Maternal Fetal Medicine Units Network

PROTOCOL

SCREENING FOR RISK FACTORS FOR SPONTANEOUS PRETERM DELIVERY

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Prepared by the

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August 1, 1994

Preface

Protocol Edition

This protocol, "Screening for Risk Factors for Spontaneous Preterm Delivery', is maintained by the MFMU Network's Biostatistical Coordinating Center (BCC) during the course of the study. The final version, dated September 18, 1992, was approved by the MFMU Network Steering Committee on September 18, 1992. Any subsequent changes, with the reasons for the changes and the dates of formal approval by the Steering Committee and the Network Advisory Board, are summarized below.

The protocol for the ancillary study being conducted by the nurse coordinators, is provided in Appendix E. It is maintained by the MFMU Network's BCC during the course of the study. The final version, dated September 18, 1992, was approved by the MFMU Network Steering Committee on September 18, 1992. Any subsequent changes, with the reasons for the changes and the dates of formal approval by the Steering Committee and the Network Advisory Board, are summarized below.

Revised - April 20, 1993

One change has been made to the protocol. Patients who are eligible but refuse consent are not to be included in an observational component of the study.

Revised - January 14, 1994

One change has been made to the protocol. Patients who have a score of ≥ 7 on the gram stain and a vaginal pH of > 4.5 will be considered positive for B.V.

Revised - August 1, 1994

A clarification has been made to the protocol to reflect the fact women are enrolled as early as 23 weeks. The primary outcome measure of delivery following preterm labor/PROM from 24-34 weeks has been revised to 23-34 weeks.

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CHAPTER 1

Introduction

1.1 Study Abstract

In this study, tests currently advocated to predict spontaneous preterm rupture of membranes (PROM) or preterm labor resulting in a preterm delivery will be evaluated individually, and in combination, to determine their effectiveness in defining women at risk for spontaneous preterm delivery. The tests to be evaluated include a demographic, behavioral, psychosocial, anthropometric and historical profile, serum and plasma levels of various proteins such as CRP, major basic protein and alpha-fetoprotein, vaginal ultrasound evaluation of the cervix, a cervical digital examination, the presence of bacterial vaginosis and trichomonas, and both vaginal and cervical fetal fibronectin.

Three thousand women enrolling for care at the participating centers will be studied. A subsidiary study of 150 twin gestation pregnancies will also be performed. The tests are administered during two major screening visits at 24 and 28 weeks gestation, and two minor visits at 26 and 30 weeks. The patients' care givers are blinded to results of the screening tests, with a few exceptions. For the singleton pregnancies, the serum test samples will be stored until after delivery when a nested case-control study will be conducted on the serum measurements for each case of spontaneous preterm delivery and two matched controls of term deliveries.

1.2 Objectives

The primary objective of this study is to find tests which define a group of patients with at least a twofold increase in risk of spontaneous preterm delivery from 23 to 34 weeks. If several tests are found which satisfy these criteria, an optimal subset (the smallest combination with the best predictive ability) will be sought.

The study will also examine the following: the predictive ability of the tests for preterm delivery from 23 to 28 weeks, reasons for indicated preterm deliveries, and associations of the screening tests with fetal growth retardation.

1.3 Purpose of the Study Protocol

This protocol describes the background, design and organization of the study and may be viewed as a written agreement between the study investigators. It is reviewed by the Data Monitoring and Safety Committee and Network Advisory Board, and approved by the Steering Committee and the Institutional Review Board of each clinical center before recruitment begins. Any changes to the protocol during the study require approval of the Steering Committee and review by the Advisory Board.

A manual of operations supplements the protocol with detailed specifications of the study procedures.

CHAPTER 2

Background of the Study

Preterm birth is the leading cause of perinatal mortality, and occurs in 4.4% to 21.5% of pregnancies, depending on the population studied [84]. Data from the Alabama March of Dimes study, confirmed in other studies, indicate that nearly 50% of white neonatal deaths and 60% of black neonatal deaths occur in premature infants weighing between 500 and 1000 grams at birth.

A considerable amount of work has been done in search of risk factors for preterm labor and preterm delivery. Most risk factors described to date are derived from retrospective analyses and for the most part have come from demographic, behavioral, anthropometric and historic areas. For example, black race is a major risk factor for preterm delivery [1,2]. Cigarette smoking, alcohol use and drug use have each been associated with preterm birth although the predictive value for each is relatively low [3,4,5,6,7]. Nevertheless, in order to place the various tests being evaluated in context, it is important to characterize the population in those terms. Other major predictors of preterm delivery are low prepregnancy weight or other indicators of maternal thinness, and low weight gain during pregnancy [8,9].

A number of medical, obstetrical history and obstetrical factors in the current pregnancy are also related statistically to preterm delivery. Obstetric history, especially a second trimester abortion, or a previous preterm birth, is a major predictor of preterm delivery in the current pregnancy [5,10,11,12]. Obstetric factors such as multiple gestation, bleeding, and placenta previa have all been related to preterm delivery. From these retrospective studies, a number of risk scoring systems have been created, the best known being the Creasy-Papiernik system [13,14,15,16]. These systems are characterized by low sensitivity and positive predictive values and a very high false positive rate. Nevertheless, most have been able to define a group of patients with approximately a 2 to 2 1/2 fold increase in relative risk of preterm delivery. They work poorly in primiparas and somewhat better in multiparas, mainly because, in virtually every study, the best predictor of a preterm birth in the current pregnancy is a preterm birth in a previous pregnancy. For primiparas, with the exception of multiple gestation, most scoring systems based on demographic and behavioral factors cannot define a relative risk above 2 for preterm delivery for any group of women.

There have also been a number of studies which have evaluated premature cervical ripening, especially dilation, as a predictor of spontaneous preterm delivery [17-29]. Virtually every study found a significant increase in spontaneous preterm delivery associated with cervical dilation in the late second or early third trimester, but again the relative risk has generally not been greater than 2 to 3 and the sensitivity and positive predictive value of cervical dilation as a screening test is low. Furthermore, the "attributable risk" associated with this risk factor has been relatively low. More recently, measurements of cervical length by ultrasound, and a lower uterine segment indentation have been associated with risk of preterm labor with reasonably good sensitivity and positive predictive values [30,31-43]. An information sheet dealing with endovaginal ultrasound of the cervix presented by Anderson at the 1992 SPO meeting is included in Appendix C.

There has been much research activity examining an increase in preterm contractions as a predictor of spontaneous preterm delivery, culminating in the current controversy about home uterine activity monitoring [44,45,46,47,48,49]. Nevertheless, there appears to be an association between increased levels of contractions and eventual spontaneous preterm delivery. Understanding the value of any measure of contractions to predict preterm delivery is complicated by the great variability in method of contraction determination, the time in pregnancy measured, the frequency of contractions considered a positive test, and the specific clinical endpoint. Apparently contractions increase predominantly in the 72 hour period prior to labor so that the predictive value of contraction monitoring at intervals greater than 72 hours is relatively poor. Moreover, the time to perform the various monitoring tests is substantial. Therefore, monitoring will not be part of this study.

In recent years, various types of infections and other measures of infection have been associated with spontaneous preterm delivery, although most of the results have been controversial [50,51,52,53,54,55]. The analysis of the VIP study is nearing completion and results suggest that bacterial vaginosis [56-63], trichomonas [64,65,66], and gonorrhea are associated with an increased risk for spontaneous PTD. Other markers of infection which have been suggested as screening tools for an at risk population include vaginal pH [67], C reactive protein [68,69], and various cytokines. There are few prospective studies testing the usefulness of these markers in a general population.

There are several studies which suggest that abnormal levels of fetal or placental protein in the cervix, vagina or the maternal serum may be predictive of spontaneous PTD. For example, an elevated maternal serum alpha-fetoprotein has been specifically associated with a several fold increased risk of PTD [70], as has another protein of fetal or placental origin - major

basic protein [71]. As another example, in a study of women with preterm contractions, Maymon, et al found that having a low level of placental isoferritin (up to 10 U/mL) was a sensitive predictor of preterm labor (PPV 59%, NPV 71%) [72]. Other proteins that have been statistically associated with preterm delivery or preterm labor include plasma protein A_2, HPL, and Schwangen-Schafts protein 1, but all had much lower positive predictive values. More recently, placental corticotropin releasing hormone has been found to be elevated in twin pregnancies and in pregnancies which ultimately deliver following spontaneous preterm labor. Interestingly, those pregnancies which ended in preterm delivery, but were associated with an infection, did not have elevated levels of corticotropin releasing hormone [73,74,75].

There has been a great deal of controversy related to the relationship of maternal hematocrit and preterm delivery, especially in black women. Liberman, et al claimed that a substantial part of the higher preterm delivery in black women could be explained by low hematocrit [76]. However, Klebanoff, et al and Lu, et al disagreed [77,78]. The relationship of both high and low blood pressures and spontaneous and indicated preterm delivery has been investigated previously although the results are controversial. Clearly, higher blood pressures are associated with a greater degree of "indicated" preterm delivery. However, Goldenberg, et al have recently shown that lower systolic and diastolic pressures were both associated with increasing spontaneous preterm delivery [79].

Most exciting has been the recent report of vaginal and cervical fetal fibronectin as a predictor of spontaneous PTD [80,81]. Fetal fibronectin is a glycoprotein with a molecular weight of 450,000 and is normally found in the space between the membranes, placenta and the endometrium. Its presence in the cervix and vagina in women with intact membranes in the midtrimester has been reported to predict preterm delivery in hospitalized women with uterine contractions, and in women at high risk for preterm labor. In high risk women, cervical or vaginal fetal fibronectin testing has great promise in identifying women at risk of preterm delivery. For example, with weekly sampling, the average interval between the first positive fetal fibronectin result and preterm delivery was nearly 5 weeks. Alternatively, sampling every two weeks between 24 and 34 was as effective as weekly collection in identifying women who will deliver prematurely. Cervicovaginal fetal fibronectin was expressed at least 2 weeks prior to delivery in 84% of women delivering prematurely. The sensitivity of bi-monthly sampling (88.9%) is similar to weekly sampling (92.6%) while the specificity increases (from 46.3% to 61.7%). These observations suggest that fetal fibronectin can be successfully incorporated into standard management of high risk patients and that a negative fetal fibronectin result between weeks 24 and 34 is strongly associated with continuance of pregnancy for an additional two weeks [81].

Despite the availability of these potentially interesting screening tests, it is not clear how useful any of these are clinically, or whether a combination of some of these tests into a scoring system would be useful. This study proposes to evaluate the usefulness of the most promising of these tests alone, and in combination, to define populations of women at high risk for spontaneous preterm delivery (SPTD). Furthermore, by correlating which tests are positive in relationship to one another, and which tend to be associated with PROM versus spontaneous labor, some additional information may be ascertained relating the specific test result to the event responsible for the preterm delivery.

CHAPTER 3

Study Design

3.1 Design Summary

The main study is an observational prospective study of 3000 women with singleton pregnancies at the participating MFMU Network clinical centers. All women registering for care at each of the centers before 24 weeks gestational age (with a few exceptions detailed below) are potentially eligible for the study. No attempt is made to select women at high risk for preterm delivery except in the subsidiary study, an observational prospective study of 150 women pregnant with twins.

Eligible women, who consent to participate, make four clinic visits at 24, 26, 28 and 30 weeks gestation, at which times the screening tests are conducted. Maternal and neonatal followup continues until discharge from the hospital after delivery.

The study procedures are conducted in a uniform manner throughout the participating center sites. Appendix A summarizes the key design features.

3.2 Sampling Scheme for the Singleton Study

Because there will be many more women eligible for the singleton study than can be enrolled, a selection procedure must be used. The consent rate, which is projected to be less than 50% on average, may vary considerably among different strata of the population at a center, so that even if a random selection procedure is used there is a possibility that the sample will not be representative of the population at that center. To overcome this, recruitment of women into strata defined by race (black or non-black) and parity (nulliparous or multiparous) is partially controlled. A quota is set for each of the four subgroups (black nulliparous, non-black nulliparous, black multiparous and non-black multiparous) based on the distribution of these factors and each center's enrollment. When the quota for one of the subgroups is filled, recruitment into that subgroup is stopped and recruitment efforts are then directed towards the other subgroup(s).

Each of the ten participating centers for the singleton study, regardless of size, is expected to recruit approximately an equal number of women. Recruitment will be evaluated at five 8-week intervals. Each interval, the difference between each center's recruitment goal and the number of patients each center has recruited will be assigned equally across those centers that have reached their recruitment goal.

3.3 Inclusion Criteria

1) Less than or equal to 24 weeks 6 days (181 days) gestational age based on clinical information and evaluation of the first ultrasound as described below.

2) An ultrasound performed after 15 weeks and before enrollment, i.e. no later than 24 weeks 6 days. A woman may have the ultrasound on the day of the first study visit but she cannot be considered as enrolled until it is performed.

3.3.1 Gestational Age Determination

The gestational age for the first inclusion criterion is determined in the following manner:

1) The first day of the last monthly period (LMP) is determined and a judgement made as to whether or not the patient was sure of the date.

2) If unsure, the ultrasound measurements obtained at the patient's first ultrasound examination are used to determine the project gestational age, by the standard method of ultrasound gestational age determination at that institution.

3) If the patient is sure of her LMP, and the ultrasound confirms this gestational age within ($\leq$) 10 days, the LMP derived gestational age is used to determine the project gestational age.

4) If the ultrasound determined gestational age is more than 10 days different from the LMP generated gestational age, the ultrasound is used to determine the project gestational age.

3.4 Exclusion Criteria

Patients with any of the following conditions or diagnoses are excluded from the study:

1) More than two fetuses in a multifetal gestation

2) Prenatal follow-up or delivery planned elsewhere

3) Placenta previa with the placenta covering the internal os

4) Major congenital anomalies or fetal death

5) Cerclage

6) Known AIDS or HIV positive

Enrollment in other studies, such as the multifetal gestation group in the High Risk Aspirin Study, does not necessarily preclude enrollment in this study. However, care will be taken to be sensitive to the wishes of individual women to ensure that they are comfortable in joining two studies and that compliance in the first study is not jeopardized.

3.4.1 Informed Consent Criteria

Written informed consent must be obtained before entry into the study. Each center will develop its own consent form according to the requirements of its own institutional review board (see Appendix B for "Sample Informed Consent Form", developed by the PTD Prediction Subcommittee).

3.5 Study Outcomes

Outcome data will be collected on all project participants as well as those women who were screened but refused to participate. In the primary analysis, a distinction will be made between preterm deliveries which followed spontaneous PROM and/or spontaneous preterm labor and those indicated for maternal or fetal reasons. The preterm deliveries will be categorized by gestational age groupings of 23 to 27 weeks, 28 to 34 weeks and 35 and 36 weeks for each screening test in each screening time period.

3.5.1 Primary Outcome

The primary outcome will be delivery from 23 to 34 weeks following spontaneous preterm labor or PROM.

3.5.2 Secondary Outcomes

1) Delivery following spontaneous preterm labor or PROM, classified differently from the primary outcome; i.e., from 23 to 27 weeks, 28 to 34 weeks and 35 to 36 weeks

2) Preterm delivery (< 37 weeks) as a result of spontaneous PROM

3) Preterm delivery (< 37 weeks) as a result of spontaneous labor

4) Preterm delivery (< 37 weeks) as a result of maternal or fetal indication (the indication(s) will be recorded)

5) Episodes of preterm contractions and preterm labor, how many, at what gestational age they occurred, and the nature of the treatment, if provided

6) Gestational age at PROM

7) Gestational age at delivery

8) Fetal growth retardation (defined as less than the tenth percentile for gestational age using Brenners standards [82])

9) If preterm, presence of amnionitis or post partum endometritis

10) Stillbirth and type of stillbirth

11) Apgar scores

12) Neonatal morbidity and mortality

13) Presence of congenital anomalies

3.6 Nested Case-Control Study

For efficiency, a case-control study will be set up for the singleton pregnancies after enrollment and follow-up are complete to compare laboratory test values of the serum samples and other variables between the cases (spontaneous preterm deliveries from 23 to 34 weeks) and two matched controls (term deliveries) per case. In this way, serum measurements will only be performed on approximately 315 samples.

CHAPTER 4

Study Procedures

4.1 Screening

The nurse coordinator (or her designate) will evaluate each woman enrolling for prenatal care prior to 24 weeks gestational age to determine her strata classification. The date at which she will reach 24 weeks will be noted. For each calendar week, a list, continuously updated, is generated of women approaching 24 weeks. The study staff will then select women from the list according to the quota sampling scheme (see "Sampling Scheme for the Singleton Study" on page 3.1). The chart of each woman selected is reviewed to ensure that none of the exclusion conditions "Exclusion Criteria" on page 3.3 exist and that the gestational age is appropriate for entrance into the project (see "Gestational Age Determination" on page 3.2). If she is eligible, she is contacted and informed consent requested. If an abdominal ultrasound examination has not been performed at 15 weeks or later, one must be arranged prior to entrance into the study. The results of the ultrasound must also be reviewed to check for exclusion conditions and, if it is the patient's first ultrasound, gestational age.

4.2 Requesting Consent

The informed consent document includes details regarding the study design and objectives as well as possible maternal and fetal risks. Risks associated with the study will be discussed, as will the fact that the results of only a few specific tests will be forwarded to her physician under circumstances of increased risk.

If a 15-24 week abdominal ultrasound has not been performed as part of clinical care and has to be arranged to satisfy the eligibility criteria, the ultrasound results will be made available to the patient's physician. However, the results of all other tests or findings except for fetal death, major anomalies, the presence of regular painful contractions, prolapse of membranes through the external os, oligohydramnios or polyhydramnios, placenta previa or cervical dilation of > 2 cm in a primipara and > 3 cm in a multipara will be blinded to the patient and the clinicians involved in patient care.

Upon receiving the appropriate information and electing to participate in the study, each woman will be asked to sign the appropriate informed consent form. A copy of the signed consent will be provided to the patient.

4.3 Enrollment, Data Collection and Test Schedule

After informed consent and, if applicable, the ultrasound, the patient is formally enrolled in the study at the 24 week visit (Visit 1). This visit may, in fact, take place from 23 weeks 0 days to 24 weeks 6 days, but must not be arranged any earlier or later. Each woman will be interviewed to collect demographic and behavioral data, medical history, previous pregnancy history if applicable and present pregnancy history. Maternal anthropometric data will be obtained and blood and urine specimens will be collected.

A pelvic examination will then be performed. Cervical and vaginal secretions will be collected for the measurement of fetal fibronectin and vaginal pH. A gram stain, a wet prep for trichomonas and a KOH preparation for yeast will be performed.

Upon completion of the specimen collection, a digital examination will be performed to assess the position, consistency, and dilation of the external os and, if the external os is dilated, of the internal os. Cervical length and the condition of the lower uterine segment will be recorded, as well as the station of the presenting part, if palpated.

The vaginal ultrasound examination will be performed next. Data to be collected will include cervical length, and the extent if any, to which the membranes are protruding into the cervix. A urine specimen will be obtained.

Two weeks after the first visit, at about 26 weeks, the patient will be seen for a pelvic examination to collect cervical and vaginal fetal fibronectin, to perform a KOH preparation for yeast, and to administer the psychosocial questionnaire (see Appendix E). Visit 2 may take place from 25 weeks 0 days to 26 weeks 6 days, but must not be arranged any earlier or later. If the patient presents for this visit (Visit 2) after 26 weeks 6 days, Visit 3 procedures should be substituted and Visit 2 will be considered missed.

Four weeks after the first visit, at about 28 weeks, data similar to that collected at the first visit will be obtained, except that much of the demographic and historical data will be omitted. Visit 3 should be scheduled from 27 weeks 0 days to 28 weeks 6 days. If the patient misses this appointment, Visit 3 can be rescheduled, as soon as possible, up to 31 weeks 6 days.

If Visit 3 occurs before 30 weeks 0 days, the last visit (Visit 4) should be scheduled approximately two weeks later. If the patient misses her appointment, Visit 4 may be scheduled at any time up until 31 weeks 6 days. If Visit 3 takes place from 30 weeks 0 days to 30 weeks 6 days, the last visit should be scheduled at or before 31 weeks 6 days, at an interval of at least a week. If Visit 3 occurs after 30 weeks 6 days, no Visit 4 should be scheduled. At Visit 4 the patient will be seen for a pelvic examination to collect cervical and vaginal fetal fibronectin and to perform a KOH preparation for yeast.

4.4 - Test methods

These are presented by order of performance.

1) Demographic Data

Data to be collected by interview will include:

- Date of birth
- Race
- Parity (FPAL)
- Marital status
- Family income level
- Type of job
- Source of funding for maternity care
- Educational level
- Gestational age at first prenatal visit

2) Behavioral Data

At the first visit, the following behavioral information will be gathered:

- Cigarette smoking
- Alcohol use before and during pregnancy
- Use of illicit drugs before and during pregnancy

3) Medical History

Historical medical factors such as the history of DES exposure, the presence of chronic hypertension including previous use of medication, insulin requiring diabetes, renal disease, heart disease limiting activity, hemoglobinopathy, chronic lung disease and history of pelvic inflammatory disease will be ascertained. Each of these will be defined as present or absent.

4) Previous Pregnancy History

Previous pregnancy history will be collected including total pregnancies, number of term/preterm deliveries, and stillbirth and midtrimester abortion. If preterm, information will include whether the delivery followed spontaneous preterm labor/PROM, or followed specific maternal or fetal indications for delivery such as hypertension, diabetes, or distress.

5) Current Pregnancy History

Current pregnancy history will be collected including:

- Bleeding and the gestational age at which it occurred
- Uterine anomalies or fibroids
- Occurrence of urinary tract infections
- CVS or amniocentesis performed
- Surgery performed
- Frequency of uterine contractions
- Frequency of intercourse
- Other purported symptoms of preterm labor such as pressure, discharge, back pain, etc.

Blood pressure measurements and the results of certain laboratory tests (conducted outside the study) from the pregnancy including urine, gonorrhea and chlamydia cultures and hematocrit will be recorded.

6) Anthropometric Measurements

The following physical measurements will be recorded:

- Reported pre-pregnancy weight
- Height: measured with a tape mounted securely against the wall and a block placed firmly on top of the skull. The woman will not wear shoes.

- Weight: the measurement will be taken with the woman wearing light clothing and no shoes.

Weight gain per week prior to the first visit and weight gain per week in the late second trimester will be calculated. Using the weight and the height obtained at the first study visit, the body mass index (weight (kg)/height ^{2}(m)), will be determined.

7) Blood Sampling For Proteins

At any time of the day, but prior to cervical examination and not necessarily in a fasting state, 10cc of whole blood for serum, and 10cc for plasma will be drawn from each woman at the 24 and 28 week visits. Each tube of blood will be centrifuged at room temperature and the serum or plasma will be divided into approximately 5 two cc aliquots. The serum and plasma will be labeled and frozen at -80^{o}c. No more than 2 hours will elapse between blood drawing and freezing. The serum and plasma will be stored locally until selected for analysis of proteins including:

- Serum placental isoferritin: to be measured using the anti-human placental ferritin CM-H-9 monoclonal antibody.

- Major basic protein: to be analyzed using a competitive binding radio immunoassay with a mouse monoclonal antibody (J 14 7A2). 100 micro-liters of serum will be used for each assay

- C-reactive protein

- Alpha feto-protein

- Cytokines

In addition, there is a lot of activity now being devoted to relating preterm delivery to circulating breakdown products from various basement membrane proteins, enzymes which break down basement membranes such as collagenase, elastase, etc., and antibodies to various basement membrane fractions. It is possible that when the case-control study is specifically designed, the proteins to be measured may be somewhat different than those listed above.

8) Urine Collection

At the two major visits, a 30cc urine sample will be collected and frozen. No specific studies are planned at this time.

9) Specimen Collection for Fetal Fibronectin

At each major project visit and at the two special "fibronectin visits," fetal fibronectin will be assayed by taking samples of both the cervical and posterior vaginal forniceal fluids with separate dacron swabs during a speculum examination. Each swab will be left in place for approximately 10 seconds, becoming saturated with approximately 150 micro-liters of vaginal or cervical fluid. The swab will be withdrawn and placed in 750 micro-liters of sample buffer. Therefore, the dilution of the vaginal or cervical fluid in the buffer is approximately 1:5.

Concentrations of fetal fibronectin will be measured using the fetal fibronectin specific monoclonal antibody FDC-6 from Adeza Biomedical, Sunnyvale, California with reported inter-assay and intra-assay variations of less than 10 percent and a detectable range of 0.02 to 4 mg/mL.

10) Specimen Collection for Infection

- Vaginal pH: a meter will be used for the study. Vaginal pH will be considered as a risk variable in addition to its role in the the diagnosis of B.V.

- Bacterial vaginosis (B.V.): the gram stains will be read by trained microbiology technologists. All technologists will be trained to read the gram stain using the criteria outlined by Nugent and Hillier. Those who have a score of ≥ 7 and a vaginal pH > 4.5 will be considered to have B.V.

- Trichomonas: a wet prep of vaginal fluid using saline and a glass cover slip will be made at each of the major visits. The wet prep will be defined as positive if any motile trichomonads are seen. It will be defined as negative if 20 high-power fields are examined and found to be negative for motile trichomonads.

- Yeast: a wet prep of vaginal fluid using KOH and a glass cover slip will be made at each visit. The KOH prep will be defined as positive if any yeast-like forms, budding or branching hyphae are noted. It will be defined as negative if 20 high-power fields are examined and found to be negative for yeast-like forms, budding or branching hyphae.

11) Cervical Examination - Digital

At the two major visits, a digital examination will be performed in which the following characteristics of the cervix and lower uterine segment will be determined:

- Length: The length will be estimated by digital examination from the lower uterine segment to the tip of

the cervix in cm (to the nearest 0.5 cm) along both sides of the cervix at 3 and 9 o'clock. The average of the two sides will be considered the estimated length. A paper ruler will be available to estimate the cervical length after the exam.

- Dilatation:

  External Os: using the forefinger, the transverse diameter of the external os at approximately 0.5 cm from the end of the cervix will be estimated in cm (to the nearest 0.5 cm). A paper ruler will be available to estimate the dilation after the exam.

  Internal Os: using gentle pressure, only if the external os is open, the forefinger will be advanced to the level of the internal os and the transverse diameter will be ascertained. A paper ruler will be available to estimate the cervical length after the exam.

- Station: if a presenting part is palpated, the station in relationship to cm above or below the ischial spines will be estimated.

- Position: the cervical os will be classified as positioned anteriorly, in the mid position, or posteriorly in the pelvis.

- Consistency: the cervical os will be classified as being of soft, medium, or firm consistency.

- Lower Segment: the lower uterine segment will be classified as normal or bulging.

- Membranes: the amniotic membranes will be classified as not palpated, palpated but above the level of the internal os, palpated within the cervix, or palpated outside the level of the external os.

12) Ultrasound Examination - Vaginal

After the subject empties her bladder, an endovaginal ultrasound examination will be performed with a 4MHz (or higher) vaginal probe. The internal cervical os will be visualized in the sagittal plane and the probe manipulated until the entire cervical canal can be visualized. Markers will be placed at the furthest points at which the cervical canal walls are juxtaposed and the cervical length will be measured.

The lowest portion of the cervical membranes will be identified and a judgement made whether or not the membranes

are protruding into the cervix. If so, the maximum penetration of the internal cervical os will be noted. All ultrasound examinations will be documented by the creation of a permanent image.

4.4.1 Test Procedure Risks

The potential risks to the study participants include those associated with a pelvic and cervical examination, a vaginal ultrasound, and blood drawing. While there are theoretical risks with pelvic or cervical examinations, including pelvic ultrasound during pregnancy, no report shows any statistically significant relationships between these examinations and any outcome. Multiple ultrasound examinations during pregnancy are routine and considered safe. Theoretical risks from these procedures exist as follow:

- Fetal fibronectin: potential risk includes rupturing membranes and initiating contractions.
- Cervical examination: there is potential risk of infection or rupture of the membranes.
- Ultrasound examination (vaginal): there is potential risk of rupturing the fetal membranes. Since a digital examination will precede the vaginal ultrasound exam, this risk should be minimal.

4.4.2 Patient Management Protocol

The results of all tests or findings except for fetal death, major anomalies, the presence of regular painful contractions, prolapse of membranes through the external os, oligohydramnios or polyhydramnios, placenta previa or cervical dilation of more than 2 cm in a nullipara and more than 3 cm in a multipara will be blinded to the patient and the clinicians involved in patient care.

4.5 Operational Requirements

4.5.1 Study Area

Each MFMU Network center will provide a study area where they will have the ability to perform vaginal ultrasound evaluations and pelvic examinations. They will also provide a laboratory area in which blood and urine specimens can be obtained, aliquoted and frozen, and where vaginal and cervical secretions can be processed for further analysis as well as for the diagnosis of bacterial vaginosis.

4.5.2 Training

The nursing staff assigned to this study will be trained in all aspects of the protocol in order to standardize examination methods and specimen collection and handling. Training will occur at each center through a "hands on" training session and an ultrasound teaching video tape.

Examples of training methodology:

- Cervical examinations: Training and standardization of cervical examination techniques will be performed locally at each clinical center. Each center will designate one examiner to be the "standard" to which all cervical examiners are compared. Cervical examinations will be performed on several patients by both the "standard" examiner and study nurse until there is agreement between them with respect to cervical dilation, length, station, consistency, position, and lower uterine segment distension. Ten patients will then be examined by both the "standard" examiner and study nurse and results compared for consistency. Seventy percent of the exams must be consistent (within an error range) for the study nurse to be certified.

- Ultrasound examinations: A videotape of various vaginal cervical ultrasound evaluations which emphasizes the definition and measurement of various cervical parameters will be created and evaluated by "experts." The intent is that there will be agreement as to what is being measured prior to teaching the research staff. After achieving consensus among the "experts," the videotape will be provided to each center to be used for instruction. Each nurse (or ultrasound technician) will make a videotape of each of 10 ultrasound examinations prior to the start of the study. These tapes will be used as a "post test" to evaluate the capability of each nurse (or ultrasound technician) in making the cervical observations and measurements. The videotapes will be reviewed and graded as satisfactory or unsatisfactory. A total of 8 satisfactory exams are required prior to the start of the study.

- Specimen collection: Careful attention will be paid to the detail of specimen collection and handling so that the fetal fibronectin specimens, BV evaluation and blood specimens will be collected, processed, and when appropriate, analyzed in similar ways at each institution. Training of the reading of the wet prep for trichomonas and the KOH prep for yeast will be performed locally at each center. Preps from ten patients will be examined by both the "standard" examiner and study nurse and results compared for consistency. A total of 8 exams are required to be consistent.

- Measurement standards: Emphasis will be placed on the methodology for taking the anthropometric measurements, performing cervical examinations, vaginal ultrasound examinations, etc.

Additional training will review the study design, objectives, procedures, data collection forms and use of the microcomputer system. All study personnel will be certified in test procedures, data collection procedures, forms completion, and data entry before the start of the study.

4.5.3 Pretesting

Prior to the start of the study, each participating clinical center will be asked to test study procedures at their institution. The purposes of the pretest are to familiarize the study staff with study procedures and logistics, and to determine whether any changes to the study design, operating procedures or data collection forms are warranted.

4.5.4 Quality Assurance

Attention will be paid to quality assessment of the data gathered throughout the project. Examples of quality assessment will include a requirement that a random sample of photostats from the ultrasound cervical length examinations be evaluated for consistency of measurement. All photostats of the cases with membranes protruding into the os will be examined. Additionally, periodic videotapes will be examined for demonstration of measurement techniques. If either the hard copy or the videotape shows errors in technique, further training will be arranged.

A random sample of patients will be chosen for verification of cervical exams. These patients will be examined by both the "standard" examiner and the study nurse and checked for consistency of measurements. In addition, any cervical dilations of 2 cm in primiparous or 3 cm in multiparous must be verified.

Each laboratory performing analysis on any specimen will be required to demonstrate the variability of the assay, as well as the reanalysis of standard specimens.

Study procedures will be reviewed at meetings of the nurse coordinators and at site visits.

CHAPTER 5

Data Processing

5.1 Data collection forms

Data will be collected on standardized forms, on which nearly all responses have been precoded. Each form is described briefly below.

- MP01 - Screening and enrollment (completed for all patients selected for screening).
- MP02 - Visit 1 (24 weeks - includes the baseline demographics, behavioral data, medical history, previous and current pregnancy histories in addition to recording specimens taken for analysis and the results of the cervical exam and ultrasound).
- MP03 - Visit 2/4 (26/30 weeks - records fetal fibronectin samples taken).
- MP04 - Visit 3 (28 weeks - follow-up on some behavioral and pregnancy history data, in addition to to recording specimens taken for analysis and the results of the cervical exam and ultrasound).
- MP05 - Basic maternal/neonatal outcome (to be completed for every baby born. This is the only outcome form required for term deliveries).
- MP06 - Preterm delivery maternal/neonatal outcome (for patients who delivered preterm only).
- MP07 - Incomplete Visit (to document incomplete or missed visits, withdrawal from the study and loss to follow-up. Wherever possible, outcome information will still be obtained).
- MP08 - How I Feel Survey (Nurse's Ancillary Study).
- MP09 - Laboratory Report Form(s) (Blood Catalog Form, Urine Catalog Form, Fetal Fibronectin Form and Gram Stain Form).

5.2 *Distributed Data Entry System*

The microcomputer data entry system consists of a network of microcomputers, one at each clinical center and one at the Biostatistical Coordinating Center (BCC). Data entry software corresponding to the study forms will be developed and maintained by the staff of the Biostatistical Coordinating Center. Data will be entered by clinical center staff, and transmitted weekly via telecommunications link to the BCC. Detailed instructions for entering and transmitting data are provided in the MFMU Network Distributed Data Entry System Handbook (User's Manual).

5.3 *Centralized Data Management System*

All created and updated data forms are transmitted weekly from the clinical centers to the BCC where they are uploaded to the mainframe computer and merged with the existing data base. The data are automatically edited on an intraform basis for missing, out of range and inconsistent values. After review at the BCC, edit printouts are returned to the center for correction or clarification on a weekly basis. At regular intervals, audits, which compare data across forms (interform) are run by the BCC on the entire database or on a specific subset of data. These reports are also submitted to the centers for corrections.

5.4 *Performance Monitoring*

The BCC will present regular reports to the PTD Prediction Subcommittee, the Steering Committee and the Data Monitoring and Safety Committee. These include:

Recruitment reports: Reports of the number of patients screened and enrolled into the study by month and by clinical center will be provided monthly to the Steering Committee. This report will included tabulations of the various reasons for exclusion.

Quality control reports: Reports concerning the quality of data collected, amount of missing data and adherence to study protocol by clinical center, will be submitted periodically to the Steering Committee and the Data Monitoring and Safety Committee.

CHAPTER 6

Statistical Considerations

6.1 Data Relevant to the Study Outcome

Data on the incidence of preterm births (24-34 weeks) among patients registering for prenatal care by 20 weeks gestation are available from the March of Dimes data base collected at the time of the Creasy Prematurity Prevention Program and from the (incomplete) Maternal Fetal Medicine Unit (MFMU) Network Low Risk Aspirin study.

In the March of Dimes (MOD) data base, 58% of the women registered for prenatal care <20 weeks gestational age and of these, excluding those with a multiple gestation, 9.8% delivered preterm where 4.2% followed spontaneous PTL, 3.2% followed spontaneous PROM and 2.5% were indicated.

In the following table, preterm deliveries are broken into different gestational age groups as a percentage of all deliveries.

Table 6.1: Preterm Delivery by Gestational Age Groups (March of Dimes)

	Gestational Age (weeks)			
	20-23	24-34	35-36	Total
Indicated	.18	1.42	0.93	2.5
PROM	.20	1.51	1.37	3.1
Spontaneous	.35	1.77	2.04	4.2
	.73	4.70	4.34	9.8%

Thus, while 9.8% of the total group delivered preterm, only 3.3% of all deliveries followed spontaneous labor or PROM at 24-34 weeks.

Similar data from the MFMU Network Low Risk Aspirin Study data base are shown in the table below.

Table 6.2: Preterm Delivery By Gestational Age Group (MFMU Network)

	Gestational Age (weeks)			
	20-23	24-34	35-36	Total
Indicated	.11	.84	.68	1.63
PROM	.16	1.94	1.63	3.73
Spontaneous	.11	1.63	2.89	4.63
Total	.38	4.41	5.20	9.99%

*MFMU Network Data

From the MFMU data base, 3.57% of all deliveries occurred at 24-34 weeks following spontaneous PTL or PROM, a number very similar to the 3.28% from the MOD data base. Similarly, from the MFMU data base, 8.09% of all deliveries occurred from 24-36 weeks following PROM or spontaneous PTL, a proportion roughly similar to 6.69% from the MOD data base.

Given these data, 3.5% appears to be a suitable estimate of the rate of PTD for singleton pregnancies at 24-34 weeks following PROM or spontaneous labor. Sample size calculations are made based on this assumption.

In the March of Dimes study, 35% of twins had a preterm delivery at 24-34 weeks. It is assumed that most of the twins who deliver at 24-34 weeks will do so secondary to spontaneous preterm labor or PROM, so that sample size calculations are based on a 30% event rate.

6.2 Sample Size

Since specific hypothesis tests are not being proposed, sample size calculations are based on the precision of the odds ratio estimate for a given screening test. The odds ratio is the ratio of the odds of PTD given a positive test to the odds of PTD given a negative test [85]. In the tables below, lower 95% confidence bounds are calculated for odds ratios of 2 and 2.5

assuming a PTD rate of 0.035 and positive test rates of 5%, 10%, 15% and 20% for singleton pregnancies. Likewise, lower 95% confidence bounds are calculated for odds ratios of 2 and 2.5 assuming a PTD rate of 0.30 and positive test rates of 10%, 20%, 30% and 40% for twin pregnancies.

Table 6.3: Lower 95% Confidence Bounds on Odds Ratios for Singleton Pregnancies

Odds Ratio = 2

Sample Size	Proportion Positive 0.05	0.10	0.15	0.20
500	0.41	0.59	0.69	0.74
1000	0.65	0.85	0.94	0.99
2000	0.90	1.09	1.17	1.22
3000	1.04	1.22	1.29	1.33

Odds Ratio = 2.5

Sample Size	Proportion Positive 0.05	0.10	0.15	0.20
500	0.58	0.80	0.90	0.95
1000	0.89	1.12	1.22	1.27
2000	1.20	1.42	1.50	1.54
3000	1.38	1.57	1.65	1.69

For the singleton pregnancies, a sample size of 3000 was chosen to give a 95% lower confidence limit ranging between 1.04 and 1.33 for an odds ratio of 2.0 for a given screening test, depending on the proportion of women positive for that test (5% to 20%). This will result in an estimated 105 cases.

For the twin pregnancies, a sample size of 150, gives a 95% lower confidence limit ranging between 1.08 and 1.11 for an odds ratio of 2.0 for a given screening test, depending on the proportion of women positive for that test (10% to 20%). This will result in an estimated 45 cases.

Table 6.4: Lower 95% Confidence Bounds on Odds Ratios for Twin Pregnancies

	Odds Ratio = 2 Proportion Positive			
	0.10	0.20	0.30	0.40
Sample Size				
50	0.54	0.68	0.72	0.72
100	0.79	0.94	0.98	0.97
150	0.94	1.08	1.11	1.11

	Odds Ratio = 2.5 Proportion Positive			
	0.10	0.20	0.30	0.40
Sample Size				
50	0.73	0.88	0.91	0.88
100	1.05	1.20	1.22	1.19
150	1.23	1.37	1.39	1.37

6.3 Analysis Plan

The singleton and the twins studies will be analysed separately. The primary endpoint, a number of the secondary endpoints and many of the risk factors are dichotomous variables. Thus, standard statistical methods for rates and proportions will be applied. Odds ratios and confidence intervals for single risk factors will be derived. Associations of risk factors with each other will also be examined. Multiple logistic regression will be used to evaluate the risk factors (both discrete and continuous) in conjunction with each other and the most parsimonious model will be sought. For endpoints which are continuous, appropriate techniques, such as multiple regression, will be used. Split sample, jacknife or other model validation methods may be employed.

Some of the tests which have been classified as positive or negative (such as fetal fibronectin where values equal to or greater than 0.05 mg/mL are considered positive) may also be treated as a continuous variable in data analyses. ROC curves will be used to determine the optimal cutpoint for individual screening tests.

As the sample size was determined for a single risk factor and several risk factors are being analyzed, the actual significance level may be different from 0.05. The corresponding

significance level may be adjusted utilizing Bonferroni's inequality adjustment. For example, if the 95% confidence intervals for odds ratios for three risk factors are being calculated, they will be closer to 85% confidence intervals.

CHAPTER 7

Study Administration

7.1 Organization and Funding

The Screening for Risk Factors for Preterm Labor and PROM study is being conducted by the Maternal Fetal Medicine Units Network. The Network is funded by the National Institute of Child Health and Human Development (NICHD) under cooperative agreements between twelve institutions - eleven clinical centers and the Biostatistical Coordinating Center. Each of these thirteen institutions is represented by a Principal Investigator (PI).

7.1.1 Participating Centers

The ten clinical centers participating in this study are all part of the MFMU Network. The Principal Investigators have agreed to abide by the study protocol and to have comparable staff, facilities and equipment.

The Biostatistical Coordinating Center (BCC) is responsible for all aspects of biostatistical design, analysis and data management of the study, in addition to the interim and final statistical analyses. The BCC collaborates with the Steering Committee members in preparing publications based on the study results. The Principal Investigator of the BCC reports to the Steering and Data Monitoring and Safety Committees.

7.1.2 NICHD

In addition to its role as funding agency, the NICHD participates in the activities of the cooperative agreement. NICHD staff participate in the development of protocols and coordination of the studies conducted by the Network.

7.1.3 Network Advisory Board

Appointed by the NICHD, its members include the director of the CRMC, the NICHD Network program officers, chairpersons of the Network Steering Committees and outside experts and consultants. Its role includes assisting in the identification and

prioritization of research projects and reviewing final protocols.

7.2 Committees

7.2.1 Steering Committee

This committee is comprised of fourteen members: one Principal Investigator from each of the eleven clinical centers and the Biostatistical Coordinating Center, the NICHD/CRMD/PPB Special Assistant, and the Chairman of the Steering Committee. In addition, the Center for Research for Mothers and Children, the Division of Epidemiology, Statistics and Prevention Research, and the Pregnancy and Perinatology Branch of NICHD are each represented by one non-voting member. The Chairman, a person independent of the participating institutions, is appointed by NICHD. The Steering Committee has the responsibility for identifying topics for network studies, designing study protocols, and monitoring study implementation, recruitment and protocol adherence. The committee receives recommendations from the Data Monitoring and Safety Committee and the Network Advisory Board. The Principal Investigator from each clinical center is responsible for ensuring the proper conduct of the trial at his or her center including: recruitment and treatment of patients as specified in the protocol, accurate data collection and the transmission of information to the Steering Committee.

7.2.2 Protocol Subcommittee

The PTD Prediction Subcommittee is responsible for the preparation and conduct of this study. The subcommittee will report the progress of the study to the Steering Committee.

7.2.3 Publications Subcommittee

The Publications Subcommittee is a standing subcommittee of the Steering Committee. The functions of this committee are to develop publication policies and to review all manuscripts and abstracts prior to submission. The goals of this committee are fair and appropriate authorship credit and high quality publications.

CHAPTER 8

Study Timetable

8.1 Recruitment and data collection period

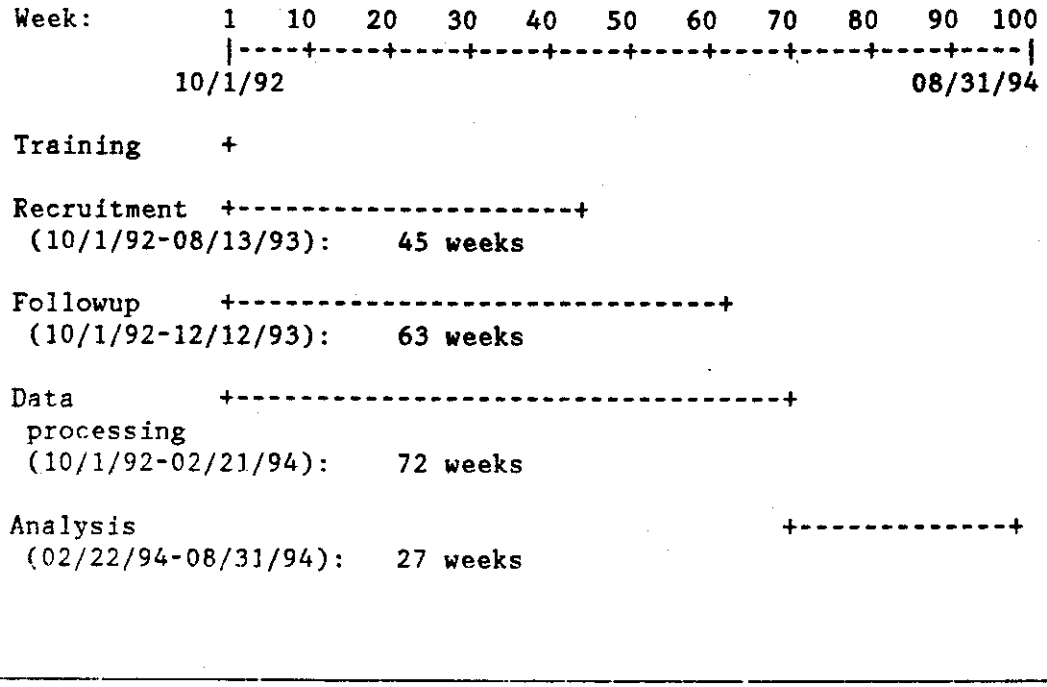
Approximately 12 weeks of startup time for staff training and testing of procedures will be needed prior to the study.

Based on the overall goal of enrolling 3000 women with singleton pregnancies equally distributed among the ten participating MFMU centers, the initial recruitment goal is 300 women from each center. Since it is expected that at least 6-7 women will be enrolled at each center per week (or 24-28/center/month), the recruitment period should last 45 weeks. Recruitment will be evaluated at five 8-week intervals. The recruitment goal for each interval is approximately 56 patients. If a center has not reached the goal for the interval, the difference between 56 and the number of patients the center has recruited will be assigned equally across those centers that have reached their recruitment goal. Every center will have the opportunity to recruit 300 women with singleton pregnancies within 45 weeks. No center will be allowed to contribute more than 20 percent of the total population.

Concurrently, 150 women pregnant with twins will be enrolled. It is expected that this will take about the same time.

Each center is expected to schedule a maximum of 20 'major' visits for the group of screening tests and 20 'minor' visits for fibronectin collection each week. The study period for the last patients recruited is 6 weeks and approximately 12 weeks will pass between the last study visit and delivery, so that the total data collection period is expected to be 63 weeks.

Table 8.1: Timetable of Activities for Screening for Risk Factors Study



8.2 Final analysis

After a two month period for completion of data entry, the data set will be locked and data will be available for analysis. Approximately six months will be required to complete final analyses of study results and to submit the study's primary reports for publication.

Appendix A

Design Summary

MATERNAL FETAL MEDICINE UNITS NETWORK

A Prospective Study of Screening for Risk Factors for Spontaneous Preterm Delivery

OBJECTIVE

To evaluate individually and in combination the screening tests currently advocated to predict spontaneous PROM or preterm labor resulting in a preterm delivery so as to determine their effectiveness in defining women at risk for spontaneous preterm delivery

ORGANIZATION

Clinical centers: * Magee, Tennessee, Alabama, Wayne State, Cincinnati, Oklahoma, Bowman Gray, Chicago, Ohio State, South Carolina

Subcommittee: * Dr. Goldenberg (Chair)

DESIGN

Type: * Observational, prospective cohort

Major Eligibility Criteria:
* Registering for prenatal care prior to 24 weeks gestation
* Informed consent

Stratification:
* Singleton pregnancy
* Twin pregnancy

Strata-Specific Sample Size:
* Singleton pregnancy:
  - Goal = 3,000
  - Assumptions:
    + Antecedent factor = screening test (+,-) (+ test rate = 5%, 10%, 20%)
    + Outcome factor = preterm delivery (24-34 weeks) (PTD rate = 3.5%)
    + 95% confidence interval on the odds of PTD given + test to the odds of PTD given - test (odds ratio = 2.0)
  - Lower bound of confidence interval is 1.04, 1.22, 1.33 for 5%, 10%, 20%
* Twin pregnancy:
  - Goal = 150
  - Assumptions:
    + Antecedent factor = screening test (+,-) (+ test rate = 20%, 30%, 40%)
    + Outcome factor = preterm delivery (24-34 weeks) (PTD rate = 30%)
    + 95% confidence interval on the odds of PTD given + test to the odds of PTD given - test (odds ratio = 2.0)
  - Lower bound of confidence interval is 1.08, 1.11, 1.11 for 20%, 30%, 40%

Total Sample Size: * Goal = 3150

Nested Case-Control Study:
* Compare laboratory values of serum samples
  - cases = spontaneous PTD (23-34 weeks)
  - controls = term deliveries

SCHEDULED EVALUATIONS/DATA COLLECTION

Visit 1 and 3 (24/28 weeks):
* Questionnaire: demographic data, substance and medication use, previous and current pregnancy history, anthropometrics
* Cervical exam: length, dilation, station, position, consistency, membranes
* Ultrasound for cervical parameters: same as above
* Specimen collection: fetal fibronectin, gram stain for bacterial vaginosis, wet mount for trichomonas, vaginal pH, KOH for yeast
* Serum collection: proteins

Visit 2 and 4 (26/30 weeks):
* Fetal fibronectin
* Psychosocial questionnaire (visit 2 only)
* KOH for yeast

Labor and Delivery:
* Maternal and neonatal outcome

OUTCOME MEASURES

Primary:
* Delivery following spontaneous preterm labor/PROM from 23-34 weeks

Secondary:
* Delivery following spontaneous preterm labor/PROM from 23-27, 28-34, 35-36 weeks
* Preterm delivery as result of maternal or fetal indications
* Fetal growth retardation
* Infant morbidity
* Gestational age at PROM/delivery

TIMETABLE

As designed:
* Recruitment: 10/92-08/93
* Data Collection: 10/92-12/93
* Closeout/Final Analysis: 12/93-08/94

Projected as of 2/94:

Singleton:
* Recruitment: 11/92-03/94
* Data Collection: 11/92-08/94
* Closeout/Final Analysis: 08/94-04/95

Twin:
* Add 10 months to above ending dates

(REV: 8/1/94)

Appendix B

Sample Informed Consent Form

SCREENING FOR RISK FACTORS FOR PRETERM LABOR AND PROM

INFORMED CONSENT

Explanation of Procedures

I understand that I am being asked to participate in a research study designed to define groups of women at high risk for preterm delivery. In addition, this study is designed to further develop particular screening tests for identifying specific characteristics and risk factors for PTD. This research study is sponsored by the National Institute of Child Health and Development (NICHD).

If I decide to participate, I understand that I will attend the UAB Specialty clinic four times during my pregnancy for study visits in addition to my regular prenatal care visits at my health center. These four visits will occur at 24, 26, 28, and 30 weeks gestational age. Most of the tests will be administered at the 24 and 28 week visits. During these visits I will be asked to answer questions about my home life, financial status, and educational background. In addition, I will have a pelvic examination performed, a vaginal ultrasound performed, and blood and urine specimens collected. At the 26 and 30 week clinic visits, only a pelvic examination will be done.

Risks and Discomforts

I understand that the potential risks to the study participants include those associated with a pelvic examination and blood drawing. While there are theoretical risks with pelvic, cervical, and ultrasound examinations during pregnancy, no report shows any significant risks between these examinations and poor outcome. Multiple ultrasound examinations during pregnancy are routine and considered safe. Risks and discomforts associated with blood drawing include pain, bleeding, infection, inflammation, and clotting in the vein.

Benefits

The benefits I will receive from participation in this study will be additional free prenatal care and possible early detection of risk factors for and the possible prevention of a preterm delivery.

Alternative Treatments

The alternative to this study is not to participate.

Confidentiality

I understand that any information about me obtained from the research, including my history, laboratory data, findings or physical examination, will be kept strictly confidential and never identified in any report or publication. The results obtained from this study will be made available only to the patients, investigators, and the NICHD.

_____
Patient's Initials

I understand that I am free to refuse to participate in this study or to withdraw at any time and that my decision will not adversely affect my care at this institution or cause a loss of benefits to which I might be otherwise entitled.

Costs to Subjects from Participation in Research

I understand there will be no costs to me for participation in this study. All examinations, ultrasounds, and specimen collection associated with this study will be free of charge.

Payment for Participation in the Research

I understand that I will not receive any monetary payment for participation in this study.

Payment for Research Related Injuries

I understand that UAB has made no provision for monetary compensation in the event of physical injury resulting from the research and in the event of such injury, medical treatment is provided, but not free of charge.

Questions

I acknowledge that I have been given an opportunity to ask questions about this research study and that they have been answered to my satisfaction. If I have further questions about this clinical research trial, Dr. Robert L. Goldenberg or one of the physicians in his division will be glad to answer them at 934-1322.

Agreement

I acknowledge that I have read and understand the above information, and that I agree to participate in this study. I have received a copy of this informed consent and realize that I am not waiving any of my legal rights by signing this consent form. My signature below indicates that I agree to participate in this study.

Signature of Participant	Date
Signature of Father (If Available)	Date
Signature of Physician	Date
Signature of Witness	Date

Appendix C

Endovaginal Ultrasound of the Cervix

ENDOVAGINAL ULTRASOUND OF THE CERVIX
H. Frank Andersen / SPO, February 1992

Reported normal cervical length (mm) by endovaginal ultrasound during pregnancy.

	N Pts.	Trimester (mean ± S.D.) First	Second	Third
Kushnir, 1990	166	43.0 ± 6	45.5 ± 8	42.3 ± 9
Andersen, 1991	177	39.8 ± 8.5	41.6 ± 10.2	32.2 ± 11.6

Kushnir: Patients delivered at term — Andersen: Patients presenting for ultrasound exam

Endovaginal Ultrasound Cervical Length Norms, 14-30 wks EGA (Andersen, 1990)

Mean ± 1 S.D.	40.9 mm ± 10.0
50'th %tile	39 mm
25'th %tile	34 mm
10'th %tile	30 mm

Term vs. Preterm Delivery - Cervical length by endovaginal ultrasound

Andersen, 1990 (Patients presenting for ultrasound, 14-30 wks EGA)

Term Delivery (n=99)	42.8 mm ± 9.9
Preterm Delivery (n=17)	34.1 ± 6.9

Iams, SGI 1991 (Patients at risk of preterm delivery)

	<20 wks	25-28 wks	33+ wks
Term Delivery (n=32)	38.2 mm ± 6.7	30.4 ± 10.7	27.3 ± 6.8
PTL, Term (n=32)	34.2 ± 6.8	27.9 ± 8.2	22.1 ± 5.4
Preterm Delivery (n=26)	31.4 ± 10.4	26.2 ± 8.1	14.6 ± 7.0

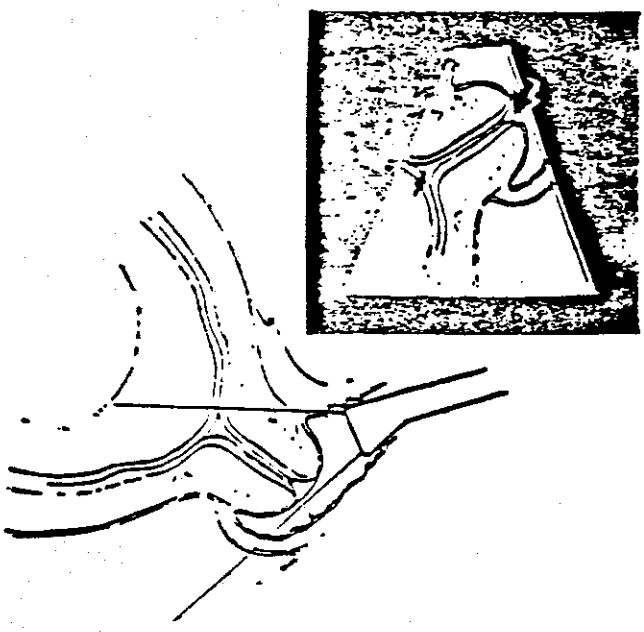


FIGURE 2. Diagram of method to visualize uterine cervix with transvaginal ultrasound. The inset represents the ultrasound picture, the small arrow points to the internal cervical os and the large arrow points to the external cervical os.

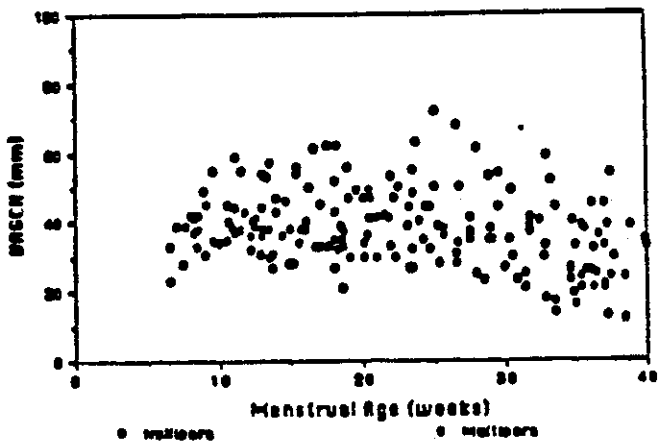
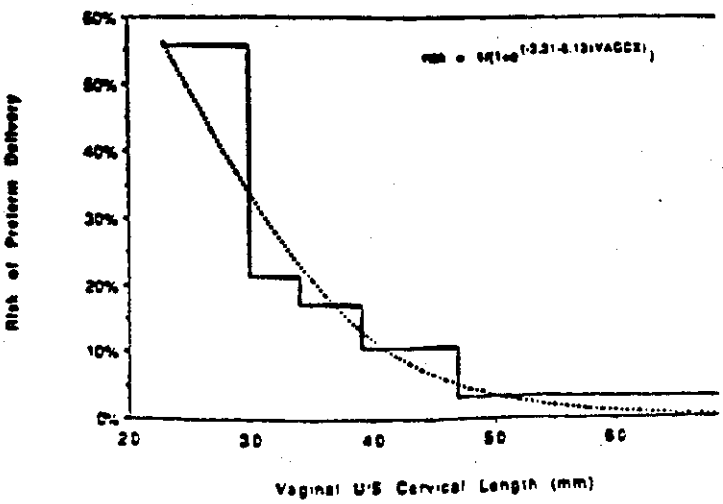


FIGURE 5. Scattergram of transvaginal ultrasound measurements of the cervix throughout gestation in 185 women. (Open circles: primiparae. Solid circles: multiparae. Crosses: 8 patients with incompetent cervix.)



Appendix D

References

1. Goldenberg RL, Cliver SP, Cutter GR et al. Black-White differences in newborn anthropometric measurements. Obstet Gynecol 1991;78:782-8.

2. Shiono PH, Klebanoff MA, Graubard BI, Berendes HW, Rhoads GG. Birth weight among women of different ethnic groups. JAMA 1986;255:48-52.

3. Kullander S, Kallen B. A prospective study of smoking and pregnancy Acta Obstet Gynecol Scand 1971;50:83-94.

2. Shiono PH, Klebanoff MA, Rhoads GG. Smoking and drinking during pregnancy. JAMA 1986;255:82-4.

5. Kramer MS. Determinants of low birth weight: methodological assessment and meta-analysis. Bull WHO 1987;65:663-757.

6. Kleinman JC, Madans JH. The effects of maternal smoking, physical stature, and educational attainment on the incidence of low birth weight. Am J Epidemiol 1985;121:843-55.

7. Wen SW, Goldenberg RL, Cutter GR. Smoking, maternal age, fetal growth, and gestational age at delivery. Am J Obstet Gynecol 1990;162:53-8.

8. Wen SW, Goldenberg RL, Cutter GR, Hoffman HJ, Cliver SP. Intrauterine growth retardation and preterm delivery: prenatal risk factors in an indigent population. Am J Obstet Gynecol 1990;162:213-8.

9. Edwards LA, Alton IR, Barrada MI, et al. Pregnancy in the underweight woman. Am J Obstet Gynecol 1979;135:297-302.

10. Goldenberg RL, Hoffman HJ, Cliver SP, Cutter GR, Nelson KG, Copper RL. The influence of previous low birthweight on anthropometric measurement in the current pregnancy. Obstet Gynecol 1992;79:February.

11. Berkowitz GS. An epidemiologic study of preterm delivery. Am J Epidemiology 1981;113:81-92.

12. Fedrick J, Anderson ABM. Factors associated with spontaneous pre-term birth. Br J Obstet Gynaecol 1976;83:342-50.

13. Creasy RK, Gummer BA, Liggins GC. System for predicting spontaneous preterm birth. Obstet Gynecol 1980;55:692-5.

14. Taren DL, Graven SN. The sensitivity and specificity of a preterm risk score for various patient populations. J of Perinatology 1991;XI:130-5.

15. Liu DTY, Blackwell RJ. The value of a scoring system in predicting outcome of pre-term labour and comparing the efficacy of treatment with aminophylline and salbutamol. Br J Obstet Gynaecol 1978;85:418-24.

16. Main DM, Gabbe SG. Risk scoring for preterm labor: Where do we go from here? Am J Obstet Gynecol 1987;157:789-93.

17. Anderson BM, Turnbull AC. Relationship between length of gestation and cervical dilatation, uterine contractility, and other factors during pregnancy. Am J Obstet Gynecol 1969;105:1207-14.

18. Blondel B, Le Coutour X, Kaminski M, Chavigny C, Breart G, Sureau C. Prediction of preterm delivery: Is it substantially improved by routine vaginal examinations? Am J Obstet Gynecol 1990;162:1042-8.

19. Bouyer J, Papiernik E, Dreyfus J, Collin D, Winisdoerffer B, Gueguen S. Maturation signs of the cervix and prediction of preterm birth. Obstet Gynecol 1986;68:209-14.

20. Hendricks CH, Brenner WE, Kraus G. Normal cervical dilatation pattern in late pregnancy and labor. Am J Obstet Gynecol 1970;106:1065-82.

21. Lenihan JP Jr. Relationship of antepartum pelvic examinations to premature rupture of the membranes. Obstet Gynecol 1984;83:33-7.

22. Leveno KJ, Cox K, Roark ML. Cervical dilatation and prematurity revisited. Obstet Gynecol 1986;68:434-5.

23. Neilson JP, Verkuyl DAA, Crowther CA, Bannerman C. Preterm labor in twin pregnancies: Prediction by cervical assessment. Obstet Gynecol 1988;72:719-23.

24. Papiernik E, Bouyer J, Collin D, Winisdoerffer G, Dreyfus J. Precocious cervical ripening and preterm labor. Obstet Gynecol 1986;67:238-42.

25. Parikh MN, Mehta AC. Internal cervical os during the second half of pregnancy. Obstet Gynaecol 818-21.

26. Schaffner F, Schanzer SN. Cervical dilatation in the early third trimester. Obstet Gynecol 1966;27:130-3.

27. Stubbs TM, Van Dorsten P, Miller MC. The preterm cervix and preterm labor: Relative risks, predictive values, and change over time. Am J Obstet Gynecol 1986;155:829-34.

28. Wood C, Bannerman RH, Booth RT, Pinkerton JH. The prediction of premature labor by observation of the cervix and external tocography. Am J Obstet Gynecol 1965;91:396-402.

29. Copper RL, Goldenberg RL, Davis RO et al. Warning symptoms, uterine contractions, and cervical examination findings in women at risk of preterm delivery. Am J Obstet Gynecol 1990;162:748-54.

30. Andersen HF, Nugent CE, Wanty SD, Hayashi RH. Prediction of risk for preterm delivery by ultrasonographic measurement of cervical length. Am J Obstet Gynecol 1990;163:859-67.

31. Ayers JWT, DeGrood RM, Compton AA, Barclay M, Ansbacher R. Sonographic evaluation of cervical length in pregnancy: Diagnosis and management of preterm cervical effacement in patients at risk for premature delivery. Obstet Gynecol 1988;71:939-44.

32. Bernstine RL, Lee Sh, Crawford WL, Shimek MP. Sonographic evaluation of the incompetent cervix. J Clin Ultrasound 1981;9:417-20.

33. Bowie JD, Andreotti RF, Rosenberg ER. Sonographic appearance of the uterine cervix in pregnancy: The vertical cervix. AJR 1983;140:737-40.

34. Brook I, Feingold M, Schwartz A, Zakut H. Ultrasonography in the diagnosis of cervical incompetence in pregnancy - A new diagnostic approach. Br J Obstet Gynaecol 1981;88:640-3.

35. Confino E, Mayden KL, Giglia RV, Vermesh M, Gleicher N. Pitfalls in sonographic imaging of the incompetent uterine cervix. Acta Obstet Gynecol Scand 1986;65:593-7.

36. Lorenz RP, Comstock CH, Bottoms SF, Marx SR. Randomized prospective trial comparing ultrasonography and pelvic examination for preterm labor surveillance. Am J Obstet Gynecol 1990;162:1603-10.

37. Michaels WH, Schreiber FR, Padgett RJ, Ager J, Pieper D. Ultrasound surveillance of the cervix in twin gestations: Management of cervical incompetency. Obstet Gynecol 1991;78:739-44.

38. Michaels WH, Thompson HO, Schreiber FR, Berman JM, Ager J, Olson K. Ultrasound surveillance of the cervix during pregnancy in diethylstilbestrol-exposed offspring. Obstet Gynecol 1989;73:230-9.

39. Michaels WH, Montgomery C, Karo J, Temple J, Ager J, Olson J. Ultrasound differentiation of the competent from the incompetent cervix: Prevention of preterm delivery. Am J Obstet Gynecol 1986;154:537-46.

40. Podobnik M, Bulic M, Smiljanic N, Bistricki J. Ultrasonography in the detection of cervical incompetency. J Clin Ultrasound 1988;13:383-91.

41. Vaalamo P, Kivikoski A. The incompetent cervix during pregnancy diagnosed by ultrasound. Acta Obstet Gynecol Scand 1983;62:19-21.

42. Varma TR, Patel RH, Pillai U. Ultrasonic assessment of cervix in 'at risk' patients. Acta Obstet Gynecol Scand 1986;65:147- 152.

43. Zemlyn S. The length of the uterine cervix and its significance. J Clin Ultrasound 1981;9:267-269.

44. Brustman L, Langer O, Damus K, Anyaegbunam A, Merkatz IR. A longitudinal study: Uterine contractility patterns in patients after an episode of preterm labor. SPO Eighth Annual Mtg. Las Vegas Nevada 1988.

45. Eden RD, Sokol RJ, Sorokin Y, Cook HJ, Sheeran G, Chik L. The mammary stimulation test - A predictor of preterm delivery? Am J Obstet Gynecol 1991;164:1409-19.

46. Katz M, Newman RB, Gill PJ. Assessment of uterine activity in ambulatory patients at high risk of preterm labor and delivery. Am J Obstet Gynecol 1986;154:44-7.

47. Main DM, Grisso JA, Wold T, Snyder ES, Holmes J, Chiu G. Extended longitudinal study of uterine activity among low-risk women. Am J Obstet Gynecol 1991;165:1317-22.

48. Main DM, Katz M, Chiu G, Campion S, Gabbe SG. Intermittent weekly contraction monitoring to predict preterm labor in lowrisk women: A blinded study. Obstet Gynecol 1988;72:757-61.

49. Zahn V. Uterine contractions during pregnancy. J Perinat Med 1984;12:107-13.

50. Thomsen AC, Morup L, Hansen KB. Antibiotic elimination of group-B streptococci in urine in prevention of preterm labour. Lancet 1987;591-3.

51. Hastings MJG, Easmon CSF, Neill J, Bloxham B, Rivers RPA. Group B streptococcal colonisation and the outcome of pregnancy. J Infection 1986;12:23-9.

52. Smeltzer J, Cruikshank D, Lewisx J, Van Dorsten P. Group B streptococcal colonization is an independent risk factor for premature birth. SPO 8th Annual Mtg., Las Vegas, Nevada 1988.

53. McGregor JA, French JI, Richter R et al. Antenatal microbiologic and maternal risk factors associated with prematurity. Am J Obstet Gynecol 1990;163:1465-73.

54. Toth M, Witkin SS, Ledger W, Thaler H. The role of infection in the etiology of preterm birth. Obstet Gynecol 1988;71:723-6.

55. Romero R, Oyarzun E, Mazor M, Sirtori M, Hobbins JC, Bracken M. Meta-analysis of the relationship between asymptomatic bacteriuria and preterm delivery/low birth weight. Obstet Gynecol 1989;73:576-82.

56. Eschenbach DA, Hillier S, Critchlow C, Stevens C, DeRouen T, Holmes KK. Diagnosis and clinical manifestations of bacterial vaginosis. Am J Obstet gynecol 1988;158:819-28.

57. Gravett MG, Nelson HP, DeRouen T, Critchlow C, Eschenbach DA, Holmes KK. Independent associations of bacterial vaginosis and chlamydia trachomatis infection with adverse pregnancy outcome. JAMA 1986;256:1899-1903.

58. Iams JD, Clapp DH, Contos DA, Whitehurst R, Ayers LW, O'Shaughnessy W. Does extra-amniotic infection cause preterm labor? Gas-liquid chromatography studies of amniotic fluid in amnionitis, preterm labor, and normal controls. Obstet Gynecol 1987;70:365-8.

59. Krohn JA, Hillier SL, Eschenbach DA. Comparison of methods for diagnosing bacterial vaginosis among pregnant women. J Clin Microbiology 1989;27:1266-71.

60. Martius J, Eschenbach DA. The role of bacterial vaginosis as a cause of amniotic fluid infection, chorioamnionitis and prematurity - a review. Arch Gynecol Obstet 1990;247-1-13.

61. Schoonmaker JN, Lunt BD, Lawellin DW, French JI, Hillier SL, McGregor JA. A new proline aminopeptidase assay for diagnosis of bacterial vaginosis. Am J Obstet Gynecol 1991;165:737-42.

62. Spiegel CA, Amsel R, Holmes KK. Diagnosis of bacterial vaginosis by direct gram stain of vaginal fluid. J Clin Microbiology 1983;18:170-7.

63. Thomason JL, Gelbart SM, Anderson RJ, Walt AK, Osypowski PJ, Broekhuizen FF. Statistical evaluation of diagnostic criteria for bacterial vaginosis. Am J Obstet Gynecol 1990;162:155-60.

64. Krieger JN, Tam MR, Stevens CE et al. Diagnosis of Trichomoniasis. JAMA 1988;259:1223-7.

65. Lossick JG, Kent HL. Trichomoniasis: Trends in diagnosis and management. Am J Obstet Gynecol 1991;165:1217-22.

66. Cotch MF, Pastorek JG, Nugent RP et al. Demographic and behavioral predictors of trichomonas vaginalis infection among pregnant women. Obstet Gynecol 1991;78:1087-92.

67. Ernest JM, Meis PJ, Moore ML, Swain M. Vaginal pH: A marker of preterm premature rupture of the membranes. Obstet Gynecol 1989;74:734-8.

68. Dodds WG, Iams JD. Maternal C-reactive protein and preterm labor. J Reprod Med 1987;527-30.

69. Watts DH, Krohn MA, Wener MH, Eschenbach DA. C-reactive protein in normal pregnancy. Obstet Gynecol 1991;77:176-80.

70. Davis RO, Goldenberg RL, Boots L. Elevated levels of midtrimester maternal serum alpha-fetoprotein are associated with preterm delivery not intrauterine growth retardation. Am J Obstet Gynecol In Press.

71. Coulam CB, Wasmoen T, Creasy R, Siiteri P, Gleich G. Major basic protein as a predictor of preterm labor: A preliminary report. Am J Obstet Gynecol 1987;156:790-6.

72. Maymon R, Bahari C, Moroz C. Placental isoferritin measured by a specific monoclonal antibody as a predictive marker for preterm contraction outcome. Obstet Gynecol 1989;74:597-9.

73. Goland RS, Wardlaw SL, Blum M, Tropper PJ, Stark RI. Biologically active corticotropin-releasing hormone in maternaland fetal plasma during pregnancy. Am J Obstet Gynecol 1988;159:884-90.

74. Warren WB, Goland RS, Strak RI, Fox HE, Conwell IM. Elevated maternal plasma corticotrpin releasing hormone levels in twin gestation. J Peri Med 1990;18:39-44.

75. Warren WB, Patrick SL, Goland RS. Elevated maternal plasma corticotropin releasing hormone concentrations in preterm labor. Presented at the Annual Meeting of the Am Gynecol Obstet Society, 1991.

76. Lieberman E, Ryan KS, Minon RR, Schoenbaum SC. Association of maternal hematocrit with premature labor. Am J Obstet Gynecol 1988;159:107-14.

77. Klebanoff MA, Shiono PH, Berendes HW, Rhoads GG. Facts and artifacts about anemia and preterm delivery. JAMA 1989;262:511-5.

78. Lu ZM, Goldenberg RL, Cliver SP, Cutter GR, Blankson M. The relationship between maternal hematocrit and pregnancy outcome. Obstet Gynecol 1991;77:190-194.

79. Goldenberg RL, Cliver SP, Wu Wen S, Cutter GR, Davis RO, Hoffman HJ. Bolled pressure, growth retardation, and preterm delivery. In Press International J Technology Assessment in Health Care. 1992.

80. Lockwood CJ, Senyei AE, Dische R et al. Fetal fibronectin in cervical and vaginal secretions as a predictor of preterm delivery. New Engl J Med 1991;325:669-74.

81. Nageotte MP, Hollenbach KA, Vanderwahl BA, Hutch KM. Oncofetal fibronectin in patients at increased risk for preterm delivery. Society Perinatal Obstetricians. Oral presentation at the 1992 12th Annual Meeting in Orlando, Florida.

82. Brenner WE, Edelman DA, Hendricks CH. A standard of fetal growth for the United States of America. Am J Obstet Gynecol 1976;126:555-64.

83. Friedman L. Furberg C, DeMets D. Fundamentals of clinical trials (2nd ed.). Boston: Wright-PSC, 1985.

84. Rush RW, Keirse MJNC, Howat P, Baum JD, Anderson ABM, Turnbull AC. Contribution of preterm delivery to perinatal mortality. Br Med J 1976;2:965-968.

85. Phillips AN and Pocock SJ. Sample size requirements for prospective studies, with examples for coronary heart disease. J Clin Epidemiol 1989;42:639-648.

Appendix E

Nurses Ancillary Study

E.1 Study Abstract

In this study, a 28 item psychosocial questionnaire will be administered to the 3,000 women enrolled in the study, "Screening For Risk Factors for Spontaneous Preterm Delivery." The study will determine if psychosocial characteristics such as stress, anxiety and depression are related to spontaneous preterm delivery from 24 to 34 weeks and fetal growth retardation.

E.2 Hypotheses

The study is designed to test the following hypotheses:

1) Women who demonstrate increased measures of stress, anxiety, depression, poor self esteem, and poor coping ability will have higher rates of spontaneous preterm delivery.

2) Women with increased measures of poor psychosocial well-being will demonstrate increased rates of maladaptive health behaviors such as smoking, drug and alcohol use and cervicovaginal infections, factors which have been associated with increased rates of spontaneous preterm delivery.

E.3 Introduction

The ability to accurately predict and/or prevent the occurrence of spontaneous preterm delivery has been the subject of intense study for many years [1-6]. Numerous variables have been examined and many risk scoring systems [4,7] have been created. Nevertheless, the ability to predict spontaneous preterm delivery has been poor [7].

Various maternal psychosocial characteristics such as stress, depression and poor social support have been related to pregnancy complications especially low birthweight [8-11],

although the mechanisms for these associations are speculative at best. Both direct and indirect causal pathways have been postulated. For example, psychosocial variables may be related to low birthweight directly by the reduction of blood flow and subsequently fetal nutrients (calories and oxygen) due to vasoconstriction precipitated by the release of catecholamines during periods of stress. Psychosocial factors such as depression, anxiety and low self esteem have also been known to result in higher incidences of maladaptive health behaviors. Zuckerman et. al, reported that depressive symptoms during pregnancy were associated with smoking, substance abuse and poor weight gain during pregnancy [12]. These factors have previously been related to low birthweight and other pregnancy complications [1,13-19]. Indeed, the ability of health care providers to change a pregnant woman's adverse behavior may not be dependent on the counselling itself; rather, counselling regarding adverse health behaviors may be ineffective due to the woman's psychosocial affect [16].

E.4 Background

Previous studies of psychosocial characteristics in relationship to pregnancy outcome are difficult to interpret. A variety of tools created to evaluate stress, depression, self esteem, and other psychosocial characteristics have been utilized. However, in most studies only one psychosocial characteristic has been evalutated and, furthermore, specific definitions of pregnancy complications have been vague. There have been several reports of well-validated measures of anxiety and stress relating to pregnancy outcome. Gorsuch and Key (1974) related anxiety and life change to pregnancy outcome and found that these variables contributed independently to abnormalities of pregnancy [20]. Newton, et. al, [21], used modified life events inventory and found that women with high levels of psychosocial stress delivered more preterm infants. Norbeck and Tilden related stress and emotional disequilibrium to pregnancy complications [22].

Recently, in an epidemiologic study of fetal growth retardation [23] numerous psychosocial characteristics were measured independently by the use of previously validated tools to measure social support [24], anxiety [25], self esteem [26], mastery [27], depression [28], and stress [29]. After controlling for other factors known to influence birth weight, these scales were found to be predictive of fetal growth retardation, both alone as well as when combined [30]. These factors were further evaluated in a model controlling for maternal size and smoking habit. The authors concluded that psychosocial status and smoking were associated with fetal growth retardation predominantly in thin women [31].

However, the administration of five or six psychosocial questionnaires is time consuming. The benefit gained from identifying a subgroup of women at risk for low birthweight may be compromised by the amount of health care provider time invested in the interview process. The five scales utilized in the epidemiologic study of fetal growth retardation cumulatively contained 59 items, far too many items to comfortably administer in an interview session. Therefore, a preliminary study aimed at reducing the item number without compromising efficacy is underway at the University of Alabama at Birmingham. This preliminary work has been made available to this interim subcommittee for use in a prospective study.

E.5 Questionnaire Development

The psychosocial scales used in the preliminary work include: the Speilberger Trait Anxiety Inventory [25], and CES-D Scale [28] for the measurement of anxiety and depression, respectively. Both instruments are in common use, well standardized, and have been previously used in lower socio-economic patients. The Pearlin Mastery Scale [27] was used to measure the patient's perception of her ability to control events and cope with situations. This scale is also well standardized, and has validity and reliability measurements. Because the patient's self esteem has been related to pregnancy outcome, the Rosenberg Scale [26] was used to measure this characteristic. This scale has been used extensively in pregnancy [22]. Finally, a four item questionnaire to measure stress was utilized in addition to these previously mentioned scales. The stress questionnaire was modified from the work of Schar, et. al [29].

To obtain a questionnaire of acceptable length, the first step was to assess the relationship of these scales to one another. Secondly, the entire pool of 59 items was analyzed to address the primary factors that emerged unrelated to pregnancy outcome (to reduce redundency inherent in using five separate scales). A correlation of non-redundent factors with gestational-age specific birthweight was performed and finally, a principal factor analysis with Varimax rotation was performed which revealed seven primary factors (subscales) accounting for 99.9 % of the variance in item scores between subjects. Based upon this principal factor analysis, a short 28 question form was developed (Gotlieb, 1992, in manuscript).

The 28 item questionnaire is constructed in a Likert format, with responses ranging from one ("agree" or "always") to five ("disagree" or "never"). Positively and negatively worded items are arranged at random in the questionnaire to prevent the patient from attempting to "second guess" the desired answer.

E.6 Questionnaire Administration

The 28 item psychosocial questionnaire (MP08) will be administered at Visit 2 (26 weeks gestational age) to all women enrolled in the prospective observational study "Screening for Risk Factors for Spontaneous Preterm Delivery". If a patient missed Visit 2 or had no Visit 2 scheduled, the questionnaire will be administered at Visit 3 (28 weeks gestational age). If unable to administer at Visit 3, then the questionnaire will be considered missed.

Flash cards containing the two answer formats will be utilized to assist the patients in responding to each question.

ALMOST ALWAYS	OFTEN	SOMETIMES	RARELY	NEVER

STRONGLY AGREE	SOMEWHAT AGREE	UNDECIDED	SOMEWHAT DISAGREE	STRONGLY DISAGREE

E.7 Scoring

Responses are arranged on the data sheet using the following scoring system:

Response	Score
Almost always	1
Often	2
Sometimes	3
Rarely	4
Never	5
Strongly agree	1
Somewhat agree	2
Undecided	3
Somewhat disagree	4
Strongly disagree	5

A "high" score reflects a "good" psychosocial status and a "low" score reflects "poor" psychosocial status.

Because the questions are both positively and negatively worded, the positive questions will be reverse scored *by the computer* so that cumulative scores reflect good psychosocial status.

Positively worded statements include:

1 - I feel pleasant

4 - I am happy
6 - I feel secure
9 - I am calm . . .
15 - I feel hopeful about the future
18 - I am content
19 - I take a positive attitude . . .
24 - I am able to do . . .
26 - I feel I have good qualities
27 - I feel I am equal . . .

E.7.1 Example

Although the form reflects:

Question 1 - I feel pleasant

1) Almost always 2) Often 3) Sometimes 4) Rarely 5) Never

the actual scoring of this question will be

1) Never 2) Rarely 3) Sometimes 4) Often 5) Almost always

Therefore, a patient who chooses to answer Question 1 "almost always" will be scored a "5" rather than a "1".

E.8 Analysis Plan

The total psychosocial score will be calculated. The positively worded items ("I am happy") will be reversed scored {"always"(5) - "never"(1)} such that when added, a low score is indicative of poor psychosocial status. The psychosocial score will be categorized into "good" psychosocial status and "poor" psychosocial status.

Scores will be evaluated as additional "risk factors" for spontaneous preterm delivery and fetal growth retardation. The primary endpoint, a number of the secondary endpoints and many of the risk factors are dichotomous variables so standard statistical methods for rates and proportions will be applied. Multiple logistic regression will be used to control for other risk factors. The occurrence of spontaneous preterm delivery as well as growth retardation will be correlated with the total psychosocial score as well as scores from each of the individual subscales. Both univariate and multivariate analyses, controlling for other risk factors for the outcome in question, will be performed.

E.9 Study Advantages

The advantages of inclusion of this 28 item questionnaire in the study, "Screening for Risk Factors for Spontaneous Preterm Delivery" are many. First, the psychosocial status of otherwise "low risk" women may impart valuable insight to any independent role of stress in preterm delivery. Certain psychosocial characteristics may also affect patient's acceptance of many of these test procedures. Knowledge of this information may provide some insight to the complex issue of patient compliance to suggested health care interventions. Finally, the questionnaire in its reduced form will be relatively simple to administer, thus we will be able to evaluate maternal psychosocial status in an efficient manner.

E.10 References

1. Wen SW, Goldenberg RL, Cutter GR, Hoffman HJ, Cliver SP. Intrauterine growth retardation and preterm delivery: prenatal risk factors in an indigent population. *Am J Obstet Gynecol,* 162: 213-218, 1990.

2. Berkowitz, GS. An epidemiologic study of preterm delivery. *Am J Epidemiology,* 113:: 81-92, 1981.

3. Fedrick J, Anderson ABM. Factors associated with spontaneous preterm birth. *Br J Obstet Gynecol,* 83: 342-350, 1976.

4. Creasy RK, Gummer BA, Luggins GC. System for predicting spontaneous preterm birth. *Obstet Gynecol,* 55: 692-695, 1980.

5. Liu DTY, Blackwell RJ. The value of a scoring system in predicting outcome of pre-term labour and comparing the efficacy of treatment with aminophylline and salbutamol. *Br J Obstet Gynecol,* 85: 418-424, 1970.

6. Main DM, Gabba SG. Risk scoring for preterm labor: where do we go from here? *Am J Obstet Gyencol,* 157: 789-793, 1987.

7. Taren DL, Graven SN. The sensitivity and specificity of a preterm risk score for various patient populations. *J of Perinatology,* XI130-135, 1991.

8. Gaziano EP, Freeman DW, Allen TE. Antenatal prediction of women at increased risk for infants with low birth weight: a discriminant analysis of maternal characteristics known at 24 weeks. *Am J Obstet Gynecol,* 140: 99-107, 1981.

9. Brooke OG, Anderson HR, Bland JM, Peacock JL, Stewart CM. Effects on birthweight of smoking, alcohol, caffeine, socioeconomic factors, and psychosocial stress. *Br Med J,* 298: 795-801, 1989.

10. McAnarney ER, Stevens-Simon C. Maternal psychosocial stress/depression and low birth weight: is there a relationship? *AJDC,* 144: 789-92, 1990.

11. Berkowitz GS, Kasl SV. The role of psychosocial factors in spontaneous preterm delivery. *J Psychosomatic Res,* 27: 283-290, 1983.

12. Zuckerman B, Amaro H, Bauchner R, Cabral H. Depressive symptoms during pregnancy: relationship to poor health behaviors. *Am J Obstet Gynecol,* 160: 1107-1111, 1989.

13. Kullander S, Kallen B. A prospective study of smoking and pregnancy. *Acta Obstet Gynecol Scand,* 50: 83-94, 1971.

14. Shiono PH, Klebanoff MA, Rhoads GG. Smoking and drinking during pregnancy. *JAMA,* 255: 82-84, 1986.

15. Kramer MS. Determinants of low birth weight: methodological assessment and meta-analysis. *Bull WHO,* 65: 663-757, 1987.

16. Kleinman JC, Madans JH. The effects of maternal smoking, physical stature, and educational attainment on the incidence of low birth weight. *Am J Epidemiol,* 121: 843-855, 1985.

17. Wen SW, Goldenberg RL, Cutter GR, Hoffman HJ, Cliver SP, Davis RO, DuBard MB. Smoking, maternal age, fetal growth, and gestational age at delivery. *Am J Obstet Gynecol,* 162: 53-58, 1990.

18. Edwards LE, Alton IR, Barrada MI, Hackanson EY. Pregnancy in the underweight woman. *Am J Obstet Gynecol,* 135: 297-302, 1979.

19. McCormick MC, Brookes-Gunn J, Shorter T, Holmes JH, Wallace CY, Heagarty MC. Factors associated with smoking in low-income pregnant women: relationship to birth weight, stressful life events, social support, health behaviors and mental distress. *J Clin Epidemiol,* 43: 441-448, 1990.

20. Gorsuch RL, Key MK. Abnormalities of pregnancy as a function of anxiety and life stress. *Psychosom Med,* 36: 352- , 1974.

21. Newton RW, Webster PAC, Binu, et al. Psychosocial stress in pregnancy and its relation to the onset of premature labor. *Br Med J,* 2: 411-414, 1979.

22. Norbeck JS, Tilden VP. Life stress, social support, and emotional disequilibrium in complications of pregnancy: a prospective, multivariate study. *J Hlth Soc Beh,* 24: 30-46, 1983.

23. Bergsjo P, Hoffman HJ, Davis RO, et al. Preliminary results from the collaborative Alabama and Scandinavian study of succesive small for gestational age births. Report from a Symposium in Trondheim, June 9, 1988. *Acta Obstet Gynecol Scand,* 68: 19-25, 1989.

24. Pascoe JM, Ialongo NS, Horn WF, Reinhart MA, Perradatto D. The reliability and validity of the maternal social support index. *Family Medicine,* 20: 271-279, 1988.

25. Spielberger CD, Gorsuch RL, Lushene RE. Manual for the state-trait anxiety inventory. Consulting Psychology Press, Palo Alto, CA 1970.

26. Rosenberg M. Society and adolescent self image. Princeton University Press, Princeton NJ 1965.

27. Pearlin LI, Liberman MA, et al. The stress process. *J Hlth Soc Beh,* 22: 337-356, 1981.

28. Radloff LS. The CES-D scale: a self report depression scale for research on a general population. *Applied Psychol Measurement,* 1: 385-401, 1977.

29. Schar M, Reeder LG, Kirken JM. Stress and cardiovascular health: an international cooperative study - II. The male population of a factory in Zurich. *Soc Sci Med,* 7: 585-603, 1973.

30. Goldenberg RL, Cliver SP, Cutter GR, Hoffman HJ, Copper RL, Gotlieb S, Davis RO. Maternal psychosocial characteristics and intrauterine growth retardation. *Pre- and Peri-Natal Psychology Journal,* 6: 129-134,1991.

31. Cliver SP, Goldenberg RL, Cutter GR, Hoffman HJ, Copper RL, Gotlieb S, Davis RO. The relationship among psychosocial profile, maternal size, and smoking in predicting fetal growth retardation. *Obstet Gynecol,* In press, 1992.