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Effects of Hyperprolactinemia on Ornithine Decarboxylase Activity and Polyamine Levels in Seminal Vesicles of Genetically Prolactin-Deficient Adult Dwarf Mice[1]

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ABSTRACT

Prolactin (PRL) has been shown to exert many different actions in various biological systems. Polyamines are known to influence the growth and function of the seminal vesicles (SV). Furthermore, ornithine decarboxylase (ODC) is considered a key enzyme in the biosynthesis of polyamines and is regulated by PRL in certain target tissues. Adult Ames dwarf mice (df/df), genetically deficient in PRL, were used for this study. The experimental groups were as follows: Group 1, pituitary-grafted; Group 2, sham-operated; Group 3, castrated + testosterone propionate (TP)-treated (25 μg/mouse, 3 times/wk, s.c.) + grafted; and Group 4, castrated + TP as above. The animals were killed 40 days later, and polyamines and ODC activity in SV and liver were determined. Serum PRL, FSH, and testosterone (T) were also measured. In the grafted groups, there were significant elevations in serum PRL and FSH levels. In the gonad-intact, pituitary-grafted group, animals exhibited an elevation in plasma T levels, and similar levels were achieved in the castrated, androgen-replaced groups. In hyperprolactinemic mice, the weights of SV were significantly greater than in the corresponding control groups. The relative weights of the SV showed a similar pattern. An increase in ODC activity was observed in both SV and liver in hyperprolactinemic groups. In those animals in which serum T levels were held constant, an increase in the enzyme activity in SV was detected in hyperprolactinemic group whereas in liver, no significant difference was observed. Concentrations of polyamines in the SV were increased in hyperprolactinemic, castrated, TP-treated mice. The present results indicate that PRL can exert a direct stimulatory effect on the growth, ODC activity, and polyamine levels in the SV.

INTRODUCTION

The ability of prolactin (PRL) to affect the growth of male accessory reproductive glands was described several decades ago (for a review, see Bartke [1]), and this earlier suggestion received strong support from the demonstration of specific PRL receptors in the prostate gland, the seminal vesicles (SV), and the coagulating glands [2–5].

It is not known how the PRL-receptor interaction transmits its signal to elicit intracellular metabolic changes. Since intracellular second messengers are usually involved in the action of polypeptide hormones, the possibility remains that the effect of PRL involves some intracellular mediators. Previous studies suggested several possible candidates, including cyclic GMP, calcium ions, prostaglandins, and polyamines [6].

Ornithine decarboxylase (ODC) is considered to be the key enzyme in the biosynthesis of polyamines. The regulation of ODC is apparently complex, and its activity can be influenced by alterations in the rate of synthesis and turnover, by conversion of active to inactive forms, as well as by interactions with an anti-enzyme, a specific protein inhibitor of ODC [7, 8].

Mice with hereditary dwarfism (Ames dwarf, df/df) described by Schaible and Gowen [9] are PRL- and growth hormone-deficient [10, 11]. Because of their congenital PRL deficiency, dwarf mice are particularly useful in defining the role of this pituitary hormone.

The structural and functional integrity of the SV has been shown to be maintained by PRL and androgens [12–14]. In the present study, we have examined the effects of experimentally induced long-term hyperprolactinemia in adult dwarf mice on the growth, polyamine content, and ODC activity of the SV, and on several parameters of pituitary-testicular function. In addition, another PRL target organ, the liver, was studied for comparison.

MATERIALS AND METHODS

Chemicals

L[1-^{14}C]ornithine (sp. act. 49–56 mCi/mmol) and 1, 2, 6, 7-^{3}H(N)-testosterone (sp. act. 85–105 mCi/mmol) were purchased from New England Nuclear Corp. (Boston, MA). The purity of the steroid was regularly checked by thin-layer chromatography and [^{14}C]ornithine was purified as described previously [15]. L-Ornithine hydrochloride, dithiothreitol (DTT), Tris, EDTA, pyridoxal 5-phosphate, and

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testosterone propionate (TP) were obtained from Sigma Chemical Co. (St. Louis, MO). All other chemicals and reagents were of analytical grade.

Animals and Treatments

Ames dwarf mice (df/df) and their normal littermates (DF/−) were produced in our breeding colony (SIU, Carbondale) by mating heterozygous normal carriers of the *df* gene [11]. All animals were maintained in a room with controlled illumination (14L:10D) and temperature (22 ± 2°C) with free access to commercial food (TekLad, Madison, WI) and tap water.

Adult mice were divided into four groups and were treated as follows: Group 1: two pituitaries from young adult females of the same strain were implanted under the capsule of the right kidney of individual male mice; Group 2: animals were sham-operated; Group 3: the animals were castrated through a midline incision, grafted as described before, and immediately treated with TP in corn oil (25 μg/mouse, 3 times/wk, s.c.); Group 4: the animals were castrated and treated with TP as above. Forty days later, 16 h after the last injection, the animals were weighed, blood was obtained via cardiac puncture under ether anesthesia, and the animals were killed by overexposure to ether, followed by cervical dislocation. Plasma was obtained and frozen until assayed for PRL, FSH, and testosterone (T). The SV were removed, blotted, and extruded before weighing. In addition, a fragment from the central region of the liver was removed and weighed. Tissues were kept at −20°C until processing.

Preparation of Tissue Extracts

The frozen tissues were thawed, cut with scissors into small pieces, and homogenized; and 20 000 × *g* supernatants were prepared for the enzyme assay as previously described [15].

ODC Assay

ODC activity was measured in duplicates according to the original method [16], as previously described [15]. Detectable levels of ODC activity were found when 0.5–1.0 mg protein [17] per sample was used. The enzymatic activity was expressed in terms of pmol $^{14}CO_2$ released/mg protein/h, pmol/g tissue or pmol/organ.

Polyamine Measurements

Polyamine concentrations in SV were determined by thin-layer chromatography as described previously [18].

Hormone Assays

Plasma levels of FSH were determined by a heterologous RIA system utilizing rat reference preparation (rFSH-RP-2) and rat FSH antiserum (rFSH-A/S-S-11). We have described this RIA method [19] and validated this assay for measuring FSH levels in mice [20]. Plasma PRL levels were measured by a specific homologous RIA using mouse reference preparation (AFP-6476C) and mouse PRL antiserum (AFP-131078), kindly donated by Dr. A.F. Parlow, as described previously [20]. Plasma T concentrations were also measured by an RIA system [21, 22]. The sensitivities of these assays were as follows: FSH, 0.25 ng/tube; PRL, 0.1 ng/tube; and T, 5 pg/tube. All samples for measuring FSH and PRL levels were included in the same assay, and the intraassay coefficients of variation were FSH, 5.3%, and PRL, 3.8%. The intraassay and interassay coefficients of variation for the T assay were 6.4% and 7.4%, respectively.

Other Methods

Protein concentration was measured by the method of Lowry et al. [17] with BSA as a standard.

Statistical analysis was carried out using nonparametric Mann-Whitney U-test (one-tail), or by using Student's *t*-test.

RESULTS

Hormone Levels in Dwarf Mice

As expected, PRL was undetectable in sham-operated, gonad-intact dwarf mice. Plasma FSH and T levels were detectable in these animals. Treatment of male dwarf mice with PRL-producing ectopic pituitary homografts caused the expected increase in plasma PRL levels, and a significant elevation in plasma FSH levels in both intact and castrated mice (Table 1). An elevation in plasma T levels was also

TABLE 1. Effects of ectopic pituitary transplants on plasma PRL, FSH, and T levels in adult Ames dwarf mice (df/df).*

Treatment	No. of mice	PRL (ng/ml)	FSH (ng/ml)	T (ng/ml)
Graft	9	51.7 ± 6.4**	4.7 ± 0.4*	4.1 ± 1.7*
Sham	9	ND†	3.1 ± 0.4	1.8 ± 0.1
Castrated + TP + graft	8	59.2 ± 5.5**	7.9 ± 0.9*	5.4 ± 0.9
Castrated + TP + sham	12	ND†	3.5 ± 0.2	5.3 ± 0.5

*Results are expressed as mean ± SE. For more details of experimental groups, see *Materials and Methods*.

*,**Asterisks denote different levels of significance: *$p < 0.05$, **$p < 0.005$, compared to the corresponding groups of controls.

†ND: not detectable.

detected in the gonad-intact, grafted mice in comparison to the sham-operated animals.

In dwarf mice that were castrated, treated with TP, and grafted or sham-operated, plasma T levels were similar to those measured in the gonad-intact, grafted group and higher than the values found in intact dwarf mice or in normal adult male mice (data now shown).

Effects of PRL on SV Weight

Absolute weights of SV in both hyperprolactinemic groups were significantly greater than in the corresponding groups of control animals (Table 2). However, the increase in absolute SV weight was more pronounced in intact, grafted mice than in castrated, TP-treated, grafted mice. Relative weight of the SV was similarly increased in the hyperprolactinemic groups (Table 2).

Effect of PRL on ODC Activities in the SV and Liver

Hyperprolactinemia produced a significant elevation in ODC activity in SV relative to that in sham-operated mice (358 ± 30 vs. 232 ± 39 pmol/mg protein/h, $p < 0.05$) (Fig. 1). A similar increase was observed in castrated and androgen-treated groups in which peripheral T levels did not differ (pituitary grafted + castrated + TP-treated: 366 ± 50 vs. castrated + TP-treated: 255 ± 32 pmol/mg protein/h, $p < 0.05$). The same pattern was obtained if the ODC activity was expressed as pmol/mg protein/h, pmol/mg tissue or pmol/organ.

Treatment of dwarf mice with pituitary grafts for 40 days produced a significant increase in ODC activity in the liver as compared to values measured in sham-operated mice (Fig. 1: 180 ± 25 vs. 81 ± 14 pmol/mg protein/h, $p < 0.005$). However, in castrated, androgen-treated mice, no difference was detected in the enzyme activity between groups with pituitary graft (102 ± 8) vs. without pituitary graft (110 ± 9 pmol/mg protein/h). The same pattern was obtained if the ODC activity was expressed as pmol/mg prot/h or pmol/mg tissue.

Effect of PRL on Polyamines in SV

Treatment of TP-injected, castrated dwarf mice with ectopic pituitary transplants significantly increased putrescine

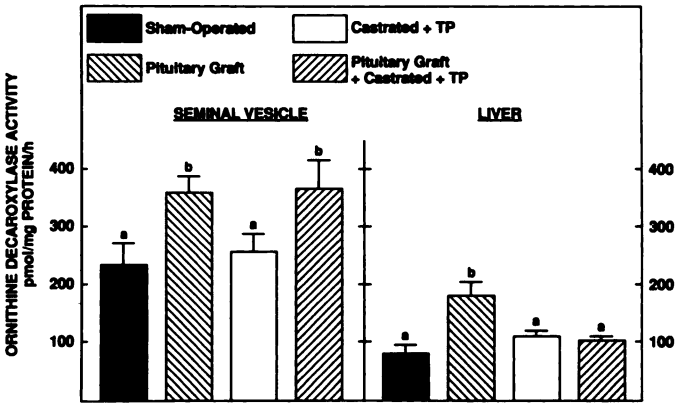

FIG. 1. Effects of PRL-secreting ectopic pituitary transplants on ODC activity in SV and liver of adult dwarf mice. Values are expressed as mean ± SEM (n = 5–6 mice/group). Values without the same letter differ at a significance level of at least $p < 0.05$.

(0.49 ± 0.06 vs. 0.32 ± 0.05 nmol/mg SV; $p < 0.005$) and spermidine (0.56 ± 0.1 vs. 0.40 ± 0.09 nmol/mg SV; $p < 0.01$) levels in SV relative to those in SV of TP-treated, castrated, non-grafted dwarf mice (Fig. 2).

DISCUSSION

One of the physiological roles of PRL in male animals is concerned with the growth and function of the reproductive and other androgen-sensitive organs [23]. A complete deficiency in PRL and growth hormone and reduced levels of thyroid-stimulating hormone and gonadotropins have been demonstrated in Ames dwarf mice (df/df) [10, 24]. We have treated these animals with ectopic pituitary tissue because it secretes biologically active PRL [25, 26].

The observed increase in plasma FSH levels in both intact and castrated, TP-treated male dwarf mice implanted with pituitary grafts confirms previous findings that PRL may stimulate FSH release in the male mouse [27, 28]. In addition, the present results suggest that the positive feedback of T on FSH may be reduced when PRL levels are undetectable (androgen-treated, castrated animals without the pituitary graft). Apparently, hereditary PRL deficiency in dwarf mice is associated with altered physiological input to the hypothalamic centers responsible for the increase of synthesis and/or release of pituitary FSH. Moreover, neither

TABLE 2. Effects of ectopic pituitary transplants on SV and body weights (BW) in adult Ames dwarf mice (df/df).*

Treatment	No. of mice	BW (g)	SV weight (mg)	SV weight mg/100 g BW
Graft	9	24 ± 2	134 ± 9**	582 ± 62*
Sham	9	20 ± 3	72 ± 6	393 ± 37
Castrated + TP + graft	8	23 ± 1	85 ± 6*	339 ± 42*
Castrated + TP + sham	12	24 ± 2	64 ± 4	256 ± 15

*Results are expressed as mean ± SE. For more details of experimental groups, see *Materials and Methods.*

*,**Asterisks denote different levels of significance: *$p < 0.05$, **$p < 0.005$, compared to the corresponding groups of controls.

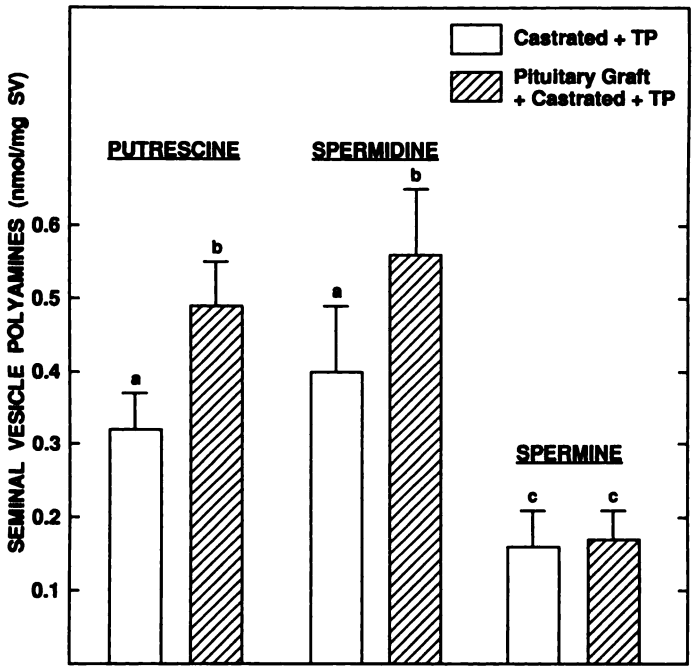

FIG. 2. Effects of PRL-secreting ectopic pituitary transplants on polyamine levels in SV of adult dwarf mice. Values are expressed as mean ± SEM (n = 5–6 mice/group). Values without the same letter differ at a significance level of at least $p < 0.05$.

endogenously increased nor exogenously administered T suppressed plasma FSH levels in pituitary-grafted dwarf mice. The ability of PRL to increase hypothalamic noradrenergic activity and, thus, presumably LHRH release in this species [29] provides a plausible explanation for these effects. In gonad-intact dwarf mice, it is also possible that PRL influenced production of testicular products other than T to account for these effects. For example, reduced secretion of inhibin may have contributed to the observed increase in plasma FSH levels [30, 31].

The significant elevation of peripheral T levels seen in grafted mice is probably due to PRL increasing gonadotropin release [27–29], and the ability of the testis to produce androgens [23] as has been shown in PRL-injected immature rats [5].

The true growth of any organ involves a concomitant increase in total organ weight and DNA content [32]. The structural and functional integrity of male sexual accessory organs, such as SV, are stimulated and maintained by androgens [33, 34]. Testosterone exerts, at least in part, its androgenic trophic effects through 5α-reduction to dihydrotestosterone (DHT). The androgen growth response in adult mice is imprinted by neonatal endogenous androgens [35], which exhibit a developmental pattern described previously [36]. Part of such biological response could be mediated by specific androgen receptors. These binding sites decrease with aging independently from tissue androgen levels, leading to a diminished sensitivity to androgens in adult mice [36]. A more recent observation from the same group [37] confirmed lack of a significant correlation between androgen levels and concentration of androgen-binding sites. Therefore, at present, the mechanism by which androgens regulate the growth of SV is incompletely understood.

Apart from androgens, PRL also influences the SV in a number of species, most likely through specific binding sites [2, 38], and these effects appear to be particularly important in mice [29].

Prolactin action on male accessory reproductive glands appears to require the presence of androgens [39, 40], since hyperprolactinemia alone does not induce trophic effects in the SV of adult castrated rats [12] or mice [13]. However, recent data indicate that in mice, hyperprolactinemia alone can have a stimulatory effect on DNA synthesis in the epithelial cells of the SV and that it can delay the involution of the SV until 30 days following castration plus adrenalectomy [14]. The present results are in agreement with these recent findings. Plasma T levels were similarly increased in the gonad-intact, pituitary-grafted, and castrated T-treated mice; yet, SV weight was increased only in the first group. The effect of pituitary transplants on SV weight was greater in gonad-intact than in castrated, T-replaced mice in spite of very similar plasma PRL levels. This raises a possibility that stimulation of SV growth in gonad-intact mice may have involved androgen metabolite(s) other than T. Testosterone metabolism and its disposition are probably different in intact as compared to T-replaced, castrated mice [37, 41]. It is also possible that various intratesticular factors, such as testicular peptides, may influence the effect of T on SV growth.

Forty days after grafting, the relative SV weight was moderately but significantly increased. This increase was comparable to that described in the C57BL PRL-deficient mice 1 or 2 mo after receiving pituitary grafts at the age of 30 or 90 days [42, 43]. In addition, these authors indicated that the displastic epithelium of the SV of untreated mice was normalized by pituitary isografts. These and the observations presented here strongly suggest that PRL displays a specific trophic effect on SV.

Ornithine decarboxylase (EC 4.1.1.17) is the initial and rate-limiting enzyme in polyamine biosynthesis under normal conditions [44]. Activation of ODC and polyamine production are associated with cell growth, differentiation, and proliferation, whereas reduction in ODC activity and polyamine levels results in a decrease in growth and differentiation of cells [45, 46]. Measurements of ODC have been used as the most sensitive marker for assessing androgen activity in prostatic [47] and epididymal tissue [15], as well as in SV [48, 49].

Since PRL has been shown to increase the nuclear concentration of DHT in perfused male accessory reproductive glands [50] and specific PRL receptors have been described in SV [51], the stimulation of ODC activity was considered a valuable tool in evaluating the PRL-androgen interactions

and as particularly pertinent to the present experimental model. The effects of pituitary grafts in dwarf mice in the present study suggest that PRL exerts a clear-cut effect upon growth and ODC activity of the SV.

Rat liver contains specific binding sites for lactogenic hormones [52], which can be induced by pituitary transplants [53], PRL injections [54], or stimulation of endogenous PRL release [55]. Moreover, PRL has been reported to stimulate ODC activity in the liver [56] and, although the regulation of ODC activity is very complex, the magnitude of this response appears to correlate well with the number of receptor sites [57]. To further assess the organ specificity of PRL action and the efficacy of T, the ODC activity was also measured in the liver in the present study. The observed increase in the specific ODC activity in the liver was probably due to the increase in plasma T levels induced by PRL rather than to the actions of PRL itself. In support of this conclusion, hepatic ODC activity was not affected by the grafts in animals that had been castrated and given T replacement. In contrast to these observations, Grahn et al. [58] reported that T injections failed to increase ODC activity in the liver. These discrepancies could be ascribed to the fact that Grahn et al. [58] used intact females rather than castrated males and a different protocol of administration.

In summary, results obtained in our experimental model under the conditions described allow us to conclude that in SV of adult mice PRL produces a trophic response that differs from the response seen in the liver in terms of growth and ODC activity.

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REFERENCES

1. Bartke A. Role of PRL in reproduction in male mammals. Fed Proc 1980; 39:2577–2581.
2. Rajaniemi H, Oksanen A, Vanha Perttula T. Distribution of ^{125}I-prolactin in mice and rats. Studies with whole body and microautoradiography. Horm Res 1974; 5:6–10.
3. Charreau EH, Attramadal A, Torjesen PA, Calandra RS, Purvis K, Hansson V. Androgen stimulation of PRL receptors in rat prostate. Mol Cell Endocrinol 1977; 7:1–8.
4. Barkey RJ, Shani J, Barzilai D. Regulation of PRL binding sites in the seminal vesicle, prostate gland, testis and liver of intact and castrated adult rats: effect of administration of testosterone, 2-Bromo, α-ergocriptine and fluphenazine. J Endocrinol 1979; 81:11–17.
5. Baranao JLS, Tesone M, Oliveira-Filho RM, Chiauzzi VA, Calvo JC, Charreau EH, Calandra RS. Effects of PRL on prostate androgen receptors in male rats. J Androl 1982; 3:281–288.
6. Rillema JA. Mechanism of PRL action. Fed Proc 1980; 39:2593–2598.
7. Canellakis, ES, Kriakidis DA, Pawlak JW. The complexity of regulation of ornithine decarboxylase. Med Biol 1981; 59:279–283.
8. Pegg AE, McCann PC. Polyamine metabolism and function. Am J Physiol 1982; 243:C212–C218.
9. Schaible R, Gowen JW. A new dwarf mouse. Genetics 1961: 46:896.
10. Bartke A. Prolactin deficient mice. In: Alexander NJ (ed.), Animal Models for Research on Contraception and Fertility. Hagerston: Harper and Row; 1978: 360–365.
11. Bartke A. Genetic models in the study of anterior pituitary hormones. In: Shire JGM (ed.), Genetic Variation in Hormone Systems, vol I. Boca Raton: CRC Press; 1979: 113–126.
12. Negro-Vilar A. Prolactin and the growth of prostate and seminal vesicles. In: Spring Mill L, Hafez ESE (eds.), Male Accessory Sex Glands. New York: Elvesier North Holland Bio Med Press; 1980: 223–233.
13. Keenan EJ, Klase PA, Thomas JA. Interactions between PRL and androgens in the accessory sex organs of male mice. J Endocrinol 1981; 90:323–330.
14. Ayata M, Yamana T, Okamoto S, Li W, Terada N, Matsumoto K. Increase in epithelial cell growth by hyperprolactinemia induces delay of castration-induced involution of mouse seminal vesicle. J Steroid Biochem 1989; 32:719–724.
15. de las Heras MA, Calandra RS. Androgen-dependence of ornithine decarboxylase in the rat epididymis. J Reprod Fertil 1987; 79:9–14.
16. Pegg AE, Lockwood DH, Williams Ashman HG., Concentrations of putrescine, polyamines and their enzyme synthesis during androgen-induced prostatic growth. Biochem J 1970; 117:17–31.
17. Lowry OH, Rosenbrough NJ, Farr AL, Randale RJ. Protein measurement with the Folin phenol reagent. J Biol Chem 1951; 193:265–275.
18. Seiler N. Thin-layer chromatography and thin-layer electrophoresis of polyamines and their derivatives. Methods Enzymol 1983; 94:3–9.
19. Chandrashekar V, Bartke A, Sellers K. Prolactin modulates the gonadotropin response to the negative feedback effect of testosterone in immature male rats. Endocrinology 1987; 120:758–763.
20. Chandrashekar V, Bartke A, Wagner TE. Endogenous human growth hormone [GH] modulates the effect of gonadotropin-releasing hormone on pituitary function and the gonadotropin response to the negative feedback effect of testosterone in adult male transgenic mice bearing human GH gene. Endocrinology 1988; 123:2717–2722.
21. Chandrashekar V, Bartke A. Influence of endogenous prolactin on the luteinizing hormone stimulation of testicular steroidogenesis and the role of prolactin in adult male rats. Steroids 1988; 51:559–576.
22. Chandrashekar V, Bartke A. The influence of short photoperiod on testicular and circulating levels of testosterone precursors in the adult golden hamster. Biol Reprod 1989; 40:300–306.
23. Bartke A. Pituitary-testis relationships. In: Hubinont PO, L'Hermite M, Schowers J (eds.), Progress in Reproductive Biology, vol 1. Karger: Basel; 1976: 136–152.
24. Slabaugh MB, Lieberman ME, Rutledge JJ, Gorski J. Growth hormone and PRL synthesis in normal and homozygous Snell and Ames dwarf mice. Endocrinology 1981; 109:1040–1046.
25. Everett JW. Functional corpora lutea maintained for months by autografts of rat hypophysis. Endocrinology 1956; 58:786–796.
26. Adler RA. The anterior pituitary-grafted rat. A valid model of chronic hyperprolactinemia. Endocr Rev 1986; 7:302–313.
27. Bartke A, Goldman BD, Bex F, Dalterio S. Effects of prolactin (PRL) on pituitary and testicular function in mice with hereditary PRL deficiency. Endocrinology 1977; 101:1760–1766.
28. Klemcke HG, Bartke A. Effects of chronic hyperprolactinemia in mice on plasma gonadotropin concentrations and testicular human chorionic gonadotropin binding sites. Endocrinology 1981; 108:1763–1768.
29. Bartke A, Morgan WW, Clayton RN, Banerji TK, Brodie AM, Parkening TA, Collins TJ. Neuroendocrine studies on hyperprolactinemic male mice. J Endocrinol 1984; 112:215–220.
30. Chowdhury M, Steinberger A, Steinberger E. Inhibition of de novo synthesis of FSH by the Sertoli cell factor (SCF). Endocrinology 1978; 103:644–657.
31. Franchimont P, Verstraelen-Proyard J, Hazee-Hagelstein MT, Renard Ch, Demoulin A, Bourguignon JP, Hustin J. Inhibin: from concept to reality. Vitam Horm 1979; 37:243–302.
32. Coffey DS. The effect of androgens on DNA and RNA synthesis in sex accessory tissues. In: Brandes D (ed.), Male Accessory Sex Organs. Structure and Function in Mammals. New York: Academic Press; 1974: 307–315.
33. Brandes D. Hormonal regulation of fine structure. In: Brandes D (ed.), Male Accessory Sex Organs. Structure and Function in Mammals. New York: Academic Press; 1974: 184–222.
34. Dadaune JP. Functional morphology of the seminal vesicle epithelium. Prog Reprod Biol Med 1985; 12:18–35.
35. Jean-Foucher Ch, Berger M, Gallon C, De Turckheim M, Veyssiere G, Jean Cl. Imprinting of male sex tissues by neonatal endogenous androgens in mice. Molecular alterations following exposure to cyproterone acetate. Horm Res 1986; 24:38–45.
36. Jean-Foucher Ch, Berger M, De Turckheim M, Veyssiere G. Developmental pat-

terns of plasma and testicular testosterone in mice from birth to adulthood. Acta Endocrinol 1978; 89:780–788.
37. Gallon Ch, Veyssiere G, Berger M, Jean-Foucher Ch, De Turckheim M, Jean Cl. Age related changes in the concentration of cytosolic androgen receptors in the epididymis, vas deferens and seminal vesicle of maturating male mice. J Androl 1989; 10:188–194.
38. Costlow ME, McGuire WL. Autoradiographic localization of the binding of ^{125}I-labelled PRL to rat tissues in vitro. J Endocrinol 1977; 75:221–227.
39. Thomas JA, Keenan EJ. Prolactin influences upon androgen action in male accessory sex organs. Adv Sex Horm Res 1976; 2:425–470.
40. Calandra RS, Baranao JLS, Bertini L, Calvo JC, Charreau EH, Chiauzzi VA, Suescun MO, Tesone M. Effects of PRL on male gonad and sex accessory organs. In: DeNicola AF, Blaquier J, Sota RJ (eds.), Physiopathology of Hypophysial Disturbances and Diseases of Reproduction. New York: Alan R. Liss; 1982: 87–116.
41. Djoseland O. Androgen metabolism by rat epididymis. 3. Effect of castration and anti-androgens. Steroids 1976; 27:47–64.
42. Nomura M, Hashimoto K, Hrigaya T, Hashimoto H, Yoshida O. Effects of hyperprolactinemia on reporduction in male mice. J Endocrinol 1985; 107:71–76.
43. Grew FC. Genotype specific effects of pituitary isografts on the reproductive system in male mice. University of Essex; 1988. Thesis.
44. Tabor CW, Tabor H. Polyamines. Annu Rev Biochem 1984; 53:749–790.
45. Russell DH, Durie BGM. Polyamines as Markers of Normal and Malignant Growth. New York: Raven Press; 1978.
46. Russell DH. Ornithine decarboxylase: a key enzyme in normal and neoplastic growth. Drug Metab Res 1985; 16:1–88.
47. Marchetti B, Poulin R, Plante M, Labrie F. Castration levels of plasma testosterone have potent stimulatory effects on androgen-sensitive parameters in the rat prostate. J Steroid Biochem 1988; 31:411–429.
48. Piik K, Rajamaki P, Guha SK, Janne J. Regulation of L-ornithine decarboxylase in rat ventral prostate and seminal vesicle. Biochem J 1977; 168:379–385.
49. Fjosne HE, Strand H, Ostensen MA, Sunde A. Ornithine decarboxylase and s-adenosylmethionine decarboxylase activity in the accessory sex organs of intact, castrated and androgen-stimulated castrated rats. The Prostate 1988; 12:309–320.
50. Baker HWG, Worgul TJ, Santen RJ, Jefferson LS, Bardin CW. Effect of PRL on nuclear androgens in perfused male accessory sex organs. In: Troen P, Nankin HR (eds.), the Testis in Normal and Infertile Men. New York: Raven Press; 1977; 379–385.
51. Barkey RJ, Shani J, Amit RJ, Barzilai D. Specific binding of PRL to seminal vesicle, prostate and testicular homogenates of immature, mature and aged rats. J Endocrinol 1977; 74:163–173.
52. Posner BI, Kelly PA, Friesen HG. Induction of lactogenic receptor in rat liver: influence of estrogens and the pituitary. Proc Natl Acad Sci USA 1974; 71:2407–2410.
53. Posner BI, Kelly PA, Friesen HG. Prolactin receptors in rat liver: possible induction by PRL. Science 1975; 188:57–59.
54. Calvo JC, Finocchiaro L, Luthy IA, Charreau EH, Calandra RS, Engstrom B, Hansson V. Specific PRL binding in the rat adrenal: its characterization and hormonal regulation. J Endocrinol 1981; 89:317–325.
55. Luthy IA, Tesone M, Oliveira-Filho RM, Somoza GM, Charreau EH, Libertun C, Calandra RS. Effect of median eminence lesions and hormonal replacement on the PRL receptors in the adrenal gland and Langerhans islets from ovariectomized rats. J Recept Res 1985; 5:105–119.
56. Hurley TW, Kuhn CM, Schanberg SM, Handwerger S. Differential effects of placental lactogen, growth hormone and PRL on rat liver ornithine decarboxylase activity in the perinatal period. Life Sci 1980; 27:2269–2272.
57. Richards JF. Ornithine decarboxylase activity in tissues of PRL treated rats. Biochem Biophys Res Commun 1975; 63:292–298.
58. Grahn B, Heninngsson SSG, Kahlson G, Rosengren E. Alterations in the activities of ornithine and histidine decarboxylases provoked by testosterone in mice. Br J Pharmacol 1973; 48:113–120.