt van de timing van blootstelling in de zwangerschap. Daarnaast lijken de verschillen met de bevindingen van de Indonesische vrouwen erop te wijzen dat cultuur en dieetgewoonten, maar ook lengte van de vastenduur (deels) een rol spelen.

Hoofdstuk 5 betreft het verband tussen in-utero Ramadan blootstelling en verschillende geboorte uitkomsten. Wij analyseerden data van 1.987.124 pasgeborenen uit 11 geboortecohorten van de Perinatale Registratie Nederland (Perined), waarvan 139.322 konden worden geclassificeerd als moslims. Wij vonden geen enkele klinisch relevante invloed van blootstelling aan Ramadan op zwangerschapsuitkomsten, inclusief geboortegewicht, Apgar score, congenitale afwijkingen, geslachtsratio, zwangerschapsduur en perinatale sterfte.

In deel 2 richtten wij ons op de effecten van maternale voedingstoestand in de zwangerschap op de uitkomsten van de zwangerschap. Wij beschrijven de verbanden tussen hyperemesis gravidarum als een model voor mogelijke verstoring van normale voedingsinname, op de ontwikkeling van disfunctioneren van de placenta in **Hoofdstuk 6**. Wij maakten gebruik van ons prospectieve cohort in Jakarta, Indonesië, en vonden dat vrouwen die ernstige hyperemesis gravidarum hadden gehad kinderen kregen met significant lager geboortegewicht. Hyperemesis was niet van invloed op de ontwikkelingen van afwijkingen op basis van placentadysfunctie (zwangerschapshypertensie, preeclampsie, dood geboorte, en miskramen), en ook niet op andere gezondheidskenmerken van de pasgeborene (SGA, laag geboortegewicht, Apgar score en duur zwangerschap) en afmetingen van de placenta (placentagewicht en placentagewicht-geboortegewicht ratio).

In **Hoofdstuk 7** gaan we in op het effect van de body mass index (BMI) van vrouwen voor de zwangerschap op hun bloeddruk tijdens de zwangerschap. Gebruik makend van onze prospectieve cohort studie in Jakarta, Indonesië, laten we zien dat BMI voor de zwangerschap bepalend is voor het systolische en diastolische bloeddrukniveau in de zwangerschap. Een hogere BMI was ook gerelateerd aan een hogere kans op zwangerschapshypertensie en preeclampsie. Deze verbanden impliceren dat (cardiovasculaire) factoren voor de zwangerschap sterk van invloed zijn op de bloeddruk van vrouwen tijdens de zwangerschap. Deze bevindingen geven ook het belang aan van preventie van zwangerschapscomplicaties voor de zwangerschap.

Deel 3 (**Hoofdstuk 8**) beschrijft een onderzoeksprotocol van een gerandomiseerde trial over het optimaliseren van borstvoeding (BRAVO) die momenteel loopt in Jakarta, Indonesië. Het doel van de studie is om de korte en lange termijn gezondheidseffecten, vooral cardiovasculair en metabool, na te gaan van borstvoeding. Daartoe worden zwangeren die niet of nauwelijks van plan zijn om te gaan borst voeden random gealloceerd naar gebruikelijke zorg of naar een add-on intensief borstvoeding optimalisatie programma dat zich uitstrekt over de antenatale, perinatale en postnatale periode, inclusief speciale ondersteuning voor werkende vrouwen. De primaire uitkomsten zijn borstvoeding gewoonten, long functie, en bloeddruk gedurende het eerste levensjaar en vasculaire en cardiale eigenschappen van de kinderen op de leeftijd van 4 a 5 jaar. Tot op heden is de BRAVO studie succesvol verlopen met bevredigend complete follow-up.

Tot slot geeft **Hoofdstuk 9** (algemene discussie) een discussie van onze bevindingen in het licht van implementeerbare interventie maatregelen, zowel in de context van maternale als kind gezondheid, ter preventie van verminderde gezondheid later. Hoewel bevindingen van etiologische studies in dit proefschrift over maternale voeding, inclusief Ramadan (vasten) blootstelling in de zwangerschap, hyperemesis gravidarum, BMI voor de zwangerschap, en borstvoeding, niet direct kunnen worden vertaald naar de praktijk, leveren ze wel indicaties voor preventie en richtingen voor verder onderzoek op dit terrein.

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Curriculum vitae

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