

15 JUL 2000



**THE RELATIONSHIPS BETWEEN PROTEIN INTAKE  
DURING THE SECOND AND THIRD TRIMESTERS  
OF LOW WEIGHT GAIN PREGNANT WOMEN  
AND FETAL OUTCOMES**

**SUWALEE LOWIRAKORN**

อธิษฐานทนาย

จาก

ศิริราชวิทยาคาร ม.มหิดล

**A THESIS SUBMITTED IN PARTIAL FULFILLMENT  
OF THE REQUIREMENTS FOR  
THE DEGREE OF DOCTOR OF PUBLIC HEALTH  
FACULTY OF GRADUATE STUDIES, MAHIDOL UNIVERSITY**

**2000**

**ISBN 974-664-076-3**

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was submitted to the Faculty of Graduate Studies, Mahidol University  
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## ACKNOWLEDGEMENTS

I would like to express my sincere gratitude and deep appreciation to Dr. Mandhana Pradipasen, my principal supervisor for guidance, in valuable advice supervision and encouragement throughout. I am equally grateful to Dr. Junya Pattaraarchachai and Dr. Rewadee Chongsuwat, my co-advisors for their contents, supervision and encouragement.

I would like to thank Dr. Dusanee Suttapreyasri, Dr. Montharop Chakkaphak, Dr. Prapapen suwan and Dr. Somchit Padumanonda for their kindness and consultation for this study.

I would like to thank the staff of ANC and delivery unit of Maternal and Child Hospital, Khon Kaen Regional Hospital, Srinakarin Hospital, Khon Kaen University and Health Promotion Center Region 6 for their co-operation of data collection.

I would like to thank Mrs.Jitra Gosi and students of Vocational College for helping in collecting data.

I also thank Miss Bongkot Noppon and Dr. Kriengsak Kisawadkorn, who contributed their valuable time to correct my English language in this study.

I wish to thank the staff of the Department of Nutrition, Faculty of Public Health, Khon Kaen University for their co-operation and generous assistance.

I wish to thank the Faculty of Graduate Studies, Mahidol University for partially supporting research grant.

I would like to thank Mr. Sikhin Rattanathip for his help in typing and some consultation for this study.

Finally, I would like to thank my family, my father, Miss Suwaluck Lowirakorn, Sittipong Lowirakorn, Anuwat Lowirakorn, Pinittaya Lowirakorn for their helps and moral support, which enable me to undertake this study.

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KEY WORDS : PREGNANT WOMEN / PROTEIN INTAKE / BIRTH WEIGHT  
BIRTH LENGTH / GESTATIONAL AGE

SUWALEE LOWIRAKORN: THE RELATIONSHIPS BETWEEN  
PROTEIN INTAKE DURING THE SECOND AND THIRD TRIMESTERS OF  
LOW WEIGHT GAIN PREGNANT WOMEN AND FETAL OUTCOMES. THESIS  
ADVISORS: MANDHANA PRADIPASEN, Dr.P.H., REWADEE CHONGSUWAT,  
Ph.D., JUNYA PATTARAARCHACHAI, Sc.D. 217 P ISBN 974-664-076-3

The study is aimed to determine the effect of quantity and quality of protein intake during the second and third trimesters in pregnant women with low weight gain before the 20<sup>th</sup> week of gestation. The study design was a prospective cohort study. An interview of dietary intake was conducted twice during the second and the third trimesters. Participants of this study were healthy pregnant women who had low weight gain at the time of recruitment and attended an antenatal care clinic regularly until delivery at Maternal and Child Hospital, Khon Kaen Regional Hospital Srinakarin Hospital, Khon Kaen University, from March 1998 to July 1999. One hundred and seventy subjects were sampled.

The maternal socioeconomic and health-related information and delivery data were collected using an interview and from medical records. The interview of dietary intake was employed with food frequency, 24-hour dietary recalls and food records. Gestational weight gain was collected from the initial pregnancy until delivery.

This study showed that there were no statistical differences of birth weight, birth length and gestational age among different socioeconomic characteristics. Energy intake and essential nutrient intakes were adequately consumed, compared with 75% RDA except for calcium and iron intake. Mean birth weight and birth length were not significantly different among groups of different protein intake. Gestational age was significantly different among the protein intake groups. There were no significant differences in birth weight, birth length and gestational age among groups of different percentage caloric intake from protein. The protein quality was also compared. No significant difference was found. A regression analysis was employed and results showed that total weight gain and prepregnancy weight were correlated positively with birth weight and birth length. The higher vegetable protein in the second trimester and the less vegetable protein intake in the third trimester were found to have better birth weight and length of infants. In addition, the higher of total protein intake in the second trimester, the longer gestational age was. Therefore, the pregnant women are recommended to consume high protein intake in the second trimester and moderate protein intake in the third trimester.

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สุวดี โลวีรกรรม : ความสัมพันธ์ระหว่างการบริโภคโปรตีนในไตรมาสที่สองและสามของหญิงตั้งครรภ์ที่มีการเพิ่มน้ำหนักน้อยกว่าทารกแรกเกิด (THE RELATIONSHIPS BETWEEN PROTEIN INTAKE DURING THE SECOND AND THIRD TRIMESTERS OF LOW WEIGHT GAIN PREGNANT WOMEN AND FETAL OUTCOMES) คณะกรรมการควบคุมวิทยานิพนธ์: มันทนา ประทีปะเสน, Dr.P.H., เรวดี จงสุวัฒน์, Ph.D., จรรยา กัทรอาชาชัย, Sc.D. 217 หน้า ISBN 974-664-076-3

การศึกษานี้มีวัตถุประสงค์ เพื่อศึกษาการบริโภคโปรตีนทั้งปริมาณและคุณภาพในช่วงไตรมาสที่สองและสามของหญิงตั้งครรภ์ที่มีการเพิ่มน้ำหนักน้อยในช่วงก่อน 20 สัปดาห์ของการตั้งครรภ์ เป็นการศึกษาติดตามระยะยาว โดยสัมภาษณ์การบริโภคอาหารในไตรมาสที่สองและสามไตรมาสละ 2 ครั้ง หญิงตั้งครรภ์ที่ศึกษาเป็นผู้มีสุขภาพดี แต่มีการเพิ่มน้ำหนักน้อยและมาฝากครรภ์สม่ำเสมอ และคลอดที่โรงพยาบาลแม่และเด็ก โรงพยาบาลศูนย์ขอนแก่น และโรงพยาบาลศรีนครินทร์ มหาวิทยาลัยขอนแก่น ตั้งแต่เดือนมีนาคม 2541 ถึงเดือนกรกฎาคม 2542 จำนวนตัวอย่าง 170 คน

ข้อมูลสถานภาพทางเศรษฐกิจ-สังคม และสุขภาพจนถึงคลอดได้จากการสัมภาษณ์หญิงตั้งครรภ์และแบบบันทึกของโรงพยาบาล การบริโภคอาหารเก็บโดยวิธีการสัมภาษณ์ความถี่ในการบริโภคอาหาร การบริโภคอาหารย้อนหลัง 24 ชั่วโมงและการบันทึกการบริโภคอาหารโดยหญิงตั้งครรภ์ การเพิ่มน้ำหนักระหว่างตั้งครรภ์จะเก็บตั้งแต่เริ่มฝากครรภ์จนถึงคลอด

การศึกษานี้พบว่า ไม่มีความแตกต่างของน้ำหนัก ความยาวทารก และอายุครรภ์ ในสถานภาพเศรษฐกิจ-สังคมที่ต่างกัน พลังงานและสารอาหารที่บริโภคในไตรมาสที่สองและสามเพียงพอเมื่อเปรียบเทียบกับ 75% RDA ยกเว้นเหล็กและแคลเซียม ค่าเฉลี่ยของน้ำหนักและความยาวของทารกไม่แตกต่างกันในกลุ่มที่มีการบริโภคโปรตีนที่แตกต่างกัน ยกเว้นอายุครรภ์ และ ไม่มีความแตกต่างของน้ำหนักและความยาวทารก อายุครรภ์ ในกลุ่มที่มีร้อยละของพลังงานได้จากโปรตีนและคุณภาพของโปรตีนที่ต่างกัน จากการวิเคราะห์การถดถอย พบว่า น้ำหนักก่อนการตั้งครรภ์และน้ำหนักที่เพิ่มขึ้นตลอดการตั้งครรภ์ มีความสัมพันธ์ในทางบวกกับน้ำหนักและความยาวของทารก การบริโภคโปรตีนจากพืชมากในไตรมาสที่สองและลดลงในไตรมาสที่สามสัมพันธ์กับน้ำหนักและความยาวของทารกที่เพิ่มมากขึ้น ส่วนการบริโภคสัตว์รวมในไตรมาสที่สองเพิ่มมากขึ้นสัมพันธ์กับอายุครรภ์ที่เพิ่มขึ้น ดังนั้น หญิงตั้งครรภ์ควรบริโภคโปรตีนสูงในไตรมาสที่สองและบริโภคโปรตีนปานกลางในไตรมาสที่สาม

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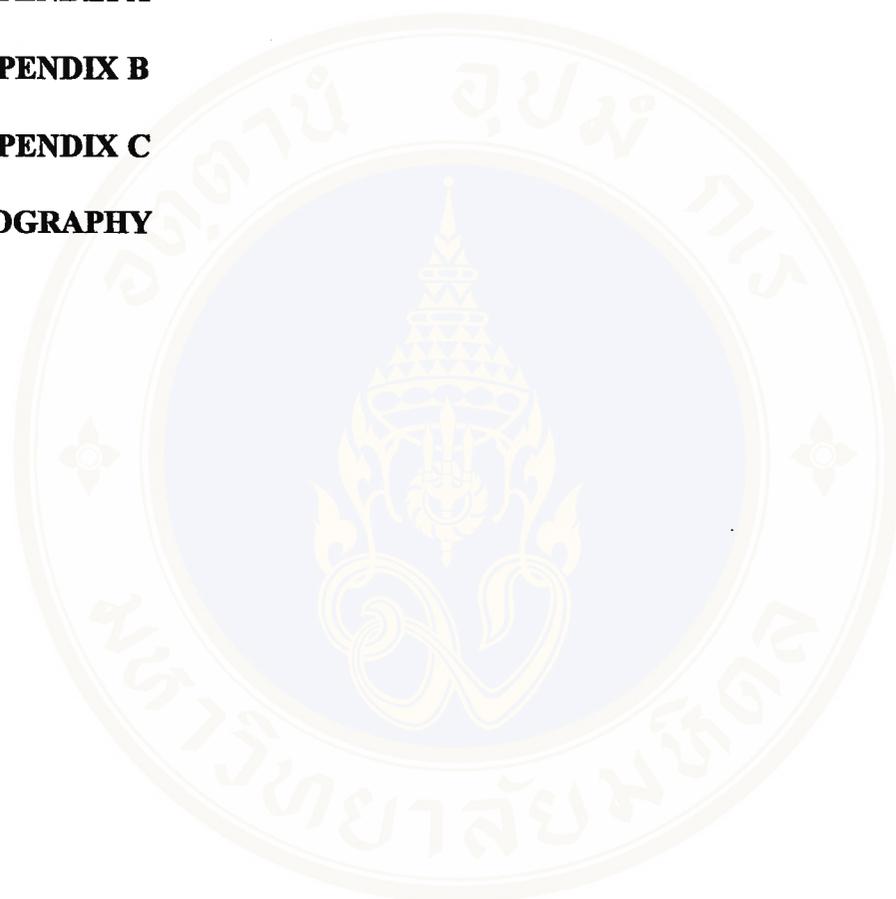
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## **CHAPTER I**

### **INTRODUCTION**

#### **BACKGROUND AND RATIONALE**

Food is essential to life and growth. Environment and genetics are the major factors that influence human growth and development. The environmental factors stimulate the development of a child after birth. Nutritional status is one of the most important environmental factors affecting the health of pregnant women and their infants. The other factor, genetics, which involves and affects the fetus is an internal control. The genetics factor is what the child inherited from the parent since fertilization and it can hardly be changed. For the environmental factor especially nutrition and health, it has a parallel effect on the development of the child. The pregnant women with good nutritional status and adequate dietary intake during pregnancy will give birth to infants in good condition with normal weight. On the other hand, the mothers who receive inadequate dietary intake and have low weight gain are at high risk of complication during pregnancy, which will increase morbidity and mortality (1).

The indicators for nutritional status of pregnant women are infant birth weight and length and gestational age. Malnutrition mothers are at high risk group for delivery of low birth weight infants pre-term delivery (birth prior at the 37<sup>th</sup> week of gestation) and intrauterine growth retardation (IUGR, delivery of a term infant weighting less than

2500 gram). The most salient cause of low birth weight in most developing countries is IUGR whereas the most significant cause contributed to low birth weight in a developed country is pre-term delivery. From many studies, maternal weight gain and gestational duration are important determinants of infant birth weight, and maternal weights are gained by increasing dietary intake especially energy and protein intake (2,3).

During pregnancy, additional nutrients are required for maternal tissue growth. In humans, dietary intake restricted in protein and energy is more relevant than vitamins and minerals, and affects maternal nutritional status and fetal growth. All animals need energy and protein in the similar way but the needs of vitamins and minerals are their specific functions differing from species to species. In humans, the highest intake during pregnancy is reached in the second trimester median intake of calories, carbohydrate, and fat decreases while protein intake remains high in term. The deposit of protein is not necessarily linear throughout pregnancy. During early pregnancy the fetal component is minimal, whereas the maternal volume expansion and tissue growth may be substantial. Late in pregnancy, the fetus may account for the major increase in protein needs (4).

According to the goals of the Seventh Five-Year National Health Development Plan, the target of the birth weight of Thai infants should be more than 3000 gram. No more than 7 percent of infants birth weight are less than 2500 gram and no less than 70 percent of infants birth weight are equal or more than 3000 gram. However, the Ministry of Public Health reported there were 8.69 and 33.28 percent of infants whose birth weight was less than 2500 gram and 3000 gram respectively. The Northeastern

part is a particular area where there is high incidence of low birth weight infants. There were 8.15 percent of infants whose birth weight was less than 2500 gram and 32.17 percent of infants whose birth weight was less than 3000 gram (5). The incidence rate of low birth weight infants at Srinakarin Hospital, Khon Kaen Regional Hospital and Maternal and Child Hospital, Khon Kaen Province were 7.15, 10.3, and 8.10 percent respectively. The incidence of prematurity from the hospitals' report was high in Srinakarin Hospital, and Maternal and Child Hospital; there were 8.64 and 7.02 percent of preterm infants respectively (6,7), but there were only 1.1 percent in Khon Kaen Regional Hospital (8). However, there were no national data for the incidence of prematurity. One of the main reasons of low birth weight and prematurity is health status of pregnant mothers which is related to dietary intake. In human and animal experiments, the protein intake of pregnant women is highly correlated with infants birth weight and length. The higher protein intake, the heavier birth weight and longer birth length, but with lower protein intake, fetus tends to encounter premature delivery. From these results, the Ministry of Public Health tries to promote the health of pregnant women and infants by providing health education including nutrition education and supplementary program to improve the health behavior and health practice before and during pregnancy. Health workers usually advise the pregnant women to eat more than usual especially protein. The high energy and protein supplement is more efficient for malnourished than normal pregnant women as reported in previous studies (9,10,11). Increasing dietary intake is good for pregnant women. However, there are some conflicts regarding protein supplement: too much protein intake and unbalanced diet may have negative effects on pregnancy course and outcomes. Jongsongsrerm's study

(12) showed that pregnant women with maternal low weight gain during the second trimester had shortening gestational age which may be due to dietary intake. The dietary intake of these pregnant women with premature labor was inadequate during the second trimester, which resulted in low weight gain. However, the tremendous increase in the amount of dietary intake after the second trimester resulted in normal weight gain during the last trimester. As a common practice in the antenatal care clinic, when the mothers first attend the antenatal care clinic late in the second trimester with abnormally low weight gain, they are advised to improve their nutritional status by increasing the dietary intake especially protein. With a successful dietary practice, pregnant women's weight increased in the third trimester but the finding was supported by Rush's study (13) in which there were 3 groups of low weight gain pregnant women who received supplement with high protein diet, supplement with balanced protein-calories diet, and no supplement, the gestational age was shortest in high protein supplement group. Riopelle's study (14) had a similar finding that the deprived monkeys on high protein diet had shorter length of gestation at delivery. In light of these findings, they suggested that supplementation of maternal diets with protein in amount of about 25 percent of total calories (100-150 g), with imbalance calorie intake may lead to lower weight gain, infant birth mass, but increase in perinatal morbidity and mortality and cognitive impairment (15). Therefore, this study intended to investigate the dietary intake during the second and third trimesters especially protein intake of the pregnant women who had inadequate weight gain during the first trimester and whether different quantities of protein intake at different time would affect fetal

outcomes. The results of this study will be applied in nutritional education to improve the health status, birth weight, birth length and gestational age of infants in the future.

## **RESEARCH QUESTIONS**

1. Do the different quantity and quality of protein intake during the second and third trimesters in pregnant women who have low weight gain affect infants' birth weight, birth length and gestational age ?
2. Is high protein intake during the second and third trimesters of pregnant women who have low weight gain harmful for the fetal outcomes?
3. Is the animal protein intake of pregnant women more efficient for increasing infants' birth weight, birth length than vegetable protein intake?

## **OBJECTIVES**

### **General Objective**

To determine the effect of different quantity and quality of protein intake during the second and third trimesters in pregnant women with low weight gain on infants' birth weight, birth length and gestational age.

### **Specific Objectives**

1. To determine the effect of quantity of protein intake in pregnant women in terms of gram unit and percentage of caloric intake on infants' birth weight, birth length and gestational age.
2. To determine the effect of animal protein and vegetable protein intake in pregnant women on infants' birth weight, birth length and gestational age.

3. To determine the relationships between protein intake, energy intake, and maternal weight gain in the second and third trimesters of pregnancy and fetal outcomes.

## **HYPOTHESES**

1. In low weight gain pregnant women, the quantity of protein intake in the second and third trimesters correlate negatively with infants' birth weight, birth length and gestational age.

2. The pregnant women with low quantity of maternal protein intake in both the second and third trimesters will give birth to lower infants' birth weight and birth length including shorter gestational age compare with other groups.

3. The pregnant women with low quantity of protein intake in the second trimester but high quantity of protein intake in the third trimester have shorter infants' gestational age than other groups.

4. The pregnant women who have good quality of protein intake will have heavier infant's birth weight, longer infants' birth length and gestational age than the pregnant women who have poor quality of protein intake.

## **ASSUMPTIONS UNDER THIS STUDY**

1. Most of the pregnant women were in the second trimester of pregnancy when they first visited antenatal care clinic and were followed up through the third trimester until delivery. Thus, it was assumed that they did not receive any vitamins or minerals supplement before they came to antenatal care clinic.

2. This study did not limit the quality and quantity of the participants' dietary intake. All participants had their ordinary diets. However, participants might have changed their nutritional behavior because of the health education in antenatal care clinic.

### **SCOPE OF THE STUDY**

The participants of this study were confined to low weight gain pregnant women who attended antenatal care clinic regularly in a common ward and delivered at Maternal and Child Hospital, Khon Kaen Regional Hospital and Srinakarin Hospital, Khon Kaen University, Khon Kaen province.

### **LIMITATION OF THE STUDY**

1. All participants in the study were selected from the pregnant women who were in low and lower middle socioeconomic status because they were the general patients in the common ward. They did not represent the whole population, but the results from this study could represent the population who had the same characteristics.

2. The interview of pregnant women began from the 20<sup>th</sup> week of gestational age so the information before pregnancy regarding the health status, food behavior was retrospectively collected.

3. Since antenatal care clinic was open from 8 am. to 12 am. on weekdays and pregnant women visited only once a month so 24-hour dietary recalls and food records were used for dietary assessment.

4. The majority of the pregnant women had their first antenatal care visit in their second trimester so there were some errors for accurate prepregnancy weight.

## DEFINITION OF TERMS

**Fetal outcomes** : the outcomes specify only infant birth weight, and birth length and gestational age.

**The pregnant women** : the second and third parity pregnant women who had low weight gain at the time of recruitment and attended antenatal care clinic regularly at Maternal and Child Hospital, Khon Kaen Regional Hospital and Srinakarin Hospital, Khon Kaen University.

**Weeks of gestation** : the time calculated from the first day of the last menstrual period to the interview date.

**The second and third trimesters** : the second trimester was defined between 16 and 27 weeks of gestation and the third trimester was defined between 28 and 40 weeks of gestation. This study collected only 4 periods beginning from the 20<sup>th</sup> week of gestation to 35<sup>th</sup> week of gestation.

### The second trimester

4.1. interview during the 20<sup>th</sup> - 23<sup>rd</sup> weeks of gestation.

4.2. interview during the 24<sup>th</sup> - 27<sup>th</sup> weeks of gestation.

The third trimester

4.3. interview during the 28<sup>th</sup> - 31<sup>st</sup> weeks of gestation.

4.4. interview during the 32<sup>nd</sup> - 35<sup>th</sup> weeks of gestation.

**Average weight gain regarding trimester (kg/wk)** : weight gain (kg) per week of pregnancy during each trimester.

$$\text{weight gain} = \frac{wt_t - wt_{t-1}}{ga_t - ga_{t-1}}$$

where wt was weight, ga was gestational age in weeks, t was time of most recent measurement, and t-1 was the time of previous measurement (16).

**Low weight gain** : minimal accumulation weight gain in the first trimester about 1 kg (17). In this study, low weight gain during the first trimester defines less than 1.5 kgs. Weekly weight gain of less than 0.4 kg during the second and third trimesters for women of normal BMI before pregnancy, less than 0.5 kg for those who were underweight, and less than 0.3 kg for overweight women (16).

**Body Mass Index (BMI) ( kg/m<sup>2</sup> )** : prepregnancy weight of pregnant women (kg) was divided by square of height (m<sup>2</sup>) of pregnant women. It refers to nutritional status in prepregnancy and were classified into 3 groups as follows (18) :

$$7.1 \text{ BMI} < 18.5 \text{ kg/m}^2 = \text{underweight before pregnancy.}$$

$$7.2 \text{ BMI } 18.5 - 24.9 \text{ kg/m}^2 = \text{normal nutrition status before pregnancy.}$$

$$7.3 \text{ BMI} > 25 \text{ kg/m}^2 = \text{overweight before pregnancy.}$$

**Gestational age at delivery** : the time calculated from the first day of the last menstrual period to the day of delivery (expressed in days).

**Preterm delivery** : the delivery within the 20<sup>th</sup> - 36<sup>th</sup> weeks of gestation (or 140 - 258 days) from the first day of the last menstrual period (LMP) (19).

**Full term delivery** : the delivery within the 37<sup>th</sup> - 42<sup>nd</sup> weeks of gestation (or 259 - 294 days) from the first day of the last menstrual period (LMP) (19).

**Infant birth weight and birth length** : the first weight (grams) and length (centimeters) of infant after delivery.

**The adequate energy intake** : the average energy intake in the second and third trimesters from 24-hour dietary recall, compared to 100 % Thai recommended daily dietary allowance for pregnant women, 1989 (2300 Kcal), and were divided higher and lower than 100% RDA (20).

**The energy distribution** : energy is derived from carbohydrate, fat and protein. The optimal ratio for percentage of energy intake from carbohydrate, fat and protein should be 55%, 30%, 15% respectively as recommended for Thai population (20).

**The maternal protein intake** : the average protein intake from 24-hour dietary recall method.

#### 14.1 The quantity of protein intake

##### Using RDA in terms of gram unit

The protein intake was divided into 3 levels:

1. Low protein intake (<51 gram)
2. Moderate protein intake ( 51 -100 gram)

### 3. High protein intake ( $> 100$ gram)

A hundred percent of Thai recommended daily dietary allowance for pregnant women, 1989( 51 g) was used as criteria to divide the level of protein intake into low and moderate protein intake (20). From the literature review, the pregnant women who received protein more than 100 gram had negative effects on fetal outcomes (15) so this level was used as criteria to divide the level of protein intake into moderate and high protein intake.

#### Using RDA in terms of percentage of caloric intake

The protein intake was divided into 3 levels:

1. Low protein intake ( $<15\%$ )
2. Moderate protein intake ( 15-20 %)
3. High protein intake ( $> 20\%$ )

Thai recommendation for Thai population of protein intake as percentage of energy intake about 15% was used as criteria to divide the level of protein intake into low and moderate protein intake (20). From the literature review, the pregnant women who received more than 20% of energy intake from protein were associated with retarded fetal growth (21) so this level was used as criteria to divide the level of protein intake into moderate and high protein intake.

#### 14.2 The quality of protein intake

The good quality protein intake : the ratio of animal protein and vegetable protein was 1:2 or more (21).

The poor quality protein intake : the ratio of animal protein and vegetable protein was less than 1:2 (21).

**Antenatal care:** prenatal care visits at the hospital in the second trimester and follow-up until delivery at the following schedule:

- once every months until the 28<sup>th</sup> - 32<sup>nd</sup> weeks of gestation.
- once every 2 weeks until the 36<sup>th</sup> week of gestation.
- once a week up to the onset of labor.

**Parity:** the status of women with regard to number of times she had given birth.

**Birth or pregnancy interval:** the time between the previous birth and conception of the current pregnancy.

**Moderate energy expenditure:** pregnant women with moderate activity during pregnancy were included in this study. The moderate activity at work per day for females was referenced as follows:

Moderate activity :

Sleeping (1 kcal/min) 8 hours

Occupational activities (2.7 kcal/min) 7 hours

Discretionary activities:

- Socially desirable and household tasks (3 kcal/min) 2 hours

Residual time (1.4 kcal/min) 7 hours

Activities derivation of average values of the energy cost was shown in appendix B (22).

**Low and lower middle socioeconomic status:** average household income was approximately 99,144 baht per year (23) so this study defined low and lower middle socioeconomic status equal to less than 10,000 baht per month.

**Marital status:** pregnant women who were married and/or lived with husbands were included in this study.



## CHAPTER II

### LITERATURE REVIEW

Pregnancy is a period of increasing nutritional needs. Proper health care and dietary intake are essential for pregnant women. Generally, both genetics and environment may influence embryo and fetus any time during development. This chapter will review selected literature related to the present study under the following headings: (1) Factors determining infant birth weight and length: prepregnancy factors that affect infant birth weight and length including maternal height and prepregnancy weight; demographic information such as maternal age, socioeconomic status, marital status; parity and birth interval; and (2) Factors that affect infant birth weight and length during pregnancy including nutritional factors such as dietary intake, gestational weight gain, energy expenditure, hemoglobin, health practice such as antenatal care, smoking, alcohol and caffeine consumption and the complication during pregnancy. The headings of factors determining gestational age are similar to those determining infant birth weight and length.

## **I. Factors Determining Infant Birth Weight and Length**

### **1. Prepregnancy factors that affect infant birth weight and length**

#### **1.1 Maternal stature**

##### **1.1.1 Maternal height**

Numerous studies found that the taller women, the heavier their infants. The results of Love (24) were similar to those of Nondasuta (25) that there were associations between infant birth weight and the height of the mother. The height of mother was significantly and positively correlated with infant birth weight. Chumnijarakij et al. (26) reported that mother whose height were under 150 cm had 1.41 times greater risk to give low birth weight infants than taller mothers did. Maternal height above 160 cm ceased to be a low birth weight risk factor. Similar finding was treated by Fedrick and Adelstein (27) that low birth weight was almost 3 times higher in shorter women ( less than 1.57 m ) than in taller women ( more than 1.67 m).

Roman (28) reported that one of the factors which contributed to low birth weight in low income groups was the short maternal stature. With the maternal height increase, there was a significant increase in infant birth weight. Ratakallio et al. (29) found that low prepregnancy weight and short stature predicted an increased risk in both cohorts on small for gestational age. However, the size of the newborn is not paternal height. This may be a natural survival mechanism to protect small mothers from having difficulties during labor (30).

### 1.1.2 Maternal prepregnancy weight

Maternal weight is one of the important parameters measured during antenatal visits. Prepregnancy weight provides adequate baseline data for computation of weight gain during pregnancy. Gormican et al. (31) found that women who had heavier prepregnancy weight tended to have heavier babies. The underweight pregnant women delivered infants of low birth weight than normal weight women did (32). Also Chumnijarakij et al. (26) noted that maternal weight prior to delivery was statistically associated with low birth weight. Mothers whose weight was under 45 kg had 1.3 times greater risk of giving low birth weight infants than mothers whose weight was in the Thai standard range (45 to 54.9 kg). On the other hand, mothers whose weight was over 55 kg had less risk of low birth weight than mothers with weight in the Thai standard range. Similar findings by Varma (33) showed that the incidence of low birth weight infant was higher when maternal weight was 50 kg or less.

Ratakallio et al. (34) had two cohort studies in North Finland and found that low prepregnancy weight and short stature of mothers predicted an increase risk on low birth weight. The mothers who had prepregnancy BMI under 20 had 1.37 times risk of giving low birth weight infants than women in the normal range (24,25). The incidence of low birth weight was lowest among overweight women and the obese women, BMI 30-35, in the later cohort. Mavalankar et al. (35) found that women who weighed 40 kg or less were 2.6 times greater risk of small for gestation as compared to those who weighed more than 50 kg.

The maternal body size had an effect on fetal size. Small mothers tended to give birth to small offsprings than the larger mothers did. Therefore, in the present study,

the critical cut-off point for risk of poor infant outcomes were maternal height of <148 cm and prepregnancy weight of <45 kg (36).

## **1.2 Sociodemographic factors**

### **1.2.1 Maternal age**

Several studies have shown that maternal age was related to infant birth weight. The optimal age of pregnant women is between 20 and 30 years old (37,38,39). Many studies have indicated that maternal age is an important factor for physiological and gynecological maturity. The mothers who were younger than 20 years old had high risk of low birth weight because of gynecological immaturity. Mother who were older than 30 years old had high risk also because of ovarian function impairment.

O'Sullivan et al. (40) studied in Boston City Hospital about the relationships of infant birth weight and maternal age. A rise in age was found to be associated with an increase in mean birth weight. Kaltreider and John (41) found that about 20.3% and 5.8% of low birth weight infants delivered from mothers under 19 years and older mothers respectively. Similar findings revealed that mothers who were under 19, between 35 and 39, and over 40 years old were 1.80, 1.32 and 3.62 respectively of relative risk to give low birth weight infants as compared to mothers aged 19 to 34 (26).

Eisner et al. (42) noted that women who had first pregnancy at age 35 and over had increased risk of delivering low birth weight infants compared to mothers age 18 - 34 years, and the odd ratios ranged from 2.0 to 8.86. Multigravida over 35 years of age also had an increase risk but not so great as for primigravida.

The optimal age for healthy pregnant women to delivery normal birth weight infants was between 20 - 35 years old.

### 1.2.2 Socioeconomic status

Socioeconomic factors including occupation, income and education were related to proper dietary intake of pregnant women. Poor and low educated women tend to be unhealthy before pregnancy and were at high risk to give low birth weight infants.

Suttipreyasri et al. (43) studied in Thai pregnant women and found that mothers from low socioeconomic group were at higher risk of having low birth weight infants than those from high socioeconomic one across every category of weight gain during pregnancy. Mothers from the low socioeconomic group were also inferior in terms of knowledge, attitude and practice concerning nutrition during pregnancy.

Chumnijarakij et al. (26) found that mothers of lower educational attainment (less than high school) had greater risk of low birth weight than mother with college education or higher. Mothers who had uneducated, school attainment of 4, or 9 years were at risk of low birth weight about 1.67, 1.33, and 1.39 times respectively compared with mothers who had school attainment up to 12 years. However, the results of Wildschut's study (44) showed that social class did not predict low birth weight in either primiparous or multiparous women.

Several studies indicated that socioeconomic including maternal education, occupation, labor force participation and household income were strongly inter-related and these factors affected food intake, proper nutrition and prenatal care.

### **1.2.3 Maternal marital status**

Maternal marital status is closely linked to socioeconomic status and psychological mechanism such as stress which affects intrauterine growth. Numerous studies found that maternal marital status was associated with low birth weight. Unplanned or unwanted pregnancy especially in teenagers and unmarried women tended to have low prenatal care which led to health problems (45).

Chumnijarakij et al. (26) studied in Thai women and found that mothers who were not registered were 1.4 times at greater risk of low birth weight than mothers with registered marriages. Mothers who were divorced, widowed or separated had higher risk of low birth weight than currently married mothers.

## **1.3 Obstetric factors**

### **1.3.1 Maternal parity**

Maternal parity was reported to influence the size of the newborn at birth. In general primiparous women gave birth to infants who were smaller than those of multiparous women (27,46).

Suttipreyasri et al. (43) studied in Thai pregnant women and revealed that the incidence of low birth weight was high in the first parity groups. These data were similar to Malvalankar's study (35) that first births were high risk of small for gestation.

Chumnijarakij et al. (26) reported that mothers of first parity had a relative risk of low birth weight of 1.72 compared with mothers of second or third parity. Mothers of fourth or greater parity had 33 times the risk of low birth weight than mothers of

second or third parity. Koetsawang et al. (47) studied in healthy middle-class Thai women and had a similar finding that mean infants' birth weight in the first parity was significantly lower than in subsequent parity for both male and female infants.

Selvin and Garfinel (48) studied more than 1.5 million single births in New York and found that when the influence of age was ignored the number of low birth weight was greatest for birth order 1 and 6<sup>+</sup>. Maternal parity is one of the important factors considered to affect pregnant women in delivering low birth weight infants especially for birth order 1 or more than 4.

### 1.3.2 Birth or pregnancy interval

A short interval of pregnancy might lead to poor fetal outcomes. The birth interval < 6 months is a risk factor for low birth weight. Eisner et al.(42) found that higher rate of low birth weight was in women with shorter pregnancy interval. The high level of significance was associated with pregnancy interval of <6 months. Papiernik et al. (49) found that lower birth weight were observed for births following a pregnancy interval <1 years.

Rawling et al. (50) studied in 1,922 white and black women and found that a short interval between pregnancies was a risk factor for low birth weight and such intervals were more common among black than white women. The interpregnancy interval of less than nine months and three months were associated with a significantly greater prevalence of low birth weight among black and white women respectively. Klebanoff et al. (51) found that mean birth weight increased from 3,101 g for intervals of < 3 months to 3,193 g for interval of 15-17.9 months.

## **2. Factors that affect infant birth weight and length during pregnancy**

### **2.1 Nutritional factors**

#### **2.1.1 Maternal dietary intake**

Food availability is an important factor that can affect maternal-fetal exchange. The placenta grows faster than the fetus and for a significant period it is actually larger than fetus. During the last half of gestation, however, the fetus grows considerably faster (52). These nutrients are acquired from mother via the placenta. The maternal-fetal nutritional relationship is the process by which appropriate quantities of these nutrients, derived from food, are assimilated by mother and reach their ultimate destination to support cell division, replication and growth of the functional organ systems in fetal placental unit and ultimately, the baby, while enhancing the maternal tissues and preparing her for lactation. The most rapid period of fetal growth is between the 12<sup>th</sup> and 26<sup>th</sup> weeks of gestation. Between the 32<sup>nd</sup> and 36<sup>th</sup> weeks the rate of fetal weight gain reaches its peak at 200-225 g/week (53). So extra energy, protein, minerals and vitamins are needed. The average extra energy cost of this typical pregnancy has been calculated to be about 335 MJ (80,000 kcal) over the 9 months period. The pregnancy needs extra 630 MJ /day (150 kcal/day) during the first trimester and 1465 MJ/day (350 kcal/day) during the second and third trimesters (51). The quality of diet has long been known to affect the fetal condition, a better maternal diet being associated with better condition of the offspring. During the first 12 weeks of gestation, women's food intake either remains at prepregnancy level or in a high proportion may even decrease because of nausea and vomit. In subsequent weeks

appetite increases and most women perceive that they are eating more than before pregnancy, but the magnitude of this increase and its pattern remain undefined.

Numerous studies have suggested that inadequate maternal dietary intake during the second and third trimesters may exert the largest impact on fetal growth because the fetus grows most rapidly during the third trimester. During the second trimester, the additional caloric cost of pregnancy is reflected primarily in growth of maternal compartment and the third trimester expenditures are reflected mainly in fetal and placental growth (52).

All nutrients are essential for pregnant women but energy and protein are more relevant. Protein plays a major role in pregnancy outcome since it is necessary for maternal tissue growth and fetal growth. During early pregnancy the fetal component is minimal whereas the requirement for maternal volume expansion and tissue growth may be substantial. Late in pregnancy, the fetus may account for the major increase in protein needs. The fetus receives a continuous supply of amino acid through the placenta. Most amino acid and all the neutral amino acid cross the placental by active transport and present in fetal blood in higher concentrations than in maternal blood. The safe levels of additional protein computed in the manner are 1.2 g, 6.1 g and 10.7 g per day in the first, second and third trimesters respectively. During fetal growth not only an increase in the amount is needed, but also the quality of protein in the diet is important for fetal growth. Animal protein foods are recommended for much of protein intake, although use of the incomplete plant protein is appropriate. Protein foods are utilized most efficiently when they are available distributed throughout the day. Maternal protein restriction alone and in combination with energy restriction,

results in consistently decreased fetal growth in many species. It is not only decreased body weight and growth but also decreased number of cells and a variety of biochemical change. The balance energy and protein diet should be considered because the dietary intake with high protein will be harmful for pregnant women (16).

In Dutch famine study (54), the pregnant women consumed less than 1,000 kcal per day, 11 g animal protein, the average maternal weight gain during this period fell about 2 kg and all birth weight was less than 2,250 g. When dietary intake improved during the third trimester after the famine the birth weight increased.

Buke et al. (55) studied the newborns of 216 Boston mothers with a wide range of dietary habits. In this series the better nourished mothers clearly gave birth to healthier offspring. An increase in birth weight and length accompanied each increment in maternal protein intake from less than 45 g to greater than 85 g daily.

Kanittasen (56) studied the relationships between protein intake of Thai pregnant women who had adequate energy intake in the third trimester and found that there was no significant difference in infants' birth weight, birth length in the third trimester between higher and lower of the average maternal protein intake groups. However, all the average value of fetal outcomes of the higher group was better.

Wolff et al. (57) studied the relationships between maternal diet and infant birth weight in 459 Mexican mothers. Seven distinct eating patterns were identified nutrient dense, traditional, transitional, nutrient dilute, protein rich, high fat dairy, and mixed dish. The results indicated that the nutrient dense (fruits, vegetables, low fat dairy, etc.) and protein rich (low fat meats, processed meats, dairy desserts, etc.) eating pattern were associated with increased birth weight and the transitional eating

pattern( fat and oils, bread, high fat meat, sugar, etc.) was associated with decreased birth weight.

Because energy and protein are essential for pregnant women to weight gain, supplementation of diet to achieve a given daily nutritive value during pregnancy has been attempted in several locations as a consequence. The various results of those studies are as follows:

Briend et al. (58) found that the energy supplement during pregnancy was likely to be affective only for mothers with lean body mass deficit. For other women, any supplement that results in only fat deposition increase seemed to be unlikely to have a beneficial effect on fetal growth.

Viegas et al. (59) studied in Asian mothers who received one of the three supplements from the 18<sup>th</sup> - 38<sup>th</sup> weeks of pregnancy: (1) vitamin only (Vi) - vitamin C 30 mg daily , iron 3 mg daily; (2) energy (EnVi) - 10,000-19,000 kcal/ trimester, all from carbohydrate plus vitamin; and (3) protein energy (PrEnVi) - energy and vitamins as before, but with 11% energy from milk protein. By the 28<sup>th</sup> week the mothers who received the protein energy supplement had more weight and more fat than vitamin groups only. Neither protein energy nor energy supplement alone enhanced intrauterine growth. Later (9) they studied in 45 mothers who were at the 28<sup>th</sup> week of gestation and nutritionally at risk received one of three supplements during third trimester: (1) vitamin only - multivitamin containing vitamin A, B, C and D; (2) energy (10,000 - 30,000 kcal ), all from carbohydrate, plus vitamins; and (3) protein energy - energy and vitamins as before but with 5-10% of energy from milk protein. Eighty-three mothers regarded as adequately nourished at the 28<sup>th</sup> week of

gestation also received one of the three supplements. In the nutritionally at-risk mothers, the protein energy supplement was associated with a heavier birth weight and heavier weight for gestational age. Supplementation did not lead to improved intrauterine growth in those mothers who were adequately nourished.

Metcoff (60) did a prospective randomized controlled nutrition intervention study. Four hundred and ten women were enrolled at midpregnancy. The participants received vouchers for supplements of milk, eggs and cheese, intended to provide 40-50 g protein and 900-1000 kcal daily. This was intended as an addition to the regular diet (1.1 g protein/kg/d, 28 kcal /kg/d). The mean infant birth weight in supplement group was higher than unsupplement group.

Mardones-Santander et al. (61) studied the effect of powdered milk-based fortified product (V-N) and powder milk (PUR) on pregnancy outcomes in a group of underweight gravidas. They found that women in the V-N group had greater weight gain and mean birth weight ( 3,178 vs 3,105 g,  $p < 0.05$  ) than those in the PUR group.

Tontisirin et al. (61) gave supplementation in pregnant women and found that among undernourished mothers, supplementation of as little as 13 g protein and 350 kcal daily in the last trimester significantly improved maternal weight gain and birth weight of newborns.

Kardtjati (63) provided two different supplement formulas in pregnant women at the 26<sup>th</sup> –28<sup>th</sup> weeks of gestation: (1) HE supplement ( 50% fat, 40% glucose, 10% casein ) provided 465 kcal and 7.1 g protein; and (2) LE supplement ( 28% glucose, 50% casein ) provided 52 kcal and 6.2 g protein. Supplement intake was variable. Testing of the effect by treatment and compliance was thus done by subcategories (HE

1-3 and LE 1-3, corresponding to <45, 45-89, and > 90 packets of supplement consumed). Analysis of variance did not show significant difference among the six subcategories in the third trimester weight gain. The participants' supplementation for short duration of the last 90-110 days of pregnancy was not sufficient to improve maternal nutrition as judged by anthropometry.

Adams et al. (64) gave supplementation in healthy young women. They were randomly assigned to one of the three dietary regimens: (group 1) normal diet plus a high-protein beverage supplement; (group 2) normal diet plus a low-protein beverage supplement; and (group 3) normal diet plus a vitamin-mineral preparation. The protein beverages were supplied in 8-oz cans. Each woman in groups 1 and 2 was expected to drink two cans of these beverage cans each day. Women in group 3 were provided prenatal vitamin-mineral capsules, with instruction to take one each day. Women in all three groups were prescribed iron supplement. Each daily allowance of two cans of high-protein beverage contained 40 g protein (casein and soy isolate), 470 kcal, and vitamins and minerals. The daily amount of low-protein beverage was similar except that it contained only 6 g protein (of the same type) and total of 320 kcal. They found that the mean birth weight was similar in all three groups.

Many studies of caloric and/or protein supplement in Montreal, Bogota, Guatemala, and Taiwan (10,11) found that mothers with supplementation had infants whose birth weight was higher than those from no supplementation mothers. Many mothers in those studies were at least in moderately undernourished women so the supplement should be beneficial for them and their infants.

Besides human research, some research works were done in animals such as: Morgan et al. (65) studied in pregnant rats. Those rats were ad libitum fed diets of varying casein content from conception. The control rats were given 25% casein plus 2.5% DL methionine after 17 days of gestation until delivery. The second experimental group was given 25% casein plus 2.5% DL methionine throughout pregnancy. A protein supplement given in late pregnancy (control group) at the time of the fetal exponential growth phase was found to increase the weight.

Lederman and Rosso (66) did the study on the effect of equal amounts of carbohydrate or protein supplement in rats by given protein supplement from day 15 of gestation after a 50% food restriction for the previous 10 days. Maternal body weight and composition, fetal and placental weights of supplemented rats were compared with control, fed ad libitum (well balanced pelleted diet) and with rats maintained on a 50% food restriction until term. The control group had the highest gain in net maternal weight followed by protein supplemented and the carbohydrate supplemented rats. Both types of supplements increased fetal weight equally compared with the restricted group but not to the control group.

Some research works showed that high protein intake had adverse effect as Phillip et al. (67) found that low levels of iron and protein intake led to heavier babies, or conversely that high levels caused smaller babies.

The overview research about effect of energy and protein intake by Kramer (68) suggest that a balanced increase in energy and protein intake result in modest increase in weight gain and fetal growth. Surprisingly, these increases do not appear to be

beneficial in undernourished women, nor do they seem to confer long-term benefits to the child in terms of growth or neurocognitive development.

The dietary intake during pregnancy should be a balanced energy-protein diet for increasing infant birth weight and length. High protein may have an adverse effect on fetal outcomes.

### **2.1.2 Gestational weight gain**

Maternal weight gain during pregnancy is an essential component of normal growth and development of mother and her fetus. Maternal prepregnancy weight and weight gain indicates maternal nutritional status during pregnancy. Gestational weight gain is a potential cause of maternal and fetal outcomes. Energy is the major nutrient determinant of gestational weight gain other than specific nutrient deficiencies which may restrict weight gain.

Rosso et al. (69) found that the protein restricted rats had a significant reduction in body weight, fetal and placental weight and the major cause of the fetal growth retardation was associated with maternal malnutrition.

Anderson et al. (70) divided the subjects into 3 groups: (1) 50% dietary restriction during the anabolic phase ( days 15-20 ) of gestation; (2) 50% dietary restriction during the catabolic phase; and (3) 50% dietary restriction throughout gestation. They found that restriction of dietary intake during the anabolic phase resulted in decreased maternal weight gain. Feeding an adequate diet during catabolic phase following restriction during anabolic phase caused only slight decrease in net maternal weight. Dietary restriction during catabolic phase or throughout gestation

caused increase in net maternal weight loss. Average term placental weight was lowest only in the group fed with the restricted diet during the catabolic phase of pregnancy.

Besides animal experiments, there were many studies in humans about gestational weight gain and fetal outcomes as follows:

Varma (33) studied in 3,002 pregnant women who delivered at St. George's Hospital and found that the incidence of infants with low birth weight was higher if the maternal weight gain was less than 6 kg. Eastmann and Jackson (71) studied in white and black women, the 39<sup>th</sup> - 42<sup>nd</sup> weeks of gestation with no complication and normal labor. The subjects were divided into 2 groups depending on prepregnancy weight: (1) low group ( body weight <120 lb ); and (2) high group ( body weight >120 lb ).

Maternal weight gain in each weight group was also divided into 2 groups: (1) high weight gain ( >13 lb ); and (2) low weight gain ( <11 lb ). Average birth weight was compared among combined prepregnancy weight status and weight gain groups. They found that average birth weight was lowest among low prepregnancy weight and low weight gain. Average birth weight of infants born to low prepregnancy weight and high weight gain women was 411g higher than that of infants born to low prepregnancy weight and low weight gain women. The highest average birth weight was shown among high prepregnancy weight and high weight gain in both white and black women.

Brown et al. (32) studied prenatal and postpartum care of 654 women divided into 3 groups: (1) underweight women weighed less than 80% standard weight; (2) moderate underweight women weighed 80-90% standard weight; and (3) normal weight women weighed 90-120% standard weight. They found that prepregnancy

weight status was not associated with the amount of weight gain during pregnancy and underweight women who gained the same amount of weight as normal weight women delivered infants of low birth weight and length.

Rosso (72) studied in 262 singleton pregnancy and full term delivery by dividing the subjects into 3 groups: (1) underweight ( $<89\%$  standard weight); (2) normal ( $90-110\%$  standard weight); and (3) overweight ( $>110\%$  standard weight). The finding was that pregnancy weight gain had an influence on mean birth weight in both prepregnancy underweight and normal weight but no significant effect on the overweight group so that the heavier women were advised to increase their initial weight proportionally less than normal and underweight subjects. The similar findings by Frentzen (73), who studied two groups of pregnancy were: (1) those who were average weight ( $90\%$  to  $120\%$  of standard weight for height); and (2) highly overweight ( $>135\%$  of standard weight for height). He found that pregnancy weight gain in the average weight group affected birth weight but the highly overweight group did not significantly affect birth weight. Infant outcomes for both groups were similar.

Edwards et al. (74) revealed that to optimize fetal growth weight gain of 7-11.5 kg for obese women and 11.5-16 kg for normal weight women appeared to be appropriate.

Tilon et al. (75) studied in obesity pregnancy and found that the mean birth weight of the infants of obese women was 163 g greater than that of control subjects, no difference was observed in infant length.

Mitchell and Lerner (75) had a retrospective study in 1,080 singleton pregnancies of middle class women. It was found that 1 lb gain in maternal weight was associated with a 6 g increase in birth weight. And later (77) they studied in 362 pairs of underweight women ( 90% Metropolitan Relative Weight ) and normal weight (90 % to 110%) women who were matched by age, occupation, height, parity, race and smoking habit. They found that the infants of underweight women who gained <9 kg had mean birth weight 361 g less than the mean birth weight of infants of underweight women who gained more weight ( >9 kg).

Picone et al. (78) studied in urban hospital clinic. The subjects were assigned to one of the four groups depending on their predicted weight gain. These group were: (1) low weight gain ( LWG <15 lb ) & smokers; (2) low weight gain & nonsmokers; (3) adequate weight gain ( AWG >15 lb ) & smokers; and (4) adequate weight gain & nonsmokers. They found that weight gain of women in the LWG groups was significantly less than that of those in the AWG groups during each of the three trimesters. Weight gain was not significantly correlated with prepregnancy weight. They concluded that low weight gain was associated with lower food intake. In contrast, smoking and stress may cause low weight gain by reducing the utilization of calories for weight gain.

Gueri et al. (79) noted that average increment of weight during pregnancy was 20 % of the prepregnancy weight for normal birth weight. The average weight increase was estimated to be approximately 12 kg, of which about 3.5 kg (29 %) represented the weight of the fetus at term, the uterus, and amniotic fluid; the placenta accounted for about 2 kg; and the breast for 1 kg. One and one-half kg were fluid

retention and 4 kg were fat deposit. They suggested that during the second and third trimesters of pregnancy weight gain should be about 0.4 kg /week.

Abrams et al. (80) studied in nonobese, white women and found that fetal size was correlated positively with the rate of weight gain in each trimester. Another Abrams' study (81) found that specific patterns of maternal weight gain, particularly weight gain during second trimester were related to infants' birth weight. Jackson et al. (81) found that mothers of infants with birth weights between 5.5 and 8.4 lb gained an average of total 14.6 lb (6.62 kg) and about 7.0 net lb over the third trimester compared to the average overall gain of 13.3 lb (6.03 kg) for the entire sample. Beal et al. (4) studied in 95 pregnancies, in which 38 were primiparous and 57 were multiparous, and found that birth length was significantly correlated with maternal preconception weight and with weight gain in the third trimester, women with large weight gain during pregnancy tended to bear infants who were both longer and heavier.

Ancrì et al. (83) noted that the correlation between maternal weight gain and infant birth weight was not significant but Naeye (84) found that maternal weight gain had its greatest correlation with the outcomes of pregnancy when offspring were male.

It's known that total weight gain during normal pregnancy range from 10-12 kg were classified as reference groups. For the underweight women who gain weight less than 6 kg at term, significant fetal growth retardation occurs.

### **2.1.3 Energy expenditure during pregnancy**

The working condition and timing in any activities during pregnancy is an essential factor because it is related to physical and psychological stress that affects the fetus.



Heavy physical work, maternal emotion and stress were harmful to the fetus during pregnancy. During pregnancy additional nutrients are required for maternal needs and fetal growth. The mothers who work hard during pregnancy require increasing energy over mothers who are less active. Fedrick and Adelstein (27) found that increasing low birth weight infants delivered from mothers who worked outdoors compared with the unworked mothers. These data were supported by Tafari et al. (85) which studied in 181 pregnant women in Ethiopia. Maternal intake of calories and protein were similar in the high and low physical activity groups which were below 70 percent of WHO / FAO recommended standards. The mothers who engaged in heavy physical labor during pregnancy had much smaller pregnancy weight gain, and decreased the weight of their infants than the less active mothers did.

The kinds of work that the pregnant women had done were an essential point to study so Naeye and Peter (86) studied 7,722 women who could be placed in one of the three pregnancy work categories: (1) did not work outside the house; (2) employment outside that required sitting most of the time; and (3) employment outside that required standing most of the time. The results showed that gestation was not shortened but newborns of women who worked in the third trimester weighed 150-400 g less than newborns of mothers who remained at home. The frequency of large placental infarcts progressively increased when women continued stand-up work into late gestation. The association of the prolonged standing required by certain jobs with the rate of low birth weight deliveries was examined by Teilelman et al. (87). The job categories were defined according to the following criteria: (1) standing - jobs which required standing more than 3 hours per day predominantly in one position without much activity; (2)

active - jobs which involved continuous or intermittent walking with active range of motion; and (3) sedentary - jobs which required less than 1 hour of standing per day and less than 1 hour of active motion per day. They found that low birth weight rate was higher among women in standing group (5.5%) compared with those in the sedentary (4.0%) and active groups (4.0%), but this association was not significant when confounding factors were controlled.

Manshande et al. (88) studied the influence of resting versus hard work on the fetal growth and had a supporting data that the duration of rest had a strong influence on birth weight and length in the newborn females but to less extent in the newborn males.

#### **2.1.4 Hemoglobin level**

The epidemiological evidence suggests that anemia during pregnancy could be harmful to the fetus. Anemia in pregnancies are also one of the risk factors of low birth weight. Among healthy human beings, pregnant women and rapidly growing infants are most vulnerable to iron deficiency. During pregnancy, more iron is needed primarily to supply the growing fetus and placenta and to increase the maternal red mass. The higher hemoglobin concentration as a result of an improving iron supply not only increases the oxygen-carrying capacity but also provides a buffer against the blood loss that will occur during delivery. Hemoglobin and hematocrit levels decrease during pregnancy are called physiologic anemia but if hemoglobin level is below 11 g/dl during the first and third trimesters or below 10.5 g/dl during the second trimester it is defined as anemia (16). Iron deficiency is common among pregnant women. Maternal anemia

will cause low birth weight. Chumnijarakij et al. (26) revealed that an hematocrit of under 30% had a relative risk of low birth weight of 1.56.

Edward et al. (89) had studied in 354 underweight patients compared with matched control subjects of normal weight and found that underweight women particularly if they were anemia had a higher incidence of low birth weight infants despite adequate weight gain.

## **2.2 Health practice factors**

### **2.2.1 Antenatal care**

The antenatal care is essential for pregnant women because it reveals that mothers who lack ANC have a high risk of low birth weight infants. Antenatal care has two major components: (a) health promotional activities and prophylaxis aims at improving the health and nutrition of all pregnant women; and (b) early detection and prompt treatment of obstetric problems. Early identification, registration and regular follow-up of pregnant women are critical to both components of antenatal care. It is also increasingly recognized that quality of care is as important as coverage of pregnant women if adverse obstetric outcomes are to be averted (90).

From the study of Showstock (91), it was found that prenatal care was adequate if it was started in the first trimester and adequate prenatal care was associated with an increase in birth weight of 126 g for black babies and 105 g for white babies.

Chumnijarakij et al.(26) found that lack of prenatal care or having equal or less than four visits increased the mothers' risk of low birth weight by 2.26 and 1.59 respectively.

### 2.2.2 Maternal smoking

Smoking during pregnancy has a detrimental effect on fetal growth. Maternal smoking is associated with small placentas and thickening of the villous membrane in early pregnancy. In late pregnancy there is also a reduction in the capillary volume of the villi and at both stages of pregnancy necrosis of the syncytiotroblast is found. The villi also show a thickened basement membrane and increased collagen. These presumable processes contribute to reduction in birth weights (16). The mothers who have smoked heavily tend to have lower birth weight infants (92,93).

Nilson et al. (94) showed the hazardous effect of smoking, by which nutrient-carrying capacity of plasma was reduced and viscosity of the maternal blood was increased which reduced uteroplacental circulation.

Haste et al. (95) studied the effect of smoking on dietary intake of pregnant women. They found that the nonsmokers had higher intake of almost all nutrient than the smokers did and the nutrient density of their diet was greater. Dietary intake was reduced in pregnancy particularly in smokers. However, Harworth et al. (96) noted that fetal growth retardation due to smoking was not caused by the dietary intake.

Roquer's results (97) confirmed that smoking in pregnant women reduced weight and length at birth. Weight reduction in term newborns was 458 g between mothers smoking more than 10 cigarettes/day and those who did not smoke.

Smoking behavior has definitely been to retard fetal weight gain. The reduction in potential birth weight was proportional to the number of cigarettes smoked. The higher number of cigarettes, the lower the birth weight. However, very few Thai women generally smoke as compared to Western women.

### 2.2.3 Maternal alcohol consumption

Numerous studies indicated that alcohol drinking during pregnancy will increase risk of stillbirth, low birth weight, and physical malformations. The evidence is clear that women who drink heavily during pregnancy place their unborn children at substantial risk for fetal damage, and physical and mental growth retardation in infants. This phenomenon is known as Fetal Alcohol Syndrome. The children born with Fetal Alcohol Syndrome may be below average in birth weight and length. The current hypothesis is that high alcohol level built up in the fetus produces a direct toxic effect that is most severe in the early phases of pregnancy during blastogenesis and cell differentiation. During the second and third trimesters alcohol consumption may affect fetal brain development and have greater effect on fetal size (98,99). Another theory is the predicted effect of alcohol which may be caused by maternal malnutrition (21).

The risk from drinking alcohol increased with the high amount consumed. This was supported by the study of Wright et al. (100) which was investigated in 900 pregnant women. The results showed that women who drank more than 100 grams of alcohol a week ( heavy drinkers ) had double the risk of delivering a baby on or below the 10<sup>th</sup> percentile compared with women who drank less than 50 grams a week ( light drinkers ). Kennedy (101) also found that the placental weight was reduced with increasing alcohol intake.

Rosett et al. (102) speculated that fetal growth retardation probably resulted from the multiple effects of high ethanol concentration in the maternal placental fetal system. These included effect on fetal metabolism and endocrine function, transport of amino acid across the intestinal mucosa, and altered hepatic metabolism in the mothers.

#### **2.2.4 Maternal caffeine consumption**

Another factor of maternal behavior during pregnancy apart from smoking and alcohol drinking is drinking coffee or tea, which is widely preferred among Thai people now (26). Hinds et al. (103) found that total caffeine consumption of > 300 mg (about 3 cups of coffee) daily during pregnancy had been associated with infant birth weight.

Martin et al.(104) studied in 3,891 women at Yale-New Haven Hospital and demonstrated a dose response of caffeine intake in increasing risk of low birth weight in single deliveries after 36 weeks. The adjusted relative risk of low birth weight delivery was 1.4 for mothers consumed 1-150 mg of caffeine per day, 2.3 for mothers consumed 151-300 mg of caffeine daily and 4.6 for mothers consumed >300 mg daily. Corresponding decreased in mean birth weight were 6 g, 31 g, and 105 g respectively.

#### **2.2.5 The complication during pregnancy**

The maternal complication during pregnancy will be harmful for mothers and infants. During pregnancy, if pregnant women have some physical disorder such as hypertension, diabetes mellitus, heart disease, chronic renal disease, severe pre-eclampsia, etc., these will affect fetal growth.

Metcoff et al. (105) reported that maternal complication observed during pregnancy may influence infant birth weight and length. Infections, including urinary tract infection, vaginitis, endometritis, etc., had a negative effect on birth weight. Pre-eclampsia and toxemia of pregnancy were associated with significant reduction in birth

weight. In addition, maternal bleeding which resulted in fetus oxygen insufficiency caused low birth weight infants in many studies (106,107).

## **II. Factors determining the gestational age**

### **1. Prepregnancy factors that affect the gestational age**

#### **1.1 Maternal stature**

##### **1.1.1 Maternal height**

Short mothers tend to have shorter gestational age than normal height mothers. Baird et al. (45) studied Aberdeen primigravida and classified their height into 3 groups: (1) 64 inch and over ( tall ); (2) 63 inch to 61 inch ( medium ); and (3) < 61 inch ( small ). It was found that the incidence of premature delivery was 4.9, 7.7 and 12.1% respectively. The stunted growth of mothers gave rise to high rate of prematurity. Nondasuta et al. (25) found that gestational age increased with the increase in maternal height.

##### **1.1.2 Maternal prepregnancy weight**

Mothers who are underweight and have inadequate weight gain during pregnancy tend to have premature births. Riopelle et al. (108) found that heavy mothers tended to have long gestations. Wen et al. (39) found that low prepregnancy weight was statistically related to the preterm delivery.

## **1.2 Sociodemographic factors**

### **1.2.1 Maternal age**

Younger pregnant women (under 20 years old) and older pregnant women (over 35 years old) have high risk of preterm delivery. Zlatnik and Burmister (109) had similar findings as Baird (45) who found that primigravida mothers younger than 20 years old had high rate of prematurity because the uterus may somehow structurally or functionally be less able to carry a fetus to term and the uterine vasculature was less well-developed in those young women.

Wen et al. (39) studied in younger (<17 years old) and older (>30 years old) pregnant women and found that maternal age was related to preterm delivery.

### **1.2.2 Socioeconomic status**

Poverty and low education of pregnant women were associated with premature labor. Baird et al. (45) found that mothers in high socioeconomic class had the lowest rates of prematurity. Ericson et al. (110) showed that high, medium, low socioeconomic status had relative risk of preterm delivery of 1.0, 1.3 and 1.8 respectively. Arbuckle and Sherman (111) found that maternal education and family income were associated with preterm delivery.

### **1.2.3 Maternal marital status**

Kramer et al. (112) reported that unmarried mothers tend to have preterm delivery than married mothers. This factor is an indirect factor related to other factors which are associated with preterm delivery.

### **1.3 Obstetric factors**

#### **1.3.1 Birth or pregnancy interval**

Papiernik and Kaminski (49) examined the effect of pregnancy interval on gestational age and found that no significant increase of short pregnancy interval (1 year) among women who gave birth to premature infants.

Lang et al. (113) found that the highest risk of prematurity occurred in women who had an interval of 3 months or less, 8.3% of those women gave birth to premature infants. The risk of prematurity decreased with increasing interpregnancy interval up to approximately 24 months. However, the effect of birth or pregnancy interval on gestational duration had inadequately been studied.

## **2. Factors that affect the gestational age during pregnancy**

### **2.1 Nutritional factors**

#### **2.2.1 Maternal dietary intake**

Maternal dietary intake not only affect fetal size but also affected gestational age. Delgado et al.(114) had supplemented two types of liquid diet to pregnant women in four villages. In two villages, a high protein calorie supplement called "atole" contained 11.5 g of protein and 163 kcal per 180 ml was made available. The other two villages were provided with a fruit-flavored drink called "fresco" which contained no protein and supplied only 59 kcal per 180 ml. Energy intake from the supplement, whether accompanied by protein as in atole group or not as in fresco group, was significantly associated with the length of gestational during the first and second , but not the third trimesters of pregnancy. Supplement intake during the

first trimester of pregnancy was strongly more related to length of gestation than intake during the rest of the pregnancy.

Rush et al. (13,115) did a study in poor black pregnancy in New York whose gestational age was less than 30 weeks of gestation at the time of interview. At least one of the following additional criteria also had to be fulfilled: (1) low prepregnancy weight ( under 110 pounds at conception ); (2) low weight gain at time of recruitment; (3) at least one previous low birth weight infant; and (4) a history of protein intake less than 50 g in 14 hours presiding registration, as calculated by the nutritionist from a 24-hour qualitative dietary recall. The three study groups received one of the diets:(1) supplement consisting of two 8-oz cans of beverage daily that provided a daily total of 40 g of animal protein, 470 kcal and array of vitamins and minerals; (2) complement consisting of two 8-oz cans of beverage daily that provided a daily total of 6 g of animal protein, 322 kcal and the same amount of vitamins and minerals; and (3) non-intervention ( control group ). They found that with balanced protein calories supplementation, length of gestation was increased, the proportion of low birth weight infant reduced and mean birth weight raised by 41 g (not statistically significant). With high protein supplementation, there was excess of very early premature births associated with neonatal death, and there was significance with retardation up to 37 weeks of gestation. The significant effect of the high protein supplement on maternal weight gain was conditional enrollment for prenatal care 15 weeks of gestation. There were significantly fewer preterm deliveries in the complement group and this was reflected by increasing in the size of decidual cells, an index associated with placental aging. Several other characteristics of the placentas

of the complement group may have been more directly associated with improved perinatal outcome. There was no evidence of any placental change associated with the increase in preterm delivery and highly significant depressed birth weight among preterm deliveries in the supplementation group. From this study, the researchers noted that three indices of maternal weight gain ( total, average, early ) were significantly related to indices of dietary intake, but only among women recruited before 15 weeks of gestation. Among them the expected gradient in weight gain across treatment groups was found, with the high protein supplement having the maximum effect and indeed responsible for virtually all significant effects. Among premature deliveries recruited early the high protein supplement was associated with high early weight gain and depressed average and total weight gains. It seems likely that the supplement, after accelerating weight gain at the outset among all women recruited weight gain among a group that delivered prematurely.

Riopelle (14,116) who experimented with rhesus monkeys provided some analogies with Rush's results. Riopelle studied in forty-five pregnancies of deprived rhesus monkeys which maintained in semioutdoor environment and fed about 1, 2 or 4 g of protein/kg of body weight each day from the 30<sup>th</sup> day of pregnancy. The liveborn infants born to 1 g and 2 g mothers who were fed with 1 g and 2 g protein/ kg of body weight were, on average, as large as those in the 4 g group. The males appeared to be slightly larger than the females. Moreover, the monkeys fed with the high protein had shorter length of gestation at delivery. Heavy mothers tended to have long gestation. Another research was done by Riopelle (108) in young adult female rhesus monkeys maintained in a seminatural environment. All animals were

initially adapted to the 4 g diet for at least 3 months prior to mating which began 11 days after the onset of menstruation and lasted 4 days. They were maintained on that diet for 1 more month, where upon they were palpated for pregnancy determination. If they concluded that the monkeys were pregnant and then assigned them to one of the three diets. The diet made isocaloric (balanced semisynthetic diet) by addition of carbohydrates to replace the missing casein, supplied 4, 2 or 1 g protein/kg/day if the animals ate 120 kcal /kg/day. They found that weight gain of the highest protein group was greater than the other groups.

The suggestion of a reduced risk of preterm births associated with balanced energy and protein supplementation and premature infants appear to be most vulnerable to high maternal protein supplement. It is not yet clear whether high protein intake deteriorates to the fetus throughout the course of pregnancy or only during a particular trimester. However, low weight gain pregnant women with high protein tend to have short gestational length in previous studies.

### **2.1.2 Gestational weight gain**

Maternal prepregnancy weight and weight gain which indicate nutritional status during pregnancy are associated with preterm delivery. Brown et al (32) found that women who gained as the same amount as normal weight gain had shorter gestational age than normal. Van den Berg and Oecheli (117) reported that an increase weight less than 0.23 kg/week after 20 weeks of pregnancy associated with preterm delivery. Papiernik and Kaminski (49) showed a significant association between preterm and maternal weight gain of less than 5 kg (11 lb) at 32 weeks of gestation.

Abrams et al. (118) studied the relationships between maternal weight gain and preterm delivery in 1,263 pregnant women. Categories of rate of weight gain were defined as follows: (1) low rate of gain = less than 0.27 kg / week; (2) average rate of gain = 0.27 - 0.52 kg / week; and (3) high rate of gain = more than 0.52 kg / week. It was found that 37.3 % of the preterm mothers gained less than 0.27 kg / week, compared with only 24.2 % of the term group. Women with a low rate of gain were more than twice as likely to experience a preterm delivery as those with a high rate of gain, the odd ratio was 2.54 ( 95 % CI 1.49 to 4.8 ). The mean weekly weight gain of preterm mothers (  $0.33 \pm 0.19$  kg / week ) was significantly lower than that of full-term mothers (  $0.39 \pm 0.17$  kg/week ). This difference in weight gain appeared after 20 weeks of gestation. The similar findings were found by Jongsongsrern (12) who studied in Thai women. She found that the mean weekly weight gain in mothers of preterm was  $0.29 \pm 0.08$  kg and that of full-term mothers was  $0.33 \pm 0.09$  kg. There was a statistically significant difference. For the average weight gain in each trimester of pregnancy, the significant difference was found only during the second trimester. Additional study supporting these data were done by Sririkulchayanonta (119).

Siege-riz et al. (120) found that women who were underweight before pregnancy ( BMI < 19.8) had the greater risk of delivering preterm. Inadequate weight gain during the third trimester as opposed to excessive gain (high weight gain as compared to prepregnancy weight status) was predictive of preterm birth. Another study of Siega-riz (121) found that women who delivered preterm had pattern of weight gain similar to women delivering term infant. Inadequate weight gain

in the third trimester defined as  $<0.34$ ,  $0.35$ ,  $0.30$  and  $>0.30$  kg/wk for underweight, normal weight, overweight and obese women respectively, increased the risk by a similar magnitude. In addition, numerous studies showed that the pregnant women with low weight gain during the second trimester increased the risk of preterm delivery.

### 2.1.3 Energy expenditure during pregnancy

Henriksen et al. (122) studied in 4,259 pregnant women who worked at the 16<sup>th</sup> week of gestation and found that women who stood more than five hours per week day had an odd ratio (OR) for preterm delivery of 1.2 ( 95% CI 0.6 to 2.4 ) compared to women who stood two hours or less. For walking the OR for preterm delivery was 1.4 (95 % CI 0.7 to 2.5 ). The conclusion of this study suggested that standing and walking at work during the second trimester may present a particular risk for preterm delivery. Teilelman et al. (87) studied in 1,206 women in Connecticut and found that the rate of preterm was higher among with jobs requiring prolong duration (7.7%) compared with those sedentary (4.2%) or active jobs (2.8%).

Mamelle (123) studied in 3,437 women in France to determine the elements of fatigue in occupations which constituted possible risk factors for the course of a pregnancy particularly those would cause premature birth and found that the mothers who continued to work after mid-pregnancy had gestation no shorter than the mothers who remained at home, even if they had stand-up jobs. Pregnant women who worked hard after mid-pregnancy tended to deliver premature infants.

#### **2.1.4 Hemoglobin level**

Anemia during pregnancy is one of the risk factors for premature birth. Hudono et al. (124) found that mothers who had hemoglobin below 10 gm % tend to have low birth weight infants. However, Funderburk (125) noted that the pregnant women who had haemoglobin below 7 gm % was at high risk to deliver preterm infants. Scholl et al. (126) found that there were >2.5 fold increase of preterm delivery in pregnant women with iron deficiency.

### **2.2 Health practice factors**

#### **2.2.1 Antenatal care**

First antenatal care visit and number of antenatal care have an impact on infant gestational age. McCarthy et al. (127) found that the absence of prenatal care during pregnancy increased the risk of preterm delivery, infants born after no prenatal care had six times higher neonatal mortality rate than those born to mothers who had prenatal care. Ketterlinus et al. (128) noted that gestational age at first attendance for antenatal care was significantly associated with occurrence of preterm delivery.

#### **2.2.2 Maternal smoking**

Smoking during pregnancy is one of the risk factors for premature labor. Harriot et al. (129) found that smoking was most common among lower social class. The wives of semi-skilled and unskilled manual workers tended to continue smoking during pregnancy. The habit was least common in the wives of professional

and managerial group who frequently reduced their smoking during pregnancy. In the babies of mothers who smoked, not only the mean birth weight was lower but also prematurity rate was higher than those of babies of mothers who did not smoke. Smokers had a slightly shorter gestation period.

Wen et al. (130) studied the relationships between smoking and their combined effect on birth weight. It was found that smoking in older women was also associated with more cease of preterm delivery and a lower mean gestational age when compared to women of 25 years old or younger.

### **2.2.3 Maternal alcohol consumption**

Maternal alcohol consumption was found to be associated with shorter gestation in pregnant women. Adams (131) showed that alcohol drinkers were 2.3 times (95 % CI = 1.1 - 4.8) more likely to deliver during 33-36 weeks compared with nondrinkers.

### **2.2.4 Maternal caffeine consumption**

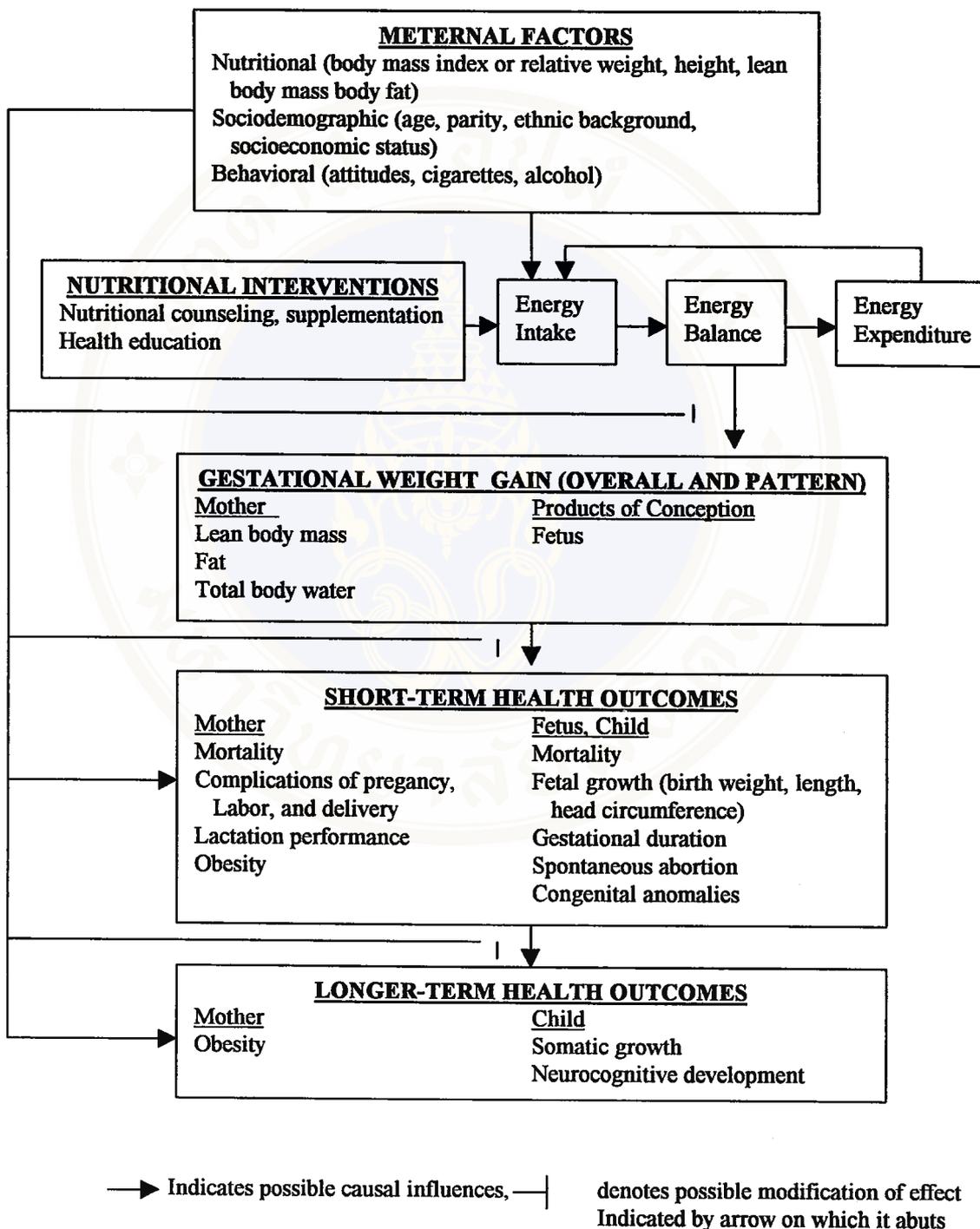
Pastore et al. (132) studied in women consisting of 408 cases and 490 controls. They found that both the first and second trimesters caffeine consumption of 1-150 mg/day were associated with a modestly increased risk of preterm delivery, while no association was found at higher consumption levels and Fortier et al. (133) had a similar finding.

Berkowitz et al. (134) who designed a case-control retrospective study reported that pregnancy outcomes who consumed caffeine beverage during the third trimester and increased quantity of caffeine up to 3 cups had highest risk of preterm delivery.

But the pregnant women who consumed more than 3 cups risk for preterm delivery reduced but McDonald (134) found that the highest risk for preterm delivery if they consumed up to 10 cups of caffeine beverage. Hinds et al. (103) and Fenster (136) reported that gestational age was not related to caffeine consumption. The relationships between maternal caffeine consumption and preterm delivery remained controversial.

In addition to the important factors mentioned above, the other factors such as previous abortion or preterm delivery, obstetric complication during pregnancy, fetal abnormality of placenta (137,138) may affect preterm delivery.

**Theoretical Frame work**



**Figure 1** Determinants, consequences, and effect modifier for gestational weight gain and outcomes.  
 Source: Institute of Medicine(16). Nutrition during pregnancy. Washington D.C: National Acedemy Press;1990:33.

The schematic summary of the potential determinants, consequences and effect modifiers for gestational weight gain ( Figure 1) in the report of the Committee on Nutritional Status during Pregnancy and Lactation, Institute of Medicine (16) was used as the conceptual framework for this study.

In this scheme maternal factors including nutritional, sociodemographic and behavioral were direct determinants of energy intake and indirect determinants of gestational weight gain. Nutritional interventions such as nutrition counseling supplementation and health education did not directly influence maternal weight gain; nor did changing maternal attitudes have a direct impact on gestational weight gain. Gestational weight gain was a consequence of energy intake during pregnancy and a determinant of maternal and fetal outcomes.

In this model, the term “consequence” implies causality. Any association between gestational weight gain and subsequent maternal and child health outcomes are the most important to the extent that gestational weight gain is a cause of those outcomes.

This model is relevant for the present study because it has a variety of factors influence gestational weight gain and consequently affect fetal outcomes.

As illustrated in Figure 2. the model “ Factors determining fetal outcomes from the literature review ” was modified by separating protein intake from energy intake because protein intake during pregnancy had shown to affect fetal outcomes and protein intake could be served as source of energy intake when energy intake was inadequate (21). In addition, the model was modified by adding obstetric factors such as parity and birth interval, and antenatal care to the health practice factor

because those variables could have a beneficial impact on fetal growth or gestational duration. Maternal smoking, alcohol consumption, caffeine consumption, complication during pregnancy such as hypertension, diabetes mellitus, heart disease, etc., and hemoglobin level were variables to include in this model because these maternal behavior, complications and anemia during pregnancy could be harmful to the fetus (46).

In the study framework ( Figure 3), the variables which affected uterine ischemia were controlled so these variables were not illustrated in this framework and the other variables such as maternal stature, sociodemographic, antenatal care, obstetric factors and energy expenditure were controlled by selecting the participants in the same characteristics .

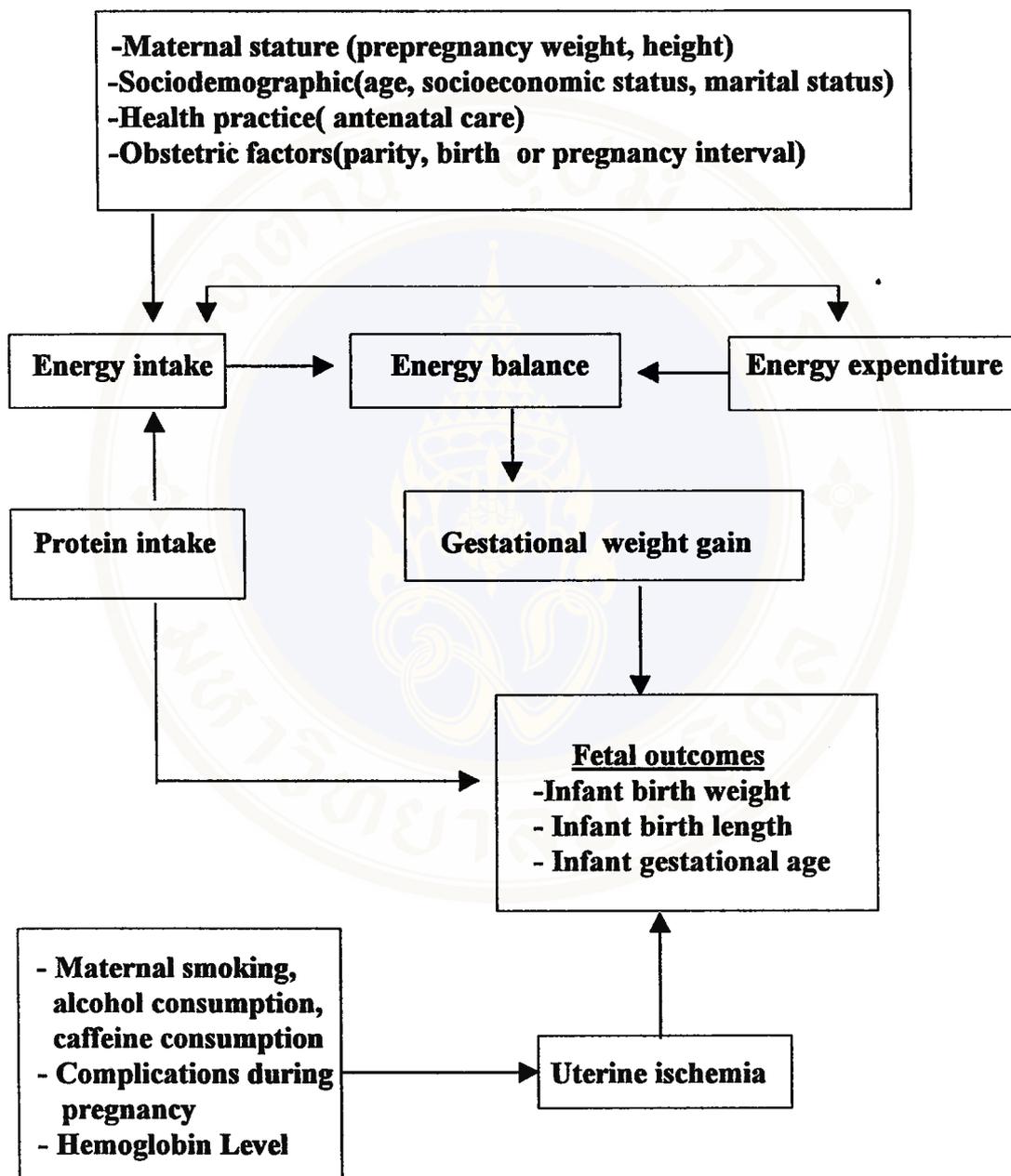
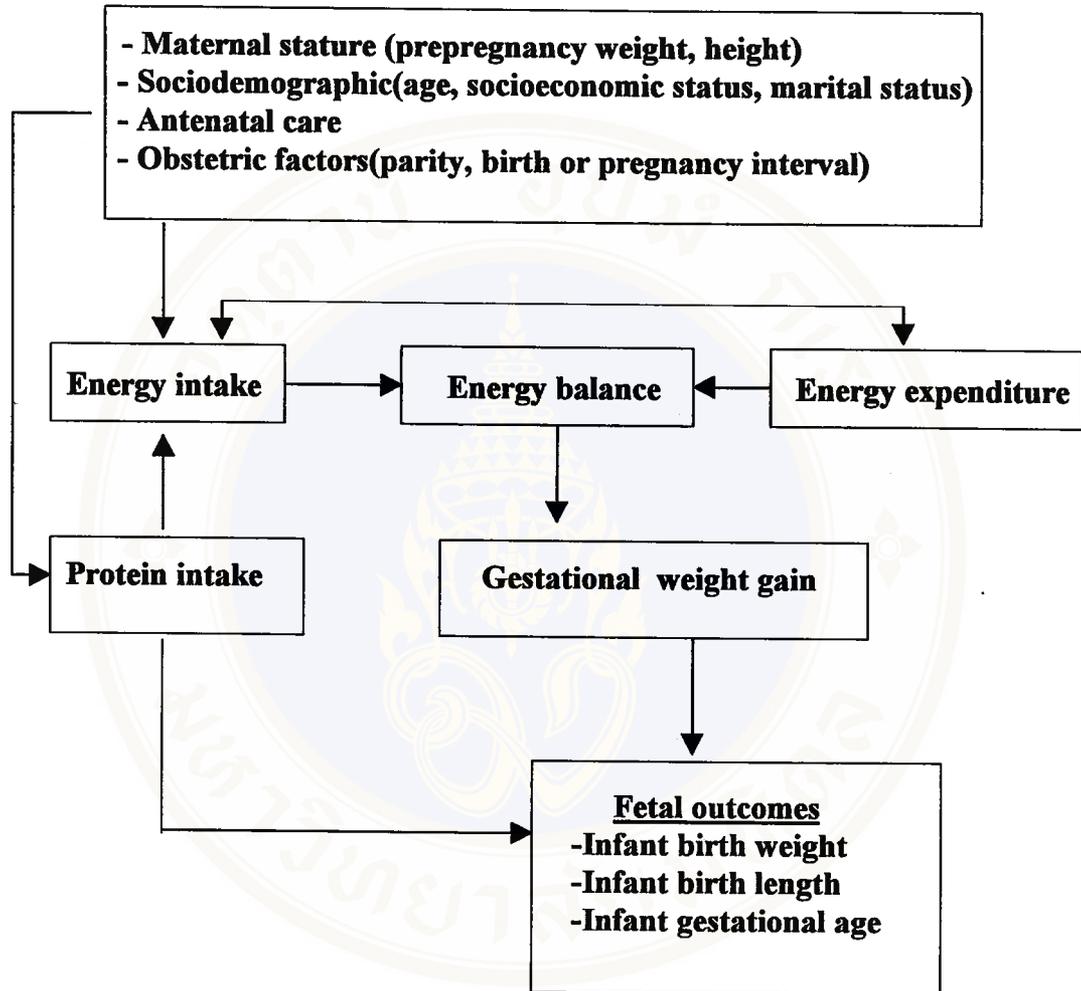


Figure 2 Factors determining fetal outcomes from the literature review



**Figure 3** Factors determining fetal outcomes in the study

## CHAPTER III

### MATERIALS AND METHODS

#### STUDY DESIGN

This study determined the effect of protein intake of pregnant women with inadequate weight gain on infant birth weight, birth length and gestational age during the second and third trimesters. The protein intake of the participants was measured at the 20<sup>th</sup> - 23<sup>rd</sup> weeks of gestation, follow up until delivery to determine the effect of protein intake on the fetal growth and gestational age. The research design was a prospective cohort study from March, 1998 to July, 1999. The dietary intake interviews were done twice during the second trimester and twice during the third trimester as follows:

#### The second trimester

1<sup>st</sup> interview the 20<sup>th</sup> - 23<sup>rd</sup> weeks of gestation

2<sup>nd</sup> interview the 24<sup>th</sup> - 27<sup>th</sup> weeks of gestation

#### The third trimester

3<sup>rd</sup> interview the 28<sup>th</sup> - 31<sup>st</sup> weeks of gestation

4<sup>th</sup> interview the 32<sup>nd</sup> - 35<sup>th</sup> weeks of gestation

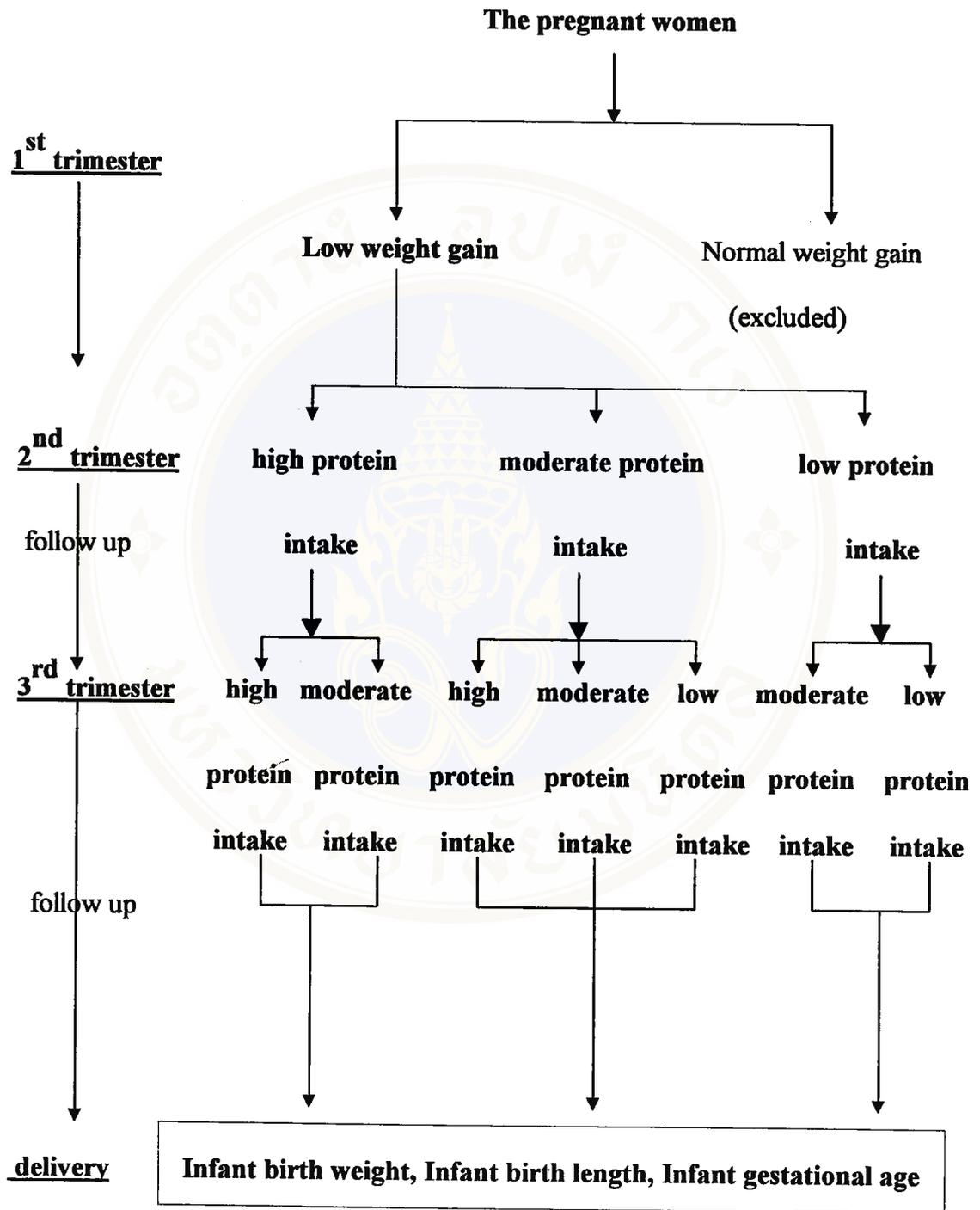


Figure 4 Study design in this study

## THE STUDIED PARTICIPATIONS

The participants of this study were pregnant women who were the 20<sup>th</sup> week to 23<sup>rd</sup> week of gestation and attended antenatal care clinic regularly ( at least 5 times from the 20<sup>th</sup> week of gestation until delivery) at Maternal and Child Hospital, Khon Kaen Regional Hospital and Srinakarin Hospital, Khon Kaen University. The recruitment of participants was based on the following criteria:

1. Low gestational weight gain at the time of recruitment
2. 20 - 34 years of age
3. Not less than 148 cm of height
4. Not less than 45 kg of prepregnancy weight
5. No complication during pregnancy
6. No history of chronic illness
7. Not less than 11 gm % of hemoglobin or not less than 33 % of hematocrit during pregnancy
8. Not alcohol, or caffeine beverage drinkers or cigarette smokers
9. Singleton pregnancy
10. No abnormality of either the placenta or the new born
11. Low and lower middle socioeconomic status
12. Moderate activity during pregnancy
13. Less than 6 months of birth interval
14. Pregnant women who were married and/or lived with husbands
15. Only the second and third parity

**SAMPLE SIZE (139,140)**

Sample size calculation from sample size in the analysis of variance (Table 1, Appendix A)

$\alpha$  = the chance of asserting that a difference exists when the true difference is zero (type 1 error).

$\beta$  = the chance of asserting that a difference exists when the true difference is non-zero (type 2 error).

$\Delta$  = true difference in mean clearance for two different speeds.

$$= \max(\mu_i) - \min(\mu_i)$$

$\sigma$  = standard deviation of the probability distribution of dependence variable.

For this design:

$$\alpha = .01$$

$$\beta = .05 \text{ or power} = 1 - \beta = .95$$

In this study maternal protein intake levels were divided into 3 groups (high, moderate, low)

Entering table 1 for  $\frac{\Delta}{\sigma} = 1.0$ , no of level = 3,  $\alpha .01$

$$n = 43 * 3$$

$$\text{sample size} = 129$$

$$\text{participants in this study} = 129 \quad \text{cases}$$

## **Sampling Scheme**

Interviewing of pregnant women and using hospital records were integrated to select subjects by inclusion criteria. The willingness of pregnant women to participate in this study was the main concern. Subjects were interviewed and followed up until their deliveries. More than 250 samples were collected. After a certain period of time, there was a drop-out of some pregnant women that might be attributable to following factors:

1. Did not visit their antenatal care clinic in that particular hospital. Some changed their antenatal care visit to a nearby hospital or health center;
2. Did not regularly visit the antenatal care clinic of which contributed to an incomplete dietary intake data;
3. Delivered at other hospitals;
4. Complications during the follow up time;
5. Abortions;
6. Migrated to other places.

Therefore only 170 cases completed the data

## **INSTRUMENTS**

1. Questionnaire Forms:

1.1 Demographic and health-related information (Form A) were used to screen participants of the study. This information included the detail of parity, present gestational age, height, prepregnancy weight, health status, health behavior,

socioeconomic status, energy expenditure level, obstetrical history and delivery data ( Appendix B ).

1.2 Food frequency form (Form B) was used to record general dietary pattern during the second and third trimesters (Appendix B ).

1.3 Twenty-four hours dietary recall form (Form C) was used to collect the dietary intake twice during the second trimester and twice during the third trimester and gestational weight gain was recorded on the day of 24-hour dietary recall interview. Dietary weight scales, standard measuring cups and spoons, food models, fresh and cooked foods, and two-dimensional food models were used to help the participants to estimate the portion size and amount of foods consumed (Appendix B ).

1.4 Food records form (Form D) was used to collect the dietary intake twice during the second trimester and twice during the third trimester. The pregnant women recorded these data.

1.5 Fetal outcomes information form (Form E) was used to record fetal outcomes including the details of gestational age at delivery, infant birth weight, infant birth length, and infant sex (Appendix B ).

2. Weighing scale “Detecto” of 0.1 kilogram interval was used to measure the weight of pregnant women. They should be weight with the minimum of clothing and without shoes.

3. Height measuring scale of 0.1 centimeter interval was used to measure the height of pregnant women. They should stand straight and look straight ahead with shoulder blades, buttocks and heel touching measurement board. Arms should be hanging loosely at the sides with palms facing the thighs.

## DATA COLLECTION

The study was conducted with the permission from Maternal and Child Hospital, Khon Kaen Regional Hospital and Srinakrin Hospital to collect the data from the pregnant women who were the 20<sup>th</sup> - 23<sup>rd</sup> weeks of gestation and attended the antenatal care clinic regularly until delivery as follows:

1. The out patient records and the screening form were used to select the participants who were recruited in the study before the 20<sup>th</sup> week of gestation.

2. The interview for dietary intake was conducted by using food frequency, 24-hour dietary recalls and food records method .

- 2.1 Food frequency was conducted to investigate the dietary pattern of pregnant women during the second and third trimesters.

- 2.2 24-hour dietary recall was conducted when pregnant women visited antenatal care clinic. The 24-hour dietary interview was done twice during the second trimester and twice during the third trimester. The pregnant women were interviewed the first food or drink consumed in the morning of previous day and proceeded systematically throughout the day including snacks and beverage.

- 2.3. Food record forms were distributed to the pregnant women when they visited the antenatal care clinic. They recorded their own food intake 1-2 days at home similar to the 24- hour dietary recall and returned the forms in the next ANC visit. The record had to be interviewed again for checking the kind and quantity of food.

3. Data concerning the health of pregnant women and weight gain were recorded from the first visit at ANC until delivery.

4. Data concerning the infant birth weight, birth length and gestational age at birth was collected from the data of the hospital record upon the pregnant women delivery at the hospital.

## **DATA ANALYSIS**

Nutritionist III program (INMUCAL) was used to analyze 24-hour dietary recall. Data from the 24-hour dietary recall were converted into raw weight. All food items were coded before being entered into computer. The software, Nutritionist III, was a data base for Thai foods which were used to analyze the composition of the diet. The 24-hour dietary recall and food record were collected twice during the second trimester and twice during the third trimester. The dietary intake during each trimester was averaged.

Variables for analysis as follows:

### **1. Independent variables**

#### **1.1 Quantity of energy intake (kilocalories )**

The average kilocalories of energy intake in each trimester was used for analysis.

#### **1.2. Quantity of protein intake (gram and percentage)**

This variable was measured as gram of protein intake and percentage of energy intake. The protein intake in grams unit in each trimester was averaged to determine the acid loading. The protein intake was averaged into percentage of total energy intake in each trimester to determine the function of protein.



### 1.3 Quality of protein intake

The protein intake was averaged and calculated into animal and vegetable protein intake ratio ( A:V ratio) in each trimester to determine the quality of protein intake.

### 1.4 Quantity of nutrients intake

The essential nutrients intake that related to fetal outcomes such as calcium and phosphorus were also analyzed

### 1.5 Gestational weight gain (kilogram)

The total weight gain and average weight gain per week of pregnancy in each trimester was used for analysis.

1.6 Some maternal obstetric variables such as prepregnancy weight, height, prepregnancy body mass index (BMI) and maternal age were used.

1.7 Socioeconomic variables such as maternal education, occupation and family income which determined the effect on dependent variables and protein intake were used.

## 2. Dependent variables

2.1 infant birth weight (gram)

2.2 infant birth length (centimeter)

2.3 infant gestational age (week)

## STATISTICAL ANALYSIS

The data were processed and analyzed as follows:

1. Descriptive statistics such as percentage (%), mean ( $\bar{x}$ ), standard deviation (S.D) and range were used to describe background characteristics of participants, the food patterns from food frequency of pregnant women in the second and third trimesters, the nutrient intake and fetal outcomes of pregnant women with different quantity and quality of protein intake.

2. t-test was used to compare the means of infants birth weight, birth length and gestational age between two groups of different protein levels.

### 3. Analysis of variance approach

One way ANOVA was used to compare the means of infants birth weight, birth length and gestational age among various obstetric factors and socioeconomic characteristics groups.

### 4. Multivariate analysis of variance approach

4.1 The Manova was used to compare the means of all three dependent variables: infants birth weight, birth length and gestational age among three protein intake level groups (low, moderate, high). The participants might keep in the same protein intake level group or shifted from one protein intake level group in the second trimester to another in the third trimester.

4.2 Analysis of covariance was used to determine the effect of different protein intake on birth weight, birth length and gestational age with energy intake as a covariate.

### 5. Regression analysis approach

Multiple regression was used to explain the relationships between all dependent variables. Each model was including interesting factors and one dependent variable. The dependent variables in the study were birth weight, birth length and gestational age. Potential factors from research hypotheses were acting as independent variables in regression. Statistical significant level was considered at 0.05. The independent variables were incorporated in the equation by using entering method. The variables were added to the regression equation according to time order.

## CHAPTER IV

### RESULTS

Of the 16 months' data collections, 4 consecutive rounds of the 24-hour dietary recall and food record were commenced in the 20<sup>th</sup> week gestational age women. Weight gain of pregnant women until delivery was also included in this study. There were 170 pregnant women who met the criteria from 3 different hospitals: Maternal and Child Hospital, Khon Kaen Regional Hospital, and Srinakarin Hospital, Faculty of Medicine, Khon Kaen University. The results of this study were divided into the following:

1. General description of the subjects;
2. Maternal obstetric factors and infant birth weight, birth length and gestational age;
3. Maternal socioeconomic characteristics and infant birth weight, birth length and gestational age including protein intake;
4. General nutrient intake, food behavior and food frequencies;
5. Protein intake in different groups during the second and third trimesters and infant birth weight, birth length and gestational age of the mothers;
6. Relationships between nutrient intake, weight gain and infant birth weight, birth length and gestational age of the mothers;

## **1. General description of subjects**

Among the 170 pregnant women, 42.4 %, 24.1 % and 33.5 % delivered their child at Maternal and Child Hospital, Khon Kaen Regional Hospital and Srinakarin Hospital, Khon Kaen University respectively (see Table 1).

### **Education of the subjects**

The educational level of the pregnant women was divided into primary education (Prathomsuksa), secondary education (Mathayomsuksa-MS including MS 3 and MS 6) and higher than secondary education (higher than MS 6). The majority of subjects completed their primary education (63.5 %). The highest education of the subjects was 27.6 %, 7.1 % and 1.8 % for MS 3, MS 6 and higher than MS 6 respectively.

### **Age of the subjects**

The 20-34 year-old mothers were selected for this study. Forty percent of them aged between 20-25, while 44.1 % were 26-30 years old and 15.9 % aged between 31-34.

### **Child parity**

Only the second and third parity pregnancies were included in the present study. The majority of them were in their second parity (82.8 %).

### **Occupation of subjects**

Approximately half of the pregnant women were housewives (42.9 %). There were 25.9 %, 19.4 % and 9.4 % of pregnant women who were farmers, laborers and traders respectively. Only 2.4 % were officers, dressmakers and baby-sitters.

### **Family income**

Most of the family income was 2501-5000 baht per month (46.5%). There were 29.4% of pregnant women whose family income was more than 7500 baht per month.

**Table 1** General description of subjects ( n =170)

<b>Variables</b>	<b>n</b>	<b>%</b>
<b><u>Place</u></b>		
Maternal and Child Hospital	72	42.4
Khon Kaen Regional Hospital	57	33.5
Srinakarin Hospital	41	24.1
<b><u>Maternal education</u></b>		
Prathomsuksa	108	63.5
Mathayomsuksa 3	47	27.6
Mathayomsuksa 6	12	7.1
Higher than Mathayomsuksa 6	3	1.8
<b><u>Age of pregnant women (years)</u></b>		
20-25	68	40.0
26-30	75	44.1
31-34	27	15.9
<b><u>Parity of pregnant women</u></b>		
Second	151	82.8
Third	19	11.2

**Table 1** General description of subjects (continued)

Variables	n	%
<b><u>Occupation of pregnancy</u></b>		
Farmer	44	25.9
Laborer	33	19.4
Trader	16	9.4
Housewife	73	42.9
Others	4	2.4
<b><u>Family income(baht per month)</u></b>		
<2500	17	10.0
2501-5000	79	46.5
5001-7500	24	14.1
>7500	50	29.4

## **2. Maternity obstetric factors and infant birth weight, birth length and gestational age of the mothers**

The majority of subjects were 26-30 years old (44.1 %). Sixty seven percent of the subjects weighed between 45 to 55 kg and 51.8 % of them were 148-155 cm in height. Most of them (79.4 %) had their prepregnancy body mass index (BMI) between 18.5 to 24.9 (see Table 2).

### **Age of mothers**

The mean birth weight, birth length and gestational age of the mothers were analyzed using one way analysis of variance. The results, however, indicated no significant difference ( $p = 0.863, 0.454$  and  $0.996$ ).

### **Prepregnancy weights**

Prepregnancy weights were categorized into 3 groups: 45-55 kg, 56-65 kg and 66-80 kg. There were significant differences of the birth weight and the birth length among the 3 groups ( $p = 0.019$  and  $0.018$ ) but not of the gestational age. The 45-55 kg pregnancy weight group tended to have lower birth weight and birth length than other groups.

### **Height of mothers**

Height of the pregnant women was divided into 2 categories: 148-155 cm, and 156-170 cm. There was no significant difference among both groups ( $p = 0.704$ ,  $0.375$  and  $0.889$ ).

### **Prepregnancy body mass index (BMI) of pregnant women**

Body mass index (BMI) was categorized into 3 groups following the criteria of WHO including BMI  $< 18.5 \text{ kg/m}^2$ ,  $18.5\text{-}24.9 \text{ kg/m}^2$  and  $\geq 25 \text{ kg/m}^2$ . No significant differences were found among the 3 groups ( $p = 0.091$ ,  $0.204$  and  $0.583$ ).

### **Total weight gain of pregnant women**

Total weight gain of normal pregnant women was 10-12 kg so that total weight were categorized into 3 groups:  $<10 \text{ kg}$ ,  $10\text{-}12 \text{ kg}$  and  $>12 \text{ kg}$ . Approximately 35.2%, 32.9%, 31.7% of pregnant women had total weight gain  $<10 \text{ kg}$ ,  $10\text{-}12 \text{ kg}$  and  $>12 \text{ kg}$  respectively. There were significant differences of birth weight among 3 groups ( $p = 0.001$ ) but not of birth length and gestational age. The higher total weight gain, the higher birth weight was.

**Table 2 Mean infant birth weight, birth length and gestational age by maternal**

obstetric factors				
Maternal		birth weight(g)	birth length(cm)	gestational age(day)
obstetric	%(n)	Mean±SD	Mean±SD	Mean±SD
factors		(Min, Max)	(Min, Max)	(Min, Max)
<b><u>Maternal age groups (years)</u></b>				
20-25	40.0(68)	3129.56±427.39 (2200, 4150)	49.94±2.47 (45, 55)	272.41±9.46 (252, 304)
26-30	44.1(75)	3154.27±428.56 (2000, 4720)	49.71±2.29 (43, 55)	272.44±10.84 (245, 301)
31-34	15.9(27)	3106.48±353.81 (2410, 3980)	50.37±2.71 (46, 55)	277.22±12.71 (230, 295)
p – value		0.863	0.454	0.996
<b><u>Maternal prepregnancy weight (kg)</u></b>				
45-55	67.6(115)	3080.09±366.25 (2000, 4160)	49.56±2.29 (43, 54)	271.76±10.32 (245, 304)
56-65	25.3(43)	3287.21±360.56 (2650, 4720)	50.70±2.23 (46, 55)	274.40±11.35 (230, 301)
66-80	7.1(12)	3141.25±571.59 (2200, 4000)	50.42±2.87 (46, 55)	271.33±10.78 (259, 291)
p – value		0.019*	0.018*	0.359
<b><u>Maternal height (cm)</u></b>				
148-155	51.8(88)	3125.06±456.27 (2000, 4720)	49.75±2.57 (43, 55)	272.28±10.20 (245, 301)
156-170	48.2(82)	3149.39±368.49 (2000, 4150)	50.07±2.13 (46, 55)	272.51±11.11 (230, 304)
p – value		0.704	0.375	0.889

**Table 2** Mean infant birth weight, birth length and gestational age by maternal obstetric factors (continued)

Maternal obstetric factors	% (n)	birth weight(g)	birth length(cm)	gestational age(day)
		Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
<b>Maternal prepregnancy BMI (kg/m<sup>2</sup>)</b>				
<18.5	4.1(2)	2964.29±180.17 (2680, 3200)	49.00±1.73 (46, 51)	269.86±9.32 (259, 284)
18.5-24.9	79.4(135)	3116.37±391.76 (2000, 4720)	49.82±2.36 (43, 55)	272.19±10.94 (230, 304)
≥25	16.5(28)	3278.39±528.78 (2200, 4150)	50.54±2.47 (46, 55)	274.19±9.34 (259, 295)
p – value		0.091	0.204	0.583
<b>Total weight gain (kg)</b>				
<10	35.2(60)	3023.75±375.53 (2000, 4150)	49.55±2.31 (43, 55)	271.67±12.52 (230, 304)
10-12	32.9(56)	3101.79±389.12 (2200, 3980)	49.68±2.37 (45, 54)	271.93±9.03 (253, 296)
>12	31.7(54)	3298.70±439.48 (2430, 4720)	50.54±2.33 (46, 55)	273.69±9.89 (245, 301)
p – value		0.001*	0.056	0.555

\*p-value < 0.05

In addition to obstetric factors, sex of infant was also compared. Approximately 47.1% and 52.9% of infants were male and female respectively. The average infant birth weight, birth length and gestational ages were not significantly different between male and female babies ( $p = 0.838, 0.256$  and  $0.234$ ) (see Table 3).

**Table 3** Mean infant birth weight, birth length and gestational age by sex

Infant sex	%(n)	Birth weight(g)	Birth length(cm)	Gestational age(day)
		Mean±SD	Mean±SD	Mean±SD
		(Min, Max)	(Min, Max)	(Min, Max)
Male	47.1(80)	3143.75±444.94 (2000, 4720)	50.13±2.34 (43, 55)	273.43±11.32 (230, 304)
Female	52.9(90)	3130.61±389.30 (2200, 4110)	49.71±2.39 (45, 55)	271.48±9.92 (245, 295)
p – value		0.838	0.256	0.234

### 3.Socioeconomic characteristics

#### 3.1 Socioeconomic characteristics versus infant birth weight, birth length and gestational age

The socioeconomic characteristics of the mothers were classified according to their educational levels, occupations and family income (see Table 4).

##### Educational levels

The education of pregnant mothers was categorized into 5 groups: Prathomsuksa 4 and 6, Mutthayomsuksa 3 (MS 3), Mutthayomsuksa 6(MS 6), and higher than Mutthayomsuksa 6(> MS 6). There was no significant difference imposing on birth weight, birth length and gestational ages among the different educational groups ( $p=0.520, 0.445$  and  $0.411$ ). In spite of the fact that majority of them completed their primary education.

### Occupations

The common occupations in this study were farmers, laborers, traders, housewives and others. The majority of pregnant mothers were housewives (42.9 %). Mean infant birth weight, birth length and gestational age were not significantly ( $p = 0.729, 0.848$  and  $0.794$ ) differed among the various occupational groups.

### Family income

The monthly family income of the subjects was divided into 4 groups: income  $\leq 2,500$  baht, 2,501-5,000 baht, 5,001-7,500 baht and  $> 7,500$  baht respectively. Most infants (46.4 %) were born from mothers whose family income was 2,501-5,000 baht/month. Considering infant mean birth weight, birth length and gestational age, there was no significant difference among the three groups ( $p = 0.142, 0.903$  and  $0.338$ ).

**Table 4** Mean infant birth weight, birth length and gestational age by socioeconomic characteristics

Socioeconomic characteristics		birth weight(g)	birth length(cm)	gestational age(day)
	%(n)	Mean $\pm$ SD (Min, Max)	Mean $\pm$ SD (Min, Max)	Mean $\pm$ SD (Min, Max)

#### Maternal educational levels

Prathomsuksa	63.5(108)	3148.75 $\pm$ 431.55 (2000, 4720)	50.11 $\pm$ 2.45 (43, 55)	272.64 $\pm$ 10.46 (245, 304)
MS. 3	27.6(47)	3157.02 $\pm$ 413.17 (2250, 4110)	49.66 $\pm$ 2.19 (45,55)	273.04 $\pm$ 9.94 (252,294)

**Table 4** Mean infant birth weight, birth length and gestational age by socioeconomic characteristics (continued)

Socioeconomic characteristics	%(n)	birth weight(g)	birth length(cm)	gestational age(day)
		Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
MS. 6	7.1(12)	2967.50±294.10 (2570, 3620)	49.25±2.34 (45, 53)	267.42±14.97 (230, 280)
>MS.6	1.8(3)	3066.67±75.72 (2980, 3120)	49.00±1.73 (47, 50)	273.29±10.60 (273, 274)
p – value		0.520	0.445	0.411
<b><u>Maternal occupations</u></b>				
Farmer	25.9(44)	3113.18±508.94 (2000, 4720)	49.98±2.70 (43, 55)	272.50±10.52 (255, 304)
Laborer	19.4(33)	3080.61±394.88 (2250, 4100)	49.70±1.94 (46, 54)	271.18±11.00 (230, 295)
Trader	9.4(16)	3121.56±465.73 (2250, 4160)	50.00±2.53 (45, 54)	270.19±10.06 (258, 291)
Housewife	42.9(73)	3185.75±360.69 (2400, 4110)	50.00±2.30 (45, 55)	273.30±10.93 (245, 296)
Others	2.4(4)	3027.50±103.72 (2950, 3180)	48.75±2.87 (47, 53)	273.50±5.80 (266, 280)
p – value		0.729	0.848	0.794
<b><u>Family income</u></b>				
≤2500	10.0(17)	3064.71±518.73 (2200, 4720)	49.88±2.23 (46, 54)	269.47±12.19 (250, 301)

**Table 4** Mean infants birth weight, birth length and gestational age by socioeconomic characteristics (continued)

Socioeconomic characteristics	%(n)	birth weight(g)	birth length(cm)	gestational age(day)
		Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
2501-5000	46.5(79)	3188.35±421.09 (2000, 4100)	50.01±2.39 (43, 55)	273.59±10.37 (252, 304)
5001-7500	14.1(24)	3220.42±399.66 (2650, 4150)	50.00±2.25 (45, 55)	273.42± 8.83 (252, 288)
>7500	29.4(50)	3039.70±361.08 (2410, 4160)	49.70±2.47 (45, 55)	271.00±11.16 (230, 291)
p – value		0.142	0.903	0.336

### 3.2 Socioeconomic characteristic and protein intake

In comparison of protein intake with socioeconomic characteristics, the results indicated no significant difference for all groups of education, occupation and family income (see Table 5). Protein intake increased in the third trimester of pregnancy in all groups. Exception was made to occupation and family income. Furthermore, the source of protein intake was also analyzed (see Table 6 and 7).

#### Maternal educational levels

Subjects who completed Prathomsuksa 4 and 6 consumed less animal protein. They rather consumed high vegetable protein. Subjects who completed their education higher than MS 6 tended to have higher intake of animal protein and lower intake of vegetable protein than other groups. Mean intake of animal and vegetable protein in

various educational levels during the second and third trimesters was not statistically different ( $p > 0.05$ ).

### **Maternal occupations**

Farmers consumed less animal protein than other groups. On the contrary, they preferred vegetable protein than other groups during both trimesters of their pregnancy. Various occupation groups were compared; there was no significant difference of animal protein intake in both trimesters ( $p = 0.086$  and  $0.054$ ) and vegetable protein in the third trimester ( $p = 0.219$ ). There was significant difference for vegetable protein intake during the second trimester ( $p = 0.043$ ).

Employing a multiple comparison test, LSD (Least Significant Differential Test), the results indicated that farmers were statistically different from other groups.

### **Family income**

The group with income more than 7,500 baht/month consumed lower animal protein during their second trimester. However, they increased their animal protein consumption and were the group that consumed highest animal protein during the third trimester. Considering vegetable protein intake, there was a similar amount of consumption among various family income groups. Comparing mean intake of animal protein and vegetable protein as a whole, there was marginal significant difference only for animal protein in the third trimester ( $p = 0.046$ ). Multiple comparison (LSD test) was also analyzed for family income in the third trimester; the results indicated that the lowest income group significantly differed from others.

**Table 5** Mean total protein intake by socioeconomic characteristics

Socioeconomic characteristics	Total protein intake(g)			
	The second trimester		The third trimester	
	Mean±SD	P	Mean±SD	P
<b><u>Maternal educational levels</u></b>				
Prathomsuksa	72.25±19.28		75.99±20.29	
MS. 3	73.95±20.54	0.966	74.59±19.29	0.871
MS. 6	72.18±18.77		80.28±31.59	
Higher than MS.6	71.34±19.42		75.65±22.05	
<b><u>Maternal occupations</u></b>				
Farmer	71.93±24.95		73.43±19.71	
Laborer	73.06±16.28		75.10±17.10	
Trader	76.21±17.10	0.818	69.15±17.99	0.390
Housewife	71.76±18.0		78.97± 23.61	
Others	81.38±8.29		80.79±14.12	
<b><u>Family income</u></b>				
≤2500	75.45±30.04		65.06±20.57	
2501-5000	71.46±18.55	0.775	75.30±16.83	0.084
5001-7500	75.38±7.74		77.24 ±23.98	
>7500	72.70±19.45		79.90±24.11	

**Table 6** Mean animal protein intake by socioeconomic characteristics

Socioeconomic characteristics	Animal protein intake(g)			
	The second trimester		The third trimester	
	Mean±SD	P	Mean±SD	P
<b><u>Maternal educational levels</u></b>				
Prathomsuksa	42.02±15.14		44.55±17.13	
MS. 3	44.55±14.88	0.796	45.91±14.62	0.956
MS. 6	42.91±15.61		45.82±21.25	
Higher than MS.6	46.15±23.86		47.70±21.33	
<b><u>Maternal occupations</u></b>				
Farmer	38.99±19.30		39.90±14.45	
Laborer	44.97±12.43		45.33±13.47	
Trader	46.94±13.97	0.086	40.72±13.29	0.054
Housewife	42.54±13.67		48.76±19.02	
Others	57.27±14.02		49.81±19.59	
<b><u>Family income</u></b>				
≤2500	44.18±23.91		36.59±16.99	
2501-5000	44.85±14.27	0.825	43.82±12.83	0.046*
5001-7500	44.94±12.57		46.98±16.10	
>7500	42.98±14.89		49.01±20.98	

\*p- value &lt; 0.05

**Table 7** Mean vegetable protein intake by socioeconomic characteristics

Socioeconomic characteristics	Vegetable protein intake(g)			
	The second trimester		The third trimester	
	Mean±SD	P	Mean±SD	P
<b><u>Maternal educational level</u></b>				
Prathomsuksa	30.20±7.98		31.45±8.72	
MS. 3	29.45±9.61	0.758	28.70±9.05	0.152
MS. 6	29.52±7.54		34.54±13.09	
Higher than MS.6	25.30±7.43		27.94±1.09	
<b><u>Maternal occupations</u></b>				
Farmer	32.94±10.17		33.53±9.88	
Laborer	28.07±6.13		29.76±8.02	
Trader	29.22±6.35	0.043*	28.39±9.86	0.219
Housewife	29.26±8.13		30.20±9.02	
Others	24.19±7.88		31.74±7.50	
<b><u>Family income</u></b>				
≤2500	31.27±10.16		28.48±7.52	
2501-5000	29.57±8.30	0.845	31.47±8.34	0.531
5001-7500	30.55±8.44		30.25±10.98	
>7500	29.49±8.02		30.94±10.10	

\*p - value &lt; 0.05

## Conclusion

Comparing means of birth weight, birth length and gestational age with obstetric factors and socioeconomic characteristics, no statistical difference was encountered in all instances. Exception was for the prepregnancy weight ( $p < 0.05$ ). Taken into account the protein intake over socioeconomic characteristics, no significant difference was found. However, animal protein intake during the third trimester, the significant difference existed between animal protein intake over family income during the third trimester and vegetable protein intake over occupation during the second trimester ( $p < 0.05$ ).

#### **4. General nutrient intake, food consumption behavior and food frequency**

##### **4.1 Nutrient intake during the second and the third trimester of pregnancy**

The average nutrient intake was calculated from 24-hour dietary recalls and food records for 4 consecutive rounds in each trimester. Comparing quantity of food intake during weekdays and weekends, results showed no difference in food consumption among most subjects. Energy and nutrient intakes were shown in Table 8.

The average energy intake and other nutrients were slightly higher in the third trimester than the second trimester except for vitamin C (as shown in Table 8).

Comparing the energy and nutrient intakes with the recommended daily dietary allowance (RDA) for healthy Thais (1989), the energy and all other nutrient intakes were lower than that of RDA except for protein and vitamin C. The percentages of protein intake were 142.54 and 148.83 g per day in the second and third trimesters respectively.

Food consumption over 75 % of RDA provided sufficient energy and other nutrients to pregnant women except for iron and calcium which was lower than 75 % RDA. Pregnant women rendered sufficient amount of iron from the routine antenatal clinic. This indicated a sufficiency of iron intake. However, calcium was consumed to a lesser extent than it was required especially for pregnant women in the Northeast of Thailand. Milk consumption is not a normal practice among Thai adults. Moreover, milk is too expensive for daily consumption even the subjects consumed milk more frequently than unusual but the average intake was still low.

**Table 8** Nutrient intake of subjects in the second and third trimesters

Nutrient intake	The second trimester		The third trimester	
	Mean±SD	%RDA	Mean±SD	%RDA
Energy (Kcal)	2002.41±505.01	87.06	2080.99±520.57	90.48
Fat (g)	39.07±19.93	-	39.59±15.79	-
Carbohydrate (g)	341.45± 92.25	-	356.84±103.14	-
Total protein (g)	72.69±19.45	142.54	75.90±20.84	148.83
Animal protein (g)	42.85±19.34	-	45.07±16.71	-
Vegetable protein(g)	29.86±8.39	-	30.84±9.18	-
Total iron (mg)	15.56±5.06	34.57	15.74±4.87	34.04
Animal iron (mg)	5.98±3.03	-	6.12±2.84	-
Vegetable iron (mg)	9.61±3.41	-	9.69±3.26	-
Calcium (mg)	479.05±243.66	39.92	486.18±226.05	40.52
Phosphorus (mg)	897.99±27.13	74.84	911.71±21.21	75.98
Vitamin A (µgRE)	741.57±959.27	92.69	869.50±1113.54	108.69
Vitamin B <sub>1</sub> (mg)	1.08±0.68	76.93	1.17±0.69	83.74
Vitamin B <sub>2</sub> (mg)	1.23±0.49	82.13	1.29±0.51	86.44
Vitamin C (mg)	116.32±89.68	145.40	100.83±75.64	126.03

Proportions of the caloric distributions i.e. protein: carbohydrate: fat during the second and third trimesters were 14.62: 68.29: 17.08 and 14.69: 68.03: 17.25

respectively (see Table 9). Comparing caloric distributions with normal adults, results indicated higher carbohydrate consumption but lower fat consumption among the subjects with the proportion of 15: 55: 30. As for food consumption pattern of the Northeastern villagers, They preferred rice as their staple food and utilized small amount of oil for cooking. In this instance, pregnant women had adequate protein intake; therefore, caloric distribution from protein was also sufficient.

**Table 9** Mean Caloric distribution of the subjects in the second and third trimesters

Source of caloric intake	The second trimester	The third trimester
	Mean±SD	Mean±SD
Protein	14.62±2.09	14.69±2.19
Carbohydrate	68.29±7.36	68.03±6.95
Fat	17.08±6.51	17.25±5.91

Considering protein quality, 2.4 % (n = 4) and 1.8 % (n = 3) of pregnant women consumed poor quality protein (animal protein: vegetable protein <1:2) in the second and third trimesters respectively. The majority of them consumed good quality protein (animal protein: vegetable protein ≥ 1: 2 or more). There were 97.6% (n = 166) and 98.2% (n = 167) who consumed good quality of protein intake.

#### **4.2 Food consumption behavior of pregnant women**

In early pregnancy, 64.7 % (n = 110) of pregnant women usually had nausea and vomiting in the morning which affected their food intake during the first 3 months. Only 2.94 % of subjects continued having nausea and vomiting until 4-5 months of pregnancy when they came for the first interview (4-5 months) and 32.35 % (n = 55) had no symptom of nausea and vomiting during pregnancy.

Most pregnant women had 3 meals a day (95.25 %). Eight pregnant women (4.71 %) had 4 meals a day. However, three of them consumed more in the afternoon while the rest consumed more before bedtime.

Taken into account likes and dislikes of food, the subjects were interviewed for their food preference and the data were presented in Table 10. It showed that pregnant mothers preferred more fruits during pregnancy than their normal practice. Moreover, sour fruit was consumed more during their pregnancy. Approximately 9 % drank milk while 2.4 % disliked it. Nearly 15 % of pregnant women stated that they disliked hot papaya salad during their pregnancy especially its taste and odor. 42.4 % of subjects had no preference toward food choices. As for meat, pregnant women disliked beef compared to other kinds of meat.

**Table 10** The percentages of food likes and dislikes in pregnant women

Food items	Likes	Dislikes
	%(n)	%(n)
General fruit	21.2(36)	4.7(8)
Sour fruit	17.1(29)	-
Milk	8.8(15)	2.4(4)
Dessert	8.2(14)	3.0(5)
Noodles	8.2(14)	2.4(4)
Northeastern foods / papaya salad	6.5(11)	14.6(25)
Oily food	5.8(10)	4.2(7)
Meat	5.3(9)	5.8(10)
Fish	2.9(5)	6.4(11)
Fermented / raw food	-	7.0(12)
Strong smell vegetable	-	1.8(3)
No special food	11.2(19)	42.4(72)
Others	2.4(4)	2.4(4)

Half of the subjects preferred mild taste food regardless of their preference of tasty food before they became pregnant (see Table 11).

**Table 11** Taste preference of subjects before and during their pregnancy

Taste	Before pregnancy	During pregnancy
	%(n)	%(n)
Sour	24.7(42)	16.5(28)
Sweet	5.9(10)	10.6(18)
Mild	5.3(9)	50.0(85)
Hot and spicy	32.9(56)	8.8(15)
Mixed	31.2(53)	14.1(24)

As shown in Table 12, most subjects did the cooking by themselves regardless of their physical status (pregnant or non- pregnant).

**Table 12** The percentages of cooking patterns before and during their pregnancy

Cooking pattern	Before pregnancy	During pregnancy
	%(n)	%(n)
Cooking themselves	88.8(151)	89.4(152)
Buying cooked food	11.2(19)	10.6(18)

During pregnancy, food is an important factor for both mothers and fetuses. In normal practices, pregnant women increased their food intake. They were very selective in food items for their consumption. In the present study, both special food items and taboo food items were also interviewed. One third of pregnant mothers

stated that no special food items were introduced during their pregnancy. However, others stated that milk, fruit and meat were special for them where their consumption for these items had been increasing during their pregnancy period. On the contrary, the majority of pregnant mothers explained that they had no food taboo or restriction during their pregnancy. In the health education session, they were educated by health officials concerning healthy food. Few of them had some food taboo. A list of food restriction items was, also illustrated, i.e., fermented food, bamboo shoot, pond-snail and black glutinous rice.

#### **4.3 Frequency of food consumption**

Food consumption frequencies were measured using food frequency questionnaires. All subjects were interviewed during their second and third trimesters. The results were shown in Tables 2 and 3, Appendix A).

##### **Meat, poultry, fish and their products**

Approximately 51.8 %, 67 %, 47.6 % and 54.1 % of subjects consumed pork, poultry, beef and fresh-water fish respectively. One to three times a week was the frequency that was most stated and in the quantity of a half cup.

In the view of seafood, 41.8 % of mothers consumed 1-3 times a week. Fifty one percent of them did not have seafood and their products during the interview period. Some mothers mentioned that they disliked seafood and that it was not available for them either. Mackerel was mostly consumed since it was usually available for them in their community. Approximately 69.4 % of subjects consumed mackerel 1-3 times a week and in the amount of one third cup.

Eggs were the most preferable items for pregnant women. Forty-four percent of them consumed eggs daily and 36.5 % consumed 1-3 times a week. Fried eggs were mostly consumed in the amount of one piece per meal.

Other protein food, e.g., dry fish, squids, traditional sausages, meatballs and animal organs, was consumed 1-3 times a week. Various quantities of food were stated by the mothers and were presented in Table 3, Appendix A. The first five order of food in this group which the pregnant women liked to consume could be prioritized as follows: eggs, pork, fresh-water fish, poultry and mackerel respectively.(as shown in Tables 2 and 3, Appendix A and for calculation method, see in Table 4, Appendix B)

#### **Legumes and their products**

Rural Northeasterners disliked soybean curd, therefore, 88.2 % of mothers did not consume soybean curd. Other legumes that mothers were not familiar with were cowpea and mungbean, for instance. Peanuts, both steamed and roasted, were the most preferable among pregnant mothers. Fifty-two percent of pregnant mothers consumed 1-3 times a week and in the amount of approximately a half cup (48.2 %). Soybean milk in plastic package was consumed owing to its low cost and locally available.

#### **Cereals, grain products, starchy roots and tubers**

It was found that pregnant mothers consumed glutinous rice more frequently (89.4 %) than ordinary rice (25.9 %) daily, indicating that most pregnant mothers who came from other parts or worked in town were the group that consumed more ordinary rice. Thirty five percent consumed one cup of ordinary rice while 35.3 % consumed 2 cups while forty eight percent consumed 1 cup of glutinous rice. However, they consumed more ordinary rice in comparison.

More than half of the pregnant mothers consumed noodles (55.2 %) and rice noodles (55.9 %) (local name, Kanom jean) 1-3 times a week. The amount of one cup per meal was the amount consumed, i.e., 64.7 % and 40.6 % respectively.

Less than half (43.5 %) of the subjects disliked instant noodles. However, approximately 34.7 % of them consumed 1-3 times a week. The quantity that was mostly consumed was 1 pack (53.5 %). Occasionally, they had boiled instant noodles with no addition of other ingredients, neither vegetables nor meat. However, they added some eggs and vegetables in some instances.

Considering mung bean noodles, they normally consumed half to one cup for 1-3 times a week. In addition, other dishes of mung bean noodles were to go with chicken. This was very common in the Northeast (local name, Tom Sen or Tome Wun Sen). More than half of the pregnant mothers did not consume bread, sponge cakes, crackers or donuts, i.e., 53.5 %, 50.5 %, 65.9 % and 67.1 % respectively. Table 2, Appendix A indicated that 45.9 % and 57.7 % of subjects consumed taro, sweet potato and corn for 1-3 times a week with varying quantity. The first five order of food in this group which the pregnant women liked to consume could be prioritized as follows: glutinous rice, ordinary rice, rice noodles (Kanom jean), corn and noodles respectively (as shown in Tables 2 and 3, Appendix A and for calculation method, see in Table 4, Appendix A).

### **Vegetables**

Pregnant mothers frequently consumed green vegetables that were available in their community. Pumpkin, as a source of carotene, was consumed 1-3 times a week but not usually found in their community. The average quantity of vegetable intake was 1 cup. The popular vegetable dish was papaya salad (Som Tum) which was

consumed among most mothers. Up to 1-3 times a week and half to one cup was consumed.

### **Fruit**

During their pregnancy, pregnant mothers consumed more fruit. Forty three percent had a daily consumption and 48.2 % had a 1-3 times consumption per week. Approximately one cup or more was the mean quantity intake of fruit. The variety of fruit consumed depended on seasons. The most common fruit consumed was banana, orange, pineapple, watermelon, guava and ripe papaya.

### **Fat and oil**

Approximately 43.5 % of subjects used oil for daily cooking. Twenty-two percent used 1-3 times per week. They normally used vegetable oil for eggs, meat and stirred-fry vegetables. The subjects stated that they took 1 teaspoon fat per meal. Forty-nine percent did not utilize coconut milk in their food. Coconut milk was preferred more in a dessert or sweets. Generally, a half cup per meal was consumed.

### **Dessert**

Frequency of dessert consumption was 1-3 times a week in the amount of 1 cup each time. Approximately 29 % of mothers did not have dessert during their pregnancy. The most popular dessert with coconut milk was banana, pumpkin, lod-chong, sago (tapioca pearl), taro and native melon respectively. Other dessert that went with ice was also consumed.

Frequency of ice cream consumption was 1-3 times a week in the amount of a half cup each time. Most of ice cream they consumed was coconut milk ice cream.

**Beverages**

Easy delivery was the reason for the consumption of coconut juice among most mothers. Approximately 37.6 % of the subjects had 1-3 times per week of carbonated beverages. Limited consumption of fruit juice existed among pregnant women.

**Milk and milk products**

Daily milk consumption was evidenced in approximately half of the subjects (47.6 %). Varieties of milk such as drinking yogurt and whole milk both sweet and natural flavored were consumed among pregnant mothers. The pregnant women drank whole milk more frequently than other kinds of milk (as shown in Table 2, Appendix A).



## Conclusion

Most nutrient intake was adequately consumed among pregnant women comparing with 75 % RDA. The exception was in the case of iron and calcium. Pregnant women normally cooked for themselves. Mild taste dishes were their preferences. Fruit, meat, milk and eggs were the items that they consumed more frequently than usual because they knew that these kinds of food were important for themselves and their infants.

## 5. Protein intake during the second and third trimesters

Protein intake was categorized into 3 levels: low (< 51 g), moderate (51-100 g) and high (> 100 g) respectively. There were 9.4 %, 82.4 % and 8.2 % of subjects falling in the low, moderate and high protein intake groups in the second trimester. Considering the third trimester, 8.8 %, 83.0 % and 8.2 % of subjects were categorized into low, moderate and high protein intake categories.

In the second trimester, high protein intake group tended to have higher weight gain than other groups. Low protein intake group had shortest gestational age. There was significant difference among gestational age except birth weight and birth length of subjects (see Table 13). Nevertheless, significant difference was found between low and high protein intake over gestational age using LSD test (see Table 5, Appendix A ). There was a significant difference for overall dependent variables using Manova analysis.

In the third trimester, high protein intake group tended to have shorter birth length and gestational age. Employing Anova analysis, no significant difference was encountered concerning birth weight and birth length except for gestational age. There was no significant difference for overall dependent variables using Manova analysis. Results were shown in Table 14 (see Table 6, appendix A ).

**Table 13** Average birth weight, birth length and gestational age by protein intake (g) in the second trimester

Level of protein intake T <sub>2</sub>		Birth weight(g)	Birth length(cm)	Gestational age(day)
	%(n)	Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Low	9.4(16)	3088.13±486.28 (2250, 2980)	50.50±2.31 (46, 54)	264.44±12.51 (230, 283)
Moderate	82.4(140)	3117.82±377.00 (2000, 4160)	49.84±2.34 (43, 55)	273.16±10.06 (245, 304)
High	8.2(14)	3382.14±614.03 (2670, 4720)	49.86±2.77 (45, 55)	272.39±10.74 (259, 301)
p – value (Anova)		0.066	0.575	0.006*
p – value (Manova)				
Wilks'lambda			0.002*	

\*p-value < 0.05

**Table 14** Average birth weight, birth length and gestational age by protein intake (g) in the third trimester

Level of protein intake T <sub>3</sub>		Birth weight(g)	Birth length(cm)	Gestational age(day)
	%(n)	Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Low	8.8(15)	3049.33±358.70 (2250, 3690)	49.67±2.09 (46, 52)	271.00±16.94 (230, 296)

**Table 14** Average birth weight, birth length and gestational age by protein intake (g) in the third trimester (continued)

Level of protein intake T <sub>3</sub>		Birth weight(g)	Birth length(cm)	Gestational age(day)
	%(n)	Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Moderate	83.0(141)	3151.31±427.60 (2000, 4720)	50.05±2.37 (43, 55)	273.18±9.66 (252, 304)
High	8.2(14)	3084.29±345.65 (2680, 3850)	48.71±2.33 (45, 53)	266.00±9.90 (245, 282)
P – value (Anova)		0.591	0.120	0.046*
P – value (Manova)				
Wilks' lambda			0.055	

\*p-value < 0.05

Aside from protein intake, the percentage of caloric intake from protein was also considered in the present study. Caloric distribution from protein was divided into 3 levels: low (< 15 %), moderate (15-20 %) and high (> 20 %). It was found that only 2 levels were encountered in either the second or third trimester. Approximately 60.6% and 39.4 % of subjects were categorized into low and moderate groups during the second and third trimesters. Fifty-seven percent and 42.9 % were classified as low and moderate groups during the third trimester.

No significant difference was found comparing mean birth weight, birth length and gestational age of both groups during the second and third trimesters.

**Table 15** Average birth weight, birth length and gestational age over the percentage of caloric intake from protein in the second trimester

%Caloric intake		Birth weight(g)	Birth length(cm)	Gestational age(day)
from protein T <sub>2</sub>	%(n)	Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Low	60.6(103)	3144.27±387.62 (2250, 4150)	49.87±2.25 (46, 55)	271.61±10.12 (245, 304)
Moderate	39.4(67)	3125.30±457.19 (2000, 4720)	49.96±2.55 (43, 55)	273.60±11.31 (230, 301)
p – value		0.117	0.413	0.386

**Table 16** Average birth weight, birth length and gestational ages over the percentage of caloric intake from protein in the third trimester

%Caloric intake		Birth weight(g)	Birth length(cm)	Gestational age(day)
from protein T <sub>3</sub>	%(n)	Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Low	57.1(97)	3140.88±459.29 (2000, 4720)	50.14±2.48 (43, 54)	272.63±10.50 (250, 304)
Moderate	42.9(73)	3131.37±351.21 (2540, 4160)	49.59±2.18 (45, 55)	272.08±10.84 (230, 296)
p – value		0.441	0.065	0.370

A comparison was made on birth weight, birth length and gestational age among subjects with different patterns of protein intake shifting during the second and third

trimesters. Therefore, protein intake in both trimesters was accounted for in the present study. The protein intake was divided into 7 groups: low-low, low-moderate, moderate-low, moderate-moderate, moderate-high, high-moderate and high-high protein intake group. None of the pregnant women was in low-high and high-low groups. The majority of subjects were categorized into moderate-moderate group (70.6 %). Considering fetal outcomes, results indicated that low-low group gave birth to the lowest birth weight and shortest gestational age. However, this group was found to have longest birth length. The high-moderate group had highest birth weight while moderate-high group had shortest birth length. In addition, moderate-low group had longest gestational age. In accordance with gestational age, protein intake shifted from lower to higher protein intake group; for example, low-moderate and moderate-high groups tended to have short gestational age. On the contrary, when protein intake shifted from higher to lower, i.e., high-moderate and moderate-low, they tended to have long gestational age. Only gestational age was a significant difference by Anova method (see Table 17). LSD test indicated a significant difference of high-moderate protein intake group except for the high-high protein intake group. Low-low protein intake group statistically differed from other groups. Exception was for moderate-high protein intake group (see Table 7, Appendix A). Using Manova analysis, there was a significant difference for overall dependent variables ( $p$ -value = 0.05).

**Table 17** Average birth weight, birth length and gestational age by protein intake level in the second and third trimesters

Level of protein intake T <sub>2</sub> -T <sub>3</sub>		Birth weight(g)	Birth length(cm)	Gestational age(day)
	%(n)	Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Low-low	2.4(4)	3012.50±265.00 (2650, 3280)	51.00±0.82 (50, 52)	255.50±21.98 (230, 283)
Low-moderate	7.0(12)	3113.33±548.21 (2250, 3980)	50.33±2.64 (46, 54)	267.42± 6.54 (252, 276)
Moderate-low	6.5(11)	3062.73±397.90 (2250, 3690)	49.18±2.23 (46, 52)	276.64±11.22 (259, 296)
Moderate-moderate	70.6(120)	3127.96±381.98 (2000, 4160)	50.01±2.33 (43, 55)	273.58±9.64 (252, 304)
Moderate- high	5.3(9)	3050.00±300.46 (2680, 3600)	48.44±2.19 (48, 53)	263.33±9.57 (245, 278)
High-moderate	5.3(9)	3513.33±676.98 (2670, 4720)	50.22±2.86 (45, 55)	275.56±11.53 (265, 301)
High- high	2.9(5)	3146.00±447.58 (2730, 3850)	49.20±2.77 (45, 52)	270.80±9.52 (259, 282)
p – value (Anova)		0.196	0.367	0.000*
p – value (Manova)				
Wilks' lambda			0.001*	

\*p-value < 0.05

Using energy intake in the second and third trimesters as covariate for analysis of covariance, the adjusted birth weight, birth length and gestational age were lower. However, results were similar to unadjusted data. Shortest birth length shifted from moderate-high to moderate-low protein intake group. At the same time, shortest gestational age shifted from low-low to moderate-moderate protein intake group. Multiple comparison was also calculated; there was no statistical difference of birth weight, birth length and gestational age. The results were illustrated in Table 18

**Table 18** Averagely adjusted birth weight, birth length and gestational age in the second and third trimesters

Group of protein intake T <sub>2</sub> -T <sub>3</sub>	Birth weight(g)	Birth length(cm)	Gestational age(day)
	Mean	Mean	Mean
Low-low	2990.13	50.96	254.05
Low-moderate	3161.11	50.57	265.96
Moderate-low	3017.41	46.68	276.08
Moderate-moderate	3128.99	50.02	243.58
Moderate- high	3138.63	49.37	263.60
High-moderate	3433.07	49.48	277.21
High- high	3019.01	48.94	273.18

Aside from protein intake (g), the percentage of caloric intake from protein in the second and third trimesters was also employed in the present study. Caloric intake from protein was categorized into 4 groups, i.e., low-low, low-moderate, moderate-low and moderate-moderate respectively. There were 38.8%, 21.8%, 18.2% and

21.2%) of pregnant women in low-low, low-moderate, moderate-low and moderate-moderate groups respectively. High caloric intake was not evidenced in the second and third trimesters. This may be due to selection criteria where low and lower middle socioeconomic classes were recruited. To the fact that meat and milk were too much to afford for these groups, no significant difference among various groups was demonstrated in this instance using Anova and Manova analysis (see Table 19). The multiple comparison using LSD test was shown in Table 8, Appendix A

**Table 19** Average birth weight, birth length and gestational age on percentage of caloric intake from protein in the second and third trimesters

%Caloric intake from protein T <sub>2</sub> -T <sub>3</sub>	% (n)	Birth weight(g)	Birth length(cm)	Gestational age(day)
		Mean±SD	Mean±SD	Mean±SD
		(Min, Max)	(Min, Max)	(Min, Max)
Low-low	38.8(66)	3143.48±425.32 (2250, 4150)	50.20±2.43 (46, 55)	271.76±10.53 (250, 304)
Low-moderate	21.8(37)	3145.68±314.80 (2680, 4110)	49.30±1.78 (46, 54)	271.35±9.49 (245, 290)
Moderate-low	18.2(31)	3135.32±532.02 (2000, 3160)	50.03±2.63 (43, 54)	274.48±10.36 (259, 301)
Moderate-moderate	21.2(36)	3116.27±389.06 (2500, 4160)	49.89±2.53 (45, 55)	272.83± 12.16 (230, 296)
p – value (Anova)		0.990	0.317	0.606
p – value (Manova)				
Wilks' lambda			0.503	

### Protein intake quality

Mean birth weight, birth length and gestational age in each trimester were assessed. It was found that no significant difference was evidenced among subjects who had good (animal protein: vegetable protein  $\geq 1:2$ ) and poor (animal protein: vegetable protein  $< 1:2$ ) qualities of protein intake except for gestational age in the third trimester ( $p = 0.026$ ). Good quality of protein intake in the third trimester had longer gestational age than poor quality of protein intake in the second trimester. (see Table 20-21).

**Table 20** Average birth weight, birth length and gestational age on quality of protein intake in the second trimester

Quality of protein T <sub>2</sub>	%(n)	Birth weight(g)	Birth length(cm)	Gestational age(day)
		Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Good quality	97.6(166)	3131.77±413.29 (2000, 4720)	49.86±2.35 (43, 55)	272.45±10.63 (230, 304)
Poor quality	2.4(4)	3262.50±543.71 (2700, 4000)	51.75±2.63 (48, 54)	270.25±11.18 (259, 283)
p – value		0.271	0.057	0.342

**Table 21** Average birth weight, birth length and gestational ages on quality of protein intake in the third trimester.

Quality of protein T <sub>3</sub>	% (n)	Birth weight(g)	Birth length(cm)	Gestational age(day)
		Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Good quality	98.2(167)	3141.83±413.19 (2000, 4720)	49.88±2.36 (43, 55)	272.60±10.54 (230, 403)
Poor quality	1.8(3)	2856.67±526.91 (2250, 3200)	51.33±2.52 (49, 54)	260.67±9.61 (252, 271)
p – value		0.120	0.146	0.026

Considering protein quality in both trimesters (Table 22), it was found that none had poor protein quality. There were 2.4%, 1.7% and 95.9% of pregnant women in poor-good, good-poor and good-good levels of quality of protein intake respectively. Subjects who had a poor quality protein in the second trimester converted to good protein quality in the third trimester (poor-good group). They also had higher birth weight and birth length compared with other groups. The good-poor quality protein intake group had lower birth weight and shorter gestational age than other groups. The good-good quality protein group had shortest birth length but longer gestational age. LSD test indicated no significant difference for birth weight, birth length and gestational age among pregnant women in both trimesters (See Table 9, Appendix A).

**Table 22** Average birth weight, birth length and gestational age on protein quality in the second and third trimester

Quality of protein T <sub>2</sub> -T <sub>3</sub>	% (n)	Birth weight(g)	Birth length(cm)	Gestational age(day)
		Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Poor-good	2.4(4)	3262.50±543.71 (2700, 4000)	51.75±2.63 (48, 542)	270.25±11.18 (59, 283)
Good-poor	1.7(3)	2856.67±526.91 (2250, 3200)	51.33±2.62 (49, 54)	260.67±9.61 (252, 271)
Good-good	95.9(163)	3138.87±411.22 (2000, 4720)	49.83±2.34 (43, 55)	272.66 ±10.56 (230, 403)
p – value (Anova)		0.422	0.159	0.358
p – value (Manova)				
Wilks' lambda			0.085	

In addition to protein intake for birth weight, birth length and gestational age, energy intake was also assessed. The significant difference was found for birth weight, birth length and gestational age by Anova and Manova analysis (see Table 23). Results indicated that low-low protein intake group had less energy in both trimesters. The increase and decrease of protein intake were in a similar pattern of energy intake.

Considering the energy intake with 75% Recommended Daily Dietary Allowance (1725 Kcal), the pregnant women in low-low, low-moderate (only the second trimester) and moderate-low groups had energy intake under requirement. The other groups from moderate-moderate to high-high had adequate energy intake.

Employing LSD method for multiple comparison, it showed that energy intake of low-low, low-moderate and moderate-low protein intake groups did not statistically differ among these three groups. However, energy intake of these groups was significantly different from moderate-moderate, moderate-high, high-moderate and high-high groups. Other significantly different groups were presented in Table 10, Appendix A.

**Table 23** Average energy intake in the second and third trimesters for the protein intake categories

Level of protein intake T <sub>2</sub> -T <sub>3</sub>	Energy intake T <sub>2</sub> (Kcal)	Energy intake T <sub>3</sub> (Kcal)
	Mean±SD	Mean±SD
Low-low	1307.58±358.92	1199.66±170.31
Low-moderate	1369.80±288.41	1826.55 ±285.95
Moderate-low	1664.99±175.34	1376.41±269.46
Moderate-moderate	2007.55±394.67	2094.46±423.44
Moderate- high	2211.37±385.80	3004.66±342.99
High-moderate	2694.31±303.51	2134.03±344.92
High- high	3074.00±796.36	2865.31±421.21
p – value (Anova)	0.000 *	0.000*
p – value (Manova)		
Wilks' lambda	0.000*	

Comparing energy intake and fetal outcomes with 100 % RDA as a cut-off point of energy intake (2300 kcal), the energy intake was categorized into 4 groups, i.e.,

low-low (58.2%), low-high (15.9%), high-low (10.6%) and high-high(15.3%) respectively. No significant difference was encountered among birth weight, birth length and gestational age among the subjects using Anova and Manova analysis ( see Table 24).

**Table 24** Average birth weight, birth length and gestational age among different energy intake in the second and third trimesters

Energy intake in T <sub>2</sub> -T <sub>3</sub>	% (n)	Birth weight(g)	Birth length(cm)	Gestational age(day)
		Mean±SD (Min, Max)	Mean±SD (Min, Max)	Mean±SD (Min, Max)
Low-low	58.2(99)	3105.81±413.95 (2000, 4720)	49.92±2.29 (43, 55)	273.49±11.26 (230, 403)
Low-high	15.9(27)	3076.67±353.71 (2570, 3850)	49.44±2.34 (46, 54)	269.93±11.41 (245, 294)
High- low	10.6(18)	3292.22±478.17 (2670, 4160)	50.72±2.42 (45, 55)	272.28±7.12 (261, 288)
High-high	15.3(26)	3209.62±421.34 (2430, 4100)	49.77±2.60 (45, 55)	270.85±9.01 (258, 294)
p – value (Anova)		0.218	0.355	0.382
p – value (Manova)				
Wilks' lambda			0.238	

In addition of energy intake, total weight gain was also considered. The low-low protein intake group had the lowest total weight gain. The moderate-high protein intake group had the highest total weight gain. However, the total weight gain was not

significantly different among the different protein intake groups (  $p = 0.422$  ), as shown in Table 25

**Table 25** Total weight gain among protein intake categories

Level of protein intake T <sub>2</sub> -T <sub>3</sub>	Total weight gain (Kg)	
	Mean±SD	(Min, Max)
Low-low	9.30 ±1.73	(7.0, 11.2)
Low-moderate	9.72±1.53	(7.0, 12.0)
Moderate-low	10.55±3.03	(5.5, 16.5)
Moderate-moderate	11.25 ±2.93	(5.0, 20.0)
Moderate- high	11.64 ±1.91	(8.8, 15.0)
High-moderate	11.12±2.68	(6.8, 14.0)
High- high	10.56±3.08	(7.4, 15.4)
p- value	0.422	

## Conclusion

Gestational age was significantly different among the groups with different protein intake. However, this was not the case for birth weight and birth length. Comparing birth weight, birth length and gestational age among protein intake in both trimesters, The results showed that low-low protein intake tended to give birth of lowest birth weight and shortest gestational age but longest birth length. In accordance with gestational age, protein intake shifted from lower to higher protein intake tended to have short gestational age while protein intake shifted from higher to lower protein intake tended to have long gestational age. The percentage of caloric intake from protein was not significantly different for birth weight, birth length and gestational age. In addition, birth weight, birth length and gestational age were not significantly different in groups with different quality of protein intake in both trimesters.

## **6. The association of nutrient intake, weight gains and birth weight, birth length and gestational age**

The correlation matrix of dependent variables, i.e., birth weight, birth length and gestational age, and maternal nutrition variables, i.e., nutrient intakes and weight gain were presented in Tables 11, 12, Appendix A.

In addition of variables in each trimester, changing vegetable protein intake between the second and third trimesters was also done. Changing protein intake was calculated by subtracting the protein intake in the second trimester (numerator) by protein intake in the third trimester. The correlation matrix of changing protein intake, changing energy intake between the second and third trimesters and dependent variables were shown in Table 13, appendix A.

### **Birth weight**

Vegetable protein intake in the second trimester, total weight gain, prepregnancy weight, prepregnancy BMI, gestational age, changing energy intake between the second trimester and third trimesters, changing total protein intake between the second and third trimesters and changing vegetable protein intake between the second and third trimesters correlated positively with birth weight.

### **Birth length**

Prepregnancy weight, prepregnancy BMI, total weight gain, changing energy intake between the second and third trimesters, changing total protein intake between the second and third trimesters, and changing vegetable protein intake between the second and third trimesters positively correlated with birth length. Other variables which negatively correlated with birth length including energy, animal protein, total protein, phosphorus and calcium intake in the third trimester.

### **Gestational age**

Rate of weight gain in the third trimester correlated negatively with gestational age.

Prepregnancy weight and total weight gain were the variables correlated positively with birth weight and birth length.

Considering total weight gain during pregnancy, it was found that energy and fat intake in both the second and third trimesters correlated positively with total weight gain. In other words, the pregnant women who consumed high calorie and fat intake had higher weight gain.

In addition of correlation matrix, the partial correlation by controlling energy intake was also considered. The unique effect of protein intake was also scrutinized. Since energy comes from carbohydrate, fat and protein whenever energy intake is inadequate, protein will be converted to energy and lessened its other functions.

After controlling energy intake in the second trimester, vegetable protein lost its correlation with birth weight as shown in Table 26.

The same pattern of changing total protein intake between the second and third trimester and changing vegetable protein intake between the second and third trimesters were found after controlling energy intake in the second and third trimesters as shown in Table 27.

For birth length, the controlling of energy intake in the third trimester, calcium and phosphorus were changed from significantly correlated to non-significantly with birth length. However, the effect of protein intake and animal protein still existed on birth length when controlling for the effect of energy intake in the third trimester as shown in Table 26.

After controlling energy intake in the second and third trimesters, changing total protein intake between the second and third trimesters, and changing vegetable protein intake between the second and third trimesters lost its correlation with birth length as shown in Table 27.

Considering gestational age, simple correlation showed that no correlation existed between gestational age and total protein intake in the second trimester, and gestational age and animal protein intake in the second trimester in pregnant women. After controlling energy intake in the second trimester as shown in Table 26, it was found that total protein intake in the second trimester significantly correlated with gestational age, and so did the animal protein intake in the second trimester. The correlation of total protein intake in the second trimester and gestational age increased from  $r = 0.094$  to  $0.165$  and animal protein intake in the second trimester increased from  $r = 0.131$  to  $0.159$ . Hence, the higher the total protein and animal protein intake in the second trimester was, the longer the gestational age.

**Table 26** The partial correlation among protein, calcium, phosphorus and birth weight, birth length, gestational age when controlling energy in each trimester

Variables	Second trimester (T <sub>2</sub> )				Third trimester (T <sub>3</sub> )							
	birth weight		birth length		birth weight		birth length		Gestational age			
	Simple correlation	Controlling energy T <sub>2</sub>	Simple correlation	Controlling energy T <sub>2</sub>	Simple correlation	Controlling energy T <sub>3</sub>	Simple correlation	Controlling energy T <sub>3</sub>	Simple correlation	Controlling energy T <sub>3</sub>		
Total protein T <sub>2</sub>	0.146	0.057	-0.009	-0.079	0.094	0.165*	-0.039	-0.015	-0.229*	-0.162*	-0.062	-0.026
Total protein T <sub>3</sub>												
Animal protein T <sub>2</sub>	0.089	0.007	-0.072	-0.121	0.131	0.159*	-0.026	-0.008	-0.218*	-0.155*	-0.019	0.015
Animal protein T <sub>3</sub>												
Vegetable protein T <sub>2</sub>	0.169*	0.100	0.108	0.141	-0.019	-0.049	-0.041	-0.017	-0.126	0.046	-0.105	-0.113
Vegetable protein T <sub>3</sub>												
Ca T <sub>2</sub>	0.034	-0.035	0.085	-0.117	0.035	0.036	0.008	0.031	-0.158*	-0.085	0.070	0.117
Ca T <sub>3</sub>												
Phosphorus T <sub>2</sub>	0.033	-0.048	-0.023	-0.052	0.067	0.075	0.007	0.027	-0.192*	-0.106	0.071	0.157
Phosphorus T <sub>3</sub>												

\*p – value < 0.05

**Table 27** The partial correlation among changing protein intake between the second and third trimesters and birth weight, birth length and gestational age when controlling energy intake in both trimesters

Variables	Birth weight		Birth length		Gestational age	
	Simple correlation	Controlling energy T2-T3	Simple correlation	Controlling Energy T2-T3	Simple correlation	Controlling energy T2-T3
<b>Changing Total Protein T<sub>2</sub>-T<sub>3</sub></b>	0.172*	0.055	0.217*	0.086	0.148	0.149
<b>Changing animal Protein T<sub>2</sub>-T<sub>3</sub></b>	0.101	0.009	0.142	0.049	0.129	0.113
<b>Changing Vegetable Protein T<sub>2</sub>-T<sub>3</sub></b>	0.204*	0.116	0.235*	0.092	0.091	0.057

p-value<0.05

Considering the regression model, several curvilinear regression models were tried but no additional effect was found so only linear regression models were presented in this study. Many variables which associated with birth weight, birth length and gestational age were selected in the model of regression equation by using the following criteria:

1. Potential factors that were selected in the equation should be answering the hypotheses;
2. P-value of overall F test for the equation should be statistically significant;
3. P- value of T test for regression coefficient ( $\beta$ ) of each variable should be significant. However, some variables might be included in the equation even though they did not correlate with dependent variables but had an effect on the correlation between dependent variables and other variables in the equation;

4. Coefficient of determination ( $R^2$ ) should be the highest;
5. The equation should be explainable in term of biological plausibility.

### 6.1 Nutrient intake, weight gain and birth weight

Considering birth weight, gestational age became an independent variable because the gestational age of mothers also influenced infant birth weight, i.e., the longer the gestational age, the heavier the birth weight was. The regression model was presented in Table 14, Appendix A and the variables were screened into equation from the correlation matrix in Table 28. Considering the above criteria, Model 4 was the most appropriate model that associated with birth weight ( $R^2 = 0.250$ ). Prepregnancy weight, changing vegetable protein intake between the second and third trimesters, total weight gain and gestational age were the variables in the model. All variables correlated positively with birth weight. This indicated the increase of prepregnancy weight, changing vegetable protein intake between the second and third trimesters, total weight gain and gestational age with the increase of infant birth weight. Simple correlation and partial correlation of this model were presented in Table 29.

The variables in Model 4 which affected infant birth weight could be prioritized as follows: total weight gain, gestational age, prepregnancy weight and changing vegetable protein intake between the second and third trimesters respectively (Beta = 0.293, 0.266, 0.184 and 0.156).

It was noticeable for changing vegetable protein intake between the second and third trimesters and birth weight that correlated with vegetable protein intake in the second trimester and birth weight at  $r = 0.169$  ( $p < 0.05$ ) but not with vegetable protein intake in the third trimester.

**Table 28** The correlation matrix of some variables in birth weight model

Variables	Prepregnancy weight	Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	Vegetable protein T <sub>2</sub>	Energy T <sub>2</sub>	Total weight gain	Birth weight
<b>Prepregnancy Weight</b>	/					
<b>Changing Vegetable Protein T<sub>2</sub>-T<sub>3</sub></b>	0.089	/				
<b>Vegetable protein T<sub>2</sub></b>	0.105	0.429*	/			
<b>Energy T<sub>2</sub></b>	0.028	0.302*	0.850*	/		
<b>Total Weight Gain</b>	0.051	0.025	0.147	0.173*	/	
<b>Birth Weight</b>	0.229*	0.204*	0.169*	0.137	0.321*	/

\* p- value < 0.05

**Table 29** Regression analysis in Model 4 of birth weight.

Variables	$\beta$	R <sup>2</sup>	Simple correlation	Partial correlation	Sig. T
1. Constant	-701.344				0.346
2. Prepregnancy weight	9.731		0.229*	0.207	0.007*
3. Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	7.373		0.204*	0.176	0.023*
4. Total weight gain	43.596		0.321*	0.320	0.000*
5. Gestational age	10.418	0.250	0.308*	0.292	0.000*

\* p – value < 0.05

## 6.2 Nutrient intake, weight gain and birth length

Similar process of model selection was done for birth length as shown in Table 15, Appendix A. The variables were screened into the equation from the correlation matrix as shown in Table 30. Gestational age was also considered as independent variable of birth length but no correlation was found as demonstrated in the correlation matrix. Considering the above criteria, Model 3 was the most appropriate model that associated with birth length because this model was the best model to explain the variance of birth length. Prepregnancy weight, changing vegetable protein intake between the second and third trimesters, animal protein intake in the third trimester, and total weight gain were the variable in this model ( $R^2 = 0.211$ ). Simple correlation and partial correlation were shown in Table 31.

The significant variables in this model which affected infant birth length were prioritized as follow: total weight gain, prepregnancy weight, animal protein intake in the third trimester and changing vegetable protein between the second and third trimesters respectively (Beta = 0.252, 0.228, -0.221, and 0.181).

Models 3, 4, 5 and 6 had equal coefficient of determination ( $R^2 = 0.211$ ). Although calcium and phosphorus intakes in the third trimester were in the model, the coefficient of determination did not increase. It was noticeable that the partial correlation of total weight gain was larger than simple correlation. When there was deletion of calcium and animal protein intake in the third trimester, the partial correlation changed to smaller than simple correlation. Correlation matrix showed that animal protein intake in the third trimester did not correlate with total weight gain ( $r = 0.126$ ) but calcium intake in the third trimester correlated with total weight gain at  $r = 0.213$  ( $p < 0.05$ ). Animal protein intake in the third trimester correlated with calcium

intake in the third trimester correlated highly at  $r = 0.660$  ( $p < 0.05$ ). The changing of partial correlation between total weight gain and birth length is a net suppression of calcium intake in the third trimester on total weight gain as shown in Table 31. Model 1 whereas that induced by animal protein intake in the third trimester might be due to the high correlation between animal protein intake in the third trimester and calcium intake in the third trimester. Eventhough calcium intake seemed to be more important in this suppression effect but animal protein intake in the third trimester contributed higher  $R^2$  to the equation.

Table 30 The correlation matrix of some variables in birth length model

Variables	Prepregnancy weight	Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	Animal protein T <sub>2</sub>	Calcium T <sub>2</sub>	Phosphorus T <sub>2</sub>	Animal protein T <sub>3</sub>	Calcium T <sub>3</sub>	Phosphorus T <sub>3</sub>	Total weight gain	Birth length
Prepregnancy weight	/									
Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	0.089	/								
Animal protein T <sub>2</sub>	-0.015	0.049	/							
Calcium T <sub>2</sub>	-0.046	0.011	0.642*	/						
Phosphorus T <sub>2</sub>	0.051	0.142*	0.628*	0.592*	/					
Animal protein T <sub>3</sub>	-0.030	0.124	0.373*	0.333*	0.247*	/				
Calcium T <sub>3</sub>	-0.126	-0.183	0.385*	0.506*	0.275*	0.660*	/			
Phosphorus T <sub>3</sub>	-0.078	-0.277*	0.495*	0.520*	0.358*	0.772*	0.844*	/		
Total weight gain	0.051	0.025	0.108	0.126	0.120	0.126	0.213*	0.184*	/	
Birth length	0.264*	0.235*	-0.072	-0.085	-0.023	-0.218*	-0.158*	-0.192*	0.240*	/

\* p – value < 0.05

**Table 31** Regression analysis in some models of birth length**Model 3 ( Selected model)**

Variables	$\beta$	$R^2$	Simple correlation	Partial correlation	Sig. T
1. Constant	45.294				0.000*
2. Prepregnancy weight	0.069		0.264*	0.248	0.001*
3. Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	0.049		0.235*	0.197	0.011*
4. Animal protein T <sub>3</sub>	-0.031		-0.218*	-0.237	0.002*
5. Total weight gain	0.213	0.211	0.240*	0.270	0.000*

**Model 4 (Addition of calcium in the third trimester)**

Variables	$\beta$	$R^2$	Simple correlation	Partial correlation	Sig. T
1. Constant	45.305				0.000*
2. Prepregnancy weight	0.068		0.264*	0.245	0.001*
3. Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	0.048		0.235*	0.195	0.012*
4. Animal protein T <sub>3</sub>	-0.031		-0.218*	-0.179	0.021*
5. Calcium T <sub>3</sub>	-0.00008		-0.158*	-0.006	0.936 <sup>NS</sup>
6. Total weight gain	0.214	0.211	0.240*	0.269	0.001*

**Table 31** Regression analysis in some models of birth length (continued)**Model 1 (Deletion of animal protein intake in the third trimester)**

Variables	$\beta$	$R^2$	Simple correlation	Partial correlation	Sig. T
1. Constant	44.820				
2. Prepregnancy weight	0.065		0.264*	0.299	0.000*
3. Changing vegetable protein intake T <sub>2</sub> -T <sub>3</sub>	0.049		0.235*	0.193	0.012*
4. Calcium T <sub>3</sub>	-0.002		-0.158*	-0.158	0.041*
5. Total weight gain	0.217	0.185	0.240*	0.267	0.000*

**Deletion of calcium and animal protein in the third trimester**

Variables	$\beta$	$R^2$	Simple correlation	Partial correlation	Sig. T
1. Constant	44.072				0.000*
2. Prepregnancy weight	0.070		0.264*	0.247	0.001*
3. Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	0.056		0.235*	0.221	0.004*
4. Total weight gain	0.189	0.164	0.240*	0.237	0.002*

\* p – value &lt; 0.05, NS = Not Significant

### 6.3 Nutrient intake, weight gain and gestational age

The variables were screened into the equation from correlation matrix as shown in Table 32. When model fitting was done in gestational age (Table 16, Appendix A), it was found that Model 1 was the most appropriate model that associated with gestational age. Although Model 4 had the highest coefficient of determination, this model was not selected because total weight gain did not induce longer gestational age. On the contrary, longer gestational age induced higher total weight gain, so in this study total weight gain was not an independent variable of gestational age.

Energy intake in the second trimester, total protein intake in the second trimester and the rate of weight gain in the third trimester were variables in the model 1 ( $R^2 = 0.061$ ). In simple correlation, total protein intake in the second trimester did not correlate with gestational age. After the entry of energy intake in the second trimester, the correlation of total protein intake in the second trimester and gestational age was changed from non-significant to significant. From correlation matrix, total protein intake in the second trimester correlated positively with energy intake in the second trimester at  $r = 0.847$  ( $p < 0.05$ ) but energy intake in the second trimester did not correlate with gestational age. The correlation with these variables was classical suppression that energy intake in the second trimester increased the variance, accounted for in gestational age by suppression of some variance in total protein intake in the second trimester. (Table 33)

There was a similar finding of changing total protein intake between the second and third trimesters and animal protein intake in the third trimester on gestational age. Changing total protein intake between the second and third trimesters correlated with animal protein intake in the third trimester at  $r = -0.571$  ( $p < 0.05$ ) while both of

variables did not correlate with gestational age. After entering of animal protein intake in the third trimester, the correlation of changing total protein intake between the second and third trimesters and gestational age was changed from non-significant to significant as shown in Table 34.

Factors that affected gestational age in Model 1 were prioritized as follows: total protein intake in the second trimester, energy intake in the second trimester, and rate of weight gain in the third trimester respectively (Beta = 0.324, -0.241, and -0.185).

Table 32 The correlation matrix of some variables in gestational age model

Variables	Animal Protein T <sub>2</sub>	Total Protein T <sub>2</sub>	Energy T <sub>2</sub>	Changing total protein T <sub>2</sub> -T <sub>3</sub>	Animal protein T <sub>3</sub>	Total protein T <sub>3</sub>	Energy T <sub>3</sub>	Rate of weight gain T <sub>3</sub>	Gestational age
Animal protein T <sub>2</sub>	/								
Total protein T <sub>2</sub>	0.913*	/							
Energy T <sub>2</sub>	0.615*	0.847*	/						
Changing total protein T <sub>2</sub> -T <sub>3</sub>	0.448*	0.478*	0.374*	/					
Animal Protein T <sub>3</sub>	0.373*	0.344*	0.242*	-0.571*	/				
Total protein T <sub>3</sub>	0.395*	0.447*	0.409*	-0.012	0.903*	/			
Energy T <sub>3</sub>	0.368*	0.470*	0.528*	-0.372*	0.534*	0.817*	/		
Rate of weight gain T <sub>3</sub>	0.102	0.139	0.141	-0.129	0.210*	0.261*	0.232*	/	
Gestational age	0.131	0.094	0.008	0.489	-0.019	-0.062	-0.058	-0.174*	/

\* p - value < 0.05

**Table 33** Regression analysis in Model 1 of gestational age

Variables	$\beta$	$R^2$	Simple correlation	Partial correlation	Sig. T
1. Constant	274.868				0.000*
2. Energy T <sub>2</sub>	-0.005		0.008 <sup>NS</sup>	-0.131	0.091 <sup>NS</sup>
3. Total protein T <sub>2</sub>	0.177		0.094 <sup>NS</sup>	0.175	0.023*
4. Rate weight gain T <sub>3</sub>	-11.368	0.061	-0.174*	-0.186	0.016*

\* p-value <0.05 , NS = Not Significant

**Table 34** Regression analysis in Model 3 of gestational age

Variables	$\beta$	$R^2$	Simple correlation	Partial correlation	Sig. T
1. Constant	273.848				0.000*
2. Changing total protein T <sub>2</sub> -T <sub>3</sub>	0.101		0.148 <sup>NS</sup>	0.168	0.030*
3. Animal protein T <sub>3</sub>	0.085		-0.019 <sup>NS</sup>	0.110	0.154 <sup>NS</sup>
4. Rate weight gain T <sub>3</sub>	-10.812	0.058	-0.174*	-0.175	0.023*

\* p – value < 0.05, NS = Not Significant

## **Conclusion**

Total weight gain in pregnant women mostly derived from energy and fat intake. Prepregnancy weight and changing vegetable protein intake between the second and third trimesters correlated positively with birth weight and birth length. However, animal protein intake in the third trimester correlated negatively with birth length. Multiple regression showed that after the effects of other variables were partialized, the correlation of changing vegetable protein intake between the second and third trimesters on birth weight and birth length still existed with the direction that decreasing vegetable protein intake in the last trimester produced higher birth weight and birth length. In addition of gestational age, the total protein intake in the second trimester correlated positively with gestational age while the rate of weight gain in the third trimester correlated negatively with gestational age.

### **Conclusion of the results for each hypothesis**

**Hypothesis 1**, in low weight gain pregnant women, the quantity of protein intake in the second and third trimesters correlated negatively with infants' birth weight, birth length and gestational age.

Total protein intake correlated negatively only with birth length in the third trimester. Considering sources of protein intake, it was found that animal protein intake in the third trimester correlated negatively with birth length. Vegetable protein intake in the second trimester correlated positively with birth weight. This correlation supported the changing vegetable protein intake between the second and third trimesters in regression analysis results. With the higher vegetable protein intake in the second trimester and less vegetable in the third trimester, the higher birth weight and the higher birth length were found. Subjects who consumed of animal protein intake in the third trimester tended to have a shorter birth length infant. The total protein and animal protein intake in the second trimester correlated positively with gestational age.

**Hypothesis 2**, the pregnant women with low quantity of protein intake in both the second and third trimesters will give birth to lower infants' birth weight and birth length including shorter gestational age compare with other groups.

Pregnant women who consumed low protein intake in both trimesters tended to deliver low birth weight infants. However, they delivered higher birth length infants and their gestational age tended to be shorter than other groups.

**Hypothesis 3**, the pregnant women with low quantity of protein intake in the second trimester but high quantity of protein intake in the third trimester have shorter infants' gestational age than other groups.

Pregnant women who consumed protein intake shifted from lower to higher protein intake groups such as low-moderate and moderate-high group tended to have short gestational age.

**Hypothesis 4**, the pregnant mothers who have good quality protein intake will have heavier infants' birth weight, longer infants' birth length and gestational age than the pregnant women who have poor quality of protein intake.

Few subjects had low protein quality intake in both trimesters compared to other groups and no significant difference ( $p > 0.05$ ) was found among these groups. With the higher vegetable protein intake in the second trimester and the less vegetable protein intake in the third trimester, birth weight and birth length tended to increase. However, with higher animal protein intake in the second trimester, gestational age also increased.

## CHAPTER V

### DISCUSSION

Many studies on pregnant women indicated several factors influencing maternal anthropometric status and fetal outcomes. These factors were obstetric factors, maternal socioeconomic characteristics and nutritional status of prepregnancy and during pregnancy, smoking, alcohol consumption and complication during pregnancy.

The purpose of this study was to identify the effects of pregnancy protein intake in the second and third trimesters on birth weight, birth length and gestational age only for low weight gain pregnant women before the 20<sup>th</sup> week of gestational age.

Because many factors influenced birth weight, birth length and gestational age, this study would try to reduce the confounding variables which affected fetal outcomes and to select the pregnant women in similar characteristics. Maternal obstetric factors and socioeconomic characteristics were specified at the initial selection of subjects and may affect dependent variables in a narrow range. In order to clear any doubt about these factors affecting dependent variables, the dependent variables were compared with each factor.

#### **Maternal obstetric factors and fetal outcomes**

Considering prepregnancy weight and total weight gain, this study collected the data beginning from the 20<sup>th</sup> week of gestational age so the limitation of this study was that almost all pregnant women knew their approximate prepregnancy weight but

not precise weight. There might be some error in their prepregnancy weight. It was found that birth weight and birth length were significantly different among various prepregnancy weight groups. Only birth weight was significantly different among various total weight gain groups. This result was supported by multiple regression that prepregnancy weight and total weight correlated positively with birth weight and birth length, similar to the previous studies (46,72,80,141). However, the pregnant women who did not know their prepregnancy weight were not included in this study.

Nutritional factors that influenced their pregnancy outcomes were nutritional status before pregnancy and during pregnancy period. The nutritional status before pregnancy included pregnancy weight and height. The maternal nutritional status during pregnancy is the best indicated by weight gain during pregnancy.

Newborn size is related to maternal but not paternal height. This may be a nature survival mechanism to protect small mothers from having difficulties during labor, especially when they have a child of a larger father (30). Women who had heavier prepregnancy weight tended to have heavier babies. Women who had pregnancy and were underweight tended to deliver infants of low birth weight (31-39). Therefore, pregnant women who had prepregnancy weight under 45 kg were not included in this study. Pregnant women who had pregnancy weight under 55 kg tended to have lower birth weight and birth length than other groups. Nevertheless, comparing among prepregnancy Body Mass Index (BMI) groups, there was no significant difference among pregnant women. Most of the pregnant women were in the normal range of prepregnancy Body Mass Index (BMI).

Weight gain during pregnancy was a direct indicator that influenced fetal outcomes as stated before. Obstetricians monitored maternal weight gain for several

reasons. The main assumption was that weight gain was influenced by diet (11). However, total weight gain correlated positively with energy and fat intake in this study. This result was supported by a previous study (46) that maternal caloric intake during pregnancy was closely related to gestational weight gain. The maternal caloric intake and nutritional status (mostly fat) were the sole source for fetal energy requirements that weight gain during pregnancy would be expected to affect intrauterine growth. There was statistical difference only in birth weight among various total weight gain groups. The lower total weight gain, the lower was birth weight. The lowest weight gain was 5 kg and the highest was approximately 20 kg. In spite of low weight gain in the first trimester, later the majority of subjects could gain their weight during the second and third trimesters into a proper weight. This finding agreed with the previous studies (52,81) that low weight during the first trimester did not affect fetal outcomes. The last half of gestation, the fetus grew considerably faster while the appropriate quantity and quality of nutrients had to increase their proper weight and length. However, some studies conflicted (142) with this result that was causal relation between the poor first trimester growth and low birth weight; it may be that a suboptimal environment in the first trimester limited fetal growth for the remainder of pregnancy. Alternatively, poor growth in the first trimester may be secondary to a disorder of placentation that was manifested throughout pregnancy by suboptimal transfer of nutrients to fetus. It also seemed likely that some fetuses may be physiologically small throughout pregnancy.

### **Maternal socioeconomic characteristics and protein intake**

The significant difference of fetal outcomes among various maternal socioeconomic characteristics did not occur, but in view of protein intake, there was no significant difference in total protein intake and socioeconomic variables. Considering the source of protein intake, results showed that there was statistical difference in animal protein intake in the third trimester ( $p = 0.046$ ) among various family incomes. This indicated that the lower the family income, the lower the animal protein intake. Comparing animal protein intake in both trimesters, the women whose family income  $\leq 2500$  baht per month consumed animal protein in the second trimester higher than the third trimester while the women whose family income  $> 7500$  baht per month consumed animal protein in the second trimester, less than the third trimester. However, vegetable protein intake among various family incomes in both trimesters was not different. From food consumption pattern, it showed that the higher family income group consumed meat, milk and dairy products more than the lower family income one.

The pregnant women with various occupations had significant difference in vegetable protein intake in the third trimester ( $p = 0.043$ ). Farmers consumed more vegetable protein intake than other groups. However, there were no differences in total protein intake including vegetable and animal protein intake in the third trimester. Rice is the main source of vegetable protein of these subjects. From food frequency pattern, the farmer group consumed rice slightly higher in quantity than other groups. However, fetal outcomes among various socioeconomic statuses were not significantly different while the previous studies (57,143) found that meat and their products correlated positively to birth weight.

### **Food consumption and nutrient intakes**

Dietary intakes including 24-hour dietary recall and food records were done at least 4 times in each trimester for dietary assessment. The repeated 24-hour dietary recalls and food records in each trimester were reliable enough for this study. Additionally, the subjects in this study were in low and lower middle socioeconomic status. The majority of them came from rural areas so the dietary intakes were not complicated for dietary assessment.

The mean intake of the macro and micronutrients studied was above 75% of Thai Recommended Dietary Allowance (RDA) except for calcium and iron intake. However, all the subjects had normal haemoglobin because pregnant women were assigned to take obimin AF, 1 tablet per day in routine antenatal care clinic. The obimin AF tablet contains vitamins A, D, C, B<sub>1</sub>, B<sub>2</sub>, B<sub>6</sub>, B<sub>12</sub>, niacinamide, cal pantothenate, folic acid, ferrous fumarate, cal acid, acid lactate, cobalt, copper, dimethyl polysiloxane and fluoride (144). Therefore, pregnant women had medical nutrients supplement from the daily food intake.

Results of nutrient intakes were similar to those of Thailand National Nutrition Survey in 1995. The energy and protein intakes in the study were compared with those of Thailand National Nutrition Survey that was conducted in Northeastern pregnant women over 3 months of gestational age (145). The mean energy intake of Thailand National Nutrition Survey was 2124.1±829 Kcal (92.4% RDA). The mean energy intake of this study was 2002.4±505.0 Kcal (87.1% RDA) and 2081.0±520.6 Kcal (90.5% RDA) during the second and third trimesters respectively. Mean total protein intake of Thailand National Nutrition Survey was approximately 73.5±32.5g (144% RDA), approximately 45.2±56.1g (61% of total protein intake) which derived from

animal protein. The present study showed a similar finding in that the protein intake was equal to  $72.9 \pm 19.5$ g (142.5% RDA) and  $75.9 \pm 20.8$ g (148.8% RDA) in the second and third trimesters respectively. This quantity derived from animal protein about 42.9 g (58.8 % of total protein) and 45.1 g (59.4% of total protein) in the second and third trimesters respectively.

Energy distribution pattern in this study also corresponded to energy distribution of Northeastern pregnant women found in Thailand National Nutrition Survey in 1995. Energy distribution showed a typical pattern of intake of Northeastern part of Thailand. Carbohydrate provided the highest calorie, followed by fat and protein respectively. The percentage of the energy distribution of Thailand National Nutrition Survey was 67.8 carbohydrate: 17.7 fat: 14.5 protein. In present study, carbohydrate: fat: protein ratio was 68.3: 17.1: 14.6 and 68.0: 17.3: 14.7 during the second and third trimesters. Birth weight, birth length and gestational age were not significantly different when considered from the percentage of caloric intake from protein because the majority of subjects consumed the similar pattern of food intake. Previous studies (15,68) suggested that protein intake from vegetable and animal combined must keep below approximately 25-30% of their total daily energy intake to protect their health and wellbeing of the developing fetus. The protein intake >25% of total energy intake in the unbalanced diet may impair fetal growth. The protein intake in this study rarely approached this threshold. Furthermore, no effect of protein intake in subjects was from this phenomenon.

Considering food consumption behavior, frequency of glutinous rice intake was found to be the highest among starchy items. Pork, eggs and fresh-water fish were the main source of animal protein. Vegetable protein sources such as soybean, mungbean

were not common for them. Steamed or roasted peanuts were usually consumed as snack food. Vegetable oil and coconut milk were not consumed frequently. Normally, vegetable oil was used for frying eggs, meat and fish. Nevertheless, boiling and roasting were typical cooking methods employed in the normal practice of the Northeasterners (146). For milk intake frequency, approximately half of the subjects consumed whole, sweetened UHT milk and drinking yogurt. Soybean milk in plastic bags was one of their alternatives. In Maternal and Child Hospital, soybean milk was provided free to the pregnant women who attended the antenatal care clinic. In addition, fruit and vegetables were consumed more than usual during pregnancy. However, they reduced the consumption of certain food such as fermented food, hot and spicy food, raw food, etc. Information concerning this knowledge derived from the health education service class at the antenatal care clinic.

### **Protein intake and fetal outcomes**

During pregnancy, the increment of energy and protein were required for both maternal needs and fetal growth. In the case of insufficient energy intake, protein requirement for fetal and mother growth was metabolized to energy (77,147). The energy of subgroups of protein intake was significantly different among subjects. The pregnant women with low protein had also low caloric intake so protein might be used as energy in these subjects. These pattern outcomes showed as a result, that low protein group tended to have low birth weight and short gestational age because maternal protein intake was related to intrauterine growth or gestational duration (46). The moderate protein intake had energy intake more than 75% RDA. The increase of

protein intake indicated the increase of energy intake. Several studies found that fetal growth improved significantly with increasing maternal caloric intake (148, 149, 150).

A woman's protein needs increase during pregnancy, and these requirements must be met in order for her to produce a viable, healthy offspring. Extremely low maternal protein intakes, be low about 5-6% of calories, may be detrimental to the health of the fetus. As maternal protein intakes increase above this minimum threshold, several studies suggest that supplementation of maternal diets with protein in excess of about 25% of total calories, even in diets that are otherwise balanced and calorically adequate, may lead to declines, not continued gain, in infant birth mass (13). This phenomenon is not yet clear whether high protein intakes are deleterious to the fetus throughout the course of pregnancy or only during a particular trimester. Because the developing fetus is particularly susceptible to teratogenic substance during the first trimester, the period of embryonic organogenesis, this may also be the time in a pregnancy when excessive protein intake would be most problematic. However, no one has yet examined these practices to determine whether they are in effect throughout the course of a pregnancy or only during a particular trimester. Despite the many uncertainties, these factors together provide very tentative evidence that high levels of protein may be deleterious to the fetus primarily or exclusively during the first trimester of pregnancy (15). This study tried to determine the effect of protein intake of pregnant women during the second and third trimesters on fetal outcomes. Therefore, protein intake was classified into 3 levels, i.e., low (<51g), moderate (51-100g) and high (>100g) using Thai RDA as well as exiting literatures for high protein intake which affect fetal outcomes. Regardless of the high cut-off point of high protein which was 200% of Thai RDA, results showed a different pattern

among the protein intake groups. If the cut-off point of protein intake was less than 100 g, the different of the pattern would not be found.

From the literature review, the pregnant women who had low weight gain in the first two trimesters and consumed high protein intake in the third trimester had a negative effect on fetal outcomes as presented in Hypothesis 1 in this study. The following findings were reported:

### **Birth weight**

Total protein intake in both trimesters did not correlate with birth weight but vegetable protein intake in the second trimester was found to correlate positively with birth weight. However, when energy intake in the second trimester was controlled, no unique effect of vegetable protein on birth weight was found. The redundancy of the effect of these two variables on birth weight might be due to the fact that source of vegetable protein comes from cereal which is also the main source of energy.

From the regression analysis (Model 4) prepregnancy weight, changing vegetable protein intake between the second and third trimesters, gestational age and total weight gain were the variables which significantly explained the birth weight variance in Regression Model 4. Several studies ( 46, 71, 80, 81, ) found that all these variables, except for changing vegetable protein intake between the second and third trimesters, correlated positively with birth weight. On the contrary, Negger (143) found that the increase in energy and other nutrient intakes during pregnancy had no association with the increase in infant birth weight.

After controlling for the effect of other variables in the model, changing vegetable protein intake between the second and third trimesters correlated positively

with birth weight. It indicated that pregnant women who consumed high vegetable protein intake in the second trimester and less vegetable protein in the third trimester delivered an infant with higher birth weight. Since the correlation between vegetable protein and animal protein intake in the third trimester was 0.231( $p < 0.05$ ), the reduction of not only vegetable protein and total protein in the third trimester compared to that of the second trimester might also be the reason for improving the infant's birth weight. The amount of protein intake (preferably vegetable protein) in the second trimester might have set a high threshold of the placenta for acid loading effect. A reduction of serum amino acid in the third trimester might have helped reduce the rate of deterioration of placenta function before delivery. This finding appeared to contradict an earlier report (57,141) that meat intake, a high protein food group, was associated with increasing birth weight.

### **Birth length**

Total protein and animal protein intake in the third trimester had a significant negative correlation with birth length as demonstrated in the correlation matrix. But variables which best explained the variance of birth length including prepregnancy weight, changing vegetable protein intake between the second and third trimesters, animal protein intake in the third trimester and total weight gain (Regression Model 3). This finding showed that prepregnancy weight and total weight gain each had a unique effect on birth length, consistently with the results found in many studies (72, 80, 81).

Regression Model 3 showed that changing vegetable protein intake between the second and third trimesters had positive correlation with birth length; in other words,

the higher vegetable protein intake and the less vegetable protein intake in the third trimester, the higher was birth length. The effect of changing vegetable protein intake between the second and third trimesters on birth length might be explained in the same manner as its effect on birth weight.

Animal protein intake in the third trimester was one of the variables in the regression model but the result showed that the higher of animal protein in the third trimester, the shorter was birth length. On the basis of theory, calcium was the important variable that correlated with birth length as shown in Regression Model 1. For calcium intake in the third trimester in Regression Model 4, its negative effect was lost in the multiple regression due to a redundancy with the effect of other variables, which likely to be that of animal protein intake in the third trimester. This effect might be due to the fact that the resource of calcium comes from milk, small fish, etc. which is also the main source of animal protein. The negative effect of a calcium intake and animal protein in the third trimester on birth length were an unexpected finding and opposed to those found by Kanithsen (56), who studied pregnant women during the third trimester. In that study, calcium and animal protein intake showed positive correlation with birth length whereas phosphorus showed a classical suppression effect on the correlation between calcium and birth length. However, at the time of that study, milk drinking was not a habit for pregnant women so the calcium: phosphorus ratio might not be suitable for calcium retention in the body. In the present study milk drinking became a habit during the third trimester of pregnant women. The appropriate ratio of calcium: phosphorus in food might be the reason for phosphorus to lose its significance on birth length. At the same time the amount of protein intake of pregnant women over 10 years ago was not great enough to induce

high serum acidity. In the present study, subjects consumed a large amount of animal protein which induced high acidity and resulted in higher excretion of calcium in the urine. Various studies (151,152,153,154) found that animal diet had been hypothesized to increase calcium requirement because of the increase in metabolic acid products due to sulfur-containing amino acid. This acid loading leads to increase urinary calcium losses and resorption. The vegetable diet tends to have alkaline ash residue despite the presence of sulfur-containing amino acid, it would be predicted to produce lower urinary calcium loss.

With results of multiple regression, the partial correlation of total weight gain was larger than simple correlation. This effect might be due to the net suppression (155) of calcium intake or animal protein intake in the third trimester with an effect on the total weight gain as shown in Regression Models 1,3 and 4. (Total weight gain alone correlated 0.240 with birth length, but in the multiple regression analysis its partial correlation was 0.270 and 0.267 in Regression Model 3 and 1). Total weight gain correlated with calcium at  $r = 0.213$  ( $p < 0.05$ ) but did not significantly correlate with animal protein intake ( $r = 0.126$ ) while calcium and animal protein intake correlated at  $r = 0.660$  ( $p < 0.05$ ). This finding showed that animal protein intake might exert its effect on birth length through its correlation with calcium intake so that total weight gain would induce even greater birth length if either calcium or animal protein intake in the third trimester was taken into consideration.

### **Gestational age**

The result indicated that the high total protein intake especially animal protein in the second trimester increased gestational age when controlling for energy intake.

From the multiple Regression Model 1, energy intake, total protein intake in the second trimester and rate of weight gain in the third trimester were the variables included in the model. Simple correlation showed that energy intake in the second trimester did not correlate with gestational age but when it was added in the model, it made the variance of gestational age, explained by total protein intake in the second trimester, become significant. This classical suppression (155) was due to the relationships between both variables. Theoretically, protein can be converted in the body to be used as energy whenever the energy inadequacy occurs. When the energy intake was controlled, the unique effect of total protein intake on gestational age was enhanced. This phenomenon explained the change of finding in this study: after energy intake in the second trimester was controlled, the partial correlation of total protein intake increased.

From this finding, it could be hypothesized that pregnant women who consumed high protein intake during the second trimester had higher threshold for the effect of amino acid on placenta so that in the third trimester the placenta could endure higher loading acidity than those who consumed less protein in the second trimester. Therefore, there was no premature degeneration of the placenta which resulted in no premature labor. On the contrary, the pregnant women who consumed low animal protein intake in the second trimester and increased high protein intake in the third trimester, the gestational age tended to be shorter due to acid loading effect on the placenta.

The pregnant women of low protein intake in both trimesters will give birth to lower birth weight, birth length including short gestational age compared with other groups as shown in Hypothesis 2

The pregnant women who consumed low protein in both trimesters tended to deliver an infant with lower birth weight and shorter gestational age than other groups in the present study. However, they delivered an infant with longer birth length than other groups. The similar finding (46) of birth weight and gestational age was reported that low protein and energy intake were found to delivery low birth weight or premature deliver but the contrary was found for birth length. This may be due to less calcium loss, which occurred when they consumed low protein intake so the group tended to have longer birth length.

With this reason, low protein intake in both trimesters had longer birth length than other groups because of the threshold setting of placenta for acid loading effect. The similar serum amino acid in both trimesters, the placenta could properly function. Furthermore, the increase of protein intake in the third trimester might increase amino acid with an effect on placenta threshold. The increase rate of deterioration of placenta might arise due to the poor function; related to the reduction of nutrients transferring including calcium so birth length did not increase.

Although the low-low protein intake group tended to deliver infant with the lowest birth weight and gestational age, the average birth weight was still equal or more than 3000 gram but some of them were preterm delivery (<259 days). However, no significant difference from that of other groups was found, because low protein intake level was classified with protein less than 51 g which was the cut off point of 100% Thai Recommended Dietary Allowance for protein intake in pregnant women. The similar finding by Adams (64) was that the average energy and protein intake in high protein group was much higher than low protein but the mean birth weight was similar. Nevertheless, a trend of increased birth weight with higher protein intake was

observed. Birth mass may also decline when mother's total caloric intake was restricted but the decline appeared to be most extreme when the diet was low in both energy and protein. However, low protein intake in the second trimester and high protein intake in the third trimester tended to have short gestational age, which corresponded to Hypothesis 3. From this finding, the pregnant women in the low-moderate and moderate-high groups tended to have short gestational age than other groups. On the contrary, the moderate-low and high-moderate tended to have longer gestational age. Multiple Regression Model 3 of gestational age confirmed this hypothesis that if the subjects consumed the same amount of animal protein in the third trimester, the unique effect of changing total protein intake between the second trimester and third trimesters was enhanced. Therefore, the higher total protein intake in the second trimester and the less total protein intake in the third trimester, the longer was gestational age. From the observation on animal experiments (14), the high protein diet led to premature infant among the group on the high protein diet. Another study (13) supported that high protein diet led to maternal weight gain; this weight gain was not incorporated into birth weight. Furthermore, deliveries were premature among the group on the high protein diet.

Not only the quantity but also the quality of protein intake were considered. The quality of protein intake would be discussed in terms of animal protein and vegetable protein ratio, and source of protein intake. The animal protein was generally accepted as the good quality protein. From the findings, few subjects were classified in low quality protein group. Pregnant women in this study consumed high protein intake (approximately 140% Thai RDA) and approximately 80% RDA was from animal source. No significant difference among the quality protein intake groups was found.

Considering protein intake from source of protein, the higher infants' birth weight and birth length occurred along with higher vegetable intake in the second trimester. In addition, the higher animal protein intake in the third trimester, the shorter was birth length.

From these findings, together with finding in the first part of analysis, it should be emphasized that the undesirable effect of protein intake on fetal outcomes occurred only when the amount of protein intake in the third trimester was relatively closer to higher than that of the second trimester. This result added more information to Hypothesis 3: high protein intake should be aimed for during the second trimester. Furthermore, for higher birth weight and birth length, the kind of protein recommended in the second trimester was vegetable protein, the finding of which rejected Hypothesis 4. However, the low protein intake in both trimesters was the lowest birth weight and gestational age which supported Hypothesis 2.

## CHAPTER VI

### CONCLUSION AND RECOMMENDATION

#### CONCLUSION

This study was aimed to determine the effect of quantity and quality of protein intake during the second and third trimesters in pregnant women with low weight gain before the 20<sup>th</sup> week of gestation. The study design was prospective cohort study. An interview of dietary intake was conducted during the second and third trimesters at Maternal and Child Hospital, Khon Kaen Regional Hospital and Srinakarin Hospital, Khon Kaen province. The maternal socioeconomic, health-related information and delivery data were collected using an interview and also from medical record. The interview of dietary intake was employed with food frequency questionnaire, 24-hour dietary recalls and food records respectively. Gestational weight gain was collected from the initial pregnancy until delivery.

Considering demographic characteristics of pregnant women, the majority of them completed primary education, age between 20-30 years old, family income less than 7500 baht per month and 40% were housewives. No significant differences were found among various socioeconomic characteristics.

For obstetric factors, about 60% of subjects had prepregnancy weight between 45-55 kg. The majority of them were in 18.5-24.9 kg/m<sup>2</sup> of prepregnancy Body Mass Index (BMI). Approximately 56% of subjects had properly total weight gain. There were significant differences of birth weight, birth length among various prepregnancy

weight levels. Only birth weight with significant difference was found for total weight gain.

Considering protein intake with socioeconomic characteristics, it showed that the significant difference existed between animal protein intake in the third trimester over family income and vegetable protein intake in the second trimester over occupation.

The protein intake was classified into 3 levels: low (<51g), moderate (51-100 g) and high (>100 g). Mean birth weight and birth lengths were not significantly different among the different protein intake levels in both trimesters except for gestational age. However, for overall analysis, the protein intake levels were significantly different among all dependent variables. The women who had low-low protein intake tended to give birth of lowest birth weight and shortest gestational age but longest birth length. In accordance with gestational age, protein intakes shifting from lower to higher protein intake tended to have short gestational age while protein intake shifting from higher to lower protein intake tended to have long gestational age. The percentage of caloric intake from protein and quality of protein intake were not significantly different for fetal outcomes.

From multiple regression analysis, prepregnancy weight and total weight gain correlated positively with birth weight and birth length. In order to improve birth weight and birth length, pregnant women take high vegetable protein intake in the second trimester and then less vegetable protein in the third trimester. High animal protein intake in the third trimester should be avoided in order to improve birth length. When controlling for energy intake in the second trimester, the high total protein intake in the second trimester increased gestational age.

## **RECOMMENDATION**

### **I. Recommendation for the application of the study results**

1. Early antenatal clinic attendance should be encouraged for pregnant women to promote their food intake in order to achieve proper weight gain from the first trimester throughout pregnancy.

2. Low protein intake in this study was classified as less than 51g which was the cut-off point of 100% Thai RDA. The majority of pregnant women still yield proper fetal outcomes but the pregnant women who consumed protein less than 51 g in both trimesters had tendency to give birth with low birth weight and shorter gestational age than other groups. In order to have better fetal outcomes, the pregnant women are recommended to consume high protein (>100 g) in the second trimester and then decrease to moderate level (51-100g) in the third trimester. From the source of protein intake, the ratio of animal protein to vegetable protein should be 1:2. The example for food consumption per day is demonstrated in Appendix C.

3. Pregnant women who had low protein intake in the second trimester are advised against abruptly high protein intake in the third trimester. Their low threshold to acid loading might increase the rate of deterioration of placenta function. It is advised that they gradually increase protein intake with emphasis on vegetable protein intake while maintain adequate amount of animal protein to prevent essential amino acid deficiency.

4. The previous Thai RDA for pregnant women on equal amount of protein intake throughout pregnancy should be adjusted by increasing the quantity of protein intake in the second trimester rather than other trimesters.

## **II. Recommendation for future studies**

1. For the relevant data, the prospective study from the first trimester until delivery should be done if possible.

2. Both normal and low weight gain pregnant women during the first trimester should be compared.

3. The selected subjects should be generalized in various socioeconomic characteristics for better representing the population.

4. The intervention of high protein intake in the second trimester and then less protein intake in the third trimester in pregnant women is recommended for further research.

5. Regular antenatal care clinics until delivery, this issue should be aware for drop out rate in follow-up study.

6. From dietary intake, the pregnant women do not keep the same level of protein and shift to another group. The duration of sample collection should be longer and cover more areas than this study so that sample size will be large enough for categorizing protein intake in many levels for better interpretation of finding.

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### APPENDIX A

**Table 1** Minimum sample size needed to ensure the power  $1-\beta = 0.95$

No. of Levels	$\Delta\sigma$															
	1.0								1.25							
	$\alpha$								$\alpha$							
	.5	.4	.3	.25	.2	.1	.05	.01	.5	.4	.3	.25	.2	.1	.05	.01
2	11	13	15	16	18	23	27	38	7	9	10	11	12	15	18	25
3	13	16	18	20	22	27	32	43	9	10	12	13	14	18	21	29
4	15	18	21	22	25	30	36	47	10	12	14	15	16	20	23	31
5	16	19	23	25	27	33	39	51	11	13	15	16	18	22	25	33
6	18	21	24	26	29	35	41	53	12	14	16	17	19	23	27	35
7	19	22	26	28	30	37	43	56	12	14	17	18	20	24	28	36
8	20	23	27	29	32	39	45	58	13	15	18	19	21	25	29	38
9	21	24	28	30	33	40	47	60	14	16	18	20	22	26	30	39
10	21	25	29	32	34	42	48	62	14	16	19	21	22	27	31	40
11	22	26	30	33	36	43	50	64	15	17	20	21	23	28	33	42
13	24	28	32	35	38	46	53	68	15	18	21	23	25	30	34	44
16	26	30	35	38	41	49	57	*	17	20	23	24	26	32	37	*
21	29	33	39	42	45	55	63	*	19	22	25	27	29	35	41	*
25	31	36	42	45	48	61	66	*	20	23	27	29	31	39	43	*
31	34	40	46	49	53	63	71	*	22	26	30	32	34	41	46	*

No. of Levels	$\Delta\sigma$															
	1.5								1.75							
	$\alpha$								$\alpha$							
	.5	.4	.3	.25	.2	.1	.05	.01	.5	.4	.3	.25	.2	.1	.05	.01
2	5	6	7	8	9	11	13	18	4	5	6	6	7	8	10	14
3	6	7	9	9	10	13	15	20	5	6	7	7	8	10	12	16
4	7	8	10	11	12	14	17	22	6	6	7	8	9	11	13	17
5	8	9	11	11	13	15	18	23	6	7	8	9	10	12	14	18
6	8	10	11	12	13	16	19	25	6	7	9	9	10	12	14	19
7	9	10	12	13	14	17	20	26	7	8	9	10	11	13	15	19
8	9	11	13	14	15	18	21	27	7	8	10	10	11	14	16	20
9	10	11	13	14	15	19	22	28	7	9	10	11	12	14	16	21
10	10	12	14	15	16	19	22	29	8	9	10	11	12	15	17	21
11	10	12	14	15	17	20	23	29	8	9	11	11	12	15	17	22
13	11	13	15	16	17	21	24	31	8	10	11	12	13	16	18	23
16	12	14	16	17	19	23	26	*	9	10	12	13	14	17	19	*
21	13	15	18	19	21	25	29	*	10	12	13	14	15	19	21	*
25	14	17	19	21	22	28	30	*	11	12	14	15	17	21	23	*
31	16	18	21	22	24	29	33	*	12	14	16	17	18	21	24	*

Table 1 Minimum sample size needed to ensure the power  $1-\beta = .95$  (continued)

No. of Levels	$\Delta/\sigma$															
	2.0								2.5							
	$\alpha$								$\alpha$							
	.5	.4	.3	.25	.2	.1	.05	.01	.5	.4	.3	.25	.2	.1	.05	.01
2	3	4	4	5	5	7	8	11	2	3	3	4	4	5	6	8
3	4	5	5	6	6	8	9	12	3	3	4	4	5	6	7	9
4	4	5	6	6	7	9	10	13	3	4	4	5	5	6	7	9
5	5	6	6	7	8	9	11	14	3	4	5	5	6	7	9	10
6	5	6	7	7	8	10	11	15	4	4	5	5	6	7	8	10
7	5	6	7	8	8	10	12	15	4	4	5	5	6	7	8	10
8	6	7	8	8	9	11	12	16	4	5	5	6	6	7	8	11
9	6	7	8	8	9	11	13	16	4	5	5	6	6	8	9	11
10	6	7	8	9	9	11	13	17	4	5	6	6	6	8	9	11
11	6	7	8	9	10	12	14	17	4	5	6	6	7	8	9	12
13	7	8	9	10	10	12	14	18	5	5	6	7	7	8	10	12
16	7	8	10	10	11	13	15	*	5	6	6	7	7	9	10	*
21	8	9	10	11	12	15	17	*	5	6	7	8	8	10	11	*
25	8	10	11	12	13	16	18	*	6	7	8	8	9	11	12	*
31	9	11	12	13	14	17	19	*	6	7	8	9	9	11	13	*

No. of Levels	$\Delta/\sigma$							
	3.0							
	$\alpha$							
	.5	.4	.3	.25	.2	.1	.05	.01
2	2	2	3	3	3	4	5	6
3	2	3	3	3	4	4	5	7
4	3	3	3	4	4	5	5	7
5	3	3	4	4	4	5	6	7
6	3	3	4	4	4	5	6	8
7	3	3	4	4	4	5	6	8
8	3	4	4	4	5	5	6	8
9	3	4	4	4	5	6	6	8
10	3	4	4	5	5	6	7	8
11	3	4	4	5	5	6	7	9
13	4	4	5	5	5	6	7	9
16	4	4	5	5	6	7	8	*
21	4	5	5	6	6	7	8	*
25	4	5	6	6	6	8	9	*
31	5	5	6	6	7	8	9	*

\* Values not computed

Source: Bratcher TL, et al.(140) Table of sample sizes in the analysis of variance. J quality technology 1970;2(3): 160.

**Table 2** Frequency of food intake of the subjects during the second and third trimesters

Food items	everyday %(N)	4-6 times/week %(N)	1-3 times/week %(N)	<1 time/week %(N)	not consumed %(N)	food frequency score
<b>1. Meat, poultry, fish, and other products</b>						
Beef	5.9(10)	3.0(5)	47.6(81)	17.6(30)	25.9(44)	23.5
Pork	28.8(49)	10.6(18)	51.8(88)	2.9(5)	5.9(10)	51.9
Poultry	19.4(33)	7.7(13)	67.0(114)	1.2(2)	4.7(8)	45.0
Fresh-water fish	26.4(45)	10.0(17)	54.1(92)	4.2(7)	5.3(9)	49.9
Marine products	0.6(1)	-	41.8(71)	6.5(11)	51.1(87)	2.6
Mackerel	11.1(19)	6.5(11)	69.4(118)	4.2(7)	8.8(15)	36.8
Egg	44.7(76)	12.3(21)	36.5(62)	2.4(4)	4.1(7)	64.4
Dried-fish /squid	3.0(5)	-	54.7(93)	7.6(13)	34.7(59)	19.9
Thai sausage	2.4(4)	1.2(2)	38.2(65)	2.4(21)	45.8(78)	15.6
Meatball	10.6(18)	5.9(10)	56.5(96)	7.6(13)	19.4(33)	32.1

Table 2 Frequency of food intake of the subjects during the second and third trimesters (continued.)

Food items	everyday %(N)	4-6 times/week %(N)	1-3 times/week %(N)	<1 time/week %(N)	not consumed %(N)	food frequency score
Meat organ	4.1(17)	4.1(7)	55.3(94)	8.2(14)	28.3(48)	24.2
<b>2. Legumes and their products</b>						
Peanut	7.1(12)	2.9(5)	52.4(89)	14.7(25)	22.9(39)	25.8
Soybean curd	0.6(1)	-	8.8(15)	2.4(4)	88.2(150)	3.4
Soybean milk(UHT)	7.1(12)	2.4(4)	20.6(35)	3.0(5)	67.1(114)	15.4
Soybean milk	15.9(27)	2.9(5)	18.3(31)	4.1(7)	58.8(100)	23.7
<b>3. Cereals, grain products, starchy roots and tubers</b>						
Glutinous rice	89.4(152)	1.2(2)	5.9(10)	-	3.5(6)	92.0
Ordinary rice	25.9(44)	5.9(10)	48.8(83)	11.1(19)	8.3(14)	45.4
Noodle	7.1(12)	1.8(3)	55.2(94)	16.5(28)	19.4(33)	26.2
Kanom jean	11.7(20)	7.1(12)	55.9(95)	11.7(20)	13.6(23)	34.2

**Table 2** Frequency of food intake of the subjects during the second and third trimesters (continued.)

Food items	everyday %(N)	4-6 times/week %(N)	1-3 times/week %(N)	<1 time/week %(N)	not consumed %(N)	food frequency score
Instant noodle	1.2(2)	1.2(2)	34.7(59)	19.4(33)	43.5(74)	14.0
Mungbean noodle	1.8(3)	2.4(4)	39.4(67)	18.2(31)	38.2(65)	16.6
Bread	5.9(10)	4.7(8)	30.0(51)	3.9(10)	53.5(91)	18.5
Sponge cake	2.4(4)	3.0(5)	34.1(58)	10.0(17)	50.5(86)	15.4
Cracker	2.9(5)	1.8(3)	22.9(39)	6.5(11)	65.9(112)	11.6
Donut	0.6(1)	1.2(2)	23.9(40)	7.6(13)	67.1(114)	9.1
Taro/sweet potato	1.8(3)	4.1(7)	45.9(78)	9.4(16)	38.8(66)	19.2
Corn	10.6(18)	4.7(8)	57.7(98)	14.1(24)	12.9(22)	32.1
<b>4. Vegetables</b>						
Green vegetable	52.3(89)	9.4(16)	36.5(62)	1.2(2)	0.6(1)	69.9
Other vegetables	42.9(93)	44.7(76)	7.6(13)	2.4(4)	2.4(4)	76.8
Pumpkin	4.1(7)	2.4(4)	47.6(81)	17.1(29)	28.8(49)	21.3

Table 2 Frequency of food intake of the subjects during the second and third trimesters (continued.)

Food items	everyday %(N)	4-6 times/week %(N)	1-3 times/week %(N)	<1 time/week %(N)	not consumed %(N)	food frequency score
Papaya salad	29.4(50)	7.7(13)	40.6(69)	4.1(7)	18.2(21)	47.3
<b>5. Fruits</b>						
Fruits	43.5(74)	5.3(9)	48.2(82)	3.0(5)	-	61.9
<b>6. Fat and oils</b>						
Oil	43.5(74)	8.2(14)	41.8(71)	3.6(6)	2.9(5)	6.2
Coconut milk	7.1(12)	2.9(5)	33.0(56)	7.6(13)	49.4(84)	19.5
<b>7. Dessert</b>						
Dessert	10.6(18)	4.7(8)	47.1(80)	8.8(15)	28.8(49)	28.6
Ice cream	15.2(26)	6.5(11)	52.4(89)	3.0(5)	22.9(39)	35.7
<b>8. Beverage</b>						
Carbonated Beverage	6.5(11)	1.8(3)	37.0(63)	11.8(20)	42.4(73)	35.7
Fruit juice/Soft drink	9.4(16)	4.7(8)	41.2(70)	7.6(13)	37.1(63)	19.7

**Table 2** Frequency of food intake of the subjects during the second and third trimesters (continued.)

Food items	everyday %(N)	4-6 times/week %(N)	1-3 times/week %(N)	<1 time/week %(N)	not consumed %(N)	food frequency score
Coconut juice	7.1(12)	51.2(87)	2.9(5)	18.2(31)	20.6(35)	45.1
<b>9. Milk and their products</b>						
Whole milk(UHT)	47.6(81)	3.0(5)	32.9(56)	0.6(1)	15.9(27)	59.6
Drinking yogurt	9.4(16)	1.8(3)	37.6(64)	1.8(3)	49.4(84)	22.1
Milo /Ovaltine	14.7(25)	3.5(6)	18.8(32)	1.8(3)	61.2(104)	22.9

Table 3 Food consumption quantity per meal

Food items	2 cups %(N)	1 1/2 cups %(N)	1 cup %(N)	3/4 cup %(N)	1/2 cup %(N)	1/3 cup %(N)	1/4 cup %(N)	not consumed %(N)
<b>1. Meat, poultry, fish and their products</b>								
Beef	-	-	1.8(3)	1.8(3)	35.9(61)	13.5(23)	21.2(36)	25.9(44)
Pork	-	-	2.4(4)	2.4(4)	44.7(76)	17.6(30)	27.1(46)	5.9(10)
Poultry	-	0.6(1)	4.7(8)	0.6(1)	55.9(95)	12.4(21)	21.2(36)	4.7(8)
Fresh-water fish	-	-	11.8(20)	1.2(2)	46.5(79)	15.3(26)	20.0(34)	5.3(9)
Marine products	-	-	7.1(12)	-	25.9(44)	7.1(12)	8.8(15)	51.1(87)
Dried fish / squid	-	-	2.9(5)	1.2(2)	8.8(15)	22.9(39)	29.4(50)	34.7(59)
Thai sausage	-	-	2.4(4)	0.6(1)	16.5(28)	4.7(8)	30.0(51)	45.8(78)
Meatball	-	1.2(2)	9.4(16)	9.4(16)	15.9(27)	10.6(18)	34.1(58)	19.4(33)
Meat organ	-	-	6.5(11)	2.4(4)	22.4(38)	20.0(34)	20.6(35)	28.2(48)

**Table 3** Food consumption quantity per meal (continued.)

Food item	3 pieces %(N)	2 pieces %(N)	1½ pieces %(N)	1 piece %(N)	½ piece %(N)	not consumed %(N)		
Egg	1.2(2)	25.3(43)	-	69.4(118)	-	4.1(7)		
Mackerel	-	11.8(20)	1.8(3)	68.8(117)	8.8(15)	8.8(15)		
Food items	2cups %(N)	1½cups %(N)	1cup %(N)	¾cup %(N)	½cup %(N)	⅓cup %(N)	¼cup %(N)	not consumed %(N)
<b>2. Legumes and their products</b>								
Peanut	-	-	2.4(4)	2.4(4)	48.2(82)	11.2(9)	12.9(22)	22.9(39)
Soybean curd	-	-	-	1.8(3)	5.3(9)	1.8(3)	2.9(5)	88.2(150)
Soybean milk	-	-	41.2(70)	-	-	-	-	58.8(100)
Food items	250 cc.	200 cc.	not consumed					
Soybean milk(UHT)	32.9(56)	-	-	67.1(114)				

Table 3 Food consumption quantity per meal (continued.)

Food items	≥2cups %(N)	1½cups %(N)	1cup %(N)	¾cup %(N)	½cup %(N)	⅓cup %(N)	¼cup %(N)	not consumed %(N)
<b>3. Cereals, grain products, starchy roots and tubers</b>								
Glutinous rice	18.8(32)	15.9(27)	48.8(83)	0.6(1)	12.4(21)	-	-	3.5(6)
Ordinary rice	44.7(76)	5.9(10)	35.3(60)	-	4.7(8)	0.6(1)	0.6(1)	8.2(14)
Noodle	9.4(16)	0.6(1)	64.7(110)	-	-	0.6(1)	5.3(9)	19.4(33)
Kanom jean	10.0(17)	33.5(57)	40.6(69)	-	2.4(4)	-	-	13.5(23)
Mungbean noodle	6.5(11)	-	26.5(45)	-	24.7(42)	1.8(3)	2.4(4)	38.2(65)
Sponge cake	-	1.8(3)	14.1(24)	-	33.5(57)	-	-	50.6(86)
Cracker	-	-	7.1(12)	-	18.2(31)	2.4(4)	6.5(11)	65.9(112)
Taro/sweet potato	2.4(4)	-	26.5(45)	-	23.5(40)	3.5(6)	5.3(9)	38.8(66)

**Table 3** Food consumption quantity per meal (continued.)

	≥4 pieces	3 pieces	2 pieces	1 piece	not consumed
	%(N)	%(N)	%(N)	%(N)	%(N)
Instant noodle	-	-	2.9(5)	53.5(91)	43.5(74)
Bread	2.9(5)	3.5(6)	30.0(51)	10.0(17)	53.5(91)
Corn	6.5(11)	11.2(19)	32.4(55)	37.1(63)	12.9(22)
Donut	2.3(4)	4.1(7)	10.6(18)	15.9(27)	67.1(114)

	≥2cups	1 1/2 cups	1 cup	3/4 cup	1/2 cup	1/3 cup	1/4 cup	not consumed
	%(N)	%(N)	%(N)	%(N)	%(N)	%(N)	%(N)	%(N)
<b>Food items</b>								
<b>4. Vegetables</b>								
Green vegetable	4.7(8)	1.8(3)	50.6(86)	0.6(1)	37.6(64)	2.4(4)	1.8(3)	0.6(1)
Other vegetables	6.5(11)	-	63.5(108)	0.6(1)	21.2(36)	2.9(5)	2.9(5)	2.4(4)
Pumpkin	4.9(10)	-	39.4(67)	-	21.8(37)	0.6(1)	3.5(6)	28.8(49)
Papaya salad	1.8(3)	-	32.4(55)	1.8(3)	28.8(49)	4.1(7)	12.9(22)	18.2(31)

Table 3 Food consumption quantity per meal (continued.)

Food items	≥1cup %(N)	<sup>3</sup> / <sub>4</sub> cup %(N)	<sup>1</sup> / <sub>2</sub> cup %(N)	<sup>1</sup> / <sub>3</sub> cup %(N)	<sup>1</sup> / <sub>4</sub> cups %(N)	not consumed %(N)
<b>5. Fruits</b>						
Fruits	88.3(150)	-	9.4(16)	-	2.4(4)	-
Food items	3 tea spoons %(N)	2 tea spoons %(N)	1 tea spoons %(N)	1 tea spoon %(N)	not consumed %(N)	not consumed %(N)
<b>6. Fat and oil</b>						
Oil	14.1(24)	20.6(35)	62.4(106)	2.9(5)		
Food items	1cups %(N)	<sup>3</sup> / <sub>4</sub> cups %(N)	<sup>1</sup> / <sub>2</sub> cups %(N)	<sup>1</sup> / <sub>3</sub> cups %(N)	<sup>1</sup> / <sub>4</sub> cups %(N)	not eat %(N)
Coconut milk	4.7(8)	-	35.9(61)	2.4(4)	7.6(13)	49.4(84)
<b>7. Dessert</b>						
Dessert	62.4(106)	-	0.6(1)	8.2(14)	-	28.8(49)

**Table 3** Food consumption quantity per meal (continued.)

Food items	2cups %(N)	1½cups %(N)	1cup %(N)	¾cup %(N)	½cup %(N)	⅓cup %(N)	¼cup %(N)	not consumed %(N)
Ice cream	-	0.6(1)	4.1(7)	-	72.4(123)	-	-	22.9(39)
<b>8. Beverage</b>								
Fruit juice/soft drink	-	-	61.8(105)	-	1.2(2)	-	-	37.1(63)
Coconut juice	5.3(9)	0.6(1)	60.0(102)	0.6(1)	9.4(16)	2.9(5)	0.6(1)	20.6(35)
Food items	1 bottle %(N)	1 bottle %(N)	½ bottle %(N)	not consumed %(N)				
Carbonated beverage	41.2(70)	15.9(27)	42.9(73)	not consumed				
Food items	250 cc. %(N)	145 cc. %(N)	not consumed %(N)					
<b>9. Milk and milk products</b>								
Whole milk (UHT)	82.4(140)	1.8(3)	15.9(27)	15.9(27)	15.9(27)	15.9(27)	15.9(27)	15.9(27)

**Table 3** Food consumption quantity per meal (continued.)

Food items	1 bottle %(N)	not consumed %(N)
Drinking yogurt	50.6(86)	49.4(84)
Food items	1 cups %(N)	not consumed %(N)
Milo/ovaltin	38.8(66)	61.2(104)

**Table 4** Calculation method of food frequency score

From Table 2, Appendix A, the food frequency in each food item could be calculated for food frequency score as follows:

	everyday	4-6 times/week	1-3 times/week	<1 time/month	not consumed
<b>Step 1</b>					
Average food frequency					
per unit	7 times/week	$\frac{4+6}{2} = 5$ times/week	$\frac{1+3}{2} = 2$ times/week	$\frac{1+3}{2} = 2$ times/month	0
<b>Step 2</b>					
Average food frequency					
per day	$\frac{7}{7} = 1$	$\frac{5}{7} = 0.7$	$\frac{2}{7} = 0.3$	$\frac{2}{7} = 0.03$	0
<b>Step 3</b>					
Multiply the average food frequency per day by the	$1x(\%) = A$	$0.7x(\%) = B$	$0.3x(\%) = C$	$0.03x(\%) = D$	0

percentage of the subjects in  
 each column of time,  
 for example (beef)  $1 \times 5.9 = 5.9$        $0.7 \times 3.0 = 2.1$        $0.3 \times 47.6 = 14.3$        $0.03 \times 17.6 = 1.2$       0

**Step 4**

Sum of total food frequency

For example (beef)

$$A + B + C + D = F$$

$$5.9 + 2.1 + 14.3 + 1.2 = 23.5$$

**Table 5** Multiple comparison of birth weight, birth length and gestational age among protein intake groups in the second trimester using LSD

Dependent Variable	Protein intake		Mean Difference (I-J)	Std. Error	Sig.
	(I)	(J)			
Birth weight	1.00	2.00	-29.70	108.452	.785
		3.00	-294.02	150.397	.052
	2.00	1.00	29.70	108.452	.785
		3.00	-264.32*	115.195	.023
	3.00	1.00	294.02	150.397	.052
		2.00	264.32*	115.195	.023
Birth length	1.00	2.00	.66	.626	.295
		3.00	.64	.867	.460
	2.00	1.00	-.66	.626	.295
		3.00	-1.43E-02	.664	.983
	3.00	1.00	-.64	.867	.460
		2.00	1.43E-02	.664	.983
Gestational age	1.00	2.00	-8.72*	2.734	.002
		3.00	-9.42*	3.792	.014
	2.00	1.00	8.72*	2.734	.002
		3.00	-.70	2.904	.810
	3.00	1.00	9.42*	3.792	.014
		2.00	.70	2.904	.810

protein intake group :

1 = low

2 = moderate

3 = high

**Table 6** Multiple comparison of birth weight, birth length and gestational age among protein intake groups in the third trimester

Dependent Variable	Protein intake		Mean Difference (I-J)	Std. Error	Sig.
	(I)	(J)			
Birth weight	1.00	2.00	-101.98	113.082	.368
		3.00	-34.95	154.730	.822
	2.00	1.00	101.98	113.082	.368
		3.00	67.03	116.675	.566
	3.00	1.00	34.95	154.730	.822
		2.00	-67.03	116.675	.566
Birth length	1.00	2.00	-.38	.638	.549
		3.00	.95	.873	.277
	2.00	1.00	.38	.638	.549
		3.00	1.34*	.658	.044
	3.00	1.00	-.95	.873	.277
		2.00	-1.34*	.658	.044
Gestational age	1.00	2.00	-2.18	2.848	.446
		3.00	5.00	3.897	.201
	2.00	1.00	2.18	2.848	.446
		3.00	7.18*	2.938	.016
	3.00	1.00	-5.00	3.897	.201
		2.00	-7.18*	2.938	.016

protein intake group :

1 = low

2 = moderate

3 = high

**Table 7** Multiple comparison of birth weight , birth length and gestational age among protein intake groups in the second and third trimesters using LSD

Dependent Variable	Protein intake		Mean Difference (I-J)	Std. Error	Sig.
	(I)	(J)			
Birth weight	1.00	2.00	-100.83	237.804	.672
		4.00	-50.23	240.491	.835
		5.00	-115.46	209.348	.582
		6.00	-37.50	247.514	.880
		8.00	-500.83*	247.514	.045
		9.00	-133.50	276.303	.630
	2.00	1.00	100.83	237.804	.972
		4.00	50.61	171.932	.769
		5.00	-14.63	124.705	.907
		6.00	63.33	181.625	.728
		8.00	-400.00**	181.625	.029
		9.00	-32.67	219.244	.882
	4.00	1.00	50.23	240.491	.835
		2.00	-50.61	171.932	.769
		5.00	-65.23	129.756	.616
		6.00	12.73	185.130	.945
		8.00	-450.61*	185.130	.016
		9.00	-83.27	222.156	.708
	5.00	1.00	115.46	209.348	.582
		2.00	14.63	124.705	.907
		4.00	65.23	129.756	.616
		6.00	77.96	142.351	.585
		8.00	-385.38*	142.351	.008
		9.00	-18.04	188.000	.924
	6.00	1.00	37.50	247.514	.880
		2.00	-63.33	181.625	.728
		4.00	-12.73	185.130	.945
		5.00	-77.96	142.351	.585
		8.00	-463.33*	194.166	.018
		9.00	-96.00	229.740	.677
	8.00	1.00	500.83*	247.514	.045
		2.00	400.00*	181.625	.029
		4.00	450.61*	185.130	.016
		5.00	385.38*	142.351	.008
		6.00	463.33*	194.166	.018
		9.00	367.33	229.740	.122
9.00	1.00	133.50	276.303	.630	
	2.00	32.67	219.244	.882	
	4.00	83.27	222.156	.708	
	5.00	18.04	188.000	.924	
	6.00	96.00	229.740	.677	
	8.00	-367.33	229.740	.112	

protein intake group :

1 = low-low

2 = low-moderate

4 = moderate-low

5 = moderate-moderate

6 = moderate-high

8 = high-moderate

9 = high-high

**Table 7** Multiple comparison of birth weight, birth length and gestational age among protein intake groups in the second and third trimesters using LSD (continued)

Dependent Variable	Protein intake		Mean Difference	Std. Error	Sig.	
	(I)	(J)	(I-J)			
Birth length	1.00	2.00	.67	1.363	.625	
		4.00	1.82	1.378	.189	
		5.00	.99	1.200	.410	
		6.00	2.56	1.418	.073	
		8.00	.78	1.418	.584	
		9.00	1.80	1.583	.257	
	2.00	1.00	-.67	1.363	.625	
		4.00	1.15	.985	.244	
		5.00	.33	.715	.650	
		6.00	1.89	1.041	.071	
		8.00	.11	1.041	.915	
	4.00	1.00	-1.82	1.378	.189	
		2.00	-1.15	.985	.244	
		5.00	-.83	.743	.268	
		6.00	.74	1.061	.488	
		8.00	-1.04	1.061	.328	
	5.00	1.00	-1.82E-02	1.273	.989	
		2.00	-.99	1.200	.410	
		4.00	-.33	.715	.650	
		6.00	.83	.743	.268	
		8.00	1.56	.816	.057	
	6.00	1.00	-.21	.816	.793	
		2.00	.81	1.077	.454	
		4.00	-2.56	1.418	.073	
		5.00	-1.89	1.041	.071	
		8.00	-.74	1.061	.488	
	8.00	1.00	-1.56	.816	.057	
		2.00	-1.78	1.113	.112	
		4.00	-1.78	1.113	.112	
		5.00	-.76	1.316	.567	
		6.00	-.78	1.418	.584	
	9.00	1.00	-.11	1.041	.915	
		2.00	1.04	1.061	.328	
		4.00	.21	.816	.793	
		5.00	1.78	1.113	.112	
		6.00	1.02	1.316	.439	
	9.00	1.00	-1.80	1.583	.257	
		2.00	-1.13	1.256	.368	
		4.00	1.82E-02	1.273	.989	
		5.00	-.81	1.077	.454	
		6.00	.76	1.316	.567	
			8.00	-1.02	1.316	.439

**Table 7** Multiple comparison of birth weight, birth length and gestational age among protein intake groups in the second and third trimesters using LSD  
(continued)

Dependent Variable	Protein intake		Mean Difference (I-J)	Std. Error	Sig.
	(I)	(J)			
Gestational age	1.00	2.00	-11.92*	5.792	.041
		4.00	-21.14*	5.857	.000
		5.00	-18.07*	5.099	.001
		6.00	-7.83	6.029	.196
		8.00	-20.06*	6.029	.001
		9.00	-15.30*	6.730	.024
	2.00	1.00	11.92*	5.792	.041
		4.00	-9.22*	4.188	.029
		5.00	-6.16*	3.037	.044
		6.00	4.08	4.424	.357
		8.00	-8.14	4.424	.068
		9.00	-3.38	5.340	.527
	4.00	1.00	21.14*	5.857	.000
		2.00	9.22*	4.188	.029
		5.00	3.06	3.160	.334
		6.00	13.30*	4.509	.004
		8.00	1.08	4.509	.811
		9.00	5.84	5.411	.282
	5.00	1.00	18.07*	5.099	.001
		2.00	6.16*	3.037	.044
		4.00	-3.06	3.160	.334
		6.00	10.24*	3.467	.004
		8.00	-1.98	3.467	.569
		9.00	2.77	4.579	.545
	6.00	1.00	7.83	6.029	.196
		2.00	-4.08	4.424	.357
		4.00	-13.30*	4.509	.004
		5.00	-10.24*	3.467	.004
		8.00	-12.22*	4.729	.011
		9.00	-7.47	5.596	.184
	8.00	1.00	20.06*	6.029	.001
		2.00	8.14	4.424	.068
		4.00	-1.08	4.509	.811
		5.00	1.98	3.467	.569
		6.00	12.22*	4.729	.011
		9.00	4.76	5.596	.397
	9.00	1.00	15.30*	6.730	.024
		2.00	3.38	5.340	.527
		4.00	-5.84	5.411	.282
		5.00	-2.77	4.579	.545
		6.00	7.47	5.596	.184
		8.00	-4.76	5.596	.397

**Table 8** Multiple comparison of birth weight, birth length and gestational age among percentage of caloric intake from protein in the second and third trimesters using LSD

Dependent Variable	Energy intake form		Mean Difference (I-J)	Std. Error	Sig.
	(I)	(J)			
Birth length	1.00	2.00	.90	.485	.065
		3.00	.16	.514	.749
		4.00	.31	.489	.530
	2.00	1.00	-.90	.485	.065
		3.00	-.73	.575	.203
		4.00	-.59	.553	.286
	3.00	1.00	-.16	.514	.749
		2.00	.73	.575	.203
		4.00	.14	.578	.805
	4.00	1.00	-.31	.489	.530
		2.00	.59	.553	.286
		3.00	-.14	.578	.805
Birth weight	1.00	2.00	-2.19	86.010	.980
		3.00	8.16	91.188	.929
		4.00	26.82	86.772	.759
	2.00	1.00	2.19	86.010	.980
		3.00	10.35	101.971	.919
		4.00	29.01	98.042	.768
	3.00	1.00	-8.16	91.188	.929
		2.00	-10.35	101.971	.919
		4.00	18.66	102.615	.856
	4.00	1.00	-26.82	86.772	.758
		2.00	-29.01	98.042	.768
		3.00	-18.66	102.615	.856
Gestational age	1.00	2.00	.41	2.188	.853
		3.00	-2.73	2.320	.242
		4.00	-1.08	2.207	.627
	2.00	1.00	-.41	2.188	.853
		3.00	-3.13	2.594	.229
		4.00	-1.48	2.494	.553
	3.00	1.00	2.73	2.320	.242
		2.00	3.13	2.594	.229
		4.00	1.65	2.610	.528
	4.00	1.00	1.08	2.207	.627
		2.00	1.48	2.494	.553
		3.00	-1.65	2.610	.528

energy intake from protein group :

1 = low-low      2 = low-moderate      3 = moderate-low      4 = moderate-moderate

**Table 9** Multiple comparison of birth weight, birth length and gestational age among protein quality in the second and third trimesters using LSD

<b>Dependent Variable</b>	<b>Protein (I)</b>	<b>intake (J)</b>	<b>Mean Difference (I-J)</b>	<b>Std. Error</b>	<b>Sig.</b>
Birth weight	2.00	3.00	405.83	317.373	.203
		4.00	123.63	210.303	.557
	3.00	2.00	-405.83	317.373	.203
		4.00	-282.20	242.109	.245
	4.00	2.00	-123.63	210.303	.557
		3.00	282.20	242.109	.245
Birth length	2.00	3.00	.42	1.796	.817
		4.00	1.92	1.190	.109
	3.00	2.00	-.42	1.796	.817
		4.00	1.50	1.370	.276
	4.00	2.00	-1.92	1.190	.109
		3.00	-1.50	1.370	.276
Gestational age	2.00	3.00	9.58	8.062	.236
		4.00	-2.41	5.342	.652
	3.00	2.00	-9.58	8.062	.236
		4.00	-12.00	6.150	.053
	4.00	2.00	2.41	5.342	.652
		3.00	12.00	6.150	.053

protein quality group :

2 = poor-good

3 = good-poor

4 = good-good

**Table 10** Multiple comparison of energy intake among protein intake groups in the second and third trimesters using LSD

Dependent Variable	Protein intake		Mean Difference (I-J)	Std. Error	Sig.	
	(I)	(J)				
Energy T <sub>2</sub>	1.00	2.00	-62.2167	224.608	.782	
		4.00	-357.4048	227.146	.118	
		5.00	-699.9658*	197.731	.001	
		6.00	-903.7842*	233.779	.000	
		8.00	-1386.724*	233.779	.000	
		9.00	-1766.420*	260.970	.000	
		2.00	1.00	62.2167	224.608	.782
			4.00	-295.1881	162.391	.071
			5.00	-637.7492*	117.785	.000
	6.00		-841.5675*	171.547	.000	
	8.00		-1324.507*	171.547	.000	
	9.00		-1704.203*	207.078	.000	
	4.00	1.00	357.4048	227.146	.118	
		2.00	295.1881	162.391	.071	
		5.00	-342.5611*	122.556	.006	
		6.00	-546.3794*	174.857	.002	
		8.00	-1029.319*	174.857	.000	
		9.00	-1409.015*	209.828	.000	
	5.00	1.00	699.9658*	197.731	.001	
		2.00	637.7492*	117.785	.000	
		4.00	342.5611*	122.556	.006	
		6.00	-203.8183	134.452	.131	
		8.00	-686.7583*	134.452	.000	
		9.00	-1066.454*	177.568	.000	
	6.00	1.00	903.7842*	233.779	.000	
		2.00	841.5675*	171.547	.000	
		4.00	546.3794*	174.857	.002	
		5.00	203.8183	134.452	.131	
		8.00	-482.9400*	183.391	.009	
		9.00	-862.6353*	216.992	.000	
	8.00	1.00	1386.7242*	233.779	.000	
		2.00	1324.5075*	171.547	.000	
		4.00	1029.3194*	174.857	.000	
		5.00	686.7583*	134.452	.000	
		6.00	482.9400*	183.391	.009	
		9.00	-379.6953	219.992	.082	
	9.00	1.00	1766.4195*	260.970	.00	
		2.00	1704.2028*	207.078	.000	
		4.00	1409.0147*	209.828	.000	
		5.00	1066.4537*	177.568	.000	
		6.00	862.6353*	216.992	.000	
		8.00	379.6953	216.992	.082	

protein intake group :

1 = low-low

5 = moderate-moderate

9 = high-high

2 = low – moderate

6 = moderate-high

4 = moderate - low

8 = high-moderate

**Table 10** Multiple comparison of energy intake among protein intake groups in the second and third trimesters using LSD (continued)

Dependent Variable	Protein intake		Mean Difference (I-J)	Std. Error	Sig.
	(I)	(J)			
Energy T <sub>3</sub>	1.00	2.00	-626.8925*	229.036	.007
		4.00	-176.7464	231.624	.447
		5.00	-894.8021*	201.630	.000
		6.00	-1805.004*	238.388	.000
		8.00	-934.3744*	238.388	.000
		9.00	-1665.652*	266.116	.000
	2.00	1.00	626.8925*	229.036	.007
		4.00	450.1461*	165.593	.007
		5.00	-267.9096*	120.107	.027
		6.00	-1178.112*	174.929	.000
		8.00	-307.4819	174.929	.081
		9.00	-1038.759*	211.161	.000
	4.00	1.00	176.7464	231.624	.447
		2.00	-450.1461*	165.593	.007
		5.00	-718.0557*	124.972	.000
		6.00	-1628.258*	178.304	.000
		8.00	-757.6281*	178.304	.000
		9.00	-1488.906*	213.965	.000
	5.00	1.00	894.8021*	201.630	.000
		2.00	267.9096*	120.107	.027
		4.00	718.0557*	124.972	.000
		6.00	-910.2024*	137.103	.000
		8.00	-39.5724	137.103	.773
		9.00	-770.8499*	181.069	.000
	6.00	1.00	1805.004*	238.388	.000
		2.00	1178.1119*	174.929	.000
		4.00	1628.2581*	178.304	.000
		5.00	910.2024*	137.103	.000
		8.00	870.6300*	187.007	.000
		9.00	139.3524	221.270	.530
	8.00	1.00	934.3744*	238.388	.000
		2.00	307.4819	174.929	.081
		4.00	757.6281*	178.304	.000
		5.00	39.5724	137.103	.773
		6.00	-870.6300*	187.007	.000
		9.00	-731.2776*	221.270	.001
	9.00	1.00	1665.6520*	266.116	.000
		2.00	1038.7595*	211.161	.000
		4.00	1488.9056*	213.965	.000
		5.00	770.8499*	181.069	.000
		6.00	-139.3524	221.270	.530
		8.00	731.2776*	221.270	.001

Table 11 The correlation matrix of variables in the second trimester

Total protein	Animal protein	Vegetable Protein	Animal Vegetable Protein	Carbohydrate	Fat	Energy	Calcium	Phosphorus	Prepreg-nancy weight	Prepreg-nancy BMI	Total weight gain	Rate weight gain	Birth Weight	Birth length	Gestational age
/	/	/	/	/	/	/	/	/	/	/	/	/	/	/	/
0.913*	0.294*	0.658*	0.647*	0.344*	0.605*	0.615*	0.642*	0.628*	-0.015	-0.094	0.108	0.056	0.089	-0.072	0.131
0.658*	0.647*	0.313*	0.344*	0.925*	0.198*	0.850*	0.205*	0.312*	0.105	0.054	0.147	0.135	0.169*	0.108	-0.019
0.313*	0.647*	-0.439*	0.344*	0.925*	0.198*	0.850*	0.205*	0.312*	0.105	0.054	0.147	0.135	0.169*	0.108	-0.019
0.667*	0.344*	0.925*	-0.344*	0.249*	0.386*	0.198*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*
0.560*	0.605*	0.198*	0.386*	0.249*	0.386*	0.198*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*	0.249*
0.847*	0.615*	0.850*	-0.058	0.911*	0.570*	0.479*	0.911*	0.570*	0.479*	0.570*	0.479*	0.570*	0.479*	0.570*	0.479*
0.593*	0.642*	0.205*	0.152	0.237*	0.568*	0.479*	0.237*	0.568*	0.479*	0.568*	0.479*	0.568*	0.479*	0.568*	0.479*
0.628*	0.628*	0.312*	0.314*	0.352*	0.494*	0.536*	0.352*	0.494*	0.536*	0.352*	0.494*	0.536*	0.494*	0.536*	0.494*
0.036	-0.015	0.105	-0.081	0.070	0.080	0.028	-0.046	0.051	/	/	/	/	/	/	/
-0.049	-0.094	0.054	-0.132	0.021	0.117	-0.044	0.087	0.032	0.845*	/	/	/	/	/	/
0.147	0.108	0.147	-0.010	0.100	0.198*	0.173*	0.126	0.120	0.051	0.015	/	/	/	/	/
-0.007	0.056	0.135	0.249*	0.135	0.109	0.011	0.061	0.071	-0.052	-0.092	0.071	/	/	/	/
0.146	0.089	0.169*	-0.149	0.119	0.061	0.137	0.034	0.033	0.229*	0.201*	0.321*	-0.042	/	/	/
-0.009	-0.072	0.108	-0.057	0.080	-0.049	0.039	-0.085	-0.023	0.264*	0.202*	0.240*	-0.025	0.593*	/	/
0.094	0.131	-0.019	0.112	-0.038	0.092	0.008	0.035	0.067	0.061	0.038	0.055	0.051	0.308*	0.101	/

\* p – value < 0.05

Table 12 The correlation matrix of variables in the third trimester

	Total Protein	Animal Protein	Vegetable Protein	Animal Vegetable Protein	Carbohydrate	Fat	Energy	Calcium	Phosphorus	Prepreg-nancy weight	Prepreg-nancy BMI	Total weight gain	Rate weight gain	Birth Weight	Birth length	Gestational age
Total protein	/															
Animal protein	0.903*	/														
Vegetable protein	0.627*	0.231*	/													
Animal protein	0.319*	0.668*	0.487*	/												
Vegetable protein																
Carbohydrate	0.616*	0.262*	0.919*	-0.390*	/											
Fat	0.589*	0.638*	0.179*	0.409*	0.198*	/										
Energy	0.817*	0.534*	0.883*	-0.142	0.914*	0.517*	/									
Calcium	0.626*	0.660*	0.219*	0.405*	0.295*	0.489*	0.510*	/								
Phosphorus	0.807*	0.772*	0.429*	0.354*	0.277*	0.726*	0.701*	0.844*	/							
Pregpregnancy weight	-0.019	-0.030	0.011	-0.054	-0.038	-0.038	-0.028	-0.126	-0.078	/						
Pregpregnancy BMI	-0.080	-0.061	-0.072	-0.042	-0.133	-0.043	-0.105*	-0.121	0.123	0.845*	/					
Total weight gain	0.150	0.126	0.110	-0.011	0.092	0.207*	0.157*	0.213*	0.184*	0.051	0.015	/				
Rate weight gain	0.261*	0.210*	0.210*	-0.042	0.207*	0.078	0.232*	0.675*	0.205	0.045	0.107	0.608*	/			
Birth weight	-0.039	-0.026	-0.041	-0.104	0.031	-0.017	-0.037	0.008	-0.007	0.229*	0.201*	0.321*	0.146	/		
Birth length	-0.229*	-0.218*	-0.126	-0.016	-0.094	-0.124	-0.167*	-0.158*	-0.192*	0.264*	0.202*	0.240*	-0.080	0.593*	/	
Gestational age	-0.062	-0.019	-0.105	0.034	-0.072	0.059	-0.058	0.070	0.071	0.061	0.038	0.055	-0.174*	0.308*	0.101	/

\* p - value < 0.05

Table 13 The correlation matrix of variables in both the second and third trimesters

	Changing total protein T <sub>2</sub> -T <sub>3</sub>	Changing animal protein T <sub>2</sub> -T <sub>3</sub>	Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	Changing energy T <sub>2</sub> -T <sub>3</sub>	Total protein T <sub>2</sub>	Animal protein T <sub>2</sub>	Vegetable protein T <sub>2</sub>	Energy T <sub>2</sub>	Total protein T <sub>3</sub>	Animal protein T <sub>3</sub>	Vegetable protein T <sub>3</sub>	Energy T <sub>3</sub>	Birth weight	Birth length	Gestational age
Changing total protein T <sub>2</sub> -T <sub>3</sub>	/														
Changing animal protein T <sub>2</sub> -T <sub>3</sub>	0.913*	/													
Changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	0.546*	0.157*	/												
Changing energy T <sub>2</sub> -T <sub>3</sub>	0.768*	0.493*	0.846*	/											
Total protein T <sub>2</sub>	0.478*	0.459*	0.223*	0.368*	/										
Animal protein T <sub>2</sub>	0.448*	0.506*	0.049	0.238*	0.913*	/									
Vegetable protein T <sub>2</sub>	0.287	0.136	0.429*	0.418*	0.658*	0.294*	/								
Energy T <sub>2</sub>	0.374*	0.299*	0.302*	0.462*	0.847*	0.615*	0.850*	/							
Total protein T <sub>3</sub>	-0.573*	-0.502*	-0.348	-0.439*	0.447*	0.395*	0.332*	0.409*	/						
Animal protein T <sub>3</sub>	-0.571*	-0.611*	-0.124	-0.312*	0.344*	0.373*	0.123	0.242*	0.903*	/					
Vegetable protein T <sub>3</sub>	-0.260	-0.026	-0.564*	-0.427*	0.388*	0.221*	0.504*	0.488*	0.627*	0.231*	/				
Energy T <sub>3</sub>	-0.372*	-0.182*	-0.517*	-0.509*	0.409*	0.368*	0.425*	0.528*	0.817*	0.534*	0.883*	/			
Birth weight	0.172*	0.101	0.204*	0.177*	0.146	0.089	0.169*	0.137	-0.039	-0.026	-0.041	-0.037	/		
Birth length	0.217*	0.142	0.235*	0.214*	-0.009	-0.072	0.108	0.039	-0.229*	-0.218*	-0.126	-0.167*	0.593*	/	
Gestational age	0.148	0.129	0.091	0.069	0.094	0.132	-0.019	0.008	-0.062	-0.019	-0.105	-0.058	0.308*	0.101	/

\*p – value < 0.05

**Table 14** The effect of nutrient intake and weight gain on birth weight

Model	Regression Coefficient ( $\beta$ ) of Variables in the Equation						R <sup>2</sup>	P - value
1	constant	prepregnancy weight	vegetable protein T <sub>2</sub>	total weight gain	gestational age	gestational age	0.239	0.000
2	1039.669 <sup>NS</sup>	9.814*	5.633 <sup>NS</sup>	41.544*	11.083*			
3	constant	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	total weight gain	gestational age	gestational age	gestational age	0.217	0.000
4	-288.825 <sup>NS</sup>	8.096*	44.850*	10.787*				
5	constant	prepregnancy BMI	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	total weight gain	gestational age	gestational age	0.244	0.000
6	-743.409 <sup>NS</sup>	22.494*	7.113*	44.589*	10.619*			
7	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	total weight gain	gestational age	gestational age	0.250	0.000
8	-701.344 <sup>NS</sup>	9.731*	7.373*	43.596*	10.418*			
9	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	total weight gain	gestational age	gestational age	0.180	0.000
10	2076.890*	10.450*	8.443*	45.571*				
11	constant	prepregnancy BMI	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	total weight gain	gestational age	gestational age	0.171	0.000
12	2105.898*	23.437*	8.211*	46.690*				

\*p - value < 0.05, NS= Not significant

Table 15 The effect of nutrient intake and weight gain on birth length

Model	Regression Coefficient ( $\beta$ ) of Variables in the Equation					R <sup>2</sup>	P - value
1	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	calcium T <sub>3</sub>	total weight gain	0.185	0.000
	44.820*	0.065*	0.049*	-0.002*	0.217*		
2	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	calcium T <sub>3</sub>	phosphorus T <sub>3</sub>	total weight gain	0.000
	45.288*	0.067*	0.043*	-0.0002 <sup>NS</sup>	-0.001 <sup>NS</sup>	0.219*	
3	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	animal protein T <sub>3</sub>	total weight gain	0.211	0.000
	45.294*	-0.069*	0.049*	-0.031*	0.213*		
4	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	calcium T <sub>3</sub>	total weight gain	0.211	0.000
	45.305*	0.068*	0.049*	-0.00008 <sup>NS</sup>	0.214*		
5	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	animal protein T <sub>3</sub>	phosphorus T <sub>3</sub>	total weight gain	0.000
	45.295*	-0.068*	0.049*	-0.031*	-0.000006 <sup>NS</sup>	0.213*	

\* p - value &lt; 0.05, NS= Not significant

Table 15 The effect of nutrient intake and weight gain on birth length (continued)

Model	Regression Coefficient ( $\beta$ ) of Variables in the Equation							R <sup>2</sup>	p - value
6	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	animal protein T <sub>3</sub>	calcium T <sub>3</sub>	phosphorus T <sub>3</sub>	total weight gain	0.211	0.000
	45.281*	0.068*	0.048*	-0.031*	-0.0002 <sup>NS</sup>	0.00009 <sup>NS</sup>	0.214*		
7	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> -T <sub>3</sub>	phosphorus T <sub>2</sub>	animal protein T <sub>3</sub>	calcium T <sub>3</sub>	total weight gain	0.213	0.000
	45.412*	0.069*	0.051*	-0.0003 <sup>NS</sup>	-0.030*	0.00002 <sup>NS</sup>	0.216*		
8	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> - T <sub>3</sub>	calcium T <sub>2</sub>	animal protein T <sub>3</sub>	calcium T <sub>3</sub>	total weight gain	0.213	0.000
	45.382*	0.068*	0.049*	0.0004 <sup>NS</sup>	-0.031*	0.0002 <sup>NS</sup>	0.214*		
9	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> - T <sub>3</sub>	calcium T <sub>2</sub>	phosphorus T <sub>2</sub>	animal protein T <sub>3</sub>	total weight gain	0.214	0.000
	45.343*	0.068*	0.053*	-0.0003 <sup>NS</sup>	-0.0002 <sup>NS</sup>	-0.033*			
	calcium T <sub>3</sub>	phosphorus T <sub>3</sub>	total weight gain					0.214	0.000
	-0.0003 <sup>NS</sup>	0.0004 <sup>NS</sup>	0.215*						

\*p - value < 0.05, NS= Not significant

**Table15** The effect of nutrient intake and weight gain on birth length (continued)

Model	Regression Coefficient ( $\beta$ ) of Variables in the Equation					R <sup>2</sup>	p - value
10	constant	prepregnancy weight	changing vegetable protein T <sub>2</sub> - T <sub>3</sub>	animal protein T <sub>2</sub>	calcium T <sub>2</sub>	phosphorus T <sub>2</sub>	
	45.339*	0.069*	0.053*	0.0004 <sup>NS</sup>	-0.0003 <sup>NS</sup>	-0.0002 <sup>NS</sup>	
	animal protein T <sub>3</sub>	calcium T <sub>3</sub>	phosphorus T <sub>3</sub>	total weight gain			
	0.033*	-0.0001 <sup>NS</sup>	0.0004 <sup>NS</sup>	0.215*			0.000
11	constant	changing energy T <sub>2</sub> -T <sub>3</sub>	calcium T <sub>3</sub>	total weight gain			
	48.260*	0.0007*	-0.002*	0.232*			0.000
12	constant	prepregnancy weight	vegetable protein T <sub>2</sub>	total protein T <sub>3</sub>	total weight gain		
	45.236*	0.069*	0.041 <sup>NS</sup>	-0.035*	0.214*		0.000

\*p - value < 0.05, NS= Not significant

**Table 16** The effect of nutrient intake and weight gain on gestational age

Model	Regression Coefficient ( $\beta$ ) of Variables in the Equation						R <sup>2</sup>	p - value
1	Constant	energy T <sub>2</sub>	total proteinT <sub>2</sub>	rate weight gain T <sub>3</sub>			0.061	0.015
	274.868*	-0.005 <sup>NS</sup>	0.177*	-11.368*				
2	Constant	energy T <sub>2</sub>	total proteinT <sub>2</sub>	energy T <sub>3</sub>	total proteinT <sub>3</sub>	rate weight gain T <sub>3</sub>	0.066	0.046
	276.033*	-0.050 <sup>NS</sup>	0.193*	0.0003 <sup>NS</sup>	-0.047 <sup>NS</sup>	-10.403*		
3	Constant	changing total proteinT <sub>2</sub> -T <sub>3</sub>	animal protein T <sub>3</sub>	rate weight gain T <sub>3</sub>			0.058	0.019
	273.848*	0.101*	0.085 <sup>NS</sup>	-10.812*				
4	Constant	rate weight gain T <sub>3</sub>	total weight gain				0.071	0.002
	270.943*	-20.157*	0.966*					

\*p - value < 0.05, NS= Not significant

## APPENDIX B

### Gross energy expenditure in specified activities

Preliminary assessment of data (expressed in terms of the basal metabolic rate multiplied by metabolic constant)

**Female- Developed and developing societies**

Sleeping	1.0	(i.e, BMR*1)
Lying	1.2	
Sitting quietly	1.2	
Sitting activities		
sewing clothes	1.4	
sewing pandanus mat	1.5	
weaving carry bag	1.5	
preparing rope	1.5	
Standing	1.5	
Walking		
“around” or strolling	2.4	
slowly	3.0	
at normal pace	3.4	
with load	4.0	
uphill: at normal pace	4.6	
fast	6.6	
with load	6.0	
downhill: slowly	2.3	
at normal pace	3.0	
fast	3.4	
with load	4.6	
Household tasks		
light cleaning	2.7	
moderate cleaning (polishing, window cleaning, etc.)	3.7	
sweeping house	3.0	
sweeping yard	3.5	
washing cloths	3.0	
ironing cloths	1.4	
washing dishes	1.7	
cleaning house	2.2	
child care	2.2	
fetching water from well	4.1	
chopping wood with machete	4.3	
preparing tobacco	1.5	
deseeding cotton	1.8	
beating cotton	2.4	
spinging cotton	1.4	

**Gross energy expenditure in specified activities(continued)**

<b>Food preparation and cooking</b>	
Cooking	1.8
collecting leaves for flavoring	1.9
catching fish by hand	3.9
grinding grain on millstone	3.8
pounding	4.6
stirring porridge	3.7
making tortillas	2.1
removing beans from pod	1.5
breaking nuts(like peanuts)	1.9
squeezing coconut	2.4
peeling taro	1.7
peeling sweet potato	1.4
loading corn	1.3
loading earth oven with food	2.6
<b>Office work</b>	1.7
<b>Light industry</b>	
bakery work	2.5
brewery work	2.9
chemical industry	2.9
electrical industry	2.0
furnishing industry	3.3
laundry work	3.4
machine tool industry	2.7
<b>Agriculture (non-mechanized)</b>	
clearing ground	3.8
digging ground	4.6
digging holes for planting	4.3
planting root crops	3.9
weeding	2.9
hoeing	4.4
cutting grass with machete	5.0
sowing	4.0
threshing	5.0
binding sheaves	4.2
harvesting root crops	3.1
picking coffee	1.5
winnowing corn or rice	1.7
cutting fruit from tree	3.4
<b>Recreations</b>	
sedentary (playing cards, etc.)	2.1
light (billiards, bowls, cricket, golf, sailing, etc.)	2.1-4.2
moderate (dancing, swimming, tennis, etc.)	4.2-6.3
heavy ( football, athletics, jogging, rowing, etc.)	>6.3

No. □□□□

Form A

Screening Subject Form

1. Srinakarin Hospital  2. Maternal and Child Hospital  3. Khon Kaen Regional Hospital

Date of interview..... Name of interviewer.....

Name of pregnant woman.....

Hospital Number.....

Address.....

.....

1. Age.....years

2. Prepregnancy weight.....kg , Height.....cm.

3. Parity ( ) 2. ( ) 3.

4. Present gestational age.....weeks , weight.....kg

5. Weight gain during pregnancy

Date.....week of gestational age..... , weight.....kg

6. Estimate delivery date(EDC).....

7. First day of the last menstrual period.....



- ( ) 5 Diploma
- ( ) 6 Bachelor's degree
- ( ) 7 others, specify.....

18. Occupation during pregnancy.

- ( ) 1 Farmer
- ( ) 2 Laborer, type of work.....
- ( ) 3 Trader, type of trade.....
- ( ) 4 Officer, position.....
- ( ) 5 Housewife
- ( ) 6 Others, specify.....

19. Marital status

- ( ) 1. Married
- ( ) 2. Widow
- ( ) 3. Divorced / separate
- ( ) 4. Others, specify.....

20. How long do you sleep at night during pregnancy?.....hours

21. How long do you work?.....hours / day

Work with standing.....hours / day

Work with sitting.....hours / day

Work with walking.....hours / day

Housework.....hours / day

Relaxing time.....hours / day

Others.....hours/ day

22. How long do you suppose to work during your pregnancy?

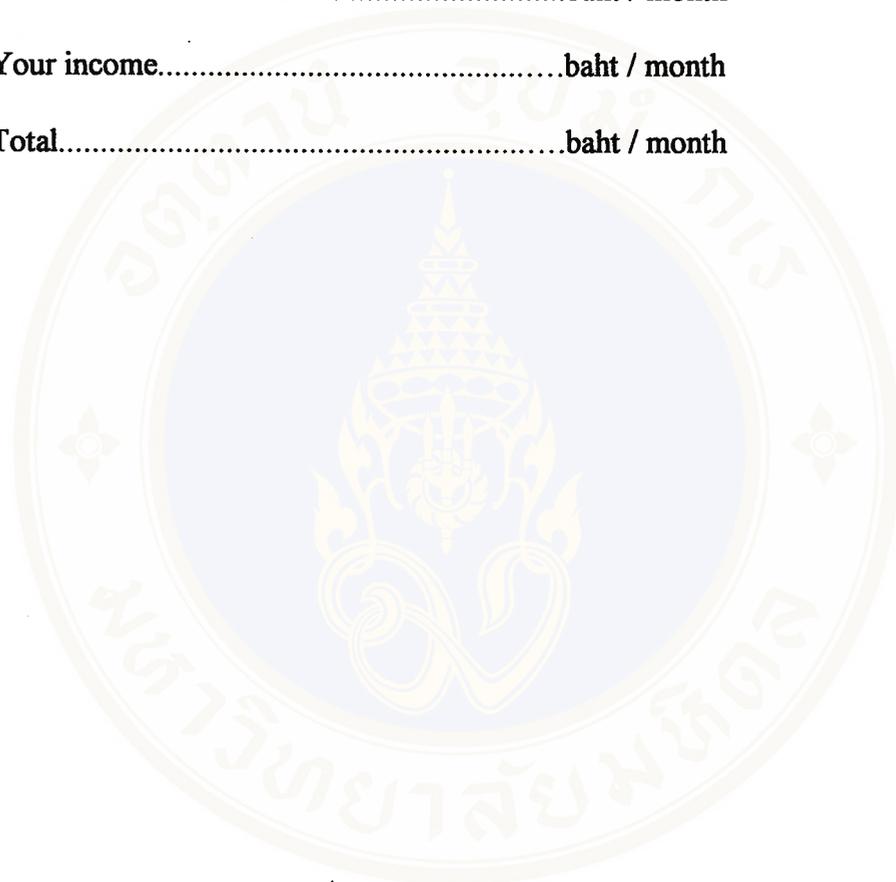
( ) 1 Until.....weeks of gestational age.

( ) 2 Until delivery

23. Your husband's income.....baht / month

Your income.....baht / month

Total.....baht / month



No

**Form B**

**Food Frequency Form**

1.Srinakarin Hospital  2. Maternal and Child Hospital  3. Khon Kaen Regional Hospital

Date of interview.....Name of interviewer.....

Name of pregnant woman.....

Hospital Number.....

1. Do you have any symptoms of morning sickness which affect your food intake?

- ( ) 1 No symptoms                      ( ) 2 During first three months
- ( ) 3 Until present interview ( ) 4 Others, specify.....

**Food habit during pregnancy**

2. How many meals do you have per day?

- ( ) Before breakfast                      ( ) Breakfast
- ( ) Between breakfast and lunch      ( ) Lunch
- ( ) Between lunch and dinner          ( ) Dinner
- ( ) Before bedtime

3. Which food do you like to eat during pregnancy?.....  
.....

4. Which food do you dislike to eat during pregnancy?.....  
.....

5. What taste of food do you like before pregnancy ?

- 1 Hot
- 2 Sour
- 3 Sweet
- 4 Mild
- 5 Others, specify.....

6. What taste of food do you like during pregnancy.?

- 1 Hot
- 2 Sour
- 3 Sweet
- 4 Mild
- 5 Others, specify.....

7. How do you manage your meal before pregnancy?

- 1 Cook by yourself
- 2 Buy cooked food
- 3 Eat out
- 4 Others, specify.....

8. How do you manage for your meal during pregnancy?

- 1 Cook by yourself
- 2 Buy cooked food
- 3 Eat out
- 4 Others, specify.....

9. What are your taboo foods during pregnancy?

.....

10. What are your special foods during pregnancy?

.....

No. □□□□

**Food Frequency of Pregnant Woman**

Name of pregnant woman.....Trimester ( ) 2 ( ) 3 Name of interviewer.....

Food items	Never	Time/ day	Time/ week	Time/ month	Usual quantity of food intake	Method of cooking (mostly)				Note	
						Deep fry	Fry	Boil	Roast		Steam
<b>Milk and products</b>											
- Sterilized milk											
- Whole milk (UHT, natural)											
- Whole milk (UHT, sweet)											
- Milk - (UHT- low fat)											
- Drinking yogurt											
- Milo / Ovaltine											
- Sweet condense milk											
<b>Meat and products</b>											
- Poultry											
- Pork											
- Beef											
- Fresh-water fish											
- Marine foods											
- Mackerel											







No. □□□□

**Form C****24 - Hour Dietary Recalls Form**

1. Srinakarin Hospital  2. Maternal and Child Hospital  3. Khon Kaen Regional Hospital

Name of pregnant woman.....

Name of interviewer.....Date of interview.....

1. Gestational age.....weeks      Weight.....kg

2. Blood pressure.....

3. Hemoglobin.....mg%

4. Sugar / protein in urine

( ) 1 Normal      ( ) 2 Abnormal

5. Are you still working?

( ) 1 Yes      ( ) 2. No

6. Do you take vitamin mineral or other supplement during pregnancy.?

( ) 1 Yes      ( ) 2. No

Specify .....

7. How about your food intake during weekdays and weekends?

( ) 1 Same quantity      ( ) 2 Different quantity

8. If different

( ) 1 Weekends > Weekdays      ( ) 2 Weekends < Weekdays



**Form D**

**Food Record of Pregnant Woman**

Name of pregnant woman.....Date of record.....No.....

Meal	Menu	Ingredient	Quantity	Note
Breakfast				
Lunch				
Dinner				

No □□□□

### Form E

### Fetal Outcomes Form

1. Srinakarin Hospital  2. Maternal and Child Hospital  3. Khon Kaen Regional Hospital

Name of pregnancy.....

Date of delivery.....

1. Gestational age.....days

2. Sex of infant.....

( ) 1 Male            ( ) 2 Female

3. Birth weight.....g.

4. Birth length.....cm.

5. Health of infant

( ) 1 Normal            ( ) 2 Abnormal

6. Type of delivery

( ) 1 Normal            ( ) 2 Caesarian section

## APPENDIX C

Based on the results, pregnant women are recommended to consume high protein (>100 g) in the second trimester and then decrease to moderate level (51-100 g) in the third trimester. The ratio of animal protein to vegetable protein should be 1:2.

The example of food consumption per day in both trimesters is as follows:

### The second trimester

Food items	Food quantity	Protein quantity(g)
<b>Animal protein</b>		
- Meat, fish, sea foods and their products	3-4 portions (30 g / portion)	21-28
- Egg	1 portion (50 g / portion)	7
- Milk and dairy products	1 portion (240 ml / portion)	8
		36-43
<b>Vegetable protein</b>		
- Cereals and their products	14-16 portions (½ cup / portion)	28-32
- Legumes and their products	2-3 portions (½ cup /portion)	14-21
- Soybean milk	2 portions (240ml/ portion)	14

**The second trimester ( continued)**

<b>Food items</b>	<b>Food quantity</b>	<b>Protein quantity(g)</b>
- Vegetables with high protein	6 portions ( $\frac{1}{2}$ cup / portion)	12
		68-79
	<b>Total</b>	<b>104-122</b>

**The third trimester**

<b>Food items</b>	<b>Food quantity</b>	<b>Protein quantity(g)</b>
<b>Animal protein</b>		
- Meat, fish, sea foods and their products	1-3 portions (30 g / portion)	7-21
- Egg	1 portion (50 g/ portion)	7
- Milk and dairy products	$\frac{1}{2}$ - 1 portion (240 ml / portion)	4- 8
		18-36
<b>Vegetable protein</b>		
- Cereals and their products	6 -12 portions ( $\frac{1}{2}$ cup / portion)	12-24
- Legumes and their products	1-2 portions ( $\frac{1}{2}$ cup /portion)	7-14
- Soybean milk	1-2 portions (240ml/ portion)	7-14

**The third trimester ( continued)**

<b>Food items</b>	<b>Food quantity</b>	<b>Protein quantity(g)</b>
- Vegetables with high protein	4-5 portions (1/2 cup / portion)	8-10 <hr/> 34-62
	<b>Total</b>	<hr/> <b>52-98</b> <hr/>

There are many varieties of foods that can be substituted for food items in the example of food consumption:

**Meat, fish, sea fish and their products;** pork, poultry, beef, fresh-water fish, shrimp, sea fish, squid, meat organ, sausage, etc.

**Milk and their products;** whole milk, yogurt, drinking yogurt, ice cream, etc.

**Cereals and their products;** rice, noodle, bread, taro, potato, corn, etc.

**Legumes and their products;** soybean, mungbean, cowpea, peanut, etc.

**Vegetables with high protein;** mushroom, bamboo shot, long bean, mung bean sprout, pumpkin, etc.

Due to the high quantity of protein intake, some food items may be consumed as dessert or snack in addition to three meals of the day, for example; legumes and beans can be cooked into dessert: cowpea with coconut milk, Tow-Suan (creamy mungbean), Taw-Pap (rice flour filled with mungbean).

## BIOGRAPHY



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