

Original Article

Prediction of spontaneous preterm birth in patients with congenital uterine anomalies using combined fetal fibronectin and cervical length

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Abstract: Objective: To evaluate cervical length (CL) and fetal fibronectin (fFN) as predictors of spontaneous preterm birth (SPTB) in patients with congenital uterine anomalies. Methods: A retrospective cohort of patients with uterine anomalies and singleton pregnancies >22 weeks delivered by one maternal fetal medicine practice from 2005-2012 who underwent routine CL and fFN testing from 22-32 weeks was identified. We excluded patients with a cerclage, and patients who had an indicated preterm birth <37 weeks. A short CL was defined as ≤ 25 mm. We compared rates of spontaneous preterm birth <37 weeks based on CL and fFN results. Results: 120 patients with uterine anomalies were identified (47 septate, 36 bicornuate, 12 unicornuate, 10 arcuate, 9 T-shaped, 6 didelphys). Seventeen (14.2%) of patients had a short CL; these patients had a higher incidence of spontaneous preterm birth <37 weeks (64.7% vs. 13.6%, $p < 0.001$). Six of 89 patients (6.7%) had a positive fFN; these patients did not have an increased incidence of spontaneous preterm birth than patients with a negative fFN. The addition of fFN to CL testing did not improve the positive or negative predictive values, nor the positive or negative likelihood ratios for spontaneous preterm birth <37 weeks. Conclusions: In women with uterine anomalies, a short CL is significantly associated with spontaneous preterm birth <37 weeks. The addition of fFN testing in these patients does not improve the prediction of preterm birth.

Keywords: Uterine anomaly, preterm birth, fetal fibronectin, cervical length, prediction

Introduction

Congenital anomalies of the uterus, or congenital müllerian anomalies, include a spectrum of uterine abnormalities caused by abnormal embryologic fusion and canalization of the müllerian ducts to form a normal uterine cavity. These anomalies are often asymptomatic and unrecognized, but have a reported prevalence of approximately 2-4% in reproductive age women [1-4], and up to 5-25% in women with adverse reproductive outcomes [4, 5]. The presence of a uterine anomaly appears to increase the risk of adverse pregnancy outcomes. Although limited by selection bias and small sample sizes, most of the data in singleton pregnancies suggest that patients with uterine anomalies are at increased risk for certain adverse pregnancy outcomes, including

preterm birth, cesarean delivery, and fetal growth restriction [6-11].

The presence of cervicovaginal fetal fibronectin (fFN) [12, 13] or a short cervical length (CL) on ultrasound [14] are both predictors of preterm birth in singleton pregnancies. In patients with uterine anomalies, a short cervix has been associated with spontaneous preterm birth [15, 16]. One study of 64 patients showed that 16% had a short CL (defined as 25 mm or less) from 14-23 6/7 weeks, and that these patients were at increased risk of spontaneous preterm birth <35 weeks [15]. Another study of 52 patients with uterine anomalies showed that women with a bicornuate uterus and a short CL (defined as less than 30 mm) from 16-30 weeks had an increased risk of spontaneous preterm birth <35 weeks. However, data regarding fetal fibro-

Cervical length and fetal fibronectin in uterine anomalies

nectin in patients with uterine anomalies is limited.

The objective of this study was to evaluate fFN, CL, and the combination of both as predictors of spontaneous preterm birth in a large cohort of asymptomatic patients with congenital uterine anomalies.

Materials and methods

After Institutional Review Board approval was obtained, a retrospective cohort of patients was obtained from patients managed in one Maternal Fetal Medicine practice between 2005 and 2012. Our study cohort included all patients with a uterine anomaly diagnosed prior to or during pregnancy who delivered in our practice after 22 weeks. The patients were identified by querying our electronic database, which was created in 2005, for all patients with a uterine anomaly. The diagnosis of a uterine anomaly was made prepregnancy either by a saline infusion sonohysterogram (SIS), MRI, hysteroscopy, laparoscopy, or a combination of the above. Some of the uterine anomalies were diagnosed in our practice; others were diagnosed by outside centers prepregnancy. For all patients with a diagnosis of a uterine anomaly made at an outside center, the medical records and imaging reports were reviewed to ensure accuracy of diagnosis. Classification of uterine anomalies was made according to the 1988 American Fertility Society classification [17]. We considered the following uterine anomalies: arcuate, septate, unicornuate, bicornuate, t-shaped, and didelphys. Patients with a uterine septum included those with an intact septum and those who underwent hysteroscopic resection prior to pregnancy. Over the course of the study period, patients with uterine anomalies underwent routine CL and fFN screening every 2-3 weeks from 22-32 weeks. We excluded patients with multiple pregnancies, patients with a cerclage, patients without either CL or fFN testing, and patients with an indicated preterm birth <37 weeks. All patients were managed in our practice for the entire pregnancy and delivery.

All CL measurements and fFN testing were done in an outpatient setting on asymptomatic patients. All tests done on labor and delivery were excluded, as they were done on symptomatic patients as part of a preterm labor evaluation. Patients and obstetricians were not blind-

ed to the CL measurements or fFN results. Gestational age was based on the last known menstrual period and confirmed by ultrasound in all patients. Patients in our practice routinely have first and second trimester ultrasound. The expected date of delivery was revised if the discrepancy was >5 days between the calculation from the last menstrual period and ultrasound scan up to 14 weeks gestation or >7 days if the dating ultrasound scan was performed after 14 weeks gestation. If the pregnancy was the result of *in-vitro* fertilization (IVF), gestational age was determined from the date of embryo transfer.

All CL measurements were measured with an empty bladder with the optimal image defined according to the criteria reported by Iams et al [14]. The shortest functional CL was used as this has been found to be the most reproducible measurement [18]. A short CL was defined as a CL \leq 25 mm any time between 22 and 32 weeks. Fetal fibronectin testing was performed using a Dacron swab without the use of a speculum according to an established protocol that has been validated previously by both our group [19] and others [20]. Fetal fibronectin testing was performed more than 24 hours from the last reported intercourse or endovaginal ultrasound. A fetal fibronectin concentration of 50 ng/mL or greater was considered to be positive. Subsequent to 2007, patients with a short CL 15 mm or less were prescribed vaginal progesterone 200 mg at night [21].

The records for each patient were reviewed. The primary outcome was the incidence of spontaneous preterm birth <37 weeks. Assuming a background risk of preterm birth <37 weeks of 40% in patients with uterine anomalies [11] and a 16% incidence of short cervix [15], in order to have 80% power to demonstrate an increase in spontaneous preterm birth from 20% to 60% with an alpha error of 5%, 120 total patients with uterine anomalies would be needed. Fisher's exact test, chi square test, and Student's t-test, were used when appropriate (SPSS for Windows 16.0, Chicago 2007). A *p*-value of <0.05 was considered significant.

Results

Over the course of the study period, we delivered 4473 singleton pregnancies \geq 22 weeks, 158 of whom had a uterine anomaly, for an

Cervical length and fetal fibronectin in uterine anomalies

Table 1. Cervical length, fetal fibronectin, and spontaneous preterm birth according to uterine anomaly

Anomaly	N	CL ≤25 mm	fFN positive	Spontaneous preterm birth <37 weeks
Unicornuate	12	3 (25.0%)	1/9 (22.2%)	4 (33.3%)
Bicornuate	36	5 (13.9%)	1/25 (4.0%)	11 (30.6%)
Didelphys	6	1 (16.7%)	1/6 (16.7%)	2 (33.3%)
Arcuate	10	1 (10.0%)	0/6 (0%)	1 (10.0%)
Septate	47	6 (12.8%)	1/36 (2.8%)	6 (12.8%)
T-shaped	9	1 (11.1%)	1/7 (14.3%)	1 (11.1%)

CL, cervical length; fFN, fetal fibronectin.

overall prevalence of 3.5%. Of the 158 patients with a uterine anomaly, the frequency of each specific anomaly in descending order of frequency was repaired septate uterus 50 (31.6%), bicornuate 46 (29.1%), unicornuate 16 (10.1%), intact septate 16 (10.1%), arcuate 14 (8.9%), t-shaped 10 (6.3%), and didelphys 6 (3.8%). One of the patients with a bicornuate uterus underwent metroplasty prior to pregnancy. We excluded 7 patients with an indicated preterm birth <37 weeks, 12 patients with a cerclage, and 19 patients who did not undergo CL screening, leaving 120 patients for analysis.

The mean age of the cohort was 30.6±6.1 years and the mean prepregnancy BMI was 24.3±5.4 kg/m². The majority (94.2%) were white. 46.7% had a prior term birth and 29.2% had a prior preterm birth. No patients had a prior LEEP or cone biopsy. 15% of patients conceived via in-vitro fertilization.

The CL and fFN results, as well as the incidence of spontaneous preterm birth <37 weeks are listed in **Table 1**, broken down by specific uterine anomaly. The incidence of spontaneous preterm birth was highest in patients with a bicornuate, unicornuate, and didelphys uterus.

Seventeen (14.2%) of patients had a short CL ≤25 mm from 22-32 weeks. These patients delivered earlier and had a higher incidence of spontaneous preterm birth <37 weeks (64.7% vs. 13.6%, p<0.001) (**Table 2**). Of the 89 patients who underwent fFN screening, 6 (6.7%) had a positive fFN from 22-32 weeks. These patients were no more like to deliver preterm than patients with a negative fFN (**Table 3**).

The testing characteristics for CL and fFN as predictors of spontaneous preterm birth <37

weeks are shown in **Table 4**. We examined testing with CL only, fFN only, combined testing with a positive test being defined as *either* a short CL or positive fFN, and combined testing with a positive test being defined as *both* a short CL and a positive fFN. The strongest predictor of spontaneous preterm birth <37 weeks with testing with CL alone, which had a positive predictive value of 64.7%, negative predictive value of 86.4%, positive likelihood ratio of 6.98, and a negative likelihood ratio of 0.60. Neither fFN alone nor combining fFN with CL improved the positive or negative predictive values or the positive or negative likelihood ratios found with CL testing alone.

Discussion

In this study, we found that in patients with congenital uterine anomalies, a short CL was predictive of spontaneous preterm birth <37 weeks, but fFN testing was not. Due to the increased incidence of preterm birth in patients with congenital uterine anomalies, it may be clinically relevant to identify which of these patients are at highest risk for preterm birth. This could assist with decisions regarding transfer of care, patient travel, and possibly provide reassurance for the majority of patients with a uterine anomaly, as only 14% of patients in our cohort had a short CL from 22-32 weeks, similar to the 16% quoted by others [15]. Unfortunately, our ability to predict preterm birth is far greater than our current ability to prevent it. Therefore, our study does not imply that CL or fFN testing will reduce the risk of preterm birth in this population. However, as certain interventions meant to reduce the risk of preterm birth are tested in patients with uterine anomalies, it might be prudent to only include patients with a short CL, as this represents the highest risk group, and the group in which benefit could most easily be demonstrated in smaller trials. For example, if studying the effect of cervical cerclage or vaginal progesterone in reducing the risk of preterm birth in this population, only including patients with a short CL would select a population with a 65% baseline risk of preterm birth, as opposed to 40%, which is the overall baseline risk in patients with congenital uterine anomalies (11%).

Cervical length and fetal fibronectin in uterine anomalies

Table 2. Gestational age at delivery, based on cervical length prior to 32 weeks in patients with a uterine anomaly

	Cervical length ≤ 25 mm N=17	Cervical length > 25 mm N=103	p
Gestational age at delivery	35.9 \pm 3.6	38.2 \pm 2.7	0.003
Spontaneous preterm birth < 37 weeks	64.7%	13.6%	< 0.001

Table 3. Gestational age at delivery, based on fetal fibronectin result prior to 32 weeks in patients with a uterine anomaly

	Positive fetal fibronectin N=6	Negative fetal fibronectin N=83	p
Gestational age at delivery	36.4 \pm 2.9	38.10 \pm 2.7	0.178
Spontaneous preterm birth < 37 weeks	33.3%	20.5%	0.604

Table 4. Testing characteristics of fetal fibronectin (fFN) and cervical length (CL) for spontaneous preterm birth < 37 weeks in patients with a uterine anomaly

	Sensitivity	Specificity	PPV	NPV	+LR	-LR
fFN positive	10.5%	94.3%	33.3%	79.5%	1.84	0.95
CL ≤ 25 mm	44.0%	93.7%	64.7%	86.4%	6.98	0.60
Either fFN positive or CL ≤ 25 mm	47.4%	88.4%	52.9%	85.9%	4.09	0.60
Both fFN positive and CL ≤ 25 mm	10.5%	97.1%	50.0%	79.8%	3.60	0.92

PPV, positive predictive value; NPV, negative predictive value; +LR, positive likelihood ratio; -LR, negative likelihood ratio.

We found that the addition of fFN testing to CL measurement did not improve the prediction of preterm birth in this population. It is possible that this was only due to us being underpowered for this specific analysis or it suggests a different underlying biophysical mechanisms resulting in the process of spontaneous preterm birth in this population. Preterm parturition can be provoked through mechanical stretch of the uterus, thrombin generation of inflammatory mediators, ascending infection secondary to various cervical factors, fetal-maternal endocrine pathways, or maternal systemic inflammation/infection. It has been proposed that the association between uterine anomalies and preterm birth may be secondary to diminished muscle mass [22]. The utility of CL vs. fFN in predicting preterm birth in patients with uterine anomalies may imply the underlying mechanism does not disrupt the maternal fetal interface but results in cervical changes and uterine activity exclusively. Also, considering that only 6.7% of patients had a positive fFN from 22-32 weeks, it is unclear how useful the test would be clinically, even if it were found to be significantly associated with preterm birth.

Prior studies found that CL is associated with spontaneous preterm birth in patients with uterine anomalies, but they did not examine fFN testing as well [15, 16]. Airoidi et al reported on CL and preterm birth < 35 weeks in 64 patients with uterine anomalies, 10 (16%) of whom had a CL of 25 mm or less prior to 24 weeks. Patients with a short CL had a 50% risk of spontaneous preterm birth < 35 weeks, as opposed to 4% of patients with a normal CL. Similar to our population, the most frequent uterine anomalies in their cohort were bicornuate and septate uterus. Crane et al reported on CL and preterm birth in 52 patients with uterine anomalies, as well as 122 controls (unaffected). Based on ROC analysis, they selected 30 mm as the cutoff for a short CL and found that a short CL was associated with spontaneous preterm birth < 35 weeks in women with a bicornuate uterus, which was the most common uterine anomaly in their cohort. Our study of 120 patients confirms their findings in a larger cohort of women with uterine anomalies, and also includes data on fFN in this population.

Limitations to this study include all the limitations inherent to a retrospective study. However,

Cervical length and fetal fibronectin in uterine anomalies

our electronic database includes the data points examined, minimizing inaccuracies in the data. The fact that patients and physicians were not blinded to the CL and fFN results could potentially introduce bias. Not all patients had CL and fFN every 2 weeks, despite our protocol. This is mostly due to patient desire not to come for prenatal visits every two weeks. Therefore, it is possible that our results would differ somewhat if all patients had all of the testing every two weeks. It is unknown if our testing interval of every two weeks is ideal, and it is possible that more or less frequent testing would be either more clinically appropriate or cost effective. Also, it is unknown if our findings could be reproduced in other populations. For these reasons, more studies are warranted.

In summary, we found that in women with uterine anomalies, a short CL is significantly associated with spontaneous preterm birth <37 weeks and that the addition of fFN testing did not improve the prediction of preterm birth in this cohort.

Disclosure of conflict of interest

None.

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Cervical length and fetal fibronectin in uterine anomalies

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