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Extra-pituitary action of gonadotropin releasing hormone: possible application

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Abstract. Chronic administration of a potent gonadotropin releasing hormone inhibits ovulation in women. The suppression of gonadal function during long term treatment with the GnRH analogues is ascribable to inhibition of gonadotropin secretion caused by the down regulatory action of the decapeptide at the pituitary level. Reduced progesterone production with premature onset of menstruation has been observed in women injected with the agonist during the midluteal phase. The decapeptide however, has no effect on *in vitro* human ovarian steroidogenesis. Specific receptors for GnRH have been located on rodent ovarian cells, but corpora lutea of rhesus monkey and human ovaries seem to lack these receptors. The luteolytic effect in women thus appears to be central in origin and not a direct effect on the corpus luteum. Recently, a superactive agonist of GnRH given around the peri-implantation period has been shown to terminate pregnancy in baboons. Monoclonal antibodies against GnRH administered during the same period in a fertile cycle also abrogated pregnancy in these animals. Using immuno-enzymatic techniques GnRH has been localized on the placenta. GnRH also exerts a stimulatory effect on hCG production by the placental villi maintained in culture. Addition of anti-luteinizing hormone releasing hormone antibodies blocks this effect completely. It seems that placenta is the only other tissue besides the pituitary where GnRH has probably a regulatory role in the human female.

Keywords. GnRH, pituitary, ovary, luteolysis, abortion, placenta.

Introduction

Gonadotropin releasing hormone (GnRH) has some unique features. The decapeptide molecular structure of the hormone is essentially conserved in evolution and the same molecule is active in a wide variety of animal species facilitating on one hand its easy testing and on the other its varied applications. This review briefly summarizes the correct state of knowledge in the area.

Effect of GnRH on ovulation

Pulsatile administration of GnRH has been used to stimulate ovulation in human (Reid *et al.,* 1981). Treatment with a potent agonist of GnRH has however, demonstrated potential as a contraceptive agent. Chronic treatment of the agonist in regularly menstruating women inhibits ovulation (Nillius *et al.,* 1978). Most of the women under chronic treatment showed cyclical estrogen production accompanied by uterine

Abbreviations used: GnRH, Gonadotropin releasing hormone; hCG, human chorionic gonadotropin.

bleedings though ovulation was suppressed. Long-term treatment with still higher doses of the agonist lead to a sustained suppression of estrogen biosynthesis in fertile women, provoking symptoms of estrogen deficiency and endometrial inactivity (Hardt *et al,* 1983). The primary effect of daily GnRH agonist administration appears to be directly on the pituitary. The agonist seems to decrease pituitary responsiveness impairing its capacity to release sufficient amounts of follicle stimulating hormone and luteinizing hormone for induction of full follicular maturation and ovulation (Dericks *et al.,* 1977; Bergquist *et al.,* 1979).

Effects on steroidogenesis

GnRH and its agonists have been reported to have direct inhibitory effects on ovarian steroidogenesis *in vitro* (Hsueh and Erickson, 1979; Clayton *et al.,* 1979; Massicotte *et al.,* 1981). Specific receptors on the surface of ovarian cells, resembling those identified on pituitary gonadotrophs appear to mediate the action of GnRH (Clayton *et al.,* 1979; Clayton and Catt, 1981). However, all these observations have been made on rodent tissues, leaving open the question whether GnRH can directly affect human ovarian function. Reports have appeared from more than one laboratory that the decapeptide or its agonist analogues can induce luteolysis in women (Casper and Yen, 1979; Koyama *et al.,* 1978; Lemay *et al.,* 1979a, b; Sheehan *et al.,* 1982). Administration of GnRH agonist during the mid luteal phase reduces progesterone production causing premature onset of menstruation. A significant cause of infertility and spontaneous abortions in monkeys (Wilks *et al.,* 1976; Zerega and Hodgen, 1981) and in women (Jones and Madrigal-Castro, 1970; Sherman and Korenman, 1974) have been recognised to be due to the occurrence of short or inadequate luteal phases. The endometrium in women with such luteal phase defect is known to develop inadequately with respect to both its morphology and function as a result of impaired secretion of estradiol and progesterone (Sheehan *et al.,* 1982). A normal predecidual reaction usually does not occur and normal nidation is thereby impeded (Tredway *et al.,* 1973). Since infertility is a dominant feature of luteal phase dysfunction, pharmacologically induced luteal phase defects may offer considerable potential as a method of contraception.

The direct effects of GnRH on the steroidogenic activity of human ovarian cells maintained in culture have been studied by two groups. Tureck *et al.* (1982) have observed that GnRH agonist (Wyeth) is able to inhibit the secretion of progesterone by granulosa cells in a dose dependent manner. Casper *et al.* (1984) on the other hand found no effect of GnRH or its agonist [(imBzl)-D-HiS^{6}-Pro^{9}-NEt] in modifying the acute steroidogenic activity of cultured human granulosa cells over a period of 6 days. The apparent discrepancy between the two observations may be due to a number of methodologic differences including the use of a different GnRH agonist. However, the fact that corpora lutea of rhesus monkey (Asch *et al.,* 1981) and human ovarian cells (Clayton and Huhtaniemi, 1982) do not bind to GnRH seem to favour the findings of Casper *et al.* (1984) that the peptide has no direct effect on human ovary. The conclusion that emerges from these experiments is that unlike in the rat, GnRH does not seem to exert direct effects on the human corpus luteum. This is further supported

by the fact that human corpus luteum do not have receptors for GnRH. The luteolytic effect of GnRH in women thus appear to be central in origin causing reduced and disordered gonadotropin secretion and not a direct effect on the corpus luteum.

Effects on pregnancy

Luteal insufficiency and shortening of the menstrual cycle by the agonist has been further exploited in a fertile cycle (Das and Talwar, 1983). GnRH agonist (Hoe 766) injected in the early luteal phase of a fertile cycle in baboons caused a significant suppression of circulating progesterone but failed to abrogate pregnancy. The agonist administered towards the end of the luteal phase around the periimplantation period, however, was successful in terminating pregnancy in these primates (Das, 1984). Using compressed pellets of two other agonists of GnRH (D-Trp^6 and D-Nal(2)^6), Vickery *et al.* (1981) could induce abortion in 2 out of 6 baboons. The timing of the treatment seems to be very important. Positive results were obtained only when the agonist was given in late luteal phase, around the time when chorionic gonadotropin was first detectable in circulation.

Khodr and Siler-Khodr (1980) have reported the presence of GnRH in human placenta and have also shown the stimulatory effect that GnRH exercises on the synthesis and secretion of human chorionic gonadotropin (hCG) (Siler-Khodr and Khodr, 1981). Using human placental villi from early pregnancy and maintaining them in culture we have also observed the stimulatory effect of GnRH on hCG secretion. Addition of anti-LHRH antibodies in the medium completely blocked this action of GnRH (Das and Talwar, 1984; Rao *et al.,* 1984). The interference of anti-GnRH antibodies with placental chorionic gonadotropic secretion is more clearly indicated by *in vivo* experiments in baboons (Talwar *et al.,* 1983). The animals were rendered pregnant and the antibodies administered at a stage when the chorionic gonadotropin was present in circulation. Two injections of antibodies were given at 24 h interval on the grounds that the rise in chorionic gonadotropin is steep in early pregnancy and the hormonal support has to be nullified for at least 48 h to influence pregnancy. In 3 out of 4 baboons tested, anti-GnRH monoclonals brought about termination of pregnancy.

It thus appears that besides its well known action on the pituitary, GnRH has also a regulatory role in the placenta. Chorionic gonadotropin is known to be essential for maintenance of early pregnancy and curtailment of the hormone at this stage leads to abortion. The *in vivo* and *in vitro* experiments indicate a possibility to block chorionic gonadotropin secretion and thus abrogate pregnancy at a very early stage by GnRH agonists and antagonists delivered locally around periimplantation period. The window of effectiveness is however, very narrow and thus detracts from the practical application of these agents.

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References

Asch, R. H., Sickle, M. V., Barnaceda, J. P., Coy, D. H. and Schally, A. V. (1981) *J. Clin. Endocrinol. Metab.,* **53,** 215.
Bergquist, C., Nillius, S. J. and Wide, L. (1979). *Contraception,* **19,** 497.
Casper, R. F. and Yen, S. S. C. (1979) *Science,* **205,** 408.
Casper, R. F., Erickson, G. F., Rebar, R. W. and Yen, S. S. C. (1982) *Fertil. Steril.,* **38,** 406.
Casper, R. F., Erickson, G. F. and Yen, S. S. C. (1984) *Fertil. Steril.,* **42,** 39.
Clayton, R. N., Harwood, J. P. and Catt, K. J. (1979) *Nature* (*London*), **282,** 90.
Clayton, R. N. and Catt, K. J. (1981) *Endocrine Rev.* **2,** 186.
Clayton, R. N. and Huhtaniemi, I. T. (1982) *Nature* (*London*), **299,** 56.
Das, C. and Talwar, G. P. (1983) *Fertil. Steril.,* **39,** 218.
Das, C. and Talwar, G. P. (1984) in *Proc. of Symp. on 'LHRH in Fertility and Malignancy',* Hyderabad, India.
Das, C. (1984) in *Indo-U.S. Symposium on Ovum Implantation,* (I.C.M.R., New Delhi) p. 150.
Dericks-Tan, J.S.E., Hammer, E. and Taubert, H. D. (1977) *J. Clin. Endocrinol. Metab.,* **45,** 597.
Hardt, W. and Schmidt-Gollwitzer, M. (1983) *Clin. Endocrinol.,* **19,** 613.
Hsuch, A. J. W. and Erickson, G. F. (1979) *Science,* **204,** 854.
Jones, G. S. and Madrigal-Castro, V. (1970) *Fertil Steril.,* **21,** 1.
Jones, P. B. C. and Hsuch, A. J. W. (1981) *Biol. Reprod.,* **24,** 747.
Khodr, G. S. and Siler-Khodr, T. M. (1980) *Science,* **207,** 315.
Koyama, T., Ohkura, T., Kumasaka, T. and Saito, M. (1978) *Fertil. Steril.,* **30,** 549.
Lemay, A., Labrie, F., Ferland, L., Reynaud, J. P. (1979a) *Fertil. Steril.,* **31,** 29.
Lemay, A., Labrie, F., Belanger, A. and Raynaud, J. P. (1979b) *Fertil. Steril.,* **32,** 646.
Massicotte, J., Borgus, J. P., Lachance, R. and Labrie, F. (1981) *J. Steriod. Biochem.,* **14,** 239.
Nillius, S. J., Bergquist, C. and Wide, L. (1978) *Contraception,* **17,** 537.
Rao, A. J., Mathialagan, N., Kotagi, S. G. and Moudgal, N. R. (1984) *J. Biosci.,* **6,** 97.
Reid, R. L., Leopold, G. R. and Yen, S. S. C. (1981) *Fertil. Steril.,* **36,** 553.
Sheehan, K. L, Casper, R. F. and Yen, S. S. C. (1982) *Science,* **215,** 170.
Sherman, B. M. and Korenman, S. G. (1974) *J. Clin. Endocrinol Metab.,* **38,** 89.
Siler-Khodr, T. M. and Khodr, G. S. (1981) *Biol. Reprod.* **25,** 353.
Talwar, G. P., Singh, V., Singh, O., Das, C, Gupta, S. K. and Singh, G. (1983) in *Hormone Receptors in Growth and Reproduction,* (eds B. B. Saxena, K. J. Catt, L. Birhbaumer and L. Martini) (New York: Raven Press) p.351.
Tredway, D. R., Mishell, D. R. and Moyer, D. L. (1973) *Am. J. Obstet. Gynaecol.,* **117,** 1031.
Tureck, R. W., Mastroianni, L. Jr., Blasco, L. and Strauss, J. F. (1982) *J. Clin. Endocrinol. Metab.,* **54,** 1078.
Vickery, B. H., Mcrae, G. I. and Stevens, V. C. (1981) *Fertil. Steril.,* **36,** 664.
Wilks, J. W., Hodgen, G. D. and Ross, G. T. (1976) *J. Clin. Endocrinol. Metab.,* **43,** 1261.
Zerega de, G. S. and Hodgen, G. D. (1981) *Endocrinol. Rev.,* **2,** 27.