CONFIDENTIAL

WORLD HEALTH ORGANIZATION ORGANISATION MONDIALE DE LA SANTE

NDP/UNFPA/WHO/WORLD BANK SPECIAL PROGRAMME OF RESEARCH,
DEVELOPMENT AND RESEARCH TRAINING IN HUMAN REPRODUCTION

FOR WHO USE ONLY
HRP Project No.:A 65097
Date received by WHO:

Form 1. Particulars of the project and investigator(s)

1. Programme thematic area for which proposal has been developed

Improving Maternal and Perinatal Health

2. Project title

WHO MULTICENTRE STUDY FOR THE DEVELOPMENT OF GROWTH STANDARDS

FROM FETAL LIFE TO CHILDHOOD: THE FETAL COMPONENT

3. Principal investigator(s)

Multicentre study

4. Institution responsible for the research project

Improving Maternal and Perinatal Health Team
UNFPA/WHO/WORLD BANK Special Programme Of Research, Development And Research Training In Human Reproduction
Department of Reproductive Health And Research
World Health Organization
Geneva, Switzerland

International Society for Ultrasound in Obstetrics and Gynecology

5. Investigator(s) and department(s) collaborating with the principal investigator(s) (if any)

- Argentina: Centro Rosarino Estudios Perinatales, Rosario
- Brazil: University of Campinas, Campinas
- Democratic Republic of Congo: University of Kinshasa, Kinshasa
- Denmark: Rigshospitalet , Copenhagen

- Egypt: Assiut University, Assiut
- France: Hopital Antoine Beclere, Paris
- Germany: University Medical Center, Hamburg
- India: Old India Institute of Medical of Science, New Delhi
- Norway: University of Bergen, Bergen
- Thailand: Kohn Kaen University, Kohn Kaen

Executive committee

Dr Larry Platt (Chairman)
Dr Kurt Hecher
Dr Guillermo Carroli
Dr Rogelio Gonzalez
Dr Torvid Kiserud

Ex officio: Dr Mario Merialdi

6. Responsible financial officer

7. Duration of project

Earliest starting date (if applicable): (Inception phase)

Total (years): 1 year at each study site. Study sites may implement activities at different times according to availability of funds.

8. Funds requested (US $)

1st year	2nd year	3rd year	**TOTAL**

9. Approval of ethics committees
For studies involving humans or human biological materials:

A. Is *institutional* ethical clearance document attached?

Yes [ ] No [x]

B. Is documentation of *national* ethical approval attached?

Yes [ ] No [x]

For studies involving the use of animals:

Is documentation of institutional or national ethical approval for animal experimentation attached?

Yes No Not required x

10. Acceptance of responsibility

If this application is accepted, I (we) declare that I (we) shall be actively engaged in, and shall be in day-to-day control of, the project; and I (we) agree to provide detailed annual progress reports to WHO of the work undertaken. I (we) agree to follow the policies of the Special Programme on the dissemination of research results

Date and signature(s) of principal investigator(s):

_____

_____

_____

_____

11. Declaration(s) of the Director of the institution (or a designated representative) and the officer responsible for the administration of WHO funds awarded (e.g. Finance Officer, Bursar, etc.)

We confirm that this application has been approved by our institution and that, if granted, the work will be administered in the institution in accordance with the general conditions of the World Health Organization. The staff gradings and salaries provided are correct and in accordance with the normal practice of the institution.

Head of the institution

Name and initials
(please print) _____
_____

Title _____
_____

Institution

_____
_____

Date _____
_____

Signature _____
_____

Administrative authority

Name and initials
(please print)

Title

Institution

Date

Signature

Form 2. Project summary

Objective: To conduct a multinational study for the development of fetal growth standards for international application by assessing fetal growth under nutritionally unrestricted conditions in populations of different ethnic and geographic backgrounds.

Global perspective: Each year approximately four million newborns and 500,000 pregnant women die due to complications related to pregnancy and childbirth. Almost all (99 %) of these deaths occur in the poorest countries of the world, and are potentially preventable through proven interventions. These figures represent one of the starkest health inequities of our time, and indicate that the most disenfranchised are often denied the basic human right to life and safe reproduction.

Current practices concerning the assessment of fetal growth highlight the disparity in available resources for maternal and newborn care between rich and poor countries. Monitoring fetal growth *in utero* by ultrasonography based on growth references derived mainly from US and European populations, for example, has become a major component of standard obstetric care worldwide. However, several reports show that these references may be inappropriate for international use given possible variances in the growth rates of fetuses from different ethnic population groups. These findings may have important implications for obstetric practice since the majority of newborn deaths occur in non-US and European populations and more than 60% of these are associated with low birth weight.

Rationale for the study: The 1995 WHO Expert Committee on Physical Status indicated as a top priority the need for the development of growth reference data suitable for international applications. In response to this call for action, the WHO conducted a multicentre study resulting in the development and release in 2006 of standard infant and child growth charts: The WHO Child Growth Standards. We propose to complement this study by extending its scope to the fetal period, thus determining if universally applicable growth standards for the intrauterine life are scientifically justified and should complement the child growth standards.

The development of reliable fetal growth standards has important clinical, research, and public health implications; and was endorsed as an imperative research need at the 2002 Meeting of Experts on Birth Weight organized by the WHO Department of Nutrition, and at the 2002 WHO Meeting of Experts on the Life Course and Health. A reliable fetal growth standard will facilitate the identification of impaired growth occurring in utero, enabling clinicians to better manage pregnancies and potentially prevent and/or reduce the risk of perinatal mortality and morbidity associated with intrauterine growth retardation. Evidence linking impaired fetal growth with the development of non-communicable diseases (e.g., cardiovascular disease and diabetes) in adulthood also suggests that early detection of restricted growth in utero may have long-term health benefits. In addition, improved measurement of fetal growth abnormalities is crucial for maternal health because these abnormalities are often a sign of pregnancy-related complications.

The results of this study will provide needed information on how normal fetuses <u>should</u> grow in ethically different populations and consequently if growth patterns differs or not across ethnic groups. These outcomes go beyond previously conducted studies that were limited to the description of patterns of growth in specific populations, settings, and times. The objectives of the study will be achieved by using a prescriptive approach to selecting the study populations which should be as much as possible free of constrains on fetal growth, involving the enrollment of pregnant women meeting the following criteria: 1) High-middle socioeconomic status with

no obvious environmental constraints on growth (adequate nutritional status, no smoking), and 2) Normal pregnancy history with no complications likely to affect fetal growth. Importantly, an international sampling frame will also be used to ensure adequate representation of diverse ethnic groups and populations located in different geographic settings.

Because adequate assessment of fetal growth is critical to monitoring the health and nutritional status of mothers, fetuses and newborns, and to monitoring progress in reducing the incidence and severity of specific obstetric and perinatal conditions, this study is relevant to the Millennium Development Goals 1, 4 and 5. The development of international standards for fetal growth will also enable inter and intra-country comparisons of fetal growth, and examination, in a standardized way, of the relationship between fetal growth patterns and maternal and perinatal health outcomes.

In view of the benefits for the pregnant population at large and the low risk associated with the use of ultrasonography in pregnancy it is felt that the proposed examination schedule of 8 examinations for research purposes is ethically justified. Monthly ultrasonographical examinations are considered necessary to have enough data points to develop growth curves that take into account the variability in growth with advancing gestational age. A four week interval is the accepted time period to detect meaningful changes in growth in non complicated pregnancy. In addition, one early examination is necessary to confirm gestational age. A reliable and certain determination of gestational age is a critical determinant of the internal validity of a study that aims at developing fetal growth standards as a function of advancing gestational age.

Study design: Multicentre study of fetal growth assessed by serial ultrasonographic examinations and newborn anthropometrical assessment. The study will apply the same research strategy in terms of sample selection procedures, data collection, and data analysis used to develop the WHO Child Growth Standards. It will follow a prescriptive approach for selecting the study population, and the collected information will also include data on maternal nutritional status and pregnancy complications that may affect fetal growth.

The study will be conducted in the centres affiliated with the WHO maternal and perinatal health research network in collaboration with the International Society of Ultrasounds in Obstetrics and Gynecology. The study team includes researchers from prestigious institutions in developing and industrialized countries, consultants of solid scientific international reputation, and WHO staff with experience in conducting important research projects in the area of maternal, newborn and child health.

15 countries have been identified as potential participants. Ultrasonographical equipment needed for the implementation of the study is already available at the centres or will be provided by a private company (through an agreement regulated by the WHO Legal Office and the legal representatives of the company).

At each centre, 140 women will be recruited between 8+0 and 12+6 weeks. Subsequently, visits for fetal biometry will be scheduled at approximately 14, 18, 24, 28, 32, 36, and 40 weeks (+/- 1 week).
It is expected that each centre will be able to recruit and follow up 140 women during an 18 month period.

If adequate funds are made available, ancillary studies will be conducted at some of the participating centres. These studies include: 1) the development of postnatal growth curves for preterm babies, 2) the determination of the relationship between gestational age at delivery, birth weight and perinatal mortality and morbidity, 3) the evaluation of fetal neurobehavioral development, 4) the analysis of socio-cultural and other contextual factors surrounding the use of ultrasonography during pregnancy and 5) descriptive studies of fetal growth in unselected or

at risk populations groups (e.g. pregnant women living in malaria endemic regions).

Investigators: Experts in:

- ultrasonographic assessment of fetal growth
- nutrition during pregnancy and epidemiological nutrition;
- perinatal epidemiology and biostatistics as applied to the development of growth reference curves;
- the design and conduct of large epidemiological studies in developing countries;
- anthropometric assessment of newborns;
- science and technology studies, and medical anthropology.

Outcome: The main outcome of the proposed study will be the development of fetal growth standards (either global or population specific) for international applications that will be integrated into the WHO Child Growth Standards. These standards will have important clinical and research implications for the prenatal and postnatal periods, as well as for maternal health. The multinational cohorts examined in the study could provide the basis for long term follow-up studies extending into adulthood to investigate the fetal origins of chronic diseases.

The growth standards developed through the study will be incorporated into obstetric practice and national health policies at country level in coordination with the activities presently conducted by WHO to implement the use of the Child Growth Standards.

Form 3. Description of the project

DESCRIPTION OF THE PROJECT RATIONALE AND ITS COMPONENTS

1. INTRODUCTION

Of the estimated 4 million neonatal deaths each year, more than 60% are associated with low birth weight due to intrauterine growth restriction and/or preterm delivery (1). Accurate prenatal assessment of fetal growth and gestational age to timely identify and adequately manage cases of growth restriction and/or preterm delivery should be considered a public health priority, especially in developing countries where 98% of the worldwide neonatal deaths occur.

Before the advent of ultrasonography, fetal growth was assessed by evaluating newborn weight as the end point of the intrauterine growth process. However, cumulative evidence indicated that this birthweight curve approach did not accurately describe the fetal growth process for newborns born before 37 weeks of pregnancy, and low birth weight babies born at term. In response to this evidence, the prenatal assessment of fetal growth and gestational age *in utero* by means of ultrasonography was adopted as standard practice in prenatal care throughout the world. The adequacy of fetal size is currently assessed by comparing the measurements of fetal anatomical parameters at a given gestational age captured through ultrasonography with reference percentiles of fetal size derived from populations of fetuses whose growth was assumed to be normal. These same reference percentiles can be used to estimate gestational age from observed fetal size. The most commonly used reference charts of size by gestational age were developed based on data from populations of fetuses in the United States or Europe. Since 1981, concerns have been raised that such charts might not be appropriate for use in other ethnic groups which may experience different patterns of fetal growth(2-4). If fetal growth is dependent upon ethnic heritage, the potential for misclassification of fetal growth abnormalities through the wide-scale application of existing growth reference charts should generate concerns regarding diagnostic and management decisions made on the basis of ultrasonographic fetal

growth assessments. Others counter-argue that environmental factors play a more critical role in fetal growth than ethnicity, and that all fetuses should undergo comparable growth patterns in the absence of environmental constraints to growth (5). The need to resolve this debate makes the development of fetal growth standards for international application both timely and a priority for improving maternal, fetal and newborn heath care worldwide (6).

Reliable standards of fetal growth are important for assessing the wellbeing of each maternal-infant dyad, determining the health status of populations, and monitoring progress in fetal and newborn health and development. Accurate fetal growth assessment is also important for maternal health. Fetal growth abnormalities are often associated with pregnancy complications such as hypertensive disorders and may affect how specific pregnancies are managed (e.g., the decision to perform a cesarean section). Thus, the results of this study will be beneficial for maternal and fetal/newborn health, will support efforts to provide a continuum of care for mothers and their infants, and will be relevant to country efforts to accelerate progress to achieving MDGs 1, 4, and 5.

The following sections briefly describe the work done and recommendations issued by the World Health Organization in the field of growth assessment that led to the development of this proposal.

1.1 The WHO Child Growth Standards

On April 27th 2006, WHO released the WHO Child Growth Standards for children between 0 and 5 years which have been obtained by the WHO Multicentre Growth Reference Study (MGRS) (7). The MGRS was implemented in 1997 after several years of careful planning and systematic review of the evidence.

Two characteristics made the MGRS unique and unprecedented as a study in its field:

- The prescriptive approach for the selection of the study populations. This approach means that the study included only children from populations with minimal environmental constraints on growth. This was achieved by recruiting children from affluent and educated parents (high education and family income have been identified as the environmental variables most likely to be associated with child growth) (8-10). In addition, eligibility criteria included absence of morbidity, adherence to MGRS feeding recommendations and absence of maternal smoking;
- International representation because the study included populations from Brazil, Ghana, India, Norway, Oman and the USA.

By virtue of these characteristics, the MGRS provided the scientifically sound foundations for developing standards that indicate how children should grow as opposed to the studies conducted before that simply described patterns of growth in specific populations, times and settings. In sum, the WHO Child Growth Standards can be used to assess the growth of all children because they indicate the adequate growth of healthy children living in environmental conditions that permit the achievement of their growth potential.

The proposed study is an extension of the MGRS to fetal life. It will be based on the same prescriptive approach for sample selection, and will ensure international representation by including populations from five continents. The design, implementation, conduct of the study, and dissemination of the study results including their incorporation into clinical practice guidelines and health care policies will also parallel the MGRS strategy. The main outcome of the study, fetal growth reference standards, will complement the MGRS by extending the WHO Child Growth Reference Charts to the pre-natal period.

1.2 The 1995 WHO Expert Committee on Physical Status Report: The use and interpretation of anthropometry in the various stages of life

In 1995, the WHO Expert Committee on Physical Status published the results of a three year collaborative effort involving more than 100 experts who reviewed the currently available data on body size and composition at the different stages of life and their interpretation in terms of nutrient intake, activity level and risk of disease.
The specific tasks of the Expert Committee were(11):

- To develop recommendations for the appropriate use and interpretation of anthropometry in individuals and populations in various operational settings;
- To identify and/or develop reference data for anthropometric indicators when appropriate;
- To provide guidelines on how these reference data should be used;
- To identity new or unresolved issues and gaps in knowledge.

Among the various life stages, the fetal, infancy and childhood periods are particularly vulnerable to malnutrition and associated diseases affecting normal growth, as reflected in anthropometrical data which show that 26 millions infants are born too small each year world wide and that 230 million children in the developing world are stunted in their growth (12). Appropriate anthropometrical reference data are necessary to identify those infants and children at risk for malnutrition and disease (12). When reference data were not available or incorrect, the WHO Expert Committee on Physical Status recommended specific research efforts to fill the identified gaps in the literature. As a follow up of these recommendations, WHO implemented and conducted the MGRS which was coordinated by Dr. Mercedes De Onis from the WHO Department of Nutrition for Health and Development. In addition, the WHO Expert Committee's also recommended the development of fetal growth reference data suitable for international applications(5). The proposed study intends to implement those recommendations as a logical extension of the MGRS.

1.3 The 2002 WHO Meeting of Experts on Life Course and Health: Life course perspectives on coronary heart disease, stroke and diabetes and the fetal origins of adult disease hypothesis.

The importance of comprehensive reference data that will span from the fetal period to childhood is not limited to perinatal and early postnatal life. There is a growing interest in the fetal origin of chronic diseases that manifest themselves in adulthood(13). It is plausible that this interest will acquire more international relevance as many of the developing countries undergo the epidemiological transition and experience an increase in the prevalence of chronic pathological conditions such as cardiovascular disease and diabetes. The true social and economic implications to developing countries of this anticipated epidemic of chronic morbidity will become more evident in the future(14). However, evaluation of available information about disease trends indicate that the burden of cardiovascular disease and diabetes is expected to rise in those countries where the resources to deal with them are most scarce (15).

The WHO is actively committed to incorporating life course perspectives into the development of policy and research and, is particularly interested in examining the possible fetal origins of coronary heart disease, stroke and diabetes. A meeting of experts on the life course and health convened by the WHO Department of Noncommunicable Diseases Prevention and Health promotion in May 2002 identified as a top research priority the "improvement of measures of intra-uterine growth retardation (alternative to low birth weight) which must reflect newborn body composition and fetal exposures that may not necessarily be expressed in birth size"(15). Recent findings relating fetal femur length as assessed *in- utero* by ultrasonography with blood pressure levels in childhood confirm that measures of fetal growth alternative to birth weight provide important information which may help clarify the potential associations between fetal growth abnormalities and postnatal risk of disease(16).

1.4 The 2002 WHO Meeting of Experts on Birth Weight

In December, 2002 the WHO Department of Nutrition organized a meeting of experts to review current knowledge and the practical implications of the interpretation of birth weight as a health outcome. Further research to develop fetal growth standards was identified by the committee as urgent.

2. FROM RECOMMENDATIONS TO RESEARCH ACTION: THE DEVELOPMENT OF AN INTERNATIONAL COLLABORATION FOR DETERMINING STANDARDS OF FETAL GROWTH

Since 2005, the Improving Maternal and Perinatal Health Team (WHO Department of Reproductive Health and Research) has been collaborating with the International Society for Ultrasound in Obstetrics and Gynecology, The WHO Department of Nutrition and Development and several institutions worldwide to:

- Develop the proposed protocol
- Identify the potential study sites
- Collaborate with the private sector to secure the provision for free of state of the art ultraonographical equipment (through a contract regulated by the WHO Legal Office)

3. STUDY PREPARATION: A TRAINING COURSE ON STUDY PROCEDURES AND RESEARCH METHODOLOGY

A critical component of the study will be the training of research staff who will correctly identify eligible study subjects, strictly follow them up to ensure compliance with research activities, recognize health and environmental conditions that may affect fetal growth, take accurate biometric and anthropometric measurements, collect other variables as described in the protocol and manual of operations, enter the data through an on-line data management system, and interact with the coordinating unit.
In order to accomplish those objectives, The International Society of Ultrasounds in Obstetrics and Gynecology, WHO, and participating institutions have organized a training programme articulated along the following activities:

- Certification of an independent training centre at the University of Parma, Italy:

1. Training and certification of one trainer to be conducted by the Thomas Jefferson University, Philadelphia (under the supervision of Dr George Bega)
2. Site visit of the centre to be conducted by Dr Wesley Lee (William Beaumont Hospital, Detroit)

- One week training and certification course to be completed by each sonographer at the independent training centre. As done in previous studies(17), sonographers participating in the study will receive specific training and will be certified as proficient under the supervision of a qualified instructor accordingly to a standard protocol. Intra-observer and inter-observer measurement errors will be assessed according to a published protocol before the initiation of the study at the training course (18). Briefly, each fetus will be scanned by one examiner and the instructor. Each examiner will obtain two images of each fetal anatomical parameter under study at approximately 5 minute intervals. Differences between the two measurements will be expressed as the percent of the measurement obtained from the technically best image of the two. Percent differences will be used to take into account the increase in the dimensions of the fetal anatomical parameters with advancing gestational age. Percent differences for each

examiner will be averaged and the mean values compared to zero and to the instructor's measurements by t-test. In addition, measurement error will be evaluated by calculating the intra-class coefficients between pairs of repeated anatomical measurements (19).

- Site visits to the study centres will be organized in order to provide lectures and update courses by experts from the The International Society of Ultrasounds in Obstetrics and Gynecology. In addition during site visits, standardization sessions will be carried out according to repeat-measure protocols to assess the accuracy and precision of the anthropometrical measurements in mother and newborns (Reference: Ulijaszek SJ. Antrhopometric measures. In: Margetts BM and Nelson M (eds.)Design Concepts in Nutritional Epidemiology. Oxford University Press. Oxford, UK 1997 pp.: 289-311)

4. THE STUDY DESIGN

4.1 Population frame

4.1.1. The international sampling frame

The 1995 WHO Expert Committee on Physical Status examined evidence from studies on fetal growth based on newborn size that showed variation in growth across populations of different ethnic and geographic backgrounds. The Committee concluded that it is difficult, due to the limitations of the available data, to determine if those differences are due to genetic characteristics, environmental factors, or methodological issues inherent to the conduct and analysis of the studies (5). The data obtained in the context of the MGRS showed that when health and environmental needs are met, children from 0 to 5 years grow very similarly, independently of ethic origin (20). Potentially, this finding could be extrapolated to the fetal period because anthropometrical measurements taken at birth were similar in the MRGS across the geographical sites included in the study.

We feel that proposing an international sampling frame for the development of a single fetal growth standard is justified given the findings of the MGRS, and as a means of resolving the debate about the role of ethnic heritage in driving fetal growth patterns. The findings of the MGRS indicated that the results of previous studies noting differences in child growth patterns in various settings were likely due to methodological issues, environmental factors, and limited adequacy of the reference data used rather than to any underlying variances in how children of different heritage grow. If we will report similar findings about fetal growth patterns across diverse populations, it would be justified to develop an universal growth standard for international use. On the contrary, if heterogeneity by centre will be detected despite selecting study populations according to the prescriptive approach, the study results will indicate the needs to develop local/ethnically specific standards.

This study uses an inclusive approach to selecting participating centres so that diverse ethnic population groups and those located in various geographic settings are adequately represented.

The following centres have been identified to participate in the study as May 2009

- Argentina: Centro Rosarino Estudios Perinatales, Rosario
- Brazil: University of Campinas, Campinas
- Democratic Republic of Congo: University of Kinshasa, Kinshasa
- Denmark: Rigshospitalet , Copenhagen
- Egypt: Assiut University, Assiut
- France: Hopital Antoine Beclere, Paris

- Germany: University Medical Center, Hamburg
- New Zealand: Centre for Clinical Research and Effective Practice, Otahuhu
- Norway: University of Bergen, Bergen
- Thailand: Kohn Kaen University, Kohn Kaen

As noted above, the participating centres were selected to ensure diverse ethnic/geographic representation. These centres were also selected based on proficiency in the use of ultrasonography. In all participating centers, pregnant women routinely receive serial ultrasonagraphical examinations during antenatal care visits.

In the rate event of a detection of a fetal malformation, women enrolled in the study will be informed of and given the opportunity to terminate the pregnancy. Women who are detected with potential fetal abnormalities will be informed immediately of the possible fetal abnormality by the attending obstetrician in charge of the woman's pregnancy who will also inform the woman of the availability of a trained counsellor providing psychological and emotional support. In addition, the attending physician will inform the woman about the nature of the abnormalities, its prognosis and the treatment options including termination of pregnancy according to the local legal regulations.

4.1.2. Sample size

- The total sample size will be 1400 pregnant women and their infants. Each country centre will collect data from 140 women. This number is sufficient to the development of local centile growth charts with high level of precision, accounting for exclusions to final analysis due to loss to follow up and occurrence of pregnancy complications (21;22).
- The sample size needed for the estimation of a specific percentile were computed using the following formula (23):

$$n = \frac{(1 + z_{\alpha}^{2}/2)}{\left(\frac{\%SE\,P_{\alpha}}{\%CV}\right)^{2}}$$

where z_{α} is the standard normal deviate corresponding to the percentile being estimated, %SE C_{α} is the expected percentage standard error of the percentile and %CV is the percentage coefficient of variation. Sample sizes were computed for 5 different parameters (biparietal diameter, abdominal and head circumference and femur and humerus length) and for 3 different percentiles: 5%, 10% and 50%. Information about the coefficient of variation was obtained from data on serial ultrasonographical examination conducted on approximately 500 pregnancies in the context of the WHO randomized trial of calcium supplementation in low intake women(24). Additionally, the expected half-width of the expected confidence interval with a sample size n = 100 was computed.

Shown in the following tables are the level of precision obtainable with a sample size of 100 women for various biometrical parameters . Reported values and half widths are in mm.

Abdominal circumference

Week	Mean	10% Centile	5% Centile	CV	%SE_50	%SE_10	%SE_5	Width_50	Width_10	Width_5
20	147.7	133.5	128.8	7.9	0.79	1.07	1.21	2.33	2.85	3.12
24	190.7	173.8	166.6	7.4	0.74	1.00	1.14	2.82	3.47	3.78
28	235.2	215.4	210.6	6.4	0.64	0.86	0.98	3.01	3.72	4.13
32	278.6	255.8	248.3	6.5	0.65	0.88	1.00	3.62	4.49	4.95

36	314.5	288.0	277.9	7.1	0.71	0.96	1.09	4.47	5.52	6.05

Bipariatal diameter

Week	Mean	10% Centile	5% Centile	CV	%SE_50	%SE_10	%SE_5	Width_50	Width_10	Width_5
20	46.7	44.0	42.0	5.6	0.56	0.76	0.86	0.52	0.67	0.72
24	59.0	56.0	55.0	4.8	0.48	0.65	0.74	0.57	0.73	0.81
28	70.8	68.0	67.0	3.9	0.39	0.53	0.60	0.55	0.72	0.80
32	80.1	77.0	75.0	3.7	0.37	0.50	0.57	0.59	0.77	0.85
36	87.1	83.0	81.0	3.7	0.37	0.50	0.57	0.64	0.83	0.92

Head circumference

Week	Mean	10% Centile	5% Centile	CV	%SE_50	%SE_10	%SE_5	Width_50	Width_10	Width_5
20	177.4	164.4	161.2	5.8	0.58	0.78	0.89	2.06	2.57	2.87
24	225.0	212.3	209.2	4.9	0.49	0.66	0.75	2.21	2.81	3.14
28	269.4	254.9	251.8	4.3	0.43	0.58	0.66	2.32	2.96	3.32
32	302.8	286.9	281.6	4.1	0.41	0.55	0.63	2.48	3.17	3.54
36	325.1	308.1	300.1	4.1	0.41	0.55	0.63	2.67	3.41	3.77

Femur length

Week	Mean	10% Centile	5% Centile	CV	%SE_50	%SE_10	%SE_5	Width_50	Width_10	Width_5
20	32.6	30.0	29.0	6.9	0.69	0.93	1.06	0.45	0.56	0.61
24	43.2	41.0	40.0	4.7	0.47	0.63	0.72	0.41	0.52	0.58
28	53.0	51.0	50.0	3.7	0.37	0.50	0.57	0.39	0.51	0.57
32	61.9	59.0	58.0	3.3	0.33	0.45	0.51	0.41	0.53	0.59
36	69.3	67.0	65.0	3.2	0.32	0.43	0.49	0.44	0.58	0.64

Humerus length

Week	Mean	10% Centile	5% Centile	CV	%SE_50	%SE_10	%SE_5	Width_50	Width_10	Width_5
20	31.4	28.0	27.0	8.9	0.89	1.20	1.37	0.56	0.67	0.74
24	40.7	38.0	37.0	6.8	0.68	0.92	1.04	0.55	0.70	0.77
28	48.6	45.0	44.0	6.3	0.63	0.85	0.97	0.61	0.77	0.85
32	55.7	52.0	51.0	5.7	0.57	0.77	0.87	0.63	0.80	0.89
36	61.2	57.0	56.0	5.4	0.54	0.73	0.83	0.66	0.83	0.93

In addition the proposed sample size of 140 women per centre will allow for testing for differences in growth patterns across centres (25).

The following table lists, for every biometrical parameter, the number of subjects needed to detect as statistically significant the smallest meaningful difference d with type I error rate of 0.05 and a power of 0.80 with values of variance (σ^2) and correlation (ρ^2) as estimated using the data from the WHO randomized trial of calcium supplementation in low intake women (26).
The presented sample sizes have been calculated by applying the formula:

$$m = \frac{2(z_\alpha + z_Q)^2 \sigma^2 (1-\rho)}{n s_x^2 d^2}$$

Where d is the minimum difference in the rate of change per unit time (week) in the five

biometrical parameters by centre, n is the number of visits (n=8) and s_x^2 is the within subject variance (25). The values of d shown in the table indicates that the sample size needed for the level of precision requested will be very conservative to detect differences in rate of change between centres.

	Sample size	Variance	Rho	D (mm)
Bipariatal Diameter	32	10.2	0.34	0.1
Head circumference	23	174	0.28	0.5
Abdominal circumference	28	263	0.43	0.5
Femur length	16	5	0.32	0.1
Humerus length	35	11	0.32	0.1

4.2 Eligibility criteria: the prescriptive approach

In terms of study subject selection, the protocol will follow the same approach of the WHO Multicentre Growth Reference Study. Specifically, this involves enrolling participants with no known health, environmental, and socio-economic constraints on fetal growth. Through this prescriptive approach to sample selection, the fetal growth references generated will approximate as much as possible a standard. Immediately below are the eligibility criteria for study inclusion:

- High socio-economic status and high parental education in order to ensure that the curves will reflect as much as possible the true growth potential of fetuses, by limiting as much as possible the influence of environmental factors. Specific cut off points in family income and education have been identified by the MGRS and will be translated into socioeconomic indicators specific to the countries participating in the proposed study (9;27;28);
- Low altitude. Only women living at altitude lower than 1,500 m will be included in the study;
- Low mobility of the target population, in order to ensure compliance with the study, follow up for the study duration, and for potential future follow-up studies in infancy, childhood and eventually adulthood;
- Local presence of collaborative institutions with proven experience in conducting prospective studies among pregnant women and newborns.

The following will be the eligibility criteria for individual study subjects:

- aged ≥ 18 years (as younger women are still growing and their babies may be smaller at birth) and ≤ 40 years
- BMI 18-30
- singleton pregnancy
- gestational age at entry between 8+0 to 12+6 weeks based in LMP (confirmed by ultraongraphy, please see 5.1.1 for more detailed description)
- no history of:
  i) health, environmental or economic constraints likely to impede fetal growth,
  ii) need for long-term medication (including fertility treatment)
  iii) smoking currently or in the previous 6 months
  iv) recurrent miscarriage
  v) any previous baby delivered pre-term (<37 weeks) or with a birth weight <2,500g (at 37 w 5% of boys and 10% of girls in low-risk pregnancies will be ≤2500 g)

In addition, there must be no evidence in the present pregnancy of congenital disease or fetal anomaly. Participation in the study will cease if a major fetal anomaly is detected or serious illness develops leading to marked IUGR; however, all mothers recruited will be followed-up until the end of the study for the purposes of describing the whole population.

5. STUDY PROCEDURE

5.1. General principles of data collection: assessment of growth and related variables

Women in the first trimester (before 12+6 weeks gestation) attending antenatal care clinics providing ultrasonographical examinations will be approached by members of the study team and asked to participate. Women will be fully informed about the study objectives and procedures. Only women who sign a consent form will be enrolled into the study.

Fetuses will be scanned in the first trimester for the estimation of gestational age and then subsequently at monthly intervals for fetal biometry (29).

The ethical review board that has approved this study requested not to provide information on the sex of the fetus. The reason for this is that this is an international study and in some parts of the world disclosure of the sex of the baby is not permitted in order to prevent selective abortion of unwanted female fetuses. Information on the sex of the fetus will be provided only if the mother will ask for it and in countries where it is legal. All infants will receive an anthropometrical assessment after delivery, including measurement of birth weight (30). All pregnant women in the study will be administered a 24-hour dietary recall at entry into the study, and at approximately 28 and 36 weeks gestation to assess maternal nutritional status. This will allow us to ensure that women enrolled in the study have an adequate diet and that nutrient intake is in agreement with current pregnancy recommendations(30;31). At each visit, the obstetric history of participating women will be updated to collect information related to pathological processes that may affect fetal growth, and blood pressure and proteinuria will be measured according to guidelines contained in the study document: *A practical guide on how to measure blood pressure and test for proteinuria.*

No additional procedures will be added to routine antenatal care provided at the study centres, with the exception of 4-5 additional ultrasonographical examinations and three repeat 24-dietray recalls.

5. 1. 1 Dating by Ultrasound

Gestational age will be confirmed by measuring the crown-rump length (CRL) between 8+0 to 12+6 weeks based in LMP. Gestational age (GA) by CRL should agree with GA by LMP to within 7 days. If LMP and CRL agree, CRL measurement will be used for dating. The average of three measurements will be used. If GA by CRL and GA by LMP differ by more than 7 days, the woman is not eligible for the study.

To acquire the CRL measurements, the midline sagittal section of the whole fetus will be visualized with the fetus horizontal on the screen at 90 degrees to the angle of insonation (32). GA will be assessed by using the reference charts published by Robinsons and Fleming (32).

5. 1. 2 Fetal biometry

The first visit (dating scan) will be between 8+0 and 12+6 weeks, and subsequent visits for fetal biometry will be scheduled at approximately 4 weekly (+/- 1 week) intervals at 14, 18, 24, 28, 32, 36, and 40 weeks.

All scanning appointments will be arranged at time of dating scan and study enrolment. Re-scans will be performed within the allocated +/- 1 week windows. In those cases where rescanning does not occur within the allotted time the volunteers will be asked to attend at their next scheduled study appointment. All participants will be scanned in the lateral

recumbent position.

The compulsory ultrasound measurements to be obtained at all visits include the following biometrical parameters:

- Biparietal diameter (BPD)
- Head circumference (HC)
- Abdominal circumference (AC)
- Femur length (FL)
- Transcerebellar diameter (TCD)
- Humerus length (HL)
- Fetal Foot length (FFL).

At each examination, all measurements are to be obtained three times from three separately generated ultrasound images and uploaded electronically (with the associated images) to the data management system. The ultrasound images of BPD, HC, AC and FL should fill at least1/3rd of the monitor screen. The mean of the three measurements of each parameter will be used for clinical management purposes as per local protocols.

In addition, a full morphological evaluation (abnormality scan) will be conducted at 18-24 weeks following standard practices at each centre. Fetuses diagnosed with any minor abnormalities will be managed according to local clinical guidelines. If the clinical decision is to continue with the pregnancy the case will remain in the study. Fetuses with major abnormalities that may affect morphometric measurements will be excluded from further study. All infants will receive an anthropometrical assessment after delivery (18).

The following measurement techniques will be used:

- Biparietal Diameter-Technique: Measured from the outer-outer (BPD 1) and outer – inner (BPD 2) edges of the parietal bones in a cross-sectional view of the fetal head at the level of the thalami and cavum septum pellucidum or cerebral peduncles. The cerebellum is not to be included. The measurement should be obtained from an image with the midline echo as close as possible to the horizontal plane with the angle of insonation of the ultrasound beam at 90 degrees.

- Head Circumference-Technique: Obtained from the same image as BPD as follows: Measurement of occipito-frontal diameter (OFD) obtained by placing calipers on the outer borders of the occipital and frontal edges of the skull at the point of the midline across the longest part of the skull. The ellipse facility will be used to calculate HC as above.

- Abdominal Circumference- Technique: The sonographer will visualize the transverse section of the fetal abdomen as “close as possible” to circular including the stomach and the junction of the umbilical vein and portal sinus. The anterior-posterior (A-P) and transverse diameters will be measured with calipers placed on the outer borders of the body outline. The A-P diameter will be measured from the spine to the anterior abdominal wall and transverse diameter at a right angle to the A-P diameter. The image should be as round as possible. The ellipse facility will be to be used to calculate AC as outlined above.

- FL-Technique: Measured from an image of the full femoral shaft in a plane as close as possible to a right angle to the ultrasound beam. The distal femoral epiphysis is to be excluded.

- Transcerebellar Diameter (TCD): the TCD can be imaged from the sub-occipito-bregmatic view of the fetal skull, and measured from the second trimester onwards. The calipers will be placed on the outer-outer margins of the cerebellar poles. Three measurements will be obtained and submitted electronically. The mean of these three measurements will be used for clinical management purposes.

- Humerus length (HL): Measured from an image of the full humeral shaft in a plane as close as possible to at right angle of insonation. Mean of 3 measurements to be used for clinical purposes.

- Fetal Foot Length (FFL): Fetal foot length will be measured from the second trimester onwards. The foot is measured from either saggital or plantar views. The measurement is taken from the skin overlying the heel (calcaneus) to the end of the longest toe. Three measurements are to be obtained with mean measurement used for clinical management purposes after electronic submission.

5.1.3 Validity of fetal biometry measurements

It has been shown that the fetal anatomical parameters we are proposing to evaluate can be measured with high level of precision (intra-observer error less than 1% and intra-class correlation coefficients between 0.98 and 0.99, and inter- observer error in the order of 1-2%)(33;34).

5.1.4 Ultrasound Volume Acquisition Protocol

The ultrasonographical equipment that will be used in the study will allow for acquiring and storing 3-dimensional images. This feature is of critical importance for data quality purposes. Stored 3-dimensional images (volumes) could be used at a later stage to retake measurements that have been identified as error.

This section describes the techniques that will be used to acquire and store 3-dimensional images.

Except for a transvaginal scan during the first research visit (8 0/7 -12 6/7 weeks), 3D volume data will otherwise be acquired using a transabdominal probe. All volumes shall be systematically labeled using the comment feature of the Voluson E8 Expert system (e.g."LA1").

5.1.4.1 Recommended Image settings:

1. At least High Quality 2
2. CRI no greater than 2
3. No speckle reduction (SRI)
4. Harmonic imaging as needed
5. Acoustic focus adjusted for the anatomic region
6. Use widest image window required to capture volume of interest depending on fetal activity
7. Adjust magnification and image depth setting to fill at least one-half of screen

5.1.4.2 Initial Ultrasound Scan

At the first scan, an endovaginal probe will be used to acquire volume data about the gestational sac and contents (RIC 6-12, GE Healthcare, Milwaukee, WI).

Two volumes will also be acquired, using a 3D abdominal transducer (RAB 4-8, GE

Healthcare or RAB 2-5 for technically difficult subjects, GE Healthcare, Milwaukee, WI):

- Two images of gestational sacs — EV1, EV2 (transveginal)
- Two images of gestational sacs — SAC1, SAC2 (transabdominal)

5.1.4.3 Subsequent Ultrasound Scans

Sonographers will be instructed to prioritize acquisition of at least one good volume dataset in each region of interest (i.e. H1, H2, CH1, AC1, UA1, UL1, LL1)
Required Volume Acquisitions - 12 total

- Two images of the head — H1, H2
- Two images of the chest — CH1, CH1 (Not necessary after 30 wks)
- Two images of the abdomen — AC1, AC2
- Two images of the upper arm — UA1, UA2
- Two images of upper leg — UL1, UL2
- Two images of the lower leg — LL1, LL2

Notes:

H1	Transverse sweep from side of head includes BPD plane
H2	Transverse sweep from front of head (orbits)
CH	Transverse sweep (spine at 3-9 o'clock position, level 4 chamber view)
AC	Transverse sweep of abdominal circumference includes conventional 2D plane
UA	Sagittal sweep upper arm - include both ends of humeral diaphyses
UL	Sagittal sweep thigh - include both ends of the femoral diaphyses
LL	Sagittal sweep lower leg - include both ends of the tibial diaphyses

Optional Volume Acquisitions
These volume datasets can be collected depending on the technical expertise and interest of each participating site:

2 STIC heart	STIC1, STIC2
2 Lower arm	LA1, LA2
1 Head post fossa	H3

STIC Fetal heart acquisitions

Beginning at 18 weeks, STIC acquisitions of the heart are ideally acquired with the cardiac apex oriented anteriorly and a 10-45 degree angle between the inter-ventricular septum and ultrasound beam. Fetal movements should be minimal and the mother should temporarily suspend breathing during the STIC acquisition. Satisfactory 4D volumes will be more difficult after 30 weeks, depending on fetal position and other technical factors.

Typical STIC settings:

Volume sweep angle	25 degrees
Acquisition time:	12.5 second sweep

LA Sagittal sweep lower arm - include both ends of the humerus/radius diaphyses with full thickness of limb

H3 Posterior fossa including vermis - difficult after approximately 30 weeks

Fetal biometry and anthropometric techniques, equipment and standardization:
All instruments and techniques to be used in all centres will be standardised, i.e. equipment and training will be provided to each of the measurement teams. Equipment specifications to be considered are the following: 1-Commercially available high quality real-time ultrasound scanner.2- Less than 2 years old. 3- T/V and abdominal probes suitable for scanning throughout pregnancy. 4- Facility for on-line transfer of measurements and associated images. 5- Facility to "blind" measurements from examiner until after data transfer.

5.1.5 Safety of ultrasongraphical assessment

Ultrasonography in pregnancy is considered a safe procedures and in more than 30 years no fetal harm has been reported with use in the low-intensity range of gray-scale imaging (no Doppler) which is the technology that will be used in the proposed study (35).
Serial ultrasonography for research purposes accordingly to schedules similar to the one we propose has been previously approved in the context of other studies (36-39).
Following a recommendation of the WHO Scientific and Ethical Review Group, we conducted a systematic review and meta-analysis to evaluate the safety of human intrauterine exposure to ultrasonography (40).
We systematically searched electronic databases, reference lists and unpublished literature according to the following criteria:

- Type of studies: Trials and observational studies that assessed short and long term effects of exposure to ultrasonography during pregnancy.
- Type of participants: Women submitted to ultrasonography in pregnancy and their offspring.
- Types of exposures: B-mode or Doppler sonography during any period of pregnancy, for any number of times, using any equipment and transducers.
- Types of outcome measures: 1) adverse maternal outcomes, 2) adverse perinatal outcomes, 3) abnormal childhood neurological development, and 4) childhood malignancies.

The electronic search identified 6716 citations and 63 were selected for full text evaluation. Additionally, 19 citations were identified from secondary sources. A total of 58 references reporting data of 38 different studies were included: 16 clinical trials, 11 cohorts, and 11 case controls.
Ultrasonography in pregnancy was not associated with adverse maternal effects, impaired physical or neurological development or increased risk for malignancies in childhood.
According to the clinical trials, there was a weak association between exposure to ultrasonography and non-right handedness in boys (OR 1.26, 95% CI 1.03-1.54) and a slight decrease in mean neonatal length (WMD -0.26 cm, CI -0.45, -0.07) and head circumference (WMD -0.15 cm, CI-0.29, -0.01).
In conclusion, according to the available evidence, exposure to diagnostic ultrasonography during pregnancy appears to be safe..

5.1.6 Neonatal anthropometrical assessment

Neonatal body composition assessment during the first 24 hours, will be used to determine the growth outcome of each pregnancy on the basis of multiple postnatal measurements in order to be able to relate pre and postnatal anthropometrical measurements. After delivery, a trained investigator will take standardized head circumference, abdominal circumference (superior border of umbilicus), and thigh circumference (at the skin crease located midway between the knee and trunk with the lower leg at about 90 degrees in relation to the thigh measured in centimeters) with non-stretchable tape. The crown-heel length will be obtained by placing the supine infant, with extended legs, on a plastic newborn length board (Statiometer, Ellard Instrumentation, Seattle, WA). Skin fold caliper measurements (Harpenden) will be used to

document soft tissue distribution (41). These skin folds will include the triceps fold, anterior thigh fold, sub-scapular skin fold, and abdominal flank skin fold that will be made at each site twice and averaged. This data will be used to estimate percent body fat and lean body mass as an index of neonatal growth outcome. Other parameters of neonatal body composition will include ponderal index and birth weight (42).

5.1.7 Nutritional assessment

Adequate nutrition is one of the major requirements for selecting populations eligible for the study. Therefore the assessment and maintenance of adequate nutritional status is considered a critical component of the study activities.

At three times during the study follow up (at entry and at approximately 28 and 36 weeks) we will assess maternal nutritional status via anthropometry (weight, height, arm circumference, head circumference, skinfolds) (Table 1), as well as assessing the dietary intake (by twenty-four-hour recall). Anthropometry will be used to assess maternal nutritional status. Anthropometry indirectly assesses the relationship between the fat and fat free components of the body. Anthropometric measures are safe, non invasive, simple, and feasible in field conditions. The study nurse/nutritionist will take the anthropometric measurements. A standardized procedure will be used and the women will be requested to empty the bladder and wear only light underclothing under an examination gown. Measurements will be carried out by a female nurse/nutritionist in a private room according to the procedures described by Gibson (43). The time of the examination will be recorded to allow for diurnal variations. Having only one person performing the measurements will minimize inter-examiner errors. The equipment required is already available at the study site.

The study nurse/nutritionist will administer a 24-hours recall at each visit. For the 24-hour recall method, a specific interview technique is used to try to estimate exactly the quality and quantity of foods eaten during the previous 24 hours (43). This method results in the attainment of estimates of the intake of single nutrients. Compliance is high because the respondent burden is low. The interview takes approximately twenty minutes. The quality of the information collected is dependent on the respondent's motivation and ability to recall and on the persistence of the interviewer (43). Repeated twenty-four-hour recalls on the same individual allow estimations of the individual's usual dietary intake over a long period of time (43). In our study the twenty-four-hour recall will be repeated three times for each woman. Briefly, this will be the procedure implemented: A nutritionist will administer a 24-hour dietary recall according to a standardized and previously published procedure and questionnaire by asking the woman to recall her exact food intake during the previous twenty-four-hour period (43). The nutritionist records a detailed description of the foods and beverage consumed. Quantities of food are estimated in household measures and entered on a data collection form. Subsequently, local food composition tables are used for the conversion of food to nutrients. This method has been successfully used in previous studies focusing on nutrition, fetal growth and other pregnancy outcomes conducted by investigators who are taking part in the proposed study.

Table 1. Description of maternal anthropometric measures.

Measure	Procedure
Weight	Weight will be measured using a beam balance with nondetachable weights. Weight will be recorded to the nearest 0.1 kg (43).
Height	Height will be measured in the standing position using a stadiometer and recorded to the nearest millimeter. If the reading falls between two values, the lowest millimeter will be

	recorded (43).
Mid-upper arm circumference	Measurements will be taken using a flexible fiberglass tape wrapped around the upper left arm, at the midpoint between the acromion process and the tip of the olecranon. Measurements will be recorded to the nearest millimeter (43).
Head circumference	A flexible fiberglass tape will be used. The tape will be placed above the supra-orbital ridges and over the part of the occiput which gives the maximum circumference. Measurements will be taken to the nearest millimeter (43).
Skinfolds	Left triceps and left scapular skinfolds will be taken using a Lange skinfold thickness caliper. The combination of one body and one limb skinfold to assess body fat is recommended by most investigators (43).

5.2 Ancillary studies

Some ancillary studies will be considered for implementation in the context of the main study. These studies have not yet been submitted for scientific and ethical review. Detailed protocols of the ancillary studies will be presented when funding will be available and the implementation of the studies will be feasible. The following is a brief description of ancillary studies under consideration to provide the reviewers with ideas about potential expansions of the main study:

5.2.1 A follow-up study of preterm infants (>26 but <37 weeks) born in the study cohort, to develop postnatal growth standards for preterm newborns;

5.2.2 A study including all live births and stillbirths over 12 months at the participating centres to determine the relationship between gestational age at delivery, birth weight and perinatal mortality and morbididy;

5.2.3 Anthropological study of science and technology as socio-cultural systems.
Based on ethnographic research methods (interviews, participant observation) the interaction of participating actors in the research will be studied with the aim of unraveling, linking and interweaving different knowledge systems that are shaping and shaped by ultrasound. Interviews with pregnant women are geared to understanding local socio-cultural notions of desirable fetal growth and pregnancy management, how these notions are influenced by ultrasonography, and women's perceptions and experiences of being research subjects in a study that will inform medical practice and guide future medical research. Interviews with the scientists performing the ultrasonography and other anthropometric measurements, and participant observation of the study procedures will be conducted to: 1.) Understand the processes of standardization, 2.) Gain insight into the viewpoints of the clinicians and scientists conducting the study on the importance of developing fetal growth reference standards, and the use of ultrasonagrophy to achieve this goal, and 3.) To observe the interactions between the women patients and scientists in terms of how these interactions are structured by power, and gender relations. Knowledge of existing ultrasound and clinical practices in the different localities will be used to inform and support a sustainable dissemination of new fetal growth standards as part of routine obstetric care based on the concept that standards are and work as 'situated' as well as distributed knowledge (44).

5.2.4 Fetal neurobehavioural development study
Fetal neurobehavioral development will be assessed by recording fetal heart rate and

movement patterns by means of computerized electronic fetal monitoring accordingly to a protocol that has already been applied in developing country and low-resource settings (45).

5.2.5 Studies of fetal growth in unselected or at-risk populations.

Fetal growth assessment will be conducted also in unselected populations in a subset of participating settings where environmental constraints are more likely to affect normal fetal growth. These studies will provide valuable insight into variations from optimal growth and the possible environmental determinants for these variations. In addition studies could be conducted in populations with specific risk factors that might affect fetal growth (e.g. endemic malaria)

5.3 Data management

Data will be collected via an internet based data management system as done with other WHO coordinated studies. All biometric measurements will be photographically documented to allow for image checking and revision.
The online data collection system will allow for real time solutions of queries or other problems in data collection, such as missing or non valid data as well as for online checking of images by the international coordination unit.

5.4 Data analysis

Population percentiles will be calculated by applying polynomial regression methods, using the generalized estimating equations method, to model the mean at specific gestational ages, taking into account correlations between repeated measurements on the same subjects (25). To estimate the standard deviations at each gestational age, we will use the method of the absolute residuals proposed by Altman (1993). This methodology takes into account the increase in variance with advancing gestation typical of fetal biometry data (46). Differences in linear growth among populations of different geographical origin (categorized by study site) will be tested by evaluating the proportion of total variability in fetal biometrical measurements attributable to sites and individuals, as well as differences among sites and the effect of excluding sites on the percentiles of the total sample, as done in the MGRS(47). The same procedure will be used to test for differences in growth related to sex of the fetus(48).

6. DISSEMINATION OF RESULTS

Dissemination of results will be achieved through the submission of articles to international and national peer review journals, a WHO publication, and presentations at national and international meetings. Results will be also presented at study centres and discussed with the local staff and community. It should be stressed that the results of the study will represent the first step towards the development of guidelines for the standardized evaluation of fetal growth in pregnant women. Post study dissemination efforts that will precede adoption of these standards within countries will include validation of the results in other population groups, and evaluation of the clinical and public health implications of using the standards in populations with risk factors for less than optimal fetal growth (e.g. malnutrition, infections, etc.).

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Form 4a. Ethical considerations

Note: Ensure all information requested in the corresponding section of Part 1 is provided

Appendix 1. Consent form[Institutional Letter Head]

[Name of PI]

World Health Organization

Information sheet for women attending the antenatal care clinic participating in the research: WHO multicentre study for the development of growth standards from fetal life to childhood: The fetal component

My name is ……and I am part of a team conducting a study on fetal growth coordinated by the World Health Organization.

Purpose:

Being able to follow how your baby is growing is very important to assure that the pregnancy is progressing fine for both you and your baby. We can measure the growth of your baby while inside the womb by ultrasound. The reason for doing this research is to understand how babies normally grow, so that if there is something wrong in the growth of the baby this can be detected by comparing his/her growth with the normal standards of growth.

Some information about ultrasounds

Ultrasounds are used normally during pregnancy to look at how babies are growing and developing. Ultrasounds have been used safely during pregnancy for more than thirty years and no side effect has been detected. As an extra precaution, we have extensively reviewed the studies that have investigated potential side effects of repeated ultrasounds examinations in pregnancy and found no evidence of harm. Usually three (specify number if different than 3) ultrasound examinations are routinely done during a normal pregnancy. In this study, a total of 8 ultrasounds will be performed.

Procedures

If you accept to participate in this project you will receive eight ultrasound examinations. The first exam will be between 8 and 12 completed weeks of pregnancy and will be performed using transvaginal ultrasounds. This is standard practice at this clinic. We will then follow you up until discharge from the hospital after delivery. The following exams will be at 14, 18, 24, 28, 32, 36 and 40 weeks of pregnancy using transabdominal ultrasounds. At each exam we will measure the growth of your baby. At this clinic three (specify number if different than 3) ultrasound examinations are routinely done during pregnancy, however we will perform eight examinations to be able to study more in detail the growth of your baby. This implies committing to ultrasound examinations more of your time during pregnancy by coming to the clinic five extra times.

We will also measure your blood pressure and test your urine for the presence of protein that may indicate the presence of pregnancy complications. These procedures are conducted routinely on all pregnant women receiving care at this clinic. We will also revise your pregnancy card to make sure that your pregnancy is progressing fine. We expect that each examination will take approximately 30-40 minutes.

At entry into this study, and at 28 and 36 weeks, you will be asked by a nutritionist about your diet. The nutritionist will give you advice on how to make sure that your diet is adequate and that your are getting all the food you need for your pregnancy. The nutritionist will also take measurements of your weight, height, arm circumference, head circumference and skinfolds) When your baby will be born, the nutritionist will weigh and measure him/her. The nutritionist will also take photographs of the baby.

If during the examinations problems are detected that need further medical care, you will be referred to the hospital where you receive antenatal care for appropriate treatment according to national guidelines. The most appropriate care will depend on the nature of the complication detected and will be decided by the attending physician in care of your pregnancy in consultation with specialists. You will be kept informed about the treatment options.

Side effects

There is no known side effect of repeated ultrasound examinations during pregnancy.

Risks and discomforts

By participating in this research you will not be exposed to risks or discomforts. Ultrasound examinations will be performed while you will be lying comfortably on a bed on your side.

Benefits

There is no direct benefit for you, however if you participate in this research, complications of pregnancy maybe found and, if you agree, you will be immediately referred for appropriate care. This study is not intended to detect fetal problems such as malformations. However should this be the case we will ensure that you will be promptly informed of the possible fetal abnormality by the attending obstetrician in charge of the your pregnancy who will also inform you of the availability of a trained counsellor providing psychological and emotional support. The study will have important benefits for all pregnant women and their babies because it will allow us to develop the standards of growth for babies during pregnancy that will help doctors to better interpret the results of ultrasounds examinations.

Incentives

You will not be provided any incentive to take part in this study. However you will be reimbursed with ……. for your travel expenses.

Confidentiality

The information that we collect from this research project will be kept confidential. Information about you that will be collected from the study will be stored in a file that will not have your name on it, but a number assigned to you instead. The name associated with the number assigned to each file will be kept under lock and key and will not be divulged to anyone except the local principal investigator (Dr……) and the doctors that are following your pregnancy at the antenatal clinic.

Right to refuse or withdraw

You do not have to take part in this research if you do not wish to do so, and refusing to participate will not affect your treatment at this centre in any way. You will still have all the benefits that you would otherwise have at this centre.

You may stop participating in this research at any time that you wish without losing any of your rights as a patient here. Your treatment at this centre will not be affected in any way.

You do not have to make a decision now. If you wish, you could take more time, take this information sheet home with you, and let us know what you decide, in your next visit

Whom to contact

If you have questions you may ask them now or later. If you wish to ask questions later, you may contact any of the following:

This proposal has been reviewd and approved by The Research Ethics Committee at the institution ……., which is a committee whose task is to make sure that research participants are protected from harm. If you wish to find more about the IRB, please contact:……

Certificate of consent

I have been invited to take part in the research project: **WHO multicentre study for the development of growth standards from fetal life to childhood: The fetal component.** I have read the foregoing information, or it has been read to me. I have had the opportunity to ask questions about it and any questions that I have asked have been answered to my satisfaction. I consent voluntarily to participate as a subject in this study and understand that I have the right to withdraw from the study at any time without in any way affecting my further medical care.Print name ……. Signature …….. Date .....

If illiterate

Print name of literate witness……..Signature of witness…… Date .....

Print name of researcher……..Signature of researcher…… Date .....

1. Describe how the research addresses a demonstrated public health need and a need expressed by women and/or men.

This issue has been addressed in the description of the project.

2. Explain how the research contributes to identifying and/or reducing inequities between women and men in health and health care?

N/A

3. Describe plans for disseminating results and sharing knowledge with the research subjects and wider community.

Please see section 6 for a description of dissemination strategies.

4. Does the nature or topic of the research make it important that the researchers are women rather than men or vice versa? Please explain. What is the sex composition of the research team and what are their duties and responsibilities in the proposed research?
N/A