Doubling time of urine human chorionic gonadotropin after assisted reproductive technology

In the present study, we measured urinary hCG after assisted reproductive technology to accurately calculate normal doubling time (DT) of hCG, and we compared DT values with prognosis of pregnancy. We clarified the correlation between normal DT values, calculated from urinary hCG levels, and viable and nonviable pregnancies. (Fertil Steril 2005;84:1040–2. ©2005 by American Society for Reproductive Medicine.)

Levels of hCG in serum and urine in early pregnancy are very useful as clinical indicators of gestation (1–4). The course of elevation of hCG levels between two time points is particularly important for prediction of the prognosis of pregnancy, and the doubling time (DT) is used as an index (5–7). Generally, the DT of hCG varies depending on the week of gestation, and many reports indicate that normal values range from 1.4 to 3.5 days (5–7). Doubling time has been established as a predictor of prognosis, with a long DT indicating poor prognosis (8, 9).

Because the date of ET is specified in assisted reproductive technology (ART), it is possible to obtain accurate dates of hCG measurement after fertilization. Thus, hCG measurement after ART allows calculation of the exact time course of hCG levels (10–12). We previously reported that prognosis could be predicted from urinary hCG levels, as well as from serum hCG levels, and that urinary hCG has an advantage over serum hCG in that it allows prognosis to be predicted noninvasively (13).

In the present study, we measured urinary hCG levels after ART, to accurately calculate normal DT of hCG, and we compared DT values with prognosis of pregnancy (i.e., viability and number of fetuses [single or multiple]), to investigate whether prognosis can be predicted from DT.

The study was conducted from April 2000 through May 2001 in our hospital. Informed consent was obtained from all patients, and the study was approved by the institutional review board of the Jichi Medical School Hospital and the Institute for Central Clinic. The subjects were 157 patients (age range, 26–43 years; mean age, 33.5 years) whose urinary hCG level was ≥2.0 IU/L at 14 days after oocyte retrieval and who had an increased urinary hCG level 21 days after oocyte retrieval (12 and 19 days after frozen embryo transplantation).

The relationship between the hCG DT and the outcome of pregnancy (normal pregnancy, abortion, multiple pregnancy) was analyzed retrospectively with statistical procedures. Patients with ectopic pregnancy or who were negative for urinary hCG at 14 or 21 days after oocyte retrieval were excluded from this analysis. We also excluded patients whose urinary hCG level at 21 days after oocyte retrieval was lower than the level obtained 14 days after oocyte retrieval.

The ART methods used were as follows: IVF, 31 patients; intracytoplasmic sperm injection, 89 patients; GIFT, 12 patients; frozen embryo transplantation, 25 patients. Random urine samples obtained during the patients’ visits to our hospital were examined. In all patients, an IM injection of hCG (10,000 U) was administered 24–48 hours before oocyte retrieval, and a P preparation (rather than an hCG preparation) was used for luteal support.

Statistical analysis of the data was performed with commercial software (StatView 4.5 for Macintosh; Abacus Concepts, Berkeley, CA).

We calculated the doubling time using the following formula (5): Doubling time (days) = (log 2) × (time interval in days)/log (hCG2/hCG1).

The urine hCG assay that we used (VIDAS assay system; bioMerieux Japan, Tokyo, Japan; 2.0 IU/L–1,500 IU/L) is a quantitative ELISA. The test includes a VIDAS immunoanalyzer and combines the sandwich immunoenzymatic method in one step with a final fluorescence detection. The reference preparation was the World Health Organization first International Reference Preparation 75/537. Samples with levels higher than this upper limit were reanalyzed after manual predilution with the diluent provided with the kit.

The patients in whom a fetal heart beat was detected at week 8 were defined as the viable group. The nonviable group comprised patients with chemical pregnancies (positive for urinary hCG but no gestational sac on day 14 and/or day 21) or clinical abortions (fetal heart beat was confirmed once, but abortion occurred subsequently; a gestational sac was observed, but no fetal heart beat was detected).
Of the 157 patients, 115 (73.2%) were classified as the viable group: single pregnancy, 78 patients; multiple pregnancy, 37 patients. The nonviable group comprised 42 patients (chemical pregnancy, 17 patients; clinical abortion, 25 patients) who were positive for urinary hCG on day 14 and had an increased urinary hCG level on day 21 but were negative for a fetal heart beat at 8 weeks.

Table 1 shows the mean values of urinary hCG levels on days 14 and 21 and the mean value of urinary hCG DT for days 14 to 21. The average hCG level was significantly lower for the nonviable group than for patients with a normal single pregnancy (P<.005) and patients with a normal multiple pregnancy (P<.005). The patients with a normal multiple pregnancy had significantly higher average hCG levels than the patients with a normal singleton pregnancy (day 14, P<.05; day 21, P<.005). However, there were no significant differences in DT between normal single pregnancies and normal multiple pregnancies (P=.378). The DT of the nonviable group was significantly longer (almost twice as long) than those of the groups with normal single pregnancies and normal multiple pregnancies (P<.005 and P<.05, respectively).

The distribution of DT (data not shown) showed that the viable rate tended to be higher for patients with a DT of ≥1.2 and <3.2. The χ² test revealed that the viable rate was significantly higher for patients with a DT of ≥1.2 and <3.2 than for patients with a DT of <1.2 or ≥3.2 (P<.005) and that the odds ratio was 5.6 (95% confidence interval 2.6–12.2). These cut-off values provided a sensitivity of 0.83, specificity of 0.52, positive predictive value of 0.83, and negative predictive value of 0.54. It is necessary to pay attention to patients outside this range because the specificity and positive predictive value are low.

The objective of the present study was to determine the normal DT of urine hCG after ART and to evaluate the prognostic value of this variable. For all subjects, measurement of the urine hCG level was performed 14 and 21 days after oocyte retrieval, and an accurate normal DT value was obtained. The mean DT was significantly longer in the nonviable group. This finding is consistent with those of previous studies (8, 9), indicating that DT is useful for prediction of prognosis.

The mean urinary hCG level correlated with the prognosis of pregnancy and the number of fetuses, but the mean DT value did not correlate with the number of fetuses. These findings suggest that the total hCG level is proportional to the volume of producer cells, but DT does not seem to be dependent on the volume of producer cells. Zegers-Hochschild et al. (9) measured serum hCG and calculated that the normal DT for a period similar to that of the present study (from 11 to 23 days after oocyte retrieval) was 1.6 ± 0.4 days for patients with a single fetus and 1.5 ± 0.3 days for patients with multiple fetuses. Their findings are roughly similar to those of the present results. They (9) also reported that although the absolute hCG level was higher in patients with multiple fetuses than in women with a single fetus, there was no difference in DT, which is also consistent with the present results.

In the present study, the mean DT was longer in the nonviable group than in the viable group, which is consistent with findings of previous studies of serum hCG (8, 14, 15). However, the present distribution results indicate that the number of nonviable cases was higher for extremely low and long DT values. Thus, we showed that a good prognosis is associated with a DT of ≥1.2 days and <3.2 days.

Doubling time varies depending on the sampling interval and weeks of gestation, and reports have shown that it is
necessary to consider these factors when examining the significance of DT (5, 16). Because the measurement interval and the week of gestation were kept constant in the present study, it was possible to accurately calculate the normal time course of hCG levels. The advantage of using the urinary hCG level, compared with the serum hCG level, for prediction of pregnancy outcome is that urine specimens can be obtained noninvasively. The results of the present study, in which the urinary hCG level was measured in 25 minutes with a commercial kit (VIDAS), also suggest that such kits are highly useful for clinical applications.

In conclusion, the present findings clarify the distribution of normal DT values of ART patients, as calculated from urinary hCG levels.

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