Effect of Epinephrine and Somatostatin-induced Insulin Deficiency on Ketone Body Kinetics and Lipolysis in Man

M. WEISS, U. KELLER, AND W. STAUFFACHER

SUMMARY

The effect of elevated plasma epinephrine concentrations (≈800 pg/ml) on ketone body kinetics was determined in postabsorptive normal subjects using primed-continuous infusions of 3-^{14}C-acetoacetate. Infusion of epinephrine (60 ng/kg/min) resulted in a transient increase in total ketone body production to a maximum of 2.5-fold the basal rate within 45 min ($P < 0.01$ versus controls). Ketone body uptake increased with a delay, compared with production, causing a 2.8-fold increase in total ketone body concentrations ($P < 0.05$ versus controls). Plasma free fatty acid (FFA) and blood glycerol concentrations increased transiently during epinephrine; their course was similar to that of ketone body production. Epinephrine administration resulted in hyperglycemia, hyperlactatemia, and a modest increase in plasma insulin and glucagon concentrations. To assess epinephrine's effect on ketone body kinetics during lack of insulin, and to avoid epinephrine-induced alterations in plasma insulin and glucagon concentrations, epinephrine was also infused combined with somatostatin (6.5 μg/kg/h). During somatostatin infusion, epinephrine administration resulted in an enhanced and sustained elevation of total ketone body production from 4.4 ± 0.8 to 15.1 ± 1.2 μmol/kg/min ($P < 0.01$ versus somatostatin alone). Ketone body concentrations increased markedly from 310 ± 63 to 1763 ± 137 μmol/L ($P < 0.01$ versus somatostatin alone); the ketonemic effect was enhanced due to a 40% decrease of the metabolic clearance rate associated with somatostatin infusion. The increase in plasma FFA and blood glycerol concentrations during somatostatin-induced insulin deficiency was transiently enhanced by epinephrine, such that they increased to 3.2- and 5.6-fold their basal values after 45 min, respectively ($P < 0.01$). Thus, elevation of epinephrine concentrations to levels that are observed in severe stress resulted, in normal man, in a transient increase in lipolysis and ketogenesis. The metabolic clearance of ketone bodies increased initially but remained uninfluenced thereafter. The transient nature of the increase in ketogenesis and lipolysis was in part due to an elevation of plasma insulin levels caused by hyperglycemia. During acute insulin deficiency, epinephrine's effect on ketogenesis was sustained, while the acceleration of lipolysis was again transient, suggesting a direct hepatic ketogenic effect. However, the major influence of epinephrine on ketone body kinetics consisted of an enhancement of ketone body production secondary to an increase in lipolysis and FFA supply for ketogenesis. **DIABETES *33*:738–744, August 1984.**

From the Departments of Internal Medicine and Research, Kantonsspital, CH-4031 Basel, Switzerland.
Address reprint requests to Dr. U. Keller at the above address.
Received for publication 1 November 1983.

In 1956, Ellis was the first to report increased ketone body concentrations during epinephrine administration in vivo and in vitro.[1] The ketonemic response to epinephrine in juvenile-onset diabetic subjects was augmented, whereas the increase in plasma free fatty acids (FFA) during epinephrine was similar in diabetic subjects compared with healthy control subjects.[2] Clutter[3] and Galster[4] reported augmented lipolysis and increased blood β-hydroxybutyrate concentrations in normal subjects during elevated epinephrine concentrations, similar to those observed during stress.[5] Diabetic ketoacidosis has been observed to be associated with markedly elevated plasma epinephrine concentrations.[6]

From these data, it appeared of interest to determine whether epinephrine influences ketone body concentrations by affecting production or peripheral utilization of ketone bodies. The importance of this distinction is emphasized by the fact that diabetic ketosis has been reported to be associated with alterations in production and peripheral clearance of ketone bodies.[7,8] In addition, the separate assessment of the effect of epinephrine on lipolysis, and thus FFA supply for ketogenesis, and on ketone body production was of interest, since it has been suggested that epinephrine exerts direct ketogenic effects on the liver.[8,9]

The present studies were, therefore, performed to deter-

Downloaded from http://diabetesjournals.org/diabetes/article-pdf/33/8/738/353322/33-8-738.pdf by guest on 01 December 2023

mine the effect of pathophysiologic concentrations of epinephrine on production and peripheral uptake of total ketone bodies in normal subjects using 3-^{14}C-acetoacetate infusions. Epinephrine's effect on lipolysis was assessed by determining plasma FFA and blood glycerol concentrations. Since the study of adrenergic influences on ketogenesis was of particular clinical relevance to insulin-deficient diabetes, the effect of epinephrine on ketone body kinetics was also determined during somatostatin-induced acute insulin lack. In addition, these studies allowed an assessment of epinephrine's effects on ketone body kinetics independent of changes in plasma insulin and glucagon concentrations, which have been reported to occur during epinephrine infusion.[10,11]

MATERIALS AND METHODS

Subjects. Twenty-eight healthy volunteers (12 men, 16 women) aged 59 ± 6 yr and weighing within 20% of their ideal body weight gave their written, informed consent to participate. An oral glucose tolerance test and an SMAC-12 profile (Technicon, Tarrytown, New York) served to exclude diabetes, kidney, or liver diseases. The study protocol was approved by the Human Ethics Committee of the Department of Medicine, University Hospital, Basel, Switzerland.

Procedures. The subjects reported to the laboratory at 7:30 a.m. after a 12-h overnight fast. A Teflon cannula was inserted into the left cubital vein for infusion, and a butterfly needle (1.1-mm inner diameter) into a dorsal hand vein for blood sampling. The latter was kept patent by a slow drip of saline (0.145 mol/L). The hand was kept in a warming box at 60°C[12] to arterialize hand venous blood. The studies were started by administering a bolus of 20 μCi of 3-^{14}C-acetoacetate. Thereafter, the volume of distribution of total ketone bodies was determined in blood samples drawn at 2-min intervals during 14 min. The volume of distribution of total ketone bodies was 0.184 ± 0.046 L/kg. After injecting a second bolus of 7.5 μCi, a continuous infusion of 150 μCi 3-^{14}C-acetoacetate was started; after 60 min of tracer equilibration, blood samples were drawn in 10- to 15-min intervals during a 30-min basal period as well as during a 170-min infusion period. The following four infusion protocols were performed in randomized order: (1) epinephrine (60 mg/kg/min), N = 6; (2) saline (0.145 mol/L, controls), N = 9; (3) epinephrine (60 ng/kg/min) and somatostatin (6.5 μg/kg/h), N = 6; and (4) somatostatin alone (6.5 μg/kg/h), N = 7. At 120 min, 5000 U of heparin (Liquemin, Roche, Basel, Switzerland) were injected to raise plasma FFA concentrations to examine epinephrine's effects on ketogenesis during substrate elevation. The subjects were connected to an EKG monitor, and arterial blood pressure was measured at each blood sampling time point using an Arteriosonde (Roche).

Infusions. Na-3-^{14}C-acetoacetate was prepared by hydrolysis of 3-^{14}C-ethyl-acetoacetate (Radiochemical Center, Amersham, United Kingdom) using NaOH.[13] After neutralization and sterilization by passing through a 0.2-μm Millipore filter, the tracer was kept at −70°C until use. Its radiochemical purity, determined according to Mayes and Felts,[14] was 77 ± 7%. The epinephrine infusions (Streuli, Uznach, Switzerland) were prepared immediately before use by diluting epinephrine in 0.145 mol/L NaCl, and by adding human albumin and ascorbic acid (Redoxon, Roche) to final concentrations of 0.5% and 20 μg/ml, respectively. Cyclic somatostatin (donated by Serono GmbH, FRG) was dissolved in 0.145 mol/L NaCl. All infusions were delivered using Harvard syringe pumps (South Natick, Massachusetts).

Analyses. Blood samples were drawn using EDTA as an anticoagulant, and immediately deproteinized by adding chilled perchloric acid 30% (1:1). Samples were neutralized within 30 min for later determination of acetoacetate,[15] β-hydroxybutyrate,[16] lactate,[17] pyruvate,[17] and glycerol[18] using microfluorometric adaptations of enzymatic methods. Blood acetone concentration was determined using head-space analysis and gas chromatography.[19] All ketone bodies and pyruvate concentrations were assayed within 4 h. Plasma FFA levels were determined using a radiochemical method.[20] Plasma was also collected with heparin as an anticoagulant for measurement of plasma catecholamines by radioenzymatic assay[21] and of plasma insulin by radioimmunoassay.[22]

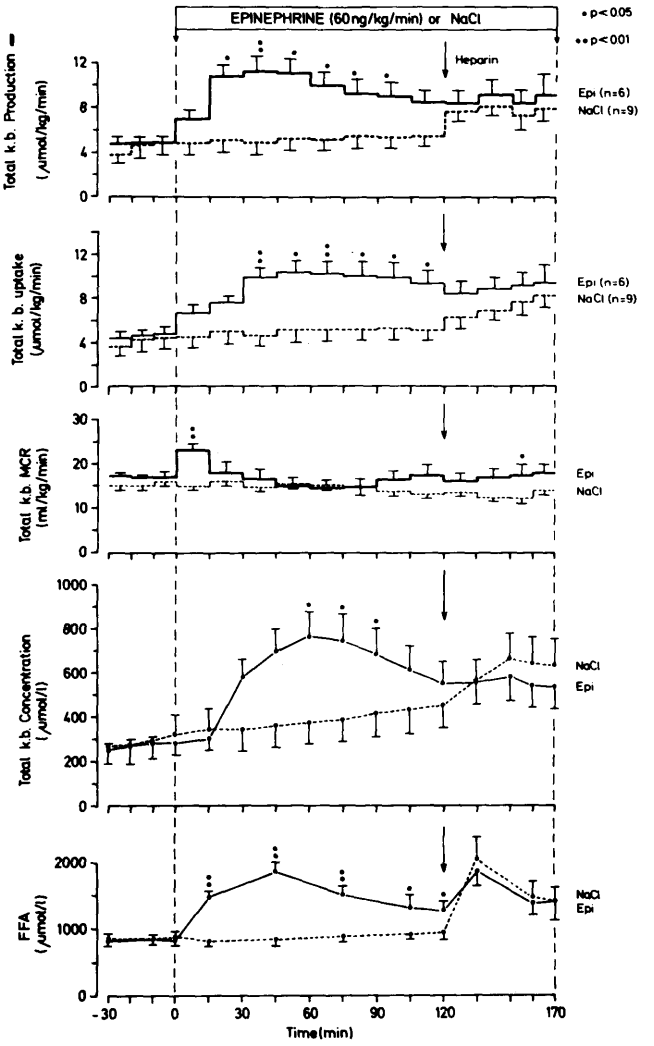

FIGURE 1. Total ketone body production, uptake, metabolic clearance rate (MCR), and concentration, and plasma FFA levels during epinephrine (solid lines) and NaCl infusion (broken lines). At 120 min, 5000 U of heparin was injected. The asterisks denote statistical differences between the two protocols.

Downloaded from http://diabetesjournals.org/diabetes/article-pdf/33/8/738/353322/33-8-738.pdf by guest on 01 December 2023

TABLE 1
Blood concentrations of acetoacetate, β-hydroxybutyrate, and acetone

	NaCl (N = 9)	Epi (N = 6)	SRIF (N = 7)	Epi + SRIF (N = 6)	Time (min)
Acetoacetate (μmol/L)	114 ± 29	148 ± 13	99 ± 21	154 ± 26	Basal§
	130 ± 30	233 ± 21*	146 ± 23	253 ± 30†	60
	157 ± 31	191 ± 29	186 ± 34	364 ± 27‡	120
	200 ± 32	187 ± 34	236 ± 35	454 ± 34‡	170
β-Hydroxybutyrate (μmol/L)	160 ± 50	109 ± 24	182 ± 48	139 ± 43	Basal§
	228 ± 61	512 ± 93*	411 ± 108	901 ± 77‡	60
	275 ± 71	335 ± 76	624 ± 125	1355 ± 111‡	120
	401 ± 82	324 ± 75	894 ± 137	1652 ± 101‡	170
Acetone (μmol/L)	15 ± 6	13 ± 4	16 ± 4	16 ± 4	Basal§
	17 ± 6	19 ± 5	21 ± 5	26 ± 5	60
	23 ± 8	22 ± 6	32 ± 7	44 ± 6	120
	29 ± 10	26 ± 7	45 ± 9	64 ± 8	170

Abbreviations used: Epi, epinephrine; SRIF, somatostatin.
*$P < 0.05$ vs. NaCl; †$P < 0.05$, ‡$P < 0.01$ vs. SRIF; and §average of 3 basal values.

Trasylol (500 IU/ml plasma) was added to the samples for glucagon determination, and immunoreactive glucagon was measured[23] using antibody 4305 donated by Dr. J. Holst.[24] ^{14}C-acetoacetate and ^{14}C-β-hydroxybutyrate were determined as previously described.[14] Recovery of added ^{14}C-acetoacetate and ^{14}C-β-hydroxybutyrate to nonradioactive blood was 90 ± 9% and 85 ± 7%, respectively. Corrections were made accordingly. The volume of distribution and rates of production, uptake, and clearance of total ketone bodies during non-steady-state conditions were calculated using a single-compartment model and the combined specific activity of individual ketone bodies.[13] The change of ketone body specific activity between two consecutive time points was used for the calculation of total ketone body production according to the formula of Steele.[25]

Statistical analyses were performed using the Mann-Whitney U test for comparison between protocols, and the Wilcoxon rank-sum test for the assessment of statistical differences between the 45- and 120-min values during the infusion period compared with the mean values of the basal period. When multiple comparisons were made with the same data, the P value obtained by either test was multiplied by the number of comparisons made (Bonferroni adjustment[26]). All results are given as mean ± SEM.

RESULTS

Effect of epinephrine on ketone body kinetics and plasma FFA concentrations in normal subjects (Figure 1, Table 1). Infusion of epinephrine resulted in a transient increase in total ketone body concentration (Figure 1); the peak level was 764 ± 111 μmol/L at 60 min ($P < 0.05$ versus controls). Total ketone body production increased within 45 min from 4.8 ± 0.6 to 11.3 ± 1.3 μmol/kg/min (2.4-fold increase, $P < 0.01$ versus controls). Total ketone body uptake also increased, but with a delay to a 2.2-fold elevation after 75 min ($P < 0.01$ versus controls). The elevation of ketone body production, uptake, and concentration was transient.

Total ketone body clearance increased rapidly during epinephrine to 23.4 ± 1.4 ml/kg/min (by 36%) at 15 min ($P < 0.02$ versus controls), but remained only slightly higher than during saline infusion at later time points. Plasma FFA concentrations increased transiently during epinephrine to 2.3-fold the basal concentrations after 45 min ($P < 0.01$ versus controls). The course of plasma FFA levels was parallel

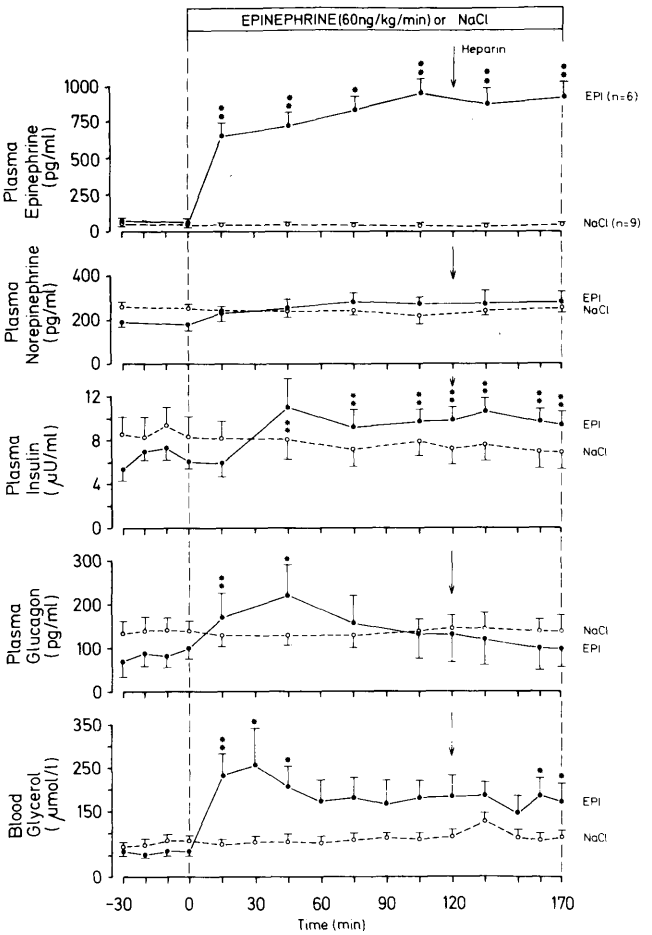

FIGURE 2. Concentrations of plasma epinephrine, norepinephrine, insulin, glucagon, and blood glycerol during epinephrine (solid line) and NaCl (broken line) in normal subjects. The asterisks indicate statistical differences between the two protocols. In the panels of plasma insulin and glucagon, asterisks refer to the comparison of their changes from basal values occurring during epinephrine versus saline infusion. *$P < 0.05$, **$P < 0.01$.

Downloaded from http://diabetesjournals.org/diabetes/article-pdf/33/8/738/353322/33-8-738.pdf by guest on 01 December 2023

TABLE 2
Plasma glucose and blood lactate and pyruvate concentrations

	NaCl (N = 9)	Epi (N = 6)	SRIF (N = 7)	Epi + SRIF (N = 6)	Time (min)
Plasma glucose (mmol/L)	5.0 ± 0.1	5.4 ± 0.2	5.3 ± 0.2	5.4 ± 0.3	Basal‖
	4.9 ± 0.1	9.6 ± 0.8†	3.9 ± 0.2	12.3 ± 0.5§	120
	4.9 ± 0.2	9.5 ± 0.8†	4.2 ± 0.3	14.1 ±, 0.7§	170
Blood lactate (mmol/L)	0.55 ± 0.06	0.58 ± 0.1	0.51 ± 0.07	0.53 ± 0.09	Basal‖
	0.43 ± 0.04	1.72 ± 0.36†	0.62 ± 0.09	1.26 ± 0.09‡	120
	0.47 ± 0.03	1.97 ± 0.41†	0.56 ± 0.09	1.55 ± 0.14‡	170
Blood pyruvate (μmol/L)	94 ± 11	102 ± 11	ND	88 ± 13	Basal‖
	74 ± 7	157 ± 19*	ND	114 ± 12	120
	73 ± 8	155 ± 19*	ND	119 ± 16	170

Abbreviations are as shown in Table 1; ND, not determined.
*$P < 0.05$, †$P < 0.01$ vs. NaCl; ‡$P < 0.05$, §$P < 0.01$ vs. SRIF; and ‖average of 3 basal values.

to that of total ketone body production. Heparin injection resulted in a further transient elevation of plasma FFA concentrations in both protocols. As a result, ketone body production increased during saline infusion but not significantly during epinephrine administration. Table 1 demonstrates that, during epinephrine infusion, there was a significant but transient increase in the concentration of acetoacetate and of β-hydroxybutyrate. Blood acetone concentrations were low compared with the other ketone bodies and increased gradually in both protocols.

Effect of epinephrine on plasma hormone and substrate levels (Figure 2, Table 2). Figure 2 demonstrates that plasma epinephrine concentrations increased to ≈ 850 pg/ml, representing a 20-fold increase compared with basal concentrations. Plasma norepinephrine levels did not change significantly during epinephrine infusion. Plasma insulin concentrations increased during epinephrine to 1.7-fold the basal concentrations after 45 min. The increase above baseline was significantly different compared with controls (4.6 ± 2.3 versus −0.6 ± 0.3 μU/ml, $P < 0.01$). Plasma glucagon concentrations increased significantly during epinephrine to 2.9-fold the basal concentrations; the increase from the basal period was 131 ± 49 versus −9.8 ± 11 pg/ml in controls ($P < 0.02$). In contrast to plasma insulin, the increase in glucagon concentrations was transient. Blood glycerol levels increased rapidly to fivefold basal concentrations within 30 min, and declined thereafter in spite of ongoing epinephrine infusion. The pattern of glycerol concentrations was similar to that of plasma FFA.

Table 2 shows that plasma glucose concentrations increased markedly and gradually to 10 ± 1 mmol/L at the end of epinephrine infusion ($P < 0.01$ versus controls). Blood lactate levels increased more than threefold during epinephrine ($P < 0.01$), whereas blood pyruvate concentration increased 1.5-fold above basal values ($P < 0.02$ compared with controls).

Effect of epinephrine during somatostatin-induced insulin deficiency on ketone body kinetics and plasma FFA concentrations (Figure 3, Table 1). Total ketone body production increased during epinephrine and somatostatin infusion within 120 min from 4.4 ± 0.8 to 15.1 ± 1.2 μmol/kg/min ($P < 0.01$ versus somatostatin alone, Figure 3); the increase was significantly greater than during infusion of somatostatin alone from 30 min onward. Total ketone body uptake increased similarly as production, but with a delay, resulting in a significant increase in total ketone body concentration to 1763 ± 137 μmol/L within 120 min compared with 842 ± 158 μmol/L during somatostatin alone ($P < 0.01$). Infusion of somatostatin alone increased production and concentration of total ketone bodies significantly ($P < 0.01$ versus somatostatin). The metabolic clearance rate of ketone bodies decreased significantly during both protocols to approximately 60% of basal rates at the end of the infusions. Again, epinephrine resulted in an early and transient increase in the ketone body metabolic clearance rate, within 15 min, to 18.1 ± 1.8 ml/kg/min, and by 4.8 ± 0.7 versus −0.9 ± 0.6 ml/kg/min during somatostatin alone ($P < 0.01$). Later on, the metabolic clearance was slightly, but not consistently, lower during epinephrine than during infusion of somatostatin alone.

Plasma FFA concentrations increased to 2832 ± 372 μmol/L at 45 min; they were significantly higher during epinephrine than during somatostatin alone (1455 ± 156 μmol/L, $P < 0.01$). The epinephrine-induced increase in plasma FFA concentrations was again transient when compared with the protocol of somatostatin infusion alone, since, after 120 min, plasma FFA were not significantly elevated by epinephrine. Heparin injection resulted in a further modest increase in plasma FFA concentration, but had no distinct effect on ketogenesis in both studies. The mean concentrations of all three individual ketone bodies (Table 1) increased gradually during infusion of somatostatin alone; the increase in blood acetoacetate and β-hydroxybutyrate was significantly augmented during addition of epinephrine infusion.

Effect of epinephrine during somatostatin-induced insulin deficiency on plasma hormone and substrate concentrations (Figure 4, Table 2). Plasma epinephrine concentrations reached values of ~750 pg/ml during epinephrine infusion, whereas plasma norepinephrine did not change significantly during both protocols (Figure 4). Plasma insulin and glucagon levels were slightly, but not significantly, higher during the basal period of the somatostatin protocol. Plasma insulin concentrations decreased rapidly and significantly in both protocols ($P < 0.05$ after 45 min) to mean values of 3 μU/ml, which were just above the detection limit of our insulin assay (2 μU/ml). Plasma glucagon

Downloaded from http://diabetesjournals.org/diabetes/article-pdf/33/8/738/353322/33-8-738.pdf by guest on 01 December 2023

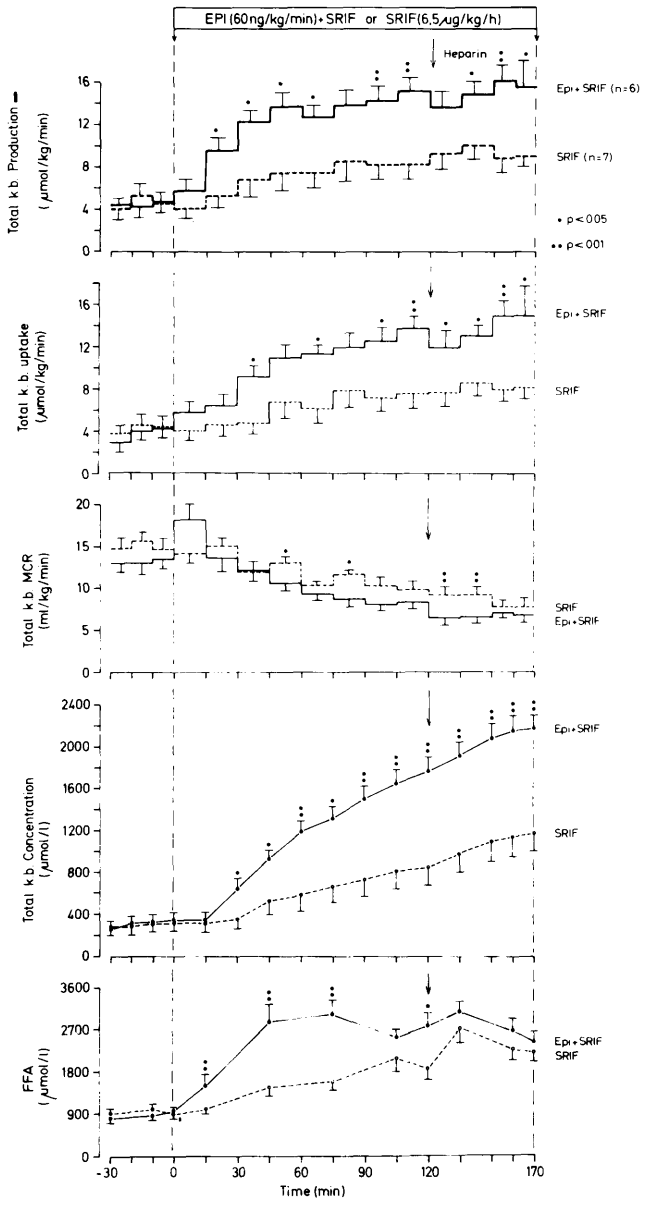

FIGURE 3. Total ketone body production, uptake, metabolic clearance rate (MCR), and concentration, and plasma FFA levels during epinephrine and somatostatin infusion (solid lines) and during somatostatin alone (broken lines). At 120 min, 5000 U of heparin was injected. The asterisks denote statistical differences between the two protocols.

declined significantly in both studies ($P < 0.05$ after 45 min); the concentrations during epinephrine administration were not significantly different compared with infusion of somatostatin alone. Blood glycerol concentrations increased to 5.6-fold the basal concentrations after 45 min, and slightly declined thereafter.

Plasma glucose concentrations (Table 2) increased progressively to 14.1 ± 0.7 mmol/L at the end of epinephrine infusion ($P < 0.01$ versus somatostatin alone). The increase in blood lactate concentration during epinephrine was statistically significant from 15 min onward compared with somatostatin alone; blood pyruvate increased significantly during epinephrine, but was less than for lactate.

Hemodynamic effects. Epinephrine administration resulted in a significant increase in heart rate from 65 ± 3 to 88 ± 4 beats/min ($P < 0.01$ versus somatostatin alone). Systolic blood pressure showed a tendency to decline (NS).

DISCUSSION

Elevation of plasma epinephrine to concentrations as are observed in severe stress[5,6] resulted, in the present study, in a transient increase in plasma FFA levels and in a parallel elevation of total ketone body production and concentration in normal subjects. During somatostatin-induced insulin deficiency, the lipolytic and ketogenic effect of epinephrine was enhanced.

Earlier studies reported increased β-hydroxybutyrate concentrations during similar elevation of plasma epinephrine levels, as was seen in the present study.[3,4,11] The present results demonstrate that epinephrine-induced hyperketonemia is primarily due to an increase in ketone body production. There was no sustained effect of epinephrine on the metabolic clearance rate of ketone bodies, in agreement with previous studies of ketone body uptake in vitro,[27] but in contrast to an earlier report suggesting diminished ketone body uptake by the human forearm during catecholamine infu-

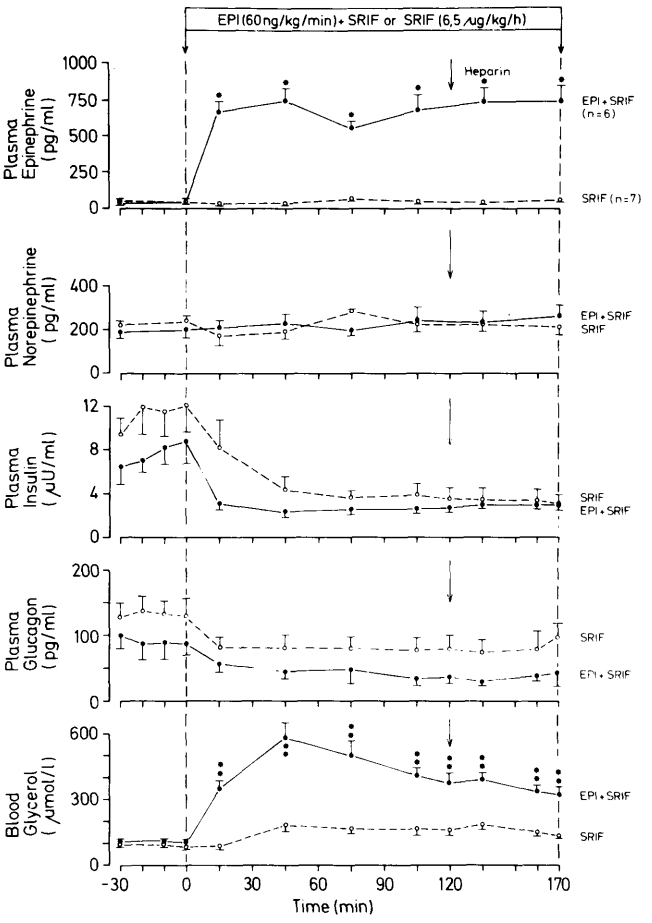

FIGURE 4. Concentrations of plasma epinephrine, norepinephrine, insulin, glucagon, and blood glycerol during epinephrine and somatostatin (solid line) and during somatostatin alone (broken line) in normal subjects. The asterisks indicate statistical differences between the two protocols. *P < 0.05, **P < 0.01.

Downloaded from http://diabetesjournals.org/diabetes/article-pdf/33/8/738/353322/33-8-738.pdf by guest on 01 December 2023

sion.[28] The present data are in agreement with a previous preliminary report.[29]

The observed increase in lipolysis and ketogenesis during epinephrine infusion in normal subjects was transient, and their return to baseline coincided with an elevation of plasma insulin concentrations. The latter most likely resulted from progressive hyperglycemia.[30] The increase in plasma insulin levels during epinephrine administration has been suggested to be the reason for the return of enhanced lipolysis to basal values.[29] However, the present results demonstrate that the epinephrine-induced enhancement of lipolysis diminishes with time, even during somatostatin-induced insulin lack. The transient nature of the epinephrine effect may be explained by the process of desensitization of lipolysis, as has been previously demonstrated of other catecholamine effects.[31–35]

In contrast to the effect of epinephrine, insulin deficiency during somatostatin infusion resulted in a gradual and sustained increase in lipolysis. It has been previously demonstrated that the increase in plasma FFA concentrations during somatostatin infusion is due to insulin deficiency;[36] it appears, therefore, that the sustained enhancement of lipolysis during insulin deficiency results from a different mechanism than during its stimulation by catecholamines.

Hepatic FFA uptake has been reported to parallel plasma FFA concentrations,[37] and hepatic fatty acid uptake was proportional to arterial plasma FFA concentrations during epinephrine infusions in dogs.[38] Epinephrine infusion resulted in an inconsistent increase in hepatic blood flow by up to 30% in a previous study in man.[39] It is, therefore, conceivable that hepatic FFA uptake increased during epinephrine in the present study due to enhanced blood flow, supporting the conclusion that epinephrine influences ketogenesis mainly by providing more fatty acid substrate for ketogenesis. However, during somatostatin-induced insulin deficiency, there was a sustained increase in ketogenesis, whereas the elevation of plasma FFA concentrations was transient; during the last 30 min of epinephrine infusion, ketone body production was still enhanced, whereas plasma FFA concentrations had reached identical levels as in the control studies. This suggests that, during somatostatin infusion, epinephrine enhanced hepatic ketogenesis directly and independently of FFA supply, as suggested previously in vitro.[8,9] A similar conclusion was reached in a previous study using norepinephrine infusions in man.[40]

The observed increase in plasma glucagon levels during epinephrine infusion was in agreement with previous studies.[10,11] The finding that epinephrine enhanced ketogenesis during somatostatin-induced glucagon deficiency does not support the hypothesis that glucagon represents an important mediator of epinephrine's effect on ketogenesis.[11]

The epinephrine-induced increase in plasma glucose concentrations observed in the present studies was similar to that seen in a previous report demonstrating that hyperglycemia resulted from increased production, and from decreased peripheral clearance, of glucose.[10] The hyperlactatemia observed during epinephrine may be due to β-receptor-mediated stimulation of glycogenolysis in muscle.[41]

The early and transient increase in the metabolic clearance rate of ketone bodies during epinephrine administration prevented an increase in ketone body concentrations during the first 15 min of epinephrine infusion, despite accelerated ketone body turnover. This increase in ketone body clearance may be related to hemodynamic effects of epinephrine, such as acceleration of heart rate and increased cardiac output.[42] At later time points, the increase in ketone body clearance may have been counteracted by developing hyperketonemia, which may have decreased the metabolic clearance of ketone bodies.[13]

Somatostatin was used to produce a model of acute insulin deficiency. Although direct effects of somatostatin on liver cell and adipocyte metabolism have not been demonstrated,[43] it cannot be excluded that glucagon or growth hormone suppression influenced the present results. However, previous studies failed to demonstrate ketogenic effects of glucagon replacement infusions during somatostatin-induced insulin deficiency in normal man.[36]

In conclusion, the present results demonstrate that epinephrine-induced hyperketonemia results mainly from increased ketogenesis, due to increased FFA supply to the liver, after stimulation of lipolysis. The increase in lipolysis and ketogenesis was transient in normal subjects; the return to basal rates was due in part to an increase in plasma insulin level, which was probably induced by hyperglycemia. Epinephrine failed to influence ketone body clearance except for an early transitory increase. During somatostatin-induced insulin deficiency, lipolysis and ketogenesis were accelerated by epinephrine. The stimulatory effect of epinephrine on lipolysis diminished with time, whereas the ketogenic effect persisted, suggesting a direct stimulatory effect of epinephrine on hepatic ketogenesis. Due to a concomitant decrease of the metabolic clearance rate, hyperepinephrinemia resulted in a marked and progressive increase in ketone body concentrations. These findings emphasize the potential role of epinephrine in the development of diabetic ketoacidosis.

ACKNOWLEDGMENTS

We thank S. Brunner and H. Niederer for expert technical assistance. The secretarial help of N. Deola is gratefully acknowledged. This study was supported by grant no. 3.988.80 of the Swiss National Science Foundation.

REFERENCES

[1] Ellis, S.: The metabolic effects of epinephrine and related amines. Pharmacol. Rev. 1956; 10:483–562.

[2] Baker, L., Kaye, R., and Haque, N.: Metabolic homeostasis in juvenile diabetes mellitus. II. Increased ketone responsiveness to epinephrine. Diabetes 1969; 18:421–27.

[3] Clutter, W. E., Bier, D. M., Shah, S. D., and Cryer, P. E.: Epinephrine plasma metabolic clearance rates and physiologic thresholds for metabolic and hemodynamic action in man. J. Clin. Invest. 1980; 66:94–101.

[4] Galster, A. D., Clutter, W. E., Cryer, P. E., Collins, J. A., and Bier, D. M.: Epinephrine plasma thresholds for lipolytic effects in man. Measurements of fatty acid transport with [1-^{13}C] palmitic acid. J. Clin. Invest. 1981; 67:1729–38.

[5] Dimsdale, J. E., and Moss, J.: Plasma catecholamines in stress and exercise. JAMA 1980; 243:340–42.

[6] Christensen, N. J.: Plasma norepinephrine and epinephrine in untreated diabetics, during fasting and after insulin administration. Diabetes 1974; 23:1–8.

[7] Miles, J. M., Rizza, R. A., Haymond, M. W., and Gerich, J. E.: Effects of acute insulin deficiency on glucose and ketone body turnover in man. Diabetes 1980; 29:926–30.

[8] Cole, R. A., and Margolis, S.: Stimulation of ketogenesis by dibutyryl cyclic AMP in isolated rat hepatocytes. Endocrinology 1974; 94:1391–96.

[9] Heimberg, M., Fizette, N. B., and Klausner, H.: The action of adrenal hormones on hepatic transport of triglycerides and fatty acids. J. Am. Oil Chem. Soc. 1964; 41:774–79.

Downloaded from http://diabetesjournals.org/diabetes/article-pdf/33/8/738/353322/33-8-738.pdf by guest on 01 December 2023

[10] Rizza, R. A., Haymond, M. W., Cryer, P. E., and Gerich, J. E.: Differential effects of epinephrine on glucose production and disposal in man. Am. J. Physiol. 1979; 237:E356–62.

[11] Johnston, D. G., Pernet, A., McCulloch, A., Blesa-Malpica, G., Burrin, J. M., and Alberti, K. G. M. M.: Some hormonal influences on glucose and ketone body metabolism in normal human subjects. *In* Metabolic Acidosis (Ciba Foundation Symp. 87) 1982:169–91.

[12] Sonnenberg, G. E., and Keller, U.: Sampling of arterialized heated hand venous blood as a non-invasive technique for study of ketone body kinetics in man. Metabolism 1982; 31:1–5.

[13] Keller, U., Sonnenberg, G. E., and Stauffacher, W.: Validation of a tracer technique to determine nonsteady-state ketone body turnover rates in man. Am. J. Physiol. 1981; 240:E253–62.

[14] Mayes, P. A., and Felts, J. M.: Determination of ^{14}C-labelled ketone bodies by liquid scintillation counting. Biochem. J. 1967; 102:230–35.

[15] Williamson, D. H., Mellanby, J., and Krebs, H. A.: Enzymatic determination of D-β-hydroxybutyric acid and acetoacetate acid in blood. Biochem. J. 1962; 82:90–96.

[16] Young, D. A. G., and Renold, A. E.: A fluorimetric procedure for the determination of ketone bodies in very small quantities of blood. Clin. Chim. Acta 1966; 13:791–93.

[17] Bergmeyer, H. U., Ed.: Methods of Enzymatic Analysis, Vols. 3 and 4, 2d edit. New York, Academic Press, 1974.

[18] Chernick, S. S.: Determination of glycerol in acyl glycerols. Methods Enzymol. 1969; 14:627–30.

[19] Van Stekelenberg, G. J., and De Bruyn, J. W.: A simple gas chromatographic determination of acetone and β-ketobutyric acid in blood serum by means of head space gas sampling. Clin. Chim. Acta 1970; 28:233–37.

[20] Ho, R. J.: Radiochemical assay of long-chain fatty acids using ^{63}Ni as tracer. Anal. Biochem. 1970; 36:105–13.

[21] Da Prada, M., and Zürcher, G.: Simultaneous radioenzymatic determination of plasma and tissue adrenaline, noradrenaline and dopamine within the femtomole range. Life Sci. 1976; 19:1161–74.

[22] Heding, L. G.: Determination of total serum insulin in insulin-treated diabetic patients. Diabetologia 1972; 8:260–66.

[23] Aguilar-Parada, E., Eisentraut, A. M., and Unger, R. H.: Pancreatic glucagon secretion in normal and diabetic subjects. Am. J. Med. Sci. 1969; 257:415–19.

[24] Holst, J. J., and Aasted, B.: Production and evaluation of glucagon antibodies for radioimmunoassay. Acta Endocrinol. 1974; 77:715–26.

[25] Steele, R.: Influences of glucose loading and of injected insulin on hepatic glucose output. Ann. NY Acad. Sci. 1959; 82:420–30.

[26] Glantz, S. A.: Primer of Biostatistics. New York, McGraw-Hill, 1981.

[27] Robinson, A. M., and Williamson, D. H.: Physiological roles of ketone bodies as substrates and signals in mammalian tissues. Physiol. Rev. 1980; 60:143–87.

[28] Wicklmayr, M., and Dietze, G.: Effect of metaproterenol on ketone body metabolism of the forearm in healthy and diabetic subjects. Horm. Metab. Res. 1979; 11:1–6.

[29] Miles, J. M., Haymond, M. W., and Gerich, J. E.: Effects of free fatty acids, insulin, glucagon and adrenaline on ketone body production in humans. Metabolic Acidosis (Ciba Foundation Symp. 87) 1982:192–213.

[30] Beard, J. C., Weinberg, C., Pfeifer, M. A., Best, J. D., Halter, J. B., and Porte, D.: Interaction of glucose and epinephrine in the regulation of insulin secretion. Diabetes 1982; 31:802–807.

[31] Mukherjee, C., Caron, M. G., and Lefkowitz, R. J.: Catecholamine-induced subsensitivity of adenylate cyclase associated with loss of β-adrenergic receptors binding sites. Proc. Natl. Acad. Sci. USA 1975; 72:1945–49.

[32] Mickey, J., Tate, R., and Lefkowitz, R. J.: Subsensitivity of adenylate cyclase and decreased β-adrenergic receptor binding after chronic exposure to isoproterenol in vitro. J. Biol. Chem. 1975; 250:5727–29.

[33] Remold-O'Donnell, E.: Stimulation and desensitization of macrophage adenylate cyclase by prostaglandins and catecholamines. J. Biol. Chem. 1974; 249:3615–21.

[34] Shear, M., Insel, P. A., Melmon, K. L., and Coffino, P.: Agonist-specific refractoriness induced by isoproterenol. J. Biol. Chem. 1976; 251:7572–76.

[35] Makman, M. H.: Properties of adenylate cyclase of lymphoid cells. Proc. Natl. Acad. Sci. USA 1971; 68:885–89.

[36] Sonnenberg, G. E., Stauffacher, W., and Keller, U.: Failure of glucagon to stimulate ketone body production during acute insulin deficiency of insulin replacement in man. Diabetologia 1983; 23:94–100.

[37] Wahren, J., Felig, P., and Hagenfeldt, L.: Effect of protein ingestion of splanchnic and leg metabolism in normal man and in patients with diabetes mellitus. J. Clin. Invest. 1976; 57:987–99.

[38] Fine, M. B., and Williams, R. H.: Effect of fasting, epinephrine and glucose and insulin on hepatic uptake of nonesterified fatty acids. Am. J. Physiol. 1960; 199:403–406.

[39] Bearn, A. G., Billing, B., and Sherlock, S.: The effect of adrenaline and noradrenaline on hepatic blood flow and splanchnic carbohydrate metabolism in man. J. Physiol. 1951; 115:430–41.

[40] Schade, D. S., and Eaton, P. R.: The regulation of plasma ketone body concentrations by counterregulatory hormones in man. I. Effects of norepinephrine in diabetic man. Diabetes 1977; 26:989–96.

[41] Day, J. L.: The metabolic consequences of adrenergic blockade: a review. Metabolism 1975; 24:987–96.

[42] Goldberg, L. J.: Drugs acting on the autonomic nervous system. *In* The Heart. Hurst, J. W., Ed. New York, McGraw-Hill, 1978:1980–83.

[43] Cherrington, A. D., Caldwell, M. D., Dietz, M. R., Exton, J. H., and Crofford, O. B.: The effect of somatostatin on glucose uptake and production by rat tissues in vitro. Diabetes 1977; 26:740–48.

Downloaded from http://diabetesjournals.org/diabetes/article-pdf/33/8/738/353322/33-8-738.pdf by guest on 01 December 2023