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# INDOMETHACIN, KETOPROFEN AND CORPUS LUTEUM REGRESSION IN THE GUINEA-PIG

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1 The accelerated regression of the corpora lutea normally produced by glass beads inserted unilaterally into the uterus of the guinea-pig early in the oestrous cycle was prevented when the beads were filled with indomethacin or ketoprofen.

2 Luteal regression was enhanced by the placement of beads in both horns of the uterus. When empty beads were placed in one horn of the uterus and drug-filled beads in the other, indomethacin blocked regression in both ovaries whereas ketoprofen was ineffective.

3 Indomethacin probably blocks luteal regression by inhibition of prostaglandin synthesis in the uterus.

## Introduction

When glass beads are placed in the uterus of the guinea-pig early in the oestrous cycle regression of the corpora lutea is accelerated (Bland & Donovan, 1966). There is evidence that release of prostaglandin $F_{2\alpha}$ from the uterus late in the cycle brings about regression of corpora lutea and it seems likely that foreign bodies placed in the uterine lumen enhance the release of this compound (Poyser, Horton, Thompson & Los, 1970, 1971; Blatchley, Donovan, Horton & Poyser, 1972). In view of the reports that anti-inflammatory drugs such as aspirin or indomethacin inhibit prostaglandin synthetases (Vane, 1971), it was of interest to determine whether the luteolytic action of glass beads placed in the uterus could be blocked by indomethacin and ketoprofen, a chemically unrelated anti-inflammatory drug which inhibits prostaglandin synthetases. To ensure a high local concentration of drug the beads were packed with the compounds under test.

## Methods

Adult guinea-pigs (475-1150 g) were used. The oestrous cycle was followed by daily inspection of the vagina and the collection of vaginal smears on those days when the vagina was open. Day 1 of the oestrous cycle was taken as the day when maximal cornification of cells was apparent before the post-ovulatory influx of leucocytes.

Glass beads were inserted into the uterus on days 3 or 4 as described previously (Donovan & Traczyk, 1962). The beads were cylindrical in shape, measuring approximately 7 x 3 mm and were sealed at one end. Immediately before insertion some beads were filled with drug by tamping. The surface of the packed down drug powder was thus accessible for solution in endometrial secretion through a caudally located orifice approximately 2 mm in diameter. The amount of indomethacin or ketoprofen packed into each bead was approximately 15 mg.

The drugs used were indomethacin (Merck, Sharp & Dohme, Ltd.) and ketoprofen (May & Baker, Ltd.). In some experiments two beads filled with drug were placed in one uterine horn, and the other left undisturbed; in parallel experiments two beads filled with drug were placed in one horn and two empty beads in the other. In this way each animal could serve as its own control. The insertion of drug laden beads was alternated between the right and left uterine horns in the guinea-pigs subjected to each mode of treatment. For control purposes two empty glass beads were inserted into each uterine horn in a further group of animals.

All animals were killed on day 12 of the oestrous cycle, eight or nine days after operation, when the genital tract was removed and the two ovaries fixed separately. Serial sections of the ovaries were prepared and the sizes of the corpora lutea determined from measurement of two diameters at right angles in the plane of the largest section of each corpus luteum and multiplication of the product by the number and thickness of the sections. Animals in which corpora lutea were present in only one ovary were discarded, and

sufficient animals prepared to bring the number of guinea-pigs in each group to ten. When the uteri were dissected some time after the experiments had been completed two empty beads were found to have been extruded from one uterine horn in an animal in which beads containing indomethacin were present on the opposite side. This group thus comprised nine animals.

## Results

Luteal regression was inhibited when glass beads were filled with indomethacin before insertion into the uterus, when compared with the effect of empty beads (Table 1). With indomethacin present, there was no significant difference between the luteal volumes in the ovaries on the 'distended' or control sides.

Although the corpora lutea were also smaller on the treated side when ketoprofen-filled beads were inserted, the degree of luteal regression was not statistically significant ($P > 0.1$). Thus both drugs blocked luteal regression.

When two empty beads were inserted into each horn of the uterus, luteal regression was potentiated in both ovaries (Table 1). The mean luteal size following bilateral introduction of beads was 1.08 in the left ovaries compared with a figure of 2.05 for the corpora lutea on the treated side following unilateral insertion of beads. This difference is significant ($P < 0.01$).

Luteal regression was inhibited bilaterally when the beads inserted into one horn of the uterus were filled with indomethacin. Mean corpus luteum size on the treated side (2.95) was not significantly different from that of the control side (2.53), but considerably greater than that of animals provided with empty beads (2.95 *versus* 1.08; $P < 0.001$). The mean luteal size in the control ovaries of the indomethacin-treated animals (2.53) was also significantly greater than that for the ovaries (right, 1.07; left, 1.08) of the animals fitted only with empty beads ($P < 0.01$).

Ketoprofen did not inhibit luteal regression on the treatment side when beads were inserted bilaterally, the corpora lutea being similarly small in both ovaries (Table 1). The luteal bodies in the ovaries on the control or treated side of the animals given ketoprofen were not significantly larger than those of animals provided with empty beads in each horn ($P > 0.3$).

The total amount of indomethacin or ketoprofen contained in the beads in a uterine horn varied between 20 and 42 mg. When the uteri were dissected after preservation in formalin solution the beads packed with indomethacin were found still to contain substantial amounts of the drug, whilst the control bead and those originally filled with ketoprofen contained only fluid and a protein coagulum.

Macroscopic amounts of decidual tissue were infrequent after bead insertion, and the incidence of decidua was not altered by the presence of indomethacin or ketoprofen.

## Discussion

Luteal regression ensued in the associated ovary when glass beads were inserted into one uterine

**Table 1** The effect of indomethacin and of ketoprofen upon the luteal regression induced by the insertion of glass beads into the uterus of the guinea-pig

| | *Mean size of corpora lutea in the ovary associated with* | | | |
|---|---|---|---|---|
| | *Control horn* | *Treated horn* | *Mean difference* | *Significance of difference*† |
| Unilateral bead insertion | | | | |
| Empty beads* | 3.74 ± 0.33 | 2.05 ± 0.33 | 1.69 | $P < 0.001$ |
| Indomethacin-filled beads | 3.90 ± 0.28 | 3.45 ± 0.29 | 0.45 | $0.2 > P > 0.1$ |
| Ketoprofen-filled beads | 3.54 ± 0.26 | 2.71 ± 0.47 | 0.83 | $0.2 > P > 0.1$ |
| Bilateral bead insertion | | | | |
| Empty beads in each horn | 1.07 ± 0.15 | 1.08 ± 0.13 | 0.01 | $P > 0.9$ |
| Indomethacin-filled beads in treated horn | 2.53 ± 0.52 | 2.95 ± 0.54 | 0.42 | $P > 0.5$ |
| Ketoprofen-filled beads in treated horn | 1.22 ± 0.38 | 1.30 ± 0.16 | 0.08 | $P > 0.6$ |

* From Bland & Donovan, 1970.
† Paired *t* tests.

horn of the guinea-pig, but the degree of regression was greater when beads were introduced bilaterally. This observation indicates that the luteolytic factor produced in one uterine horn, while primarily influencing luteal function in the neighbouring ovary, can also affect the opposite gonad. Both indomethacin and ketoprofen could block the luteolytic response to unilateral insertion of glass beads, and these findings are in accord with the concept that luteal regression is mediated by the release of prostaglandin from the uterus.

Indomethacin also inhibited luteal regression on the control side when foreign bodies were placed in both uterine horns. Thus it appears that sufficient indomethacin was released from the beads in one horn to affect prostaglandin synthesis in the whole of the uterus, despite the poor solubility of the drug. By contrast, ketoprofen did not influence luteal regression following the bilateral insertion of beads into the uterus. This finding could reflect a lesser potency of the drug in this experimental situation, particularly as ketoprofen was active when luteolysis was induced on one side only. No information is available concerning the rate, or time course, of release of indomethacin or ketoprofen from the interior of the beads. At autopsy, eight or nine days after insertion, solid ketoprofen was lacking inside the beads, but the drug content of the fluid present was not determined.

Indomethacin has extended the oestrous cycle of the guinea-pig when given systemically (Marley, 1972; Poyser, 1973), or by the intrauterine route contained in silicone rubber (Marley, 1973) or paraffin wax (Poyser, 1973; Horton & Poyser, 1973). Neither control cylindrical lengths of silicone rubber nor implants of paraffin wax caused a shortening of the oestrous cycle so that the endogenous production of prostaglandin appears not to have been enhanced by inert foreign bodies in these experiments. The marked lengthening of the oestrous cycle with indomethacin observed by Horton & Poyser (1973) was accompanied by enlargement of the corpora lutea in the ovaries and the sustained secretion of progesterone.

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