THE CONTRACEPTIVE ACTION OF IMPEDING OESTROGENS

IV. EFFECT OF OESTRADIOL ON THE ANTI-IMPLANTATIONAL ACTION OF OESTRIOL IN THE RAT

H. H. WOTIZ, S. SMITH, B. SHAPIRO AND A. SCUBLINSKY

*Department of Biochemistry, Boston University School of Medicine,
Boston University Medical Center, Boston, Massachusetts, U.S.A.*

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The unique function of oestriol as a regulatory agent was suggested by the experiments of Hisaw, Velardo & Goolsby as early as 1954. This biological antagonism was further demonstrated by Wicks & Segal (1956) and a biochemical explanation of the events was suggested by Brecher & Wotiz (1967), who recently demonstrated the competition between certain weakly oestrogenic substances and oestradiol for cytoplasmic end-organ receptor molecules.

Recent reports from this laboratory have indicated the special position and behaviour of oestriol and related substances, designated as 'impeding oestrogens', with regard to their contraceptive activity (Scublinsky & Wotiz, 1971; Wotiz & Scublinsky, 1971).

It was suggested that the major effect of oestriol was anti-implantational since it was reasonable to rule out inhibition of ovulation at the dosage used. Interference with sperm capacitation was also eliminated in view of the post-coital effect of oestriol. Further evidence for this mechanism may be derived from the effect of small doses of oestradiol on the oestriol-treated animals.

Forty-one Sprague-Dawley rats (200 to 250 g) were divided into five groups kept under 12 hr light/12 hr dark cycles and allowed unrestricted feeding. Steroids were dissolved in sesame oil, after their purity (99.99%) was ascertained by methods previously reported (Scublinsky & Wotiz, 1971). Daily injections of 12.5 µg oestriol were given from the day spermatozoa were found in the vaginal smears and were continued for 3 days. On the 4th day, varying doses of oestradiol were injected. On the 10th day after mating, the animals were killed and the uteri were examined for blastocysts. In one experiment (Exp. 5), oestriol was injected daily for 4 days to eight rats and on the last day, 0.3 µg of oestradiol was injected with the oestriol.

The results obtained when doses of oestradiol varying from 0 to 0.3 µg were injected into animals pre-treated with oestriol are recorded in Table 1.

As reported previously (Wotiz & Scublinsky, 1971), administration of 12.5 µg of oestriol for 3 days resulted in a 75% decrease of pregnant animals. All the control animals became pregnant. Administration of 0.005 or 0.01 µg of oestradiol to the oestriol-treated animals produced no change in pregnancies. However, a dose of 0.05 µg of oestradiol resulted in a pregnancy rate of 78%. It should

be noted that although seven out of nine animals became pregnant, there was a reduction in the number of implants/uterine horn and in three of the seven pregnant rats, all the implants were in the same horn. The significance of this observation is obscure and one may wonder if there is a difference between the two horns of the rat in the levels of perfusing oestriol or the ability to bind the hormone. Interestingly, in the duiker (*Cephalophus*), implantation occurs only in the right uterine horn, despite the fact that both ovaries ovulate. Administration of an even larger dose of oestradiol (0.3 μ g) resulted in a reversal of the anti-inhibitory effect noted with 0.05 μ g of this steroid.

TABLE 1
EFFECTS OF VARYING DOSES OF OESTRADIOL ON
OESTRIOL INHIBITION OF IMPLANTATION

Group	Oestriol* (μ g)	Oestradiol (μ g)	No. of pregnancies	Average no. of implantations
1	12.5	0	2/8	1.25†
2	12.5	0.005	2/8	1.0†
3	12.5	0.01	2/8	1.0†
4	12.5	0.05	7/9	3.7‡
5	12.5§	0.3	0/8	0
6	0	0	8/8	8.5

* Oestriol was injected for 3 days following proven mating.

† Seven to nine blastocysts in the two animals with implants.

‡ In three animals, all the implants were in the same uterine horn.

§ Oestriol was administered for 4 days, oestradiol was co-administered on the 4th day.

The fact that oestradiol counteracts the inhibiting (anti-implantational) activity of oestriol is further evidence for the anti-oestrogenic activity of oestriol. Oestriol prevents implantation by competing for endometrial cytoplasmic receptors, depleting them and thus making them unavailable to oestradiol and the subsequent protein synthesis, considered necessary to successful implantation. Administration of specific small doses of oestradiol re-establishes the dominance of oestradiol, removes the antagonistic effect of oestriol and implantation occurs. Excess oestradiol (0.3 μ g) was found to be ineffective, most likely because, combined with endogenous oestradiol, it became anti-implantational. This can be calculated from the work of Desaulles & Krähenbühl (1964), who showed that, at dose levels from about 0.3 μ g/200g animal/day, oestradiol will inhibit implantation.

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