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Tohoku Journal of Agricultural Research
Vol. 45 No. 1-2, September 1994
Printed in Japan

Adrenergic Effects on Pancreatic Glucagon and Insulin Secretions in Rabbits

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(Received, July 30, 1994)

Summary

Adrenergic modulation of glucagon and insulin secretions were examined using an adrenergic agonist and adrenergic antagonists in conscious rabbits. Epinephrine infusion caused increases in plasma glucagon and glucose concentrations but produced no change in the plasma insulin concentration. Plasma glucagon levels were increased by the concomitant infusion of phentolamine and epinephrine, and were not changed by propranolol and epinephrine infusion. The plasma insulin concentrations were suppressed by α-adrenergic stimulation more than β-adrenergic stimulation. Hyperglycemia was induced by both α-and β-adrenergic stimulation but the extent was greater under β-adrenergic stimulation than α-adrenergic stimulation.

It is concluded that the β-adrenergic receptor mechanism is an important component of adrenergic modulation of pancreatic glucagon secretion in conscious rabbits.

Introduction

There is general agreement that the sympatho-adrenomedullary system plays an important role in controlling the activity of insulin secretion from the pancreatic B cell and pancreatic glucagon secretion from the pancreatic A cell. It has been confirmed in various species under different physiological conditions, that β-adrenergic stimulation enhances and an α-adrenergic mechanism suppresses insulin secretion.

There are, however, conflicting results concerning the adrenergic modulation of glucagon secretion as to whether secretion was stimulated through an α-adrenergic mechanism as noted in dogs (1), calves (2), rats (5), rabbits (10), goats (14) and sheep (11, 15-17, 20), or a β-adrenergic mechanism as noted in rats (7, 13) and dogs (8, 9). It has also been reported that both α-and β-adrenergic receptors influenced glucagon secretion in rats (12). Thus, it seems conceivable that species differences in addition to some differences in experimental design may account for

the conflicting results relating to the adrenergic modulation of glucagon secretion. Adrenergic modulation of glucagon secretion could be consistently mediated through an α-adrenergic mechanism in ruminant animals, at least in sheep and in goats.

Therefore, in the present work, using rabbits, we studied in detail the effects of α-and β-adrenergic receptor activation on the secretory response to both glucagon and insulin to obtainfurther information about the adrenergic modulation of glucagon secretion under conscious states.

Materials and Methods

Animals: Japanese White male rabbits, weighing 2.6~3.5 kg, were used. The rabbits were housed in individual cages and maintained on a commercial laboratory chow (Oriental Yeast Co., LTD., RC4). Water was available continuously. Air temperature was 20 ± 5°C. At least 1 day before the experiments, a catheter (ATOM Indwelling tube, 5Fr, ATOM, Tokyo) for blood sampling was inserted into a carotid artery under general anesthesia with pentobarbital sodium (25 mg/kg).

Experimental proceduras: On the morning of an experiment, feed and water were withdrawn, and 1 hour before sampling, a polyethylene catheter for infusion was inserted into an auricle vein. All infusions were given via the auricle vein catheter at a constant rate of 0.1 ml/min using an peristatic pump. Each rabbit was used in the following experiments in the conscious state. The experiments were performed at 2-day intervals, with one infusion per day.

Epinephrine infusion: (−) Epinephrine bitartrate (Sigma, lot 100F-0356) dissolved in sterile saline was infused at a rate of 1.0 n mol/kg/min for 40 min. Sterile saline was infused for the control experiment.

Phentolamine plus epinephrine infusion: The α-adrenergic antagonist, phentolamine (Regitine, Ciba-Geigy) was infused at a rate of 100 n mol/kg/min. Phentolamine infusion was started 10 min before the onset of an epinephrine infusion, and it continued for a period of 40 min. Epinephrine was infused for 30 min at a rate of 1.0 n mol/kg/min.

Propranolol plus epinephrine infusion: The β-adrenergic antagonist, propranolol (DL-propranolol hydrochloride, ICI Pharmaceuticals) dissolved in sterile saline was infused at a rate of 200 n mol/kg/min. Propranolol infusion was started 10 min before the onset of an epinephrine infusion, and it continued for a period of 40 min. Epinephrine was infused for 30 min at a rate of 1.0 n mol/kg/min.

The timing of blood samples is indicated in figures.

Analyses: Arterial blood samples were taken in heparinized syringes and transfered into centirfuge tubes cooled in ice water. The tubes for the glucagon

samples cantained benzamidine (50 μ mol/ml blood, Sigma) as a proteolytic inhibitor. These tubes were centrifuged at 4°C. A portion of plasma was deproteinized with 50 g/liter trichloroacetic acid, and the supernatant was stored at−25°C before glucose estimation by a glucose oxidase method (6). The plasma samples were stored at−25°C for hormone assays. Insulin was assayed by the method previously reported (14), using rabbit insulin as the standard (Novo, rabbit insulin, Lot K13369). Pancreatic glucagon was assayed by a method (18) using dextran-coated charcoal and an antiserm (G42-E) which was highly specific for the C-terminal portion of glucagon (19).

Statistics: The results are expressed as mean±SE of mean (n=5). The significance of differences between preinfusion values and subsequent values were determined by Student's t-test.

Results

Effect of epinephrine infusion on glucagon and insulin secretion in rabbits

The plasma glucagon concentration was significantly increased during epine-

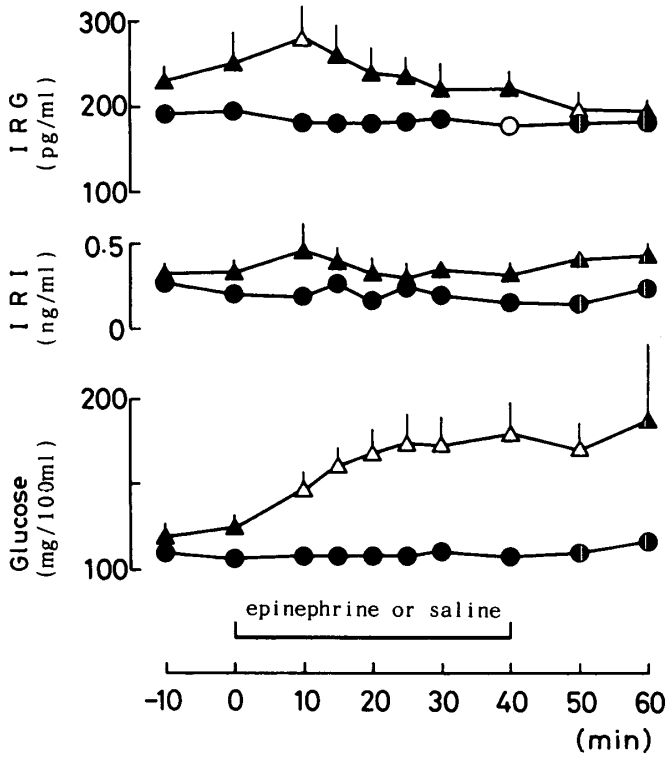

FIG. 1. Effect of epinephrine infusion on glucagon and insulin secretion in rabbits. Five rabbits each received auricle vein infusions of epinephrine (▲) at a rate of 1.0 nmol/kg/min or saline (●) for 40 min (horizontal bar). Mean values for 5 rabbits±SEM are shown. Open symbols represent significant differences between preinfusion values and subsequent values ($P<0.05$).

phrine infusion from a mean pre-infusion value of 240±27 pg/ml to 281±39 pg/ml at 10 min ($P<0.05$) and after then gradually decreased. As shown in Fig. 1, the plasma insulin concentration was maintained at pre-infusion levels (<0.46 ng/ml), and was not significantly different during the experiment. Epinephrine produced a marked increase in plasma glucose concentratins ($P<0.05$) from 122± 7.3 mg/100 ml to 187±44 mg/100 ml at 60 min.

Effects of adrenergic receptor blockade on glucagon and insulin secretion during epinephrine infusion

As shown in Fig. 2, the concentration of plasma glucagon increased during the concomitant infusion of phentolamine and epinephrine, α-adrenergic blockade, from 132±5 pg/ml to 187±17 pg/ml at 5 min ($P<0.05$) and was maintained at high levels throughout the experiment. The concentration of plasma glucagon was, however, unchanged by the concomitant infusion of propranolol and epinephrine, β-adrdnergic blockade.

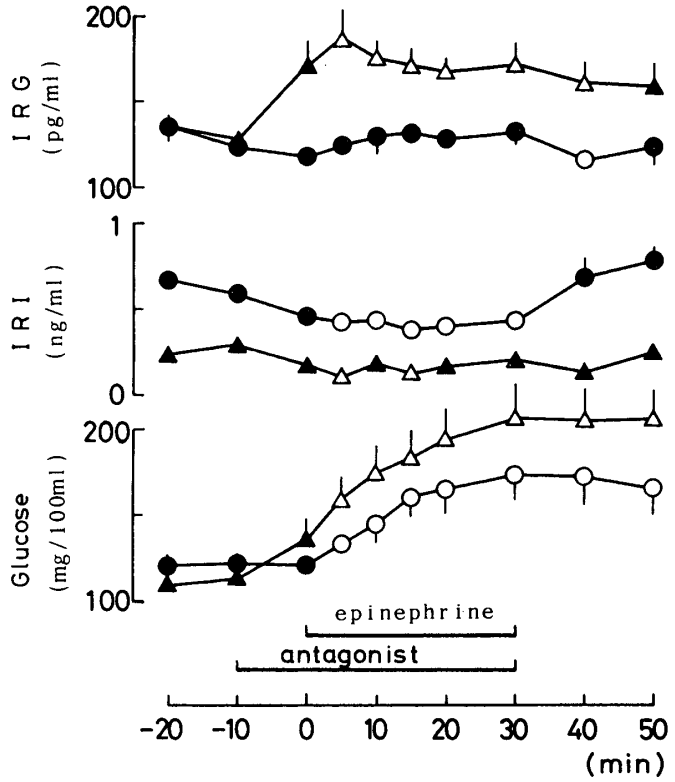

FIG. 2. Effect of concomitant infusion of antagonist and epinephrine on glucagon and insulin secretion in rabbits. Five rabbits each received auricle vein infusions of phentolamine and epinephrine (▲) at a rate of 100 nmol/kg/min and 1.0 nmol/kg/min or propranolol plus epinephrine (●) at a rate of 200 nmol/kg/min and epinephrine for 40 min (horizontal bar). Mean values for 5 rabbits±SEM are shown. Open symbols represent significant differences between preinfusion values and subsequent values ($P<0.05$).

The plasma insulin concentration was more effectively suppressed by the propranolol and epinephrine infusion than the phentolamine and epinephrine infusion.

A massive hyperglycemia was brought about by the concomitant infusion of phentolamine and epinephrine, reaching a highest value of 208 ± 20 pg/ml at 30 min ($P < 0.01$). The propranolol and epinephrine infusion also produced a significant increase ($P < 0.05$) in the plasma glucose concentration but the extent was less than that noted in the phentolamine and epinephrine infusion.

Discussion

It has been reported that epinephrine administration induces hyperglucagonemia, hypoinsulinemia and hyperglycemia in various species. However, the results of the present experiments show that pancreatic glucagon was released through β-adrenergic stimulation but was not affected by α-adrenergic stimulation in conscious rabbits. The result that glucagon secretion was increased by β-adrenergic stimulation, is in good agreement with other species, for example humans (4), conscious dogs (3), anesthetized dogs (8, 9) and rats (7, 13). Not withstanding this, it has also been reported that α-adrenergic stimulation augments glucagon secretion in ducks (21), exercised rats (5), fasted rabbits (10), hypoxic puppies (1), calves (2), goats (14) and sheep (11, 15-17, 20). It has also been reported that both α-and β-adrenergic receptors influenced glucagon secretion in rats (12). There is no general consensus about the adrenergic mechanisms regulating glucagon secretion, as to whether the secretion is mediated through α-or β-adrenergic effects. Cold stress, however, did not change the pattern of α-adrenergic stimulation of glucagon secretion in sheep (20), suggesting that α-adrenergic stimulation induced glucagon release may relate to some differences in physiological condition and stress except for in sheep.

There is general agreement that the secretion of insulin is stimulated by β-adrenergic activation and inhibited by α-adrenergic stimulation in many species including the rabbit (10). In this experiment, however, the plasma insulin concentration was more effectively suppressed by α-adrenergic stimulation than β-adrenergic stimulation. The β-adrenergic stimulation induced a slight decrease in insulin secretion. If the rabbits were under some stress, the sympathoadrenomedullary system and exogenous catecholamine might strongly suppress insulin secretion. We are unable to explain the slight suppression of insulin release by β-adrenergic stimulation in detail because the antagonists were only administered at one dose.

Hyperglycemia is caused by increasing glucagon and decreasing insulin secretions, stimulating hepatic glycogenolysis and gluconeogenesis by epinephrine and inhibiting peripheral glucose utilization. Hyperglycemia was induced by

both α-and β-adrenergic stimulation. β-adrenergic stimulation, however, caused hyperglycemia to a greater extent than the α-adrenergic stimulation. These effects were related to the no change of glucagon levels and strong suppression of insulin release by α-adrenergic activation. The hyperglycemia during the α-adrenergic stimulation was mainly induced by epinephrine on hepatic glycogenolysis and gluconeogenesis.

Acknowledgements

The authors are grateful for skillful technical assistance from Mr. Y. Ohtomo, Laboratory of Animal Physiology, Tohoku University, Sendai, Japan. We are also grateful for the comments on the manuscript by M.T. Rose, National Institute of Animal Industry, Tsukuba, Japan.

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