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Case report

Trophoblast antigen levels in the first trimester of a trisomy 22 pregnancy

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Abstract

We report trophoblast antigen (pregnancy-associated plasma protein-A, PAPP-A; free β-human chorionic gonadotrophin, FβhCG) expression in a trimosy 22 pregnancy. Maternal concentrations of these antigens were depressed prior to detection of abnormalities by ultrasonography. Immunohistochemical findings were consistent with depressed marker expression.

Keywords: Trophoblast antigen; Trisomy 22 pregnancy; Depressed marker expression

1. Introduction

Trisomy 22, characterised by deformities such as intrauterine growth retardation, cleft lip and palate, micrognatha, genital abnormalities, congenital heart defects and renal malformations, is rare in live-born infants but is relatively common in spontaneous abortions (2.9/100, [1]). To date, little is known about placental biochemistry in this abnormality.

Of the numerous trophoblast antigens, pregnancy-associated plasma protein-A (PAPP-A) and free β-subunit of human chorionic gonadotrophin (free β-hCG), perhaps show the most promise as pregnancy prognosticators. Human chorionic gonadotrophin (hCG) is synthesised by the trophoblast as two separate subunits, α and β, with β-subunit synthesis the rate-limiting step. Serial hCG measurement has been recommended for monitoring compromised early pregnancies [2]. Depressed free β-hCG mRNA expression has been demonstrated in trophoblast tissue obtained from spontaneous abortions [7], so accounting for reduced serum levels of free β-hCG and hCG.

PAPP-A, a more recently described pregnancy-associated protein, is quite distinct from other placenta-derived proteins. It is a large tetrameric protein (Mr 820 kDa), rich in carbohydrate and containing one zinc atom per monomeric subunit [3]. Clinical studies in early pregnancy have consistently demonstrated a close relationship between maternal serum PAPP-A levels and pregnancy viability [4], with depressed or undetectable PAPP-A concentrations in pregnancies which spontaneously aborted, even in the presence of normal ultrasonographic findings [2]. This study is the first to detail PAPP-A expression by lethal aneuploid placental tissue of known karyotype.

2. Materials and methods

2.1. Case history

A couple treated for primary infertility due to endometriosis conceived spontaneously and weekly blood samples were collected for hormone monitoring. The pregnancy was uncomplicated apart from some light transient vaginal blood spotting during week 6. Transvesical ultrasound performed at 7 weeks showed a single gestational sac with n rmal yolk sac and live fetus. No unusual features were detected.

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At 8 weeks gestation, hormonal support was instigated (oestradiol valerate, 2 mg three times daily, oral, and progesterone, 50 mg per day, intramuscular injection). Transvesical ultrasound findings at 9 weeks indicated a blighted ovum. Prior to complete evacuation of the uterine cavity, transvaginal chorionic villus sampling (CVS) was performed and trophoblastic villi were obtained for cytogenetic analysis and histology.

2.2. Biochemistry

Weekly venous blood samples were obtained, by routine venepuncture, from week 5 of pregnancy. Serum was stored (−20°C) until required for progesterone and oestradiol (Amerlite; Kodak Clinical Diagnostics Ltd, Sydney), total β-hCG (ACS 180 kit, Ciba Corning, USA), free βhCG (Bioclone, Aust.) and PAPP-A [5] assays.

2.3. Histology

Placental tissue was formalin-fixed (4%) and paraffin-embedded by routine pathology techniques. Sections (5 μm thickness) were deparaffinised and stained with haematoxylin and eosin or immunologically probed with antibodies to PAPP-A (MAb 25-1), intact hCG (DAKO 033A), α-glycoprotein (Bioclone 637), free β-hCG (Bioclone 375), human placental lactogen (hPL) (DAKO 054) and pregnancy-specific β1-glycoprotein (SP1) (DAKO 101A). Visualisation was achieved with a two-step avidin-biotin method (DAKO LSAB 2 peroxidase kit, DAKO Corp, USA). Normal mouse serum was used as a negative control.

3. Results

No abnormalities were ultrasonographically detected at 7 weeks of gestation. Scanning showed a single gestational sac with normal yolk sac and a live fetus. However, a subsequent scan at 9 weeks revealed a single intrauterine gestational sac (diameter = 22 mm), equivalent to 6–7 weeks gestation. No fetal pole was present. Remnants of an early yolk sac and some possible fetal parts were noted. Additional fluid had collected adjacent to the sac, just above the cervix. These ultrasound findings were consistent with a blighted pregnancy.

Due to decreasing progesterone and estradiol levels in maternal serum, hormonal support was initiated and, at week 9 of pregnancy, progesterone and estradiol levels returned to within normal limits (Table 1). Total β-hCG levels were slightly depressed from week 7 and the doubling rate, relative to normal pregnancies, was markedly prolonged. PAPP-A (Fig. 1) and free β-hCG (not shown) levels were depressed (< 0.5 MoM), from eight weeks of gestation. Both markers continued to decline in concentration.

Karyotyping of trophoblastic tissue confirmed a male karyotype with trisomy 22 (47, XY +22). Histopathology showed first trimester chorionic villi which were vascularised but showed no evidence of central cavitation. No atypical trophoblastic change was identified. By immunohistochemistry, PAPP-A, SP1 hPL, hCG and the α-subunit of hCG (α-glycoprotein) were localised only to the villus syncytiotrophoblast cells of normal (not shown) and trisomy 22 placenta (Fig. 2). Villus mesenchyme and cytotrophoblast cells were negative. Intact hCG immunoreactivity was indistinguishable between normal and trisomy 22 placental tissue. SP1, hPL and α-glycoprotein reactivity were marginally depressed and variable between villi. PAPP-A (Fig. 2D) immunoreactivity was markedly depressed, showing only weak and patchy staining. Free β-hCG reactivity (Fig. 2C) was indistinguishable from the negative control.

Table 1
Hormone levels in early trisomy 22 pregnancy

Gestation (weeks)	Progesterone (nmol/L)	Oestradiol (nmol/L)	Total β-hCG (IU/L)
5	73.9	2090	5190
6	46.1	1960	12 975
7	35.1	1840	18 380
8	22.4*	880*	21 735
9	78.0	4260	19 020

* indicates concentration below 10th centile.

4. Discussion

This report details trophoblast antigen expression in a pregnancy with a trisomy 22 fetus. In early pregnancy (<7 weeks), marker levels in maternal circulation were indistinguishable from normal, suggesting the feto-placental unit was viable. In contrast, at 8 weeks of gestation, circulating levels of trophoblast-derived proteins (PAPP-A, total β-hCG, free β-hCG) were depressed.

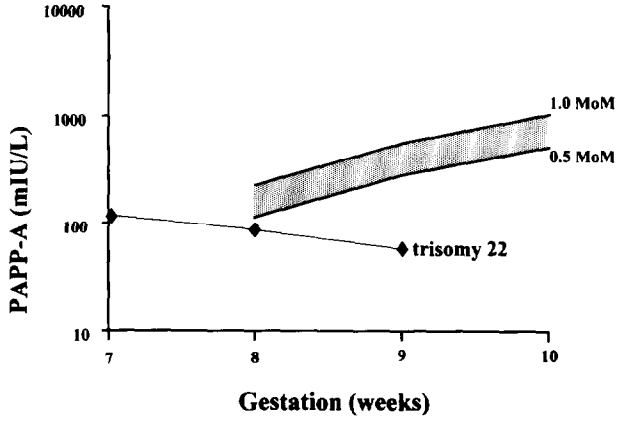

Fig. 1. Maternal serum PAPP-A concentrations in a trisomy 22 pregnancy. Shaded area represents lower limit (<0.5 MoM) of normal range.

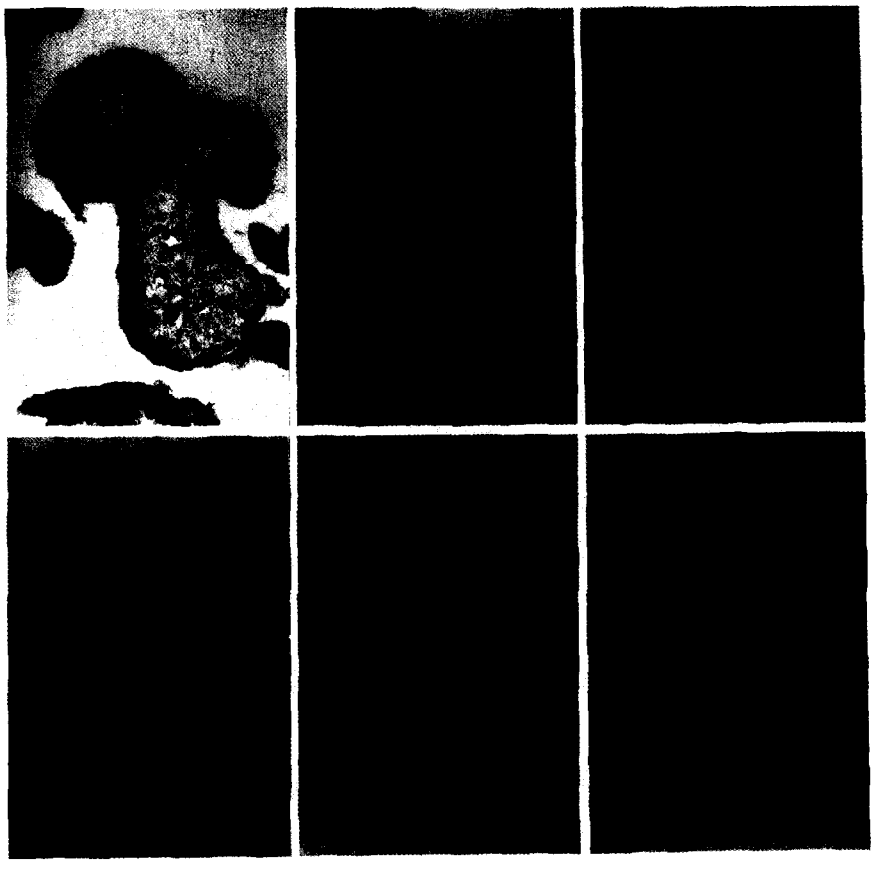

Fig. 2. Immunohistochemical analysis of trisomy 22 placenta obtained at 10 weeks of gestation (× 100), where A = Intact hCG, B = α-glycoprotein, C = free β-hCG, D = PAPP-A, E = hPL, F = SP1.

When considered in conjunction with prolonged marker doubling rates, these findings suggest the feto-placental unit was compromised from 8 weeks, or 1 week prior to ultrasound confirmation of pregnancy failure. This is consistent with earlier reports where depressed serum PAPP-A levels were detected, in ultrasonographically normal pregnancies, many weeks prior to eventual pregnancy demise [7].

By immunohistochemistry, the cellular distribution of PAPP-A, α-glycoprotein, total β-hCG, SP1 and hPL was limited to the syncytiotrophoblast layer of the placental villi. In the trisomy 22 placenta, PAPP-A, α-glycoprotein, SP1 and hPL immunoreactivity was visibly reduced. The most striking reduction was observed for PAPP-A. These findings were consistent with depressed serum marker levels. This is the first report detailing trohpoblast antigen expression by aneuploid trophoblast cells and supports earlier data suggesting PAPP-A may be a good indicator of trophoblast and, hence, pregnancy viability [4].

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