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MATURATION OF THE CONTROL OF GONADOTROPIN AND PROLACTIN RELEASE IN THE RAT

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SUMMARY

Several aspects of the maturation of the control of pituitary gonadotropin and prolactin secretion in the rat have been studied. Circulating estrogen (E_2) titers were found to be elevated in the infantile female, declining as the animal approached maturity. These high E_2 levels appear to be due to an enhanced E_2 production by the ovaries and adrenals and to a reduced metabolic degradation of the steroid. High plasma FSH titers in the presence of elevated E_2 levels in infantile rats were found to be caused at least in part by a decreased effectiveness of the estrogen negative feedback and by an increased pituitary responsiveness to LH-RH induced by a direct action of progesterone and 5α-dihydrotestosterone on the gland. Negative estrogen feedback controlling LH secretion was also decreased in infantile rats and pituitary responsiveness to LH-RH enhanced. This latter phenomenon, however, was not altered by ovariectomy-adrenalectomy or by steroid(s) replacement therapy. The decline in plasma gonadotropins observed after day 15 was found to be caused at least in part by a decrease in pituitary responsiveness to LH-RH and by an increased effectiveness of estrogen negative feedback. This feedback becomes fully competent during the days preceding the onset of puberty. At puberty estrogen secretion gradually increases to reach levels that, by exerting a positive feedback action, trigger a preovulatory peak of gonadotropins and prolactin. Pituitary responsiveness to LH-RH increased the day before the first ovulation, presumably due to the elevated E_2 levels present and to a direct effect of LH-RH that, by acting on the gland, sensitized it to its own action. Lastly, the inhibitory dopaminergic (DA) control of prolactin secretion was found to be more pronounced in female than in male rats. This difference appeared to be due to a modulatory action of estrogen initiated at a pre-pubertal age rather than to a process of neonatal hypothalamic sexual differentiation. In the female the inhibitory DA control of prolactin is initiated shortly after birth, whereas the stimulatory effect of estrogen on prolactin release develops after the third week of post-natal life, reaching a maximum around the time of puberty. It seems that in both male and female, tonic prolactin release depends on the balance between the inhibitory DA tone and the influence of stimulatory signals. Increased stimulation of prolactin release would be followed by a compensatory increase in the inhibitory DA tone to attenuate the prolactin response.

INTRODUCTION

The onset of puberty is a process that depends to a large extent upon the occurrence of numerous and complex developmental changes which appear to be causally related to each other (McCANN *et al.*, 1974 ; RAMIREZ, 1974). The study of these maturational changes, therefore, can lead us to a much better understanding of the physiological mechanisms involved in determining the transit of the individual from a stage of sexual immaturity to a phase of reproductive capacity. During development, the function of the hypothalamic-pituitary unit related to the secretion of gonadotropins and prolactin can be affected, among other factors, by hormonal signals of ovarian and adrenal origin. In the present communication, we will mainly emphasize this aspect. Likewise, although several other pituitary hormones may be involved in defining the pubertal process, only aspects relevant to the control of gonadotropin and prolactin release will be discussed herein. We will consider the changes in blood gonadotropin levels during development in the female rat, attempting to correlate them with maturational changes in steroid negative and positive feedback and with changes in pituitary sensitivity to LH-RH. Further, we will describe the alterations in blood gonadotropins and prolactin and hypothalamic LH-RH that accompany the onset of puberty in the female rat. Lastly, we will compare the developmental changes in plasma prolactin levels in female and male rats and their relation to the maturation of the hypothalamic dopaminergic and gonadal steroidal control of prolactin secretion.

GENERAL METHODS

Animals used in the present experiments belonged to the Holtzman strain. They were housed under controlled conditions of lighting (14 hr on, 10 hr off) and temperature (24°C). Tap water and Purina laboratory chow were available *ad libitum*. All animals were weaned at day 21.

Surgical procedures, namely ovariectomy and adrenalectomy, were performed using ether anesthesia. Heparinized blood samples were taken by heart puncture or by decapitation.

All steroid treatments were given subcutaneously, whereas LH-RH was administered intravenously. Doses are indicated under results. Dopaminergic receptors were blocked by Pimozide injection (0.63 mg/kg, sc).

Radioimmunoassays. Plasma LH levels were measured by the method of NISWENDER *et al.* (1968). FSH and prolactin were measured with the kits provided by the National Institute of Arthritis and Metabolic Diseases. Estradiol was measured according to the method of HOTCHKISS *et al.* (1971), and LH-RH was determined by a slight modification of the method of NETT *et al.* (1973).

RESULTS AND DISCUSSION

Plasma gonadotropin levels during prepubertal development of the female rat. Maturation of estrogen feedback control

In the female, plasma FSH rises from already elevated levels at day 5 to a peak at 12-15 days of age (KRAGT and DAHLGREN, 1972 ; OJEDA and RAMIREZ, 1972 ; DÖHLER and WUTTKE, 1974). Thereafter, it declines as the animal approaches

maturity, reaching a nadir during the days preceding the first preovulatory gonadotropin discharge (OJEDA, WHEATON, JAMESON and MCCANN, in press). Plasma LH displays a more irregular pattern, showing afternoon peaks in some animals during the first three weeks of life (DÖHLER and WUTTKE, 1974). Using some RIA systems, LH appears clearly elevated during the first 15 days of life (MEIJS-ROELOFS *et al.*, 1973 *a* ; OJEDA and RAMIREZ, 1972 ; WEISZ and FERIN, 1970 ; DÖHLER and WUTTKE, 1974), declining then to minimal values as the animal approaches puberty. Other RIA systems, such as the ovine-ovine assay used in our lab, show rather low LH levels during the first 15 days although the episodic afternoon discharges can still be observed (OJEDA, JAMESON and MCCANN, unpublished data).

The factors responsible for the patterns of plasma gonadotropin displayed in the female rat during development are still poorly understood. Nevertheless, there is evidence that they are in part caused by the post-natal age at which the pituitary-gonadal feedback relationship becomes fully operative. Subcutaneous injections

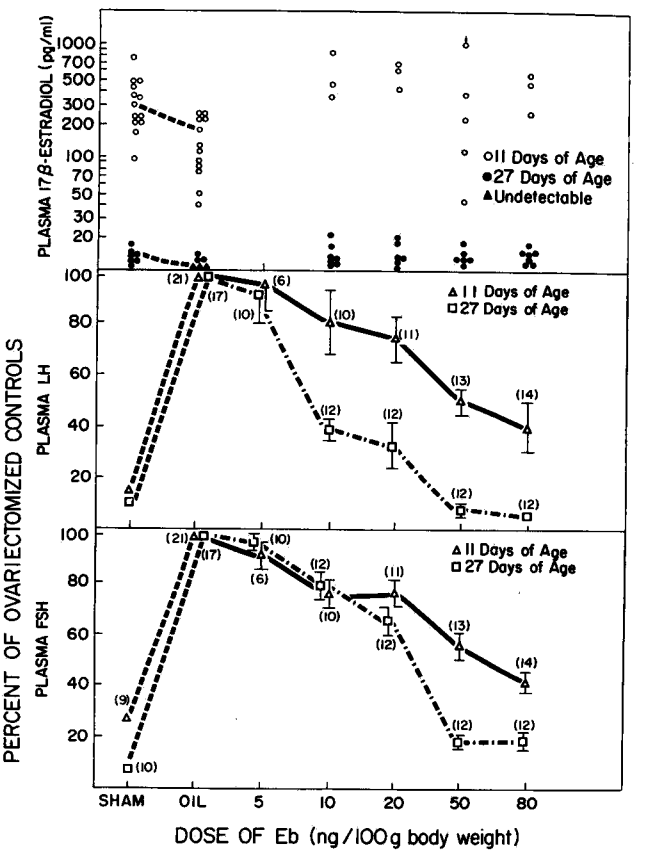

FIG. 1. — *Plasma LH, FSH and E_2 in 11 and 27 day old, ovariectomized rats treated for two days with different doses of estradiol benzoate* (Eb). Number in parenthesis represents number of animals, and for E_2, each point represents the hormone level in pooled plasma from 2-3 animals. In this and subsequent figures vertical lines represent standard error of the mean (from OJEDA *et al.*, 1975, *Neuroendocrinology*, **18**, 242-256).

of a nearly physiological single dose of estradiol benzoate in short term ovariectomized rats produced only a small decrease in post-castration plasma gonadotropin levels in rats younger than 15 days of age (OJEDA and RAMIREZ, 1973). The effectiveness of estrogen was markedly enhanced after day 15, the time at which plasma FSH normally declines. Recently, we have re-evaluated the negative feedback response to estrogen in infantile and juvenile female rats (10 and 25 days of age, respectively) (OJEDA *et al.*, 1975). The effects of the estradiol treatment administered at four different dose levels on plasma estradiol titers were monitored by radioimmunoassay. The results indicate that the negative feedback, although present, was less well developed in infantile than in the older animals (fig. 1). In these experiments, plasma estradiol levels were high in the young animals and declined by day 25, an observation that has been previously reported by other authors (MEIJS-ROELOFS *et al.*, 1973 *b* ; WEISZ and GUNSALUS, 1973). In our study, the elevated levels of estradiol were partially reduced by ovariectomy and completely eliminated by either ovariectomy-adrenalectomy or adrenalectomy alone, an observation which indicates that circulating estrogen in the infantile animal is mainly of adrenal origin. The high E_2 titers appear to be caused by an enhanced rate of production of E_2 by these glands and by a reduced metabolic clearance of the steroid. A prolonged half-life of E_2 in plasma of infantile rats is evidenced by the finding that when E_2 was injected intravenously at day 10 in intact rats to elevate plasma E_2, E_2 remained elevated when measured 30 to 120 min after its injection, but on day 25, 50 p. 100 of the injected E_2 had disappeared from plasma in 90 min.

A decreased function of negative estrogen feedback (as compared to the adult) during the first two weeks of life despite the presence of high circulating E_2 levels can be caused by the absence of specific hypothalamic cytoplasmic and nuclear sites for estrogen binding in rats younger than 20 days (KATO *et al.*, 1971 ; PLAPINGER and MCEWEN, 1973) and by the presence in high concentration in plasma of neonatal rats of a specific estradiol binding protein which becomes very low between 20 and 30 days of life (RAYNAUD *et al.*, 1971). However, the former concept has been recently challenged on the basis that the topographic hypothalamic pattern of nuclear concentration of estrogen in infantile rats as determined by autoradiography is exactly similar to that of the adult rat (SHERIDAN, SAR and STUMPF, 1974). This finding and the recent observation that estrogen implanted directly into the medial basal hypothalamus of 12 day old female rats effectively suppresses post-ovariectomy plasma FSH levels (RAMIREZ, in press) indicates that the binding of E_2 to plasma protein may be the more important mechanism which prevents the estrogen negative feedback from attaining competence at these early ages.

Relatively low but unstable LH levels in the presence of high FSH titers cannot be totally explained by the relative inability of estrogen negative feedback to operate during the first 2-3 weeks of life. Recently, GELATO and WUTTKE (1975) have shown that the administration of an anti-estradiol serum to infantile rats results in decreased LH levels, an observation that led these authors to suggest the existence of a positive feedback action of estrogen on LH secretion. However, earlier studies indicated that this mechanism, like the negative feedback of estrogen does not appear to develop fully until the fourth week of life, a time at which estrogen treatment becomes consistently able to induce a preovulatory-type surge of gona-

dotropin release (CALIGARIS *et al.*, 1972). Estrogen treatment at earlier ages failed to evoke gonadotropin release.

FSH secretion, on the other hand, appears to be controlled by estrogen only in a negative feedback manner during the first three weeks of post-natal life since anti-estrogen serum treatment elevated serum FSH titers (GELATO and WUTTKE, 1975).

The attainment of full competence by the estrogen negative and positive feedbacks after the third week of life is correlated with increasing levels of progesterone (DÖHLER and WUTTKE, 1974 ; MEIJS-ROELOFS *et al.*, 1975) which may synergize with the low estrogen titers to provide a more effective feedback signal than estrogen alone. It is likely, therefore, that the decline in FSH levels after day 15 is caused by the combined feedback action of these two steroids.

Steroidal modulation of pituitary function during the infantile phase in the female rat

The pattern of plasma FSH levels which peak at about day 12-15 without a corresponding consistent LH rise suggests that some other ovarian or adrenal steroid(s) may be acting directly on the pituitary gland to stimulate FSH secretion and/or enhance pituitary responsiveness to the action of hypothalamic LH-RH. We have evaluated this latter possibility by injecting LH-RH intravenously (25 ng/-100 g b.w.) at day 15 (13:00 hr) to intact, ovariectomized, and ovariectomized-adrenalectomized rats. Surgical operations were performed at day 10. Both LH and FSH were substantially increased by LH-RH 15 min following its injection in intact animals. However, the FSH response to LH-RH was almost completely prevented by ovariectomy or combined adrenalectomy-ovariectomy. Adrenalectomy alone was ineffective. By contrast, the LH response to LH-RH remained either unchanged or was slightly increased after ovariectomy or ovariectomy-adrenalectomy. In an attempt to determine the cause(s) of this divergent pattern of response, animals were ovariectomized and adrenalectomized at day 13 and starting the same day, they were treated with different steroids or combinations of steroids for two days (fig. 2). Injections were administered twice a day, starting in the afternoon of the day of the operation. As before, LH-RH was injected i.v. on day 15 at 13:00 hr. Among the steroids tested, surprisingly, 5α-dihydrotestosterone (DHT) and progesterone completely restored the FSH response to LH-RH, whereas estradiol and testosterone were ineffective (fig. 2). When progesterone was administered along with estradiol, the restorative effect of progesterone was significantly reduced. However, estradiol treatment did not alter the effect of dihydrotestosterone on the LH-RH-induced FSH release. The LH response to LH-RH was not altered by any of these treatments except perhaps by DHT, which in combination with estrogen depressed the response.

These findings suggest that progesterone and possibly 5α-dihydrotestosterone are involved in the physiological control of FSH secretion during the infantile phase of life by acting directly on the pituitary gland to increase its responsiveness to endogenous LH-RH. It is known that progesterone is present in plasma at this phase of development (DÖHLER and WUTTKE, 1974 ; MEIJS-ROELOFS *et al.*, 1975) and that the capacity of the pituitary gland to form 5α-dihydrotestosterone and

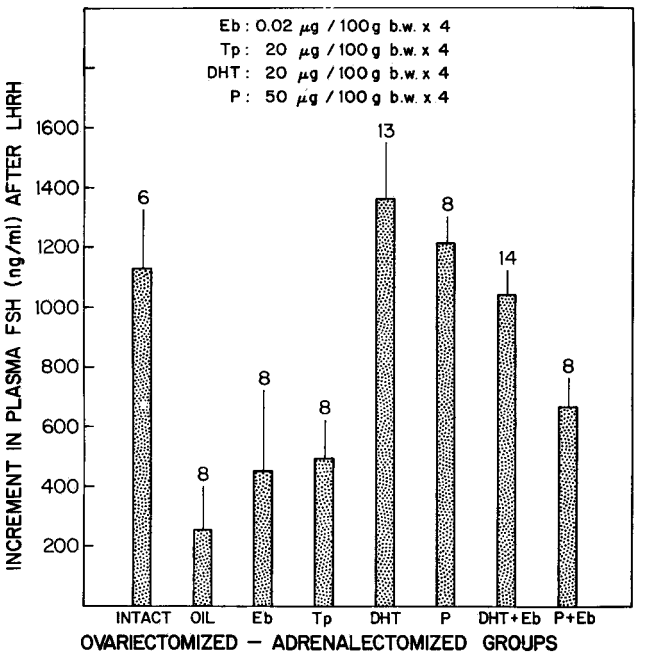

FIG. 2. — *Effect of treatment with different steroids on the pituitary FSH responsiveness to LHRH of 15 day old, ovariectomized-adrenalectomized female rats.* Number of animals are shown above each bar. Eb : estradiol benzoate, Tp : testosterone propionate, DHT : 5α-dihydrotestosterone, P : progesterone.

3α-androstanediol from testosterone during post-natal development in the female rat correlates very closely with the pattern of changes in plasma FSH found at the same period of time (DENEF *et al.*, 1974). In addition, a stimulatory effect of 5α-dihydrotestosterone on pituitary gonadotropins *in vitro* has also been described (MITTLER, 1974). Thus, if testosterone is present at those early ages, it would be readily converted into DHT.

These observations in conjunction with those of GELATO and WUTTKE (1975) make the possibility particularly attractive that in the infantile animal the maturing negative feedback action of steroids is counter-balanced by their developing stimulatory effects on the hypothalamic-pituitary unit.

The role of the adrenal glands during the infantile period remains conjectural. Although adrenalectomy removed all radioimmunoassayable circulating plasma E_2 (WEISZ and GUNSALUS, 1973 ; OJEDA *et al.*, 1975), in the present experiments adrenalectomy alone did not induce an increase in gonadotropins and did not alter pituitary responsiveness to exogenous LH-RH. Nevertheless, combined ovariectomy-adrenalectomy resulted in higher plasma LH levels than those found after ovariectomy alone, a finding that suggests the existence of an ovarian-adrenal interaction.

Pituitary responsiveness to LH-RH during the prebubertal phase

In earlier work, DEBELJUK *et al.* (1972) found that the maximal increase in plasma LH in response to a single dose level of LH-RH occurred around day 15 (the first age tested), declining to a minimum around day 35, *i.e.*, at the time of

puberty. We have restudied this problem, giving special consideration to pituitary responsiveness during the first weeks of life. LH-RH was injected i.v. at four different dose levels in different groups of rats, at intervals of 5 days beginning on day 5, and LH and FSH were measured 15 and 45 min later. The results revealed that the pituitary gland is remarkably more responsive to LH-RH during the first two weeks of life than in later phases of development (fig. 3). The LH and FSH response

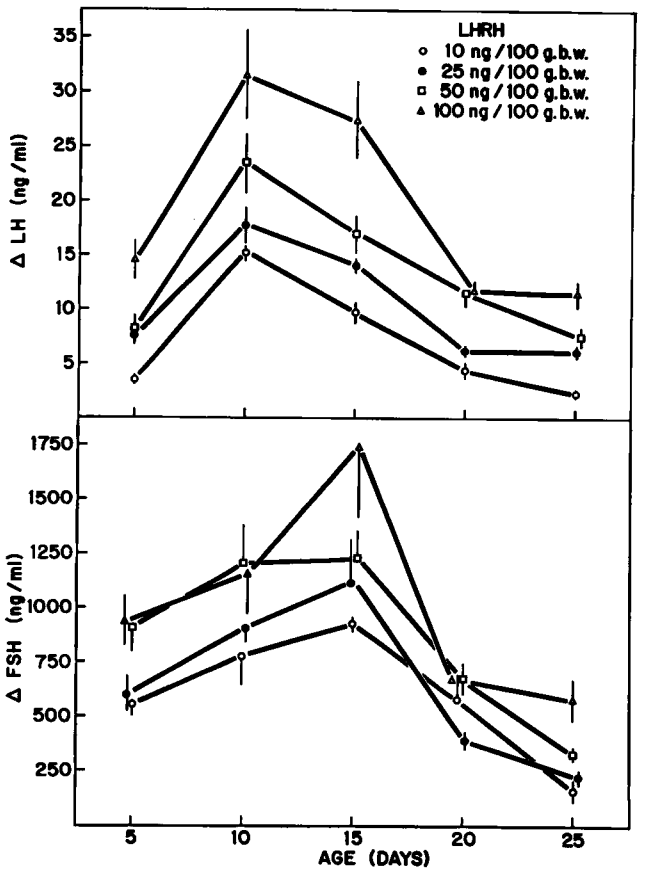

FIG. 3. — *Changes in pituitary LH and FSH responsiveness to LHRH during sexual development of female rats*

increased from day 5 to day 10-15, becoming maximal around this age. It declined at day 20, becoming even smaller at day 25, to reach a minimum shortly before the first pre-ovulatory gonadotropin discharge (CASTRO-VAZQUEZ and OJEDA, unpublished). Interestingly enough, in the infantile rat, FSH levels increased dramatically after LH-RH injection, reaching maximum levels at 15 min to decline slightly at 45 min. This is in contrast with the pattern of response observed in adult animals in which FSH is barely elevated by a single i.v. injection of LH-RH (ARIMURA *et al.*, 1972 ; COOPER *et al.*, 1974).

This increased LH and FSH response to LH-RH does not seem to be due to AM-PM variations in pituitary sensitivity since the same pattern of response was

observed when animals 10, 15, 20 or 25 days old were injected with LH-RH at 10:00, 13:00 or 16:00 hr. The presence of the ovaries and perhaps the adrenals appears to be responsible for the large FSH release induced by LH-RH in the younger animals, since, as indicated before, ovariectomy or ovariectomy plus adrenalectomy completely abolished this response, whereas treatment with DHT or progesterone restored it.

Since the LH response to the decapeptide in the infantile rat remained essentially the same after ovariectomy, adrenalectomy or steroid replacement therapy, we investigated the possibility that a reduced metabolic clearance of exogenous LH-RH in the infantile rat was responsible, at least in part, for the apparent enhancement in LH response to LH-RH observed at those early days. The decapeptide was injected i.v. at a dose of 10 ng/100 g b.w. in 10 or 28 day old rats and different groups of animals were sacrificed at 1, 3, 4, 5, 6, 8, 16 and 32 min later. Plasma LH-RH was then assayed by RIA. The results showed that in 28 day old rats, LH-RH had at 1/2 of 2.78 minutes, whereas in 10 day old rats, its at 1/2 was 3.36 minutes. Therefore, the more pronounced LH response to the decapeptide found in infantile rats cannot be readily explained by a more sustained LH-RH concentration in plasma of these animals than in the older rats. In addition, it is unclear which factors are involved in determining the observed decrease in LH response to LH-RH as the animal matures. We have found that around the time of puberty in animals of the same age, the LH response to LH-RH injected on body weight basis is less pronounced in individuals with a greater body weight than in lighter individuals (CASTRO-VAZQUEZ and OJEDA, unpublished data). The mechanism underlying this phenomenon remains to be determined.

Hormonal changes accompanying vaginal opening and first ovulation

Earlier work in which LH was measured by bioassay indicates that the first ovulation which usually coincides with vaginal opening is preceded by a preovulatory peak of LH (RAMIREZ, 1974). Moreover, other authors have reported an acute drop in pituitary FSH which presumably follows an elevation of plasma FSH just before ovulation (WATANABE and MCCANN, 1969). Similarly, VOOGT *et al.* (1970) have observed a rise in plasma prolactin levels around the time of puberty.

We have recently studied in further detail the sequence of hormonal events that accompany the onset of puberty in female rats (OJEDA *et al.*, in press). Immature rats were sacrificed by decapitation between days 32 and 38 and plasma concentrations of gonadotropins, prolactin, LH-RH and hypothalamic content of LH-RH were determined by RIA. Groups of animals were sacrificed at 10 AM and 4 PM throughout the pubertal period, the uterine weight recorded and ovaries inspected for signs of ovulation. Animals with vaginae closed were grouped according to the condition of the uterus as anestrus, early proestrus and late proestrus, the uterus being unstimulated, dilated with some fluid, or ballooned, respectively. Vaginal opening usually occurred at the end of late proestrus and was associated with ovulation. Animals were studied for up to 3 days after vaginal opening and were grouped according to vaginal cytology. It should be noted that vaginal opening was not always accompanied by ovulation, a fact that may be related to sensitivity of the vaginal epithelium to estrogen which even though not secreted in amounts sufficient to trigger gonadotropin discharge could induce vaginal opening.

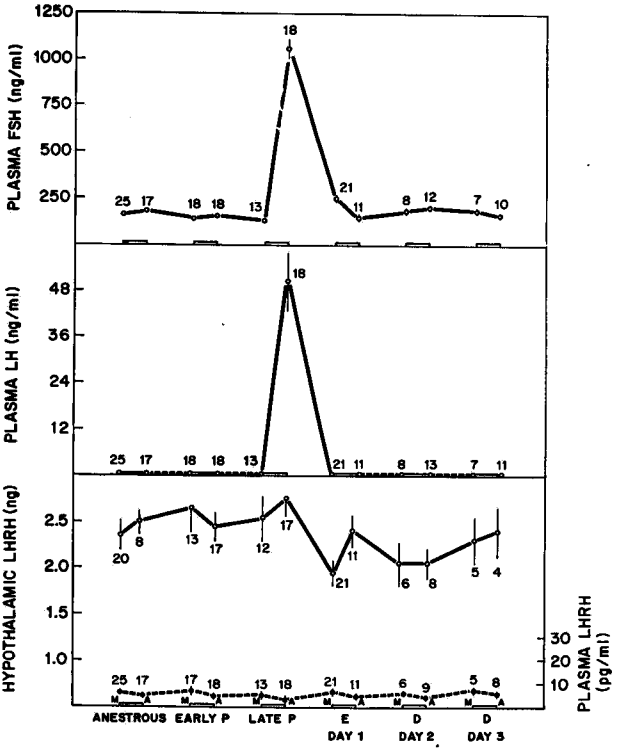

FIG. 4. — *Plasma levels of LH, FSH and LHRH and hypothalamic LHRH content around the time of puberty in the female rat*
(for details, see text)

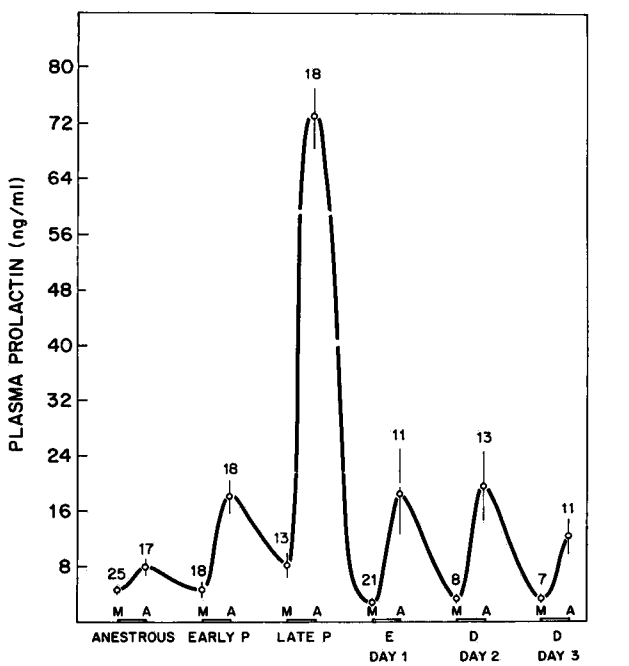

FIG. 5. — *Changes in plasma prolactin levels during puberty in the female rat*
(for details, see text)

Uterine weight, taken as an index of estrogen secretion, was low during the anestrous phase, increased during early proestrus, and reached a peak at late proestrus, declining thereafter. Plasma LH and FSH remained low through the morning of the late proestrous phase. On the afternoon of this phase, when uterine weight was maximal, both gonadotropins increased dramatically (fig. 4). The following morning (estrus), LH but not FSH had returned to basal values. FSH returned to basal levels on the afternoon of estrus. Plasma LH-RH was uniformly low throughout the entire pubertal period, whereas hypothalamic LH-RH content showed a slight decline on the morning of the day of vaginal opening (fig. 4). Plasma prolactin was low in the morning during the entire pubertal period, but showed peaks on the afternoons which reached a maximum at the late proestrous phase and declined thereafter, following the pattern of changes in uterine weight (fig. 5).

These results indicate that the onset of puberty in the female rat is brought about by a gradual increase in estrogen secretion which, acting at the CNS-pituitary level, triggers the preovulatory surge of gonadotropins and prolactin. The gradual elevation in circulating prolactin observed in the afternoon preceding the preovulatory peak may be related to an enhanced sensitivity of the prolactin-releasing apparatus to the stimulatory effect of estrogen (OJEDA and MCCANN, 1974) secreted in increased amounts by the ovaries and perhaps by the adrenals (SHAIKH and SHAIKH, 1975).

Although a change in the threshold for the negative feedback of gonadal steroids has been reported to occur at puberty in the female rat (MCCANN *et al.*, 1974 ; RAMIREZ, 1974), it is also apparent that the metabolic clearance of estrogen increases with development (de HERTOGH *et al.*, 1970) and this could, in fact, explain the apparent change in the setpoint for negative feedback observed during the pubertal process. Nevertheless, the existence of this latter factor cannot be discounted, especially in view of the recent finding that minute amounts of estrogen implanted into the medial basal hypothalamus effectively suppressed plasma FSH in prepubertal ovariectomized rats, but not in adult animals (RAMIREZ, in press).

Pituitary responsiveness to LH-RH during puberty. Initiation of the « priming » effect of LH-RH

To evaluate hypophyseal responsiveness, synthetic LH-RH (20 ng/100 g b.w.) was injected i.v. at 1:00 PM during the different phases or puberty and blood samples were withdrawn 20 min later. Following the injection of LH-RH, a small increase in plasma LH was observed on anestrus, early proestrus, the day of vaginal opening (estrus), and the second and third day after vaginal opening (diestrus 1 and diestrus 2, respectively). This response was greater in the late proestrous phase of puberty, *i.e.*, on the day on which the preovulatory gonadotropin discharge occurred (CASTRO-VAZQUEZ and OJEDA, unpublished data). However, in the anestrous phase, the more immature rats (as judged by their lower body weight) gave a response similar to that found in the late proestrous phase.

In the adult rat, LH-RH appears to exert a priming effect on the pituitary gland so that further exposure to this neurohormone results in enhanced responsiveness of the gonadotrophs (AIYER *et al.*, 1974 ; CASTRO-VAZQUEZ and MCCANN, 1975). This finding prompted us to determine the phase of sexual development at which the

priming effect of LH-RH first appears. With this purpose in mind, a second dose of LH-RH was given 60 min following the first injection and the LH response observed 20 min later was compared with the response to the first injection. The results indicate that the priming effect of LH-RH appears on the day of the first preovulatory gonadotropin discharge and, as in the adult, may depend upon the prior presence of increased circulating estrogen levels, since the LH response to the second LH-RH injection was greater than the first injection only on the late proestrous phase.

Development of the control of prolactin release

In the female rat, plasma prolactin is barely detectable in 21 day old fetuses and on the day of birth. It declines further by days 3-6, and then rises gradually to attain adult levels around day 35 (OJEDA and MCCANN, 1974). This pattern of prolactin appears to depend on the balance between the stimulatory effect of estrogen on prolactin release and the inhibitory control exerted by the hypothalamic dopaminergic system. The stimulatory effect of estrogen on prolactin release appears after the third week of life and becomes maximal around the time of vaginal opening. In contrast to the delayed development of estrogen stimulatory action on prolactin, the dopaminergic

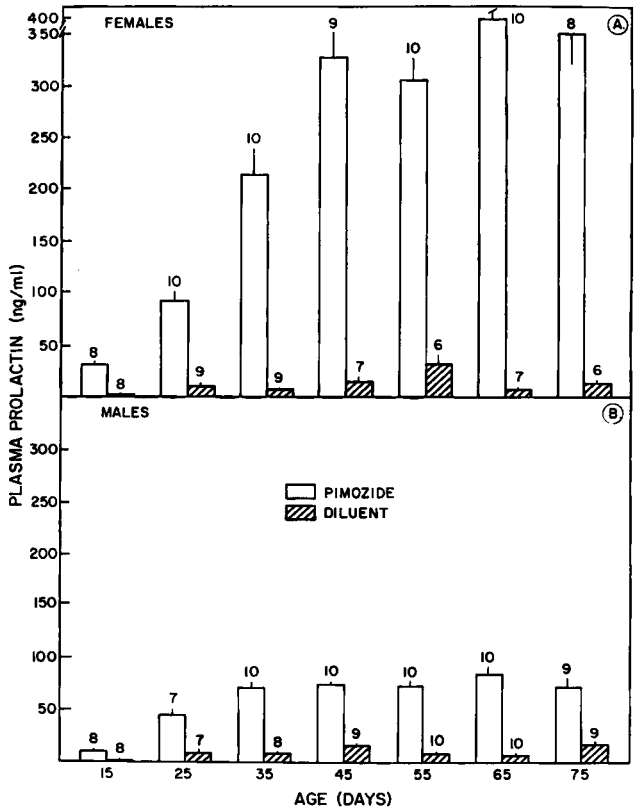

FIG. 6. — *Comparison of the effect of blockade of dopaminergic receptors with Pimozide on plasma prolactin levels of developing male and female rats*

inhibitory control of prolactin is already present by the third day of life. It seems that once the dopaminergic mechanism becomes operative, prolactin release is predominantly under an inhibitory influence until the responsiveness to estrogen develops around day 24, prolactin levels rising thereafter in response to endogenous estrogen stimulation.

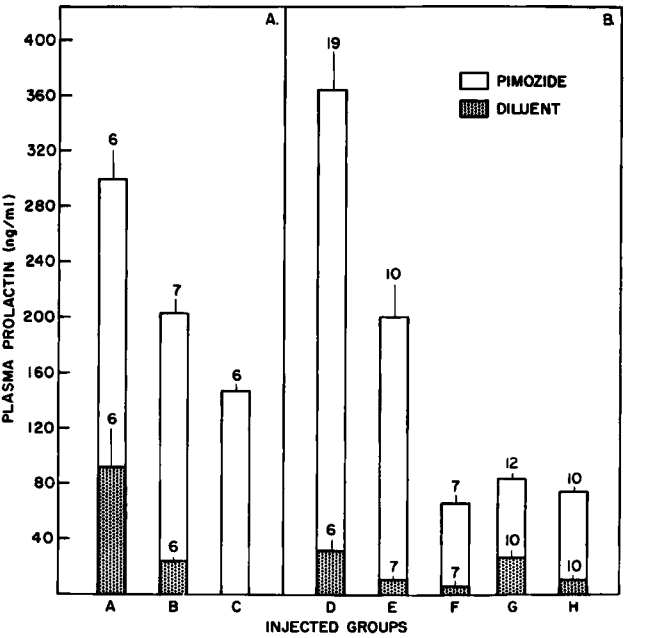

FIG. 7. — *Effect of ovariectomy at adulthood* (a) *or ovariectomy at day 21* (b) *on the prolactin response to blockade of dopaminergic receptors of normal or androgenized female rats*

A : androgenized rats treated with Pimozide.
B : androgenized rats ovariectomized 7 days before Pimozide treatment.
C : androgenized rats ovariectomized 34 days before Pimozide treatment.
D : intact normal females treated with Pimozide.
E : females castrated 7 days before Pimozide treatment.
F : females castrated at day 21 and treated with Pimozide at adulthood.
G : females castrated at day 21 and treated with Pimozide at adulthood. These animals were adrenalectomized 10 days before Pimozide.
H : normal males treated with Pimozide.

In the male rat, plasma prolactin levels are also low during the first three weeks of life (DÖHLER and WUTTKE, 1974 ; MCCANN *et al.*, 1974). Thereafter, they increase, reaching maximal values arond day 90 (NEGRO-VILAR *et al.*, 1973). We have recently found that part of this increase in prolactin levels can be attributed to an age-dependent increase in responsiveness to stress stimuli (OJEDA, JAMESON and MCCANN, 1976, in press). Animals subjected to the same stress (ether fumes for 3 min) responded with more pronounced increases in prolactin titers as they matured, with plateau values being reached around day 60-70. By contrast, decapitated rats subjected to minimum stress showed little, if any, change in prolactin levels from 20 to 70 days of age.

The blockade of the inhibitory dopaminergic control of prolactin with pimo-

zide resulted in a larger release of prolactin in female than in male rats (fig. 6). In experiments addressed to determine the cause (s) of this sex difference (OJEDA and JAMESON, unpublished data), it was found that neonatal androgenization of female rats did not reverse the female pattern of prolactin release in response to Pimozide injection. Likewise, the male type of response to Pimozide was not altered by castration at the day of birth or feminization of male fetuses with cyproterone acetate, performed according to the procedure of HAHN *et al.* (1973). In contrast, when males castrated at adulthood or feminized adult male rats were treated with estrogen, the magnitude of the prolactin release in response to DA blockade was much larger than that of male rats not given estrogen. In androgenized or intact female rats, castrated as adults, the prolactin response to Pimozide was attenuated, though it still was large than that of normal males (fig. 7). However, when female rats were castrated at day 21, the prolactin response to DA receptor blockade at adulthood was similar to that observed in males (fig. 7). Consequently, these results indicate that the more pronounced prolactin release in female than in male rats in response to blockade of DA receptors is not caused by a process of perinatal sexual differentiation at a hypothalamic level, but is rather a consequence of a modulatory action of estrogen initiated during the first four weeks of life.

Sexual Maturation, 3rd Workshop
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RÉSUMÉ

MATURATION DU CONTRÔLE DE LA SÉCRÉTION DES GONADOTROPINES ET DE LA PROLACTINE CHEZ LE RAT

On a étudié divers aspects de la maturation du contrôle de la sécrétion des gonadotropines et de la prolactine chez le Rat. Les estrogènes (E_2) circulants sont très élevés chez la jeune Ratte et diminuent quand elle approche de la maturité. Ces fortes concentrations d'E_2 semblent dues à une augmentation de la production d'E_2 par les ovaires et les surrénales et à une faible dégradation métabolique du stéroïde. Les concentrations élevées de FSH plasmatique au moment où les taux E_2 sont également importants doivent résulter au moins en partie, d'une diminution de l'efficacité de la rétroaction négative de l'estrogène et d'une augmentation de la sensibilité de l'hypophyse au LH-RH, induite par une action directe de la progestérone et de la 5α-dihydrotestostérone sur la glande. La rétroaction négative des estrogènes contrôlant la sécrétion de LH est également réduite chez la jeune Ratte et la sensibilité de l'hypophyse au LH-RH augmentée. Ce dernier phénomène n'est pas altéré par ovariectomie-surrénalectomie ou par traitement de remplacement avec des stéroïdes. La chute des gonadotropines plasmatiques observée après le 15e jour est due, au moins en partie, à une diminution de la sensibilité de l'hypophyse au LH-RH et à une augmentation de l'efficacité de la rétroaction négative des estrogènes. La rétroaction devient pleinement compétente dans les jours qui précèdent la puberté. A la puberté, la sécrétion d'estrogènes augmente progressivement pour atteindre les taux qui, en exerçant

une rétroaction positive, entraîneront la décharge préovulatoire de gonadotropines et de prolactine. La sensibilité de l'hypophyse au LH-RH augmente la veille de la première ovulation, probablement sous l'effet des niveaux élevés d'E_2 présents à ce moment et par une action directe du LH-RH qui, en agissant sur l'hypophyse, la sensibilise à sa propre action.

Enfin, le contrôle dopaminergique (DA) inhibiteur de la sécrétion de prolactine est plus important chez la femelle que chez le mâle. La différence semble due à une action modulante de l'estrogène initiée avant la puberté plutôt qu'à une différenciation sexuelle néonatale de l'hypothalamus. Chez la femelle, le contrôle DA inhibiteur de la prolactine est initié peu après la naissance, alors que l'effet stimulant des estrogènes sur la sécrétion de prolactine se met en place après trois semaines et atteint son maximum au moment de la puberté. Il semble que chez le mâle et la femelle la sécrétion tonique de prolactine dépend de la balance entre l'action inhibitrice DA et l'influence des signaux stimulants. L'augmentation de la stimulation de la sécrétion de prolactine doit être suivie d'une augmentation compensatrice de l'action inhibitrice DA pour atténuer la réponse de la prolactine.

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