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THE EFFECTS OF HUMAN CHORIONIC GONADOTROPIN (HCG) ON PROLACTIN RECEPTORS (PRLR) OF CROP SAC EPITHELIUM

M. Ahmad[1], R. Hasan[1], M. Ahmad[2], A. Qureshi[3] and A. Javaid[1]

[1]*Department of Physiology, University of Karachi, Karachi-75270. Pakistan*
[2]*Department of Physiology, New York Medical College,USA*
[3]*Westchester Hospital, Valhalla, New York, USA.*

ABSTRACT

This study deals with the effect of human chorionic gonadotropin (hCG) on crop sac prolactin receptors (PRLR). Direct hypodermal injections of 45 IU hCG per day for 4 days produced a diametric response of 2.28 cm. This indicates that hCG synergises PRL. Since hCG promotes synthesis of pigeon's milk by binding to specific receptors. Whereas, injections of vehicle only administrated control crop sacs produced a sub specific response.

Key words: hCG, prolactin prolactin receptor,

INTRODUCTION

Many drugs that affect central nervous system (CNS) can produce hyperprolactinemia (Turkington, 1972; Kleinberg, 1977). Chlorpromazine, a centrally acting drug has been used to produce increased prolactin (PRL) titres (Ahmad *et al*., 2003) and recently, the non-selective adrenergic drugs have also been reported to produce endogenous hyperprolactinemia (Ahmad *et al*., 2002a,b). This induction may be associated with dopaminergic antagonistic drugs that interfere with the regulation of (PRL) secretion by prolactin release inhibiting hormone (PRIH) with increase in TRH, and through the mediation of oral contraceptives (Macleod, 1976).

It is widely known that (PRL) is involved in a variety of physiological processes (Nicoll, 1974) and there are several recent studies reporting (Ahmad *et al*., 2001a, b; 2003) the production of milk like secretion by the lactiferous cells of crop sac epithelium. The purpose of the present study is to establish the stimulatory effects of hypodermic human chorionic gonadotropin (hCG) on the pigeon crop sac PRL receptors.

MATERIALS AND METHODS

Assay animals:

Pigeons of 8 to 10 weeks age, belonging to white race weighing 320 ± 35 g were used for crop sac assay. They were obtained from local breeders in groups of 6 to 12, housed one to a cage, fed millet and water *ad libitum.* The birds were kept in the laboratory for five days including the period of bioassay.

Drug information:

Chorionic gonadotropin (hCG) is a hormone of human pregnancy; secreted by the syncytiotrophoblasts of foetal placenta as early as 7 days after ovulation. It is absorbed into the blood in sufficient amount to sustain luteal function and forestall the next menstrual period. The secretion of LH remains suppressed because of the rising concentrations of estrogen and progesterone (Lipsett and Ross, 1978).

hCG is detectable in the urine by immunoassay several days before the first missed period, and this is the basis of the most commonly used test of pregnancy. The quantity excreted increases rapidly thereafter to a maximum of about 6 weeks after ovulation. Its urinary content declines over the next month stabilizes at a lower level for the remainder of pregnancy.

The changes in the corpus luteum in early pregnancy reflect the intense stimulation provided by the LH-like action of hCG. Furthermore, concentrations of placental lactogen increase progressively during pregnancy. It is also luteotropic and may play some role with hCG to stimulate steroid production by the corpus luteum. With the increasing secretion of estrogen and progesterone by the palcenta during the third month, the ovaries and the corpus luteum become unessential to the maintenance of gestation; but the corpus luteum does not undergo a pronounced change at this time.

Instead, there is a slow histological regression that is not complete even at the time of delivery. In the presence of flood of hCG during pregnancy; the rest of the ovary remains quiescent with no growth or maturation of follicles and no changes suggesting luteinization either of the granulosa or of thecal or stromal elements.

hCG is a water soluble glycoprotein. The study of highly purified preparations indicates that the hormone may be composed of 2 polypeptide chains. Amino acid analysis of these preparations suggests its molecular weight as 27,000. The nature of the carbohydrate chain is highly complex structure. The specific activity of the hormone is about 12,000 IU/mg. It sufficiently resembles LH in its biological activities.

Currently no purified gonadotropins, prepared from human pituitaries are available commercially. For clinical purposes, quite crude preparations are suitable, even though they contain FSH and LH as well as other active principles. The limiting factor is the scarcity of human pituitary glands and not the preparation of suitable extracts.

Dose administration:

The 45 IU hCG in solution form was administered daily to each pigeon hypodermally for 4 days. The birds were divided into two groups. One group received 45 IU hCG hypodermally and the other received pyrogen free distilled water injections for "sham" assay. Since avian normal saline or even 0.5 sodium chloride in amount as little as 0.5ml produced crop sac milk forming reaction (Lahr *et al*., 1942. Ahmad *et al*., 2001, 2002a,b).

Assay procedure:

Assay procedure was essentially the same as that of Ahmad *et al*. (2002b). Two groups consisting of 12 pigeons each were used for the "sham" and hCG test assay. Feathers were plucked off the skin overlying crop 8 hours prior to injections.

The injections were given for a period of 4 days, using a 1 ml hypodermic syringe bearing a 27 gauge needle. Each injection was given at the centre of each half of crop sac, which was marked earlier with a nontoxic dye. The injections were made with such accuracy that an intradermal bleb was always formed.

The birds were killed on day 5; a day after the last injection. The crop sac epithelium was removed and bisected. The lining of each half was rinsed with tap water and each half was stretched after removing the fat against the light of a table lamp fitted with a 100 watt bulb as a source; while someone measured the diameter of the proliferated area in cm with a caliper on stretching against light; since the proliferated epithelium appeared circularly opaque and was easily measurable.

RESULTS

The crop-sac induction following the hypodermal injections of hCG gave a minimum diametric response of 2.21cm; a maximum of 2.37cm and 2.28 cm as the mean of 24 measurements (Table 2). Whereas, the response to pyrogen free distilled water was 0.83 cm (Table 1). Since the diametric response of less than 2.0 cm and more than 3.85 cm are unreliable to rate (Ahmad et al., 2002 b), the hypodermic administration of 45 IU hCG did produce the diametric response, while injection of pyrogen free distilled water were failed to produce the diametric response. A significant difference between the mean crop sac diametric response was observed between two groups ($P<0.05$, T-test).

Table 1. Crop-sac diametric response following hypodermal injections of pyrogen free double distilled water daily for 4 days.

Pigeons No.	Pigeon's weight (g)	Diametric response (cm)*
1.	318	0.65
2.	335	0.89
3.	336	0.87
4.	340	0.80
5.	325	0.75
6.	345	0.77
7.	350	0.90
8.	316	0.88
9.	330	0.81
10.	326	0.90
11.	345	0.86
12.	348	0.84
Mean ± SD	334.5 ± 11.57	** 0.83 ± 0.07

* Each figure is the mean of right and left crop-sac diametric response.; ** The mean of 24 measurements with ± SD.

Table 2. Crop sac diametric response following direct hypodermic injections of 45 IU hCG daily to each pigeon for 4 days.

Pigeons No.	Pigeon's weight (g)	Diametric response (cm)*
1	325	2.36
2	351	2.37
3	339	2.23
4	340	2.28
5	324	2.21
6	350	2.28
7	332	2.30
8	341	2.28
9	336	2.26
10	334	2.25
11	350	2.29
12	355	2.27
Mean	339 ± 10.19	** 2.28 ± 0.05

* Each figure is the mean of right and left crop-sac diametric response.; ** The mean of 24 measurements with ± SD.

DISCUSSION

Assay of hCG and its subunits are also used to diagnose and to evaluate the treatment of trophoblastic tumors (Vaitukaitis *et al*., 1976). Quantification of secretion of the hormone by choriocarcinomas and hydatidiform moles can provide an accurate index of tumor regression or recurrence. This ability has contributed to the high rate of successful treatment of these tumors.

Moreover its effect is similar to estrogen and LH. However, its estrogenic like action on developing testes can hardly be regarded as physiological. Because the hormone gains access to the male only *in utero,* when it does cause minimal gonadal stimulation. But when injected into man, it stimulates the interstitial cells of testis to secrete androgen quite normally; if descent of testes is not achieved before the age of seven (Lattimer, 1973), it can often be accomplished by administration of hCG. Thus hCG assumes not only PRL and estrogen activity at certain positions but capably promotes a sequence of androgenic events at another position. In the end it can be said pigeon PRLR, are sensitive to hCG and can be successfully used to diagnose drugs for their lactogenic activity.

REFERENCES

Ahmad, M., I. Mahmood, R. Hasan, A. Javaid, T. Naim, H. Fatima and M. Ahmad (2001a). *In vitro* effects of ACTH on pigeon crop-sac epithelium. *Pak. J. Pharm. Sci*., 14: 19-23.

Ahmad, M., I. Mahmood, R. Hasan, H. Fatima and M. Ahmad (2001b). Effect of dexamethasone on pituitary prolactin in the lizard *Uromastix hardwickii*. *Pak. J. Pharm. Sci*., 14: 43-46.

Ahmad, M., I. Mahmood, R. Hasan, H. Fatima, A. Javaid, T. Naim and M. Ahmad (2002a). Hypoprolactinemia following intradermal administration of ouabain in *Uromastix hardwickii*. *Pak. J. Pharm. Sci*., 15: 37-40.

Ahmad, M., I. Mahmood, R. Hasan, T. Naim, A. Javaid, H. Fatima and M. Ahmad (2002b). Bioassay of pituitary prolactin of the lizard, *Uromastix hardwickii*. *Pak. J. Pharmacol*., 19: 7-11.

Ahmad, M., R. Hasan, S. Yameen and M. Ahmad (2003). The effect of centrally acting drug chlorpromazine on the secretion of prolactin in the lizard, *Uromastix hardwickii*. *Pak. J. Pharm. Sci*., 16: 79-83.

Kleinberg, D. L., G. L. Noel and A. G. Frantz (1977). Galactorrhea : a study of 235 cases including 48 with pituitary tumors. *N. Engl. J. Med*., 296: 589-600.

Lahr, E. L., O. Riddle and R. W. Bates (1942). The role of hormone that initiation of maternal behaviour in rats. *Am. J. Physiol*., 137: 299-317.

Lattimar, J. K. (1973). The optiman treatment for undescended testes. *Med. Col. Va. Q*, 9, 270- 274.

Lipsett, M. B. and G. T. Ross (1978). The ovary. In: *The endocrinology* (Ingbar, S. H. ed.) Plenum Med. Book Co. New York. p p. 233-254.

MacLeod, R. M. (1976). Regulation of prolactin secretion. In: Frontiers in neuroendocrinology, vol. 4 (Martini, L. and F. Ganong eds.) Raven Press. New York. p p. 169-194.

Nicoll, C. S. (1974). Physiological actions of prolactin. In: Hand book of Physiology. Endocrinology, section-7. Vol. II, Part-2 (Greep, R. O. and E. B. Astwood eds.) Washington D.C. American Physiol. Soc. p p. 253-292.

Turkington, R. W. (1972). Inhibition of prolactin secretion and successful therapy of the Forbes-Albright syndrome with L-DOPA. J. Clin. Endocroinol. Metab., 34: 306-311.

Vaitukaitis, J. L., G. T. Ross, G. D. Braunstein and P. L. Rayford (1976). Gonadotropins and their subunits: Basic and Clinical studies. Recent Prog. Horm. Res., 32:289-331.

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