# The Effect of Carbohydrates on Ammonium and Ketoacid Excretion during Starvation

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ABSTRACT The metabolic effects of oral ingestion of minute quantities of carbohydrate during prolonged starvation were studied in nine obese subjects. Measurements were made during a control period of total starvation, during the ingestion of 7.5 g carbohydrate daily, and finally during the ingestion of 15.0 g carbohydrate daily. Daily ketoacid excretion fell after carbohydrate ingestion and was significantly correlated ($r = 0.62$, $P < 0.01$) with the amount of carbohydrate administered. Despite this fall in ketoacids, the concentration of blood ketoacids, plasma free fatty acids, and serum insulin remained constant throughout the study. Urinary ammonium excretion, closely correlated with ketoacid output ($r = 0.95$, $P < 0.001$), also fell significantly after carbohydrate ingestion. No significant changes were present in extracellular or urinary pH. Urea nitrogen excretion did not change when urinary ammonium output fell. These results indicate that: the excretion of ketoacids and ammonium in starving man is exquisitely sensitive to minute amounts of ingested carbohydrate; the change in ketonuria appears to be due to increased renal ketoacid reabsorption after carbohydrate ingestion; and the nitrogen-sparing effect of reducing renal ammonium output in starvation can be dissociated from nitrogen sparing occurring because of changes in urine urea excretion.

## INTRODUCTION

A known consequence of starvation is a fall in blood glucose and serum insulin with a subsequent rise in plasma free fatty acids and blood ketoacids, acetoacetate and beta-hydroxybutyrate. During this state of starvation hyperketonemia, urinary acetoacetate and beta-hydroxybutyrate excretion increase markedly. The loss of these anions in the urine is matched by the loss of ammonium, which contributes significantly to the depletion of body protein stores (1).

Although previous studies have shown that ingestion of carbohydrate reduces both urinary ketoacids and ammonium excretion (2–4) no information is available regarding the dose-response relationship between ingested carbohydrate and the urinary excretion of these substances.

In this study a systematic investigation was undertaken to define the minimal amount of exogenous carbohydrate required to reduce urinary ketoacid and ammonium excretion during starvation. In addition, the effect of the ingested carbohydrate on acid-base parameters, blood glucose, serum insulin, plasma free fatty acids, and blood acetoacetate and beta-hydroxybutyrate concentrations was evaluated.

The results of the study indicate that minute amounts of carbohydrate (7.5 g ingested daily) will significantly alter the excretion of urinary ketoacids and ammonium in man after prolonged starvation.

## METHODS

*Subjects.* Nine obese volunteers were admitted to The Johns Hopkins Hospital Clinical Research Center to undergo therapeutic starvation for weight reduction (Table I). After each was fully informed as to the purposes and risks of undergoing this protocol, consent was obtained. Clinical data from these subjects are given in Table I. Intake during the starvation period consisted of a multivitamin capsule and sufficient distilled water to maintain a daily urine volume of about 800 ml.

*Protocol.* The nine subjects underwent total starvation for 15–24 days, at which time a steady state of ammonium

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TABLE I

*Clinical Data and Protocol*

| Subject | Age | Sex | Weight | Height | Deviation from pop. mean weight* | Periods of study: Pre-CHO starvation | 7.5 g CHO | 15.0 g CHO | Post-CHO starvation | Diagnoses‡ |
|---|---|---|---|---|---|---|---|---|---|---|
| | *yr* | | *kg* | *cm* | % | | | | | |
| D. P. | 18 | F | 100.93 | 167.6 | +66 | X | X | | | Obesity. |
| T. C. | 19 | F | 136.61 | 168.9 | +121 | X | X | X | X | Obesity. |
| S. B. | 23 | F | 113.71 | 166.4 | +87 | X | X | X | X | Obesity. |
| J. T. | 23 | F | 112.52 | 168.9 | +80 | X | X | | | Obesity. |
| L. S. | 28 | F | 118.96 | 181.6 | +61 | X | X | | | Obesity. |
| J. H. | 29 | F | 152.24 | 160.0 | +160 | X | X | | | Obesity. |
| B. H. | 38 | F | 102.81 | 165.1 | +60 | X | X | | | Obesity. Diabetes mellitus. Unilateral nephrectomy 6 yr before study. |
| L. C. | 43 | F | 126.58 | 170.2 | +76 | X | X | X | X | Obesity. Diabetes mellitus. |
| S. T. | 26 | M | 207.27 | 193.0 | +140 | X | | X | | Obesity. Transverse colostomy for diverticulitis, 5 months before study. |

* From Society of Actuaries, Build and Blood Pressure Study, Volume 1, Chicago, 1959.

‡ Oral glucose tolerance test (100 g) was performed before weight reduction in all subjects except L. S., who had a normal fasting glucose level. The diagnosis of diabetes mellitus was based on the criteria of Fajans (29).

excretion was achieved. At the end of this interval oral glucose (J. T., L. S., J. H., B. H., and L. C.), fructose (S. B.), or sucrose (D. P., T. C., and S. T.) was given as a 25% solution in distilled water (see Table I). Eight of the subjects were initially given 2.5 g of carbohydrate (CHO)[1] 3 times daily, whereas the ninth subject (S. T.) received 5 g of CHO 3 times daily. Observations were continued at these levels of CHO ingestion for 7–14 days, during which time a new steady state of ammonium excretion was attained. In three of the eight patients initially given 7.5 g CHO daily the quantity was subsequently increased to 15 g, given as 5.0 g 3 times daily. The duration of this second period of CHO administration was 8–12 days. Subject S. B. was initially given 7.5 g of fructose in divided daily doses. This quantity was subsequently increased to 15 g daily. Fructose was then discontinued, the patient was allowed to return to a new steady state of starvation, and then she was given 15 g of glucose in divided daily doses. In addition to this subject who was studied during a postcarbohydrate control period, similar control observations were carried out in two other subjects (T. C. and L. C.) after stopping CHO supplements and while they continued to fast, to ensure that the results obtained were not time-related.

*Blood and urine collections.* All blood specimens were obtained without the use of a tourniquet from an antecubital vein at approximately 9 a.m. During the carbohydrate ingestion periods blood specimens were obtained 1 hr after the morning CHO supplement. Blood glucose, acetoacetate (AcAc), beta-hydroxybutyrate (β-OHB), plasma free fatty acids (FFA), serum immuno-reactive insulin (IRI), pH, and plasma $CO_2$ content were determined 5, 3, and 1 day before carbohydrate supplementation and daily during the last 5 days of ingestion of 7.5 g CHO. Blood was obtained randomly from two subjects (S. B. and L. C.) during the period of administration of 15.0 g CHO and during the postcarbohydrate control period.

Daily 24 hr urine collections were obtained using mineral oil and thymol-chloroform preservatives, and refrigerated at 4°C during the collection period. On completion of the daily collection urinary pH was measured, and portions were stored at −10°C for subsequent analyses of urea, ammonium, creatinine, AcAc, β-OHB, and total organic acids.

*Chemical analyses.* Plasma $CO_2$ content was determined by the method of Van Slyke and Neill (5) while blood pH was measured anaerobically at 37°C (none of the subject's temperatures varied by more than 1° from 37°C) with a Radiometer (Radiometer A/S, Copenhagen) glass electrode coupled to an Orion (Orion Research, Cambridge, Mass.) pH meter. Plasma bicarbonate concentration and $P_{CO_2}$ were calculated from the Henderson-Hasselbalch equation using a pK′ of 6.1 and a solubility constant of 0.0301. Urinary ammonium was analyzed by the Conway microdiffusion method (6) and creatinine and urea were determined by autoanalyzer techniques (7, 8). Total urinary organic acids were determined by the Van Slyke and Palmer titration method (9) and expressed after correcting for their creatinine content. Blood and urinary AcAc and β-OHB were measured by the method of Williamson and Mellanby (10) employing a phosphate buffer (pH 8.7) in determining β-OHB rather than Tris buffer. Blood glucose was determined by the Technicon (Technicon Instruments Corp., Tarrytown, N. Y.) autoanalyzer glucose oxidase-peroxidase procedure of Hill and Kessler (11) as modified by Steiner, Goodman, and Treble (12). Plasma FFA were determined by the method of Dole and Meinertz (13). Insulin concentrations were determined by the double antibody radioimmunoassay of Mor-

[1] *Abbreviations used in this paper:* AcAc, acetoacetate; CHO, carbohydrate; GFR, glomerular filtration rate; IRI, immuno-reactive insulin; β-OHB, beta-hydroxybutyrate.

gan and Lazarow (14) as modified by Soeldner and Slone (15). Daily glomerular filtration (GFR) rates were measured by endogenous creatinine clearance. All analyses were done in duplicate except those for glucose, which were done in triplicate.

*Data analyses.* For statistical evaluation of the significance of the observed metabolic changes the last 5 days of the precarbohydrate period and the last 5 days of each carbohydrate supplementary period were taken as representing the new steady states for each period, since ammonium excretion remained constant on the day-to-day basis during these intervals. The steady state for each 5 day period was assessed by examining the interday variation in ammonium excretion by the Student $t$ test for paired samples and by the coefficient of correlation (16). Thus all subjects' urinary ammonium excretion on the 1st day of the steady state period were paired for analysis with their values on each subsequent day of this metabolic period. No significant differences were obtained.

Blood and urine data obtained during these 5-day periods were averaged and are expressed as the mean daily blood value or mean daily excretion.

Urinary ammonium, ketoacid, and urea excretion data were corrected for GFR, since the latter is known to decrease during prolonged starvation when supplementary salt is not given (1, 17). The following formula was used:

$$U_{Substance}V/GFR \times 100 = \text{Corrected value.}$$

All values were expressed as the mean ±SEM.

## RESULTS

*Urinary ketoacid excretion.* Total urinary ketoacid excretion, AcAc plus β-OHB, of 122.0±20.3 mmoles/day during the control period fell to 74.7±13.8 ($P < 0.01$) mmoles/day after the ingestion of 7.5 g CHO and 34.3±2.6 ($P < 0.05$) mmoles/day when 15.0 g of CHO (four subjects) was administered daily. The control value for total urinary ketoacid excretion per day of these four subjects was 129.1±23.0 mmoles/day, a figure similar to the control value of the entire group. When calculated as total ketoacid per 100 ml GFR the daily ingestion of 7.5 g of CHO produced a fall in total urinary ketoacid excretion from a control value of 118.1±13.2 mmoles/day to one of 86.9±13.7 mmoles/day ($P < 0.01$, Table II). When the dose of ingested CHO was raised to 15.0 g/day (four subjects) the excretion when compared to the control value fell further to 44.0±8.6 mmoles/day ($P < 0.05$, Table II).

Fig. 1 displays the relationship between the quantity of ingested CHO and ketonuria, the latter corrected for changes in GFR ($r = 0.62$, $P < 0.01$). An analysis of the response patterns of β-OHB excretion also calculated per 100 ml GFR, yields comparable results to the total ketone body excretion data. This could have been anticipated since β-OHB is the major ketone body excreted in the urine (1). The mean control β-OHB excretion was 103.8±9.9, falling significantly to 77.5 ±11.8 mmoles/day ($P < 0.01$) after ingesting 7.5 g of CHO and then to 40.6±6.5 mmoles/day ($P < 0.05$) after ingestion of 15.0 g of CHO (four subjects). The mean control AcAc excretion per 100 ml GFR was 13.2 ±2.2 mmoles/day, falling to 10.4±2.2 mmoles/day ($P < 0.05$) and 7.4[2]±1.6 mmoles/day ($P < 0.05$) with the ingestion of 7.5 and 15.0 g of CHO, respectively.

TABLE II

*Relationship between Daily Ketoacid Excretion Corrected for Changes in GFR and Carbohydrate Ingestion*

| Subject | Precarbohydrate | Carbohydrate | |
|---|---|---|---|
| | | 7.5 g CHO | 15.0 g CHO |
| | *mmoles/100 ml GFR per day* | *mmoles/100 ml GFR per day* | *mmoles/100 ml GFR per day* |
| D. P. | 128.5± 8.7 | 102.3±11.0 (S)* | — |
| T. C. | 117.6± 8.0 | 93.9± 4.9 (S) | 33.5± 3.8 (S) |
| S. B. | 86.3± 7.6 | 62.2± 6.1 (F) | 47.1±11.5 (F)‡ |
| | | | 33.4± 3.5 (G) |
| J. T. | 100.6± 8.9 | 66.4± 8.6 (G) | — |
| L. S. | 143.0±23.6 | 138.8± 8.3 (G) | — |
| J. H. | 124.1± 2.8 | 63.5± 3.8 (G) | — |
| B. H. | 50.4± 2.5 | 29.6± 6.7 (G) | — |
| L. C. | 160.0±16.8 | 138.3± 8.2 (G) | 67.0± 5.0 (G) |
| S. T. | 92.5± 5.1 | — | 28.3± 1.2 (S) |
| Mean ±SEM | 118.1±13.2 | 86.9±13.7§ | 44.0± 8.6‖ |

* F, S, and G denote fructose, sucrose, or glucose, respectively.
‡ This subject was studied with 15 g of fructose and glucose. Only the one value (F) was used to calculate the mean for the entire group.
§ $P < 0.01$ from control.
‖ $P < 0.05$ from control.

To evaluate the effect of administering even greater quantities of CHO on ketonuria subject S. T. was given 15.0 and then 30.0 g sucrose per day. The subject's control value for total ketoacid excretion of 92.5 ±11.5 fell to 28.3±1.2 ($P < 0.001$, from pre-CHO) and then to 21.2±1.1 mmoles/day per 100 ml GFR ($P < 0.01$, from 15 g) after the ingestion of 15.0 g and then 30.0 g/day.

To evaluate the effect of various carbohydrates on diminishing ketonuria, subject S. B. was given two different sugars. The subject first received 7.5 g/day of fructose which caused a fall in the mean daily ketoacid excretion rate from 86.3±7.6 to 62.2±6.1 mmoles/day per 100 ml GFR ($P < 0.05$). 15 g/day of fructose caused a further decline to 47.1±11.5 mmoles/day per 100 ml GFR ($P < 0.01$, from pre-CHO). A subsequent 14 day postcarbohydrate period of total starvation was of sufficient duration to allow the development of a new steady state of hyperketonuria. During this latter period the daily total ketoacid excretion was 97.2±8.8 mmoles/

[2] A small discrepancy exists between total daily urinary ketoacid excretion per 100 ml GFR and the sum of the daily excretion of β-OHB and AcAc per 100 ml GFR when 15.0 g CHO was administered. This is due to the fact that in subject S. T. urinary ketoacids were determined from organic acid excretion (see below), and hence separate analysis of β-OHB and AcAc excretion was not carried out.

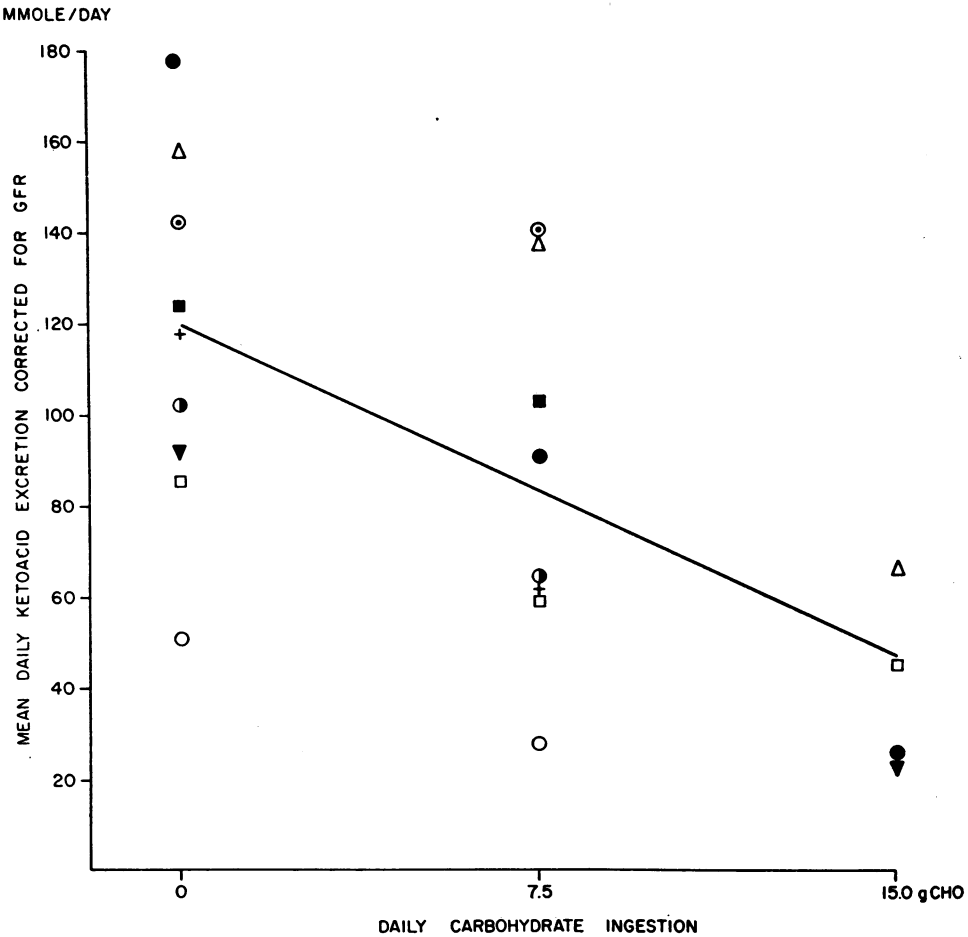


FIGURE 1 Data obtained showing changes in mean daily ketoacid excretion per 100 ml GFR vs. quantity of carbohydrate ingested daily. Each symbol represents the mean daily excretion of ketoacid over a 5 day period in one subject. The regression line is: $y = +119.6 - 4.82x$.

day per 100 ml GFR, a value unchanged from the previous pre-CHO control period. After the establishment of this new steady state the subject received 15.0 g/day of glucose. Ketoacid excretion fell to 33.4±3.5 mmoles/day per 100 ml GFR, a value similar to the one obtained during the 15.0 g/day of fructose administration.

In T. C. and L. C. ketoacid excretion rates were rising but did not reach new steady states after 5 days of post-CHO starvation. Nonetheless, the mean rate of ketoacid excretion for T. C. during these 5 post-CHO days rose from 33.5±3.8 to 228.6±18.0 mmoles/day per 100 ml GFR and for L. C. from 67.0± 5.0 to 121.6±15.0 mmoles/day per 100 ml GFR, thus showing the same tendency as seen in S. B. when total starvation was reinstituted. Fig. 2 shows daily ketoacid as well as ammonium excretion during the entire study in T. C.

Subject B. H., who had only one kidney, developed hyperketonemia indistinguishable from the other subjects. However, her control ketoacid excretion uncorrected for changes in GFR was only 29.5±2.8 mmoles/day. After ingestion of 7.5 g of CHO ketoacid excretion fell to 10.2±1.4 mmoles/day.

There was no significant correlation between ketoacid excretion and daily urine volume or urinary pH.

*Circulating ketoacids, FFA, glucose, and IRI (Tables III and IV).* The control pre-CHO blood AcAc level determined in six subjects was 1.27±0.08 mmoles/liter. It remained unchanged at 1.36±0.07 mmoles/liter when 7.5 g of CHO was ingested (Table III). Similarly, blood β-OHB levels were unchanged throughout the study. The pre-CHO control value was 5.00±0.32 mmoles/liter, and during the 7.5 g CHO period was 5.32±0.21 mmoles/liter (Table III). Blood ketoacid levels were determined in only two subjects (S. B. and L. C.) after the consumption of 15.0 g of CHO and during the post-CHO period. With the possible exception of higher blood ketoacid concentrations during the

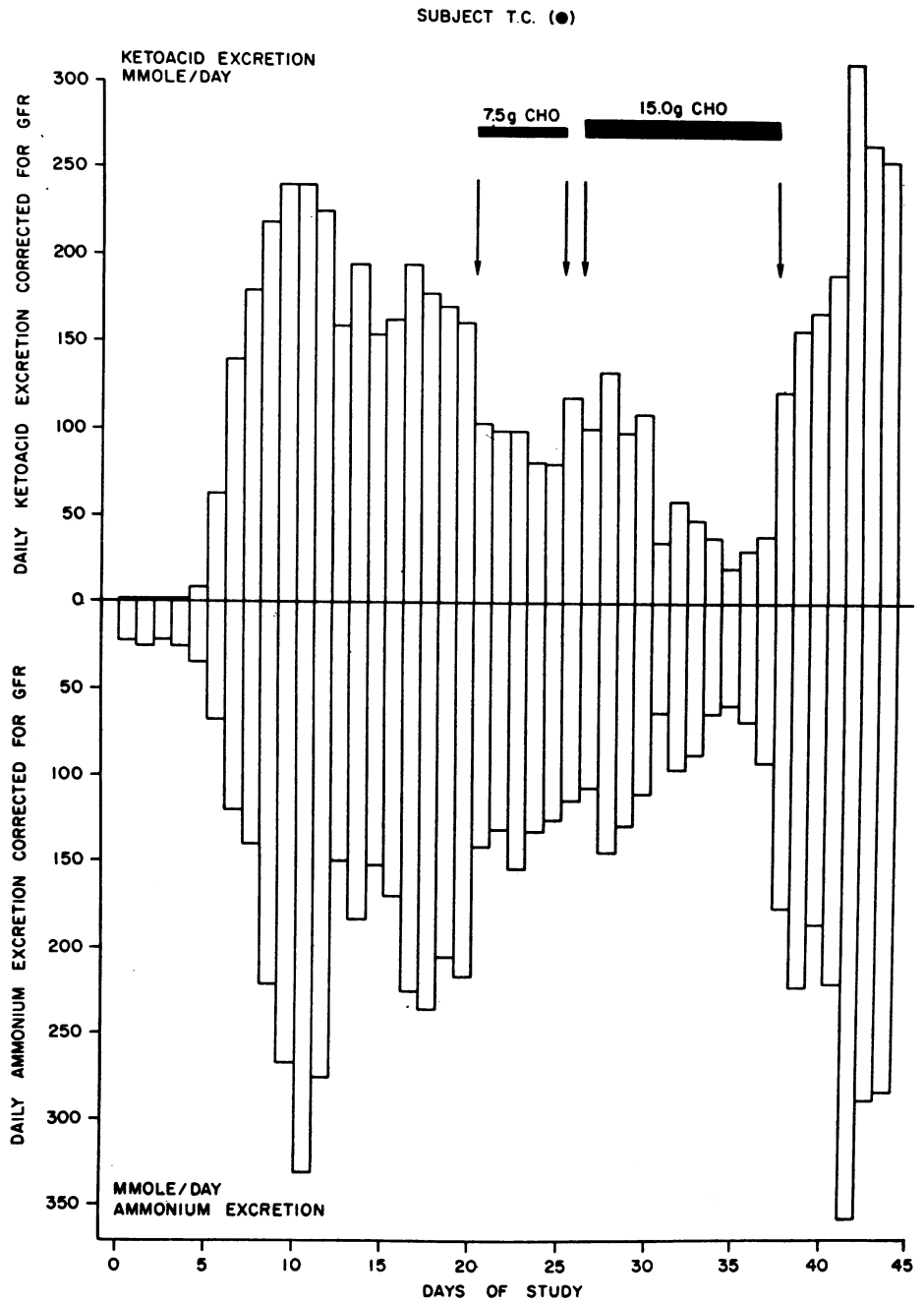


FIGURE 2 Changes in daily ketoacid and ammonium excretion expressed after correcting for changes in GFR during an entire study in subject T. C. The upper black bars denote the periods of administration of carbohydrate.

post-CHO period in L. C., all periods have similar blood ketoacid levels (Table III).

The pre-CHO control values for FFA, glucose, and IRI remained unchanged when 7.5 g of CHO was ingested (Table IV). After the ingestion of 15.0 g CHO (S. B. and L. C.) blood IRI and FFA remained unchanged while blood glucose showed a small and insignificant rise (Table IV). In the post-CHO control period (S. B. and L. C.) IRI and FFA remained constant while blood glucose fell, though this change was not significant (Table IV).

*Urinary ammonium and urea nitrogen excretion*

TABLE III

*Relationship between Blood AcAc and β-OHB Concentrations and Carbohydrate Ingestion*

| Subject | Precarbohydrate | | Carbohydrate: 7.5 g CHO | | Carbohydrate: 15 g CHO | | Postcarbohydrate | |
|---|---|---|---|---|---|---|---|---|
| | AcAc | β-OHB | AcAc | β-OHB | AcAc | β-OHB | AcAc | β-OHB |
| | *mmoles/liter* | | *mmoles/liter* | | *mmoles/liter* | | *mmoles/liter* | |
| D. P. | 1.17±0.10 | 4.65±0.44 | 1.48±0.31 | 4.95±0.64 | | | | |
| S. B. | 1.07±0.23 | 4.68±0.19 | 1.10±0.25 | 5.36±0.11 | 0.94±0.08 | 4.86±0.38 | 1.06±0.02 | 5.78±0.10 |
| L. S. | 1.10±0.61 | 3.71±0.55 | 1.47±0.14 | 4.53±0.95 | | | | |
| J. H. | 1.52±0.03 | 4.96±0.24 | 1.52±0.05 | 5.47±0.20 | | | | |
| B. H. | 1.53±0.03 | 6.11±0.29 | 1.40±0.06 | 6.11±0.25 | | | | |
| L. C. | 1.21±0.09 | 5.80±0.61 | 1.21±0.10 | 5.44±0.60 | 1.25±0.23 | 4.97±0.14 | 1.43±0.10 | 6.80±0.31 |
| Mean ±SEM | 1.27±0.08 | 5.00±0.35 | 1.36±0.07 | 5.30±0.21 | 1.10 | 4.92 | 1.25 | 6.29 |

(*Table V*). Daily ammonium excretion during the pre-CHO control period for all subjects was 153.1±21.1 mmoles/day. It fell to 107.6±14.9 mmoles/day ($P < 0.05$) with the ingestion of 7.5 g CHO/day, and in the four subjects (T. C., S. B., L. C., and S. T.) who ingested 15.0 g CHO to 56.0±9.4 mmoles/day ($P < 0.05$). When calculated per 100 ml GFR the control ammonium excretion of 148.3±11.7 mmoles/day was diminished to 127.4±12.6 mmoles/day ($P < 0.05$) after the ingestion of 7.5 g CHO daily. In the four subjects who received 15.0 g CHO/day their control excretion of 157.2±23.8 fell to 75.1±17.2 mmoles/day ($P < 0.05$, Table IV).

During the pre-CHO control period subject S. T. excreted 102.7±7.7 mmoles/100 ml GFR. After the ingestion of 15 and 30 g CHO daily, ammonium excretion fell to 38.1±2.2 and 23.5±1.8 mmoles/100 ml GFR, respectively. The administration of 15.0 g of fructose or glucose in subject S. B. produced indistinguishable declines in daily urinary ammonium excretion falling from 108.2±5.7 to 64.0±7.4 or 61.8±6.8 mmoles/100 ml GFR. In three subjects (S. B., T. C., and L. C.) followed after cessation of CHO intake the urinary ammonium excretions paralleled the increases in ketonuria.

There was a significant positive correlation between renal excretion of ammonium and ketoacids ($r = 0.95$, $P < 0.001$). There was no correlation between urinary ammonium excretion and urinary pH or volume.

The control daily urea nitrogen excretion of 2.6±0.3 g fell to 1.1±0.01 g ($P < 0.02$) when 7.5 g of CHO was consumed. It remained unchanged in the four sub-

TABLE IV

*Circulating Insulin, Substrate, and Bicarbonate Concentrations before, during, and after Carbohydrate Ingestion**

| | Precarbohydrate | Carbohydrate: 7.5 g CHO | Carbohydrate: 15 g CHO | Postcarbohydrate |
|---|---|---|---|---|
| Blood glucose, *mmoles/liter* | 3.29±0.26 | 3.41±0.21 | 4.36±1.82‡ | 3.28±0.66‡ |
| Serum IRI, *μU/ml* | 17 ±3 | 20 ±3 | 16 ±6 | 19 ±9 |
| Plasma FFA, *mmoles/liter* | 1.37±0.18 | 1.58±0.24 | 1.91±0.28 | 1.54±0.20 |
| Plasma $HCO_3^-$, *mmoles/liter* | 17.4 ±0.8 | 18.2 ±0.5 | 21.5 ±1.3§ | 17.2 ±0.7§ |

* Values expressed are mean ±SEM.

‡ The measurements of glucose, IRI, and FFA were made in only two subjects when 15.0 g CHO was ingested and when CHO supplements were discontinued.

§ Bicarbonate concentration was determined in four subjects when 15.0 g CHO was ingested and in three subjects after stopping CHO.

TABLE V

*Relationship between Daily Ammonium Excretion Corrected for Changes in GFR and Carbohydrate Ingestion*

| Subject | Precarbohydrate | Carbohydrate: 7.5 g CHO | Carbohydrate: 15.0 g CHO |
|---|---|---|---|
| | *mmoles/100 ml GFR per day* | *mmoles/100 ml GFR per day* | *mmoles/100 ml GFR per day* |
| D. P. | 137.7± 6.8 | 148.2± 7.0 (S)* | — |
| T. C. | 192.9±16.3 | 136.1± 4.9 (S) | 75.5± 6.5 (S) |
| S. B. | 108.2± 5.7 | 112.9± 6.2 (F) | 64.0± 7.4 (F)‡ |
| | | | 61.8± 6.8 (G) |
| J. T. | 135.5±12.4 | 102.3±10.3 (G) | — |
| L. S. | 193.2± 8.6 | 154.4± 7.6 (G) | — |
| J. H. | 163.9± 1.9 | 122.3± 7.4 (G) | — |
| B. H. | 97.4± 6.2 | 63.6± 5.3 (G) | — |
| L. C. | 175.6± 9.2 | 179.3±10.0 (G) | 121.5± 8.9 (G) |
| S. T.§ | 103.2± 7.7 | — | 38.1± 2.2 (S) |
| Mean ±SEM | 148.3±11.7 | 127.4±12.6‖ | 75.1±17.2‖ |

* F, S, and G denote fructose, sucrose, or glucose, respectively.

‡ Only one value (F) was used to calculate the mean for the entire group when 15.0 g CHO was given.

§ The figure for precarbohydrate ammonium excretion for subject S. T. was excluded when calculating the significance of the observed fall in ammonium output by the Student's *t* test for paired samples. The resultant mean value for daily ammonium excretion for the eight other subjects was 150.6±13.0 mmoles/100 ml GFR per day.

‖ $P < 0.05$ from precarbohydrate.

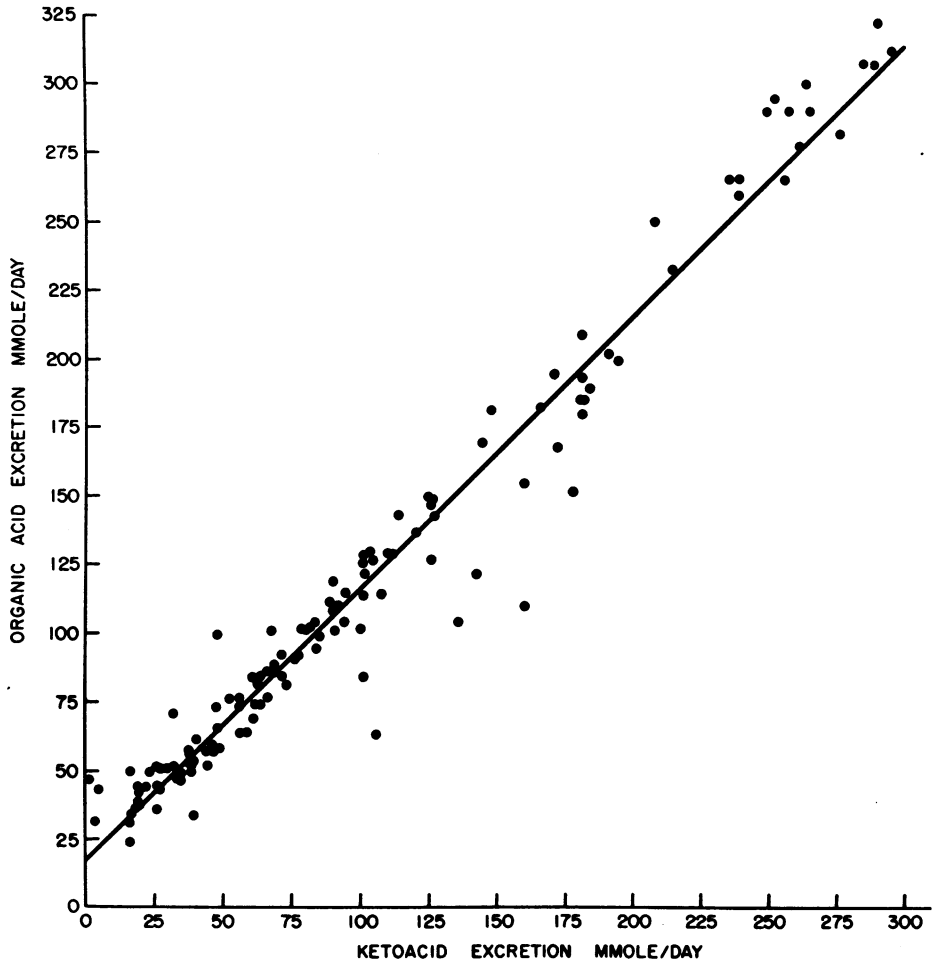


FIGURE 3 Daily organic acid vs. ketoacid excretion during an entire study in three subjects. Each point represents 1 day. The calculated regression line is: $y = +17.6 + 0.97x$.

jects studied during the administration of 15.0 g CHO/day. Similar values were obtained when daily urea nitrogen excretion was calculated per 100 ml GFR. Although the initial decrease in urea excretion after CHO ingestion is statistically significant it may represent the expected decline in urea excretion as starvation progresses (1). In the three subjects (T. C., S. B., and L. C.) studied post-CHO administration daily urea nitrogen excretion remained unchanged at 1.2±0.3 g/day, compared to 1.1±0.1 g/day in the preceding period of 15.0 g CHO ingestion.

*Venous $HCO_3$ and pH.* After the ingestion of 7.5 g CHO venous plasma bicarbonate (Table IV) remained unchanged at 18.2±0.7 mmoles/liter vs. a pre-CHO control value of 17.6±0.9 mmoles/liter. Venous pH rose during these periods from 7.32 to 7.36 ($P < 0.01$) primarily reflecting a decrease in the $P_{CO_2}$ from 33 to 31 mm Hg. When 15 g of CHO was given to four subjects (T. C., S. B., L. C., and S. T.) plasma $HCO_3^-$ rose significantly from 17.9±1.6 to 21.5±1.3 mmoles/liter ($P < 0.05$). Although the mean pH value increased from 7.31 to 7.38, this change was not statistically significant. In the post-CHO control period (three subjects) venous plasma $HCO_3$ fell to 17.2±0.7 mmoles/liter ($P < 0.02$) and the pH remained unchanged at 7.35.

*Glomerular filtration rates, urine volume, and pH.* The glomerular filtration rate measured as the creatinine clearance was 111.2±12.5 ml/min during the pre-CHO period. It was 81.2±10.0 ml/min with the ingestion of 7.5 g CHO and was 89.4±15.3 ml/min with the ingestion of 15.0 g CHO.

No consistent changes in urine pH or in daily urine volume were present during this study. Mean urine pH was 5.92, 6.09, 6.01 and corresponding urine volumes were 882±222, 732±169, and 927±227 ml/day

for the pre-CHO, 7.5, and 15.0 g CHO periods, respectively.

*Urinary organic acid excretion (Fig. 3).* The relationship between daily urinary organic acid and ketoacid excretion was examined in three patients (T. C., S. B., and L. S.) throughout the starvation and glucose administration periods. There was a highly significant correlation ($r = 0.98$, $P < 0.001$) calculated from the regression line: organic acids = 17.6 + 0.97 (ketoacids), obtained by plotting $U_{\text{organic acid}}V$/day vs. $U_{\text{ketoacid}}V$/day.

*Changes in body weights.* The weight loss for these subjects who did not receive supplementary sodium chloride during the entire study was 0.52±0.06 kg/day.[3] During the pre-CHO period the weight loss was 0.37 ±0.05 kg/day. The rate of weight loss was not significantly different after the ingestion of either 7.5 or 15.0 g CHO, being 0.30±0.04 kg/day during these periods.

## DISCUSSION

The antiketogenic and nitrogen-sparing effects of supplementary carbohydrates during caloric deprivation are well recognized (2–4). It is generally assumed that the daily administration of 50–100 g is required for these effects (3). The data presented here clearly demonstrate that the ingestion of 7.5 g carbohydrate daily during starvation causes a decrease in ketonuria and exerts a nitrogen-sparing effect by reducing urinary ammonium excretion. Increasing carbohydrate intake to 15 g daily causes a further decline in both ketoacid and ammonium excretion. Thus, starving man is exquisitely sensitive to minute amounts of ingested carbohydrates.

Although the precise mechanisms whereby small quantities of carbohydrate cause a fall in ketonuria are unclear, several conclusions can be drawn from the observed data. The diminished ketonuria seen after ingestion of 7.5 g carbohydrate cannot be explained by alterations in circulating levels of AcAc and β-OHB since no changes in the concentrations of these ketoacids were observed. Moreover, indirect evidence that a steady state of ketonuria existed can be derived from the observed circulating levels of FFA and insulin, which remained constant throughout the study period.

The fact that ketoacid excretion fell while blood AcAc and β-OHB remained constant implies that the renal reabsorption of ketoacids is accelerated by ingesting small quantities of carbohydrates. The reabsorption rates of AcAc and β-OHB/100 ml GFR (six subjects) can be calculated by using the observed mean blood levels, mean filtration rates, and mean daily excretions of these substances over the 5-day steady-state periods. This computation yields a reabsorptive rate for AcAc of 79 μmoles/min before carbohydrate administration and 90 μmoles/min after the ingestion of 7.5 g of carbohydrate. Rates for β-OHB are 279 μmoles/min and 344 μmoles/min in the precarbohydrate and carbohydrate periods, respectively. Although five of the six subjects demonstrated higher mean absorptive rates for AcAc and β-OHB after CHO ingestion, the results were not significantly different from control ($P < 0.1$) since the sixth subject's reabsorptive rate fell. Since there was no demonstrable change in blood ketoacid concentrations after carbohydrate ingestion the apparent enhanced renal reabsorption was probably coupled with either increased utilization and/or decreased production of ketoacids. The possibility that changes in either rates of hepatic production or endogenous utilization accompany the changes in renal reabsorption of ketoacids is suggested by the rise in plasma bicarbonate observed when 15.0 g carbohydrate was ingested. This increase in bicarbonate could be explained by either a decrease in the rate of ketoacid production or an increased rate of ketoacid oxidation. The question of whether blood ketoacid levels changed when 15.0 g carbohydrate was administered, cannot be definitely answered, although in the two subjects studied they appeared to remain constant.

Whether the effect of the ingested carbohydrate on renal ketoacid excretion is exerted through an action of glucose in the blood or is mediated by a hormonal agent is not clear from the present study. There was no increase in venous blood glucose 1 hr after the morning dose of 2.5 g carbohydrate had been ingested. However, a slight but not statistically significant increase was noted when blood was drawn 1 hour after the ingestion of 5.0 g of glucose. In recent publications it has been noted that ketoacid excretion is modified by the administration of hormones. Felig, Marliss, and Cahill demonstrated that the heightened ketonuria after administration of growth hormone to starving man was related to elevated blood levels of AcAc and β-OHB (18). When glucagon was administered to starving man an increased excretion of ketoacid was observed in association with an unchanged blood level of these substances (19). In contrast to glucagon, glucocorticosteroid administration during starvation diminished renal clearance of ketoacids (20). Although urinary corticosteroid excretion falls during starvation and rises with refeeding (21), circulating levels or secretion rates do not appear to change significantly (22). It is therefore unlikely that corticosteroids are responsible for altered ketoacid excretion.

[3] The higher rate of weight loss during the entire study than during the pre-CHO or CHO administration periods reflects the initial loss of edema fluid observed during the 1st wk of starvation.

Although the effect of insulin on renal handling of ketoacids during starvation has not been determined, we observed no change in serum insulin concentration after the ingestion of small amounts of carbohydrate though changes in ketonuria were clearly present.

Subject B. H., who had one kidney and a diminished glomerular filtration rate, exhibited a ketoacid excretion that at its maximum was one-quarter the value of the other subjects studied. Despite this marked decrease in ketonuria her blood ketoacid level was not different from those other subjects. This finding suggests that a mechanism is operative in starvation to modulate and keep constant the circulating ketoacid concentration (23).

The fall in daily urinary excretion of ammonium after the ingestion of 7.5 g of carbohydrate demonstrates that the nitrogen-sparing effects of carbohydrate can be exerted with a much smaller quantity than that reported by others (3, 24). In this study the decline in urinary ammonium excretion seen with the ingestion of 7.5 g carbohydrate was also accompanied by a decrease in the excretion of urea nitrogen. On the basis of a previous observation by Owen, Felig, Morean, Wahren, and Cahill, who demonstrated a progressive fall in urea excretion through the first 4 wk of starvation (1), it is unlikely that the small amount of carbohydrate ingested affected urea excretion in this study. Further support for this arises from the stability of urea excretion when the carbohydrate supplements were increased from 7.5 to 15.0 g daily or when they were discontinued. After 4 wk of total starvation ammonium excretion constitutes approximately half the daily nitrogen loss (1). During this study 15 g carbohydrate ingested daily reduced the loss of ammonium nitrogen by approximately 70% of the control value, resulting in a significant sparing effect on body nitrogen stores. It thus appears that the nitrogen-sparing effect of carbohydrates in starvation is the result of at least two separable processes. The first is due to a decrease in renal ammonium output as observed in this study. The second process, a fall in urea excretion accompanying decreased hepatic gluconeogenesis, was not seen since urea excretion was unchanged during the administration of 7.5 g or 15 g of carbohydrate and during the postcarbohydrate control period. A more pronounced fall in total urinary nitrogen presumably the result of a reduction in both urine urea and ammonium excretion was observed by Gamble (3) after the daily administration of 50–100 g of carbohydrate.

The changes in renal ammonium excretion observed during this study were not related to factors known to influence renal ammoniagenesis, namely GFR and extracellular or urinary pH (25–27). Correction of the ammonium excretion data for alterations in glomerular filtration rates did not change its significance. Moreover, there were no significant changes in plasma bicarbonate or venous pH from control values when ammonium excretion fell after the administration of 7.5 g carbohydrate, and finally urine pH did not vary significantly during the study. What other factors control the excretion of ammonium during carbohydrate administration and whether the observed changes after carbohydrate ingestion were due to a primary effect of ketonuria or renal ammoniagenesis are not evident from the present investigation. In view of recent data linking renal ammoniagenesis with gluconeogenesis (28), it is possible that the diminution seen in ammonium and nitrogen excretion was caused by alterations in the rate of renal gluconeogenesis.

The close correspondence between the observed changes in renal ketoacids and ammonium output throughout all of the study suggests that the minute quantities of ingested carbohydrate may produce a change in a common intrarenal metabolic pathway, thus resulting in the parallel decreases in ketoacid and ammonium nitrogen excretion.

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