

Serum human chorionic gonadotropin measurements may predict pregnancy outcome and multiple gestation after in vitro fertilization

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Objective: To predict pregnancy outcome and multiple gestation using a common parameter by which hCG values are made comparable independently of the day of blood sampling.

Design: Retrospective study.

Setting: University-based IVF program.

Patients: One hundred twenty IVF pregnancies conceived between November, 1995 and August, 1999.

Interventions: None.

Main Outcome Measures: Early pregnancy loss (preclinical and first trimester abortions, ectopic pregnancies) or ongoing pregnancies (singleton and multiple deliveries, second trimester abortions). Day 11 hCG levels were calculated assuming an exponential increase of hCG values in early pregnancy. Receiver-operating characteristic analysis was used to determine cut-off levels with the best sensitivity and specificity for the prediction of pregnancy outcome.

Results: Serum hCG levels in the group of early pregnancy loss were significantly lower than in ongoing pregnancies. A cut-off level of 50 IU/L predicts pregnancy outcome with a sensitivity of 75% and a specificity of 81%, while an hCG value >135 IU/L predicts a multiple ongoing pregnancy with a sensitivity of 80% and a specificity of 88%.

Conclusion: After IVF, early pregnancy loss or multiple gestation may be predicted with high sensitivity and specificity by using cut-off values of serum hCG derived from two measurements independently of the day of blood sampling. (Fertil Steril® 2002;78:540–2. ©2002 by American Society for Reproductive Medicine.)

Key words: In vitro fertilization, hCG, pregnancy outcome, multiple pregnancy

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Patients undergoing IVF are at increased risk for an adverse outcome of pregnancy compared with women who conceive naturally (1). Patients who know that their prognosis is favorable have less anxiety. In addition, closer follow-up of multiple gestations that is started as early as possible is beneficial.

Several approaches using endocrinologic markers, such as serum progesterone, E₂, inhibins, and pregnancy-specific β 1-glycoprotein, have been examined in an attempt to predict pregnancy outcome (2, 3), but the clinical role of these measurements is not yet fully established. Recently, single hCG measurements were proposed to predict pregnancy outcome (4, 5). However, no study has established

a single common variable that can be used as a cut-off value independent of the day of serum sample collection.

We sought a cut-off value for a common variable derived from two consecutive hCG measurements that could reliably predict early pregnancy loss independent of the specific day of serum sample collection. We also sought to determine whether multiple gestations could be predicted by using the same method.

We reviewed the records of all patients who conceived after IVF or ICSI and ET between November 1995 and August 1999 at our unit. In all cases, oocyte pick-up was performed after stimulation with GnRH agonist plus go-

nadotropin according to the long protocol. A maximum of four embryos per cycle were replaced 2 or 3 days after oocyte aspiration.

Of 137 cycles resulting in pregnancy, 17 were excluded from further analysis because hCG was given for luteal phase support. In the resulting group of 120 cycles, only micronized progesterone (200 mg given intravaginally three times daily) was given for luteal phase support. The study was approved by the institutional review board of the First Department of Obstetrics and Gynecology, Semmelweis University Faculty of Medicine, Budapest, Hungary.

Pregnancy was detected by two measurements of serum hCG from venous blood samples obtained 8 to 16 days after ET, with a difference of two days. Between November 1995 and May 1998, serum hCG concentrations were measured by using a microparticle enzyme immunoassay for total β -hCG (IMx; Abbott Laboratories, Abbott Park, IL). The coefficients of variation were 4.4% to 5.9% over the range of 25 to 750 IU/L. Starting in June 1998, the assay was performed by using the AxSYM system (Abbott Laboratories), which had a coefficient of variation of 4.1% to 6.6%. The sensitivity of both assays was 2 IU/L.

Pregnancies were defined by serum hCG levels ≥ 25 IU/L on either of the two assessments, and patients were classified according to pregnancy outcome. The group with early pregnancy loss included preclinical abortions (pregnancy detected by hCG measurements but no gestational sac seen on transvaginal ultrasonography), first-trimester abortions, and ectopic pregnancies. Ongoing pregnancy was defined as progression to > 12 weeks' gestation (singleton and multiple deliveries and second-trimester abortions).

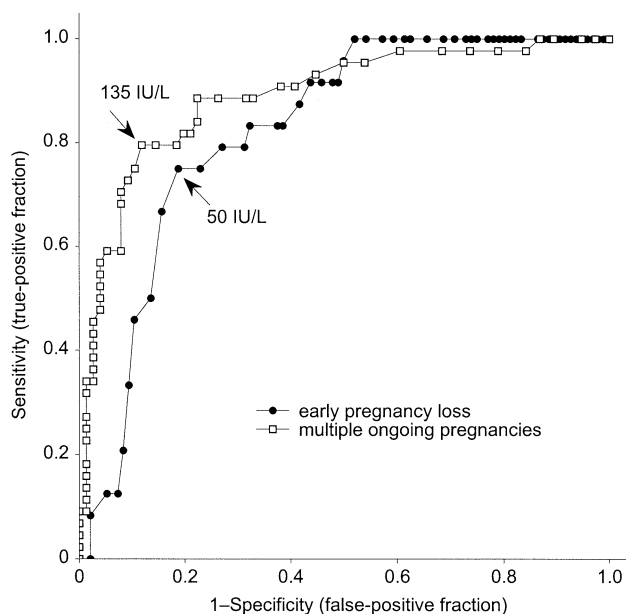
On the basis of the exponential increase of serum hCG levels in early pregnancy (6), doubling times were calculated by using the following formula: doubling time = $\log_{10} 2 \times (\text{time interval in days}) / \log_{10} (hCG_2/hCG_1)$, where hCG_1 and hCG_2 are consecutive serum hCG concentrations (7).

Although the increase in hCG levels slows down at later stages of pregnancy, it can be regarded as constant for the short interval of interest in this study. We used doubling times to calculate theoretical day 11 (post-ET) hCG levels according to the formula $hCG_{\text{day 11}} = hCG_1 \times 2^{(11 - \text{day}_1) / \text{doubling time}}$, in order to obtain a comparable measure of serum hCG concentrations. We chose post-ET day 11 values for this purpose because most of our patients had their first serum samples collected on this day and we could thus use real values for most of the cases.

Differences between groups were detected after logarithmic transformation of hCG data by using the Student two-sample *t*-test. We constructed receiver-operating characteristic (ROC) curves by using all available day 11 hCG values and classifying the values into 5 IU/L wide categories based on level. Receiver-operating characteristic curves are generated by joining points defined by sensitivity (true-positive

FIGURE 1

Nonparametric receiver-operating characteristic plots of calculated day 11 serum hCG levels to distinguish between ongoing pregnancies and early pregnancy losses (*closed circles*) and to predict multiple pregnancies (*open squares*).



Urbansek. Serum hCG in predicting pregnancy outcome. *Fertil Steril* 2002.

proportion) and corresponding values of $1 - \text{specificity}$ (false-positive proportion) for possible cut-off limits that predict a binary outcome (8). In this study, ROC curves were used to distinguish between early pregnancy loss and ongoing pregnancies and to predict multiple ongoing pregnancies.

Patients ranged in age from 23 to 43 (median age, 31) years. Infertility was due to tubal factor (49%), endometriosis (5%), or male factor (40%) or was unexplained (6%). Twenty percent (24 of 120) of pregnancies ended in early loss, and 80% (96 of 120) were ongoing.

Calculated day 11 hCG values differed significantly between women with early pregnancy loss and those with ongoing pregnancy (36.9 vs. 102.9 IU/L; $P < .0001$). We generated an ROC plot to determine which cut-off value for calculated day 11 hCG levels predicted early pregnancy loss with optimal sensitivity and specificity (Fig. 1). The plot shows that the best choice is 50 IU/L, which has a sensitivity of 75% and a specificity of 81%. The positive predictive value at this level was 50%, and the negative predictive value was 93%.

We also sought a cut-off value to predict multiple gestation with the greatest certainty. The cut-off level that had optimal sensitivity (80%) and specificity (88%) was 135 IU/L. The positive predictive value was 80%, and the negative predictive value was 88%.

A single measurement of serum hCG does not characterize the dynamics of the increase in hCG level. To compare such results, all measurements must be obtained on the same post-ET day (5), or comparisons must be made for every day (4). We used a model for comparing hCG values that is based on the exponential increase of placental hormone levels in early pregnancy (6).

Our cut-off level with the best balance between sensitivity and specificity (50 IU/L) is in good agreement with the findings of other studies of prediction of early pregnancy loss by use of hCG measurements: Sugantha et al. (9) reported a discriminatory hCG value of 50 IU/L for day 14 after oocyte retrieval, and Bjercke et al. (5) found an optimal cut-off of 55 IU/L for post-ET day 12.

Cut-off values were obtained from our sample by using the measurement protocols of our laboratory. Although ROC analysis is based on pairs of sensitivity and specificity, which are by definition independent of the prevalence of the outcome of interest, the results may be influenced by factors specific to our methods.

In conclusion, we found discriminatory serum hCG values that can predict early pregnancy loss (<50 IU/L) and multiple gestation (>135 IU/L) with the best balance between sensitivity and specificity on post-ET day 11, the earliest to date, which can be used independent of the day of serum sample collection during the interval ranging from 8

to 16 days after ET. Any serum variable measured in early pregnancy will probably help in patient management, reduce anxiety, and direct surveillance, but these results must always be interpreted in the context of the findings on transvaginal ultrasonography performed from week 5 onward.

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