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Unexplained Elevated Midtrimester Maternal Serum Levels of Alpha Fetoprotein, Human Chorionic Gonadotropin, or Low Unconjugated Estriol: Recurrence Risk and Association With Adverse Perinatal Outcome

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Objective: To determine if women experiencing an unexplained elevated maternal serum alpha fetoprotein (MSAFP; ≥2.0 MoM) or human chorionic gonadotropin (hCG; ≥2.0 MoM), or low unconjugated estriol (E3; ≤0.5 MoM) in one pregnancy are at increased risk for similar results in a subsequent pregnancy, and to determine if recurrence of these analyte extremes is associated with adverse perinatal outcome.

Methods: We identified all women delivering two consecutive singleton pregnancies at one hospital between 1992–1997 for whom second trimester trisomy 21 serum screen was performed in each pregnancy. All screens were performed in a single laboratory. Each pregnancy delivered after 20 weeks and had gestational age confirmed by ultrasound prior to 24 weeks. Subjects were excluded if a fetal anomaly or aneuploidy was present. Adverse outcomes included abruption, oligohydramnios, preeclampsia, preterm membrane rupture, preterm delivery, stillbirth, birthweight $<10^{th}$ centile, and admission to neonatal intensive care unit (NICU).

Results: A total of 538 women had 1,076 pregnancies meeting inclusion criteria; 12/515 (2.3%) of women with a normal MSAFP, 28/470 (6.0%) with a normal hCG, and 11/504 (2.2%) with a normal E3 in the first pregnancy had an anomalous result for the respective analyte in the second pregnancy. In contrast, only 4/23 (17.4%) patients with an elevated MSAFP ($P = 0.003$), 14/44 (31.8%) with an elevated hCG ($P < 0.001$), and 2/10 (20.0%) with a low E3 ($P < 0.025$) in the first pregnancy had the same analyte anomaly recur in the second pregnancy. The odds ratios for recurrent elevated MSAFP, hCG, and low E3 were 7.5, 5.3, and 9.2, respectively. Adverse perinatal outcomes occurred with similar frequency, regardless of MSAFP, hCG, or E3 results in consecutive pregnancies, using women with normal MSAFP, hCG, and E3 results in one or both pregnancies as controls.

Conclusions: Women experiencing an anomalous serum analyte in one pregnancy are at significant risk to experience the same analyte result in a subsequent pregnancy. *J. Matern.-Fetal Med. 2000;9:161–164.*

Key words: maternal serum screening; Down syndrome; pregnancy outcome

INTRODUCTION

Screening for fetal open neural tube defects using maternal serum alpha-fetoprotein (MSAFP) has been available for over two decades. A later study noted that unusually low levels of MSAFP were associated with an increased risk of fetal Down syndrome [1]. Subsequently, addition of human

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chorionic gonadotropin (hCG) and unconjugated estriol (E3) increased Down syndrome screening efficiency [2]. Several studies have noted that elevated levels of MSAFP [3–5], elevated hCG [6–8], or low E3 [9,10] were associated with an increased risk of adverse pregnancy outcome in nonanomalous fetuses.

Availability of these tests for several years has resulted in large numbers of women receiving screening in more than one pregnancy. Previous studies suggest that patients with an unexplained elevated MSAFP or hCG appear to have an increased probability for recurrence of that analyte abnormality in a subsequent pregnancy [11–13]. The recurrence risk of an anomalously low E3 has not been evaluated. In addition, the clinical significance with respect to pregnancy outcome of a recurring unexplained anomalous serum analyte is unknown. Therefore, we sought to determine if women experiencing an unexplained elevated MSAFP or hCG, or low E3 in one pregnancy are at increased risk for similar results in a subsequent pregnancy and to determine if pregnancies with a recurrent anomalous analyte are at increased risk for adverse perinatal outcome.

METHODS

We retrospectively identified all women delivering two consecutive singleton pregnancies at our facility between January 1, 1992, and December 31, 1997, for whom second trimester trisomy 21 serum screen was performed in both gestations. Each pregnancy delivered after 20 weeks gestation and had gestational age confirmed by ultrasound prior to 24 weeks gestation. Subjects were excluded if fetal anomalies or chromosomal abnormalities were present. If a patient delivered more than two pregnancies during the study period, only the last two were included, in keeping with previous reports [14].

All maternal serum analyses were performed in a single laboratory. Details of the screening program have been published previously [7,14,15]. Analytes were adjusted for weight; only MSAFP levels were adjusted for race and maternal insulin-dependent diabetes status. An anomalous MSAFP was defined as ≥2.0 multiples of the median (MoM). An elevated hCG was defined as ≥2.0 MoM, and low E3 as ≤0.5MoM, as previously described [8,12]. The recurrence risk for each abnormal analyte was calculated separately.

Adverse perinatal outcomes included placental abruption, oligohydramnios, preeclampsia (blood pressure ≥140/90mm Hg after 20 weeks' gestation, accompanied by generalized edema or proteinuria), [7] preterm premature rupture of membranes (<37 weeks' gestation), preterm delivery (<37 weeks' gestation), birthweight <10th centile, [16] admission to neonatal intensive care unit, or stillbirth. Data were obtained from the computerized records of our institution and the University of Connecticut maternal serum screening program. Women with normal MSAFP, hCG, and E3 results in both pregnancies served as the comparison group for subjects with an exclusively elevated MSAFP or hCG, or low E3 in either or both gestations. Patients experiencing more than one anomalous analyte in a single pregnancy or different anomalous analytes in consecutive pregnancies were specifically excluded from the analysis of adverse perinatal outcome.

Between-pregnancy analyte correlations were evaluated using the Pearson correlation coefficient and two-tailed tests of significance. Statistical analysis employed Fisher's exact test or chi-square, with $P < 0.05$ considered significant. 95% confidence intervals were calculated for odds ratios.

RESULTS

A total of 538 women had 1,076 pregnancies meeting inclusion criteria. Twenty-four of these women had only MSAFP results available. Our subjects consisted of 440 whites, 23 blacks, 67 Hispanics, and 8 women of other races. The mean ages at the first and second pregnancies were 28.5 ± 4.5 and 30.8 ± 4.5 years, respectively. At the time of the first study pregnancy, 385 women were nulliparous. The ranges of MoM for MSAFP were 0.37–3.23 and 0.10–2.78, for hCG 0.25–3.99 and 0.001–3.90, and for E3 0.40–2.35 and 0.16–2.87 for first and second study pregnancies, respectively. Between-pregnancy correlation coefficients for MSAFP, hCG, and E3 were 0.42 ($P < 0.001$), 0.40 ($P < 0.001$), and 0.37 ($P < 0.001$), respectively. We found that 12/515 (2.3%) of women with a normal MSAFP in the first pregnancy, 28/470 (6.0%) with a normal hCG in the first pregnancy, and 11/504 (2.2%) with a normal E3 in the first pregnancy had the respective analyte anomalous in the second pregnancy. Of the 23 patients with an elevated MSAFP in the first pregnancy, four (17.4%) also had an elevated MSAFP in the second pregnancy ($P = 0.003$). Of the 44 women with an elevated hCG in the first pregnancy, 14 (31.8%) also had an elevated hCG in the second pregnancy ($P < 0.001$). Of the 10 subjects with a low E3 in the first pregnancy, two (20.0%) exhibited a low E3 in the second pregnancy ($P < 0.025$). The relative risks for recurrent elevated MSAFP, hCG, and low E3 were 7.5 (95% CI = 2.6–21.4), 5.3 (95% CI = 3.0–9.4), and 9.2 (95% CI = 2.3–36.1), respectively (Fig. 1).

Adverse perinatal outcomes occurred with similar frequency regardless of MSAFP, hCG, or E3 results in consecutive pregnancies (Fig. 2). We did not compare the frequency of the different adverse outcomes by individual analyte, because relatively few pregnancies with anomalous analytes experienced adverse perinatal events. In both pregnancies, 401 women had normal MSAFP, hCG, and E3 results and served as the comparison group for subjects with an elevated MSAFP, hCG, or low E3 for both gestations. In all, 102/802 (12.7%) control pregnancies were characterized by an adverse outcome. Of the 18 subjects having an elevated MSAFP in only one gestation, four (22.2%) experienced a poor outcome in the affected pregnancy ($P = 0.27$). Three women had recurrent elevated MSAFP levels

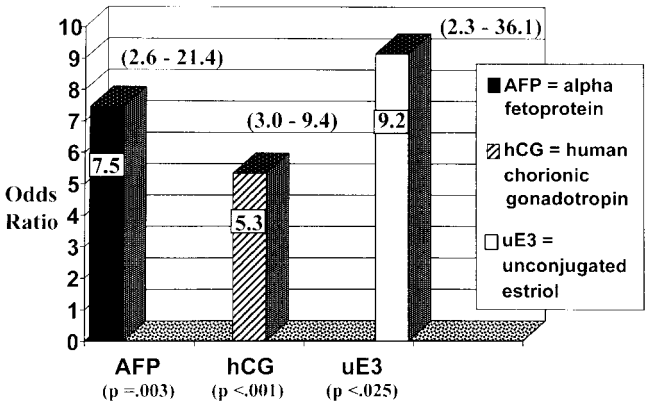

Fig. 1. Odds ratio for anomalous analyte in second pregnancy given anomalous result in first pregnancy (95% CI) CI = confidence interval.

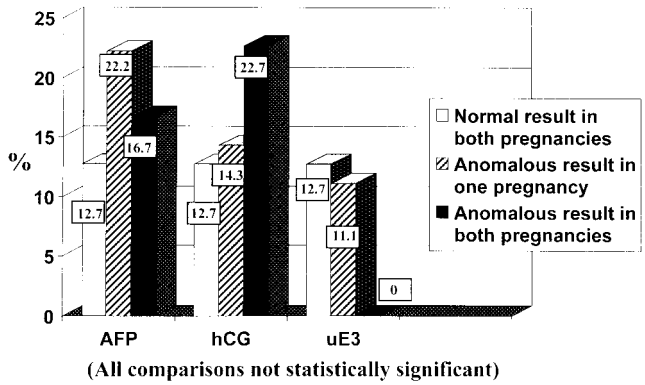

Fig. 2. Percentage of pregnancies with adverse outcomes by analyte results in consecutive pregnancies. AFP = alpha fetoprotein hCG = human chorionic gonadotropin uE3 = unconjugated estriol.

and 1/6 (16.7%) pregnancies had an adverse event (P = 0.56). Of the 49 subjects having an elevated hCG in only one gestation, seven (14.3%) experienced a poor outcome in the affected pregnancy (P = 0.75). Eleven women had a recurrent elevated hCG and 5/22 (22.2%) pregnancies had an adverse event (P = 0.19). Of the 18 subjects having a low E3 in only one gestation, two (11.1%) experienced a poor outcome in the affected pregnancy ($P > 0.90$). Two women had recurrent low E3 levels and 0/4 (0%) pregnancies had an adverse event ($P > 0.90$).

DISCUSSION

Multiple marker screening for Down syndrome has been available for almost a decade, permitting testing of women in more than one pregnancy. Investigators have examined the between-pregnancy variability of serum analytes, focusing attention on MSAFP and hCG [11,12]. Since our screening program additionally employs E3, we specifically sought to determine if women having unexplained pathologic levels of MSAFP, hCG, or E3 in one pregnancy were more likely to have similar results in their next pregnancies.

The current investigation supports reported recurrence risks for anomalously elevated MSAFP and hCG. Dar et al. [12] noted that an MSAFP ≥2.0 MoM in a first pregnancy conferred a 6.5–fold increased likelihood of a recurrent unexplained elevated MSAFP, similar to our observed 7.5–fold increased risk. We found a between-pregnancy MSAFP correlation coefficient of 0.42, corroborating reported values ranging from 0.24–0.33 [11,12,17]. Authors also found a 3.8–fold increased chance of a recurrent hCG ≥2.0 MoM, compared to the 5.3–fold likelihood in our study [12]. Published correlation coefficients for intact hCG of 0.30 and 0.38 are consistent with our results of 0.40 [12,17].

Our study demonstrates a significantly increased risk for a recurrent low E3. Women with a low E3 in one pregnancy are over nine times more likely to have similar subsequent results than women with a normal E3 in their first pregnancies. The between-pregnancy correlation coefficient of 0.37 for E3 was highly significant and consistent with the result of 0.28 reported by Holding and Cuckle [17].

The etiology of recurrent unexplained anomalous analytes remains unknown. Authors speculate that nonspecific causes such as intrinsic biological variability, genetic predisposition, or unspecified environmental factors may influence serum marker results in consecutive pregnancies [12,13]. Increasing parity alone does not appear to be responsible for these observations [18,19]. Although only a small minority of women will experience a recurrent elevated MSAFP or hCG, [11,12] or low E3, the between-pregnancy correlations of analyte values will result in higher than expected numbers of women showing recurrent false-positive Down syndrome screens [20].

The perinatal implications of consecutive-pregnancy serum marker results have not been addressed to date. Elevated levels of MSAFP, [3–5], hCG, [6–8], or low E3 [9,10] unexplained by fetal anomalies or chromosomal abnormalities have been associated with an increased risk of adverse perinatal outcome. Previous studies of serum analyte levels in consecutive pregnancies have specifically excluded [12] or not discussed [11,13,17] gestations with such complications. Thus, the significance of a recurrent unexplained anomalous marker was unknown.

Our investigation did not demonstrate an increased risk of adverse perinatal events in pregnancies characterized by a recurrent elevated MSAFP or hCG, or low E3. However, we recognize several possible explanations for these negative findings. The frequency of poor outcomes in gestations with MSAFP or hCG ≥2.0 MoM is directly related to the degree of analyte elevation [7,8,21]. Likewise, the risk for adverse outcomes in the presence of a low E3 is directly related to the degree of analyte depression [9]. Most of our subjects demonstrated only mildly elevated MSAFP or hCG values, or mildly low E3 levels. Therefore, the relative infrequency of poor perinatal outcomes is not surprising.

Alternatively, mildly elevated MSAFP or hCG, or slightly low levels of E3, may not incur additional risk for adverse pregnancy outcomes beyond that inherent in our population. Investigators failing to confirm an association between elevated MSAFP [22] or hCG [23] and adverse

perinatal outcome suggested that these markers may have little predictive value for women with additional risk factors that place them at high risk for complications. The retrospective nature of our study and information available within the respective databases precluded consideration of such potential confounders in the adverse outcomes noted. Additionally, the possibility that this study may suffer from a limited sample size cannot be excluded. Specifically, 300 subjects with adverse outcomes would be necessary for each group characterized by a single abnormal marker to attain 80% power to detect a 50% increase in adverse outcomes from 13% to 20%, with alpha = 0.05. Our investigation currently offers 12% to 25% power to detect this difference, depending on the analyte studied.

In summary, our study demonstrates a significant risk of an extreme analyte value to recur in a subsequent pregnancy. While we observed no increased risk of adverse perinatal outcome in women with recurrent anomalous serum markers, definitive conclusions must await larger series. Whether or not between-pregnancy analyte correlations can improve open neural tube defect and Down syndrome identification requires additional studies that evaluate the extent of correlation when one or both pregnancies are affected.

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