A positive feedback effect of oestradiol on LH release in the male marmoset monkey, *Callithrix jacchus*

J. K. Hodges and J. P. Hearn

M.R.C. Unit of Reproductive Biology, 2 Forrest Road, Edinburgh EH1 2QW, U.K.

Summary. Subcutaneous injections of oestradiol benzoate in oil, resulting in a sustained elevation of circulating oestradiol levels, induced an initial suppression of LH secretion, followed by a positive discharge of LH in castrated male and female and in intact male marmosets. Oestrogen-induced LH release (producing maximum LH concentrations 24 h after the injection) was observed in 75% of castrated males and females. A positive discharge of LH occurred in 50% of intact males 28–36 h after oestrogen administration.

The experimental production of a preovulatory type of LH discharge after oestradiol administration (positive feedback) in intact and ovariectomized females has been well documented for rats (Caligaris, Astrada & Taleisnik, 1971; Brown-Grant, 1974), sheep (Scaramuzzi, Tillson, Thorneycroft & Caldwell, 1971; Martensz, Baird, Scaramuzzi & Van Look, 1976), rhesus monkeys (Yamaji *et al.*, 1971; Karsch *et al.*, 1973) and man (Monroe, Jaffe & Midgley, 1972). However, it has also been shown that there is no positive feedback of LH in response to oestrogen in intact or castrated males of these species (Yamaji *et al.*, 1971; Short, 1974; Karsch & Foster, 1975; Van Look, 1976). This difference in responsiveness to oestrogen suggests that the ability to display positive feedback represents a fundamental difference in the hypothalamus of males and females. There is evidence to suggest that the male rhesus monkey (Karsch, Dierschke & Knobil, 1973; Steiner, Clifton, Spies & Resko, 1976) and men (Dörner, Rohde, Stahl, Krell & Masius, 1975; Kulin & Reiter, 1976) may differ from males of other species in showing positive feedback, although the data are inconclusive. The present communication reports a clear positive feedback response to oestradiol in both male and female marmoset monkeys.

Materials and Methods

Sexually mature marmosets were bilaterally gonadectomized at least 3 months before the study. Four males and four females were given 35 µg oestradiol benzoate (Organon Laboratories Ltd) as a single subcutaneous injection in arachis oil, and then bled at 4-h intervals for 36 h. Ten adult intact males received similar treatment, and were bled 0, 8, 24, 28, 36 and 48 h after oestradiol administration. As controls, 6 castrated males, 6 spayed females and 6 normal males received a single injection of arachis oil and were bled after 0, 8, 20, 24, 28, 36 and 48 h. Blood samples were collected from the femoral vein, centrifuged immediately, and the plasma stored at –20°C. Plasma oestradiol concentrations were measured by radioimmunoassay (Baird, Swanston & Scaramuzzi, 1976), LH concentrations were determined with a heterologous double-antibody radioimmunoassay (Welschen *et al.*, 1975) which was validated for marmoset LH by testing parallelism between dilution curves of marmoset plasma and a rat LH standard, and for cross-reaction with various pituitary hormones (Hodges, 1978). Biological validation was also obtained by measuring LH levels during the normal cycle, after LH-RH treatment, after immunization of male and female marmosets against LH-RH (Hodges & Hearn, 1977), and following gonadectomy. The detection limit of the LH assay was 20 ng NIAMDD-LH-RP1/ml plasma.

Results

The LH responses to a single injection of oestradiol benzoate in castrated animals are shown in Text-fig. 1. The oestrogen injection produced a sustained rise in peripheral plasma oestradiol concentrations to give levels similar to those (0.8–1.5 ng/ml) found in intact females immediately before

Text-fig. 1. Plasma LH and oestradiol concentrations in (a) male and (b) female gonadectomized marmosets after a single subcutaneous injection of 35 μ g oestradiol benzoate in oil (arrow). Oestradiol values are mean $\pm$ s.e.m. Individual results for LH levels in 4 males and 4 females are shown. The broken lines represent the limits of 2 standard deviations above and below the mean control LH levels in 6 males or 6 females.

the spontaneous LH surge in the normal cycle (Hearn & Lunn, 1975). As plasma oestradiol concentrations rose, LH levels initially fell (in all animals) and then increased abruptly (in 3 males and 3 females) between 12 and 24 h after the oestrogen injection. The initial decline in plasma LH levels reflects the negative feedback action of oestradiol which characteristically precedes the initiation of the oestrogen induced LH surge in the rhesus monkey. The LH surge occurs earlier in the marmoset than in the rhesus monkey, in which maximum LH concentrations were found between 36 and 48 h after the oestradiol benzoate injection (Yamaji *et al.*, 1971; Karsch *et al.*, 1973; Steiner *et al.*, 1976). In the present study positive and negative feedback has been defined as an increase or decrease respectively in circulating LH to levels exceeding 2 standard deviations from mean control values. According to this definition negative feedback was observed in all animals at 4–12 h after the oestrogen injection while positive feedback was seen in 75% of animals of each sex, with maximum LH levels occurring after 24 h. There was an increase in LH secretion following the initial negative feedback response in the remaining 2 animals, but it was not sufficient to fulfil the criterion given above.

In the intact males the LH response to oestradiol was much more varied than in castrated animals, and only 5 out of 10 responded with an increase in LH levels which could be classified as positive feedback (Text-fig. 2). In the other 5 animals the maximum LH levels remained within the defined control limits. In the animals that showed positive feedback maximum LH levels occurred 28–36 h after the oestradiol injection, slightly later than in the castrated animals. A decrease in LH secretion was observed 8 h after oestrogen administration in 8 monkeys but could be classified as a negative feedback decrease in only 4 animals. In 2 animals LH levels were undetectable before and 8 h after the

Text-fig. 2. Plasma LH and oestradiol concentrations in 10 intact male marmosets after a single subcutaneous injection of 35 µg oestradiol benzoate in oil (arrow). Individual results for LH levels are shown in two graphs. The broken lines represent the limits of 2 standard deviations above and below the mean control LH levels in 6 intact males. Oestradiol values are mean $\pm$ s.e.m.

oestrogen injection and any negative feedback action could not be observed. The injection of oestradiol benzoate produced an increase in peripheral plasma oestradiol concentrations similar to that seen in castrated animals.

Discussion

These results indicate that castrated male marmosets will release LH in response to oestrogen treatment in a manner not unlike that observed in spayed females. Furthermore, they also suggest a similar capacity in some intact males. Initial attempts (Yamaji *et al.*, 1971) to obtain a female-like gonadotrophin surge in castrated male rhesus monkeys in response to a single oestradiol challenge were unsuccessful, and it was only later (Karsch *et al.*, 1973), when gonadotrophin levels in castrates were chronically suppressed with oestrogen before the oestradiol benzoate injection, that such a response was obtained. In such a situation the fact that the animal has been so far removed from its normal endocrine state may make interpretation of the results difficult. It has been claimed that oestrogen may be capable of inducing LH release in normal and homosexual men (Dörner *et al.*, 1975; Kulin & Reiter, 1976), but the gonadotrophin rises that were described are comparable in neither magnitude nor duration to those seen in normal women.

From the present study it is difficult to compare the response to oestradiol stimulation in intact and castrated animals, because the incidence of positive feedback (50%) was considerably lower in intact males than in either male or female castrates (75%). Furthermore, the small numbers of animals used necessitates cautious interpretation of the results. Nevertheless, the data provide evidence that castrated and intact male marmosets can release LH in response to oestradiol stimulation. A similar response to oestrogen in intact males of other primate species has yet to be determined.

Positive feedback in primates and many other mammalian species was, for a long time, considered to be exclusively a female characteristic, although its adaptive significance is not obvious. Positive feedback is necessary for ovulation to occur, but the persistence of the response in the male would not interfere with normal testicular function. During the course of evolution, centres regulating positive feedback in the male may have developed differently in marmosets than in rats or sheep.

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