Introduction: 11β-Hydroxysteroid dehydrogenase activity is responsible for the interconversion of cortisol and cortisone. Its presence in granulosa-lutein (GL) cells is associated with a poor outcome in IVF (Michael *et al.*, 1993). Recently we have developed a simple rapid (<8 h) method of assessing the reductase activity of the type 1 isoform in GL cells (Knaggs *et al.*, 1996). In this pilot study we assayed the enzyme's activity in all the individual follicles aspirated from patients undergoing IVF.

Materials and methods: Cells from individual follicular aspirates (n = 107 from 20 patients) were plated (12 500 cells/well) into 96-well plates and challenged with cortisone (6×10^{-6} M) for 2 h at 37°C within 1 h of aspiration. Cortisol secretion was measured by a radioimmunoassay (intra- and interassay coefficients of variation <10 and <14% respectively). The DNA content of each well was assessed by a Hoescht fluorescent dye binding assay.

Results: The intra-assay coefficient of variation for the reductase activity, based on 107 cell preparations each assayed in quadruplicate, was 13%. It varied considerably between follicles both within and between patients (range 0.001–0.560 nmol cortisol/μg DNA). Enzymatic activity was [median (range)] 0.04 (0.0012–0.14) nmol cortisol/μg DNA in the follicles (n = 22) from the four patients who became pregnant compared with 0.05 (0.002–0.56; n = 85 follicles from 16 patients) nmol cortisol/μg DNA in those who did not conceive. Follicles which gave rise to oocytes leading to a pregnancy cycle had levels of 0.05 (0.001–0.08; n = 10) as opposed to 0.04 (0.002–0.33; n = 31) nmol cortisol/μg DNA for cells from follicles that produced a transferred embryo which failed to result in a pregnancy. However, there were no differences between the groups because of the small number of patients who were infertile for various reasons.

Conclusion: This method is suitable for assessing enzymatic activity in GL cells isolated from follicles aspirated from patients undergoing IVF. This work on a series of 20 patients forms part of a prospective study to assess whether 11β-reductase activity in GL cells from individual follicles may be used as a predictor of IVF success. At present the low patient numbers preclude a meaningful correlation between 11β-reductase activity and pregnancy rate.

References:

Knaggs *et al.* (1996) *Hum. Reprod.*, **11,** 151.
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P-220. Significance of non-placental pregnancy-specific β_1-glycoprotein in the serum of IVF cycles before and after embryo transfer

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Introduction: Non-placental pregnancy-specific β_1-glycoprotein (SP1) is a monomeric, heavily glycosylated (29%) protein of syncytiotrophoblastic origin. Its serum concentration rises steeply in early pregnancy, allowing a dating superior to that of ultrasound before 6 weeks of gestation. It appears 1–3 days after HCG, a small but significant delay which has prevented its use in IVF programmes. With a sensitive assay, small SP1 signals were recorded at around the time of oocyte retrieval and embryo transfer, i.e. at a time incompatible with a trophoblastic source. Previous observations have shown that this 'abnormal SP1 pattern' occurred in all cycles of a defined group of non-pregnant patients. The aim of this study was to retrospectively analyse SP1 versus outcome in a larger group of patients.

Materials and methods: The study included 182 consecutive conventional IVF and embryo transfer treatment cycles. Standardized serum collection was performed on day 2, before the initiation of ovarian stimulation on oocyte retrieval day −1 (OPU−1) and on embryo transfer day +14 (ET+14). Of these cycles, 45 gave rise to either a spontaneous early abortion (n = 8) or an ongoing pregnancy (n = 37). SP1 was determined using an ultrasensitive (0.02 ng/ml) microplate ELISA method developed in our laboratory.

Results: A longitudinal (paired) analysis showed no difference in serum SP1 between days 2 and OPU−1 in pregnant and non-pregnant groups. On the other hand, the concentrations at ET+14 were significantly higher (200%) than at OPU−1; again this increase did not depend on outcome and was observed consistently, even in the implantation failure group. A cross-sectional analysis showed no significant differences between outcome groups at any of the three selected time points. Nevertheless, a frequency analysis indicated that at days 2 and OPU−1 the cycles of subsequently pregnant women were homogenously distributed, while those not leading to pregnancy were scattered over a wider range. At ET+14 there was less homogeneity in both groups.

Conclusion: The SP1 frequency distribution patterns observed on days 2 and OPU−1 suggest the presence of a subpopulation of women with a 'high' serum SP1 concentration and a reduced pregnancy prognosis because none of these women became pregnant. An endometrial factor or other endocrine effect may play a role in influencing ectopic SP1 production as well as embryo implantation. This should be investigated further to identify low-chance patients. At ET+14 such an analysis carries no advantage because the pregnancy subgroup also contains women with sera with high SP1 concentrations, probably because of early or multiple implantations, and with low SP1, probably because of poor embryo quality or delayed implantation. From a clinical point of view, HCG remains superior to SP1 as an indicator of pregnancy after embryo transfer, even when HCG is used as luteal phase support.

P-221. Subclinical pregnancy loss associated with pelvic endometriosis and infectious disease

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Introduction: Subclinical pregnancy loss has been documented in normal and infertile women. We have previously reported