J. Egypt. Soc. Parasitol. (JESP), 44(2), 2014: 525 - 530

PREDICTIVE VALUE OF PHOSPHORYLATED INSULIN–LIKE GROWTH FACTOR BINDING PROTEIN-1(PIGFBP-1) (BEDSIDE TEST) IN PRETERM LABOR

By

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Abstract

This study evaluated the predictive value of phosphorylated insulin like growth factor binding protein-1 in cervical secretion as bedside test for prediction of preterm labor in symptomatic women.

A total of 57 patients with singleton pregnancy at 24-34 weeks gestation, with symptoms suggestive of preterm labor were included in this study. A rapid cervical sample for PIGFBP-1 determination (Actim partus test, Medix Biochemical, and Kaunianen, Finland) was taken by means of a polyester-tipped swab during a speculum examination of the cervix, and extracted with specimen- extraction solution. We analyzed the prevalence of preterm labor in these patients within seven days upon admission. And calculate sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) for PIGFBP-1. This study was done at Kuwait Oil Company hospital (KOC) and Taiba hospital (Kuwait) during the period between April 2011 and June 2012.

The test was positive in 50.9% of patients and negative in 49.1%, among those tests was positive 64% delivered less than a week, and among those tests was negative 35.7 delivered less than one week, with 74.3% sensitivity, 61% spesivity, 76.3% PPV and 73.6% NPV.

Key Words: phosphorylated insulin –like growth factor binding protein, bedside test, preterm labor.

Introduction

Spontaneous preterm birth (SPTB),defined as non indicated birth before 37 weeks' estimated gestational age, remains a major source of neonatal morbidity and mortality despite continuing research efforts (Alexander *et al*, 2003). It accounts for 60% of all perinatal deaths (Martin *et al*, 2008).

Quantitative fetal fibronectin (qfFN) testing has demonstrated accuracy at predicting sPTB in high-risk asymptomatic women (previous preterm birth or late miscarriage). Prediction of sPTB using qfFN in women with cervical surgery proved useful (Calvert *et al*, 2014).

Most cases of SPTB are believed to arise from the 1 or a combination of 4 classic pathways: infection/inflammation, excessive myometrial mechanical stretch (i.e., multifetal gestation), fetal or maternal stress, and chorion-decidual hemorrhage (Cheong and Tay, 2014).

In order to institute specific therapy more appropriately, it is important to have adjuvant tests to help predict who is most likely to deliver preterm. There are few methods available to reliably predict true preterm labor in women who present with symptoms of labor. Currently, transcervical ultrasound measurements and/or cervicovaginal fetal fibronectin levels are the most commonly used diagnostic tools (Gomez *et al*, 2005). These techniques both have high negative predictive values but relatively although some confusion may occur when differentiating between endometrial polyps and submucosal myoma. Postprocedural complications were mostly attributed to vigorous dilation of the cervix (Chang, 2007). Consequently, many women and their fetuses are exposed unnecessarily to tocolytic drugs and corticosteroids and are admitted to hospital (Warren *et al*, 1992).

Several other biochemical markers have been associated with an increased risk of

preterm delivery including activin, inhibin, and relaxin (Plevyak *et al*, 2003).These glycoprotein's are produced in the placenta and decidua and seem to facilitate the labor process through varying mechanisms: the former two through the release of local prostaglandins, whereas the latter seems to be involved in collagen remodeling within the amnion and chorion (Petersen *et al*, 1992). Although promising as potential markers, Plevyak *et al.* (2003) noted a significant effect with activin and inhibin levels only at 31 to 34 weeks' gestation.

The detection of phosphorylated insulin like growth factor binding protein-1 (pIGFBP-1) in the cervical secretions of women presenting with preterm labor has been shown to be associated with an increased risk of preterm delivery (Lembet *et al,* 2002). IGFBP-1 belongs to the super family of insulin like binding proteins. There are 6 subtypes of IGFBP distributed widely throughout the body. Highly phosphorylated form (PIGFBP-1) is found in decidual tissues. The process of labor is hypothesized to disturpt the chorio-decidual interfaces, releasing PIGFBP-1into cervical secretions (Kekki *et al,* 2001).

A bedside test-kit for PIGFBP-1 has been developed and is commercially available under the trade name Actim Partus (Medix Biochemical, Finland). It is an immunoenzymatic test relying on the monoclonal antibody 6303 as the detecting antibody. This antibody is specific for the phosphorylated form of IGFBP-1 and above the level of 10ug/L, which is the form found in high concentrations in the chorio-decidual interface (Kwek *et al,* 2004).

Patients, Materials and Methods

This prospective study was performed on 57 women with symptoms of preterm labor during the period between April 2011 and June2012 in departments of gynecology and obstetrics in Kuwait Oil Company (KOC) hospital and Taibahospital (Kuwait). An informed consent was obtained from all patients. The inclusion criteria were a singleton pregnancy at 24-34 weeks' gestation, with symptoms suggestive of preterm labor (pressure like symptoms (pelvic pressure), backache, and vaginal discharge, documented uterine contractions (at least 8 per hour, 30 seconds in duration and confirmed by external tocodynamometry), intact membranes and cervical dilatation of 0-3 cm (nulliparous women) or 1-3 cm (primiparous or multiparous) and 50% effacement. The exclusion criteria were >3cm dilatation of the cervix, having undergone a cervical examination or sexual intercourse less than 24h previously, vaginal bleeding, placenta previa, multiple gestations, fetal abnormalities and uterine anomalies, preterm rupture of membranes, cervical circlage, intrauterine growth restriction, choriamnionitis.

Gestational age was based on menstrual date confirmed by first or an early second-trimester ultrasound scan. When a discrepancy of more than 5 days occurred, gestational age was set only by first trimester ultrasonographic examination. The other recorded data included demographic information (age, parity, gravidity, previous preterm delivery) pre-term labor management and delivery outcomes. All women were admitted to hospital and received tocolytics (Oxytocine antagonist (atosiban) or calcium channel blockers). Fetal lung maturation was accelerated by administering 24 mg of betamethasone (12 mg intramuscularly every12 hr. per a day)

A rapid cervical sample for PIGFBP-1 determination (Actim Partus Test, Medix Biochemical, and Kauniainen, Finland) was taken by means of a polyester-tipped swab during a speculum examination of the cervix, and extracted with specimen- extraction solution. The lower end of the swab was inserted into the external cervical orifice and left in place for about 10 s, after which it was placed in a test tube containing 0.5 mL of buffer solution. The dipstick was dipped into the samples and left for 15 s to allow the liquid front to enter the results area. After removing the dipstick from the solution

and holding it for 5 min in a horizontal position, the test was interpreted as being positive, negative or invalid, respectively, when two, one or no blue lines appeared in the result area. The test is based on immunochromatography and has a detection limit of 10Ug/L.No changes were made in the treatment of the positive or negative patients. After delivery, data collection on gestational age at delivery, admission to delivery interval, mode of delivery, indication of delivery and baby status was completed. The primary outcome measure was delivery before 35 weeks gestation. The secondary outcome measure was delivery within 7 days following inclusion.

Statistical analysis: Student's t test was used to compare the average values of the continuous variables, with the values being expressed as the mean ± SD. Pearson's Chi-square test or Fisher's exact test was used to analyze the categorical variables, whose values are expressed as percentages. Univariate logistic regression analysis was performed to assess the capability of the PIGFBP-1 test to predict pre-term birth (Tab. 1).

Results

In the present study, the sensitivity, eighty symptomatic women with singleton Pregnancies were recruited into the study. Fourteen patients had to be excluded from the final analysis due to incomplete data. Five patients delivered in another hospital and thus delivery data was not available, 1 were found to be less than 24 weeks on admission, 2 had preterm prelabour ruptured of membranes, and 1 had cervical dilatation >3cm at admission. Fifty seven patients had complete data for analysis. The average maternal age of the patients was 27.40±6.1; the mean gestational age at sampling was 29.70±2.5 gestational weeks, and the mean parity was 2.91.

The demographics and clinical characteristics populations, without significant differences were observed concerning maternal, gestational age at admission and parity (Tab. 2), 29 (50.9%) tested positive for PIGFBP-1 while 28 (49.1%) tested negative for PIGFP-1. We analyze the prevalence of preterm delivery in these patients within seven days upon admission. Among those with negative PIGFBP-1, ten of them (35.7%) delivered less than one week after a test became negative and 18 of them (64% delivered after one week of a test became negative, among those with positive PIGFBP-1. 20 of them (68%) delivered less than one week after a test became positive and 9 of them (31%) delivered after one week of a test became positive (Tab. 3).

Significantly greater proportion of women who tested positive delivered less than one week in compare to those tested negative (68%VS 35.7%) also significant greater proportion of women who test negative remain undelivered after one week in compare to those test positive (64% Vs 31%).the result show that bed side test for PIGFBP-1has 74.3% sensitivity, 61% specificity, 76.3 positive predictive value and 73.6% negative predictive value for the prediction of preterm labor. The results suggested that this test may have an important role in the management of women presenting with symptoms suggestive of preterm labor. And it is consider as effective adjuvant bedside test in diagnosis of preterm labor.

Discussion

An accurate diagnosis of preterm labor is clinically difficult. Only about 20% of women presenting with signs and symptoms of preterm labor would actually deliver preterm. Various tools have been devised for the identification of women at risk of preterm delivery. These include risk scoring systems, biochemical markers of inflammation, and fetal fibronectin (**Honest *et al*, 2002**). These aim to decrease the unnecessary interventions for patients with symptoms of preterm labor and to identify patients who might benefit from aggressive therapy including tocolysis, corticosteroids, and intra-uterine transfer to a tertiary care facility. As such, it is important that we are

able to distinguish between inconsequential abdominal pain or uterine activity and true preterm labor with associated morbidity and mortality of preterm delivery, PIGFBP-1 belongs to the super family of insulin-like binding proteins and the phosphorylation status of IGFBP-1 in decidual tissues is different from that in amniotic fluid. Thus, the highly phosphorylated form (PIGFBP-1) was found in the decidual tissues and was previously designated as placental protein 1.

Table 1: Statistical analysis.

Items	Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	6.318[a]	1	.012		
Continuity Correction	5.054	1	.025		
Likelihood Ratio	6.439	1	.011		
Fisher's Exact Test				.017	.012
Linear-by-Linear Association	6.207	1	.013		
N of Valid Cases	57				

a=0 cells (.0%) have expected count less than 5.The minimum expected count is13.26. b=computed only for a 2x2 table

Table 2: demographic and clinical characteristics of populations (No significant differences)

Test			Delivery				
Valid	frequency	percent		Negative	positive	Frequency	Percent
negative	28	49.1	<1 week	10	20	30	52.6
positive	29	50.9	> 1 week	18	9	27	47.4
total	57	100	Total	28	29	57	100

Table 3: test-delivery time interval

Age	Parity	Ges. age	Test		Delivery	
			negative	positive	<1 week	> 1 week
Mean	Mean	Mean	Count	Count	Count	Count
27.40	2.91	29.79	28	29	30	27

The hypothesis is that labor disturb chorio-decidual interface, releasing PIGFBP-1 into the cervical secretions. Table (3) shows prevalence of preterm delivery in patients within seven days upon admission with significantly greater proportion of women who tested positive delivered less than one week in compare to those tested negative.

The identification of PIGFP-1 would thus be indicative of the occurrence of the labor process. Kekki *et al*. (2001) showed that women with aPIGFBP-1 concentration of at least 10ug/L in a cervical swab sample had a 10 fold risk of preterm delivery compared with women in whom the concentration of PIGFP-1 was less than 10ug/L. Lembet ***et al.*** **(2002)** carried out a prospective study on 36 women between 20 and 36 weeks of gestation with regular contractions. Eighteen patients had a positive Actim Partus test and 18 had a negative test. Among the 18 patients with a positive test, only 1 delivered at term and the other 17 patients delivered preterm (<37 weeks). Among the 18 women with a negative test, only 2 delivered preterm (P <0.05). Sensitivity, specificity, PPV and NPV of the rapid pIGFBP-1 test for preterm delivery were 89.5%, 94.1%, 94.4% and 88.9%, respectively.

In the present study, specificity, PPV & NPV for PIGFP-1 bedside test in prediction of preterm labor were in accordance with Bittar (**1999)** and Kwke *et al*. (2004) they showed that in their study reliability of PIGFP-1 in prediction of preterm labor with sensitivity 78% and PPV 66%.also our results are in accordance with Elizur, et al, they found sensitivity and specificity of PIGFP-1 in prediction of preterm labor 80 % and 77% respectively.

Adeyemi and Osoba (2010) evaluated the role of phosphorylated insulin-like growth factor binding protein-1 (IGFBP-1) test in the prediction of pre-term delivery in twin pregnancies. All the patients had a transvaginal ultrasound scan for cervical length at 24

weeks, followed by a high vaginal swab for IGFBP-1 at 26 weeks. A total of 95% of women screened negative for the IGFBP-1 test. None of these women delivered before 30 weeks; 7.50% delivered between 30 and 33+6 weeks; 87.5% delivered after 34 weeks. Two women (5.00%) screened positive for phosphorylated insulin-like growth factor binding protein-1; one had a spontaneous pre-term delivery at 30 weeks, while the other patient delivered at 38 weeks. They stated that women with twin pregnancies that have negative phosphorylated insulin-like growth factor binding protein-1 have a low risk of delivery before 34 weeks in the absence of other obstetric complications.

Brik *et al*. (2011) assessed the efficacy of cervical interleukin-6 (IL-6) compared with cervical length and its association in the prediction of preterm delivery in symptomatic women. They found that the cervical IL-6 can predict preterm delivery similarly to cervical length; when combined, it adds prognostic information to that provided by sonographic measurement of the cervical length. Riboni *et al*. (2011) evaluated the efficacy of the phosphorylated insulin-like growth factor-binding protein (phIGFBP-1) and of the fetal fibronectin test (fFN) in predicting pre-term delivery in symptomatic women. They concluded that the phIGFBP-1 test may be better that the fFN test in predicting pre-term delivery before 34 weeks' gestation.

Kallioniemi *et al*. (2013) studied vaginal fluid phosphorylated insulin-like growth factor binding protein-1 (phIGFBP-1) testing in early pregnancy to predict spontaneous preterm delivery (PTD). They found that first-trimester vaginal phIGFBP-1 performed better than cervical phIGFBP-1 in the prediction of spontaneous PTD. Rolnik *et al*. (2013) investigated the usefulness of the measurement of cervical length and of the test for phosphorylated insulin-like growth factor binding protein-1 (phIGFBP-1) performed sequentially in the prediction of preterm birth and the correlation, They concluded that both cervical length and the test for phIGFBP-1 were able to predict premature delivery, and sequential combination of both tests showed a high sensitivity and high negative predictive value.

Brik *et al*. (2014) evaluated the use of cervical interleukin 6 (IL-6) and phosphorylated insulin growth binding protein 1 (pIGFBP1) in the prediction of adverse neonatal outcome. They reported that the logistic regression analysis showed cervical IL-6 and examination-to-delivery interval as predictors of NCM in the univariate analysis, but the only independent marker of adverse neonatal outcome was the examination-to-delivery interval. and concluded that the adverse neonatal outcome is associated with increased cervical IL-6 concentrations.

Conclusion

The prediction and prevention of preterm delivery remain great challenge. To examine the presence of insulin-like growth factor binding protein 1 (IGFBP-1) in cervicovaginal secretion of pregnant women with symptoms of preterm labor is a must.

The outcome data showed that the detection of PIGFBP-1 in cervical secretion can be used as a predictor and adjuvant test in prediction of preterm labor in symptomatic women

Acknowledgement

The authors would like to express their appreciation and indebtedness to Prof. Dr. Ibrahim Anwar Abdel-Azim for his effort, continuous advice and revise this manuscript.

Conflict of interest: No actual or potential conflict of interest is related to this manuscript.

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