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Intracerebroventricular Insulin-Like Growth Factor-1 Decreases Feeding in Diabetic Rats

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Insulin-like growth factor-1 (IGF-1) is a hormone that is important in the regulation of growth processes and additionally has been demonstrated to modulate metabolic and autonomic responses. Some of its effects are mediated by the central nervous system (CNS), and there are IGF-1 receptors dispersed throughout the CNS. Both IGF-1 and insulin alter peripheral metabolic and autonomic nervous activity by a central mechanism, and the well-defined role of insulin in the regulation of feeding, especially in diabetes, led us to investigate the effect of chronic central administration of IGF-1 on metabolic and feeding parameters in normal and diabetic rats. Normal and diabetic rats with intracerebroventricular cannulas were given IGF-1, insulin (0.5 nmol/animal), or artificial cerebrospinal fluid via cannula twice daily for 4 d. Blood samples were collected on d 2 and 4, and the body weights and food intake were recorded daily. IGF-1 administered intracerebroventricularly did not alter plasma glucose, insulin, body weight, or food intake in normal rats. However, in diabetic animals, IGF-1 decreased food intake but did not alter blood glucose or plasma insulin. In correlated studies, intracerebroventricular insulin decreased food intake in both normal and diabetic animals. From these studies, we conclude that IGF-1 may act centrally to decrease food intake in the hyperphagic diabetic animals but not in normal animals. This suggests that diabetic animals have an increased sensitivity to CNS IGF-1.

Key Words: Insulin-like growth factor-1; insulin; food intake; intracerebroventricular administration.

Introduction

Insulin-like growth factor-1 (IGF-1) has been identified in nervous tissue and has been implicated in the growth of the nervous system *(1,2)*. In addition, IGF-1 receptors have been characterized in the central nervous system (CNS) and these receptors have discrete locations throughout the CNS *(3)*. IGF-1 and the IGF-1 receptor both have molecular structures similar to insulin and the insulin receptor, respectively, and insulin can bind to the IGF-1 receptor, and IGF-1 can bind to the insulin receptor, albeit with lower affinities *(4,5)*. Also, an insulin/IGF-1 hybrid receptor has been identified *(5)*. IGF-1 has been linked mostly to CNS growth regulation, and CNS insulin is associated with the regulation of energy balance *(1,6)*. Insulin infusion in the CNS has been demonstrated to have well-characterized effects on food intake and plasma insulin and glucose, and this CNS effect of insulin has been implicated in the modulation of autonomic nervous activity and cardiovascular function *(7–10)*. We have demonstrated that IGF-1 administered in the CNS mimics some of its effects on the cardiovascular system but not the sympathetic nervous responses *(11,12)*. It is also known that diabetes is associated with altered sensitivity to insulin and insulin actions, especially the feeding mechanisms *(13)*. In the present study, we investigated the effect of chronic CNS administration of IGF-1 on plasma glucose, plasma insulin, food intake, and body weight in normal and diabetic animals.

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Results

The basal food intake was determined, and it was observed that diabetic animals had a significantly higher intake when compared with normal animals. The administration of IGF-1 for 2 or 4 d in normal animals did not decrease the food intake and may have increased it (Table 1). However, intracerebroventricular administration of IGF-1 in diabetic animals resulted in a significant decrease in the food intake at 2 and 4 d (Table 1). Intracerebroventricular administration of an equimolar concentration of insulin resulted in a significant decrease in food intake in both normal and diabetic animals (Table 1). IGF-1 treatment tended to lower but not significantly decrease body weight in the diabetic animals but not in normal animals (Table 2). The body weights of insulin-treated rats were decreased after d 4 (Table 2) in normal rats. Insulin treatment also significantly enhanced weight loss in diabetic rats (Table 2). IGF-1 treatment did not alter the plasma glucose (Table 3) or plasma insulin (Table 4) in normal or diabetic rats. However, intracerebroventricular insulin decreased the blood glucose in normal and diabetic animals (Table 3) and also increased plasma

Table 1
Effect of Intracerebroventricular Administration of IGF-1, Insulin (0.5 nmol/twice daily per animal), or Artificial Cerebrospinal Fluid (ACF) on Daily Food Intake (g) for 2 or 4 d in Normal and Diabetic Rats

	IGF-1				Insulin			
	Normal		Diabetic		Normal		Diabetic	
	ACF	IGF-1	ACF	IGF-1	ACF	Insulin	ACF	Insulin
Before	24 ± 2.7 (10)	21 ± 2.2 (10)	48 ± 4.0 (10)	52 ± 6.1 (10)	25 ± 1.2 (10)	28 ± 2.0 (10)	51 ± 2.0 (10)	54 ± 6.1 (10)
Day 2	23 ± 2.6 (10)	28 ± 3.0 (10)	45 ± 3.5 (10)	18 ± 4.0[a,b] (10)	27 ± 2.0 (10)	20 ± 1.2[a,b] (10)	56 ± 5.2 (10)	21 ± 6.1[a,b] (9)
Day 4	24 ± 2.4 (10)	26 ± 1.8[a] (10)	39 ± 5.2 (10)	17 ± 5.2[a,b] (9)	28 ± 2.1 (10)	24 ± 1.8[a,b] (10)	49 ± 5.1 (9)	22 ± 3.1[a,b] (9)

[a] $p < 0.05$ vs. before.
[b] $p < 0.05$ vs. control.

Table 2
Effect of Intracerebroventricular Administration of IGF-1, Insulin (0.5 nmol/twice daily per animal), or ACF for 2 or 4 d on Body Weight (g) in Normal and Diabetic Rats

	IGF-1				Insulin			
	Normal		Diabetic		Normal		Diabetic	
	ACF	IGF-1	ACF	IGF-1	ACF	Insulin	ACF	Insulin
Before	284 ± 20 (10)	286 ± 15 (10)	248 ± 21 (10)	291 ± 16 (10)	310 ± 24 (10)	322 ± 26 (10)	275 ± 16 (10)	270 ± 14 (10)
Day 2	283 ± 18 (10)	272 ± 21 (10)	284 ± 20 (10)	274 ± 14 (10)	292 ± 28 (10)	296 ± 21 (10)	260 ± 12 (10)	240 ± 12[a] (9)
Day 4	282 ± 17 (10)	281 ± 10 (10)	274 ± 18 (10)	266 ± 14 (9)	301 ± 21 (10)	274 ± 16[a] (10)	271 ± 20 (9)	234 ± 10[a,b] (9)

[a] $p < 0.05$ vs. control.
[b] $p < 0.05$ vs. before.

insulin (Table 4) in normal and diabetic rats when compared with controls (Tables 3 and 4).

Discussion

We have observed that the central (intracerebroventricular) administration of IGF-1 decreased food intake in diabetic but not normal animals. IGF-1 did not affect the glycemic status or the plasma insulin levels in normal or diabetic animals. The failure of IGF-1 to influence food intake, blood glucose, or insulin levels in normal animals is consistent with previous observations *(14)*. The enhanced sensitivity of the hyperphagic diabetic animals suggests that they are more sensitive to the anorexiant effect of IGF-1 than normal animals. Our studies also confirm the demonstrated insulin suppression of food intake *(7–9)* in normal or diabetic animals. However, some responses are not always consistent *(15,16)*. IGF-1 and insulin were administered in equimolar concentrations and although IGF-1 did not alter food intake in normal rats, their suppression of food intake in diabetic animals was approximately the same (60 vs 59% for IGF-1 and insulin, respectively). The CNS insulin was also associated with an increase in plasma insulin and decreased blood glucose in normal animals. This was consistent with previous studies in which CNS insulin was demonstrated to increase peripheral insulin secretion *(8)*. IGF-1, unlike insulin, did not alter plasma insulin or glucose.

Although the mechanism of action of IGF-1 in the suppression of food intake is not readily apparent, our observation is consistent with the possibility that IGF-1 acts indirectly by the insulin receptor *(17)*. First, IGF-1 can bind to the insulin receptor, albeit with a lower affinity *(5)*. Insulin receptors are abundant in the arcuate nucleus in the hypothalamus *(3,18)*. The arcuate nucleus, a major site in the regulation of food intake has been shown to be sensitive to insulin, especially insulin regulation of neuropeptide Y (NPY) *(13,19)*. NPY is thought to be the major stimulator of feeding behavior, and it is significantly increased in the arcuate nucleus in diabetes and is suppressible by insulin *(19,20)*. Second, peripheral IGF-1 has also been associated

Table 3
Effect of Intracerebroventricular Administration of IGF-1, Insulin (0.5 nmol/twice daily per animal), or ACF for 2 or 4 d on Plasma Glucose (mg/dL) Levels in Normal and Diabetic Rats

	IGF-1				Insulin			
	Normal		Diabetic		Normal		Diabetic	
	ACF	IGF-1	ACF	IGF-1	ACF	Insulin	ACF	Insulin
Before	151 ± 4 (10)	151 ± 3 (10)	501 ± 22 (10)	520 ± 33 (10)	169 ± 7 (10)	147 ± 12 (10)	510 ± 35 (10)	480 ± 51 (10)
Day 2	147 ± 4 (10)	147 ± 5 (10)	635 ± 50 (10)	546 ± 29 (10)	161 ± 8 (10)	109 ± 10[a,b] (10)	490 ± 42 (9)	400 ± 40 (9)
Day 4	152 ± 2 (10)	149 ± 6 (10)	536 ± 34 (9)	488 ± 51 (9)	143 ± 4 (10)	104 ± 16[a,b] (10)	517 ± 45 (9)	275 ± 35[a,b] (9)

[a] $p < 0.05$ vs. control.
[b] $p < 0.05$ vs. before.

Table 4
Effect of Intracerebroventricular Administration of IGF-1, Insulin (0.5 nmol/twice daily per animal), or ACF for 2 or 4 d on Plasma Insulin (uU/mL) Levels in Normal and Diabetic Rats

	IGF-1				Insulin			
	Normal		Diabetic		Normal		Diabetic	
	ACF	IGF-1	ACF	IGF-1	ACF	Insulin	ACF	Insulin
Before	54 ± 16 (10)	62 ± 14 (10)	42 ± 12 (10)	53 ± 8 (10)	47 ± 13 (10)	38 ± 12 (10)	72 ± 3.0 (10)	75 ± 7.0 (10)
Day 2	64 ± 12 (8)	77 ± 15 (9)	47 ± 8 (10)	61 ± 12 (9)	38 ± 16 (10)	79 ± 12[a,b] (10)	59 ± 6.1 (8)	61 ± 8.2 (9)
Day 4	60 ± 8 (8)	87 ± 11 (10)	39 ± 6 (8)	61 ± 12 (9)	41 ± 16 (10)	80 ± 21[a,b] (10)	68 ± 7.1 (8)	51 ± 9.1[b,c] (9)

[a] $p < 0.01$ vs. control.
[b] $p < 0.05$ vs. before.
[c] $p < 0.05$ vs. control.

with enhanced insulin sensitivity *(21,22)*. Since IGF-1 receptors are not abundantly expressed in the arcuate nucleus, an action via the insulin receptor is suggested *(23)*. However, it is also possible that these observations can be attributed to IGF-1 acting directly on the IGF-1 receptor. This is supported by previous work from our laboratory demonstrating that intracerebroventricular IGF-1 lowers sympathetic nervous activity whereas intracerebroventricular insulin increases it *(24)*. The process of lowering sympathetic activity could account for the lack of a significant decrease in body weight. A lower energy expenditure would not result in a rapid weight loss. IGF-1 receptors have been demonstrated to be upregulated in food-deprived states and diabetes *(25)*. Our observed response is consistent with nutrient deficiency and enhanced sensitivity in diabetics. Another explanation could be that IGF-1 exerted its influence by binding to the IGF-2 receptor. IGF-2 has been demonstrated to decrease food intake in normal animals *(26,27)*. However, we did not confirm any effect of IGF-1 in normal animals.

An additional aim of the present study was to determine whether central IGF-1 had any influence on plasma glucose or insulin. Insulin has been demonstrated to act centrally to increase plasma insulin levels, and this insulin could affect plasma glucose *(8,9)*. We confirmed the observation that intracerebroventricular insulin can increase plasma insulin *(10)*. However, IGF-1 did not have any effect. It would be expected that if we administered IGF-1 and insulin in equimolar concentrations and they acted via the insulin receptors, they would have similar effects. However, it has been shown that central IGF-1 can alter a number of factors such as growth hormone secretion, leptin production, and other hormones that may indirectly minimize the insulin or sympathetic nervous response *(28–30)*.

We conclude from this study that central IGF-1 can influence food intake, and this effect is most evident in the hyperphagic state of diabetes. Also, this effect may be mediated by the higher sensitivity owing to increased numbers of insulin or IGF-1 receptors. The lack of effect of IGF-1 on plasma glucose, body weight, and insulin is probably associ-

ated with IGF-1's capacity to decrease sympathetic activity and autonomic responses.

Materials and Methods

All animal procedures were approved by the Wayne State University Animal Care and Use Committee. The studies were conducted in compliance with principles expressed in the National Institutes of Health, US PHS, *Guide for Care and Use of Laboratory Animals*. Male Wistar rats (Harlan) were maintained on a 12-h light/12-h dark cycle and given food and water ad libitum. Rats were made diabetic using streptozotocin (50 mg/kg intravenously via tail vein) and maintained for 4 wk as just described. A blood glucose >300 mg/dL confirmed diabetes. Intracerebroventricular cannulae were implanted in the lateral ventricle in both normal and diabetic rats following the induction of anesthetics using ketamine (8 mg/kg) and xylazine (5.0 mg/kg). The animals were placed in a Kopf stereotaxic apparatus, and a midsaggital incision was made in the scalp. A 22-gage stainless steel guide cannula (Plastic Products, Roanoke, VA) was placed in the lateral ventricle using bregma as a reference point as described previously *(31)*. Stainless steel anchoring screws were mounted in the skull, and an acrylic cap was built to hold the cannula in place. A removable "dummy" cannula was then attached to the guide cannula and the wound was closed. The animals were maintained in individual cages and allowed to recover for 4–7 d before the start of the experiments.

Rats were administered IGF-1 (0.5 nmol/animal) via the intracerebroventricular cannula twice daily, and food intake and body weight were determined daily. Blood samples were collected before the start of IGF-1 administration and on d 2 and 4 two hours following the last injection. The animals were sacrificed after d 4 and blood samples were collected for assay. For comparison purposes, normal and diabetic animals prepared as already described were treated with insulin (0.5 nmol/animal) and blood samples were collected, along with food intake and body weight determinations.

Plasma glucose was measured using the glucose diagnostic kit (Sigma, St. Louis, MO), and plasma insulin was assayed with a radioimmunoassay kit (ICN) using a rat insulin standard.

Following the experiment, all animals were sacrificed by decapitation, the brains were removed, and the placement of the cannula was verified by visual inspection. Data are reported as mean ± SEM of absolute numbers or percent change. When comparing values each animal served as its own control. Student's *t*-test was used to compare pairs of means. One way analysis of variance was used to evaluate several means.

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