Intravenous Niacin Acutely Improves the Efficiency of Dietary Fat Storage in Lean and Obese Humans

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Spillover of fatty acids released by lipoprotein lipase hydrolysis of meal triglycerides may be a major contributor to the free fatty acid (FFA) pool. We studied lean (n = 6) and overweight and obese (n = 5) subjects during continuous feeding on two occasions: during intravenous infusion of niacin (2.8 mg/min) and saline. After establishment of steady-state chylomicronemia and suppression of adipose tissue lipolysis with a liquid meal, spillover was measured with infusions of [U-^{13}C]oleate and [^{3}H]triolein. Total FFA concentrations were lower during niacin infusion in both lean (50 ± 4 vs. 102 ± 7 μmol/L; $P < 0.002$) and obese (75 ± 6 vs. 143 ± 13 μmol/L; $P < 0.01$) subjects. Oleate appearance was lower during niacin infusion than during saline infusion in both lean (21 ± 2 vs. 32 ± 5 μmol/min; $P = 0.07$) and obese subjects (25 ± 3 vs. 46 ± 8 μmol/min; $P < 0.02$). Spillover was lower during niacin infusion than during saline infusion in lean (21 ± 4 vs. 29 ± 3%) and obese (21 ± 2 vs. 29 ± 5%) subjects ($P <$ 0.03 for both). In summary, during meal absorption, niacin produces additional suppression of lipolysis and a reduction in fractional spillover compared with saline in both normal and obese subjects. Infusion of intravenous niacin provides a model for acutely improving dietary fat storage, perhaps by suppressing lipolysis in visceral adipose tissue. ***Diabetes*** **61:3172–3175, 2012**

Abnormalities in free fatty acid (FFA) metabolism contribute to insulin resistance, type 2 diabetes, and dyslipidemia (1). Dyslipidemia is thought to be driven by increased visceral lipolysis (2). It has become apparent in recent years that spillover of fatty acids from the action of lipoprotein lipase (LPL) on triglyceride (TG)-rich lipoproteins, especially chylomicrons, may be an important contributor to circulating plasma FFA concentrations (3,4). In overweight and obese humans, the majority of systemic spillover appears to occur in the splanchnic bed (5), where there is a strong correlation between spillover and intracellular lipolysis (6). This suggests that spillover and intracellular lipolysis are linked and that improvement in spillover may depend on suppression of intracellular lipolysis. We have recently found that hyperinsulinemia produced by an insulin infusion during continuous meal ingestion had no effect on spillover in dyslipidemic overweight and obese volunteers (7). Visceral fat is relatively resistant to the antilipolytic effects of insulin (8) and is known to contain receptors for niacin (9). Therefore, this study was undertaken to determine whether suppression of lipolysis with intravenous niacin decreases spillover during continuous feeding in lean and dyslipidemic obese nondiabetic humans.

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RESEARCH DESIGN AND METHODS

A total of 16 volunteers gave informed consent. Four individuals were studied using a pilot protocol to optimize the feeding regimen and the rate of niacin infusion. Lean subjects (n = 6) and overweight or obese subjects with mild to moderate dyslipidemia (n = 5) were studied on two occasions. Subjects with dyslipidemia had fasting TG levels >150 mg/dL, and all but one had an HDL-cholesterol level <40 mg/dL. All had normal blood pressure and normal function of the thyroid, liver, and kidney; all were screened with a 2-h glucose tolerance test to rule out diabetes. None were taking lipid-modifying medications. A sixth obese individual did not complete the protocol because of a superficial thrombophlebitis after the first study.

Study protocol. Subjects were studied according to a protocol approved by the Mayo Institutional Review Board. Beginning 5 days before admission to the Clinical Research Unit, subjects followed a controlled diet for weight maintenance containing 55% carbohydrate, 30% fat, and 15% protein. The diet was continued for the duration of the study.

Subjects were admitted to the Clinical Research Unit the afternoon before the study. Visceral and total abdominal fat were determined with single-slice computed tomography scan, and total body fat was measured with dual-energy X-ray absorptiometry on the first study day only. The participants then consumed an evening meal containing 10 kcal/kg. After an overnight fast, a catheter was placed in a forearm vein for isotope infusion. A second catheter was placed in a retrograde fashion into a vein of the dorsum of the hand contralateral to the infusion catheter. The hand was placed in a heated Plexiglas box maintained at 55°C to allow sampling of arterialized blood.

A liquid meal made from chocolate-flavored Ensure Plus with additional canola oil to match the macronutrient distribution in the controlled diet was prepared and sonicated with a Sonicator XL ultrasonic probe (Misonix Inc., Farmingdale, NY) to ensure thorough mixing. An ~80 mL bolus of the meal was administered at 0 min, followed by ~20 mL doses every 15 min beginning 120 min after the bolus and continuing until 15 min before the conclusion of the study. For each individual, the meal provided one-third of the estimated daily basal energy expenditure (Harris-Benedict equation). This protocol results in steady-state chylomicrons TG and FFA concentrations (10). An infusion of [9,10-^{3}H]triolein (~1.2 μCi/min) in a commercial lipid emulsion (4,10) was started at 270 min and continued until blood sampling was completed. An infusion of [U-^{13}C]oleate (0.5 nmol · kg^{-1} · min^{-1}) was initiated no later than 270 min and continued to the end of the study. At 270 min, an infusion of niacin (at an initial rate of 0.6 mg/min and increased gradually to 2.8 mg/min over 30 min, as described previously [11]) or normal saline, in random order on the two study days, was initiated and continued to the end of the study. The total niacin dose was ~285 mg for each subject. Blood samples were taken every 15 min from 330 to 390 min for measurement of plasma TG, chylomicron TG, and FFA concentrations and [^{3}H]oleate-specific activity and [^{13}C]oleate enrichment.

Participants underwent the second study after a 2-week washout period, using a protocol identical to the first study except that subjects who received niacin at the first visit received a saline infusion and vice versa.

Methods. Niacin was obtained from Spectrum Chemical Manufacturing Corporation (New Brunswick, NJ) and administered intravenously under the U.S. Food and Drug Administration's IND #77,935. Niacin was dissolved in 0.9% sodium chloride and titrated to pH 7.06 with a solution of 1.2 N sodium hydroxide containing 0.01 mol/L potassium phosphate. It was then filtered through a 0.22-μ filter into 500 mg unit dose vials; sterility was confirmed by culture and pyrogen testing before use.

Plasma FFA concentration and specific activity were determined by high-performance liquid chromatography (12). [^{13}C]Oleate enrichment was determined by liquid chromatography–mass spectrometry (13). Chylomicrons were isolated by ultracentrifugation (10). Total TG and chylomicron TG concentrations were determined on a Cobas Integra autoanalyzer (Roche

TABLE 1
Subject characteristics and screening laboratory data

	Lean subjects (n = 6)	Obese subjects (n = 5)
Age (years)	31 ± 3	39 ± 3
Sex (female/male)	1/5	2/3
Weight (kg)	72 ± 4	88 ± 9
BMI (kg/m^2)	22.7 ± 0.6	30.1 ± 1.2**
Total body fat (%)	22 ± 2	35 ± 2**
Total abdominal fat (cm^2)	147 ± 39	388 ± 69*
Visceral fat (cm^2)	54 ± 17	175 ± 41*
Total cholesterol (mg/dL)	160 ± 6	185 ± 19
HDL cholesterol (mg/dL)	59 ± 4	38 ± 3*
LDL cholesterol (mg/dL)	85 ± 5	100 ± 16
TG (mg/dL)	76 ± 12	238 ± 19**
Fasting glucose (mg/dL)	86 ± 1	91 ± 4

Data are mean ± SEM unless otherwise indicated. *$P < 0.05$ vs. lean subjects. **$P < 0.002$ vs. lean subjects.

Diagnostics, Indianapolis, IN). Glucose concentrations also were measured on the Integra analyzer.

Data analysis. The averages of the measured data from the final five time points were used for calculations of total oleate rate of appearance and fractional spillover (from the chylomicron precursor pool) under steady-state conditions, as described previously (4). Data are expressed as mean ± SEM. Between-group comparisons were made with a t test for unequal variance, and within-group comparisons were made with a paired t test. All comparisons required $\alpha < 0.05$ for significance. Statistical calculations were done using the data analysis package in Microsoft Excel (Microsoft Corp., Redmond, WA).

RESULTS

Subject characteristics are shown in Table 1. Based on enrollment criteria, BMI and TGs were higher, and HDL cholesterol was lower in overweight and obese subjects. Total body, abdominal, and visceral fat were all significantly greater in the overweight and obese group. Insulin concentrations were borderline higher in overweight and obese subjects (P = 0.06). Niacin infusion was well tolerated; mild, mostly asymptomatic flushing during the first 30 min of niacin infusion was the only side effect.

Plasma glucose, insulin, TG, and FFA concentrations are shown in Table 2. Glucose concentrations were higher in obese volunteers than in lean volunteers during continuous feeding. TG concentrations were higher in obese subjects than in lean subjects at baseline and during feeding on both study days. Plasma FFA concentrations were higher in obese subjects compared with lean subjects at baseline and during feeding on both study days, and they were lower during niacin infusion than during saline infusion in both groups of subjects.

As can be seen in Fig. 1, steady-state oleate concentrations were achieved during the continuous feeding period. Plasma oleate [U-^{13}C] enrichments and [^{3}H] specific activities also were at steady state (Supplementary Fig. 1). Oleate rate of appearance was lower during niacin infusion compared with saline infusion in both lean (with borderline significance, P = 0.07) and obese subjects (Table 2). Oleate was 42 ± 1% of meal fat, and oleate ingestion rate was 93 ± 7 μmol/min. Fractional spillover was lower during niacin infusion in both lean (21 ± 4 vs. 29 ± 3%; P = 0.028) and obese (21 ± 2 vs. 29 ± 5%; P = 0.027) subjects (Fig. 2).

DISCUSSION

Spillover of fatty acids derived from LPL hydrolysis of chylomicron TGs is a potential major source of circulating FFA and reflects inefficient dietary fat storage (4). However, little information is available concerning regulation of the spillover process. In this study, spillover was measured in lean volunteers and in overweight and obese individuals with dyslipidemia during continuous feeding on two occasions: once during infusion of saline and once during infusion of niacin. Niacin infusion produced a nearly 30% decrease in fractional spillover in both groups.

The majority of systemic spillover appears to occur in the splanchnic bed (5), and for this reason it is tempting to speculate that niacin has a major effect on splanchnic spillover. If this were the case, niacin could very well exert such an effect via potent inhibition of intracellular lipolysis in visceral fat because spillover in that tissue correlates strongly with intracellular lipolysis (6). We have recently conducted studies of dyslipidemic overweight and obese individuals in whom an insulin infusion had no effect on

TABLE 2
Average plasma glucose, insulin, total TG, and chylomicron TG concentrations and oleate rate of appearance at baseline and during continuous feeding, before and during infusion of saline or niacin in lean and obese subjects

	Lean subjects		Obese subjects	
	NaCl	Niacin	NaCl	Niacin
Plasma glucose (mg/dL)				
Baseline	82 ± 4	82 ± 3	94 ± 4†	93 ± 5
330–390 min	95 ± 2**	88 ± 4	107 ± 3**†	103 ± 5**†
Serum insulin (μU/mL)				
Baseline	4.4 ± 1.0	3.6 ± 0.8	10.1 ± 2.6	12.0 ± 2.5†
330–390 min	10.2 ± 0.9*	10.3 ± 2.5*	38.6 ± 13.7	42.9 ± 4
Total plasma TGs (mg/dL)				
Baseline	77 ± 15	74 ± 12	145 ± 10††	137 ± 15†
330–390 min	106 ± 15*	89 ± 15	219 ± 19**††	197 ± 25**††
Chylomicron TGs (mg/dL), 330–390 min	31 ± 8	20 ± 5	23 ± 9	20 ± 9
Plasma FFA (μmol/L)				
Baseline	250 ± 15	226 ± 25	357 ± 30†	336 ± 13††
330–390 min	102 ± 2**	50 ± 4**∞	143 ± 13**†	75 ± 6**∞††
Oleate R_a, 330–390 min	32 ± 5	21 ± 2	46 ± 8	25 ± 3∞

R_a, rate of appearance. *$P < 0.02$ vs. baseline. **$P < 0.01$ vs. baseline. ∞$P < 0.01$ vs. saline. †$P < 0.05$ vs. lean subjects. ††$P < 0.01$ vs. lean subjects.

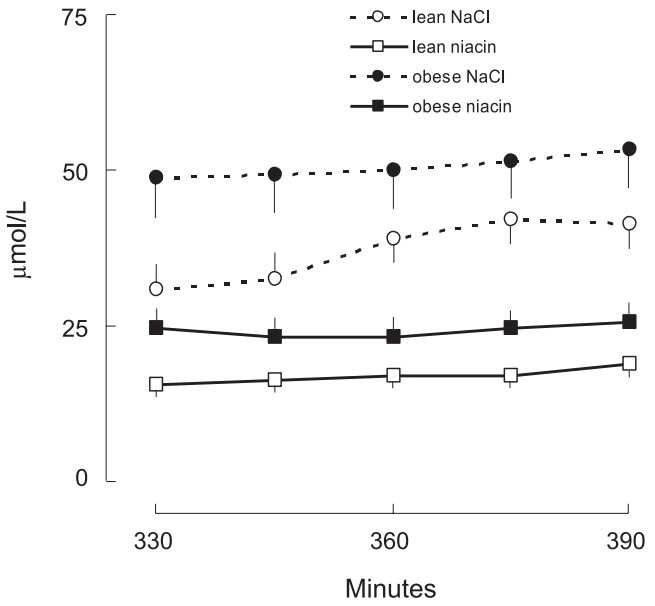

FIG. 1. Plasma oleate concentrations during continuous feeding and infusion of niacin or saline in lean and overweight or obese subjects.

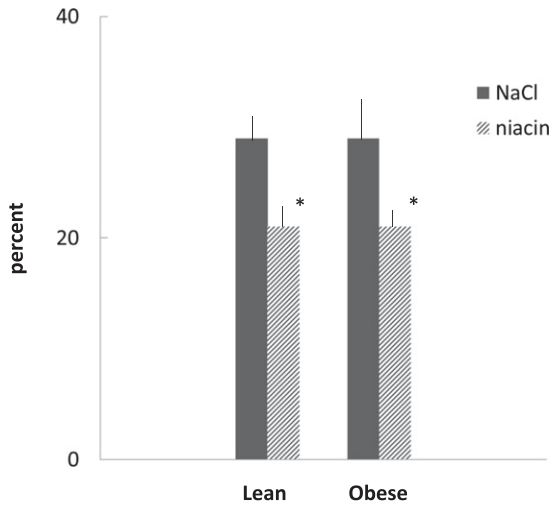

FIG. 2. Fractional spillover of [^{3}H]oleate during continuous feeding before and during infusion of niacin or saline in lean and overweight or obese subjects. *$P < 0.03$ vs. baseline.

systemic spillover in spite of a ~40% suppression of plasma FFA concentrations (7). The failure of insulin to produce a decrease in spillover may be attributed to the well-known resistance of visceral adipose tissue to the antilipolytic effects of that hormone (8,14). To our knowledge, no in vivo data are available on the effects of niacin on visceral lipolysis.

These experiments were conducted in the context of continuous feeding. This design produces suppression of intracellular lipolysis similar to that occurring in response to bolus ingestion of a mixed meal, but it allows observations under steady-state conditions, which is desirable when studying precursor-product relationships with tracers (15). Infusion of niacin produced an ~50% decrease in plasma FFA concentrations in addition to that achieved by the meal compared with saline infusion, accompanied by a significant decrease in spillover.

Niacin is known to suppress adipose tissue lipolysis in individuals with atherogenic dyslipidemia, improving hypertriglyceridemia and elevated plasma FFA concentrations (16). Treatment with niacin (nicotinic acid) and its analogs has been shown to reduce FFA concentrations and flux (16), reduce TG concentrations (17,18), and acutely improve insulin sensitivity (19). However, the mechanism by which niacin lowers TGs is far from clear. In fact, the suppressive effect on lipolysis of crystalline niacin given chronically is so evanescent—there is a rebound in FFA concentrations to levels above baseline within 6 h of an oral dose (J.M.M., W. Isley, and W. Harris, unpublished data)—that it produces a net increase in around-the-clock FFA availability and actually worsens insulin resistance (16,18). This observation, together with the known ability of niacin to reduce VLDL TG synthesis in the liver (20), suggests that either niacin has a sustained antilipolytic effect in visceral fat that is not reflected in peripheral FFA concentrations or that marked suppression of portal venous FFA concentrations in the early postprandial phase has an inordinate and sustained effect on intrahepatic fatty acid trafficking.

The labeled lipid emulsion used to trace meal fat disposition does not contain cholesterol esters or the apolipoprotein C2 required for LPL to hydrolyze TG-rich lipoprotein TGs. However, it rapidly acquires the latter when it is infused (21). Moreover, the regional distribution of tracer uptake (6) is very similar to that seen when native chylomicrons are labeled (22). Considering the possibility that there may be temporary sequestration of dietary fat in enterocytes after meals (23), the lipid emulsion actually may be preferable to labeling of meal fat for the measurement of spillover.

The combined effect of sustained suppression of visceral lipolysis (if it indeed occurs) and a reduction in spillover in visceral fat, where ~14% of systemic disposal of meal fat takes place (6), could have an important effect on FFA delivery to the liver, with implications for hepatic insulin sensitivity, VLDL production, and hepatic steatosis (24,25). Additional studies are needed to investigate the pharmacodynamics of niacin in different adipose tissue depots.

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R.H.N. was involved in every aspect of the project, conducted the studies, analyzed data, and cowrote the manuscript. D.V. and A.S. helped with planning and executing the study and data analysis, edited the manuscript, and contributed to the discussion. J.M.M. was involved in study design and data analysis and cowrote the manuscript. J.M.M. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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