Effect of Glucagon on Intravenous Glucose Tolerance

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The plasma insulin response to intestinally administered glucose is greater than that elicited by the same quantity of glucose given intravenously (McIntyre *et al.*, 1965). McIntyre and his colleagues suggested that the most likely explanation is the release from the intestine of a humoral factor which stimulates insulin secretion. The identity of this hormone remains speculative, but the intestinal hormones secretin (Pfeiffer *et al.*, 1965; Dupré *et al.*, 1966; Unger *et al.*, 1966b), pancreaticozym (Ketterer *et al.*, 1966), and glucagon (Samols *et al.*, 1965a, 1965b) have all been reported to stimulate the release of insulin.

Glucagon is an attractive possibility. Its presence in the gastrointestinal tract has been indicated by biological (Kenny and Say, 1962; Makman and Sutherland, 1964) and immunochemical techniques (Samols *et al.*, 1966; Unger *et al.*, 1966a). Samols *et al.* (1965a) demonstrated that the injection of 1 mg. of glucagon into normal fasting humans produced a rise in plasma insulin that could not be accounted for by elevation of blood glucose. In addition, glucagon has been shown to stimulate the release of insulin from isolated pancreatic tissue (Candela *et al.*, 1963; Turner and McIntyre, 1966). Furthermore, a rise in plasma glucagon levels was found after the oral administration of 100 g. or more of glucose (Samols *et al.*, 1965b; Lawrence, 1966), though Unger and Eisentraut (1964), Lawrence (1966), and Young and Jenkinson (1966) have observed a fall in plasma glucagon after the ingestion of 50 g. (or 1 g./kg.) of glucose.

McIntyre *et al.* (1965) found that the higher insulin response to intestinal glucose was associated with improved glucose tolerance; arterial blood glucose levels were lower during intestinal infusion than during intravenous infusion of the same quantity of glucose. This effect was also demonstrated in two patients after end-to-side portacaval anastomosis, and could not therefore be accounted for by the removal of ingested glucose from the portal circulation on its initial passage through the liver, as suggested by Scow and Cornfield (1954).

If the intestinal release of glucagon is responsible for the enhanced insulin secretion associated with oral glucose it might also be responsible for the superiority of oral over intravenous glucose tolerance. Such an effect would at first sight seem unlikely, as glucagon administration normally produces glycogenolysis and hyperglycaemia. However, Samols *et al.* (1965a) found that the high plasma insulin levels which resulted from the injection of 1 mg. of glucagon were associated with an increased rate of assimilation of a simultaneously administered rapid intravenous injection of glucose. It seemed possible that a similar effect of glucagon on glucose disposal might be seen with more prolonged infusions of glucose and glucagon, simulating the relatively slow absorption of glucose after oral administration. This possibility was tested in the following studies.

Methods

Eight healthy volunteers were given two separate intravenous infusions of the same quantity of 10% glucose after an overnight fast. On one occasion the glucose was given alone, and on the other glucagon was added to the glucose to give a rate of glucagon infusion of 0.6-1.2 µg. per minute. The actual quantities given to each subject are shown in the Tables. Glucose was given at a rate of 900 or 600 mg. per minute by means of a constant infusion pump.

Arterial blood was sampled through an indwelling Riley needle. Blood glucose was measured by an automated glucose oxidase method (Cramp, 1967) and plasma insulin by radioimmunoassay (Samols and Bilkus, 1964). Plasma samples were normally stored at -15° C. before insulin assay; those from the glucose-plus-glucagon infusion in Subject 6 were inadvertently left at room temperature and insulin assays were not performed on these samples.

Several subjects experienced nausea at the end of the glucagon infusion. A reaction characteristic of hypoglycaemia was also seen after cessation of the infusion; relief with glucose was rapid.

Results

Plasma insulin results are presented in Table I. In all seven studies in which plasma insulin was measured the levels were higher during the infusion of glucose plus glucagon than with glucose alone. The difference was evident at five minutes and continued to increase during the whole period of sampling. The greatest effect of glucagon on insulin secretion was seen in those subjects who received the largest dose of glucagon. This was particularly striking in Subject 2, in whom the insulin rose very rapidly with the glucagon load and reached a level of 204 µu./ml. at 20 minutes while the equivalent level with glucose alone was only 36 µu./ml.

The blood glucose values are presented in Table II. All subjects showed a more rapid rise in blood glucose during the first 30 minutes of the infusion of glucose plus glucagon than during the first 30 minutes of the infusion of glucose alone, while two of the subjects (Nos. 1 and 6) showed higher levels during the first 90 minutes. In the other six subjects the curves crossed and the blood glucose levels were lower at the end of the glucagon plus glucose infusion. In five of the six subjects the cross-over occurred between 30 and 70 minutes, while in the sixth it did not occur until after 105 minutes.

The magnitude of the difference in glucose levels during the first 30 minutes was, like the insulin response, related to the dose of glucagon administered. In the three subjects (Nos. 6-8) who were given 0.6 or 0.8 µg. of glucagon per minute the blood glucose did not rise more than 17 mg./100 ml. above the corresponding level which resulted when glucose alone was infused. In those subjects who received 2 µg. per minute or more the maximum difference for each individual during the first 30 minutes was not less than 41 mg./100 ml.

Discussion

In these studies glucagon did not produce an early improvement in glucose tolerance, despite the higher plasma insulin levels obtained when glucagon was infused with glucose. Glucose levels were actually higher during the first 30 to 45 minutes of the infusion of glucose plus glucagon than during

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TABLE I.—Plasma Insulin Response to Slow Intravenous Infusion of 10% Glucose With and Without Infusion of Glucagon

Subject	Age	Rate of Glucose Infusion (ml./min.)	Rate of Glucagon Infusion (μg./min.)	Plasma Insulin (μu./ml.)													
				Fasting		5'	10'	15'	20'	30'	45'	60'	75'	90'	105'	120'	
1	22	6.0	0 12.0	8	8	14	—	—	17	13	21	26	24	34	41	41	
				11	11	29	38	46	57	77	111	128	172	230	205	250	
2	21	9.0	0 10.0	13	15	33	35	34	36	46	48	62	62	71	83	90	
				11	8	60	130	169	204	260	299	307	373	424	478	461	
3	22	9.0	0 10.0	7	8	19	17	15	23	26	24	27	56	50			
				9	10	24	32	45	54	89	155	200	223				
4	22	9.0	0 2.0	10	18	29	25	29	23	38	27	61	69	75	80	88	
				14	15	45	50	63	62	56	76	100	155	174	200	190	
5	27	9.0	0 2.0	8	16	49	52	51	60	76	88	104	144	155			
				20	18	78	81	75	103	128	232	324	336	404			
6	26	6.0	0 0.8	—	—	—	—	—	—	—	—	—	—	—	—	—	
				—	—	—	—	—	—	—	—	—	—	—	—	—	
7	21	6.0	0 0.6	14	8	21	19	43	42	44	53	60	61	94			
				16	20	42	65	86	91	113	142	174	155				
8	21	6.0	0 0.6	16	16	24	29	28	31	34	35	38	56	62			
				14	14	26	35	43	45	53	86	90	150	189			

TABLE II.—Blood Glucose Response to Slow Intravenous Infusion of 10% Glucose With and Without Infusion of Glucagon

Subject	Rate of Glucose Infusion (ml./min.)	Rate of Glucagon Infusion (μg./min.)	Arterial Blood Glucose (mg./100 ml.)													
			Fasting			5'	10'	15'	20'	30'	45'	60'	75'	90'	105'	120'
1	6.0	0	76	80	82	120										
		12.0	100	94	90	137	160	184	146	165	175	225	225	240	275	270
2	9.0	0	50	78	75	126	130	160	170	190	195	200	240	230	198	180
		10.0	75	78	75	117	173	190	191	207	196	170	205	190	195	170
3	9.0	0	80	70	71	120	135	160	176	195	230	251	255	300		
		10.0	75	81	85	140	168	195	210	240	255	236	230			
4	9.0	0	66	66	66	99	107	123	146	163	184	191	212	176	173	162
		2.0	69	73	66	113	137	165	187	198	194	182	186	147	149	137
5	9.0	0	62	62	62	94	125	147	159	176	214	245	258	264	273	272
		2.0	66	66	68	110	147	195	220	258	275	277	286	280	264	
6	6.0	0	75	75	77	95	100	106	115	130	145	145	140	138	140	
		0.8	80	80	80	95	110	115	122	145	160	150	145	140	115	100
7	6.0	0	90	92	90	130	152	160	180	180		205	198	168	160	121
		0.6	90	95	92	130	155	172	187	192	170	160	160	140	121	86
8	6.0	0	90	90	94	116	125	130	140	155	165	165	182	180	140	100
		0.6	85	90	90	110	125	136	150	168	182	175	164	150	115	85

the infusion of glucose alone. It would seem that the hyperglycaemic-glycogenolytic effect of glucagon initially overcame the glucose-lowering effect which should have resulted from the increased insulin levels. In four of the subjects there was a change in the shape of the glucose curves during the infusion of glucose plus glucagon; the arterial blood glucose level fell and reached values below those found during the infusion of glucose alone. This suggests that when the short-lived glycogenolytic effect was over the effect of high insulin levels on peripheral glucose uptake could become manifest.

The glycogenolytic effect of glucagon seen in these studies suggests that it is unlikely that glucagon is the intestinal beta-cell stimulant suggested by McIntyre *et al.* (1965). A clear enhancement of insulin secretion can be obtained with 50 g. of oral glucose; elevation of plasma glucagon levels, however, has been consistently demonstrated only with the ingestion of larger doses. In addition, no glycogenolytic effect appears to occur with oral glucose; such an effect was presumably the cause of the higher blood glucose levels which we observed when glucagon was added to an infusion of glucose. Though we made no measurement of plasma glucagon the small amounts of glucagon infused should have produced plasma levels similar to those obtained with oral glucose (Samols *et al.*, 1965b).

Further evidence against the role of intestinal glucagon has been presented by Samols *et al.* (1966). They have claimed that glucagon of intestinal and pancreatic origin can be differentiated immunologically and that the rise in plasma glucagon which occurs after oral glucose is due to pancreatic glucagon. They suggest that stimulation of insulin secretion after oral glucose is enhanced by a high local concentration of glucagon following its release from the alpha cells of the islets of Langerhans. If this hypothesis proves to be correct it will leave unexplained the mechanism by which the signal for enhancement of glucagon, and thus insulin, secretion is trans-

mitted from the intestine after the administration of oral glucose.

Summary

When glucagon and glucose were infused intravenously the rise in plasma insulin was greater than that found when glucose was infused alone. Blood glucose levels were also higher during the first 45 minutes of glucose and glucagon infusion, indicating that under these conditions the blood glucose level was more affected by the glycogenolytic action of glucagon than by the greater insulin release due to glucagon. It is thus unlikely that glucagon-induced elevation of plasma insulin levels is responsible for the superiority of oral over intravenous glucose tolerance.

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