Afr. J. Med. med. Sci. (1997) 26, 75-78

The role of adrenergic receptors in the increased glucose uptake by the canine gut

A.R.A. Alada and D.D.O. Oyebola *

Department of Physiology, College of Medicine, University of Ibadan, Ibadan, Nigeria

Summary

The study was carried out on fasted anaesthetized dogs. A vein draining a segment of the upper jejunum was cannulated for blood flow measurement. Arterial blood and venous blood from the upper jejunal segment, were obtained for measurement of glucose content. Glucose uptake was calculated as the product of jejunal blood flow and the difference between arterial and venous glucose levels. Prazosin, (0.2 mg/kg) had no effect on the glucose uptake induced by insulin and glucagon injections and glucose infusion. However, prazosin reduced the glucose uptake by about 50% during adrenaline-induced hyperglycaemia. Pretreatment of the animal with propranolol (0.5 mg/kg) significantly reduced the increased glucose uptake caused by adrenaline, glucagon and glucose hyperglycaemia. Propranolol also reduced by half the negative glucose uptake during insulin-hypoglycaemia. Since both the alpha and beta adrenergic receptor blocking agents did not abolish the induced glucose uptake, it was concluded that the effects of adrenaline, glucagon, glucose and insulin on intestinal glucose uptake are mediated in part by beta adrenergic receptors.

Résumé

L'étude a été faite sur des chiens a Jeune et anesthesié. Une veine alimentant un segment superieur du Jejunum a été canulé, afin de mesurer le debit sanguin. Le sang arteriel et veineux du segment superieur du segment jejunal, a été obtenue afin d'y mesuré le taux de glucose. Le passage du glucose dans le sang a été calculé comme produit du flux sanguin jejunal, et la difference entre les taux de glucose arteriel et veineux. La prazosin (0.2 mg/kg) ná eut aucun effet sur la penetration du glucose induite par l'insuline, sur les injections de glucagons, et l'infusion de glucose. Cependant, la prazosin a reduit la pénétration de l'ordre de 50% pendant l'hypoglycemie induite par l'adrenaline. Le prétraitement des animaux avec du propanolol (0.5 mg/kg) a réduit de manière significative la forte pénétration de glucose causé par l'adrenaline, le glucogon, et l'hyperglycemie du glucose. Le propranolol a aussi reduit de moitié, la non-penetration du glucose pendant l'hyperglycemie due a l'insuline. D'autant plus que, les agents bloquant les recepteurs alpha et Beta adrenergique n'ont pas abolit l'induction de la penetration du glucose, il a été conclut que les éffets de l'adrenaline, du glucagon, du glucose, et de l'insuline sur la pénétration du glucose intestinal sont due a la mediation de ces recepteurs adrenergique beta.

Introduction

It is now well established that the gastro-intestinal tract (g.i.t.) is capable of taking up large quantities of glucose [1,2,3 and 4]. The glucose uptake by the g.i.t. was found to be far in excess of what can be accounted for by oxidative metabolism [1,2]. Also, glucose uptake by the g.i.t. was increased several fold by adrenaline-induced hyperglycaemia [1,2 and 4]. In a recent study, Alada and Oyebola [5] showed that increased glucose uptake was exhibited by the canine intestine not only in response to adrenaline-induced hyperglycaemia, but also in response to hyperglycaemia caused by administration of glucagon or by glucose infusion. In addition, Alada and Oyebola [5] showed that the canine gut released glucose into the blood stream when blood glucose level was reduced to below normal by insulin injection. The conclusion was reached in that study that the gastro-intestinal tract plays a role in blood glucose homeostasis.

Although Grayson and Oyebola [2,3] showed that alpha and beta adrenergic receptors mediated the increased glucose uptake in response to adrenaline-induced [2] and nicotine-induced [3] hyperglycaemia, Alada and Oyebola [5] did not report on the mechanisms of the increased glucose uptake in response to hyperglycaemia induced by adrenaline, glucagon or glucose; or the negative glucose uptake (i.e., glucose release into circulation) in response to insulin-induced hypoglycaemia. Could the adrenergic receptors also be involved in the latter responses? The need to provide an answer to this question warranted the present study.

Correspondence: Prof. D.D.O. Oyebola, Department of Physiology, College of Medicine, University of Ibadan, Ibadan Nigeria.

Materials and methods

The experimental procedure is similar to that described earlier [5]. Mongrel dogs of either sex weighing 11-15 kg were used for the study. Each animal was fasted for 18-24 h before the start of an experiment. Anaesthesia was induced by intravenous (i.v.) sodium pentobarbitone, 30 mg/kg. Light anaesthesia was maintained with supplementary does of i.v. sodium pentobarbitone as necessary. The trachea was intubated using a Y-piece cannula and the animal was allowed to breathe room air (temp. 25°C) spontanepusly. A cannula was placed in the carotid artery to monitor arterial blood pressure (BP) using a pressure transducer connected to a dynograph recorder (Beckman R511A). Cannulae were also placed in the right femoral vein and right femoral artery. The latter was advanced to the level of the superior mesenteric artery.

Through a midline laparotomy, a vein draining the proximal segment of the jejunum was cannulated using a 1.8 mm (i.d) polyethylene tubing (B.E. 260). The jejunal vein cannula was moved into an extra-corporeal position and a non-crushing clamp was applied to its free end. Sodium heparin, 300 units/kg was administered i.v. to prevent blood clotting. The abdomen was closed in two layers with interrupted sutures.

Following surgery, a period of 60 min was allowed for stabilization in all animals. Blood pressure and jejunal segment blood flow were continuously monitored. Jejunal blood flow was determined by timed collection as previously described by Oyebola and Durosaiye (4). Arterial and venous blood samples for glucose estimation were obtained from the femoral artery and jejunal venous cannulae, respectively.

Experimental procedures

Pretreatment with alpha adrenoceptor blockers (Group 1)

Five dogs were first given prazosin before adrenaline injection. Each dog was injected i.v. with prazosin, 0.2 mg/kg. Forty minutes was allowed for the drug to take effect. Then, basal recording of blood pressure, blood flow and collection of arterial and venous blood samples for glucose estimation were made. After the basal recordings and blood sample collection, adrenaline, 5 μg/kg was given i.v. as a bolus injection. The blood pressure, blood flow, arterial and venous blood glucose were similarly monitored at intervals for 90 min during the post-injection observation period. The

experiment was repeated in another three subgroups (with five dogs per subgroup) using glucagon injection (8 μg/kg), glucose infusion (20 mg/kg/min) or insulin injection (8 iu./kg) in place of adrenaline.

Pretreatment with B-adrenoceptor blockers (group II)

Five dogs were also first injected with propranolol before adrenaline injection. Each animal was given i.v. injection of propranolol, 0.5 mg/kg. After forty minutes and basal recording of blood pressure, jejunal blood flow and sample collection for arterial and venous blood glucose, adrenaline, 5 μg/kg was given i.v. as a bolus injection. Similar measurements to those used in group 1 were made. Again, another three subgroups (of five dogs per subgroup) were studied with propranolol pretreatment but using glucagon injection (8 μg/kg), glucose infusion (20 mg/kg/min) or insulin injection (8 iu/kg), respectively, instead of adrenaline.

Pretreatment with combined α—and B-adrenoceptor blockers (group III)

Five dogs were first given a combination of prazosin and propranolol before injection of adrenaline. Each animal was given an i.v. injection of both prazosin, 0.2 mg/kg and propranolol, 0.5 mg/kg. Forty minutes was allowed for the drug to take effect and then basal recordings and measurements were made as in the other groups. Then, adrenaline injection, 5 ug/kg was given i.v. and the blood pressure, blood flow and arterial and venous blood glucose were monitored during the postinjection observation period. The experiment was repeated on another two subgroups (with five dogs in each subgroup) using glucagon injection or glucose infusion instead of adrenaline.

Blood glucose was determined by the glucose oxidase method as modified by Trinder (6). Glucose uptake (mg/min) was calculated as the product of the arterio-venous glucose difference (A-V) and the jejunal blood flow per minute.

All values given are the mean ± S.E. of the variables measured. Significance was assessed by the Student's *t*-test for two means of independent variables. *P* values of 0.05 or less were taken as statistically significant.

Results

Injections of adrenaline, glucagon, and an infusion of glucose caused great increases in blood glucose levels (arterial and venous), blood flow, and glucose uptake while insulin injection caused an increase in blood flow, a reduction in blood glucose levels (arterial and venous), and a negative glucose uptake. The magnitude and time course of these effects are essentially the same as have been published in an earlier communication (5).

The effects of alpha blockade, beta blockade, and combined alpha and beta blockade on glucose uptake in the canine gut are shown in figures 1,2,3 and 4. Table 1 shows the blood glucose changes in the untreated and adrenoceptor blocker treated subgroups given adrenaline injections.

Effect of α—adrenergic blocker

Prazosin, 0.2 mg/kg significantly reduced the peak of adrenaline induced hyperglycaemia (Table 1). It also reduced the increase in intestinal glucose uptake following adrenaline injection (Figure 1a). Prazosin, however, had no effect on the increase in blood glucose induced by glucagon injection or glucose infusion. Prazosin also had no effect on the increased glucose uptake by the gut induced by glucagon or glucose administration. Pretreating the animal with prazosin had no effect on insulin-induced hypoglycaemia and negative glucose uptake by the intestine.

Effect of B-adrenergic blocker

Propranolol, 0.5 mg/kg increased the resting blood glucose levels (Table 1). It also increased the resting arterio-venous (A-V) glucose difference and glucose uptake.

Propranolol significantly reduced the adrenaline-induced hyperglycaemia. Adrenaline induced increase in glucose uptake by the intestine was also significantly reduced by propranolol. The increase in glucose uptake decreased from about 700% in the untreated animal to about 90% following propranolol pretreatment (Fig.1b). Propranolol had no effect on blood glucose increase produced by glucagon. It however caused a significant decrease in glucagon-induced increase in glucose uptake (Fig. 2a).

Table 1: Effect of iv injection of adrenaline (5 μg/kg) on arterial glucose levels in untreated, α - blocked, β - blocked, and α - and β - blocked dog (N = 5)

	0 min	5 min	10 min	15 min	20 min	25 min	30 min	45 min	60 min	75 min	90 min
Untreated (mg/dl)	107.50 ± 11.16	106.50 ± 15.11	113.00 ± 14.33	179.25 ± 12.78	182.25 ± 10.99	173.00 ± 12.08	115.25 ± 15.57	138.50 ± 8.92	127.25 ± 16.20	118.25 ± 10 36	115.50 ± 8.22
Alpha-block (mg/dl)	106.80 ± 8.76	105.00 ± 10.21	108.53 ± 11.84	***113.82 ± 10.66	***128.09 ± 11.43	***130.55 ± 11.68	126.52 ± 10.75	132.31 ± 10.75	126.01 ± 11.24	119.20 ± 11 50	116.77 ± 9.82
Beta block (mg/dl)	*119.30 ± 3.66	*119.54 ± 5.50	112.63 ± 8.24	***121.70 ± 10.67	***133.20 ± 9.18	***125.41 ± 8.31	***118.28 ± 8.03	122.00 ± 7.95	119.16 ± 8.64	120.50 ± 6.10	113.36 ± 7.48
Alpha and Beta Block (mg/dl)	106.20 ± 8.04	106.00 ± 10.60	107.10 ± 6.50	***110.35 ± 7.75	***108.30 ± 6.20	***111.62 ± 10.89	***106.40 ± 7.25	***108.64 ± 9.70	** 106.48 ± 5.18	110.37 ± 7 29	107.54 ± 5.50

* - $P < 0.05$, ** - $P \leq 0.01$, *** - $P < 0.001$

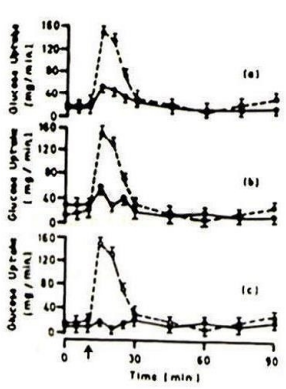

Fig. 1: Effects of i.v. injection of adrenaline on intestinal glucose uptake during (a) - block (b) B-block and (c) and B-blockade. (0--0- untreated, (0-0- pretreated with blocker.

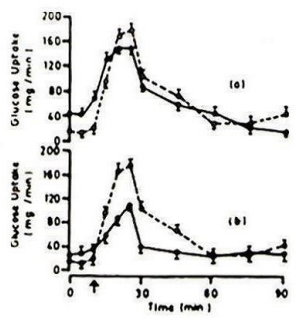

Fig. 2: Effect of i.v. injection of glucagon on intestinal glucose uptake during (a) B-blockade and (b) -and B-blockade. (0--0. -- untreated, M---M - pretreated.

Although the intestinal glucose uptake in propranolol-treated animals increased from 24.71 ± 4.17 mg/min to 72.81 ± 9.34 mg/min (about 300% increase) after infusion of glucose, this increase is significantly lower than that produced in untreated animals (Figure 3a). That is to say, propranolol significantly reduced the glucose uptake induced by glucose infusion.

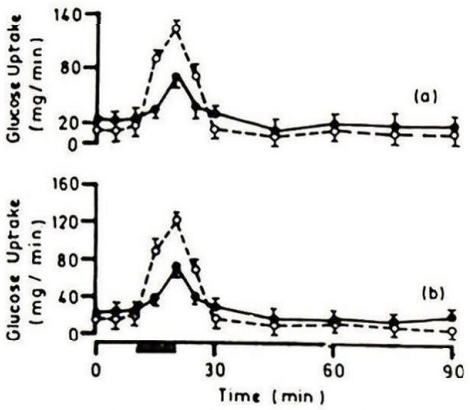

Fig. 3: Effects of intravenous infusion of glucose on intestinal glucose uptake during (a) B-blockade and (b) and B-blockade. (0--0 - untreated, M---M - pretreated).

The effect of insulin on intestinal glucose uptake after propranolol pretreatment is shown in Figure 4. Propranolol reduced significantly ($P < 0.001$) but did not abolish the insulin-induced negative glucose uptake. From a maximum value of - 70.78 ± 6.16 mg/min, the negative glucose uptake was reduced to - 34.15 ± 4.01 mg/min.

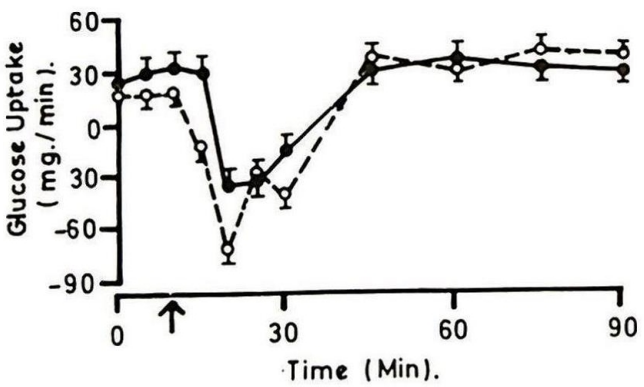

Fig. 4: Effect of i.v. injection of insulin before (0--θ) and after B-blockade on intestinal glucose uptake

Effects of combined α and B-adrenergic blockers

A combination of prazosin, 0.2 mg/kg and propranolol 0.5 mg/kg completely abolished adrenaline-induced increase in blood glucose levels (Table 1) and intestinal glucose uptake (Figure 1c). However, the combined and B-adrenergic receptor blockers only reduced but did not abolish glucagon or glucose-induced increase in intestinal glucose uptake. (Figures 2b and 3b). A combination of prazosin and propranolol did not affect the increase in blood glucose produced by glucagon or glucose.

Discussion

The effects of adrenaline, glucagon, glucose and insulin on intestinal blood flow, blood glucose levels and glucose uptake have been well discussed in our recent study (5). That the hyperglycaemia caused by adrenaline injection, glucagon injection and glucose infusion induced an increase in the gut's glucose uptake and that insulin-induced hypoglycaemia produced a negative glucose uptake by the gut, i.e., the intestine pushed out glucose in response to the hypoglycaemia is consistent with our recent findings (5).

The mechanism of the increased intestinal glucose uptake in this study is partly similar to that reported in earlier studies. Thus propranolol caused an increase in resting jejunal glucose uptake; a result similar to those earlier reported [2,4]. The effect of adrenaline after propranolol-treatment in the present study is similar to the observation reported in the canine jejunum [2] and in the canine terminal ileum [4].

The significant reduction of the large hyperglycaemic response to adrenaline injection by prazosin was a matter of great interest. Although studies in normal hypertensive patients receiving prazosin have not revealed abnormalities in carbohydrate metabolism [7,8,9], there is evidence which indicates that hepoatic glycogenolysis in rats and some other animal species is mediated by alpha adrenoceptors [10,11,12]. Other workers have shown that in man [13] and cat [14] the hyperglycaemic response to adrenaline was abolished by a combination of alpha and beta blocking agents. The latter observations are consistent with the results of the present study, whereby a combination of prazosin and propranolol abolished the effects of adrenaline on blood glucose and intestinal glucose uptake. The mechanism of adrenaline-induced hyperglycaemia and increased glucose uptake by the gut thus seem to involve alpha and beta adrenoceptors. This is consistent with earlier reports [2,3]. From the results of the present study, alpha and beta receptors are almost equipotent in their effects on adrenaline-induced hyperglycaemia (Table 1) and glucose uptake (Figure 1a and b).

The reduction by propranolol of the increase in glucose extraction and intestinal glucose uptake produced by glucagon injection suggests the involvement of beta adrenoceptors in the effect of glucagon on glucose uptake by the bowel. Failure of alpha blockers to alter these variables shows the non-involvement of alpha-adrenoceptors in the response to glucagon.

Perhaps the most relevant results in this study on the mechanism of increased glucose uptake by the bowel is the effect of B-blockade. While prazosin had no effect on the glucose-induced hyperglycaemia and increased glucose uptake, propranolol significantly reduced same. These findings suggest that the increase in intestinal glucose uptake in response to high blood glucose levels, irrespective of its cause is mediated partly by beta-receptors. Some other receptors are most probably involved since B-blockade alone did not abolish increased uptake. Alpha-receptors, unlike in the adrenaline response, are not involved in the increased uptake induced by glucagon and glucose infusion. Also, the findings in the insulin experiments suggest that the beta adrenergic receptors mediated in part the increased negative glucose uptake induced by insulin. Again, alpha receptors are not involved.

In conclusion, the present study showed that alpha receptor blockers have no effect on the increased glucose uptake in response to hyperglycaemia induced by glucagon injection and glucose infusion but it reduced the glucose uptake in adrenaline-induced hyperglycaemia.

The present study however clearly demonstrated that the increased glucose uptake by the gastro-intestinal tract during hyperglycaemia induced by injection of adrenaline, glucagon or glucose infusion and during insulin-induced hypoglycaemia is mediated in part through B-adrenoceptors.

References

1. Durotoye AO and Grayson J. Heat production in the gastrointestinal tract of the dog. J Physiol (1971); 214:417-426.

2. Grayson J and Oyebola DDO. Effect of catecholamines on intestinal glucose and oxygen uptake in the dog. J Physiol (Lond.) 1983; 343:311-322.
3. Grayson J and Oyebola DDO. Effect of nicotin on blood flow, oxygen consumption and glucose uptake in canine small intestine. Brit J Pharmacol 1985; 85:797-804.
4. Oyebola DDO and Durosaiye GO. Effect of adrenaline and propranolol on glucose uptake in the canine terminal ileum. Nig J Physiol Sci 1988; 4:31-37.
5. Alada ARA and Oyebola DDO. Evidence that the Gastrointestinal tract is involved in Glucose Homeostasis. Afr J Med and med Sci 1996; 25: 243-249.
6. Trinder P. Determination of blood glucose using 4-aminophenazone as oxygen acceptor. J Clin Path 1969; 22:246-248.
7. Thulin T, Sactre H, Vickesdahl O, Warmenius Co. S, Persson G, and Sherston B. Multicentre study of the antihypertensive effect of prazosin hydrochloride on wild and moderate hypertension. Excepta medica Int Congre Sci 1974; 331:126-127.
8. Pitts NE. The clinical evaluation of prazosin hydrochloride, a new antihypertensive agent. Excepta Medica Int Congr Sci 1974; No. 17, 331:149-163.
9. Wibell L, Berne C, Waern AU and Litheu H. Prazosin influence on carbohydrate and lipoprotein metabolism. R Soc Med Int Congr Ser 1980; No. 41:85-89.
10. Sherline P, Lynch A and Glinsman WH. Cyclic-AMP and adrenergic receptor control of rat liver glycogen metabolism. Endocrinology 1972; 91:680-690.
11. Hutson NJ, Brumley FI, Assimacoponlos FD, Harpers C and Extor JH. Studies on alpha adrenergic activation of hepatic glucose output. J Biol Chem 1976; 251:5200-5208.
12. Blair JB, James ME and Foster JL. Adrenergic control of glucose output and ademosin $3^{1}5^{1}$ monophosphate levels in hepatocytes from juvenile and adult rats. J Biol Chem 1979; 254:7579-7584.
13. Antonis A, Clark ML, Hodge RL, Moloney M and Pilkington TRE. Receptor mechanism in the hyperglycaemia response to adrenaline in man. The Lancet 1967, 1:1135-1137.
14. Al-Jibonri LM, Furman, BL and Parrat JR. Blockade of adrenaline-induced hyperglycaemia in the anaesthetised cat by continuous of infusion of phentolamine and propranolol. Br J Pharmacol 1980; 68:461-466.