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Predictive Value of Maternal Serum Level of Procalcitonin in Diagnosing Chorioamnionitis in Mothers with Preterm Premature Rupture of Membrane (PROM)

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ABSTRACT

Background: Premature rupture of membrane (PROM) is the rupture of fetal membranes at least 1 hour before the onset of labor pain. In this study, we sought to determine the predictive value of maternal serum level of procalcitonin (PCT) in the early diagnosis of chorioamnionitis in women with preterm PROM (PPROM). **Methods:** In this prospective cohort study, owing to limited financial resources, 48 patients with PPROM in the Kosar Ward of Motahari Hospital in Urmia, Iran were selected to comprise the sample. Inclusion criteria were amniotic fluid leak, positive ferning and nitrazine tests, gestational age 28-33 weeks, and lack of fetal tachycardia. Exclusion criteria were chronic and congenital heart disease and use of non-steroidal anti-inflammatory drugs. Data were analyzed using SPSS v. 19 software; descriptive statistics, independent t-tests, and the Pearson test were also performed. **Results:** We studied 48 pregnant women and their neonates. Approximately 40% of women had chorioamnionitis; approximately 60% of women did not have chorioamnionitis. Moreover, approximately 69% of the neonates had a 5-minute Apgar score ≥7. A significant correlation was observed between women who had histopathology confirmed chorioamnionitis and neonate hospitalization in the neonatal intensive care unit (P<0.001). The sensitivity, specificity, and positive and negative predictive values of PCT inflammatory indices were 100%, 79%, 57.5%, and 100%, respectively, for the histopathologic diagnosis of chorioamnionitis. **Conclusion:** In pregnant women, we found a significant correlation between PCT index at delivery and histopathologic chorioamnionitis.

INTRODUCTION

Premature rupture of membrane (PROM) is the rupture of fetal membranes at least 1 hour before the onset of labor pain; it occurs in approximately 10% of pregnancies. A third of PROM cases occur before week 37 of pregnancy and are referred to as preterm PROM (PPROM) (1). In a study of PROM in Vali-e-Asr Hospital in Tehran, Iran, Nili and Ansari reported an incidence rate of 7% (2). PPROM is divided into three groups: 1) non-viable fetus (before week 23 of pregnancy); 2) rupture of membranes from fetal viability to week 31 of pregnancy; and 3) from week 32 to week 36 of pregnancy (3). The cause of PROM has not yet been determined (4); however, important risk factors for PROM include gestational age, low maternal body mass index (<19.8 kg/m^2), low social and economic status of the mother, history of cervical conization, smoking, genital infections, excessive elongation of the membranes (due to polyhydramnios or multiple pregnancies), history of PPROM, nutritional factors (deficiency of copper and ascorbic acid), cervical incompetence, cervical cerclage, and placental detachment (second and third trimester bleeding) (5). The complications of PROM before the onset of labor pain include maternal infections (e.g., endometriosis or sepsis), chorioamnionitis (9% in term PROM and 13-60% in preterm PROM), placental detachment, fetal infections, neonatal death, low Apgar score, umbilical cord compression and prolapse, pulmonary hypoplasia, and low birth weight (6, 7). Infections causing PROM include chlamydia trachomatis, trichomonas vaginalis, Neisseria gonorrhea, and Group-B streptococcus (5). The mean duration of hospitalization of PROM and PPROM neonates increases by 20% and 25%, respectively, compared to other neonates, and their imposed costs on the health system increase by 31% and 60%, respectively (8). Chorioamnionitis is the infectious inflammation of the uterine cavity, fetal membranes (amnion and chorion), and placenta, with certain maternal and fetal complications (9, 10). In cases of chorioamnionitis, fever ≥38˚C (≥100.4˚F) and ruptured membranes are the only reliable indicators of the disease; nevertheless, it is necessary to monitor uterine tenderness, permanent maternal or fetal tachycardia, and smelly vaginal discharge (11). However, a definitive diagnosis is reached only after microscopic observations (12). Procalcitonin (PCT) is a 116-amino acid-containing polypeptide precursor of the hormone

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calcitonin that is processed to calcitonin and one 32-amino acid polypeptide via endopeptidase enzymes (13). The serum level of PCT increases within 2-6 hours after sepsis but decreases in the case of infection control (14, 15). Since chorioamnionitis is a bacterial infection that causes sepsis and since serum PCT increases in bacterial infections, PCT increase appears to be effective in the early diagnosis of chorioamnionitis, early termination of pregnancy, and fetal therapy. One study found that the sensitivity and specificity of the maternal serum level of PCT were 50% and 56%, respectively, for the histopathologic diagnosis of chorioamnionitis (16).

Because of the limited number of studies and the significant effects that early diagnosis of chorioamnionitis can have in reducing maternal and neonatal complications, we sought in this study, which was conducted in 2016, to determine the predictive value of PCT in the early diagnosis of chorioamnionitis in pregnant women with PPROM. Our purpose was to increase the knowledge in this field and reduce maternal and fetal complications of chorioamnionitis by proving the applicability of PCT in treatment interventions and patient management.

METHODS

Our study included pregnant women with PPROM who underwent conventional therapy in the Kosar Ward of Motahari Hospital in Urmia, Iran. This prospective cohort study was done during 18 months. In this prospective cohort study, owing to limited financial resources and in accordance with previous studies of patients treated in the same ward, 48 patients with PPROM were selected to comprise the sample (2, 4, 8).

Pertinent medical information was obtained from the completion of a questionnaire, from lab test results, and from histologic examinations. Demographic information was obtained from self-report dossiers, and laboratory variables were obtained from laboratory reports. The included patients were pregnant women who were hospitalized in the midwifery ward of Shahid Motahari Hospital because of rupture of membranes at the gestational age of 28-33 weeks.

Upon hospital admission, patients underwent blood sampling for screening PCT. Afterwards, they underwent treatment in the hospital. Immediately after delivery (vaginal or cesarean section), a blood sample was retaken to screen for PCT. Then, the placentas of all patients were transferred to the pathology laboratory for histologic examination for chorioamnionitis.

In the pathology laboratory, the stages of sample preparation were as follows:

1. After isolating the samples fixed in formalin, tissue-processing steps included: 1) additional fixation in formalin solution; 2) dehydration in 75-, 85-, and 96-degree alcohol and absolute alcohol; 3) clearing in xylene solution; and 4) saturation in molten paraffin via a tissue processor.
2. Sample molding by paraffin and paraffin blocks.
3. Cutting the blocks to a thickness of 4 microns by a micro-thermometer and transferring the tissues.
4. Paraffinizing the isolated tissues in a dry heat or oven.
5. Hematoxylin and eosin staining as follows:
   - 5.1. Hydrating tissues in 96-, 85-, and 75-degree alcohol
   - 5.2. Washing with water
   - 5.3. Staining with hematoxylin
   - 5.4. Washing with water
   - 5.5. De-staining (decoloring) in alcoholic acid
   - 5.6. Washing with water
   - 5.7. Alkalizing tissues by bicarbonates
   - 5.8. Washing with water
   - 5.9. Staining with eosin
   - 5.10. Washing with water
   - 5.11. Dehydrating with reverse alcohol
   - 5.12. Clearing in xylene solution
   - 5.13. Laminating with Entellan Merck

Notably, chorioamnionitis is pathologically characterized as a leukocyte infiltration with neutrophilic priority and inflammatory infiltration on the fetal membranes (16).

Finally, the patients were divided into three groups:

1. Pregnant women with clinical symptoms of chorioamnionitis
2. Pregnant women with histopathologic chorioamnionitis
3. Pregnant women with none of the above symptoms (clinical or histologic chorioamnionitis)

Patients underwent clinical examination for fever and fetal tachycardia using non-skid tape 24 hours before the onset of delivery. The antibiotic treatment was used to prevent neonatal sepsis in the patients under observation.

Next, the inflammatory indices were compared in each group. Serum levels of PCT higher than 0.06 ng/ml were considered an increased amount. This amount was selected because of the use of different cutoff points in the reviewed articles, the population proximity of the neighboring country, Turkey, to Iran, and the novelty of our study (8, 17). A cobas e411 analyzer and a Roche diagnostic kit (Lot 187-424-02) were used to measure the level of PCT.

A two-way blindness process was used between the histology group and the laboratory responsible for monitoring the maternal serum level of PCT. To evaluate the value of PCT in diagnosing chorioamnionitis, PCT levels were compared with clinical symptoms, histologic results, specificity index, sensitivity index, positive and negative predictive values, and Youden's index. Inclusion criteria included proof of amniotic fluid leak via speculum examination, positive nitrazine and ferning test, gestational age of 28-33 weeks that was approved to be less than 20 weeks us ultrasonography, lack of fever, uterine contractions, abnormal vaginal discharge on the first day of hospitalization, absence of vaginal bleeding, and lack of fetal tachycardia. Exclusion criteria included congenital heart disease, heart valve surgery, chronic vascular disease (nephritic syndrome or lupus), use of corticosteroids, or use of nonsteroidal anti-inflammatory drugs.

This study was approved by the Ethics Committee of Urmia University of Medical Sciences. All patients were informed about the research process. Researchers did not

intervene in the conventional process, management, or treatment of patients.

Data were analyzed using SPSS v. 19 software; descriptive statistics, independent t-tests, and the Pearson test were also performed. *P* values less than 0.05 were considered statistically significant.

RESULTS

In this study of 48 women with PPROM, we sought to determine the predictive value of maternal serum level of PCT in diagnosing chorioamnionitis. Patient demographic characteristics are shown in Table 1.

Twenty-three patients (47.9%) tested positive for PPROM based on clear amniotic fluid leak, and 25 patients (52.1%) tested positive for PPROM based on the nitrazine and ferning test. The gestational age of nine patients (18.8%) had valid last menstrual period (LMP), 34 (50%) was before week 12 based on ultrasonography, and 15 (39.9%) was before week 20 based on ultrasonography. In 48 patients, the minimum gestational age at PROM was 28 weeks and 5 days; the maximum gestational age at PROM was 33 weeks. The mean time of PPROM was 31 weeks and 6 days (± 1 week and 2 days). Moreover, the minimum gestational age at delivery was 29 weeks and 5 days; the maximum gestational age at delivery was 34 weeks. The mean delivery time was 33 weeks and 2 days (± 1 week). In terms of 5-minute Apgar score, three neonates (6.2%) scored 4, seven (14.6%) scored 5, five (10.4%) scored 6, and 33 (68.8%) scored 7. The minimum birth weight was 1200 g; the maximum birth weight was 2200 g. The mean birth weight was 1927.1 g ± 232.31 g. In terms of amniotic fluid index in the last ultrasonography before delivery, the minimum and maximum indices were 0 mm and 38 mm, respectively. The mean fluid index was 19.42 mm ± 9.91 mm.

As shown in Table 2, in terms of PCT inflammatory index upon hospitalization, the minimum and maximum values were 0.02 and 0.1, respectively, and the mean index was 0.041 ± 0.017. The minimum and maximum PCT indices upon delivery were 0.95 and 0.03, respectively, and the mean index was 0.104 ± 0.175. No statistically significant correlation was observed between PCT level upon hospitalization and delivery time (R^2=0.04 and P<0.176).

As shown in Table 3, all women with a PCT index ≥0.06 upon hospitalization had chorioamnionitis. Moreover, of the women with a negative PCT index upon hospitalization, 17 (37%) had chorioamnionitis and 29 (63%) did not have chorioamnionitis. According to the chi-squared test and the Pearson correlation test, no significant correlation was observed between PCT index upon hospitalization and infection with chorioamnionitis (P=0.077).

As shown in Table 4, of the women with a PCT index ≥0.06 upon delivery, 16 (66.7%) had chorioamnionitis and eight (33.3%) did not have chorioamnionitis. Moreover, of the women with a negative PCT index upon delivery, three (12.5%) had chorioamnionitis and 21 (87.5%) did not have chorioamnionitis. According to the chi-squared test and the Pearson correlation test, a significant correlation was observed between PCT index upon delivery and infection with chorioamnionitis (P=0.0). The odds ratio (OR) of chorioamnionitis in women with increased PCT index upon delivery was infinite (not defined) (OR=∞).

Table 1. Demographic characteristics of the pregnant women

Variable	Frequency	Percentage
G≤2	33	68.7
G≥3	15	31.3
PPROM	6	12.5
Abortion	6	12.5

G: Gestational age, PPROM: Preterm Premature Rupture of Membrane

DISCUSSION

Histologic chorioamnionitis (i.e. infection of fetal membranes and amniotic fluid) is commonly observed in pregnant women with PPROM and may cause neonatal sepsis, preterm birth, pulmonary diseases, and neonatal brain injury. Therefore, early diagnosis of chorioamnionitis is very important (18, 19). PCT is a peptide precursor of the calcitonin hormone, which is typically produced by monocytes and hepatocytes (20). The serum level of PCT rapidly increases in response to bacterial endotoxin within 3-4 hours, peaks within 18-24 hours, and remains at its elevated serum level within 24-48 hours (20).

We found a significant correlation between maternal infection with histologic chorioamnionitis and the inflammatory index of PCT upon delivery. However, we did not find a significant correlation between maternal infections and chorioamnionitis.

As reported in a clinical study, maternal serum level of PCT upon hospitalization of women with PPROM (up to 4 hours after rupture of membrane) was not a reliable index to diagnose histologic chorioamnionitis, fetal infection, time

Table 2. Minimum and maximum inflammatory indices upon hospitalization and delivery

Variable	No.	Minimum	Maximum	Mean	SD
PCT upon hospitalization	48	0.02	0.10	0.041	0.017
PCT upon delivery	48	0.03	0.95	0.104	0.175
Last measured index of amniotic fluid	48	0	38	19.42	9.91
Days of hospitalization until delivery	48	2	30	10.29	5.52

PCT: Procalcitonin, SD: Standard deviation

interval between PROM and delivery, or intrauterine infection. However, the maternal serum level of PCT in women with PPROM was higher than the maternal serum level of PCT in women without PPROM (21). A prospective study found that the increased maternal serum level of C-reactive protein (CRP) and PCT upon delivery significantly correlated with histologic chorioamnionitis regardless of membrane status (22). Oludaq et al. showed that the maternal serum level of PCT in women with PPROM who also had histologic chorioamnionitis was significantly higher than the maternal serum level of PCT in women with PPROM who did not have chorioamnionitis. Moreover, the sensitivity and specificity of PCT were 92.3% and 68.4%, respectively, at 0.054 ng/ml serum level for the diagnosis of chorioamnionitis (17).

In our study, the causes of neonatal death included pulmonary hemorrhage, acute respiratory distress syndrome (ARDS), and cerebral hemorrhage. An observational study found that cognitive disorders, neuro-developmental disorders, and mortality were significantly higher in neonates with a gestational age of ≤27 weeks who were born to mothers with chorioamnionitis than in other neonates (23). As reported in a retrospective study, prenatal corticosteroid administration was associated with a significant decrease in neonatal mortality, ARDS, neonatal seizure, and cerebral hemorrhage in neonates born to mothers with histologic chorioamnionitis (24). Another retrospective study showed that prenatal cortisone administration reduced the mortality rate before the age of 3 years in children of mothers who had histologic chorioamnionitis, as compared to children of non-infected mothers; however, prenatal cortisone administration did not have any effect on the neuro-developmental rate of children born to mothers in both infected and non-infected groups. Therefore, corticosteroids are recommended to mothers with chorioamnionitis (25).

A retrospective study of neonates with a low birth weight of ≤1500 g born to mothers with chorioamnionitis found that histologic chorioamnionitis was associated with an increased risk of chronic pulmonary disease and neonatal sepsis; however, histologic chorioamnionitis did not correlate with cerebroventricular hemorrhage, periventricular leukomalacia, cerebral palsy, vision impairment, or neonatal death before discharge (26, 27).

Recently, a novel antibiotic regimen based on ceftriaxone, clarithromycin, and metronidazole has been used for the treatment of chorioamnionitis; this regimen has resulted in less maternal infection with histologic chorioamnionitis as compared to the ampicillin regimen with or without cephalosporin. Accordingly, the rates of cerebral palsy and ventricular hemorrhage were significantly lower in neonates treated with the new regimen than in neonates treated with the ampicillin (28, 29).

Limitations of our study included the high costs and limited financial resources to measure the serial serum level of PCT and to compare the intended patients with healthy non-PPROM pregnant women.

Table 3. Mean±standard deviation and frequency of inflammatory indices with respect to women with chorioamnionitis

Variable	Chorioamnionitis		Total
	Yes	No	
PCT>0.06 upon hospitalization			
Positive			
Frequency	2	0	2
Percent	100	0	100
Negative			
Frequency	17	29	46
Percent	37	63	100
Total			
Frequency	19	29	48
Percent	39.6	60.4	100

PCT: Procalcitonin

CONCLUSION

We found a significant correlation between PCT index at delivery time and histopathologic chorioamnionitis. We also found a significant correlation between the inflammatory indices of CRP during the delivery time and histopathologic chorioamnionitis.

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AUTHOR CONTRIBUTIONS

All authors contributed equally to this study.

Table 4. Mean and standard deviation (SD) of inflammatory indices with respect to women with chorioamnionitis

Variable	Chorioamnionitis	No.	Mean	SD
PCT>0.06 upon hospitalization	Positive	19	0.045	0.021
	Negative	29	0.038	0.012
PCT>0.06 upon delivery	Positive	19	0.18	0.265
	Negative	29	0.056	0.018

PCT: Procalcitonin, SD: Standard deviation

REFERENCES

1. Svigos JM, Dodd JM, Robinson JS. Prelabor rupture of the membranes. Chapter 62.P 1091 In James, Steer, Weiner, Gonik, Crowter, Robson, High risk pregnancy, 2011
2. Nili F, Shams Ansari AA. Neonatal complications of premature rupture of Membrane.ActaMedicaIranica.2003; 41:175–179.
3. Kavak SB, Kavak E, Ilhan R, Atilgan R, Arat O, Deveci U, et al. The efficacy of ampicillin and Lactobacillus caseirhamnosus in the active management of preterm premature rupture of membranes remote from term.Drug Des DevelTher. 2014 Aug 30; 8:1169-73.
4. Cunningham FG, Leveno KJ, Bloom SL, Spong CY, Dashe JS, Hoffman BL, et a. preterm labor, chapter 42, section 11, Williams Obstetrics,2014, McGraw-Hill, P 836.
5. Mercer BM. Premature rupture of the membranes. Chapter 42, P 663In Creasy RK, Resnik R, Maternal-Fetal-Medicine, 2011.
6. Mercer BM. Premature rupture of the membranes. Chapter 42, P 664 in Creasy RK, Resnik R, Maternal-Fetal-Medicine, 2011.
7. Nakubulwa S, Kaye DK, Bwanga F, Tumwesigye NM, Mirembe FM.Genital infections and risk of premature rupture of membranes in Mulago Hospital, Uganda: a case control study.BMC Res Notes. 2015 Oct 16; 8:573.
8. Kariman N, Hedayati M, AlaviMajd Sh.The diagnostic power of cervico-vaginal fluid prolactin in the diagnosis of premature rupture of membranes.Iran Red Crescent Med J. 2012 Sep; 14(9):541-8.
9. Szukiewicz D, Kochanowski J, Mittal TK, Pyzlak M, Szewczyk G, Cendrowski K.Chorioamnionitis (ChA) modifies CX3CL1 (fractalkine) production by human amniotic epithelial cells (HAEC) under normoxic and hypoxic conditions.J Inflamm (Lond). 2014 May 13; 11:12.
10. Erdemir G, Kultursay N, Calkavur S, Zekioğlu O, Koroglu OA, Cakmak B, et al.Histological chorioamnionitis: effects on premature delivery and neonatal prognosis. PediatrNeonatol. 2013 Aug; 54(4):267-74.
11. Cunningham FG, Leveno KJ, Bloom SL, Spong CY, Dashe JS, Hoffman BL, et a. preterm labor, chapter 42, section 11, Williams Obstetrics,2014, McGraw-Hill, P 848.
12. Xie A, Zhang W, Chen M, Wang Y, Wang Y, Zhou Q, et al. Related factors and adverse neonatal outcomes in women with preterm premature rupture of membranes complicated by histologic chorioamnionitis.Med Sci Monit. 2015 Feb 3; 21:390-5.
13. Yousefimanesh H, Robati M, Malekzadeh H, Jahangirnezhad M, GhafourianBoroujerdnia M, Azadi K. Investigation of The Association between Salivary Procalcitonin Concentration and Chronic Periodontitis.Cell J. 2015 Fall; 17(3):559-63.
14. Pieralli F, Vannucchi V, Mancini A, Antonielli E, Luise F, Sammicheli L, et al.Procalcitonin Kinetics in the First 72 Hours Predicts 30-Day Mortality in Severely Ill Septic Patients Admitted to an Intermediate Care Unit.J Clin Med Res. 2015 Sep; 7(9):706-13.
15. Pantelidou IM, Giamarellos-Bourboulis EJ.Can procalcitonin monitoring reduce the length of antibiotic treatment in bloodstream infections?Int J Antimicrob Agents. 2015 Dec; 46Suppl 1:S10-2.
16. Thornburg LL, Queenan R, Brandt-Griffith B, Pressman EK.Procalcitonin for prediction of chorioamnionitis in preterm premature rupture of membranes.J Matern Fetal Neonatal Med. 2015 Sep 3:1-6.
17. Oludag T, Gode F, Caglayan E, Saatli B, Okyay RE, Altunyurt S.Value of maternal procalcitonin levels for predicting subclinical intra-amniotic infection in preterm premature rupture of membranes.J ObstetGynaecol Res. 2014 Apr; 40(4):954-60.
18. Kim SA, Park KH, Lee SM. Non-Invasive Prediction of Histologic Chorioamnionitis in Women with Preterm Premature Rupture of Membranes. Yonsei Med J. 2016 Mar; 57(2):461-8.
19. Ocheke AN, Agaba PA, Imade GE, Silas OA, Ajetunmobi OI, Echejoh G, et al. Chorioamnionitis in pregnancy: a comparative study of HIV-positive and HIV-negative parturients. Int J STD AIDS. 2016 Mar; 27(4):296-304.
20. Altunhan H, Annagür A, Örs R, Mehmetoğlu I. Procalcitonin measurement at 24 hours of age may be helpful in the prompt diagnosis of early-onset neonatal sepsis. Int J Infect Dis. 2011 Dec; 15(12):e854-8.
21. Torbé A. Maternal plasma procalcitonin concentrations in pregnancy complicated by preterm premature rupture of membranes. Mediators Inflamm. 2007; 2007:35782.
22. Greksova K, Parrak V, Chovancova D, Stencl P, Oravec J, Marsik L, et al. Procalcitonin, neopterin and C-reactive protein in diagnostics of intrauterine infection and preterm delivery. Bratisl Lek Listy. 2009; 110(10):623-6.
23. Pappas A, Kendrick DE, Shankaran S, Stoll BJ, Bell EF, Laptook AR, et al. Chorioamnionitis and early childhood outcomes among extremely low-gestational-age neonates. JAMA Pediatr. 2014 Feb; 168(2):137-47.
24. Karimi-Zarchi M, Mortazavizadeh MR, Soltani-Gerdefaramrzi M, Rouhi M, Yazdian-Anari P, Ahmadiyeh MH. Investigation of risk factors, stage and outcome in patients with gestational trophoblastic disease since 2001 to 2011 in Iran-Yazd. International journal of biomedical science: IJBS. 2015;11(4):166.
25. Miyazaki K, Furuhashi M, Ishikawa K, Tamakoshi K, Ikeda T, Kusuda S, et al. The effects of antenatal corticosteroids therapy on very preterm infants after chorioamnionitis. Arch Gynecol Obstet. 2014 Jun; 289(6):1185-90.
26. Miyazaki K, Furuhashi M, Ishikawa K, Tamakoshi K, Hayashi K, Kai A, et al. Long-term outcomes of antenatal corticosteroids treatment in very preterm infants after chorioamnionitis. Arch Gynecol Obstet. 2015 Dec; 292(6):1239-46.
27. Miyazaki K, Furuhashi M, Ishikawa K, Tamakoshi K, Hayashi K, Kai A, et al. Impact of chorioamnionitis on short- and long-term outcomes in very low birth weight

preterm infants: the Neonatal Research Network Japan. J Matern Fetal Neonatal Med. 2016; 29(2):331-7.
28. Lee J, Romero R, Kim SM, Chaemsaithong P, Park CW, Park JS, et al. A new anti-microbial combination prolongs the latency period, reduces acute histologic chorioamnionitis as well as funisitis, and improves neonatal outcomes in preterm PROM. J Matern Fetal Neonatal Med. 2016 Mar; 29(5):707-20.
29. Mojgan Karimi-Zarchi SMRM, Nasrollah Bashardust, Neda Zakerian, Mahbube Zaidabadi, Pouria Yazdian-Anari, Soraya Teimoori. The Clinicopathologic Characteristics and 5-year Survival Rate of Epithelial Ovarian Cancer in Yazd, Iran. Electronic Physician. 2015;7(6):1399-406.