Glutamine and Alanine Metabolism in NIDDM

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Gluconeogenesis is increased in NIDDM. We therefore examined the metabolism of glutamine and alanine, the most important gluconeogenic amino acids, in 14 postabsorptive NIDDM subjects and 18 nondiabetic volunteers using a combination of isotopic ([6-^{3}H]glucose (20 μCi, 0.2 μCi/min), [U-^{14}C]glutamine (20 μCi, 0.2 μCi/min), [3-^{13}C]alanine (99% ^{13}C, 2 mmol, 20 μmol/min), [ring-^{2}H$_5$]phenylalanine (99% ^{2}H, 2 μmol/kg, 0.03 μmol · kg^{-1} · min^{-1}), and limb balance techniques. Alanine turnover (4.54 ± 0.24 vs. 5.64 ± 0.33 μmol · kg^{-1} · min^{-1}), de novo synthesis (3.00 ± 0.25 vs. 4.01 ± 0.33 μmol · kg^{-1} · min^{-1}), and conversion to glucose (1.02 ± 0.09 vs. 1.56 ± 0.17 μmol · kg^{-1} · min^{-1}) were increased in NIDDM subjects (all $P < 0.01$), while its forearm release (0.45 ± 0.04 vs. 0.39 ± 0.04 μmol · kg^{-1} · min^{-1}) was unaltered. Although glutamine turnover (4.81 ± 0.23 vs. 4.40 ± 0.31 μmol · kg^{-1} · min^{-1}) was unaltered in NIDDM, its conversion to glucose (0.57 ± 0.04 vs. 1.08 ± 0.10 μmol · kg^{-1} · min^{-1}) and to alanine (0.10 ± 0.01 vs. 0.34 ± 0.04 μmol · kg^{-1} · min^{-1}) (both $P = 0.001$) was increased while its oxidation (2.84 ± 0.27 vs. 1.84 ± 0.15 μmol · kg^{-1} · min^{-1}, $P = 0.03$) and forearm release (0.77 ± 0.05 vs. 0.62 ± 0.09 μmol · kg^{-1} · min^{-1}, $P < 0.008$) were both reduced. Our results thus demonstrate that there are substantial alterations of glutamine and alanine metabolism in NIDDM. Conversion of both amino acids to glucose and the proportion of their turnover used for gluconeogenesis are increased; release of both amino acids from tissues other than skeletal muscle seems to be increased. Finally, the reduction in glutamine oxidation, possibly the result of competition with glucose and free fatty acids as fuels, makes more glutamine available for gluconeogenesis without a change in its turnover. *Diabetes* 45:863–868, 1996

In patients with NIDDM, there is increased release of glucose into the circulation (1,2). Since loss of glucose carbon to CO_2, i.e., glucose oxidation, is generally normal in NIDDM (3) while there is increased loss of glucose carbon due to glucosuria, new carbons (i.e., carbons other than those originating from plasma glucose and recycling back to it) must be continuously added to the glucose pool to maintain fasting hyperglycemia. There are four main sources for this new carbon: hepatic glycogen, lactate derived from glycogen, glycerol derived from triglycerides, and amino acids derived from proteins.

Recent studies suggest that glutamine is the most important amino acid gluconeogenic precursor in humans for adding new carbon to the glucose pool (4,5). In animals with experimental diabetes, increased glutamine gluconeogenesis has been found (6). It is likely therefore that alterations in glutamine metabolism may be important in providing carbon to maintain fasting hyperglycemia in NIDDM.

The present studies were therefore undertaken to determine whether glutamine metabolism was altered in NIDDM and to compare its contribution with the increased gluconeogenesis found in this condition (7–10) with that of alanine, another important amino acid gluconeogenic precursor.

RESEARCH DESIGN AND METHODS

Subjects. Informed written consent was obtained from 14 NIDDM subjects and 18 volunteers with normal glucose tolerance tests and no family history of diabetes. Their characteristics are shown in Table 1. Data from six of the nondiabetic subjects have been previously reported (4). All had been on a weight-maintaining diet containing at least 200 g carbohydrate and had abstained from alcohol for at least 3 days before the study. Sulfonylurea medication had been withdrawn from diabetic patients 3 days before the study. The protocol was approved by the local institutional review board.

Protocol. Subjects were admitted to the General Clinical Research Unit between 4:00 and 6:00 P.M. in the evening before experiments; they consumed a standard meal (10 kcal/kg, 50% carbohydrate, 35% fat, and 15% protein) between 6:00 and 8:00 P.M. and fasted overnight until experiments were completed.

At approximately 4:00 A.M., an antecubital vein was cannulated, and primed-continuous infusions of [6-^{3}H]glucose (20 μCi, 0.2 μCi/min), [U-^{14}C]glutamine (20 μCi, 0.2 μCi/min), [3-^{13}C]alanine (99% ^{13}C, 2 mmol, 20 μmol/min), and [ring-^{2}H$_5$]phenylalanine (99% ^{2}H, 2 μmol/kg, 0.03 μmol · kg^{-1} · min^{-1}) were begun. At the same time, a bolus of 50 μCi of [^{14}C]sodium bicarbonate was given. At 7:00 A.M., the ipsilateral radial artery (five normal and two NIDDM subjects) or dorsal hand vein (13 normal and 12 NIDDM subjects) and a contralateral deep antecubital vein were cannulated for blood sampling. When the dorsal hand vein was used to obtain arterialized venous blood, the hand was maintained in a thermoregulated Plexiglas box at 65°C. Several previous studies have found no difference between simultaneously sampled arterial and arterialized venous glutamine and alanine concentrations (11,12). After these findings were confirmed in the initial studies, arterialized venous sampling was used in place of arterial sampling, and for simplicity, all data are referred to as arterial.

After allowing ~4 h to achieve isotopic steady state, four blood samples were collected simultaneously from the radial artery (or dorsal hand vein) and the deep antecubital vein at 20-min intervals for determination of plasma substrate and hormone concentrations, specific activities, and stable isotope enrichments. During this interval, total CO_2 production was measured twice using a metabolic cart (Sensormedics, Anaheim, CA). Before and after these measurements, breath samples were collected by directing expiratory air through a solution of 0.5 mol/l hyamine hydroxide in methanol (VWR, Cerritos, CA).

Analytical procedures. All blood samples were immediately placed in a 4°C ice bath, and plasma was separated within 30 min by centrifuga-

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FFA, free fatty acid; STZ, streptozocin.

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TABLE 1
Patient characteristics

	Control subjects	NIDDM patients	*P*
n	18	14	
Sex (M/F)	9/9	9/5	
Age (years)	48.4 ± 1.7	54.8 ± 2.3	NS
BMI (kg/m^2)	27.7 ± 1.0	28.1 ± 1.0	NS
Body weight (kg)	81.5 ± 2.8	87.7 ± 3.1	NS
Forearm blood flow ($ml \cdot 100\ ml^{-1} \cdot min^{-1}$)	3.1 ± 0.2	2.7 ± 0.1	NS
HbA_{1c}	6.1 ± 0.1	10.0 ± 0.6	<0.001

Data are means ± SE. Data from six of the control subjects have been previously reported (4).

tion at 4°C. For subsequent measurement of glutamine, glutamate, and alanine concentrations, an internal standard (25 nmol of *p*-fluorophenylalanine) was added to 3 ml plasma. The plasma was deproteinized with 3 ml ice-cold perchloric acid (7% wt/vol). After centrifugation, the supernatant was processed according to the method of Smith and Panico (13), by adding 0.6 ml 4 mmol/l sodium acetate buffer, pH 4.8, and adjusting the pH between 4.8 and 5.0 with 5 N potassium hydroxide and were frozen at −20°C until the time of analysis.

Plasma glucose was determined with a glucose analyzer (YSI, Yellow Springs, OH). Plasma glutamine, glutamate, and alanine concentrations and the specific activities of glutamine and alanine were determined by high-performance liquid chromatography using a modification (14) of the method of Smith and Panico (13). Plasma [^{3}H]- and [^{14}C]glucose specific activities were determined using ion-exchange chromatography (15). Plasma [^{13}C]glucose and alanine enrichment and plasma [^{2}H]phenylalanine enrichments were measured by chemical ionization and selected ion monitoring gas chromatography-mass spectrometry of penta-acetate (glucose) and *N*-acetyl-*N*-propyl ester derivatives (alanine and phenylalanine), as previously described (15,16). Plasma free fatty acid (FFA) concentrations were determined by an enzymatic method (17). Glucagon, C-peptide, and insulin concentrations were determined using standard radioimmunoassays (18).

Calculations. At steady state, the rate of appearance (R_a) of a substrate in plasma equals its rate of disappearance (R_d) from plasma (19), which is collectively referred to as turnover. Plasma glucose and glutamine turnover were calculated by dividing the [6-^{3}H]glucose and [^{14}C]glutamine infusion rates (disintegrations per minute per kilogram per minute) by the arterial plasma [6-^{3}H]glucose and [^{14}C]glutamine specific activities (disintegrations per micromole), respectively (19). Plasma alanine and phenylalanine turnover were calculated according to the following equation (19): $R_a = I(E_i/E_p - 1)$, where I is the tracer infusion rate (micromoles per kilogram per minute), E_i is the enrichment of the infused tracer (mole percent excess), and E_p is the arterial isotopic enrichment of plasma tracer (mole percent excess). The systemic clearances of glutamine and alanine were calculated by dividing their turnover by their arterial plasma concentration.

The percentage of plasma glucose and alanine derived from plasma glutamine and the percentage of plasma glucose derived from plasma alanine were calculated, respectively, as:

$$\frac{[^{14}C]\ \text{alanine SA} \times 100 \times 5}{[^{14}C]\ \text{glutamine SA} \times 3},$$

$$\frac{[^{14}C]\ \text{glucose SA} \times 100 \times 5}{[^{14}C]\ \text{glutamine SA} \times 6}, \text{ and}$$

$$\frac{[^{13}C]\ \text{glucose enrichment} \times 100}{[^{13}C]\ \text{plasma alanine enrichment} \times 2}$$

The rates at which plasma glucose was derived from carbons of glutamine and alanine were calculated as the product of the rate of appearance in plasma of glucose and the percentage of its derivation from these precursors. It is appreciated that the calculated amounts of glucose derived from alanine and glutamine represent minimal estimates because of carbon exchange in the Krebs cycle (15,20,21).

The rate at which plasma glucose was derived from plasma alanine was corrected for the alanine derived from glutamine by multiplying the uncorrected rate of glucose derived from alanine by (100 − the percentage of alanine derived from glutamine).

The rate of appearance in plasma of glutamine and alanine due to their release from protein was calculated by a modification of the approach described by Darmaun et al. (16). This approach is based on the concept that since phenylalanine is an essential amino acid that cannot be synthesized in vivo, its only source in the postabsorptive state is its release from protein. Thus, the rate of appearance of phenylalanine in plasma can be used as an index of proteolysis and release of glutamine and alanine directly from protein can be estimated using the rate of phenylalanine appearance in plasma and the relative contents of phenylalanine, glutamine, and alanine in body protein with the equation $R_{a\ p}(k_{NE}/k_p)$, where $R_{a\ p}$ is the phenylalanine appearance rate, k_{NE} is the proportion of amino acids in protein accounted for by either alanine or glutamine, and k_p is the proportion of amino acids accounted for by phenylalanine. Values of 3.4, 8.2, and 4.1% were used for the proportions of phenylalanine, alanine, and glutamine, respectively, based on the average proportions found in eight tissue and circulating proteins [myosin light chain 1, myosin light chain 3, myosin heavy chain, actin, collagen α_2(IV), fibrinogen α_1 and α_2, and albumin] as determined by their gene sequence obtained from the SWISS-PROT 13 data bank (22).

Plasma glutamine oxidation (micromoles per kilogram per minute), as represented by the incorporation of ^{14}C into breath CO_2, was calculated as:

$$\frac{[^{14}CO_2]\ \text{SA} \times CO_2\ \text{production}}{[^{14}C]\text{glutamine SA} \times 0.7}$$

where CO_2 production is in micromoles per kilogram per minute and 0.7 is used to correct for incomplete recovery of CO_2 in breath (23). It is recognized that this represents production of labeled CO_2 from labeled glutamine carbon and not necessarily oxidation of the glutamine molecule.

Net forearm balance of glucose, glutamine, and alanine was calculated by multiplying arteriovenous differences in substrate concentrations by forearm blood flow. Forearm fractional extraction of [^{14}C]glutamine was calculated as the arteriovenous difference in [^{14}C]glutamine concentration divided by the arterial [^{14}C]glutamine concentration. Forearm alanine fractional extraction was calculated in an analogous manner using [^{13}C]alanine concentrations. Forearm glutamine and alanine uptake was calculated as the product of their arterial concentration, blood flow, and fractional extraction (15). Forearm release was calculated according to the equation: release = uptake − net balance (15).

Data are given as means ± SE. Data of control subjects and of NIDDM subjects were compared with the Mann-Whitney *U* test using the mean of four basal time points (24). $P < 0.05$ was considered statistically significant.

RESULTS

Plasma substrate and hormone concentrations. Fasting plasma glucose was 11.0 ± 0.8 mmol/l in NIDDM subjects and 5.2 ± 0.1 mmol/l in normal volunteers ($P < 0.001$) (Table 2). Plasma glutamine, glutamate, alanine, FFA, insulin, and C-peptide concentrations were not significantly different in the two groups. Plasma glucagon, however, was greater in diabetic patients (92 ± 6 vs. 126 ± 9 ng/l, $P = 0.006$).

Glutamine, alanine, and glucose metabolism. Plasma arterial substrate concentrations, specific activities, enrichments, and breath $^{14}CO_2$ specific activities were stable during the sampling period, indicating that an isotopic steady state

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TABLE 2
Arterial plasma glutamine, glutamate, glucose, alanine, FFA, insulin, C-peptide, and glucagon concentrations

	Control subjects	NIDDM subjects	*P* value
Plasma glutamine (mmol/l)	0.61 ± 0.03	0.58 ± 0.03	0.59
Plasma glutamate (mmol/l)	0.066 ± 0.004	0.077 ± 0.006	0.52
Plasma glucose (mmol/l)	5.2 ± 0.1	11.0 ± 0.8	<0.001
Plasma alanine (mmol/l)	0.33 ± 0.02	0.37 ± 0.02	0.16
Plasma FFA (μmol/l)	707 ± 46	751 ± 38	0.27
Plasma insulin (pmol/l)	60 ± 8	72 ± 13	0.28
Plasma C-peptide (nmol/l)	0.42 ± 0.04	0.52 ± 0.05	0.10
Plasma glucagon (ng/l)	92 ± 6	126 ± 9	0.006

Data are means ± SE.

had been reached (Tables 3 and 4). The rate of plasma glutamine appearance was similar in normal volunteers and NIDDM subjects, but that of alanine was about 25% greater in NIDDM subjects. The rate of appearance of the essential amino acid phenylalanine was not significantly different in NIDDM and normal subjects (0.64 ± 0.03 and 0.68 ± 0.05 $\mu mol \cdot kg^{-1} \cdot min^{-1}$, respectively, $P = 0.53$). These data were used to calculate de novo synthesis of alanine and glutamine. De novo alanine synthesis was increased ~33% in NIDDM subjects, whereas that of glutamine was not significantly different in the two groups. The release of both amino acids from protein was similar. Glucose turnover was significantly greater in NIDDM subjects (10.9 ± 0.4 vs. 20.1 ± 1.3 $\mu mol \cdot kg^{-1} \cdot min^{-1}$, $P < 0.001$). Moreover, glucose produced from both amino acids was significantly greater in NIDDM subjects (0.48 ± 0.04 vs. 0.90 ± 0.08 $\mu mol \cdot kg^{-1} \cdot min^{-1}$, $P = 0.001$, for glutamine and 0.50 ± 0.04 vs. 0.78 ± 0.08 $\mu mol \cdot kg^{-1} \cdot min^{-1}$, $P = 0.008$, for alanine).

Regarding the fate of these amino acids, the rate of conversion to glucose of glutamine and alanine was 2- and 1.5-fold greater in NIDDM subjects. The rate of conversion of glutamine to alanine was three times higher in NIDDM subjects, whereas the rate of glutamine oxidation was significantly decreased. Thus, in normal subjects, glutamine conversion to glucose and glutamine oxidation accounted for 11.7 ± 0.6 and 57.3 ± 2.7% of glutamine turnover, respectively, and for 24.7 ± 1.9 and 44.6 ± 4.9%, respectively, in diabetic subjects.

Forearm glutamine, alanine, and glucose metabolism. Forearm fractional extraction of glutamine but not of alanine was significantly lower in NIDDM subjects (Table 5). There was no difference in forearm uptake of these amino acids between the two groups. Forearm glutamine net balance and glutamine release were both significantly decreased in NIDDM subjects. Forearm alanine net balance, alanine release, and forearm glucose net balance were not significantly different in the two groups.

DISCUSSION

The present studies were undertaken to determine whether glutamine metabolism was altered in NIDDM and to compare its contribution with the increased gluconeogenesis found in this condition with that of alanine. Previous studies have shown that glutamine concentration (25–27), its turnover (28), and net splanchnic extraction (27) are normal in

TABLE 3
Specific activities and enrichments of metabolic substrates in normal and NIDDM subjects

	0 min	20 min	40 min	60 min
[^{3}H]Glucose specific activity (dpm/μmol)				
Normal subjects	449 ± 46	438 ± 48	443 ± 50	444 ± 47
NIDDM subjects	162 ± 13	155 ± 13	160 ± 13	165 ± 12
[^{14}C]Glucose specific activity				
Normal subjects	62 ± 4	60 ± 4	64 ± 4	62 ± 4
NIDDM subjects	56 ± 5	56 ± 4	57 ± 4	58 ± 5
[^{13}C]Glucose enrichment				
Normal subjects	0.58 ± 0.06	0.51 ± 0.06	0.51 ± 0.06	0.52 ± 0.06
NIDDM subjects	0.38 ± 0.03	0.37 ± 0.04	0.40 ± 0.05	0.37 ± 0.05
[^{14}C]Glutamine specific activity				
Normal subjects	1,239 ± 78	1,230 ± 76	1,239 ± 78	1,235 ± 78
NIDDM subjects	1,098 ± 57	1,067 ± 46	1,063 ± 48	1,075 ± 44
[^{13}C]Alanine enrichment				
Normal subjects	4.7 ± 0.4	4.8 ± 0.3	4.6 ± 0.3	4.7 ± 0.4
NIDDM subjects	3.9 ± 0.4	4.1 ± 0.5	3.9 ± 0.4	4.2 ± 0.5
[^{14}C]Alanine specific activity (dpm/μmol)				
Normal subjects	36 ± 9	35 ± 5	36 ± 6	37 ± 10
NIDDM subjects	74 ± 9	73 ± 13	76 ± 16	74 ± 10
[^{2}H]Phenylalanine enrichment				
Normal subjects	5.7 ± 0.1	5.7 ± 0.7	5.7 ± 0.7	5.9 ± 0.7
NIDDM subjects	4.8 ± 0.3	4.9 ± 0.3	4.9 ± 0.2	4.8 ± 0.3
$^{14}CO_2$ Specific activity				
Normal subjects	—	20.9 ± 0.8	—	20.8 ± 0.9
NIDDM subjects	—	11.1 ± 1.1	—	11.7 ± 1.1

Data are means ± SE.

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TABLE 4
Glutamine and alanine metabolism

	Glutamine	Alanine
Overall turnover (μmol · kg^{-1} · min^{-1})		
Control subjects	4.81 ± 0.23	4.54 ± 0.24
NIDDM subjects	4.40 ± 0.31	5.64 ± 0.33
P value	0.41	0.02
De novo synthesis (μmol · kg^{-1} · min^{-1})		
Control subjects	4.04 ± 0.22	3.00 ± 0.25
NIDDM subjects	3.59 ± 0.29	4.01 ± 0.33
P value	0.25	0.03
Formation from protein (μmol · kg^{-1} · min^{-1})		
Control subjects	0.77 ± 0.04	1.54 ± 0.07
NIDDM subjects	0.82 ± 0.06	1.63 ± 0.11
P value	0.53	0.45
Conversion to glucose (μmol · kg^{-1} · min^{-1})		
Control subjects	0.57 ± 0.04	1.02 ± 0.09
NIDDM subjects	1.08 ± 0.10	1.56 ± 0.17
P value	0.001	0.008
Conversion to alanine (μmol · kg^{-1} · min^{-1})		
Control subjects	0.10 ± 0.01	—
NIDDM subjects	0.34 ± 0.04	—
P value	0.001	—
Formation from glutamine (μmol · kg^{-1} · min^{-1})		
Control subjects	—	0.17 ± 0.02
NIDDM subjects	—	0.57 ± 0.07
P value	—	0.001
Conversion to CO_2 (μmol · kg^{-1} · min^{-1})		
Control subjects	2.84 ± 0.27	—
NIDDM subjects	1.84 ± 0.15	—
P value	0.03	—

Data are means ± SE.

patients with IDDM. Comparable plasma glutamine concentrations have been found previously in NIDDM subjects and normal volunteers (29). Although we found the plasma glutamine concentration and turnover to be normal in our NIDDM subjects, the conversion of glutamine to glucose and alanine was significantly increased.

The lack of change in glutamine turnover despite its increased conversion to glucose and alanine could be explained by the decrease in glutamine oxidation observed in the NIDDM subjects. The mechanism for reduction in glutamine oxidation remains to be explained but may reflect altered fuel homeostasis in NIDDM. Although the increase in plasma FFA concentrations in NIDDM subjects was not statistically significant in the present study, increased plasma FFA levels are usually associated with this condition (30). It is possible that FFAs and glucose, in view of the prevailing hyperglycemia, could have substituted for glutamine as an oxidative fuel in certain tissues. Octanoate and ketone bodies have been shown to reduce glutamine utilization by rat enterocytes (31), and glutamine uptake by the small intestine is reduced in streptozocin (STZ)-induced diabetic rats (6,31). This tissue normally uses glutamine as its major oxidative fuel (32).

We also found that forearm tissues released less glutamine in our NIDDM subjects. This finding, in the face of an unchanged overall appearance of glutamine in plasma, suggests that in NIDDM sources other than skeletal muscle release increased amounts of glutamine. It is of interest,

TABLE 5
Forearm glutamine, alanine, and glucose metabolism

	Glutamine	Alanine	Glucose
Fractional extraction (%)			
Control subjects	23.7 ± 1.5	29.8 ± 2.7	—
NIDDM subjects	20.8 ± 2.3	27.0 ± 4.0	—
P value	0.017	0.16	—
Net balance (μmol · kg^{-1} · min^{-1})			
Control subjects	−0.33 ± 0.03	−0.15 ± 0.02	0.76 ± 0.08
NIDDM subjects	−0.19 ± 0.04	−0.14 ± 0.02	0.76 ± 0.13
P value	0.02	0.78	0.99
Uptake (μmol · 100 ml^{-1} · min^{-1})			
Control subjects	0.44 ± 0.04	0.29 ± 0.03	—
NIDDM subjects	0.43 ± 0.08	0.26 ± 0.04	—
P value	0.92	0.43	—
Release (μmol · 100 ml^{-1} · min^{-1})			
Control subjects	0.77 ± 0.05	0.45 ± 0.04	—
NIDDM subjects	0.62 ± 0.09	0.39 ± 0.04	—
P value	0.008	0.35	—

Data are means ± SE.

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therefore, that human adipose tissue has been demonstrated to release glutamine (33). Conceivably, there could be increased glucose-glutamine cycling (5) in this and other tissues due to hyperglycemia.

In contrast with glutamine, alanine turnover and conversion to glucose were both increased in the NIDDM subjects, as has been previously reported (10). The increase in alanine turnover was wholly due to its observed greater de novo synthesis since its release into plasma as a direct result of proteolysis was normal. Increased alanine de novo synthesis could only be explained partially by increased conversion of glutamine to alanine. Another possible explanation is increased formation from plasma glucose, since there is evidence for increased carbon cycling in diabetes (34,35). Our finding of unaltered alanine release from forearm tissues confirms a previous report (10) and suggests that, like glutamine, there was increased release of alanine from tissues other than skeletal muscle.

In the present study, glucose formed from glutamine exceeded that formed from alanine (0.90 ± 0.05 vs. 0.78 ± 0.08 $\mu mol \cdot kg^{-1} \cdot min^{-1}$). However, net glutamine uptake by the human liver is small relative to that of alanine (36,37). Reported values of net hepatic glutamine uptake in normal volunteers (30 $\mu mol/min^{-1}$) (37) could not account for the glutamine conversion to glucose observed in either our normal volunteers (46 $\mu mol/min^{-1}$) or our NIDDM subjects (95 $\mu mol/min^{-1}$). In contrast, reported values for net hepatic alanine uptake (102 $\mu mol/min^{-1}$)(37) could account for alanine conversion to glucose in our normal volunteers (83 $\mu mol/min^{-1}$). Since splanchnic alanine uptake is increased in NIDDM (38), hepatic alanine uptake could probably also account for alanine conversion to glucose in our NIDDM subjects (137 $\mu mol/min^{-1}$).

Since net splanchnic glutamine extraction has been reported not to be altered in human diabetes (27), we are in a quandary to explain the increased glutamine gluconeogenesis in our NIDDM subjects. One possibility is that incorporation of label into glucose from labeled glutamine simply represents carbon exchange analogous to the situation in which label from fatty acids ends up in glucose (39). Another possible explanation is that in NIDDM, there may have been increased release or decreased uptake of glutamine by nonhepatic splanchnic tissues. In STZ-induced diabetic rats, there is decreased uptake of glutamine by the small intestine and increased uptake by the kidney (6,31,40).

The latter is of interest in accounting for the increased glutamine conversion to glucose in our NIDDM subjects since glutamine is a major gluconeogenic precursor in the kidney (41). Net uptake of glutamine by the normal human kidney (109 $\mu mol/min^{-1}$) is more than three times that of the liver (37). Moreover, activities of renal gluconeogenic enzymes (42–45) and renal gluconeogenesis are increased in diabetic animals (46–49). It is thus possible that renal gluconeogenesis, which recently has been reported to account for ~25% of systemic glucose appearance in nondiabetic humans (50), may be increased in NIDDM and may provide an explanation for increased glutamine conversion to glucose in this condition.

Forearm glucose uptake was similar in our NIDDM and normal subjects, as has been previously reported (1), whereas overall systemic glucose disposal was increased about twofold. Glycosuria, which was not measured in the present study, would be expected to explain only a small portion of the increased glucose disposal since the renal threshold for glycosuria is ~10 mmol/l and our NIDDM subjects plasma glucose averaged ~11 mmol/l. Thus, in NIDDM, it is likely that there is increased glucose uptake in tissues other than muscle.

In conclusion, the present studies demonstrate that in NIDDM, there are substantial alterations in glutamine and alanine metabolism: conversion of glutamine to glucose is increased (more so than that of alanine). Moreover, conversion of glutamine to alanine is increased while oxidation of glutamine and its release from muscle are decreased. Although plasma alanine turnover is increased, its release from forearm tissues is unaltered. These observations suggest that in NIDDM there is increased release of both these amino acids from tissues other than skeletal muscle. Finally, we suggest that the reduced oxidation of glutamine in NIDDM may be the result of increased use of glucose and FFAs as fuels in certain tissues, and that as a consequence, more glutamine may be made available for gluconeogenesis without a change in its overall turnover.

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