Vaginal progesterone reduces the rate of preterm birth in women with a sonographic short cervix: a multicenter, randomized, double-blind, placebo-controlled trial

Sonia S. Hassan, MD[1,2]; Roberto Romero, MD[1,3]; Dommeti Vidyadhari, MD, DGO, MBBS[4]; Shalini Fusey, MD, DGO, MBBS[5]; Jason Baxter, MD, MSCP[6]; Meena Khandelwal, MD[7]; Jaya Vijayaraghavan, MD, DGO, MBBS[8]; Yamini Trivedi, MD, DGO, MBBS[9]; Priya Soma-Pillay, MBChB, FCOG[10]; Pradip Sambarey, PhD, MD, MBBS[11]; Ashlesha Dayal, MD[12]; Valentin Potapov, MD, PhD[13]; John O'Brien, MD[14]; Vladimir Astakhov, MD, PhD[15]; Oleksandr Yuzko, MD, PhD[16]; Wendy Kinzler, MD[17]; Bonnie Dattel, MD[18]; Harish Sehdev, MD[19]; Liudmila Mazheika, MD, PhD[20]; Dmitriy Manchulenko, MD[21]; Maria Teresa Gervasi, MD[22]; Lisa Sullivan, PhD[23]; Agustin Conde-Agudelo, MD, MPH[1]; James A. Phillips, DrPH[24]; and George W. Creasy, MD[25], for the PREGNANT Trial

[1]Perinatology Research Branch, *Eunice Kennedy Shriver* National Institute of Child Health and Human Development/National Institutes of Health/Department of Health and Human Services, Bethesda, MD and Detroit, MI, USA; [2]Department of Obstetrics and Gynecology, Wayne State University/Detroit Medical Center, Hutzel Women's Hospital, Detroit, MI, USA; [3]Center for Molecular Medicine and Genetics, Wayne State University, Detroit, MI, USA; [4]Department of Obstetrics and Gynecology, MediCiti Institute of Medical Services, Andhra Pradesh, India; [5]Department of Obstetrics and Gynecology, Government Medical College and Hospital, Maharashtra, India; [6]Thomas Jefferson University, Department of Obstetrics and Gynecology / Maternal Fetal Medicine, Philadelphia, PA, USA; [7]Cooper University Hospital, Camden, NJ, USA; [8]Department of Obstetrics and Gynecology, Shri Ramchandra Medical College and Hospital, Tamil Nadu, India; [9]Department of Gynecology, Sheth L.G. Hospital, Gujarat, India; [10]Department of Obstetrics and Gynaecology, Steve Biko Academic Hospital, Pretoria, South Africa; [11]Department of Obstetrics and Gynecology, B.J. Medical College and Sassoon General Hospital, Maharashtra, India; [12]Albert Einstein Hospital, Bronx, NY, USA; [13]Department of Obstetrics and Gynecology of Dnepropetrovsk State Medical Academy, Municipal Establishment "City Maternity Hospital # 1", Dnepropetrovsk, Ukraine; [14]Perinatal Diagnostic Center, Central Baptist Hospital, Lexington, KY, USA; [15]Department of Obstetrics and Gynecology of M. Gorky Donetsk National Medical University, Municipal Hospital "6th Central City Clinical Hospital", Donetsk, Ukraine; [16]1st Department of Obstetrics and Gynecology of Ukrainian State Institute of Human Reproductology of P.L. Shupik National Academy of Postgraduate Education, Pechersk regional "1st Antenatal out-patients' clinic", Kiev, Ukraine; [17]Winthrop University Hospital, Clinical Trials Center, Mineola, NY, USA; [18]Eastern Virginia Medical School, Norfolk, VA,

USA; [19]University of Pennsylvania Health System, Pennsylvania Hospital, Philadelphia, PA, USA; [20]Public Health Services Establishment Minsk "1st City Clinic", Minsk, Republic of Belarus; [21]Department of Antenatal Day-Hospital of Municipal health care establishment "City Maternity Hospital # 1", Chernovtsy, Ukraine; [22]U.O. Ostetricia / Ginecologia, Azienda Ospedaliera di Padova, Padova, Italy; [23]Boston University, School of Public Health, Boston, MA, USA; [24] Sage Statistical Solutions, Inc. Elfand, NC, USA; [25] Columbia Laboratories, Inc., Livingston, NJ, USA.

Correspondence to: Dr. Roberto Romero, Chief, Perinatology Research Branch, Program Director for Perinatal Research and Obstetrics Intramural Division, NICHD, NIH, DHHS; Professor of Molecular Obstetrics and Genetics Center for Molecular Medicine and Genetics, Wayne State University; Professor of Epidemiology Michigan State University, 3990 John R, Detroit, MI 48201, romeror@mail.nih.gov

Keywords: pregnancy, preterm labor, uterine cervix, progestogens, preterm delivery, progestins, vaginal administration, respiratory distress syndrome, transvaginal ultrasound.

ABSTRACT

Background: Women with a sonographic short cervix in the mid-trimester are at increased risk for preterm delivery. This study was undertaken to determine the efficacy and safety of micronized vaginal progesterone gel to reduce the risk of preterm birth and associated neonatal complications in women with a sonographic short cervix.

Methods: This was a multicenter, randomized, placebo-controlled trial that enrolled asymptomatic women with a singleton pregnancy and a sonographic short cervix (10 to 20 mm) at 19 - 23 6/7 weeks of gestation. Women were randomly allocated to receive vaginal progesterone gel or placebo daily from 20 – 23 6/7 weeks until 36 6/7 weeks, rupture of membranes, or delivery, whichever occurred first. Randomization sequence was stratified by center and history of a previous preterm birth. The primary endpoint was preterm birth before 33 weeks of gestation. Analysis was by intention to treat.

Findings: Of 465 women randomized, seven were lost to follow-up and 458 (vaginal progesterone gel, n=235; placebo, n=223) were included in the analysis. Women allocated to receive vaginal progesterone had a lower rate of preterm birth before 33

weeks than those allocated to placebo (8.9% [n=21] vs 16.1% [n=36], relative risk [RR] 0.55, 95% confidence interval [CI] 0.33-0.92, p=0.02). The effect remained significant after adjustment for co-variables (adjusted RR 0.52, 95% CI 0.31-0.91, p= 0.02). Vaginal progesterone was also associated with a significant reduction in the rate of preterm birth before 28 (5.1% vs 10.3%, RR 0.50, 95% CI 0.25-0.97, p=0.04) and 35 weeks (14.5% vs 23.3% RR 0.62, 95% CI 0.42-0.92, p=0.02), respiratory distress syndrome (3.0% vs 7.6% RR 0.39, 95% CI 0.17-0.92, p=0.03), any neonatal morbidity or mortality event (7.7% vs 13.5% RR 0.57, 95% CI 0.33-0.99, p=0.04), and birth weight <1500 g (6.4% [15/234] vs 13.6% [30/220], RR 0.47, 95% CI 0.26-0.85, p=0.01). There were no differences in the incidence of treatment-related adverse events between the groups.

Interpretation: The administration of vaginal progesterone gel to women with a sonographic short cervix in the midtrimester is associated with a 45% reduction in the rate of preterm birth before 33 weeks of gestation, and improved neonatal outcomes.

Funding: *Eunice Kennedy Shriver* National Institute of Child Health and Human Development/National Institutes of Health and Columbia Laboratories, Inc.

Introduction

Preterm birth is the leading cause of perinatal morbidity and mortality, and its prevention is an important healthcare priority.[1] In 2005, 12.9 million births worldwide were preterm.[2] A sonographic short cervix is a powerful predictor of preterm delivery,[3-21] yet implementation of a screening program of all pregnant women requires the availability of a clinical intervention able to prevent preterm delivery and improve neonatal outcome.[22] Strategies that have been considered include progesterone administration,[23] cervical cerclage,[24-30] and a pessary.[31]

A randomized clinical trial of vaginal progesterone capsules to prevent preterm delivery (<34 weeks of gestation) in women with a short cervix (defined as 15 mm or less) reported a 44% reduction in the rate of preterm delivery (19.2% vs. 34.4%; relative risk [RR], 0.56; 95% confidence interval [CI], 0.36-0.86), although this was not associated with a significant improvement in neonatal outcome.[23] In addition, secondary analyses of a randomized clinical trial[32] of vaginal progesterone in patients with a history of preterm birth showed that progesterone administration was associated with delayed cervical shortening[33] as pregnancy progressed, a lower rate of preterm birth, a lower frequency of newborn admission to the intensive care unit and a shorter length of neonatal stay.[34]

This study was undertaken to determine the efficacy and safety of vaginal progesterone gel in reducing the rate of preterm birth before 33 weeks in asymptomatic women with a mid-trimester sonographic short cervix.

Methods

Study design and participants

This was a Phase III, prospective, randomized, placebo-controlled, double-masked, parallel-group, multi-center, international trial. The study was conducted from March 2008 – November 2010 and was approved by the institutional review board of each participating center. Participants provided written informed consent to study coordinators or investigators prior to participation in the trial. Women between 19 0/7 and 23 6/7 weeks of gestation were eligible for screening. During the screening visit, cervical length and gestational age were determined. Women were eligible for the study if they met the following criteria: 1) singleton gestation; 2) gestational age between 19 0/7 and 23 6/7 weeks; 3) transvaginal sonographic cervical length between 10 and 20 mm; and 4) asymptomatic; without signs or symptoms of preterm labor. Subjects were randomly allocated to receive vaginal progesterone gel or placebo beginning at 20 – 23 6/7 weeks. Gestational age calculation was based on the participant's reported last menstrual period and fetal biometry.[35]

Exclusion criteria included: 1) planned cerclage; 2) acute cervical dilation; 3) allergic reaction to progesterone; 4) current or recent progestogen treatment within the previous four weeks; 5) chronic medical conditions that would interfere with study participation or evaluation of the treatment (e.g. seizures, psychiatric disorders, uncontrolled chronic hypertension, congestive heart failure, chronic renal failure, uncontrolled diabetes mellitus with end-organ dysfunction, active thrombophlebitis or a thromboembolic disorder, history of hormone-associated thrombophlebitis or thromboembolic disorders, active liver dysfunction or disease, known or suspected malignancy of the breast, or

genital organs); 6) major fetal anomaly or known chromosomal abnormality; 7) uterine anatomic malformation (bicornuate uterus, septate uterus); 8) vaginal bleeding; or 9) known or suspected clinical chorioamnionitis.

All sonographers involved in sonographic cervical length measurements were required to participate in a training program and to obtain certification before screening patients for the trial. Moreover, the sonographic images of patients enrolled into the trial were reviewed by a Central Sonologist for quality assurance. An independent Data Coordinating Center was responsible for randomization and data management. Clinical research monitors (Venn Life Sciences [Canada] and PharmOlam International [Texas]) conducted planned, regular site visits at each center beginning with a site initiation visit and continuing until study completion to independently assess compliance with the study protocol, timely collection of data, quality control, data completeness, and data accuracy according to International Committee on Harmonization (ICH) and Food and Drug Administration (FDA) guidelines for Good Clinical Practice (GCP).[36, 37] This study included 44 centers in 10 countries.

Randomization and masking

The randomization allocation was 1:1 (vaginal progesterone gel and placebo) and was accomplished using a centralized interactive voice response (IVR) system. Randomization was stratified according to: a) center and b) risk strata (previous preterm birth between 20-35 weeks or no previous preterm birth) using a permuted blocks strategy with a block size of four (i.e., two placebo and two vaginal progesterone gel).

Contact with the IVR system required the input of subject characteristics and center number after which the IVR system assigned a treatment for the specific subject based on the strata to which the subject belonged and the next assignment within the randomization block.

Allocation concealment was accomplished in three ways. First, subject drug kits at each study site were numbered independently from the treatment assignments in the randomization blocks to avoid identification of dispensing patterns. Second, the IVR system (upon generating a treatment assignment for a new subject) specified which kit number was to be dispensed to the subject. Third, the study drug packaging, applicators, and their contents (vaginal progesterone and placebo) were identical in appearance.

Procedures

All drug required throughout the treatment interval for a randomized woman was included in drug kits to be assigned to each patient at each study visit to prevent dispensing errors. Prior to dispensing the assigned treatment, demographic, medical and obstetrical history, and physical examination data were collected from each participant. Treatment was to be initiated between 20 0/7 and 23 6/7 weeks gestational age. Women self-administered the study drug once daily in the morning.

Study participants were instructed to return to the study center every two weeks. During each visit, subjects were interviewed to determine the occurrence of adverse events, use of concomitant medications, and compliance with study drug. Women were asked to

return unused study drug from the previous two weeks, and determination of compliance was based on the amount of the study drug not used.

Study drug was continued until 36 6/7 weeks gestational age, rupture of membranes, or delivery, whichever occurred first. Both the vaginal progesterone gel (Prochieve® 8%, also known as Crinone® 8%) and placebo were supplied by Columbia Laboratories, Inc. (Livingston, New Jersey, USA) as a soft, white to off-white gel, in a single use, one piece, white disposable polyethylene vaginal applicator with a twist-off top. The progesterone and placebo gels were identical in appearance. Each applicator delivered 1.125 grams of gel containing 90 mg of progesterone or placebo, and was wrapped and sealed in unmarked foil over-wrap. Both the active drug and placebo were supplied in boxes of 14 applicators and were labeled with a unique kit number. Patients received a two-week supply at randomization and at each subsequent visit. Subjects also received a one-week emergency supply kit at the time of randomization and were resupplied during the treatment period if additional applicators were required before attending the next visit.

Patients who developed preterm labor during the study were treated according to the standard of practice of the participating institutions, e.g., admission to the hospital, bed rest, intravenous fluids, tocolytic therapy, and steroid administration, etc. if clinically indicated. Administration of the study drug was to be continued during treatment for preterm labor, until delivery (in the absence of preterm rupture of membranes). Maternal and neonatal outcome were recorded throughout study participation and after delivery and discharge using a standardized electronic reporting template.

An emergency cerclage was allowed after randomization if the following criteria were met: 1) 21 – 26 weeks gestational age; 2) cervical dilation > 2 cm; 3) membranes visible; 4) intact membranes; and 5) absence of uterine contractions, clinical chorioamnionitis, or significant vaginal bleeding.

The primary outcome of this study was preterm birth before 33 weeks of gestation. The key secondary outcomes were neonatal morbidity including RDS, bronchopulmonary dysplasia, grade III or IV intraventricular hemorrhage, periventricular leukomalacia, proven sepsis, necrotizing enterocolitis, and perinatal mortality (fetal death or neonatal death). Four composite outcome scores were also used to assess perinatal mortality and neonatal morbidity (any event, two 0-4 scales, and a 0-6 scale). The definitions for individual outcomes and composite scores are provided in the Supplementary material (S1). The scores (0-4; 0-6) assigned ordinal values based upon the number of morbid events from 0-3 or 0-5; the highest number, 4 or 6, was assigned to a mortality event. For one of the 0-4 scores, NICU days was also used for the assignment of the ordinal value. Other pre-specified secondary outcomes included preterm birth before 28, 35, and 37 weeks of gestation, neonatal length, weight, and head circumference at birth, and incidence of congenital abnormalities. The frequency of adverse events related to treatment was also assessed (see Supplementary material S2 for definition of adverse events). All outcomes were determined and the database was locked prior to the unsealing of the randomization code.

Statistical analysis

We estimated that a sample size of 450 women (225 per treatment group) would have a >90% power (two-tailed alpha level of 0.05) to detect a 55% reduction in the rate of preterm birth before 33 weeks of gestation, from 22% in the placebo group to 9.9% in the vaginal progesterone group.

The analysis of the trial was conducted in three different analysis sets:

1) Intent-to-treat (ITT) analysis set: all patients randomized to either vaginal progesterone or placebo. Subjects without a delivery date were excluded from this analysis set;

2) "Treated patient analysis set": patients who took at least one dose of either placebo or progesterone Women who were allocated to receive placebo with no documented delivery date were considered as if they had a delivery at term (37 weeks of gestation). For subjects who received vaginal progesterone gel and had no documented delivery date, the date of last contact was used as the delivery date; and

3) Compliant analysis set: patients who used at least 80% of study medication, did not have a cerclage and were not lost to follow-up.

The primary endpoint of the study, preterm birth before 33 weeks, was analyzed using the Cochran-Mantel-Haenszel (CMH) test. The p-value was assessed at the 2-sided significance level of 5%. Analysis of the primary efficacy endpoint was also performed using multivariable logistic regression in which the following variables were included: treatment group, pooled study site, risk strata, gestational age at first dose, maternal age,

cervical length, body mass index (BMI), and race. Relative risk (RR) with a 95% confidence interval (CI) was used as the measure of effect. The CMH test was also used for the analysis of the ordinal composite scores described in S1. For this analysis, a modified ranking procedure (modified ridits) was used to calculate the sum of the expected values for each of the ordinal categories for each of the treatment groups. This ranking procedure is equivalent to non-parametric van Elteren scores. The RR for the primary endpoint was calculated unadjusted, partially adjusted (for pooled study site and risk strata), as well as fully adjusted using multivariable logistic regression. We also calculated the number needed to treat (NNT),[38] with 95% CIs for the primary outcome and the most common complication of preterm birth, RDS. All analyses were performed with SAS® 9.2 on a Windows 2003 operating system.

An independent Data and Safety Monitoring Board (DSMB) reviewed unblinded data relevant to safety (not efficacy) after approximately 50% of the subjects had delivered. The observed frequency of adverse events did not exceed what was expected and as stated in the informed consent. The DSMB recommended the study continue without modification of the protocol or informed consent. This trial is registered with ClinicalTrials.gov, number NCT00615550.

Results

Of the 32,091 women who underwent sonographic measurement of cervical length between 19 0/7 and 23 6/7 weeks of gestation, 2.3% (733/32,091) were reported to have a cervical length 10-20 mm. Four hundred sixty-five women agreed to participate and were randomized; seven patients were lost to follow-up (progesterone n=1, placebo n=6); 458

women were included in the ITT analysis set (vaginal progesterone, n=235; placebo, n=223). Figure 1 shows the participant flow diagram (See Supplementary material S3 for further details regarding patient disposition). The trial ended on the date of the last delivered participant. Sixteen percent (72/458) of the women had a history of a previous preterm birth between 20-35 weeks of gestation.

Baseline maternal characteristics were similar between the placebo and the progesterone groups (Table 1). There were no differences in median duration of treatment (14.3 weeks for vaginal progesterone and 13.9 weeks for placebo) or mean study drug administration compliance reported by the investigator (93.3% standard deviation [SD] +/-13.1 for vaginal progesterone and 94.0% SD+/-12.7 for placebo) between the two groups. A history of cervical surgery was present in 9.4% (22/235) of patients allocated to receive progesterone and 12.6% (28/223) of those allocated to the placebo group (p=0.20). Sixteen women (ten in the vaginal progesterone group and six in the placebo group; p=0.46) underwent an emergency cervical cerclage after randomization.

Patients allocated to vaginal progesterone gel had a significantly lower rate of preterm birth before 33 weeks of gestation compared to those who were allocated to placebo, (8.9% [n=21] versus 16.1% [n=36], RR 0.55, 95% CI 0.33-0.92, p=0.02; adjusted [pooled study site and risk strata] RR 0.54, 95% CI 0.33-0.89, p=0.01). Fourteen women with cervical length between 10 and 20 mm would need to be treated with vaginal progesterone to prevent one case of preterm birth before 33 weeks of gestation (95% CI, 8-87). Even after adjustment for pooled study site, risk strata, treatment group, gestational

age at first dose, maternal age, cervical length, BMI, and race using multivariable logistic regression analysis, the effect of vaginal progesterone remained significant for the primary endpoint (adjusted RR 0.52; 95% CI, 0.31-0.91; p=0.02). No interaction between treatment and pooled study site was detected (p=0.2). In women without a history of preterm birth (84% of the population), vaginal progesterone administration was associated with a significant reduction in the rate of preterm birth before 33 weeks, (7.6% [15/197] vs 15.3% [29/189], RR 0.50 95% CI, 0.27-0.90, p=0.02). However, the reduction in the rate of preterm birth in women with a prior history of preterm birth between 20-35 weeks of gestation did not reach statistical significance. (15.8% [6/38] vs 20.6% [7/34], RR 0.77, 95% CI 0.29-2.06, p=0.60).

Vaginal progesterone was also associated with a significant reduction in the rate of preterm birth before 35 weeks (14.5% [n=34] vs 23.3% [n=52], RR=0.62 95% CI 0.42-0.92, p=0.02) and before 28 weeks of gestation (5.1% [n=12] vs 10.3% [n=23], RR=0.50, 95% CI 0.25-0.97, p=0.04). Figure 2 displays the survival analysis for patients in the entire ITT analysis set (Figure 2A), patients with no prior preterm delivery (Figure 2B), and patients with a prior preterm delivery (Figure 2C). The curves demonstrate a separation between patients allocated to receive progesterone and those in the placebo group. However, there is no difference in the proportion of patients who delivered at <37 weeks. This is because the curves converge and overlap at this point. One interpretation of this is that the administration of vaginal progesterone shifted the proportion of patients who would have delivered very preterm to a later gestational age. In addition, vaginal progesterone was associated with a significant reduction in the rate of neonatal birth

weight less than 1500 g (6.4% [15/234] vs 13.6% [30/220], RR 0.47, 95% CI 0.26-0.85, p=0.01; (Table 2).

In terms of infant outcome, neonates born to women allocated to receive progesterone had a significantly lower frequency of RDS than those born to women allocated to receive placebo (3.0% [n=7] versus 7.6% [n=17], RR 0.39, 95% CI 0.17-0.92, p=0.03); NNT for benefit 22, 95% CI 12-186. This effect remained significant after adjustment for pooled study site and risk strata (RR 0.40, 95% CI 0.17 to 0.94, p=0.03). The other neonatal outcomes are listed in Table 2. Pre-specified composite scores to assess perinatal mortality/neonatal morbidity were calculated. The rate of any morbidity or mortality was significantly lower in the neonates of subjects allocated to receive progesterone compared to those allocated to receive placebo (7.7% [n=18] vs 13.5% [n=30], RR 0.57, 95% CI 0.33-0.99, p=0.04). The composite scores '0-4 scale without NICU' and '0-6 scale without NICU' were also significantly lower in the progesterone group when compared to placebo ($p<0.05$ for both comparisons). After adjustment for pooled study site and risk strata, the effect of vaginal progesterone on composite perinatal mortality/neonatal morbidity scores 'any morbidity/mortality event', '0-4 scale without NICU,' and '0-6 scale without NICU' continued to show trends towards improvement (p=0.054, 0.065, and 0.065, respectively). The frequency of distributions for the perinatal mortality/neonatal morbidity composite scores can be found in the Supplementary material (S4).

Adverse events were comparable between patients who received vaginal progesterone gel and those who received placebo. The rate of adverse events related to study treatment was not significantly different in women who received vaginal progesterone gel compared to those who received placebo (12.8% [n=30] versus 10.8% [n=24]; RR 1.19, 95% CI 0.72-1.96, p=0.51); the most frequently reported adverse events related to study treatment occurred in two percent of women and included vaginal pruritus, vaginal discharge, vaginal candidiasis, and nausea. Furthermore, no fetal or neonatal safety signal[39] was detected for vaginal progesterone gel. Regarding labor and delivery data, there were no meaningful differences in method of delivery. There was one case of a congenital anomaly in the progesterone group and three in the placebo group (RR 0.32, 95% CI 0.03-3.02; p=0.29). Median 1 and 5 minute Apgar scores were comparable between study groups.

Results of the "treated patient analysis set**":**

Four hundred sixty-five women were randomized; 459 women received at least one dose of study drug, were included in the analysis set (vaginal progesterone gel, n=235; placebo, n=224) and represent the "treated patient analysis set". Sixteen percent (71/459) of the women had a history of a previous preterm birth between 20-35 weeks gestation.

There were no differences in the baseline patient characteristics, median duration of treatment (14.3 weeks for vaginal progesterone and 13.9 weeks for placebo), or mean study drug administration compliance reported by the investigator (93.3% SD +/- 13.1 for

vaginal progesterone and 94.5% SD +/- 10.9 for placebo) between the two groups. See table 3 for results of primary and secondary outcomes.

After adjustment for study site and risk strata (history of preterm birth) the effect of vaginal progesterone remained significant for the reduction in the primary endpoint of the rate of preterm birth before 33 weeks of gestation (8.9% [21/235] vs. 15.2% [34/224], RR 0.56, 95% CI 0.33-0.93, p=0.02) as well as the rate of RDS (3.0% [7/235] vs. 7.1% [16/224], RR 0.42, 95% CI 0.18-0.97, p=0.04). Pre-specified composite scores to assess perinatal mortality/neonatal morbidity were calculated as previously described: '0-4 scale without NICU,' '0-4 scale with NICU', and '0-6 scale without NICU' (p=0.113, 0.103, and 0.113, respectively, for vaginal progesterone versus placebo).

Adverse events were comparable between patients who received vaginal progesterone gel and those who received placebo. The rate of adverse events related to study treatment was not significantly different in women who received vaginal progesterone gel compared to those who received placebo (13% [30/235] versus 11% [24/224], RR 1.14, 95% CI 0.72-1.80), p=0.59); the most frequently reported adverse events related to study treatment occurred in up to two percent of women and included vaginal pruritus, vaginal discharge, vaginal candidiasis, and nausea. Furthermore, no fetal or neonatal safety signal was detected for vaginal progesterone gel. Regarding labor and delivery data, there were no differences in method of delivery. There was one case of a congenital anomaly in the progesterone group and three in the placebo group. Median 1 and 5 minute Apgar scores were comparable between study groups. Women allocated to receive vaginal

progesterone gel had a lower rate of neonates born at < 1500g when compared to those in the placebo group (6.4% [15/234] vs 13.3% [29/218], RR 0.49, 95% CI 0.27-0.88, p=0.01).

Results of the "compliant analysis set":

A pre-specified analysis (n=387) was conducted in a subgroup of the treated patient analysis set, excluding those who had <80% treatment compliance (n=53), those who did not have a documented delivery date (n=4), or who had a cerclage (n=17). One subject had <80% compliance and a cerclage and one subject had no delivery date and had received a cerclage.

This compliant analysis set (84% [387/459] of the treated patient analysis set) showed for unadjusted analyses that patients allocated to vaginal progesterone gel had a significantly lower frequency of preterm birth than those allocated to placebo for delivery < 28 weeks of gestation (3.1% [6/194] versus 7.8% [15/193], RR 0.40, CI 0.16-1.00, p=0.04), < 33 weeks gestation (5.7% [11/194] vs 13.0% [25/193], RR 0.44, CI 0.22-0.86, p=0.01), and < 35 weeks gestation (10.3% [20/194] vs 20.2% [39/193], RR 0.51, CI 0.31-0.84, p<0.01) weeks. There was no significant difference in the rate of preterm delivery before 37 weeks of gestation (26.8% [52/194] vs 30.6% [59/193], RR 0.88, CI 0.64-1.20, p=0.41). See table 4 for results of primary outcome and secondary outcomes, RDS, and any morbidity/mortality event.

After adjustment for study site and risk strata, the effect of vaginal progesterone gel remained significant for the reduction in the primary endpoint of the rate of preterm birth

before 33 weeks of gestation (RR 0.42, 95% CI 0.22-0.82, p<0.01), and < 35 weeks (RR 0.50, 95% CI 0.31-0.82, p<0.01). Pre-specified composite scores to assess perinatal mortality/neonatal morbidity were calculated as previously described: '0-4 scale without NICU,' '0-4 scale with NICU', and '0-6 scale without NICU' showed trends towards significance (p=0.058, 0.049, and 0.058, respectively).

In summary, there was no evidence of a safety signal and the evidence for the efficacy of vaginal progesterone gel was demonstrated in a substantially similar manner for both of these additional analysis sets as was demonstrated for the intent-to-treat analysis set.

Discussion

Principal findings of the study: Administration of vaginal progesterone to women with a short cervix (10-20 mm) was associated with: 1) a substantial reduction in the rate of preterm delivery <33 (primary endpoint), <35 and <28 weeks of gestation; 2) a significant decrease in the rate of RDS; 3) a similar rate of treatment-related adverse events in patients allocated to progesterone or placebo gel; and 4) no evidence of a "safety signal".

Clinical implications of the study: The prevention of preterm birth is a major healthcare priority. The ultimate purpose of interventions designed to reduce preterm birth is improvement in infant outcome. To date, no intervention in an asymptomatic patient with a risk factor has demonstrated both a reduction in preterm birth and an improvement in infant outcome, without a safety signal.[40] The results of this trial indicate that a combined

approach in which transvaginal sonographic cervical length was used to identify patients at risk for preterm delivery and administration of vaginal progesterone gel from the midtrimester of pregnancy until term reduced the rate of both preterm birth before 33 weeks of gestation and respiratory distress syndrome, the most common complication of preterm neonates. In addition to the primary and secondary endpoints related to gestational age, administration of vaginal progesterone gel was associated with a significant reduction in the proportion of infants with any morbidity/mortality event, and demonstrated a significant improvement in neonatal outcome through two additional composite scores as well as a significant reduction in birth weight <1500 g. Of note, vaginal progesterone gel was well-tolerated and compliance was substantial (>90%).

Results in the context of other studies: The primary result of this trial is similar to that reported by Fonseca *et al*[23] who found that vaginal progesterone (200 mg vaginal capsules) administered to women with a cervical length ≤15 mm at a median gestational age of 23 weeks reduced the rate of spontaneous preterm (<34 weeks) delivery by 44%. In our trial, there was a 45% reduction in the rate of preterm delivery before 33 weeks. This finding is robust because it was supported by a significant 38% reduction in the rate of preterm birth <35 weeks, a 50% reduction at <28 weeks, and a 53% reduction in the rate of birth weight <1500 g. In addition, the reduction in preterm birth observed in this trial translated into the improvement of clinically important neonatal outcomes such as respiratory distress syndrome and three composite perinatal mortality/neonatal morbidity scores.

Both the study by Fonseca[23] and the current trial used a similar approach to identify the patients at risk, namely, screening with transvaginal sonography to diagnose a short cervix. Differences between the trials are that: 1) our study excluded twin gestations, which have not been shown to benefit from the prophylactic administration of progesterone[41] or 17 alpha-hydroxyprogesterone caproate[42, 43]; 2) the cervical length for entry into our study was 10 – 20 mm. Patients with a cervical length of 10 mm or less have a higher rate of intra-amniotic infection/inflammation[44] and are less likely to benefit from progesterone administration than are patients with a longer cervix. We also extended the upper limit of cervical length to 20 mm to explore whether vaginal progesterone gel would have a beneficial effect beyond 15 mm, and therefore, expand its therapeutic range; 3) the treatment protocol in our study called for initiation of vaginal progesterone as early as 20 weeks of gestation continuing until 36 6/7 weeks, while Fonseca *et al*[23] began at 24 weeks and stopped at 34 weeks. It is possible that earlier treatment may confer more beneficial effects; and 4) the formulation of vaginal progesterone was different. Fonseca *et al*[23] used oil capsules containing 200 mg of progesterone while we employed a bioadhesive gel with 90 mg of progesterone. The vaginal gel preparation has been shown to be biologically active in supporting pregnancies in the first trimester undergoing assisted reproductive technology, and despite the lower dose of progesterone, our current trial results indicate that the dose was sufficient to reduce the rate of preterm delivery. We postulate that this is attributable to the bioadhesive nature of the preparation, which may enhance bioavailability.

Strengths and limitations of the study: The strengths of this study are that it was a multicenter, placebo-controlled, double-masked, randomized trial with rigorous standards for the allocation of treatment and concealment of the identity of the treatment. The placebo and vaginal progesterone gel preparations were identical in appearance, and procedures were in place to reduce the risk of other biases. In addition, we performed an additional sensitivity analysis in the ITT analysis set to provide a "worst case" scenario in which women lost to follow-up who received vaginal progesterone were considered as if they had a preterm birth before 33 weeks gestation whereas women lost to follow-up who received placebo were considered as if they had a term delivery (≥37 weeks of gestation). Even in this "worst case" scenario of the ITT analysis set, the beneficial effect of vaginal progesterone on the rate of preterm birth before 33 weeks of gestation remained significant (9.3% [22/236] vs 15.7% [36/229], RR 0.59, 95% CI 0.36-0.98, p=0.04).

Another strength of this study is its apparent external validity, supported by the following: 1) our primary results were consistent with those of a similar trial[23] that tested the effects of progesterone capsules in women with a short cervix and reported a similar effect size; 2) the preterm delivery rate in the placebo arm was similar to that reported in studies in the literature;[8, 13, 45] 3) there was no treatment by site interaction albeit with the necessity to pool sites for this test; and 4) the multi-national nature of the trial in which there was substantial representation (approximately 30%) for each of the following ethnic groups: African-American, Asian and Caucasian. A limitation of the study is that the primary endpoint is a surrogate for infant outcome. The use of surrogate endpoints is common in clinical trials because of the pragmatic challenges in the execution of trials

where infant outcome is the primary outcome of interest. Our study was not powered to detect differences in the outcome according to risk strata (presence or absence of a previous preterm birth).

Sonographic cervical length to identify the patient at risk for preterm delivery: It is now well-established that the shorter the sonographic cervical length in the midtrimester, the higher the risk of preterm delivery.[8, 10-19, 21] Indeed, it is possible to assign an individualized risk[46] for preterm delivery using sonographic cervical length and other maternal risk factors, such as maternal age, ethnic group, body mass index and previous cervical surgery. Among these factors, sonographic cervical length is the most powerful predictor for preterm birth in the index pregnancy, and is more informative than is a history of previous preterm birth.[10, 13] Selecting patients for prophylactic administration of progestogens based only on a history of a previous preterm birth[32, 47, 48] would have an effect (albeit limited) on the prevention of preterm delivery worldwide, because most women who deliver preterm neonates do not have this history. Moreover, such strategy cannot be implemented in nulliparous women; therefore, universal risk assessment (primigravidas and parous women) is possible with transvaginal cervical ultrasound. A pharmacoeconomic study is in progress to address the issue of cost-effectiveness, based on the observations of this study.

The effect of progesterone on the uterine cervix: Although the original focus of the effect of progesterone in pregnancy maintenance was on the myometrium,[49-58] it is now clear that this hormone exerts biological effects on the chorioamniotic membranes[59-62]

and the uterine cervix.[63-84] Indeed, progesterone is considered key in the control of cervical ripening. The precise mechanism by which progesterone prevents preterm delivery in women with a short cervix has not been established. A local effect is likely, given the high concentrations of circulating progesterone in pregnant women.[85, 86]

Differences among progestogens: The term "progestogen", like "progestin", includes both "natural progesterone" and synthetic compounds with progesterone-like actions. The compound used in this study is identical to natural progesterone, as was in the study by Fonseca *et al*.[23] Progesterone is currently approved to support pregnancies in the first trimester in patients undergoing assisted reproductive technologies in the U.S.,[87] Europe and other countries. The safety profile of this preparation is well-established. In contrast, there are no data to date to support the use of 17-alpha hydroxyprogesterone caproate, a synthetic progestogen, to prevent preterm birth in women with a short cervix.

Future studies: Additional studies are necessary to determine if treatment of women with a short cervix in the early second trimester may further reduce the rate of preterm delivery.[88] Moreover, it is important to determine if women with twin gestations who have a short cervix may also benefit from vaginal progesterone. The previous negative results of a randomized clinical trial in twin gestations could be attributed to the inclusion of patients with a long cervix, and thus, who may not have benefitted from vaginal progesterone. The optimal treatment of patients with a cervical length of <10mm remains a challenge. Similarly, whether vaginal progesterone may modify the effect of vaginal cerclage remains to be determined.

Importance of the findings: The potential impact of this intervention in clinical practice can be surmised from the estimate that 14 patients need to be treated to prevent one preterm birth before 33 weeks of gestation. Moreover, 22 patients need to be treated to prevent one episode of RDS. These figures compare well with those of two interventions widely used in obstetrics in which 100 patients with preeclampsia need to be treated with magnesium sulfate to prevent one case of eclampsia,[89] and 13 women at high risk of preterm birth need to receive antenatal corticosteroids to prevent one case of RDS.[90]

Implications for clinical practice: The main implication of this study for clinical practice is that universal screening of women with transvaginal cervical length in the midtrimester to identify patients at risk can now be coupled with an intervention – the administration of vaginal progesterone gel – to reduce the frequency of preterm birth and improve neonatal outcome. This can be accomplished safely and conveniently.

Contributors

SSH, RR, DV, SF, JB, MK, JV, YT, PSP, PS, AD, VP, JO, VA, OY, WK, BD, HS, LM, DM, MTG, GWC contributed to the conception, design, management and interpretation of data, drafting and critically revising the manuscript for important intellectual content, and approval of the final version to be published. JAP, LS, ACA contributed to data analysis and interpretation, as well as drafting and critically revising the manuscript for important intellectual content, and approval of the final version to be published. All authors have seen and approved the final draft.

Investigators participating in the study

The authors would like to acknowledge the investigators that participated in this study.

Belarus, Republic of L. Mazheika, S Zanko ***Chile*** J Ortiz Castro, E Oyarzun ***Czech Republic*** P Calda, ***India*** S Fusey, P Sambarey, Y Trivedi, D Vidyadhari, J Vijayaraghavan ***Israel*** A Bashiri, Y Hazan, I Hendler ***Italy*** MT Gervasi ***Russia*** A Mikhailov ***South Africa*** P Soma-Pillay ***Ukraine*** V Astakhov, D Manchulenko, V Potapov, A Senchuk, O Yuzko ***United States*** R Artal, J Balducci, J Baxter, M Beall, L Bracero, B Dattel, GW Creasy, A Dayal, SS Hassan, B Howard, J Hwang, G Kazzi, M Khandelwal, W Kinzler, J Kipikasa, J O'Brien, A Odibo, K Porter, R Quintero, R Romero. H Sehdev, A Sheikh, C Weiner, D Wing, YC Yang. **The investigators would like to thank the following study coordinators of the trial:** J Bieda, S Krafft, JR Parella, K Zubovskiy.

Role of the funding source

The study investigators listed as authors were responsible for the study design, data collection, and interpretation of the results of the data analysis. The Perinatology Research Branch of the National Institute of Child Health and Human Development (NICHD)/National Institutes of Health (NIH) was responsible for the writing of the report, and the decision to submit the paper for publication. The funding sources (NICHD/NIH and Columbia Laboratories, Inc.) were not involved in writing the report or the decision to submit the paper for publication.

Conflicts of Interest

SSH, RR, MTG, ACA, WK, and LS have no conflicts of interest. Author-Investigators DV, SF, JB, MK, JV, YT, PS-P, PS, AD, VP, JO'B, VA, OY, BD, HS, LM, and DM conducted this study with the support of grants awarded by Columbia Laboratories, Inc. for the specific purpose of conducting this trial. The terms and conditions for the awarding of the grants were consistent with what is customary for this type of industry sponsored trial and all payments were independent of the outcome of the trial. In addition, JB and JO'B have also received consulting fees and travel related to Preterm Birth Advisory Committee meetings related to the project. JO'B is an inventor on a patent for the use of progesterone in the prevention of preterm birth. JAP received compensation as a statistical consultant to Columbia Laboratories, Inc. GWC is an employee of Columbia Laboratories, Inc.

Acknowledgements

This clinical trial was conducted pursuant to a Clinical Trials Agreement between the *Eunice Kennedy Shriver* National Institute of Child Health and Human Development (NICHD) of the National Institutes of Health (NIH), Department of Health and Human Services (DHHS) of the United States, and Columbia Laboratories, Inc., Livingston, New Jersey, USA. The research was supported, in part, by the Intramural Program of NICHD. We would like to thank the patients who participated in this trial, as well as the coordinators, physicians, nurses, sonographers and administrative staff who assisted in the conduction of this study.

References

1. Preterm Birth Causes, Consequences, and Prevention, Committee on Understanding Premature Birth and Assuring Healthy Outcomes, Board on Health Sciences Policy; . Washington, D.C.: Institute of Medicine of the Academies, The National Academies press, Washington D.C. 2007.

2. Beck S, Wojdyla D, Say L, Betran AP, Merialdi M, Requejo JH, Rubens C, Menon R, Van Look PF. The worldwide incidence of preterm birth: a systematic review of maternal mortality and morbidity. *Bull World Health Organ* 2010;**88**:31-38.

3. 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**:859-867.

4. Kushnir O, Vigil DA, Izquierdo L, Schiff M, Curet LB. Vaginal ultrasonographic assessment of cervical length changes during normal pregnancy. *Am J Obstet Gynecol* 1990;**162**:991-993.

5. Okitsu O, Mimura T, Nakayama T, Aono T. Early prediction of preterm delivery by transvaginal ultrasonography. *Ultrasound Obstet Gynecol* 1992;**2**:402-409.

6. Tongsong T, Kamprapanth P, Srisomboon J, Wanapirak C, Piyamongkol W, Sirichotiyakul S. Single transvaginal sonographic measurement of cervical length early in the third trimester as a predictor of preterm delivery. *Obstet Gynecol* 1995;**86**:184-187.

7. Hasegawa I, Tanaka K, Takahashi K, Tanaka T, Aoki K, Torii Y, Okai T, Saji F, Takahashi T, Sato K, Fujimura M, Ogawa Y. Transvaginal ultrasonographic cervical assessment for the prediction of preterm delivery. *J Matern Fetal Med* 1996;**5**:305-309.

8. Iams JD, Goldenberg RL, Meis PJ, Mercer BM, Moawad A, Das A, Thom E, McNellis D, Copper RL, Johnson F, Roberts JM. The length of the cervix and the risk of spontaneous premature delivery. *N Engl J Med* 1996;**334**:567-572.

9. Berghella V, Tolosa JE, Kuhlman K, Weiner S, Bolognese RJ, Wapner RJ. Cervical ultrasonography compared with manual examination as a predictor of preterm delivery. *Am J Obstet Gynecol* 1997;**177**:723-730.

10. Heath VC, Southall TR, Souka AP, Elisseou A, Nicolaides KH. Cervical length at 23 weeks of gestation: prediction of spontaneous preterm delivery. *Ultrasound Obstet Gynecol* 1998;**12**:312-317.

11. Taipale P, Hiilesmaa V. Sonographic measurement of uterine cervix at 18-22 weeks' gestation and the risk of preterm delivery. *Obstet Gynecol* 1998;**92**:902-907.

12. Watson WJ, Stevens D, Welter S, Day D. Observations on the sonographic measurement of cervical length and the risk of premature birth. *J Matern Fetal Med* 1999;**8**:17-19.

13. Hassan SS, Romero R, Berry SM, Dang K, Blackwell SC, Treadwell MC, Wolfe HM. Patients with an ultrasonographic cervical length < or =15 mm have nearly a 50% risk of early spontaneous preterm delivery. *Am J Obstet Gynecol* 2000;**182**:1458-1467.

14. Heath VC, Daskalakis G, Zagaliki A, Carvalho M, Nicolaides KH. Cervicovaginal fibronectin and cervical length at 23 weeks of gestation: relative risk of early preterm delivery. *BJOG* 2000;**107**:1276-1281.

15. Hibbard JU, Tart M, Moawad AH. Cervical length at 16-22 weeks' gestation and risk for preterm delivery. *Obstet Gynecol* 2000;**96**:972-978.

16. Owen J, Yost N, Berghella V, Thom E, Swain M, Dildy GA, 3rd, Miodovnik M, Langer O, Sibai B, McNellis D. Mid-trimester endovaginal sonography in women at high risk for spontaneous preterm birth. *JAMA* 2001;**286**:1340-1348.

17. To MS, Skentou C, Liao AW, Cacho A, Nicolaides KH. Cervical length and funneling at 23 weeks of gestation in the prediction of spontaneous early preterm delivery. *Ultrasound Obstet Gynecol* 2001;**18**:200-203.

18. Guzman ER, Walters C, Ananth CV, O'Reilly-Green C, Benito CW, Palermo A, Vintzileos AM. A comparison of sonographic cervical parameters in predicting spontaneous preterm birth in high-risk singleton gestations. *Ultrasound Obstet Gynecol* 2001;**18**:204-210.

19. Matijevic R, Grgic O, Vasilj O. Is sonographic assessment of cervical length better than digital examination in screening for preterm delivery in a low-risk population? *Acta Obstet Gynecol Scand* 2006;**85**:1342-1347.

20. Crane JM, Hutchens D. Transvaginal sonographic measurement of cervical length to predict preterm birth in asymptomatic women at increased risk: a systematic review. *Ultrasound Obstet Gynecol* 2008;**31**:579-587.

21. Vaisbuch E, Romero R, Erez O, Kusanovic JP, Mazaki-Tovi S, Gotsch F, Romero V, Ward C, Chaiworapongsa T, Mittal P, Sorokin Y, Hassan SS. Clinical significance of early (< 20 weeks) vs. late (20-24 weeks) detection of sonographic short cervix in asymptomatic women in the mid-trimester. *Ultrasound Obstet Gynecol* 2010;**36**:471-481.

22. Romero R. Prevention of spontaneous preterm birth: the role of sonographic cervical length in identifying patients who may benefit from progesterone treatment. *Ultrasound Obstet Gynecol* 2007;**30**:675-686.

23. Fonseca EB, Celik E, Parra M, Singh M, Nicolaides KH. Progesterone and the risk of preterm birth among women with a short cervix. *N Engl J Med* 2007;**357**:462-469.

24. Althuisius SM, Dekker GA, Hummel P, Bekedam DJ, van Geijn HP. Final results of the Cervical Incompetence Prevention Randomized Cerclage Trial (CIPRACT): therapeutic cerclage with bed rest versus bed rest alone. *Am J Obstet Gynecol* 2001;**185**:1106-1112.

25. Rust OA, Atlas RO, Reed J, van Gaalen J, Balducci J. Revisiting the short cervix detected by transvaginal ultrasound in the second trimester: why cerclage therapy may not help. *Am J Obstet Gynecol* 2001;**185**:1098-1105.

26. To MS, Alfirevic Z, Heath VC, Cicero S, Cacho AM, Williamson PR, Nicolaides KH. Cervical cerclage for prevention of preterm delivery in women with short cervix: randomised controlled trial. *Lancet* 2004;**363**:1849-1853.

27. Berghella V, Odibo AO, Tolosa JE. Cerclage for prevention of preterm birth in women with a short cervix found on transvaginal ultrasound examination: a randomized trial. *Am J Obstet Gynecol* 2004;**191**:1311-1317.

28. Alfirevic Z. Cerclage: we all know how to do it but can't agree when to do it. *Obstet Gynecol* 2006;**107**:219-220.

29. Owen J, Hankins G, Iams JD, Berghella V, Sheffield JS, Perez-Delboy A, Egerman RS, Wing DA, Tomlinson M, Silver R, Ramin SM, Guzman ER, Gordon M, How HY, Knudtson EJ, Szychowski JM, Cliver S, Hauth JC. Multicenter randomized trial of cerclage for preterm birth prevention in high-risk women with shortened midtrimester cervical length. *Am J Obstet Gynecol* 2009;**201**:375. e371-378.

30. Simcox R, Seed PT, Bennett P, Teoh TG, Poston L, Shennan AH. A randomized controlled trial of cervical scanning vs history to determine cerclage in women at high risk of preterm birth (CIRCLE trial). *Am J Obstet Gynecol* 2009;**200**:623. e621-626.

31. Arabin B, Halbesma JR, Vork F, Hubener M, van Eyck J. Is treatment with vaginal pessaries an option in patients with a sonographically detected short cervix? *J Perinat Med* 2003;**31**:122-133.

32. O'Brien JM, Adair CD, Lewis DF, Hall DR, Defranco EA, Fusey S, Soma-Pillay P, Porter K, How H, Schackis R, Eller D, Trivedi Y, Vanburen G, Khandelwal M,

Trofatter K, Vidyadhari D, Vijayaraghavan J, Weeks J, Dattel B, Newton E, Chazotte C, Valenzuela G, Calda P, Bsharat M, Creasy GW. Progesterone vaginal gel for the reduction of recurrent preterm birth: primary results from a randomized, double-blind, placebo-controlled trial. *Ultrasound Obstet Gynecol* 2007;**30**:687-696.

33. O'Brien JM, Defranco EA, Adair CD, Lewis DF, Hall DR, How H, Bsharat M, Creasy GW. Effect of progesterone on cervical shortening in women at risk for preterm birth: secondary analysis from a multinational, randomized, double-blind, placebo-controlled trial. *Ultrasound Obstet Gynecol* 2009;**34**:653-659.

34. DeFranco EA, O'Brien JM, Adair CD, Lewis DF, Hall DR, Fusey S, Soma-Pillay P, Porter K, How H, Schakis R, Eller D, Trivedi Y, Vanburen G, Khandelwal M, Trofatter K, Vidyadhari D, Vijayaraghavan J, Weeks J, Dattel B, Newton E, Chazotte C, Valenzuela G, Calda P, Bsharat M, Creasy GW. Vaginal progesterone is associated with a decrease in risk for early preterm birth and improved neonatal outcome in women with a short cervix: a secondary analysis from a randomized, double-blind, placebo-controlled trial. *Ultrasound Obstet Gynecol* 2007;**30**:697-705.

35. ACOG Committee on Practice Bulletins - Obstetrics. ACOG Practice Bulletin. Clinical management guidelines for obstetricians-gynecologists. Number 55, September 2004 (replaces practice pattern number 6, October 1997). Management of Postterm Pregnancy. *Obstet Gynecol* 2004;**104**:639-646.

36. International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) adopts Consolidated Guideline on Good Clinical Practice in the Conduct of Clinical Trials on Medicinal Products for Human Use. *Int Dig Health Legis* 1997;**48**:231-234.

37. Dixon JR, Jr. The International Conference on Harmonization Good Clinical Practice Guideline. *Qual Assur* 1998;**6**:65-74.

38. Altman DG. Confidence intervals for the number needed to treat. *BMJ* 1998;**317**:1309-1312.

39. Guidance for Industry: Good Pharmacovigilance Practices and Pharmacoepidemiologic Assessment; U.S. Department of Health and Human Services Food and Drug Administration; Center for Drug Evaluation and Research (CDER); Center for Biologics Evaluation and Research (CBER). 2005 [cited; Available from: http://www.fda.gov/downloads/RegulatoryInformation/Guidances/UCM126834.pdf

40. Review by the Division of Reproductive and Urologic Products
Food and Drug Administration

August 2, 2006 [cited; Available from: (http://www.fda.gov/ohrms/dockets/ac/06/briefing/2006-4227B1-02-01-FDA-Background.pdf

41. Norman JE, Mackenzie F, Owen P, Mactier H, Hanretty K, Cooper S, Calder A, Mires G, Danielian P, Sturgiss S, MacLennan G, Tydeman G, Thornton S, Martin B, Thornton JG, Neilson JP, Norrie J. Progesterone for the prevention of preterm birth in twin pregnancy (STOPPIT): a randomised, double-blind, placebo-controlled study and meta-analysis. *Lancet* 2009;**373**:2034-2040.

42. Rouse DJ, Caritis SN, Peaceman AM, Sciscione A, Thom EA, Spong CY, Varner M, Malone F, Iams JD, Mercer BM, Thorp J, Sorokin Y, Carpenter M, Lo J, Ramin S, Harper M, Anderson G. A trial of 17 alpha-hydroxyprogesterone caproate to prevent prematurity in twins. *N Engl J Med* 2007;**357**:454-461.

43. Combs CA, Garite T, Maurel K, Das A, Porto M. 17-hydroxyprogesterone caproate for twin pregnancy: a double-blind, randomized clinical trial. *Am J Obstet Gynecol* 2011;**204**:221 e221-228.

44. Vaisbuch E, Hassan SS, Mazaki-Tovi S, Nhan-Chang CL, Kusanovic JP, Chaiworapongsa T, Dong Z, Yeo L, Mittal P, Yoon BH, Romero R. Patients with an asymptomatic short cervix (<or=15 mm) have a high rate of subclinical intraamniotic inflammation: implications for patient counseling. *Am J Obstet Gynecol* 2010;**202**:433 e431-438.

45. Berghella V, Roman A, Daskalakis C, Ness A, Baxter JK. Gestational age at cervical length measurement and incidence of preterm birth. *Obstet Gynecol* 2007;**110**:311-317.

46. Celik E, To M, Gajewska K, Smith GC, Nicolaides KH. Cervical length and obstetric history predict spontaneous preterm birth: development and validation of a model to provide individualized risk assessment. *Ultrasound Obstet Gynecol* 2008;**31**:549-554.

47. da Fonseca EB, Bittar RE, Carvalho MH, Zugaib M. Prophylactic administration of progesterone by vaginal suppository to reduce the incidence of spontaneous preterm birth in women at increased risk: a randomized placebo-controlled double-blind study. *Am J Obstet Gynecol* 2003;**188**:419-424.

48. Meis PJ, Klebanoff M, Thom E, Dombrowski MP, Sibai B, Moawad AH, Spong CY, Hauth JC, Miodovnik M, Varner MW, Leveno KJ, Caritis SN, Iams JD, Wapner RJ, Conway D, O'Sullivan MJ, Carpenter M, Mercer B, Ramin SM, Thorp JM, Peaceman AM, Gabbe S. National Institute of Child Health and Human Development Maternal-

Fetal Medicine Units Network. Prevention of recurrent preterm delivery by 17 alpha-hydroxyprogesterone caproate. *N Engl J Med* 2003;**348**:2379-2385.

49. Word RA, Cornwell TL. Regulation of cGMP-induced relaxation and cGMP-dependent protein kinase in rat myometrium during pregnancy. *Am J Physiol* 1998;**274**:C748-756.

50. Fomin VP, Cox BE, Word RA. Effect of progesterone on intracellular Ca2+ homeostasis in human myometrial smooth muscle cells. *Am J Physiol* 1999;**276**:C379-385.

51. Pieber D, Allport VC, Hills F, Johnson M, Bennett PR. Interactions between progesterone receptor isoforms in myometrial cells in human labour. *Mol Hum Reprod* 2001;**7**:875-879.

52. Mesiano S, Chan EC, Fitter JT, Kwek K, Yeo G, Smith R. Progesterone withdrawal and estrogen activation in human parturition are coordinated by progesterone receptor A expression in the myometrium. *J Clin Endocrinol Metab* 2002;**87**:2924-2930.

53. Condon JC, Jeyasuria P, Faust JM, Wilson JW, Mendelson CR. A decline in the levels of progesterone receptor coactivators in the pregnant uterus at term may antagonize progesterone receptor function and contribute to the initiation of parturition. *Proc Natl Acad Sci U S A* 2003;**100**:9518-9523.

54. Madsen G, Zakar T, Ku CY, Sanborn BM, Smith R, Mesiano S. Prostaglandins differentially modulate progesterone receptor-A and -B expression in human myometrial cells: evidence for prostaglandin-induced functional progesterone withdrawal. *J Clin Endocrinol Metab* 2004;**89**:1010-1013.

55. Condon JC, Hardy DB, Kovaric K, Mendelson CR. Up-regulation of the progesterone receptor (PR)-C isoform in laboring myometrium by activation of nuclear factor-kappaB may contribute to the onset of labor through inhibition of PR function. *Mol Endocrinol* 2006;**20**:764-775.

56. Hardy DB, Janowski BA, Corey DR, Mendelson CR. Progesterone receptor plays a major antiinflammatory role in human myometrial cells by antagonism of nuclear factor-kappaB activation of cyclooxygenase 2 expression. *Mol Endocrinol* 2006;**20**:2724-2733.

57. Merlino AA, Welsh TN, Tan H, Yi LJ, Cannon V, Mercer BM, Mesiano S. Nuclear progesterone receptors in the human pregnancy myometrium: evidence that parturition involves functional progesterone withdrawal mediated by increased expression of progesterone receptor-A. *J Clin Endocrinol Metab* 2007;**92**:1927-1933.

58. Renthal NE, Chen CC, Williams KC, Gerard RD, Prange-Kiel J, Mendelson CR. miR-200 family and targets, ZEB1 and ZEB2, modulate uterine quiescence and contractility during pregnancy and labor. *Proc Natl Acad Sci U S A* 2010;**107**:20828-20833.

59. Pieber D, Allport VC, Bennett PR. Progesterone receptor isoform A inhibits isoform B-mediated transactivation in human amnion. *Eur J Pharmacol* 2001;**427**:7-11.

60. Oh SY, Kim CJ, Park I, Romero R, Sohn YK, Moon KC, Yoon BH. Progesterone receptor isoform (A/B) ratio of human fetal membranes increases during term parturition. *Am J Obstet Gynecol* 2005;**193**:1156-1160.

61. Lee RH, Stanczyk FZ, Stolz A, Ji Q, Yang G, Goodwin TM. AKR1C1 and SRD5A1 messenger RNA expression at term in the human myometrium and chorioamniotic membranes. *Am J Perinatol* 2008;**25**:577-582.

62. Merlino A, Welsh T, Erdonmez T, Madsen G, Zakar T, Smith R, Mercer B, Mesiano S. Nuclear progesterone receptor expression in the human fetal membranes and decidua at term before and after labor. *Reprod Sci* 2009;**16**:357-363.

63. Csapo AI, Resch B, Csapo EF, Salau G. Effects of antiprogesterone on pregnancy. I. Midpregnancy. *Am J Obstet Gynecol* 1979;**133**:176-183.

64. Saito Y, Takahashi S, Maki M. Effects of some drugs on ripening of uterine cervix in nonpregnant castrated and pregnant rats. *Tohoku J Exp Med* 1981;**133**:205-220.

65. Zuidema LJ, Khan-Dawood F, Dawood MY, Work BA, Jr. Hormones and cervical ripening: dehydroepiandrosterone sulfate, estradiol, estriol, and progesterone. *Am J Obstet Gynecol* 1986;**155**:1252-1254.

66. Hegele-Hartung C, Chwalisz K, Beier HM, Elger W. Ripening of the uterine cervix of the guinea-pig after treatment with the progesterone antagonist onapristone (ZK 98.299): an electron microscopic study. *Hum Reprod* 1989;**4**:369-377.

67. Stiemer B, Elger W. Cervical ripening of the rat in dependence on endocrine milieu; effects of antigestagens. *J Perinat Med* 1990;**18**:419-429.

68. Norman J. Antiprogesterones. *Br J Hosp Med* 1991;**45**:372-375.

69. Ito A, Imada K, Sato T, Kubo T, Matsushima K, Mori Y. Suppression of interleukin 8 production by progesterone in rabbit uterine cervix. *Biochem J* 1994;**301 (Pt 1)**:183-186.

70. Mahendroo MS, Cala KM, Russell DW. 5 alpha-reduced androgens play a key role in murine parturition. *Mol Endocrinol* 1996;**10**:380-392.

71. Chwalisz K, Garfield RE. Regulation of the uterus and cervix during pregnancy and labor. Role of progesterone and nitric oxide. *Ann N Y Acad Sci* 1997;**828**:238-253.

72. Elliott CL, Brennand JE, Calder AA. The effects of mifepristone on cervical ripening and labor induction in primigravidae. *Obstet Gynecol* 1998;**92**:804-809.

73. Mahendroo MS, Porter A, Russell DW, Word RA. The parturition defect in steroid 5alpha-reductase type 1 knockout mice is due to impaired cervical ripening. *Mol Endocrinol* 1999;**13**:981-992.

74. Stenlund PM, Ekman G, Aedo AR, Bygdeman M. Induction of labor with mifepristone--a randomized, double-blind study versus placebo. *Acta Obstet Gynecol Scand* 1999;**78**:793-798.

75. Bennett P, Allport V, Loudon J, Elliott C. Prostaglandins, the fetal membranes and the cervix. *Front Horm Res* 2001;**27**:147-164.

76. Ekman-Ordeberg G, Stjernholm Y, Wang H, Stygar D, Sahlin L. Endocrine regulation of cervical ripening in humans--potential roles for gonadal steroids and insulin-like growth factor-I. *Steroids* 2003;**68**:837-847.

77. Stjernholm-Vladic Y, Wang H, Stygar D, Ekman G, Sahlin L. Differential regulation of the progesterone receptor A and B in the human uterine cervix at parturition. *Gynecol Endocrinol* 2004;**18**:41-46.

78. Tornblom SA, Patel FA, Bystrom B, Giannoulias D, Malmstrom A, Sennstrom M, Lye SJ, Challis JR, Ekman G. 15-hydroxyprostaglandin dehydrogenase and cyclooxygenase 2 messenger ribonucleic acid expression and immunohistochemical localization in human cervical tissue during term and preterm labor. *J Clin Endocrinol Metab* 2004;**89**:2909-2915.

79. Marx SG, Wentz MJ, Mackay LB, Schlembach D, Maul H, Fittkow C, Given R, Vedernikov Y, Saade GR, Garfield RE. Effects of progesterone on iNOS, COX-2, and collagen expression in the cervix. *J Histochem Cytochem* 2006;**54**:623-639.

80. Word RA, Li XH, Hnat M, Carrick K. Dynamics of cervical remodeling during pregnancy and parturition: mechanisms and current concepts. *Semin Reprod Med* 2007;**25**:69-79.

81. Andersson S, Minjarez D, Yost NP, Word RA. Estrogen and progesterone metabolism in the cervix during pregnancy and parturition. *J Clin Endocrinol Metab* 2008;**93**:2366-2374.

82. Elovitz MA, Gonzalez J. Medroxyprogesterone acetate modulates the immune response in the uterus, cervix and placenta in a mouse model of preterm birth. *J Matern Fetal Neonatal Med* 2008;**21**:223-230.

83. Xu H, Gonzalez JM, Ofori E, Elovitz MA. Preventing cervical ripening: the primary mechanism by which progestational agents prevent preterm birth? *Am J Obstet Gynecol* 2008;**198**:314. e311-318.

84. Yellon SM, Ebner CA, Elovitz MA. Medroxyprogesterone acetate modulates remodeling, immune cell census, and nerve fibers in the cervix of a mouse model for inflammation-induced preterm birth. *Reprod Sci* 2009;**16**:257-264.

85. Csapo AI, Knobil E, van der Molen HJ, Wiest WG. Peripheral plasma progesterone levels during human pregnancy and labor. *Am J Obstet Gynecol* 1971;**110**:630-632.

86. Csapo AI, Puri CP, Tarro S. Relationship between timing of ovariectomy and maintenance of pregnancy in the guinea-pig. *Prostaglandins* 1981;**22**:131-140.

87. United States Food and Drug Administration. Drugs@FDA, Crinone [cited; Available from: http://www.accessdata.fda.gov/scripts/cder/drugsatfda/index.cfm?fuseaction=Search.Overview&DrugName=CRINONE

88. Beta J, Akolekar R, Ventura W, Syngelaki A, Nicolaides KH. Prediction of spontaneous preterm delivery from maternal factors, obstetric history and placental perfusion and function at 11-13 weeks. *Prenat Diagn* 2011;**31**:75-83.

89. Altman D, Carroli G, Duley L, Farrell B, Moodley J, Neilson J, Smith D. Do women with pre-eclampsia, and their babies, benefit from magnesium sulphate? The Magpie Trial: a randomised placebo-controlled trial. *Lancet* 2002;**359**:1877-1890.

90. Sinclair JC. Meta-analysis of randomized controlled trials of antenatal corticosteroid for the prevention of respiratory distress syndrome: discussion. *Am J Obstet Gynecol* 1995;**173**:335-344.

Figure 1. Participant Flow Diagram

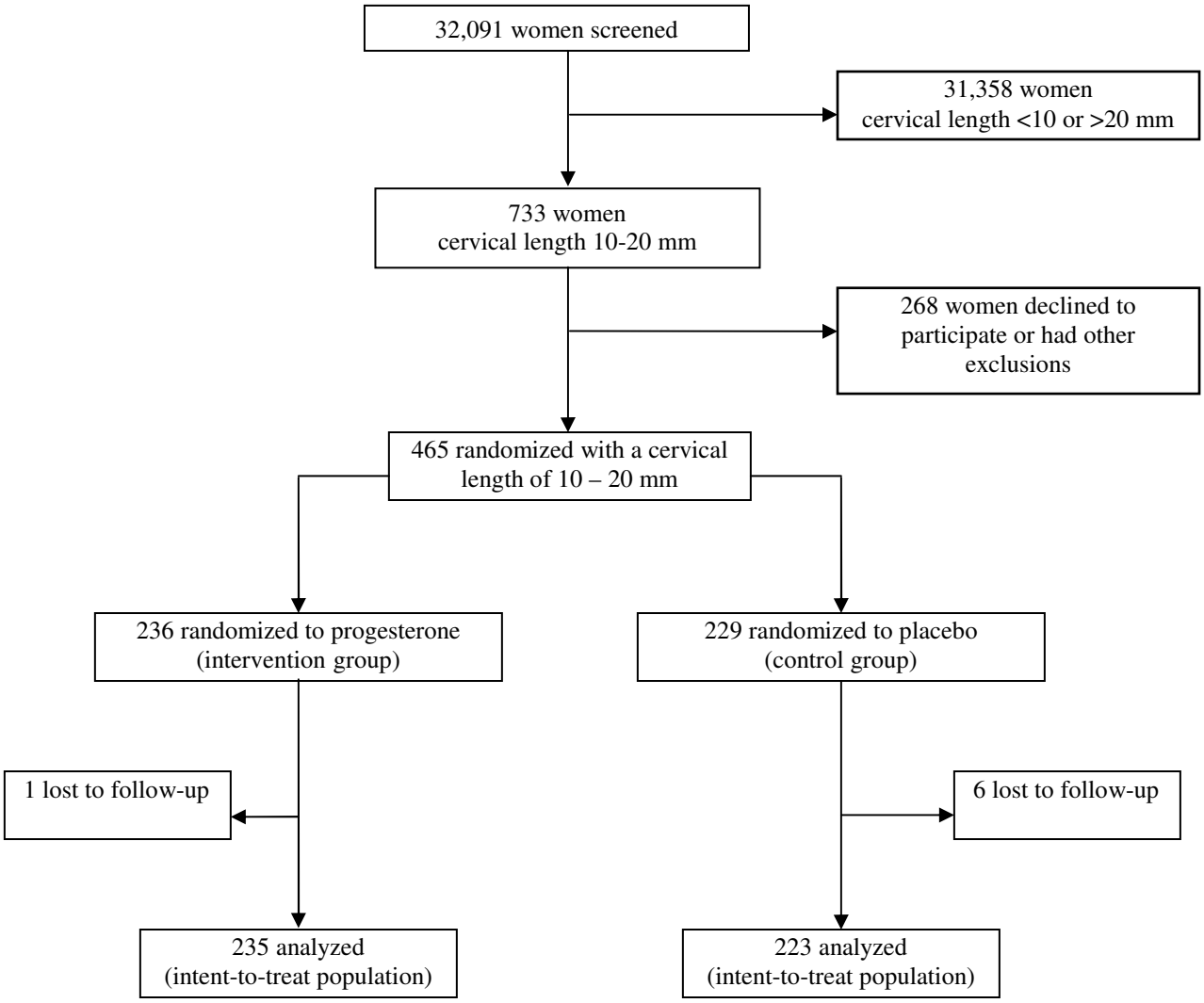
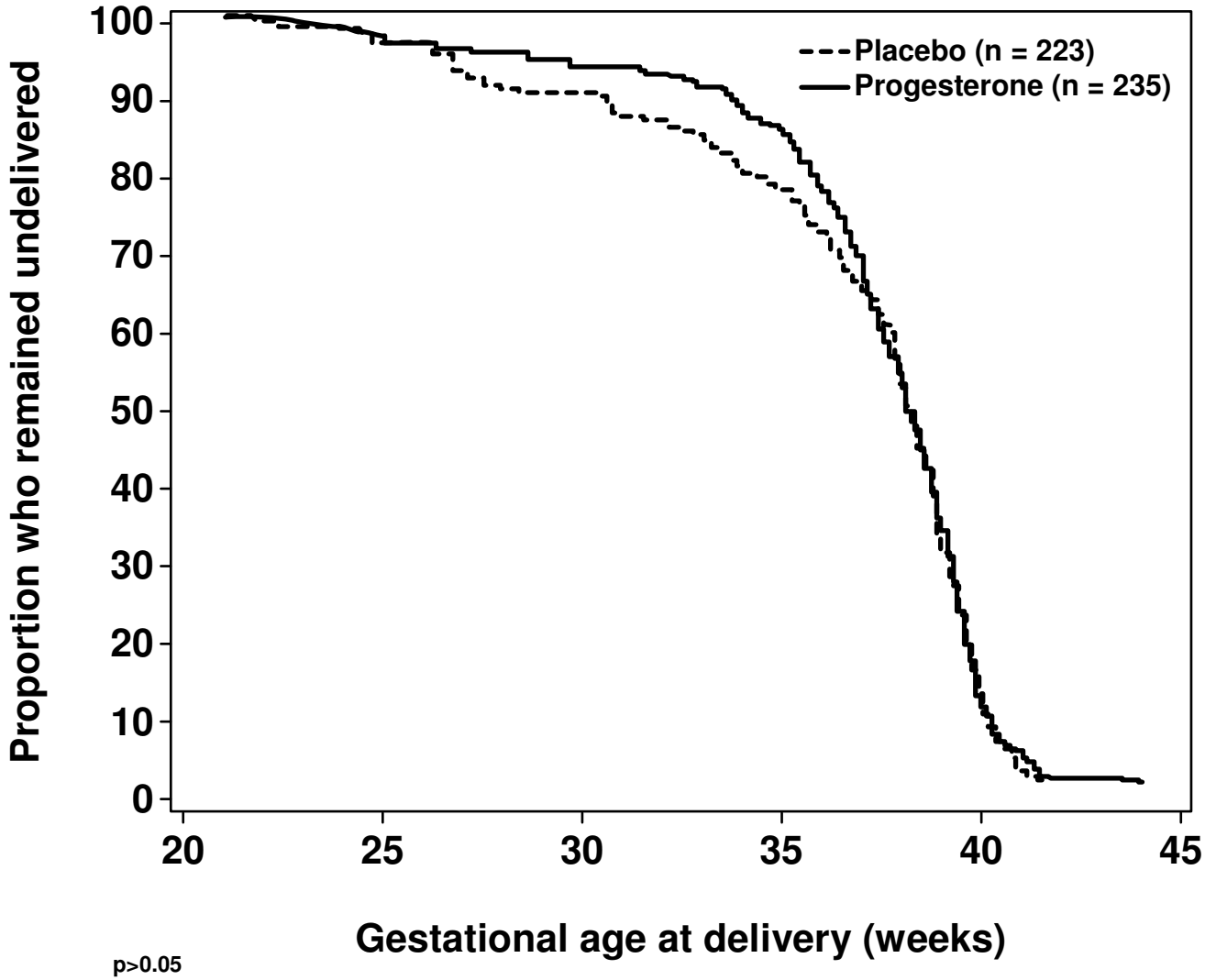
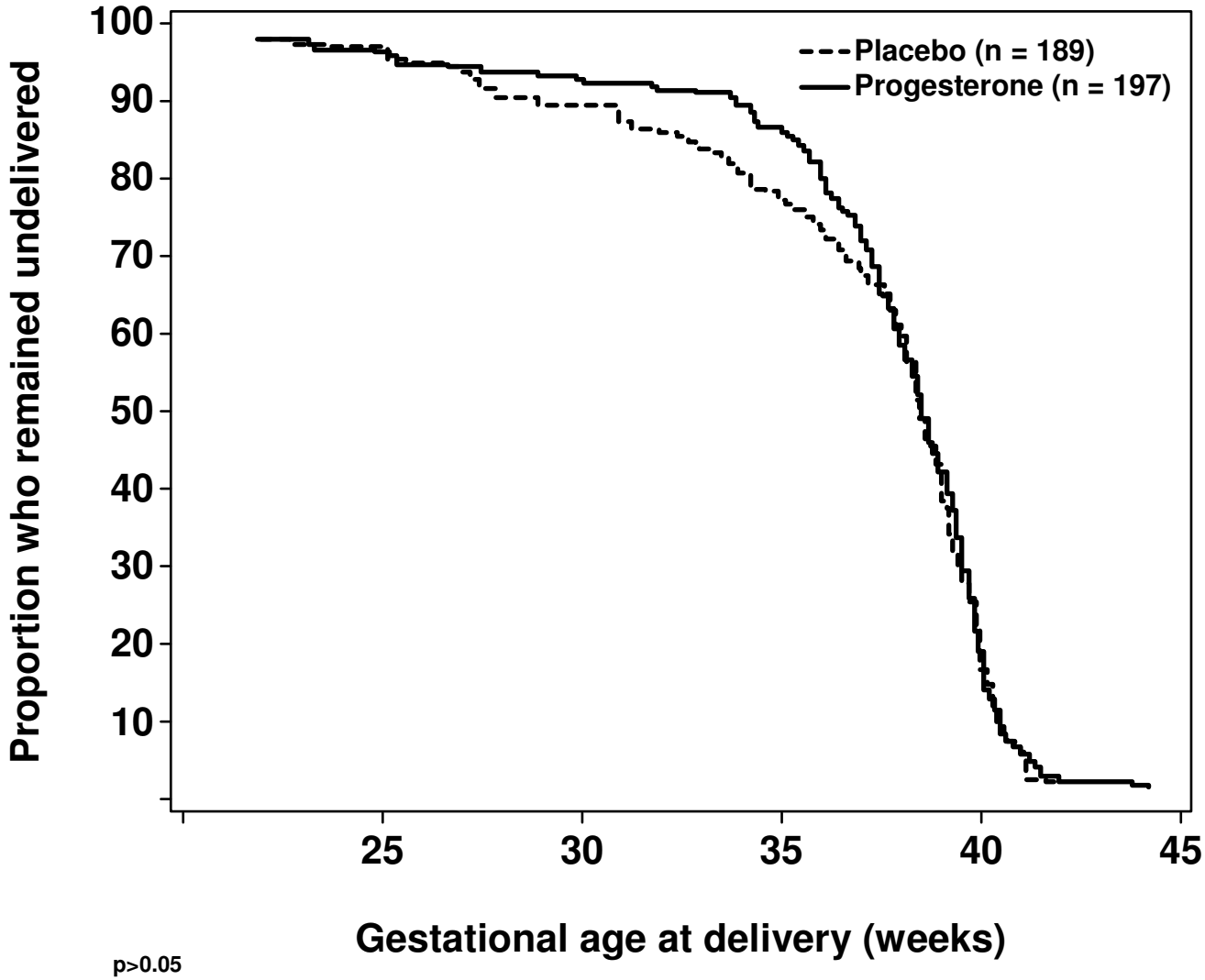


Figure 2A. Survival analysis of Intent-to-Treat Analysis Set – Proportion of Patients who remain undelivered according to treatment allocation (progesterone versus placebo)
This figure includes the Whole Population (patients with and without a prior history of preterm delivery)



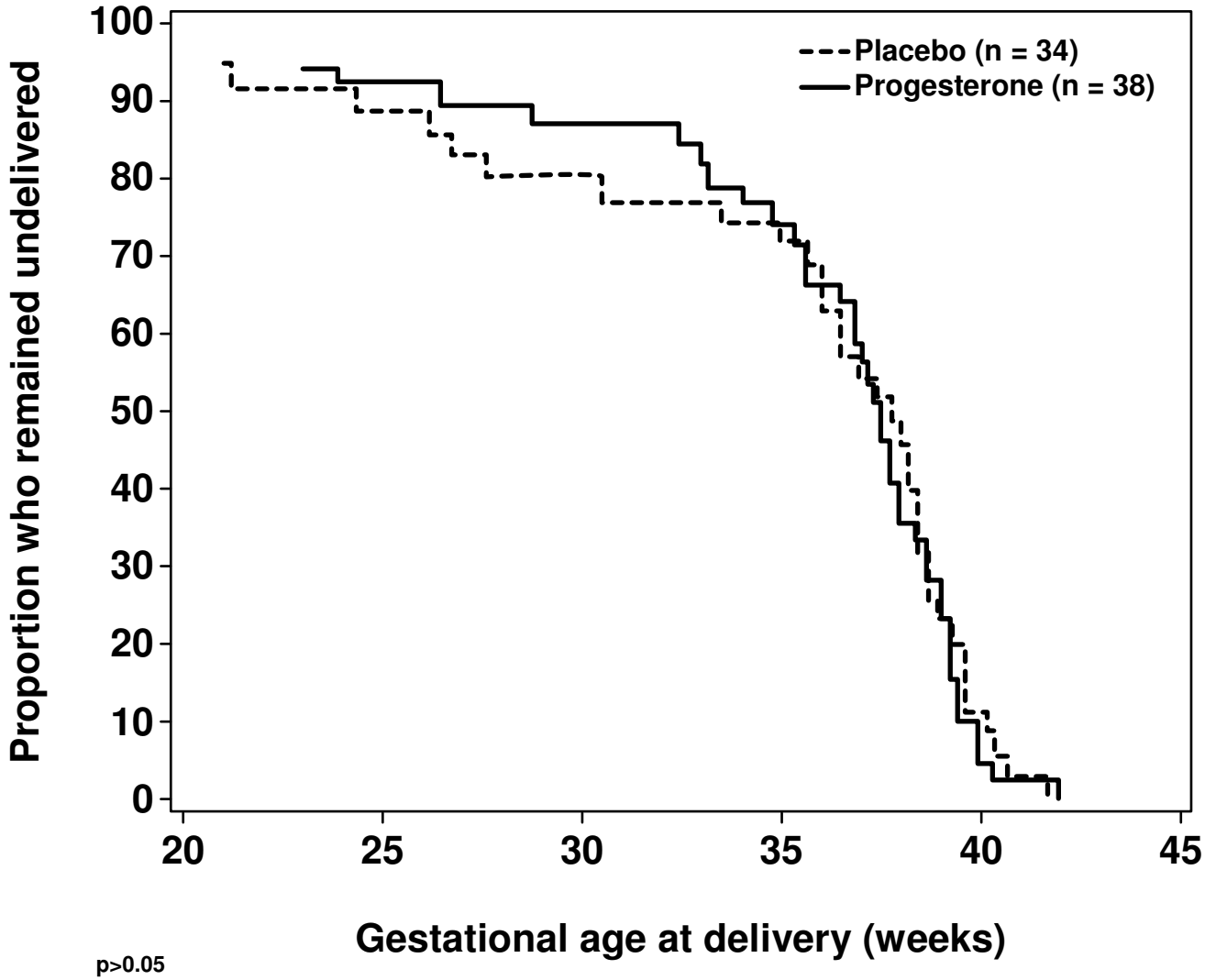
p>0.05

Figure 2B. Survival analysis of Intent-to-Treat Analysis Set - Proportion of Patients who remain undelivered according to treatment allocation (progesterone versus placebo)
This figure includes the Patients Without a Prior History of Preterm Delivery



p>0.05

Figure 2C. Survival analysis of Intent-to-Treat Analysis Set - Proportion of Patients who remain undelivered according to treatment allocation (progesterone versus placebo)
This figure includes the Patients With a Prior History of Preterm Delivery



p>0.05

Table 1 Baseline and Treatment Characteristics

	Vaginal Progesterone (n=235)	**Placebo (n = 223)**
Age (years)		
Median	25.3	25.6
Range	(18-44)	(18-41)
Interquartile Range	(21.8 –30.3)	(21.9 – 29.4)
Mean (standard deviation)	26.5 (5.8)	26.2 (5.1)
Race (n, %)		
African-American	76 (32%)	67 (30%)
Asian	76 (32%)	74 (33%)
Caucasian	73 (31%)	70 (31%)
Other	10 (4%)	12 (5%)
Body Mass Index (kg/m^2)		
Median	24.5	23.6
Range	(14-47)	(14-50)
Interquartile Range	(20.4 – 30.0)	(20.5 – 29.2)
Mean (standard deviation)	25.6 (6.3)	25.3 (6.8)
Obstetrical History (n, %)		
Nulliparous	125 (53%)	126 (57%)
No previous preterm birth (>20 weeks, <32 weeks)	204 (87%)	195 (87%)
≥ 1 previous preterm birth	31 (13%)	28 (13%)
Cervical Length (mm)		
Median	18	18
Range	(10-21)	(10-20)
Interquartile Range	(16 – 19)	(15 – 19)
Mean (standard deviation)	17 (2.5)	17 (2.8)
Gestational Age at the First Dose of Progesterone (weeks)		
Median	21.7	21.7
Range	(19-25)	(17-25)
Interquartile Range	(20.7 – 23.0)	(20.4 – 22.9)
Mean (standard deviation)	21.9 (1.4)	21.7 (1.4)
Duration of Treatment (weeks)		
Median	14.3	13.9
Range	(0-18)	(0-18)
Interquartile Range	(12.6 – 15.7)	(10.9 – 15.7)
Mean (standard deviation)	13.0 (4.2)	12.5 (4.7)
***Compliance (%)**		
Median	99.2	100
Range	6-100	0-100
Interquartile Range	(92.7 – 100)	(93.0 – 100)
Mean (standard deviation)	93.3 (13.1)	94.0 (12.7)

*Compliance was calculated using the following formula:

$$\frac{\text{Number of vaginal applicators used since last visit}}{\text{Number of vaginal applicators that should have been used since last visit}} \times 100$$

Every two weeks, a percentage of compliance was calculated and the compliance for a specific patient is based on the average of all visits. The definition of compliance is based on the formula and percentage indicated above, and a compliant patient was defined as one with an average of >80% compliance.

Table 2 Gestational age at delivery and neonatal outcome in women allocated to progesterone compared to those allocated to receive placebo **– Intent to treat analysis set**

Outcome	Vaginal Progesterone (N=235)	Placebo (N=223)	RR (95% CI)	p value
Primary Outcome				
Preterm birth <33^{+0} weeks	21/235 (8.9)	36/223 (16.1)	0.55 (0.33- 0.92)	0.020
Secondary Outcomes				
Preterm birth <28^{+0} weeks	12/235 (5.1)	23/223 (10.3)	0.50 (0.25-0.97)	0.036
Preterm birth <35^{+0} weeks	34/235 (14.5)	52/223 (23.3)	0.62 (0.42- 0.92)	0.016
Preterm birth <37^{+0} weeks	71/235 (30.2)	76/223 (34.1)	0.89 (0.68-1.16)	0.376
Respiratory distress syndrome	7/235 (3.0)	17/223 (7.6)	0.39 (0.17- 0.92)	0.026
Bronchopulmonary dysplasia	4/235 (1.7)	5/223 (2.2)	0.76 (0.21-2.79)	0.678
Proven sepsis	7/235 (3.0)	6/223 (2.7)	1.11 (0.38-3.24)	0.853
Necrotising enterocolitis	5/235 (2.1)	4/223 (1.8)	1.19 (0.32-4.36)	0.797
Grade III/IV intraventricular haemorrhage	0/235 (0.0)	1/223 (0.5)	0.32 (0.01-7.73)*	0.305
Periventricular leukomalacia	0/235 (0.0)	0/223 (0.0)	Non estimable	NA
Perinatal death	8/235 (3.4)	11/223 (4.9)	0.69 (0.28-1.68)	0.413
Fetal death	5/235 (2.1)	6 /223 (2.7)	0.79 (0.25-2.57)	0.700
Neonatal death	3/235 (1.3)	5/223 (2.2)	0.57 (0.14-2.35)	0.431
Any morbidity/mortality event	18/235 (7.7)	30/223 (13.5)	0.57 (0.33- 0.99)	0.043
0-4 without NICU†				0.048
0-4 with NICU†				0.068
0-6 without NICU†				0.048
Birth weight < 2500 grams	60/234 (25.6)	68/220 (30.9)	0.83 (0.62-1.11)	0.213
Birth weight < 1500 grams	15/234 (6.4)	30/220 (13.6)	0.47 (0.26-0.85)	0.010

* based on Logit estimator with continuity correction

Data are number (%) unless otherwise indicated

NA=not applicable

Unadjusted RR and CI calculated using the Cochran-Mantel-Haenszel (CMH) test

† Frequency of mortality/morbidity composite scores are provided in supplementary material, S4

Table 3 Gestational age at delivery and neonatal outcome in women allocated to progesterone compared to those allocated to receive placebo – **Treated Patient Analysis Set**

	Progesterone (N=235)	**Placebo (N=224)**	**Unadjusted RR (CMH)***	**p-value***	**Adjusted RR (CMH)†**	**p- value†**
Gestational Age at Delivery						
Primary Outcome						
< 33^{+0} weeks	21 (8.9)	34 (15.2)	0.59 (0.35-0.98)	0.040	0.56 (0.33 – 0.93)	0.022
Secondary Outcomes						
< 28^{+0} weeks	12 (5.1)	21 (9.4)	0.54 (0.27-1.08)	0.077	0.55 (0.28 - 1.08)	0.075
< 35^{+0} weeks	34 (14.5)	50 (22.3)	0.65 (0.44-0.96)	0.030	0.61 (0.41 – 0.90)	0.012
< 37^{+0} weeks	71 (30.2)	74 (33.0)	0.91 (0.70-1.20)	0.516	0.89 (0.68 – 1.15)	0.377
Neonatal Outcomes						
RDS	7 (3.0)	16 (7.1)	0.42 (0.17-0.99)	0.041	0.42 (0.18 – 0.97)	0.036
BPD	4 (1.7)	5 (2.2.)	0.77 (0.21-2.80)	0.683	0.78 (0.21 – 2.83)	0.701
Proven Sepsis	7 (3.0)	5 (2.2)	1.33 (0.43-4.14)	0.617	1.37 (0.45 – 4.17)	0.577
NEC	5 (2.1)	4 (1.8)	1.19 (0.32-4.38)	0.792	1.21 (0.34 – 4.30)	0.769
IVH Grade III/IV	0	1 (0.5)	0.32 (0.01-7.76)‡	0.306	0.32 (0.01-7.48)‡	0.307
PVL	0	0	Non estimable	NA	Non estimable	NA
Perinatal Death	8 (3.4)	10 (4.5)	0.76 (0.31-1.90)	0.559	0.78 (0.31 – 1.97)	0.596
Neonatal Death	3 (1.3)	5 (2.2)	0.57 (0.14-2.37)	0.435	0.57 (0.14 – 2.36)	0.436
Any morbidity/mortality event	18 (7.7)	28 (12.5)	0.61 (0.35-1.08)	0.085	0.62 (0.36 – 1.08)	0.088
Birthweight < 2500g	60/234 (25.6)	67/218 (30.7)	0.83 (0.62-1.12)	0.229	0.83 (0.62 – 1.11)	0.204
< 1500g	15/234 (6.4)	29/218 (13.3)	0.48 (0.27-0.87)	0.014	0.49 (0.27 – 0.88)	0.014

* Unadjusted RR and CI calculated using the Cochran-Mantel-Haenszel (CMH) method; P-value based on CMH test
† RR and CI calculated using the CMH method adjusted for pooled study site and risk strata ; P-value based on CMH test adjusted for pooled study site and risk strata
‡ based on Logit estimator with continuity correction
Data are number (%); NA=not applicable

Table 4 Gestational age at delivery and neonatal outcome in women allocated to progesterone compared to those allocated to receive placebo – **Compliant Analysis Set**

	Vaginal Progesterone (N=194)	**Placebo (N=193)**	**Unadjusted RR (CMH)***	**p-value***	**Adjusted RR (CMH)†**	**p-value†**
Gestational Age at Delivery						
Primary Outcome						
< 33^{+0} weeks	11 (5.7%)	25 (13.0%)	0.44 (0.22-0.86)	0.014	0.42 (0.22-0.82)	0.009
Secondary Outcomes						
< 28^{+0} weeks	6 (3.1)	15 (7.8)	0.40 (0.16-1.00)	0.043	0.40 (0.16-1.03)	0.048
< 35^{+0} weeks	20 (10.3)	39 (20.2)	0.51 (0.31-0.84)	0.007	0.50 (0.31-0.82)	0.005
< 37^{+0} weeks	52 (26.8)	59 (30.6)	0.88 (0.64-1.20)	0.413	0.85 (0.62-1.17)	0.326
Neonatal Outcomes						
RDS	7 (3.6)	14 (7.3)	0.50 (0.21-1.21)	0.114	0.48 (0.19-1.17)	0.098
BPD	3 (1.6)	4 (2.1)	0.75 (0.17-3.29)	0.698	0.85 (0.18-3.90)	0.832
Proven Sepsis	6 (3.1)	5 (2.6)	1.19 (0.37-3.85)	0.767	1.18 (0.35-3.92)	0.789
NEC	4 (2.1)	3 (1.6)	1.33 (0.30-5.85)	0.708	1.41 (0.34-5.80)	0.634
IVH Grade III/IV	0	1 (0.5)	0.33 (0.01-8.09)‡	0.316	0.39 (0.02-8.93)‡	0.355
PVL	0	0	Non estimable	NA	Non estimable	N/A
Perinatal Death	3 (1.6)	6 (3.1)	0.50 (0.13-1.96)	0.309	0.43 (0.10-1.90)	0.248
Neonatal Death	2 (1.0)	3 (1.6)	0.66 (0.11-3.93)	0.649	0.70 (0.12-4.18)	0.697
Any morbidity/mortality event	11 (5.7)	21 (10.9)	0.52 (0.26-1.05)	0.063	0.50 (0.24-1.03)	0.053
Birthweight < 2500g	45 (23.2)	54/192 (28.1)	0.82 (0.59-1.16)	0.268	0.80 (0.57-1.13)	0.210
< 1500g	8 (4.1)	22/192 (11.5)	0.36 (0.16-0.79)	0.007	0.37 (0.17-0.80)	0.008

* Unadjusted RR and CI calculated using the Cochran-Mantel-Haenszel (CMH) method; P-value based on CMH test
† RR and CI calculated using the CMH method adjusted for pooled study site and risk strata ; P-value based on CMH test adjusted for pooled study site and risk strata
‡ based on Logit estimator with continuity correction
Data are number (%); NA=not applicable

Supplementary Material

S1. Definitions of Neonatal Morbidity/Mortality*:*

Intraventricular Hemorrhage[1] (as determined by cranial ultrasound or CT)

Grade I – subependymal hemorrhage

Grade II – intraventricular hemorrhage, uncomplicated

Grade III – intraventricular hemorrhage with ventricular dilatation

Grade IV – intraventricular hemorrhage with ventricular dilatation and parenchymal extension

Necrotizing Enterocolitis[2]

Surgical – Stage III – Advanced

- Treatment was surgical

Other findings may include:

- perinatal stress
- systemic manifestations such as temperature instability, lethargy, apnea, bradycardia, occult or gross GI bleeding, abdominal distension, plus septic shock
- radiographs show: intestinal distension with ileus, small bowel separation, rigid bowel loops, pneumatosis intestinalis, portal vein gas, pneumoperitoneum

Clinical – Stage II – Definite

- Treatment was medical

Other findings may include:

- perinatal stress
- systemic manifestations such as temperature instability, lethargy, apnea, bradycardia, occult or gross GI bleeding, abdominal distension
- radiographs show: intestinal distension with ileus, small bowel separation, rigid bowel loops, pneumatosis intestinalis, portal vein gas

Other – Stage I – Suspect

- Treatment was observation

Other findings may include:

- perinatal stress
- systemic manifestations such as temperature instability, lethargy, apnea, bradycardia
- radiographs show: intestinal distension with ileus

<u>Respiratory Distress Syndrome (both diagnosis and oxygen therapy)</u>

- Clinical Diagnosis of at least RDS type I (one or more of the following):
    - tachypnoea (respiratory rate > 60 breaths per minute)
    - intercostal, subcostal, and sternal recession
    - expiratory grunting
    - cyanosis
    - diminished breath sounds
- oxygen therapy ($FiO_2 \geq 0.40$) until infant death or $\geq$ 24 hours

Retinopathy[3]

- Stage I (ophthalmoloscopic demarcation line of normal and abnormal vessels)
- Stage II (intraretinal ridge (ridge that rises up from the retina as a result of the growth of the abnormal vessels)
- Stage III (ridge with extraretinal fibrovascular proliferation (the ridge grows from the spread of the abnormal vessels and extends into the vitreous)

Bronchopulmonary Dysplasia

- Treatment with > 21% O_2 for at least 28 days, or
- O_2 dependence after 36 weeks post-conceptional age

Proven Sepsis

- Clinically ill infant with suspected infection plus
- Positive blood, CSF, or catheterized/suprapubic urine culture or cardiovascular collapse or unequivocal X-ray finding

Definitions of Composite Neonatal Outcome Scores:

1) A '0 to 4 point scale' composite neonatal morbidity/mortality score: This score was derived as an ordinal scale based upon severity. The score was defined by the following: 0=no events; 1=one event for (RDS, BPD, grade III or IV IVH, PVL, proven sepsis, or NEC) and no perinatal mortality, 2=two events and no perinatal mortality; 3=three or more events and no perinatal mortality; and 4=perinatal mortality.

2) ‘0 to 4 point scale with NICU days’: This score was defined as the following: 0=no events, 1=one event for (RDS, BPD, grade III or IV IVH, PVL, proven sepsis, or NEC) or <5 days in the NICU, and no perinatal mortality; 2=two events or between 5 and 20 days in the NICU, and no perinatal mortality; 3=three or more events or >20 days in the NICU, and no perinatal mortality; and 4=perinatal mortality.

3) A ‘0 to 6 point scale’: This score was defined as the following: 0=no events; 1=one event for (RDS, BPD, grade III or IV IVH, proven sepsis, or NEC) and no perinatal mortality, 2=two events and no perinatal mortality; 3=three events and no perinatal mortality; 4=four events and no perinatal mortality; 5=five events and no perinatal mortality; and 6=perinatal mortality.

4) Any morbidity or mortality event: (yes/no)

S2. Definition of Adverse Events

The Medical Dictionary for Regulatory Activities (MedDRA) dictionary (version 11.0) was used to classify all adverse events reported during the study by system organ class and preferred term. The incidence of treatment-emergent adverse events (TEAEs) was also determined. TEAEs were defined as those adverse events that either had an onset time on or after the start of study drug and no more than 30 days after the last dose of study drug, or were ongoing at the time of study drug initiation and increased in severity, or became closer in relationship to study drug during the treatment period.

S3. Trial Profile

This section describes patients lost to follow-up and protocol violations.

Patients lost to follow-up: There were seven patients lost to follow-up in which the investigators were not able to obtain delivery date. Six patients had been allocated to the placebo group and one to the progesterone group.

Protocol violations: This will be itemized by category:

a) One patient had a cervical length of 21 mm when the upper limit of cervical length for enrollment was 20 mm. This patient was randomised to receive progesterone.

b) One patient was enrolled despite having had a prophylactic cerclage. The protocol required that patients with a cerclage be excluded from participation. This patient was allocated to the placebo group.

c) One patient had a positive test for HIV. The protocol specified that patients testing positive for HIV should be excluded. She was allocated to receive progesterone.

d) Two patients were prescribed progesterone administration. The protocol specified that patients should not have progesterone administration. These two patients were allocated to progesterone administration in the trial.

e) A total of 55 patients began study drug before or after the planned interval of 20 – 23 6/7 weeks, as specified in the protocol, based on the date of the first dose of study drug and the accepted estimated date of confinement. The specific detail for these patients is the following:

i. 20 patients allocated to placebo began therapy before 20 weeks; range 17-19 6/7 weeks

ii. 9 patients allocated to progesterone began therapy before 20 weeks; range 19 – 19 6/7 weeks

iii. 7 patients allocated to placebo began therapy after 23 6/7 weeks; range 24 – 25 weeks

iv. 19 patients allocated to progesterone began therapy after 23 6/7 weeks; range 24 – 25 3/7 weeks

S4. Frequency Distributions for Perinatal Mortality/Neonatal Morbidity Composite Scores – ITT analysis set

0 – 4 scale

Score	Placebo n	Prochieve n
0	192	217
1	11	5
2	8	2
3	0	3
4	11	8

0 – 4 scale with NICU

Score	Placebo n	Prochieve n
0	168	194
1	11	6
2	17	19
3	15	8
4	11	8

0 – 6 scale

Score	Placebo n	Prochieve n
0	192	217
1	11	5
2	8	2
3	0	0
4	0	3
5	0	0
6	11	8

References

1. Papile LA, Burstein J, Burstein R, Koffler H. Incidence and evolution of subependymal and intraventricular hemorrhage: a study of infants with birth weights less than 1,500 gm. *J Pediatr* 1978;**92**:529–34.

2. Bell MJ, Ternberg JL, Feigin RD, et al. Neonatal necrotizing enterocolitis. Therapeutic decisions based upon clinical staging. *Ann Surg* 1978;**187**:1–7.

3. The International Classification of Retinopathy of Prematurity Revisited. *Arch Ophthalmol* 2005;**123**:991–99.