Effect of oestrogen on fetal survival in the rabbit

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Summary. Pregnant rabbits (Day 18) were treated i.m. for 3 days with 10 µg oestradiol-17β/kg. The number of dead fetuses was significantly greater than in the control animals but corpus luteum weight and peripheral progesterone concentrations were not affected. Fetal death was therefore not due to luteolysis.

Introduction

High doses of oestrogens given to the pregnant rabbit after Day 12 of gestation produce fetal death (Reynaud, 1934), and so we investigated whether this lethal effect of oestrogen was caused by luteolysis.

Materials and Methods

New Zealand White rabbits weighing 3–4 kg each were kept under standard conditions of light and temperature. They were mated with bucks of known fertility and the day of mating was designated Day 0.

Starting on Day 18, 10 µg oestradiol-17β/kg were injected intramuscularly into 7 rabbits for 3 days. The oestradiol (Progyonon: Schering) was made up in isotonic saline solution (0.154 M-NaCl). The 6 control rabbits received the vehicle only. On Day 20, all the rabbits were anaesthetized with sodium pentobarbitone. Blood was withdrawn from each ovarian vein, heparinized, and pooled for each rabbit according to treatment. Blood samples were also taken from the femoral vein. The blood was immediately centrifuged and the plasma was frozen at –20°C until assayed for progesterone. The ovaries were removed and weighed. The fetuses and placentas were removed and counted and the number of dead fetuses noted.

Progesterone concentrations were determined by the radioimmunoassay of Thorneycroft & Stone (1972). The antiserum was raised in rabbits to an 11α-succinyl-hydroxyprogesterone-bovine serum albumin conjugate. The cross-reactivity, measured and calculated as described by Thorneycroft, Tillson, Abraham, Scaramuzzi & Caldwell (1970), was 0.7% for 20α-dihydroprogesterone, 0.9% for 17α-hydroxyprogesterone, and 0.2% for pregnenolone. The labelled progesterone was from New England Nuclear (Net-208), and the progesterone standard was from Sigma; gelatin-phosphate buffer, pH 7.2, and dextran-coated charcoal were used for the assay procedure. The recovery rate was 85–90%. The intra- and inter-assay variations were 5 and 12% respectively. The sensitivity was 5 ng/ml.

The significance of the results was assessed by Student's *t* test and a value of *P* < 0.05 was taken as significant.

Results

The results are shown in Table 1. The number of dead fetuses in the oestradiol-treated rabbits was significantly higher than in the control animals. All the dead fetuses were macerated and smaller in size than the live ones. Their placentas appeared to be normal.

Table 1. The effect (mean $\pm$ s.d.) of oestradiol-17 β on various factors in pregnant rabbits

	Control rabbits (6)	Oestradiol-treated rabbits (7)
No. of fetuses/litter	10.5 $\pm$ 3.1	8.4 $\pm$ 3.2
No. of dead fetuses/litter	0.2 $\pm$ 0.4	6.1 $\pm$ 1.6*
No. of live fetuses/litter	10.3 $\pm$ 2.9	2.3 $\pm$ 0.3*
Weight of each live fetus (g)	3.1 $\pm$ 1.0	2.7 $\pm$ 0.1
Weight of each dead fetus (g)	—†	1.9 $\pm$ 0.4
Weight of placenta/live fetus (g)	3.2 $\pm$ 1.1	3.2 $\pm$ 0.6
Weight of placenta/dead fetus (g)	—†	2.7 $\pm$ 0.4
Weight of ovaries (g/pair)	1.16 $\pm$ 0.3	1.02 $\pm$ 0.4
No. of CL/2 ovaries	12.3 $\pm$ 1.4	10.3 $\pm$ 3.1
Progesterone conc. (ng/ml)		
Ovarian vein	330.3 $\pm$ 153.0	398.5 $\pm$ 220.0
Femoral vein	8.25 $\pm$ 3.51	5.57 $\pm$ 1.50

* Significantly different from control values $P < 0.001$.

† One fetus only.

Discussion

The present study confirms that a high dose of oestradiol-17 β has an adverse effect on fetal survival in rabbits. The progesterone concentrations and the similarity of luteal and ovarian weight in the treated and untreated rabbits show that there was no decline in the function of the corpus luteum.

The present results therefore support the suggestion of Makler & Morris (1971) that the detrimental effects of oestrogen on rabbit embryos is not due to luteolysis. This conclusion is not unexpected because it is known that oestrogens are luteotrophic in the rabbit (Keyes & Nalbandov, 1967; Armstrong, Jackanicz & Keyes, 1969; Hilliard, Saldarini, Spies & Sawyer, 1971). They are essential for the maintenance of secretion of progesterone by the corpus luteum, and progesterone is necessary to support gestation in this species. Small doses of oestradiol can maintain pregnancy in rabbits after hypophysectomy (Robson, 1937, 1939; Greep, 1941), treatment with antiserum to LH (Spies & Quadri, 1967), or the destruction of the follicles by X-ray irradiation (Keyes & Nalbandov, 1967). The ability of oestrogens to stimulate progesterone formation in luteal tissue *in vitro* has also been demonstrated (Fuller & Hansel, 1971). There are various possible mechanisms whereby oestrogen could exert its effect, e.g. by affecting prostaglandin and oxytocin secretion and hence myometrial activity (Blatchley *et al.*, 1971; Bedford, Challis, Harrison & Heap, 1972), by decreasing blood flow in the maternal placenta (Abdul-Karim & Bruce, 1972), or by affecting metabolic processes (Jacob, Lal & Sharma, 1975).

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