Insulin and Ketone Responses to Ingestion of Medium and Long-chain Triglycerides in Man

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SUMMARY

The effects of medium chain triglyceride (MCT), corn oil, and water on serum glucose, ketones, and immunoreactive insulin (IRI) were compared. Fourteen normal subjects ingested 1 gm. per kg. of MCT (composed principally of C8 and C10 fatty acids) and on another occasion an equal volume of water. After water ingestion, glucose, ketones, and IRI did not change appreciably up to five hours. After MCT, a significant rise in serum ketones and IRI was associated with a slight fall in serum glucose. Fourteen normal subjects ingested 1 gm. per kg. corn oil, and on another occasion an equal volume of water. Serum glucose, ketone and IRI concentrations again did not change up to five hours after ingestion of water. After corn oil, no significant rise in ketones occurred. There was a small rise in IRI and a slight fall in glucose. In the eight subjects in whom MCT, corn oil and water were compared a certain degree of insulin secretory response was obtained after MCT *and corn oil but not after water ingestion. However,* the IRI response after MCT was highest. The rise in IRI without a corresponding change in serum ketones after ingestion of the long chain triglyceride might be attributed to a gastrointestinal β-cytotropic effect. The mechanism of IRI stimulation by MCT also has been related to MCT-induced hyperketonemia and possibly to a direct effect on insulin secretion of small amounts of octanoate in peripheral blood. DIABETES *18*:96-100, February, 1969.

The hypoglycemia induced by administration of acetoacetate and other ketones has been ascribed to an insulin stimulatory effect.[1] The degree of hyperketonemia at which this stimulatory effect occurs has not been clarified. The ingestion of medium chain triglycerides can induce a modest hyperketonemia in man.[2,3] The purpose of the present study was to ascertain the insulin secretory response to medium chain triglyceride-induced hyperketonemia in normal human subjects. Also, control experiments were conducted to estimate the insulin secretory response of subjects to the ingestion of a long chain triglyceride or simply water.

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MATERIALS AND METHODS

Initially, fourteen normal subjects (ten men and four women) were studied on two separate mornings at least one week apart. All subjects were lean, healthy volunteers, eighteen to thirty-five years of age. Following overnight fast, each individual ingested either 1 gm. per kg. body weight of medium chain triglyceride (MCT) or an equivalent volume of water. The fatty acid composition of the MCT preparation was approximately 80 per cent octanoate (C8) and 17 per cent decanoate (C10) with traces of laurate (C12) and hexanoate (C6).[4]

Additional studies were made by feeding volunteers 1 gm. per kg. body weight of long chain triglyceride (corn oil). Eight of the original volunteers and six new ones were available for this purpose. The MCT or the corn oil was emulsified by ultrasonication in an equal amount of water containing 0.5 ml. of Tween 80. The same amount of Tween 80 was added to the water control "meal." Venous blood samples were obtained before and for five hours after the ingestion of either the fat or the water. After the blood was permitted to clot on ice for three hours, the serum was separated and immediately frozen until use. The serum samples were analyzed for glucose,[5,6] total ketones,[7,8] and immunoreactive insulin.[9,10] The immunoreactive insulin (IRI) was measured against a human insulin standard.* Duplicate values of the per cent activity of the bound insulin precipitates had to agree within 3 per cent to be considered valid.

*Kindly supplied by Dr. Mary Root, Lilly Laboratories, Indianapolis, Indiana.

RESULTS

The results obtained in the fourteen subjects who took MCT or water are shown in figure 1. In presentation of the results, all values represent means unless otherwise specified; those described as "baseline" represent the average of the pre-ingestion means measured at —30 and —5 min. The baseline postabsorptive serum ketone levels in the fourteen subjects studied were 0.77 ± 0.26 (S.E.) mg. per 100 ml. After water ingestion serum ketones remained at a level below 1.3 mg. per 100 ml. After MCT serum ketones rose to a peak, however, of 6.7 mg. per 100 ml. at two hours, and then were maintained at a level above 6.0 mg. per 100 ml. for five hours. Serum insulin concentrations ranged from 6.6 to 11. 2 μU. per ml. throughout the water ingestion period, the average of these means being 8.2 μU. per ml. After MCT insulin rose from a baseline level of 9.3 μU. per ml. to values of 17.7 μU. per ml. at thirty minutes ($p < .005$), 15.3 μU. per ml. at one hour ($p < .005$) and 15.4 μU. per ml. at two hours ($p < .05$). Values at four and five hours were elevated above the baseline value, but the differences were not significant. Serum glucose remained constant at about 85 mg. per 100 ml. before and after water ingestion. After MCT there was a slight fall in serum glucose values varying between 6 and 8 mg. per 100 ml. which, after one hour, were significantly lower ($p < .005$) than the preingestion values.

The results in the fourteen volunteers who ingested corn oil and then water are shown in figure 2. Serum ketones did not change significantly after water ingestion, remaining at a level below 1.2 mg. per 100 ml. After corn oil, ketones rose very slightly with the highest level of 2.0 mg. per 100 ml. recorded at five hours. Serum insulin concentrations after water did not rise but actually fell slightly below the baseline level of 10.4 μU. per ml. After corn oil, insulin rose from a baseline concentration of 10.9 μU. per ml. to values of 12.7 μU. per ml. at thirty minutes, 13.4 μU. per ml. at one hour, 13.8 μU. per ml. at two hours, diminishing again to 11.9 μU. per ml. at four hours and 10.2 μU. per ml. at five hours. With the exception of the two-hour concentration ($p < .05$), these values did not differ significantly from the baseline level. Serum glucose did not change during water ingestion, while after corn oil there was a slight fall in glucose which, at sixty minutes, was lower than the pre-ingestion values ($p < .05$).

A comparison of results obtained after water, corn oil

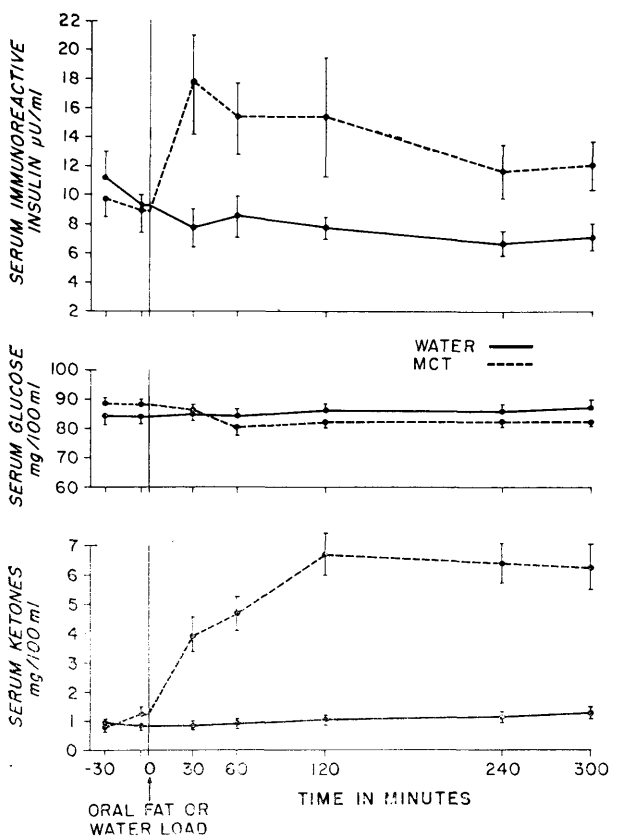

FIG. 1. Mean serum immunoreactive insulin, glucose, and ketone responses (± S.E.) to ingested water and MCT in fourteen normal subjects.

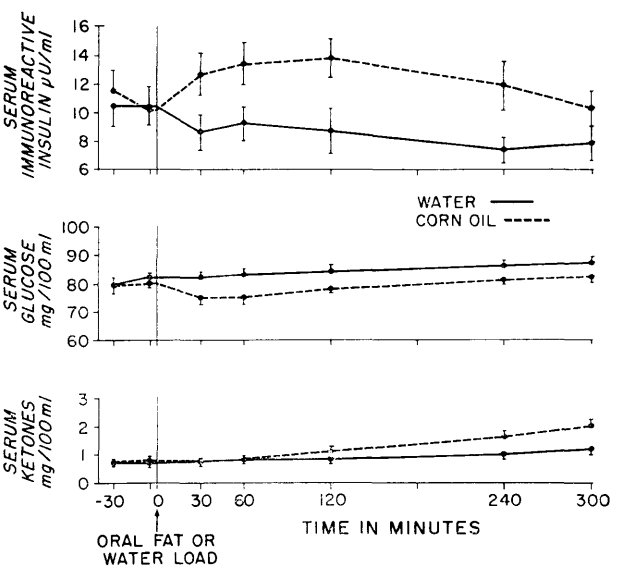

FIG. 2. Mean serum immunoreactive insulin, glucose, and ketone responses (± S.E.) to ingested water and corn oil in fourteen normal subjects.

and MCT in the eight subjects who underwent all three studies is shown in figure 3 and table 1. When the IRI responses to MCT were compared with those after water, there was a significantly ($p < 0.025$) higher IRI response throughout the period following ingestion of MCT. In contrast, when IRI responses to corn oil were compared with those obtained after water ingestion, significantly higher IRI responses were detected only at thirty and 120 minutes. When MCT was compared with corn oil, significantly higher IRI responses were obtained with MCT at thirty minutes.

DISCUSSION

MCT has been shown previously to induce modest hyperketonemia in man.[2,3,11] This phenomenon has been related to the perfusion of the liver by medium chain fatty acids which travel in the portal vein after digestion and absorption of the fatty acids and monoglyceride derivatives of the parent triglyceride.

The hypoglycemic action of ketone bodies infused into animals has been reported by a number of investigators.[12,13] The hypoglycemia induced by ketone administration in portacaval shunted dogs by Mebane and Madison[14] was ascribed entirely to a reduction in hepatic glucose output. Subsequently, Madison et al.[1]

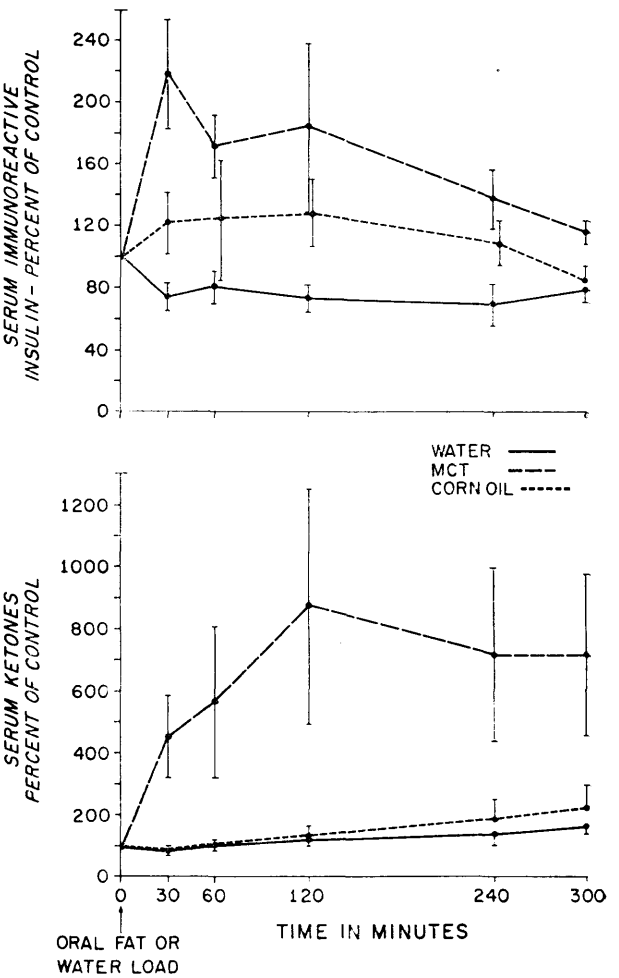

FIG. 3. Mean serum immunoreactive insulin and ketone responses (per cent of control ± S.E.) to water, corn oil, and MCT ingestion in eight normal subjects.

TABLE 1

Mean serum immunoreactive insulin (IRI), glucose, and ketone responses (± S.E.) to water, corn oil, and MCT ingestion in eight normal subjects. Values in parentheses denote per cent of control, with 100 per cent being the average of the two pre-ingestion values at —30 and —5 minutes.

Time in minutes		−30	−5	+30	+60	+120	+240	+300
	IRI	10.5±2.6	8.7±1.8	7.1±1.6 (74)	7.8±1.5 (81)	7.0±1.2 (73)	6.6±1.2 (69)	7.5±1.7 (78)
Water	Glucose	83±2.8	85±2.3	84±3.1 (100)	85±2.9 (101)	86±3.8 (102)	88±2.8 (105)	89±3.4 (106)
	Ketones	0.9±0.2	0.7±0.2	0.7±0.1 (88)	0.8±0.2 (103)	0.9±0.2 (120)	1.1±0.2 (136)	1.3±0.3 (161)
	IRI	10.9±2.3	10.1±1.7	12.8±2.5 (122)	13.0±2.5 (124)	13.3±1.5 (127)	11.4±2.7 (109)	8.9±1.7 (85)
Corn oil	Glucose	82±3.6	82±2.3	77±3.9 (94)	76±3.4 (93)	79±2.5 (96)	82±2.6 (100)	83±2.8 (101)
	Ketones	0.8±0.2	0.9±0.3	0.8±0.2 (87)	0.9±0.2 (105)	1.1±0.2 (129)	1.6±0.2 (185)	1.9±0.2 (221)
	IRI	10.7±2.1	8.4±1.6	20.8±6.5 (218)	16.4±4.5 (172)	17.6±7.2 (184)	13.1±2.8 (137)	11.1±2.2 (116)
MCT	Glucose	87±3.6	87±1.3	85±2.0 (98)	78±2.2 (90)	83±2.5 (95)	81±2.4 (93)	82±1.6 (94)
	Ketones	0.8±0.2	1.0±0.2	4.1±0.7 (450)	5.1±0.9 (561)	7.9±0.9 (870)	6.6±0.9 (712)	6.5±1.1 (718)

demonstrated enhanced insulin levels in pancreatic venous blood after ketone infusions, but these workers could not obtain a similar hypoglycemia in depancreatized dogs. The suggestion was made that the modestly increased insulin levels induced by hyperketonemia dampen further ketone production by regulating free fatty acid release from adipose tissue thereby preventing severe ketoacidosis during starvation. Since no direct ketone measurements were made by Madison et al., the degree of ketonemia at which an insulin stimulatory effect would occur was left undefined, albeit these authors estimated that an insulin secretory effect occurred at levels of 3.5 mMoles per liter (approximately 21 mg. per 100 ml. of acetone equivalent)

In the present experiments, the modest hyperketonemia induced by MCT was associated with a small rise in circulating IRI. Although serum IRI concentrations of 18 μU. per ml. would be considered within the normal postabsorptive range, the changes that occurred after MCT were not evident after water. The change in serum IRI in response to MCT may be related in part to the hyperketonemia. This interpretation is consistent with the studies in dogs by Madison et al.[1] However, Fajans et al.[15] attained plasma ketone values of 25 mg. per 100 ml. after infusing acetoacetate into normal volunteers and were unable to detect a rise in plasma IRI. Moreover, infusion of β-hydroxybutyrate into man has not been found by others[16,17] to enhance insulin secretion.

The possibility that octanoate is capable of directly stimulating release of insulin by the beta cells of the pancreas should be considered. Sanbar et al.[18] infused octanoate and found a slight increase in peripheral plasma IRI in three of the six dogs studied. Other studies of infusion of octanoate in dogs have not shown enhanced insulin secretion.[19] In any event, these studies involve direct introduction of octanoate into the systemic circulation. Under the conditions of the present study, MCT ingestion results in transport of octanoate and decanoate into the portal vein and the liver where these acids are extensively metabolized.[20] Thus, the ingestion of MCT is not associated with an appreciable rise of MCT-derived acids in the systemic circulation.[21] Whether the small rise in arterial blood octanoate that might occur after MCT ingestion could directly stimulate insulin secretion cannot be determined from the present study.

Since serum ketones exhibited virtually no rise after corn oil, it is unlikely that they played a role in the small rise in circulating IRI observed. However, the presence of ingested fat in the gastrointestinal tract may result in the release of certain intestinal hormones. Some of these hormones have been shown by Unger et al.[22] to stimulate insulin secretion. Thus, it is possible that the enhancement of insulin secretion in response to corn oil ingestion may be ascribed to a direct effect of the fat load on the gastrointestinal tract with release of β-cytotropic hormones. However, direct evidence for this possibility is not provided by the present study.

When the IRI responses were compared after MCT, corn oil, and water in the same eight subjects, it was evident that MCT induced the highest insulin response. Thus, the mechanism of IRI stimulation by MCT must be related to an effect other than a possible fat-mediated gastrointestinal hormonal release. Whether the higher degree of insulin response to MCT ingestion is induced by the hyperketonemia or the small amount of octanoate that may have escaped hepatic metabolism cannot be determined from the present study.

ACKNOWLEDGMENT

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Body Odor and Metabolic Defects

...N-Butyric/N-hexanoic acidemia is another rare condition in which the exhaled air, tissues, and body fluids have the strong odor of sweaty feet. This condition has so far been observed in two families, involving four and possibly five infants (J. B. Sidbury, Jr., W. R. Harlan, and B. Wittels, Am. J. Dis. Child. *104*:531, 1962; Sidbury, E. K. Smith, and Harlan, J. Pediat. *70*:8, 1967). This abnormality of short chain fatty acid metabolism, in which the odor is caused by butyric and hexanoic acids, is probably an autosomal recessive disorder and is due to a deficiency of the acyl dehydrogenase specific for the metabolism of four and six chain fatty acids. Feeding difficulties, lethargy, weakness, and odor developed in these children in the first days of life and were followed by severe acidosis, dehydration, and convulsions reminiscent of tetany. Death ensued in the first month from bone marrow depression with pancytopenia, a bleeding diathesis, and gram negative sepsis.

Other rare disorders which must be considered when an infant has a peculiar odor include the oasthouse syndrome, hypermethioninemia, and methionine malabsorption.

The child with oasthouse syndrome described by A. J. Smith and L. B. Strang (Arch. Dis. Child. *33*:109, 1958), had a distinctive unpleasant pungent smell reminiscent of a brewery or oasthouse. The clinical pattern was like that of phenylpyruvic oligophrenia, but differed in unresponsiveness to stimuli, muscular hypotonicity, recurrent episodes of fever, tachypnea, and edema. White hair grew at nine months and death occurred at ten months of age. A double metabolic defect existed, since phenylketonuria was also present. The smell of the urine differed from the mousy odor of phenylacetic acid, which is commonly excreted in phenylketonuria, and was thought due to an oxidation or polymerization product of alpha-hydroxybutyric acid....

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