The Effect of Progesterone on Plasma Insulin in the Rabbit

Sex steroids have been shown to influence carbohydrate metabolism in many instances in both animal and man[1]. Treatment with progesterone increased pancreatic insulin secretion in the rat[2] and in the monkey[3]. In man, the insulin response during an oral glucose test was increased after progesterone treatment[4], but this could not be confirmed during an i.v. glucose test[5]. In the rabbit, the effect of estradiol on glucose tolerance has been measured but plasma insulin was not determined[6]. We studied the effect of progesterone treatment on the insulin response after an i.v. glucose test in rabbits. The tests were performed during, as well as after, the progesterone treatment. We showed that in the rabbit – as in other species studied so far – progesterone treatment caused increased levels of plasma insulin.

Materials and methods. Two randomized groups of 6 New Zealand white rabbits (2–2.5 kg) were used. 1 group received 5 mg progesterone/kg body weight s.c. in 0.6 ml arachis oil daily. The control group was injected with arachis oil according to the same scheme. After a 2 h fast, a 35% solution of glucose was injected rapidly into the ear vein in a dosage of 1 g/kg. Blood samples from the ear veins were collected in heparinized tubes at 0, 5, 15, 30 and 45 min and placed on ice. Plasma glucose was determined with the glucose oxidase method (Boehringer test kit) and plasma insulin with the double antibody solid phase (DASP) method[7] with pig insulin as standard[8]. Plasma progesterone was determined with a competitive protein binding assay[9].

Glucose disappearance rates were calculated with the least square method from the logarithmic values of the plasma glucose concentrations[10]. The integrated insulin curves were determined by calculating the total area under the response curve above zero level.

Results and discussion. Intravenous glucose tolerance tests were performed after 5 weeks treatment with progesterone and 1 and 7 weeks after treatment had been stopped. Figure 1 shows the plasma glucose and insulin response curves of treated and control rabbits after 5 weeks treatment. Natural progesterone produced no significant change in the rate of glucose disappearance (Table I), but the mean integrated insulin response was significantly higher in the group treated with progesterone than in the control group (Figure 2). The same result was obtained 1 week after progesterone treatment had been stopped. The plasma progesterone levels were still increased 1 week after treatment (Table II).

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Table I. Mean (± SEM) glucose disappearance rates (K) following intravenous glucose

Group	n	K (mg/100 ml/min)		
		During treatment	After treatment	
		(5 weeks)	1 week	7 weeks
Placebo	6	3.00 ± 0.23	2.56 ± 0.21	2.65 ± 0.07
Progesterone	6	3.35 ± 0.20	2.94 ± 0.36	2.94 ± 0.37
		N.S.	N.S.	N.S.

Treatment of groups and calculation of K-values as described in Materials and methods.

Table II. Mean ± SEM plasma progesterone concentrations

Group	n	Progesterone (ng/ml)		
		During treatment	After treatment	
		(5 weeks)	1 week	7 weeks
Placebo	6	4 ± 1	8 ± 3	4 ± 1
Progesterone	6	67 ± 25	26 ± 7	3 ± 1
		$p < 0.005$	$p < 0.005$	N.S.

Progesterone was determined as described in Materials and methods. The P-values were calculated with the Wilcoxon test.

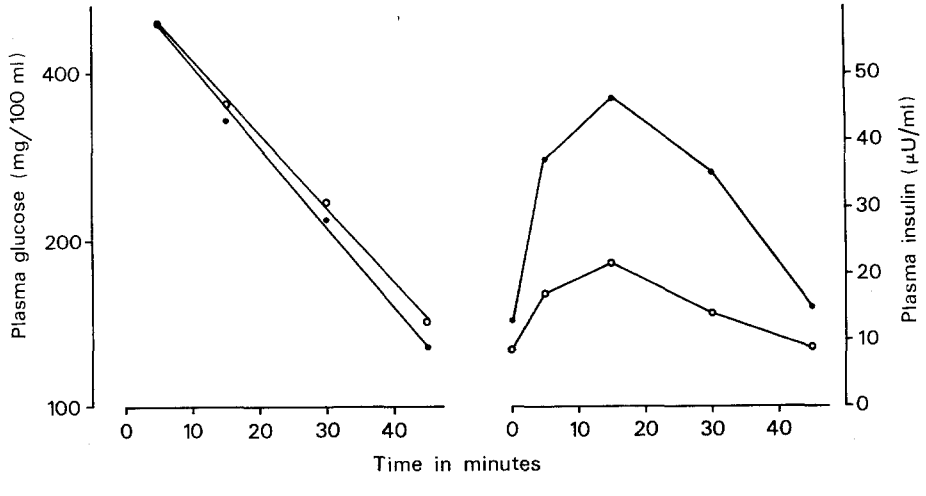

Fig. 1. Plasma glucose and insulin responses to i.v. glucose after 5 weeks progesterone treatment. ○—○, placebo group; ●—●, progesterone group.

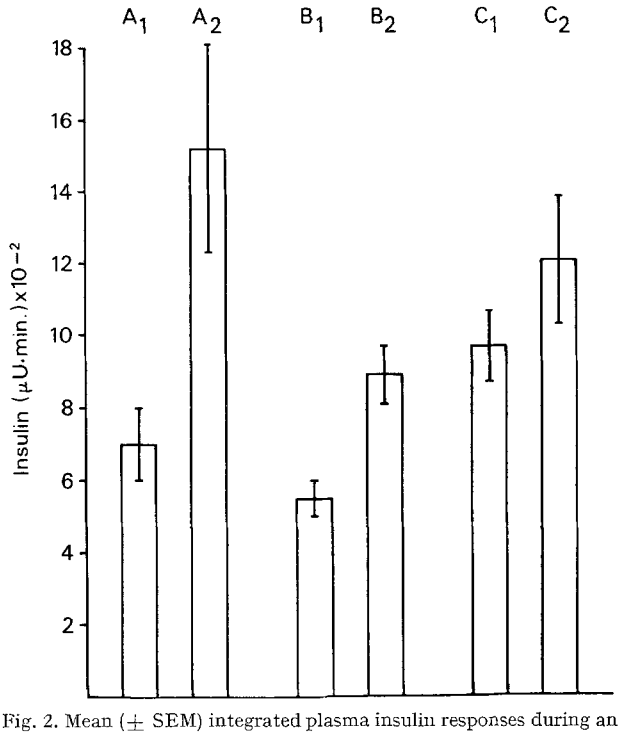

Fig. 2. Mean (± SEM) integrated plasma insulin responses during an i.v. glucose tolerance test in rabbits.
A) after 5 weeks treatment. 1. Placebo group. 2. Progesterone group ($p < 0.05$).
B) 1 week after treatment had been stopped. 1. Placebo group. 2. Progesterone group ($p < 0.01$).
C) 7 weeks after treatment had been stopped. 1. Placebo group. 2. Progesterone group (N.S.). The P-values were calculated with the permutation test[12].

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These results agree with the reported effects of progesterone on insulin responses during glucose tolerance tests[2,3]. It has been shown that progesterone treatment augments the plasma insulin response without having any apparent effect on glucose tolerance. One explanation is that progesterone induces β-cell hyperplasia while at the same time it is an insulin antagonist, causing impaired peripheral tissue utilization of glucose[2]. The fact that progesterone inhibits the insulin effect on glucose uptake of rat diaphragm in vitro[11] supports this suggestion.

Seven weeks after progesterone treatment had been stopped, there was no significant difference between the integrated insulin responses of treated and control rabbits. This shows that the progesterone effect is fully reversible. The progesterone levels were also normalized after 7 weeks (Table II). There could be a correlation between the rate in drop of progesterone levels and the normalization of the insulin responses but no attempt was made to study this question.

Résumé. Un traitement pendant 5 semaines avec de la progesterone a provoqué une augmentation significative de la réponse insulinique à l'injection i.v. de glucose chez la lapine. Une semaine après la fin du traitement, les taux de progesterone et la réponse insulinique étaient encore élevés. 7 semaines après le traitement la réponse insulinique était de nouveau normale, montrant que l'effet de la progesterone était entièrement réversible.

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11 December 1974.

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Water Deprivation in Rats: Elevated Plasma Neurophysin Levels

Water deprivation in rats is a potent stimulus to the hypothalamo-neurohypophysial system and leads to release of both oxytocin and vasopressin[1]. The availability of specific radioimmunoassays for oxytocin, vasopressin and their carrier proteins, the neurophysins, led us to reinvestigate the effects of prolonged water deprivation on the amounts of these substances present in the rat neural lobe. Plasma neurophysin levels were also determined in the same animals in order to ascertain whether they were elevated by the stimulus and in order to study, in an indirect way, the kinetics of neurohypophysial hormone secretion.

Adult, male rats of the SIV 50 strain (Sprague-Dawley derived) weighing between 230–310 g were kept 5 to a cage and allowed food ad libitum (Nafag 850 pellets). 28 animals used as controls had, in addition, free access to tap water. 10 groups of 10 animals each were deprived of drinking water for periods ranging from 1 to 10 days. At the end of the period of water deprivation, the animals were decapitated. Each neurohypophysis was rapidly removed, separated from adjacent tissue and transferred to a Pyrex tube containing 2 ml of distilled water for homogenization. Insoluble material in the homogenate was removed by centrifugation and the supernatant used for radioimmunoassay. Blood was collected from the bodies immediately after decapitation and centrifuged for blood haematocrit determination. The plasma was used for measurement of osmotic pressure and chloride concentration, as well as for determination of immunoreactive neurophysin levels.

Neurophysin levels were determined by radioimmunoassay using a cross-species reactive antibody (A_5IV) raised against bovine neurophysins; purified bovine neurophysin II served as standard[2]. Oxytocin and vasopressin were assayed using specific antibodies according to previously described methods[3].

After 1 day of water deprivation, the amounts of immunoreactive oxytocin and vasopressin present in the neural lobe did not differ greatly from those of control rats. A slight increase in vasopressin content and a decrease in oxytocin content were seen, but neither were statistically significant. With longer periods of removal of water, a progressive fall in the gland content of both

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