Increased Cell-free Fetal DNA in Plasma of Two Women with Invasive Placenta, Akihiko Sekizawa,* Masatoshi Jimbo, Hiroshi Saito, Mariko Iwasaki, Yumi Sugito, Yasuo Yukimoto, Junko Otsuka, and Takashi Okai (Department of Obstetrics and Gynecology, Showa University School of Medicine, 1-5-8, Hatanodai, Shinagawa-ku, Tokyo 142-8666, Japan; * author for correspondence: fax 81-33784-8355, e-mail sekizawa@d8.dion.ne.jp)

Invasive placenta, often involving placenta accreta, placenta increta, and placenta percreta, is a life-threatening complication of pregnancy characterized by invasion of placental villi into the underlying myometrium. Invasive placenta is usually diagnosed as a result of abnormalities encountered during removal of the placenta at delivery. Because of profuse hemorrhaging in the early postpartum period, blood transfusion or emergency hysterectomy is often required in patients with invasive placenta. Invasive placenta is also associated with high maternal morbidity and a high risk of mortality. Therefore, prenatal prediction of this disease would be of great clinical benefit.

Recently, Lo et al. (1) discovered the presence of fetal DNA in maternal plasma. They developed a real-time quantitative PCR assay with which to measure the concentration of fetal DNA within maternal plasma. In addition, they analyzed the Y-chromosome-specific SRY sequence to quantify the number of genome equivalents per milliliter of plasma in women carrying male fetuses (2). Although cell-free fetal DNA is reported to increase in maternal plasma in cases of preterm delivery (3), preeclampsia (4, 5), and pregnancy with a trisomy 21 fetus (6), it is unclear where this fetal DNA originates. In the present study, we hypothesized that trophoblasts might be destroyed by the maternal immune system after invasion of the uterine muscle in patients with invasive placenta. This might indicate that increased concentrations of fetal DNA are found within the plasma of these patients. Thus, for the retrospective trial, we measured the concentration of cell-free fetal DNA in maternal plasma.

Maternal blood samples were obtained from 20 pregnant women after written informed consent was received. Because placenta previa is a risk factor for invasive placenta, seven pregnant women with placenta previa bearing male fetuses were studied (median period of gestation, 34.0 weeks; range, 31–35 weeks). Women in the placenta previa group did not have vaginal bleeding or any symptoms of preeclampsia or preterm labor, but two cases had a history of uterine curettage. For the controls, 13 pregnant women who were also carrying male fetuses and did not have a history of uterine surgery, including cesarean section and/or uterine curettage, were studied (median period of gestation, 33.0 weeks; range, 31–36 weeks). They did not have any complications, including preeclampsia or preterm delivery, and their gestational ages were matched with those of the placenta previa group. This study was performed prospectively. Approval for this study was obtained from the ethics committee of the Showa University School of Medicine.

Maternal plasma was separated from 10 mL of maternal blood, and DNA extraction was performed with a QIAamp Blood Mini Kit (Qiagen). Subsequently, quantitative PCR of a Y-chromosome-specific DYS14 sequence was performed on maternal plasma with a LightCycler (Roche Diagnostics) as described previously (7).

In this study, two women developed invasive placenta. One had placenta accreta, which was complicated with placenta previa and was clinically diagnosed at cesarean section. The other patient belonged to the control group and was diagnosed with placenta increta. In this patient, we were unable to remove a small part of the placenta after vaginal delivery, and placenta increta was diagnosed by magnetic resonance imaging. Statistical analysis was performed with Stat View software (Ver. 5.0; SAS Institute, Inc.).

The fetal DNA concentrations of patients in the control and placenta previa groups are shown in Fig. 1. Excluding data pertaining to the two patients with invasive placenta (one from each group), median fetal DNA concentrations for the control and placenta previa groups were 184.2 (range, 126.0–398.0) genome-equivalents/mL and 294.3 (range, 191.8–484.0) genome-equivalents/mL, respectively. A significantly higher concentration of fetal DNA was observed in the maternal plasma of the placenta previa group compared with the control group ($P = 0.015$, Mann–Whitney U-test). With regard to the two cases involving placenta accreta and placenta increta, fetal DNA concentrations were 609.6 and 582.0 genome-equivalents/mL, respectively.

Previous reports have documented the ability to predict invasive placenta with the use of gray-scale ultrasonography, color Doppler imaging (8), and magnetic resonance imaging. It has also been reported that an increased concentration of creatine kinase (9) or α-fetoprotein (10) in maternal serum is a biochemical marker of the disease. However, abnormal adherence of the placenta to the uterine wall is rarely diagnosed antepartum.

Fig. 1. Fetal cell-free DNA concentration in maternal plasma in the control and placenta previa groups.
In the present study, we demonstrated that the concentration of fetal DNA within maternal plasma is increased in cases of placenta previa, especially in patients with invasive placenta. We believe that invasion of trophoblasts into uterine muscle in these patients led to increased concentrations of cell-free fetal DNA within maternal plasma. Because the concentration of fetal DNA was high in the plasma of women with invasive placenta, antenatal diagnosis might be achieved through analysis of fetal DNA concentrations within maternal plasma. Placenta previa and a previous history of uterine surgery, including cesarean delivery, are considered risk factors for invasive placenta. In women who have these risk factors, fetal DNA quantification would be especially useful in the detection of invasive placenta.

Although we observed an increase in the plasma fetal DNA concentrations of women with placenta previa, the reason for this is unclear. However, the presence of thin and dysfunctional decidua at the lower segment of the uterus might be related to this increase.

In conclusion, we report here that fetal DNA is increased in the maternal plasma of patients with invasive placenta. We propose that the concentration of fetal DNA within maternal plasma might be a useful marker by which to arrive at an antepartum diagnosis of invasive placenta.

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References