Seung Mi Lee, Roberto Romero*, Joong Shin Park, Piya Chaemsaitong, Jong Kwan Jun and Bo Hyun Yoon*

A transcervical amniotic fluid collector: a new medical device for the assessment of amniotic fluid in patients with ruptured membranes

Abstract

**Aim:** To describe a new device for the transcervical collection of amniotic fluid (AF) in patients with ruptured membranes, and to compare the concentration of proteins in fluid retrieved by transabdominal amniocentesis and the transcervical AF collector.

**Study design:** Paired AF samples were collected in patients with preterm prelabor rupture of membranes (PROM) (n=11) by transabdominal amniocentesis and with the transcervical AF collector (Yoon’s AF Collector™). Three proteins known to have high concentrations in AF [α-fetoprotein (AFP), β-human chorionic gonadotrophin (β-hCG), and prolactin] were measured.

**Results:** (1) There was a significant correlation between the concentrations of analytes in AF obtained by transabdominal amniocentesis and by the transcervical AF collector (r=0.94, P<0.001 for AFP; r=0.96, P<0.001 for β-hCG; r=0.72, P<0.05 for prolactin); (2) Bland-Altman plots showed no evidence of heteroscedasticity between transabdominal or transcervical AF concentrations of these markers.

**Conclusions:** There was a strong correlation between the concentrations of proteins in AF collected by amniocentesis or with the transcervical device.

**Keywords:** Amniocentesis; α-fetoprotein; β-human chorionic gonadotrophin; premature rupture of membranes; prolactin; PROM; pPROM; transcervical amniotic fluid collector; Yoon’s AF Collector™.

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Introduction

Rupture of membranes (ROM) occurs spontaneously during the course of labor [1]. Prelabor rupture of membranes (PROM) is defined as chorioamnionrrhexis prior to the onset of labor and complicates 10% of all pregnancies [2–20]; in 2% of the cases, it occurs in preterm gestations [1, 21–37]. Preterm PROM accounts for 30–35% of all preterm births [21, 24, 38].

Amniotic fluid analysis is used to assess the likelihood of intra-amniotic infection, intra-amniotic inflammation [32, 39–79], and fetal lung maturity [73, 80–95] in preterm PROM. Amniotic fluid can be retrieved with amniocentesis; however, this procedure is technically demanding in the context of rupture of membranes due to the decreased volume of amniotic fluid frequently observed in these cases [37, 96, 97].

We report herein a novel device designed for transcervical collection of amniotic fluid. The purpose of this article is to describe the device and to report the concentration of...
proteins in amniotic fluid collected transabdominally, as well as that obtained transcervically with the use of the device. For this, we measured α-fetoprotein (AFP), beta-human chorionic gonadotropin (β-hCG), and prolactin, which are known to be present in high concentrations in amniotic fluid [98–102].

Materials and methods

Study design

Eleven women with singleton pregnancies admitted to the Seoul National University Hospital, Seoul National University Bundang Hospital, or Dongguk University Hospital, Korea, were included in the study. All patients had the diagnosis of preterm PROM (≤35 weeks of gestation). Amniotic fluid was collected using the new medical device described herein. Transabdominal amniocentesis was also performed to examine the presence or absence of intra-amniotic infection and/or inflammation and is part of the standard of practice in our institution.

The retrieval of amniotic fluid was performed after obtaining written informed consent from the patients, and the Institutional Review Boards of the Seoul National University Hospital, Seoul National University Bundang Hospital, and Dongguk University Hospital approved the collection and use of these samples and information for research purposes.

Collection of amniotic fluid

Transcervical amniotic fluid collection was performed with the transcervical amniotic fluid collector (Yoon’s AF Collector™), which was developed and patented by the authors (patent number: Korea 10-1170053-0000). The collector was placed against the cervix during a sterile speculum exam and fixed with a silicon balloon. Amniotic fluid that leaked through the cervix pooled into the collector (Figure 1). Transabdominal amniotic fluid was obtained after transcervical amniotic fluid collection by amniocentesis under ultrasonographic guidance as previously described [78, 103, 104]. Amniotic fluid was centrifuged for 10 min, and stored in polypropylene tubes at −70°C until assayed.

Amniotic fluid analysis

Amniotic fluid was assayed for AFP, β-hCG, and prolactin by immunoradiometric assays (IRMA) with commercially available kits (AFP: Immunotech, Prague, Czech Republic; β-hCG: Shinjin, Seoul, Korea; prolactin: Shinjin, Seoul, Korea).

Statistical analysis

The concentrations of amniotic fluid proteins in samples retrieved either by transabdominal amniocentesis or using the transcervical amniotic fluid collector were compared using Spearman’s correlation and Passing and Bablok regression analysis. Bland-Altman plots were used to evaluate the differences between protein concentrations obtained from transabdominal amniotic fluid and those retrieved transcervically. We used SPSS version 20.0 for Windows (SPSS Inc., Chicago, IL, USA) and MedCalc version 12.7.1.0 (MedCalc Software, Mariaherke, Belgium). A P-value <0.05 was considered significant.

Results

The clinical characteristics of patients enrolled in the study are summarized in Table 1. None of the patients developed clinical chorioamnionitis.

Figure 2 shows the distribution of protein concentrations in fluid collected either by transabdominal amniocentesis or using the transcervical amniotic fluid collector. The transcervical collector was effective in collecting amniotic fluid from all patients. There was a significant correlation between the concentrations of analytes in transabdominal and transcervical amniotic fluid (Spearman correlation coefficients: r=0.936, P<0.001 for AFP; r=0.964, P<0.001 for β-hCG; r=0.718, P<0.05 for prolactin; Figure 3 and Table 2). Bland-Altman plots showed no evidence of heteroscedasticity between transabdominal and transvaginal amniotic fluid.
Table 1  Clinical characteristics of the study population.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>n=11</th>
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<tbody>
<tr>
<td>Agea</td>
<td>33 (29–36)</td>
</tr>
<tr>
<td>Nulliparity</td>
<td>6 (55%)</td>
</tr>
<tr>
<td>Gestational age at amniocentesisa</td>
<td>30.1 (27.3–34.1)</td>
</tr>
<tr>
<td>Positive AF culture</td>
<td>2 (18%)</td>
</tr>
<tr>
<td>Intra-amniotic infection and/or inflammation</td>
<td>3 (27%)</td>
</tr>
<tr>
<td>Gestational age at deliverya</td>
<td>32.6 (27.4–34.4)</td>
</tr>
<tr>
<td>Cesarean delivery</td>
<td>2 (18%)</td>
</tr>
<tr>
<td>Birthweighta</td>
<td>1956 (1080–2800)</td>
</tr>
<tr>
<td>Histologic chorioamnionitis</td>
<td>6/10 (60%)</td>
</tr>
<tr>
<td>Funisitis</td>
<td>3/10 (30%)</td>
</tr>
</tbody>
</table>

AF=amniotic fluid. *Median (range).

Figure 2  Protein concentrations in transabdominal amniotic fluid and those in transcervical amniotic fluid, with means and 95% confidence interval presented in solid line. (A) α-Fetoprotein (AFP) (IU/mL); (B) β-human chorionic gonadotrophin (β-hCG) (mIU/mL); (C) prolactin (ng/mL).

Discussion

Principal findings of the study

(1) We describe a new device for the transcervical collection of amniotic fluid from patients with ruptured membranes; and (2) a strong and significant relationship was observed between the concentration of proteins in amniotic fluid obtained by the transabdominal and transcervical method.
Amniotic fluid analysis in the assessment of patients with ruptured membranes

Amniotic fluid analysis has been used to study fetal lung maturity [80–82, 84, 85, 89–92, 94, 105], karyotype, intra-amniotic inflammation, and microorganisms (bacteria and viruses) [32, 39–79] in patients with preterm PROM. Fluid obtained from the vaginal pool has been employed to assess fetal lung maturity [73, 80–95, 105]. However, this fluid cannot be used for the assessment of microbial invasion of the amniotic cavity or the assessment of intra-amniotic inflammation, because bacteria are normally present in the vagina, and vaginal fluid contains a high concentration of inflammatory mediators [44, 51, 54, 56, 59–61, 75, 76]. Therefore, we devised a method to obtain amniotic fluid noninvasively, by collecting this biological material directly through the cervix before it reached the vaginal canal. Ultrasound images demonstrate that this fluid is present in the cervix of patients who have ruptured membranes, and can be seen with color Doppler in the endocervical canal [106]. Obtaining amniotic fluid noninvasively can allow assessment of patients with severe oligohydramnios in which amniocentesis is not possible. Moreover, it allows serial evaluation of patients with preterm PROM without repeat amniocentesis.

Collection of amniotic fluid by a transcervical amniotic fluid collector

We obtained amniotic fluid using a transcervical amniotic fluid collector designed for this specific purpose. This device has the following features: (1) it allows collection at the level of the external cervical os, and thereby reduces
the likelihood of dilution of amniotic fluid with vaginal discharge; (2) it prevents leakage of amniotic fluid into the vagina, and therefore, fluid can be obtained even in patients with scant vaginal pooling; and (3) placement of the device for a short period of time (60 min) allowed collection of enough fluid for analysis. Patients did not report discomfort during collection of fluid with this device (Figure 1). Importantly, there was an excellent correlation between the concentration of a set of analytes in fluid retrieved by transabdominal amniocentesis and the use of the transcervical amniotic fluid collector, suggesting that this method may be useful in assessing the composition of amniotic fluid in patients with preterm PROM. Future studies are required to determine if this material can be used to assess the likelihood of intra-amniotic inflammation and fetal lung maturity.

This was a feasibility study; therefore, the sample size was small. We measured AFP, prolactin, and hCG, all of which are known to be present in high concentrations in amniotic fluid. It would be important to assess other components of amniotic fluid, including “omics” characterization (proteomics, metabolomics, lipidomics, etc.).

**Conclusion**

We describe a new device for the collection of amniotic fluid from patients with ruptured membranes. The transcervical amniotic fluid collector might be useful in the evaluation of amniotic fluid in patients with ruptured membranes.

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Table 2

<table>
<thead>
<tr>
<th>Concentrations [mean (SD)]</th>
<th>α-Fetoprotein (IU/mL)</th>
<th>β-hCG (mIU/mL)</th>
<th>Prolactin (ng/mL)</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Transabdominal AF</td>
<td>Transcervical AF</td>
<td>Transabdominal AF</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Transcervical AF</td>
<td></td>
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<tr>
<td></td>
<td>2006 (1771)</td>
<td>2006 (1771)</td>
<td>1997 (1575)</td>
</tr>
<tr>
<td></td>
<td>1190 (1220)</td>
<td>1190 (1220)</td>
<td>1222 (1230)</td>
</tr>
<tr>
<td></td>
<td>829 (592)</td>
<td>829 (592)</td>
<td>784 (400)</td>
</tr>
<tr>
<td>Wilcoxon signed rank test (P-value)</td>
<td>0.93 (P=0.001)</td>
<td>0.96 (P=0.001)</td>
<td>0.72 (P=0.010)</td>
</tr>
<tr>
<td>Spearman correlation coefficients (P-value)</td>
<td>0.94 (P&lt;0.001)</td>
<td>0.96 (P&lt;0.001)</td>
<td>0.72 (P&lt;0.010)</td>
</tr>
<tr>
<td>Passing-Bablok regression analysis</td>
<td>Intercept [95% CI]</td>
<td>176.9 [4.5 to 2.69]</td>
<td>75.6 [7.9 to 2.69]</td>
</tr>
<tr>
<td></td>
<td>Slope [95% CI]</td>
<td>0.86 [0.62 to 1.46]</td>
<td>0.89 [0.67 to 1.30]</td>
</tr>
</tbody>
</table>

AF = amniotic fluid, AFP = α-fetoprotein, β-hCG = β-human chorionic gonadotropin.

References


The authors stated that there are no conflicts of interest regarding the publication of this article.

Article note: The device described in this article was patented by the Seoul National University in Seoul, Korea. Dr. BH Yoon, Dr. JS Park, and Dr. SM Lee are inventors.