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Relaxin concentrations in exoembryonic fluids during the first trimester

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In 15 women undergoing therapeutic termination of pregnancy (8–13 weeks), the median concentration of relaxin was 1000 ng/l in maternal serum, 122 ng/l in coelomic fluid and 9 ng/l in amniotic fluid. Its presence in coelomic fluid suggests that relaxin may be present in the fetal circulation and thus be able to influence embryonic development during the period of organogenesis.

Key words: abortion/amniotic fluid/embryonic development/first trimester/relaxin

Introduction

Relaxin has been suggested to have a role in embryopathy (Edwards and Newall, 1988; Uden and Lindhagen, 1988). Although no direct evidence for such a role exists, the higher concentrations of relaxin in the sera of diabetic pregnancies have been suggested to support this hypothesis (Steinetz *et al.*, 1992). During pregnancy circulating relaxin is derived exclusively from the corpus luteum of pregnancy (Johnson *et al.*, 1991), and has been shown to be present in amniotic fluid in increasing concentrations between 10 and 14 weeks gestation, but not to be present in the fetal circulation from 19 weeks gestation onwards (Johnson *et al.*, 1992). In order to have a causative role in embryopathy, relaxin would have to be present either in the fetal circulation or in the amniotic fluid during the period of organogenesis. Obtaining fetal blood from this period of pregnancy is not possible, although the coelomic fluid, which may be expected to give an approximation of the maternal contribution to the fetal circulation, may be obtained. Thus, in order to examine the hypothesis that relaxin may affect embryo development, we have studied relaxin concentrations in maternal serum, the coelomic fluid and amniotic fluids.

Materials and methods

In 15 women undergoing therapeutic termination of pregnancy at 8–13 weeks gestation, maternal blood was obtained prior to the procedure, and samples of coelomic fluid and amniotic fluid were obtained via two needles sited under ultrasound control, as described previously (Jauniaux *et al.*, 1991). The project was approved by the Ethics Committee of King's College Hospital.

The presence of correlations betweeen (i) the levels of relaxin in maternal serum, coelomic fluid and amniotic fluid, and (ii) gestational age and the levels of relaxin in maternal serum, coelomic fluid and amniotic fluid were investigated with a simple regression analysis.

Assay

Concentrations of relaxin in serum, coelomic fluid and amniotic fluid were measured using an enzyme-linked immunosorbent assay (ELISA) as described previously (Ferrailo *et al.*, 1991). The standards (short relaxin), from 1 to 1250 ng/l, were prepared in pooled normal male serum. The working range of the assay was between 35 and 1100 ng/l. The cross-reactivity with insulin, nerve growth factor and porcine relaxin were $<0.1\%$. The use of the ELISA for amniotic fluid samples has been validated previously (Johnson *et al.*, 1992), and a standard curve made up in coelomic fluid stripped of endogenous relaxin diluted in parallel with that made up in serum (Figure 1).

Results

The median relaxin concentration was 1000 ng/l in maternal serum, 122 ng/l in coelomic fluid and 9 ng/l in amniotic fluid (Figure 2). There appeared to be a weak correlation between

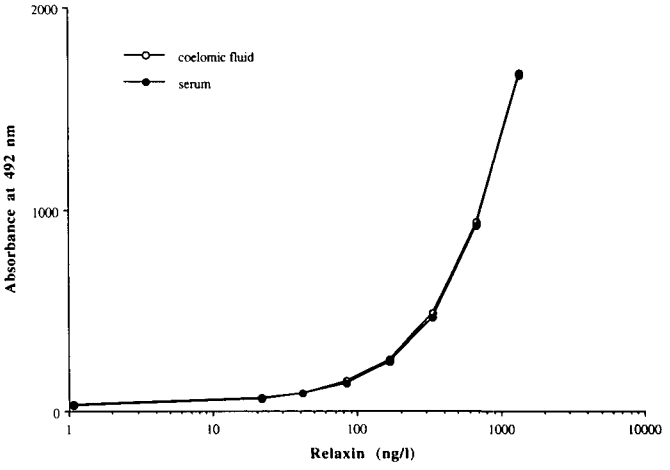

Fig. 1. Dilution curves of synthetic relaxin in serum and coelomic fluid, the latter stripped of its endogenous relaxin, measured by an enzyme-linked immunosorbent assay, read at a wavelength of 492 nm.

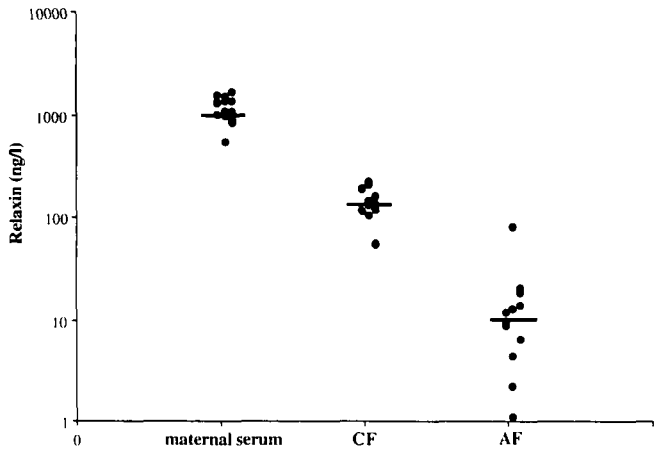

Fig. 2. The log concentration of relaxin in maternal serum, coelomic fluid (CF) and amniotic fluid (AF) in 15 normal pregnancies between 8 and 11 weeks gestation. The horizontal bar represents the median value.

relaxin concentrations in maternal serum and coelomic fluid ($r = 0.41$) but this was not significant ($P = 0.13$). There were no associations between coelomic fluid and amniotic fluid concentrations of relaxin and gestational age. However, a significant negative correlation was observed between gestational age and serum relaxin concentrations ($r = -0.58$, $P = 0.022$). This observation was probably due to the small size and limited period of gestation studied, as circulating plasma relaxin concentrations have been shown by several authors to rise to a peak at the end of the first trimester and to decline thereafter (Johnson *et al.*, 1991).

Discussion

While amniotic fluid is produced mainly by the embryo, coelomic fluid is derived from the maternal circulation, by transplacental diffusion, with contributions from the placenta and the embryo via its secondary yolk sac (Jauniaux *et al.*, 1991). Therefore, relaxin present in the coelomic fluid may be derived from the maternal circulation exclusively, or may be augmented by a contribution from the developing placenta, which is known to be able to synthesize relaxin (Sakbun *et al.*, 1990), but not to contribute to the maternal circulating pool (Johnson *et al.*, 1991).

The low concentrations of relaxin in amniotic fluid support our earlier conclusion that relaxin is unlikely to be embryopathic (Johnson *et al.*, 1992). However, the presence of relaxin in the coelomic fluid suggests that it is able to diffuse across the placenta into the circulation of the embryo. Alternatively, or in addition to this, relaxin may be absorbed from the coelomic fluid via the secondary yolk sac into the embryonic circulation. The present findings suggest that maternal relaxin is available to the embryo from as early as 8 weeks gestation and could play a role in embryogenesis and organogenesis.

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