Effect of Meal Frequency on Blood Glucose, Insulin, and Free Fatty Acids in NIDDM Subjects

JETTE BERTELSEN, MD
CHRISTIAN CHRISTIANSEN, MD
CLAUS THOMSEN, MD
PER L. POULSEN, MD
SVEND VESTERGAARD, MD
ALICE STEINOV, MD
LARS H. RASMUSSEN, MD
OLE RASMUSSEN, MD
KJELD HERMANSEN, MD

OBJECTIVE — We studied the effects of meal frequency on blood glucose, serum insulin, and FFAs in 12 NIDDM subjects.

RESEARCH DESIGN AND METHODS — Subjects were assigned in random order to two 8-hr observation periods after an overnight fast. They received isocaloric diets with similar composition either as six small or as two large meals. At the end of each study period, an IVGTT was given.

RESULTS — Two large meals induced an 84% greater maximum amplitude of glucose excursions than six small meals (6.1 ± 0.5 vs. 3.3 ± 0.5 mM, $P < 0.005$) and higher insulin responses ($P < 0.03$). The K_g response to an IVGTT did not differ in the two situations. The average FFA level was lowest in response to frequent meals ($P < 0.02$).

CONCLUSIONS — A higher meal frequency acutely subdues glucose excursions and reduces insulin and FFA levels during the daytime in older NIDDM subjects.

Individuals with NIDDM are instructed to partition their caloric intake into several small, rather than into a few large, meals in order to optimize metabolic control and enhance weight reduction efforts (1,2). Controversy exists about the metabolic impact of meal frequency in healthy volunteers (3,4). Thus, Gwinup et al. (3) found subdued blood glucose levels in response to oral glucose during a period with food intake distributed evenly throughout the day compared with a period on an isocaloric diet taken as a single, daily large meal. Jenkins et al. (4), on the other hand, found similar daily profiles of blood glucose, but subdued insulin levels in response to a 2-wk diet with many small meals (nibbling) compared with a three-meal isocaloric diet. Recently, the acute effect of the temporal pattern of caloric intake has been studied in normal and NIDDM subjects (5,6). It was found that continuous sipping of glucose elicited similar average blood glucose levels, but lower insulin and FFA levels compared with a single bolus glucose in normal subjects (5). In NIDDM, the temporal pattern of food intake did not affect the overall level of blood glucose and inconsistently influenced the insulin levels (6). In view of the potential importance of any dietary maneuver that might improve glycemic and insulinemic control, we examined the effects of two different temporal patterns of caloric intake on the levels of blood glucose, insulin, and FFA in 12 NIDDM subjects.

RESEARCH DESIGN AND METHODS

This study included 12 NIDDM subjects (8 males, 4 females) who were fully informed of the experimental nature of the study, which had received approval from the local ethical committee. The subjects' clinical characteristics are given in Table 1. Subjects with diabetic complications, except simplex retinopathy, were excluded. None of the participants was treated with insulin; 5 subjects were treated with diet alone. The 7 participants taking oral hypoglycemic agents took their prescribed medicine throughout the study.

The participants, who were randomly assigned to study A or B, were investigated on two occasions after an overnight fast with an interval of 1–3 wk. In study A, six isocaloric, small meals with an identical composition were served from 0800 in 80-min intervals. In study B, two large, isocaloric

FROM THE MEDICAL DEPARTMENT, HORSENS HOSPITAL; MEDICAL DEPARTMENT M (DIABETES AND ENDOCRINOLOGY), AARHUS KOMMUNEHOSPITAL; AND MEDICAL DEPARTMENT C (ENDOCRINOLOGY), AARHUS AMTSSYGEHUS, DENMARK.

ADDRESS CORRESPONDENCE AND REPRINT REQUESTS TO KJELD HERMANSEN, MEDICAL DEPARTMENT C, AARHUS AMTSSYGEHUS, TAGE-HANSENSGADE 2, DK-8000 AARHUS C, DENMARK.

RECEIVED FOR PUBLICATION 5 MARCH 1992 AND ACCEPTED IN REVISED FORM 18 AUGUST 1992.

NIDDM, NON-INSULIN-DEPENDENT DIABETES MELLITUS; FFA, FREE FATTY ACID; IVGTT, INTRAVENOUS GLUCOSE TOLERANCE TEST; K_g, GLUCOSE DISAPPEARANCE RATE; FBG, FASTING BLOOD GLUCOSE; ANOVA, ANALYSIS OF VARIANCE; BMI, BODY MASS INDEX.

Table 1—*Clinical data for the 12 NIDDM patients*

Sex	Duration of diabetes (yr)	Age (yr)	BMI (kg/m^2)	HbA_{1c} (%)	FBG (mM) Study A	FBG (mM) Study B
8 male/4 female	5 ± 1	64 ± 2	32.2 ± 1.3	6.6 ± 0.4	8.2 ± 2.2	8.3 ± 2.1

Values are means ± SE. Study A, six identical meals in 8 h. Study B, two identical meals in 8 h.

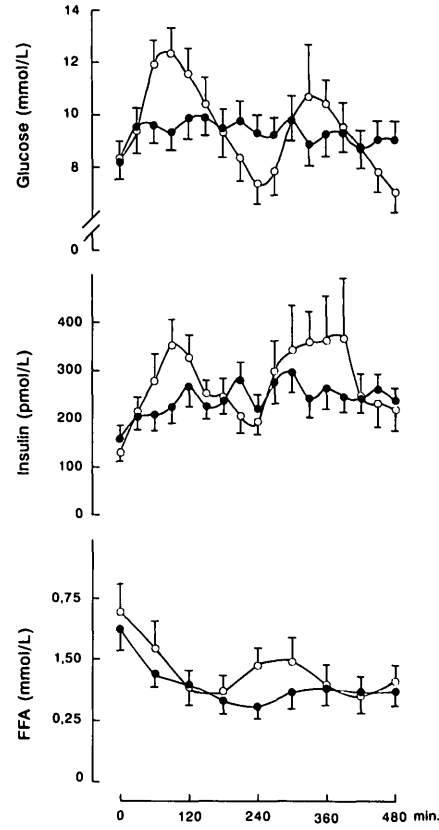

Figure 1—*Blood glucose, insulin, and FFA levels in 12 NIDDM patients after isocaloric diets taken as six meals (●) or two meals (○). Values are means ± SE.*

meals were served at 0800 and 240 min later. All meals were offered with liberal amounts of tea, coffee, or water. In both studies, blood glucose was reset after 8 h by 15 g of carbohydrate consumed as five biscuits.

To determine the K_g (5), an IVGTT was conducted 60 min later, with 10 g of glucose given as 50 ml (20%) of glucose over a 2-min interval.

Blood samples were collected through an i.v. catheter placed in a cubital vein. Before the IVGTT, an i.v. device was placed in the opposite arm. Samples for blood glucose analysis were collected every 30 min from 0730 until 1700. Serum for analysis of insulin and FFA was obtained at 0730, then hourly from 0800 to 1700. From the start of the IVGTT, samples for blood glucose measurement were obtained every 5 min, and serum for insulin measurements was obtained every 10 min for 60 min. The samples for glucose, insulin, and FFA were stored at −20°C until assayed. Urine was collected from 0800 to 1700.

Meals

On both occasions, subjects received a 3394 kJ diet comprising 200 g white bread, 15 g margarine, 30 g 30% cheese, 15 g filet, 1 egg, 1 tomato, and slices of cucumber. The diet composition was 28% fat, 18% protein, and 54% carbohydrate, according to Helms (7). The food was prepared by the dietary department at Horsens hospital.

Analytical methods

Plasma and urinary glucose levels were measured by a glucose oxidase method. Serum insulin levels were determined by a specific radioimmunossay (8). HbA_{1c} was measured by a commercial kit (Bio-Rad, Richmond, CA) (normal 3.5–5.5%). FFAs were determined by a standard enzymatic colorimetric assay method by using a commercial kit (Boehringer-Mannheim, Mannheim, Germany).

Statistical methods

FBG and insulin were expressed as the incremental area above fasting level (9). The fasting level was defined as the mean level between 0730 and 0800. Average FFA levels were calculated as the mean of the values obtained hourly during the observation periods. K_g was defined as the slope of the line calculated by least-squares linear regression with natural logarithm (ln) of the absolute blood glucose value (G, mmol) for a 15–35-min period against the time in min, and was expressed as a percentage. Results are expressed as means ± SE. Statistical analysis between FFA levels were made by ANOVA. The two-tailed Wilcoxon's rank-sum test for paired data was used for statistical analysis of the other data, and $P < 0.05$ was considered statistically significant.

RESULTS

All subjects participated in studies A and B. They adhered well to the timing, and consumed the meals completely. The K_g test was missed in 3 subjects because of problems with the i.v catheters. FBG, FFA, and insulin levels were not significantly different in the two studies.

The FBG values were similar before the meals (Table 1). Blood glucose, insulin, and FFA levels for the two diets are given in Fig. 1. During the first 240 min, the incremental blood glucose area was lower in study A, in response to frequent small meals, than in study B (328 ± 68 vs. 476 ± 70 mM × 240 min, $P = 0.02$). However, during the entire period, no difference in the blood glucose responses was observed between study A and study B (630 ± 190 vs. 745 ± 134 mM × 480 min, $P = 0.12$). The amplitude of glucose excursions in study B was 84% larger than in study A (6.1 ± 1.9 vs. 3.3 ± 1.7 mM, $P < 0.005$). The K_g was similar in study A and study B (0.32 ± 0.13 vs. 0.24 ± 0.16, $P = 0.37$). Mean insulin level in response to an IVGTT in study A and study B were similar (227 ± 29 vs. 234 ± 25 pM). Urinary glucose excretion was detected in only 5 subjects after

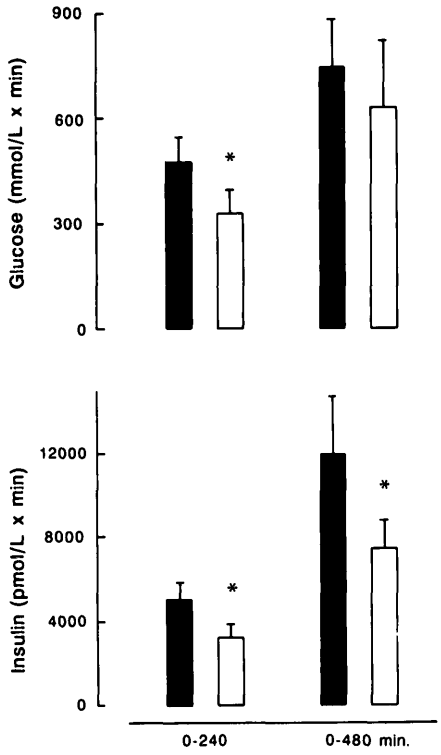

Figure 2—*Mean blood glucose and serum insulin incremental areas over 240 and 480 min after isocaloric diets taken as six meals (□) and two meals (■) in 12 NIDDM subjects. *P < 0.05.*

study A, compared with 9 in study B, with similar losses in study A and study B (1.0 ± 0.6 vs. 2.2 ± 1.0 g). The incremental insulin response was lowest to six meals in study A compared with study B (7,426 ± 1,351 vs. 11,921 ± 2,762 pM × 480 min, $P < 0.03$) (Fig. 2). Also, FFA levels remained low in response to the frequent meals, while FFA levels rose from time 180–300 min in response to few large meals. The FFA levels were lowest in study A compared with study B (0.39 ± 0.01 vs. 0.39 ± 0.01 mM, $P < 0.02$).

CONCLUSIONS

Various national diabetes associations have debated the importance of the temporal distribution of ingested carbohydrates. Thus, the Canadian Diabetes Association recommends a few large meals (10), whereas the British Diabetes Association (11) holds up frequent meals as a model. We found that the postprandial blood glucose fluctuations, insulin, and FFA levels were reduced by increasing the meal frequency in NIDDM subjects. Higher insulin levels after large meals may be attributable to either an enhanced insulin secretion, a decreasing fractional clearance of insulin, or a combination of both. In our study, however, the blood glucose response areas after an 8-h observation period and the K_g response to an IVGTT were unaffected by meal frequency. Because glucose levels were similar, but insulin levels were lower with frequent meals, one might expect this to result from a difference in insulin sensitivity, i.e., that the K_g would have been augmented in response to frequent meals. The reason why we did not find this is not known. It might have been too subtle for the IVGTT (K_g) technique to detect.

Similar results to oral glucose have been found in normal subjects (5), where bolus or continuous sipping resulted in identical 4-h blood glucose areas but reduced insulin and FFA levels. However, the K_g response to an IVGTT was increased after sipping compared with bolus in normal subjects, associated by a more rapid decline in blood glucose (5). The discrepancy in K_g of the two studies might be explained by a difference in the duration of the fasting period before the IVGTT in normal subjects (5). We avoided this in our study by means of a snack served 1 h before the IVGTT.

In a previous study of 6 NIDDM patients, Beebe et al. (6) found that serum insulin and insulin secretion rates were higher for three meals of equal energy distribution (30, 40, and 30%) than for an isocaloric pattern of three meals plus three snacks, whereas three meals of graded energy distribution (10, 20, and 70%) resulted in lower levels. In contrast, the incremental blood glucose response areas were similar after these diets. The results of Beebe et al. (6) are not comparable with ours because they measured diurnal variations versus our 8-h measurements. Recently, Jenkins et al. (12) showed that a diet of 13 snacks (nibbling diet) causes lower blood glucose, serum insulin, and C-peptide levels than a 3-meal diet in NIDDM subjects. The reason for the divergence in results, i.e., that we did not pick up any change in average blood glucose levels, may be because of the lower energy intake in our study.

Note that ingestion of a few large meals elicits higher FFA levels than smaller, frequent meals because an elevation in the plasma FFA concentration reduces insulin-mediated glucose disposal in muscle, stimulates gluconeogenesis, and increases hepatic glucose output that results in hyperglycemic NIDDM (13). Portioning the caloric intake seems, consequently, to have a beneficial effect on the lipid and, thereby, the carbohydrate metabolism. In contrast, Jenkins et al. (12) found similar FFA concentrations on the nibbling diet and 3-meal diet in NIDDM.

The clinical importance of our results, obtained in acute experiments in NIDDM subjects, is highlighted by the longterm study of Jenkins et al. (4), in healthy volunteers, that showed a 25% reduction in daytime insulin levels after many small meals compared with a few large meals. The frequent meals were associated with a fall in low-density lipoprotein cholesterol in normal subjects (4,14,15). The metabolic benefits of increasing the number of meals included economy in insulin secretion, improved insulin sensitivity, and reduced levels of contraregulatory hormones. Low insulin levels are desirable in recognition of the probable atherogenic potential and the attendant risk of cardiovascular disease and hypertension secondary to a high insulin level (16,17).

In conclusion, consuming frequent meals acutely reduces blood glucose fluctuations, and lowers average insulin and FFA levels in NIDDM subjects. Consequently, our results suggest that

increased partitioning of caloric intake may be beneficial in older NIDDM subjects, and we call for long-term experiments.

Acknowledgments— This work was supported by grants from Diabetesforeningen, Fonden til lægevidenskabens fremme i Vejle amt.

We thank the patient volunteers whose collaboration made this study possible and Eva Seier for expert technical assistance.

References

1. Fabry P, Hejl Z, Braun T, Zvolankova K: The frequency of meals, its relation to overweight, hypercholesterolemia, and decreased glucose tolerance. *Lancet* 2:614–18, 1964
2. Metzner HL, Lampiear DE, Wheeler NC, Larkin FH: The relationship between frequency of eating and adiposity in adult men and women in the Tecumseh Community Health Study. *Am J Clin Nutr* 30:712–15, 1977
3. Gwinup G, Byron RC, Roush WH, Kruger FA, Hamwi GJ: Effect of nibbling versus gorging on glucose tolerance. *Lancet* 2:165–67, 1963
4. Jenkins DJA, Wolever TMS, Vuksan V, Brighenti F, Cunnane SC, Venketeshwer R, Jenkins AL, Buckly G, Patten R, Singer W, Corey P, Josse RG: Nibbling versus gorging: metabolic advantages of increased meal frequency. *N Engl J Med* 321:929–34, 1989
5. Jenkins DJA, Wolever TMS, Ocana AM, Vuksan V, Cunnane SC, Jenkins M, Wong S, Singer W, Bloom SR, Blendis M and Josse R: Metabolic effects of reducing rate of glucose ingestion by simple bolus versus continuous sipping. *Diabetes* 39: 775–81, 1990
6. Beebe CA, Cauter EV, Shapiro ET, Tillil H, Lyons R, Rubenstein AH, Polonsky KS: Effect of temporal distribution of calories on diurnal pattern of glucose levels and insulin secretion in NIDDM. *Diabetes Care* 13:748–55, 1990
7. Helms P: *Kostvurderingstabeller*. Copenhagen, Akademisk Forlag, 1980
8. Ørskov H, Thomsen HG, Yde H: Wick chromatography for rapid and reliable immunoassay of insulin, glucagon, and growth hormone. *Nature (Lond)* 219: 193–95, 1968
9. Jenkins DJA, Wolever TMS, Tailor RH, Barker H, Fielden H, Baldwin JM, Bowling AC, Newmann HC, Jenkins AL, Goff DV: Glycemic index of foods: a physiological basis for carbohydrate exchange. *Am J Clin Nutr* 34:362–66, 1984
10. Special Report Committee of the Canadian Diabetes Association: 1980 guidelines for the nutritional management of diabetes mellitus: a special report from the Canadian Diabetes Association. *J Can Diet Assoc* 42:110–18, 1981
11. The Nutrition Subcommittee of the British Diabetes Association's Medical Advisory Committee: Dietary recommendations for diabetics for the 1980's. *Hum Nutr Appl Nutr* 36A:378–94, 1982
12. Jenkins DJA, Ocana A, Jenkins AL, Wolever TMS, Vuksan V, Katzman L, Hollands M, Greenberg G, Corey P, Patten R, Wong G, Josse RG: Metabolic advantages of spreading the nutrinet load: effects of increased meal frequency in non-insulin-dependent diabetes. *Am J Clin Nutr* 55:461–67, 1992
13. DeFronzo RA, Bonadonna RC, Ferrannini E: Pathogenesis of NIDDM: a balanced overview. *Diabetes Care* 15:318–68, 1991
14. Gwinup G, Byron RC, Roush WH, Kruger FA, Hamwi GJ: Effect of nibbling versus gorging on serum lipids in man. *Am J Clin Nutr* 13:209–13, 1963
15. Irwing MI, Freely R: Frequency and size of meals and serum lipids, nitrogen and mineral retention, fat digestibility, and urinary thiamine and riboflavin in young women. *Am J Clin Nutr* 20:816–24, 1967
16. Fabry P, Tepperman J: Meal frequency: a possible factor in human pathology. *Am J Clin Nutr* 23:1059–68, 1970
17. Ducimetiere P, Eschwege E, Papoz L, Richard JL, Claude JR, Rosselin G: Relationship of plasma insulin levels to the incidence of myocardial infarction and coronary heart disease mortality in a middle-aged population. *Diabetologia* 19:205–10, 1980