Effect of carbohydrate ingestion on glucose kinetics and muscle metabolism during intense endurance exercise

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McConnell, Glenn K., Benedict J. Canny, Marcus C. Daddo, Marcus J. Nance, and Rodney J. Snow. Effect of carbohydrate ingestion on glucose kinetics and muscle metabolism during intense endurance exercise. J Appl Physiol 89: 1690–1698, 2000.—There has been recent interest in the potential performance and metabolic effects of carbohydrate ingestion during exercise lasting ∼1 h. In this study, 13 well-trained men ingested in randomized order either a 6% glucose solution (CHO trial) or a placebo (Con trial) during exercise to exhaustion at 83 ± 1% peak oxygen uptake. In six subjects, vastus lateralis muscle was sampled at rest, at 32 min, and at exhaustion, and in six subjects, glucose kinetics was determined by infusion of [6,6-2H]glucose in both trials and ingestion of [6-3H]glucose in the CHO trial. Of the 84 g of glucose ingested during exercise in the CHO trial, only 22 g appeared in the peripheral circulation. This resulted in a small (12 g) but significant (P < 0.05) increase in glucose uptake without influencing carbohydrate oxidation, muscle glycogen use, or time to exhaustion (CHO: 68.1 ± 4.1 min; Con: 69.6 ± 5.5 min). Decreases in muscle phosphocreatine content and increases in muscle inosine monophosphate and lactate content during exercise were similar in the two trials. Although endogenous glucose production during exercise was partially suppressed in the CHO trial, it remained significantly above preexercise levels throughout exercise. In conclusion, only 26% of the ingested glucose appeared in the peripheral circulation. Glucose ingestion increased glucose uptake and partially reduced endogenous glucose production but had no effect on carbohydrate oxidation, muscle metabolism, or time to exhaustion during exercise at 83% peak oxygen uptake.

endogenous glucose production; glucose absorption; insulin; carbohydrate oxidation; muscle inosine monophosphate; humans

CARBOHYDRATE INGESTION DURING exercise at 70–75% maximal oxygen uptake (VO2max) increases time to exhaustion (6, 25). This increase in endurance is thought to be due to a maintenance of blood glucose availability and higher muscle glucose uptake (24) and carbohydrate oxidation (6) late in exercise when muscle glycogen levels are low. It has generally been considered that carbohydrate ingestion does not benefit exercise performance during more intense exercise of ∼1 h at ∼80–85% VO2max. Indeed, carbohydrate ingestion has been shown to have no effect on cycling time to exhaustion at ∼85% VO2max in trained men (28). However, several recent studies have found an improvement in exercise performance when carbohydrate was ingested during time trial-type exercise of ∼60 min of exercise at 80–90% VO2max (e.g., Refs. 2, 17). It is difficult to understand why carbohydrate ingestion would benefit such exercise because the proportional contribution of muscle glycogen to energy use far exceeds the contribution of blood glucose at these high intensities (31) and muscle glycogen is not fully depleted after such exercise (10). In addition, the absorption of exogenous glucose may be lower at ∼85% VO2max than at ∼70% VO2max, and blood glucose concentration tends to increase even when no carbohydrate is ingested during exercise at 80–85% VO2max (2, 28). Carbohydrate ingestion increases the rate of glucose uptake during exercise at 70% VO2max (24), but it is not known whether carbohydrate ingestion increases glucose uptake during more intense exercise at 80–85% VO2max. Therefore, the first aim of this study was to determine whether carbohydrate ingestion increases glucose uptake and improves exercise capacity during heavy exercise lasting ∼1 h.

Fatigue after prolonged exercise at 70–75% VO2max is associated with decreases in tricarboxylic acid cycle intermediates (TCAIs) and increased muscle IMP (26, 33, 36). Muscle IMP has been used as an indicator of transient increases in ADP and AMP and, therefore, muscle energy imbalance during exercise (26, 32). It has been shown that carbohydrate ingestion results in lower levels of muscle IMP (25, 36) and higher TCAI (36) contents late in exercise at 70–75% VO2max. This suggests that endurance at this intensity is limited by carbohydrate availability and that carbohydrate ingestion may improve performance by maintaining muscle energy balance late in exercise. The effect of carbohydrate ingestion on muscle energy balance during more intense exercise (80–90% VO2max) has not been examined and constitutes the second aim of this study.

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Abstract
Ingestion of glucose during exercise at 50–70% \(\text{VO}_2\text{max}\) in humans increases plasma glucose and insulin levels and suppresses endogenous glucose production (EGP) to resting levels (19, 24). During more intense exercise at \(\sim 85\% \text{VO}_2\text{max}\), rats, glucose infusion fails to fully suppress EGP despite large increases in plasma glucose levels (38). It was suggested that this response was due to the stimulatory effect of high circulating levels of epinephrine on EGP. It is not known whether a similar response occurs when carbohydrate is ingested during intense exercise in humans. Therefore, the third aim of this study was to examine the effect of glucose ingestion on EGP during exercise at \(\sim 80\% \text{VO}_2\text{max}\) in humans.

**METHODS**

**Subjects**

Thirteen well-trained cyclists and triathletes volunteered for this study, which was approved by Monash University Standing Committee on Ethics in Research Involving Humans. Before commencing the study, they completed a medical questionnaire and provided informed, written consent. The age, height, and weight of the subjects were \(24 \pm 1\) (SE) yr, \(183 \pm 2\) cm, and \(77.1 \pm 2.5\) kg, respectively. Approximately 2 wk before the first trial, peak pulmonary oxygen uptake (\(\text{VO}_2\text{peak}\)) was determined during continuous incremental cycling (Lode, Groningen, The Netherlands) to volitional fatigue and averaged \(5.05 \pm 0.16\) l/min (\(65.7 \pm 1.5\) ml·kg\(^{-1}\)·min\(^{-1}\)). Approximately 1 wk before the first trial, the subjects attended the laboratory for a familiarization trial, during which they cycled to exhaustion at a similar work rate (\(293 \pm 9\) W, \(82 \pm 1\% \text{VO}_2\text{peak}\)) and were encouraged to encounter in the experimental trials (\(294 \pm 9\) W, \(83 \pm 1\% \text{VO}_2\text{peak}\)). Approximately 24 h before each trial, the subjects reported to the laboratory for a 45-min cycling bout at \(70 \pm 1\% \text{VO}_2\text{peak}\). They then refrained from physical exercise and were supplied with food for the remainder of the day (\(15.4 \pm 0.5\) MJ: 65.8 ± 0.0% carbohydrate, 19.2 ± 0.1% fat, 15.0 ± 0.1% protein). The subjects were instructed to refrain from caffeine, alcohol, and tobacco intake for the 24 h before each trial. Such exercise and dietary control appears to result in reproducible preexercise metabolite and hormone levels (24). In an attempt to match the subject’s hydration status between trials, the subjects were also asked to ingest fluids at a rate sufficient to produce “clear” urine during the day before a trial and to ingest 250 ml of water \(\sim 1\) h before attending the laboratory. On arrival at the laboratory for the trial, they ingested a further 250 ml of water.

**Experimental Procedures**

**Subject involvement.** Thirteen subjects were involved in this study, with all subjects cycling to exhaustion while ingesting in one trial a glucose solution (CHO trial) and in the other trial a placebo (Con trial). It was considered unnecessary and overly costly for all of the subjects to be subjected to the glucose tracer components of the study. Therefore, six subjects were involved in this aspect. Five of these six subjects were also involved in the muscle biopsy procedures of the study. An additional one subject was muscle biopsied without being involved in the tracer aspects. The remaining six subjects were not involved in the glucose tracer or muscle biopsy components of the study.

**Common to all subjects.** The subjects reported to the laboratory after either an overnight fast (9–11 h) or a 6- to 8-h fast (afternoon trials). The trials for each subject took place at the same time of the day. The subjects voided, then a catheter (Optiva, 20 gauge) was inserted into an antecubital vein, and a blood sample was then obtained. The catheter for blood sampling was kept patent by flushing with 0.9% saline and every 30 min with \(\sim 0.5\ ml\) of saline containing 10 U/ml heparin. Blood for glucose, lactate, hemoglobin, and hematocrit determination was placed in fluoride heparin tubes; blood for nonesterified fatty acids (NEFA) analysis was placed in tubes containing EDTA; and blood for insulin, sodium, and potassium measurement was placed in lithium heparin tubes. After removal of whole blood for hemoglobin and hematocrit analysis, the tubes were spun and the plasma stored at \(-20\°\)C for later analysis. After the blood sampling, subjects rested for 60–120 min before cycling to volitional exhaustion at \(294 \pm 9\) W, which elicited \(83 \pm 1\%\) of their \(\text{VO}_2\text{peak}\). It was necessary for the subjects that were infused with tracer to rest for 120 min before commencing exercise to allow for tracer equilibrium. Therefore, in an attempt to match the preexercise conditions as closely as practicable, the subjects not involved in the tracer component of the study rested for \(\sim 1\) h before commencing exercise. Subjects ingested 7 ml/kg of fluid at room temperature immediately before exercise and then 3.5 ml/kg of fluid every 15 min of exercise. In the CHO trial, a noncommercial 6% \(\beta\)-glucose artificially flavored solution was ingested. In the Con trial, an equal volume of artificially sweetened and flavored water placebo was ingested. The rate of fluid ingestion during exercise was 1,348 ± 44 ml/h, and glucose supplementation in the CHO trial was 81 ± 3 g/h. The trials were conducted in a counterbalanced order and double blind. Fatigue was defined as the point when the subject could no longer maintain the workload despite strong verbal encouragement. Because the subjects were cycling on an electrically braked ergometer, the work rate remained constant, independent of the revolution rate chosen. The laboratory was maintained at 19–22°C, and a large fan placed in front of the subject circulated air to minimize thermal stress. Heart rate was recorded from a heart rate monitor (Accurex, Polar, Oulu, Finland) throughout exercise and at the point of fatigue. In addition, after every 10 min of exercise, expired air was collected into a Douglas bag for oxygen uptake and respiratory exchange ratio (RER) determination. A Douglas bag was also collected as close to the point of fatigue as practical (within the last 5–15 min of exercise). The use of RER to calculate carbohydrate oxidation has been shown to be valid up to \(85\% \text{VO}_2\text{max}\) in trained men (30). The subjects were asked to provide a rating of their perceived exertion (RPE) during exercise by using the 14-point Borg scale (4).

**Subjects assessed for glucose kinetics.** These subjects (six) attended the laboratory in the morning after an overnight fast. In these subjects, an additional catheter was inserted into an antecubital vein of the contralateral arm for [6,6-\(^2\)H\]glucose infusion (Intracath, 19 gauge). A blood sample was obtained, after which a primed, continuous infusion of [6,6-\(^2\)H\]glucose (Cambridge Isotope Laboratories, Andover, MA) was commenced. The bolus dose was \(54.2 \pm 2.3\) \(\mu\)mol/kg, and the infusion rate of glucose tracer (0.62 ± 0.04 \(\mu\)mol·kg\(^{-1}\)·min\(^{-1}\)) remained unchanged for the duration of the experiment (120 min of rest and throughout exercise). In the glucose ingestion trial, 1 \(\mu\)Ci [6-\(^2\)H\]glucose/g glucose was added to the ingested 6% glucose solution. Blood samples were obtained every 10 min during the last 30 min of rest and then every 5 min during the first 30 min of exercise and then every 10 min of exercise and at the point of fatigue for the measurement of plasma glucose, percent enrichment of [6,6-\(^2\)H\]glucose, and the specific activity of [6-\(^2\)H\]glucose. A por-
tion of the blood sampled was placed in specific tubes for later analysis of plasma cortisol (lithium heparin tubes), glucagon (lithium heparin tubes containing aprotinin), norepinephrine, and epinephrine (plain tubes containing EGTA and reduced glutathione). In the CHO trial, an aliquot of the ingested drink was frozen for later measurement of glucose concentration and [6-3H]glucose specific activity. In both trials, an aliquot of the infusate was frozen for later measurement of glucose concentration. The exact pump (Minipuls 2, Gilson, Villiers-le-Bel, France) infusion rate was determined at the end of each trial.

Subjects assessed for muscle metabolism. In six subjects, muscle samples were obtained from the vastus lateralis at rest, after 32 min of exercise, and immediately after exercise. Muscle sampling took place under local anesthesia by using a percutaneous needle-biopsy technique with suction. Muscle samples were frozen in liquid nitrogen within 20 s after the subjects ceased exercise. At 32 min, a standard 60-s rest period was allowed for completion of the biopsy and taping of the area. For consistency, this 60-s rest period was given to all subjects, including those who did not undergo muscle sampling. The muscle samples were analyzed for glycogen, ATP, ADP, AMP, IMP, phosphocreatine (PCr), and creatine (Cr).

Analytic Techniques

Gas analysis. Expired air samples were measured for oxygen and carbon dioxide content by using Exerstess OX21 and CO21 electronic analyzers (Clinical Engineering Solutions, Sydney, Australia). These analyzers were calibrated by using commercial gases of known composition. Expired air volume was measured by using a dry-gas meter (American Meter, Vacumed, Ventura, CA) calibrated against a Tissot spirometer.

Blood. Blood hematocrit was measured in quadruplicate by microcentrifugation, whereas hemoglobin was measured spectrophotometrically in triplicate by using the cyanmethemoglobin method. Changes in plasma and blood volume were estimated by using the Dill and Costill equation (9). Plasma glucose and lactate were determined by using an automated glucose oxidase and l-lactate oxidase method, respectively (model YSI 2300 Stat, Yellow Springs Instrument, Yellow Springs, OH). Plasma NEFA content was analyzed by an automated colorimetric procedure (NEFA-C test, Wako, Osaka, Japan). Plasma sodium and potassium were measured by using an automated ion-selective electrode method (Ciba-Corning, Essex, UK). Plasma insulin (Incstar, Stillwater, MN), plasma glucagon (1), and total plasma cortisol (3) were determined by radioimmunoassay, whereas plasma catecholamines were measured by using radioenzymatic assay (Amersham, Buckinghamshire, UK). The percent enrichment of [6,6-2H]glucose and the specific activity of [6-3H]glucose in plasma samples were determined as described previously (24). Briefly, plasma was deproteinized and then spun. The resulting supernatant was passed down an ion-exchange column. The eluant was dried and reconstituted with water, and a portion was added to scintillant before being counted on a beta counter for the determination of [6-3H]glucose. The other portion was dried and then derivatized to the pentacetate derivative. The derivatized glucose level was measured with a gas chromatography-mass spectrometer using a selected ion-monitoring mode to determine the relative abundance of the selected ions with mass-to-charge ratios of 98 and 100. Glucose kinetics at rest and during exercise were estimated by using a modified one-pool, non-steady-state model as proposed by Steele et al. (37), which has been validated by Radziuk et al. (29). We assumed a value of 0.65 as the rapidly mixing portion of the glucose pool and estimated the apparent glucose space as 25% of body weight. Rates of plasma glucose appearance (total Ra) and disappearance (Rd) were calculated by dividing glucose Rd by the plasma glucose concentration. Glucose kinetic comparisons between two successive time points (e.g., between 10 and 15 min) result in a data point that corresponds to 12.5 min, which is the midpoint of the sampling interval. The muscles of the legs account for 80–85% of tracer-determined, whole body glucose uptake during exercise at 55–60% V O2 max and probably a greater proportion during more intense exercise (19). During exercise at 50% of V O2 max workload >90% of tracer-determined glucose uptake is oxidized (19). The rate of appearance into plasma of the ingested [6-3H]glucose (gut Ra) was determined by transposition of the equation of Steele et al. (37) and the known specific activity of the drink. In the Con trial, EGP was equal to total R a, whereas in the CHO trial, EGP was calculated as total R a minus gut R a. EGP comprises glucose output from both hepatic glycogenolysis and gluconeogenesis with a possible small contribution from the kidney.

Muscle. The muscle samples were freeze dried and then crushed to a powder with any visible connective tissue removed. For muscle glycogen determination, ~1 mg of muscle was added to HCl, incubated at 100°C, then neutralized with NaOH, and analyzed for glucose units by using an enzymatic, fluorometric method (27). For analysis of the other muscle metabolites, ~2 mg of muscle were extracted according to the procedure of Harris et al. (13). Muscle lactate, PCr, and Cr were analyzed by using enzymatic, fluorometric techniques (20), whereas muscle ATP, ADP, AMP, and IMP were measured by HPLC as described by Snow et al. (35). The intra-assay coefficient of variation for these muscle analyses in our hands is as follows: glycogen = 4.3%, lactate = 4.5%, PCr = 7.4%, and Cr = 8.6% (enzymatic, fluorometric assays); and ATP = 5.5%, ADP = 5.9%, AMP = 5.2%, and IMP = 9.0% (HPLC). The content of ATP, ADP, AMP, IMP, PCr, and Cr were corrected to the peak total Cr (PCr + Cr) content for each subject to account for any nonmuscle contamination of the muscle samples.

Statistics

Data from the two trials were compared by using two-factor repeated-measures analysis of variance. The significance level for statistical analysis was set at the P < 0.05 level. If a significant interaction existed, specific differences were located by using the Fisher’s least significant difference test. Performance time was compared with a paired t-test. All data are reported as means ± SE.

RESULTS

Heart Rate, Hemoglobin, and Blood and Plasma Volume Measurements

Pretrial hydration status was likely to be similar (P > 0.05) in the two trials because resting hemoglobin levels (CHO: 14.7 ± 0.4 g/100 ml; Con: 14.7 ± 0.4 g/100 ml) and heart rate at 10 min of exercise (CHO: 165 ± 3 beats/min; Con: 162 ± 3 beats/min) were similar. There was no difference (P < 0.05) in heart rate, blood volume, or plasma volume response during exercise between the two trials. Heart rate significantly increased...
from the 10-min time point in both trials and at the end of exercise was 175 ± 4 beats/min in the Con trial and 178 ± 3 beats/min in the CHO trial. Blood volume and plasma volume decreased \((P < 0.05)\) to a similar extent in the two trials during the first 10 min of exercise and then remained essentially unchanged until the final measurement at 60 min of exercise. For example, blood volume at 60 min of exercise had decreased from rest by 5.3 ± 1.7% in the Con trial and by 7.4 ± 1.2% in the CHO trial.

**RPE, Oxygen Consumption, RER, and Carbohydrate Oxidation, and Performance Measurements**

RPE increased \((P < 0.05)\) during exercise to a similar extent in both trials and was 19 ± 0 at exhaustion in the Con trial and 18 ± 0 at exhaustion in the CHO trial. No \((P > 0.05)\) trial or time effects in oxygen consumption, RER, or estimated carbohydrate oxidation \((\text{g/min} \text{ and } \mu\text{mol} \cdot \text{kg}^{-1} \cdot \text{min}^{-1})\) were observed during exercise even approaching the point of fatigue (Table 1). Calculated carbohydrate oxidation averaged 4.49 ± 0.20 g/min \((323 ± 10 \mu\text{mol} \cdot \text{kg}^{-1} \cdot \text{min}^{-1})\) in the Con trial and 4.53 ± 0.21 g/min \((327 ± 12 \mu\text{mol} \cdot \text{kg}^{-1} \cdot \text{min}^{-1})\) in the CHO trial. The high RER of 0.96–0.97 throughout exercise indicates that over 85% of energy was derived from carbohydrate sources. The exercise time to exhaustion was not different \((P > 0.05)\) between the two trials, being 68.1 ± 4.1 min in the CHO trial and 69.6 ± 5.5 min in the Con trial \((n = 13)\). The time to exhaustion in the subgroup of subjects \((n = 6)\) who underwent muscle sampling before and during exercise \((\text{CHO}: 72.0 ± 6.4 \text{ min}; \text{Con}: 73.6 ± 9.9 \text{ min})\), and in the subgroup of subjects \((n = 6)\) involved with measurement of glucose kinetics \((\text{CHO}: 66.8 ± 6.7 \text{ min}; \text{Con}: 67.8 ± 10.7 \text{ min})\), was not significantly different from the group as a whole and not significantly different between trials. In addition, there was no significant order effect in terms of time to exhaustion \((\text{trial 1}: 71.2 ± 4.8 \text{ min}; \text{trial 2}: 66.4 ± 4.8 \text{ min})\).

**Plasma Metabolites and Hormones**

No differences were observed between trials for any of the measured plasma substrates, ions, or hormones at rest before treatment (Fig. 1, Table 2). Plasma glucose concentration was higher \((P < 0.05)\) at 15 min and throughout exercise in the CHO trial (Fig. 1A). Plasma insulin decreased significantly in both trials during exercise and was lower \((P < 0.05)\) in the Con compared with CHO trial (treatment effect, Fig. 1B). Plasma NEFA decreased \((P < 0.05)\) over the first 30 min of exercise in CHO and was lower than Con at 30 min and 60 min of exercise (Fig. 1C). Plasma sodium and potassium concentrations increased significantly during the first 30 min of exercise and then remained essentially unchanged until exhaustion with no differences between trials (data not shown). Plasma lactate was similar at rest \((\text{CHO}: 1.1 ± 0.1 \text{ mmol/l}; \text{Con}: 1.2 ± 0.1 \text{ mmol/l})\) and throughout exercise in the two trials \((6.7 ± 0.5 \text{ mmol/l} \text{ in CHO and } 7.1 ± 0.9 \text{ mmol/l} \text{ in Con})\) at 30 min and 7.8 ± 0.8 mmol/l in CHO and 7.5 ± 1.0 mmol/l in Con at exhaustion). In the subjects \((n = 6)\) involved in the glucose kinetics component of the study, there were no significant differences between trials at rest or during exercise in plasma glucagon, cortisol, epinephrine, or norepinephrine (Table 2). Plasma cortisol increased significantly during exercise in both trials. Plasma norepinephrine increased significantly throughout exercise in the two trials, whereas plasma epinephrine increased from rest to 15 min of exercise and then was essentially unchanged at 60 min of exercise (Table 2).

**Glucose Kinetics**

The pattern of the plasma glucose response during exercise in the six subjects involved with the glucose kinetics facet of the study was not significantly different from the group as a whole. The percent enrichment of \([6,6-2H] \text{glucose}\) decreased significantly during exercise even approaching the point of fatigue (Table 1).

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**Table 1. Oxygen consumption, respiratory exchange ratio, and carbohydrate oxidation during exercise to exhaustion at 83 ± 1% \(\text{V} \text{O}_{2}\) with (CHO) and without (Con) carbohydrate ingestion**

<table>
<thead>
<tr>
<th>Time, min</th>
<th>10</th>
<th>20</th>
<th>30</th>
<th>40</th>
<th>50</th>
<th>Exhaustion</th>
</tr>
</thead>
<tbody>
<tr>
<td>(\text{V} \text{O}_{2}, \text{l/min}) Con</td>
<td>4.12±0.13</td>
<td>4.13±0.13</td>
<td>4.16±0.15</td>
<td>4.17±0.15</td>
<td>4.31±0.13</td>
<td>4.17±0.13</td>
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<tr>
<td>CHO</td>
<td>4.16±0.15</td>
<td>4.16±0.14</td>
<td>4.16±0.16</td>
<td>4.20±0.15</td>
<td>4.21±0.18</td>
<td>4.23±0.16</td>
</tr>
<tr>
<td>RER</td>
<td>0.97±0.01</td>
<td>0.97±0.01</td>
<td>0.97±0.01</td>
<td>0.96±0.01</td>
<td>0.96±0.01</td>
<td>0.96±0.01</td>
</tr>
<tr>
<td>Con</td>
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<td>0.98±0.01</td>
<td>0.97±0.01</td>
<td>0.97±0.01</td>
<td>0.96±0.01</td>
<td>0.97±0.01</td>
</tr>
<tr>
<td>CHO</td>
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<td>4.50±0.20</td>
<td>4.57±0.22</td>
<td>4.43±0.22</td>
<td>4.57±0.20</td>
<td>4.33±0.20</td>
</tr>
<tr>
<td>Carbohydrate oxidation, g/min</td>
<td>4.60±0.18</td>
<td>4.59±0.16</td>
<td>4.55±0.21</td>
<td>4.47±0.20</td>
<td>4.43±0.27</td>
<td>4.57±0.23</td>
</tr>
<tr>
<td>Con</td>
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<td>328±10</td>
<td>318±11</td>
<td>324±10</td>
<td>313±11</td>
</tr>
<tr>
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<td>332±9</td>
<td>332±9</td>
<td>328±12</td>
<td>329±13</td>
<td>318±17</td>
<td>330±14</td>
</tr>
</tbody>
</table>

Values are mean ± SE; \(n = 13\) subjects. Values at exhaustion relate to data collected during the final 5–15 min of exercise. Con, control trial; CHO, carbohydrate ingestion trial; \(\text{V} \text{O}_{2}\), oxygen uptake; RER, respiratory exchange ratio.
reduction in percent enrichment of [6,6-2H]glucose was due to the fact that the tracer infusion rate was kept constant at rest and during exercise, whereas glucose appearance in the blood increased during exercise. It is likely that this fall in percent enrichment led to a small underestimation of the calculated glucose Ra that was quantitatively relatively similar in both trials. Glucose total Ra, glucose Rd, and glucose clearance rate were similar before exercise in the two trials (Fig. 2, Table 4). Exogenous glucose appearance (gut Ra) during exercise in the CHO trial was 22 g (Table 4). Therefore of the 84 g of glucose ingested in the CHO trial, only 26% appeared in the peripheral blood. Total Ra (EGP + gut Ra) increased (P < 0.05) in both trials and was significantly higher in the CHO than in the Con trial (Fig. 2A) because of the contribution of gut-derived glucose (Table 4). Total Ra increased (P < 0.05) until 17.5 min of exercise in both trials and then remained relatively unchanged for the remainder of exercise in both trials (Fig. 2A). During the first 17.5 min of exercise, EGP increased to a similar extent in both trials (Fig. 2B). In the Con trial, EGP continued to increase throughout the exercise bout, whereas in the CHO trial, EGP was significantly suppressed from 22.5 min until the end of the trial. Although EGP was suppressed in the CHO compared with the Con trial, EGP remained significantly above the preexercise level throughout exercise (Fig. 2B). Glucose Ra increased during exercise in both trials and was significantly higher in the CHO trial toward the latter stages of exercise (Fig. 2C). The total exercise glucose Rd was 45 g in the CHO trial and 33 g in the Con trial. Of the 45 g of glucose in the CHO trial, 23 g was from the exogenous source (calculated by subtracting glucose Rd from EGP). Glucose clearance rate increased throughout exercise to a similar (P > 0.05) extent in the two trials (Table 4).

Muscle Measurements

Muscle glycogen, ATP, ADP, AMP, total adenine nucleotide (TAN = ATP + ADP + AMP), IMP, PCr, Cr, and lactate were similar (P > 0.05) in the two trials at rest, after 32 min, and at exhaustion (Fig. 3, Table 5). In both trials, muscle glycogen decreased significantly from rest to 32 min, and then from 32 min until exhaustion (Table 5). Muscle glycogen use during the trials was not significantly influenced by carbohydrate ingestion during exercise (CHO: 366 ± 16 mmol/kg dry muscle, Con: 292 ± 42 mmol/kg dry muscle). No alteration in muscle ATP, ADP, AMP, or TAN content as determined by HPLC analysis was observed during exercise in either trial (Table 5). Muscle PCr decreased (P < 0.05) and muscle lactate and IMP increased (P <...
0.05) from rest to 32 min in both trials and then remained essentially unchanged until exhaustion (Fig. 3). As would be expected from the muscle PCr data, muscle Cr increased significantly from rest to 32 min and then remained unchanged until exhaustion (Table 5).

**DISCUSSION**

The major finding of this study was that, during exercise at 83% V\(\text{O}_2\) max, insufficient ingested glucose appears in the blood to have major effects on exercise metabolism. Of the 84 g of glucose ingested in the CHO trial during the 70 min of exercise, only 22 g appeared in the blood. Glucose uptake was 12 g higher during exercise when carbohydrate was ingested, but, considering carbohydrate oxidation during both trials was 270 g, it is understandable that muscle metabolism and exercise capacity were unaffected by carbohydrate ingestion. Although glucose ingestion suppressed EGP during exercise, it remained significantly above resting levels throughout exercise. Interestingly, the muscle concentrations of PCr, lactate, and IMP were similar at 32 min of exercise and at exhaustion (∼70 min) in both trials. This lack of change in muscle metabolites suggests that exercise capacity during exercise at 80–85% V\(\text{O}_2\) max may have been limited by factors other than muscle energy supply.

Only ∼25% of the ingested glucose appeared in the blood during exercise in CHO. Studies utilizing more prolonged exercise at lower intensities (50–70% V\(\text{O}_2\) max) have also found, although to a lesser extent, that a significant proportion of carbohydrate ingested during prolonged exercise remains unaccounted for (14, 24). For example, our laboratory found during exercise at 70% V\(\text{O}_2\) max that only 34% of 200 g of glucose ingested during 120 min of exercise appeared in the blood (24). Although the reason for this “disappearance” of ingested carbohydrate is not entirely clear, it is likely that a portion of it remains in the gut or is absorbed and taken up first pass by the liver (14). Even allowing for the fact that the rate of glucose ingestion was a little less in the present study than our laboratory’s previous study (24) at 70% V\(\text{O}_2\) max (65 vs. 75 g ingested before 45 min of exercise), the exogenous glucose Ra in the present study was much lower than we observed at 70% V\(\text{O}_2\) max (25 ± 6 \(\mu\)mol·kg\(^{-1}\)·min\(^{-1}\) compared with 42 ± 3 \(\mu\)mol·kg\(^{-1}\)·min\(^{-1}\) at 45 min). This is possibly due to the fact that, as exercise intensity increases, rates of gastric emptying (5) and intestinal absorption decrease, as has been shown for water (23).

Glucose ingestion during exercise increased glucose uptake by 12 g (CHO: 45 ± 8 g; Con: 33 ± 8 g; Fig. 2C). This small but significant increase in glucose uptake could theoretically have been due to the observed higher plasma glucose (39) and insulin levels (8) or to the lower plasma NEFA (12) levels during exercise in
CHO (Fig. 1). Given that the glucose clearance rate was similar in the two trials (Table 4), it appears the higher glucose uptake in the CHO trial was due to the relative hyperglycemia in CHO. Indeed, a synergistic effect of plasma glucose and exercise on glucose uptake has been shown in exercising dogs when insulin is clamped at basal levels and plasma glucose is at physiological levels (6.7 mmol/l) (39).

Given that carbohydrate oxidation during both trials was 270 g, whereas glucose uptake was 45 g in the CHO trial, it is not surprising that muscle metabolism was unaffected by glucose ingestion. During exercise at 85% V\(\text{O}_2\) max, although the absolute rate of glucose uptake at the same time points is higher than at 70% V\(\text{O}_2\) max (Fig. 2; Ref. 24), the contribution of muscle glycogen to total energy yield is much larger than during exercise at the lower intensity (31). Just before exhaustion, blood glucose accounted for 18 ± 2% (0.9 ± 0.1 g/min) of energy needs in CHO compared with 12 ± 2% (0.6 ± 0.1 g/min) in Con. Late in prolonged exercise at 70% V\(\text{O}_2\) max when muscle glycogen levels are low, blood glucose can contribute up to 50% of energy needs when carbohydrate is ingested (6).

Late in prolonged exercise at 70% V\(\text{O}_2\) max, muscle glycogen depletion occurs and carbohydrate ingestion has been shown to attenuate falls in muscle TCAI (36) and increases in IMP contents (25, 36). During the more intense exercise of the present study, however, there was little evidence of an energy imbalance at the point of fatigue in either trial, because muscle IMP, lactate, and PCr contents were similar at fatigue and at 32 min of exercise (Fig. 3, Table 5). On the basis of the muscle and plasma lactate levels, which were generally maintained from 30 min of exercise until exhaustion, the rate of muscle glycogenolysis appeared to have remained high throughout exercise (Fig. 3). In addition, the rate of carbohydrate oxidation was well maintained in both trials throughout exercise, even approaching fatigue (Table 1). In contrast, at 70% V\(\text{O}_2\) max, muscle and blood lactate levels tend to decline late in exercise (e.g., Ref. 33), indicating a slowing of muscle glycolysis as muscle glycogen levels become depleted. All of the above suggests that exercise capacity was limited by factors other than muscle energy supply or carbohydrate availability in the present study.

On the basis of our findings, it is somewhat surprising that some studies have found improvements in exercise performance when carbohydrate is ingested during exercise at 80–85% V\(\text{O}_2\) max, lasting ~60 min (2, 17). It should be kept in mind, however, that carbohy-

### Table 4. Rate of gut glucose appearance and glucose clearance rate before and during exercise to exhaustion at 83 ± 2% \(\text{VO}_2\)peak during CHO and Con trials

<table>
<thead>
<tr>
<th>Time, min</th>
<th>0</th>
<th>17.5</th>
<th>35</th>
<th>45</th>
<th>Exhaustion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gut Ra, (\mu\text{mol}\cdot\text{kg}^{-1}\cdot\text{min}^{-1})</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Con</td>
<td>19.2 ± 7.0</td>
<td>24.6 ± 5.7</td>
<td>25.2 ± 5.0</td>
<td>25.9 ± 5.6</td>
<td></td>
</tr>
<tr>
<td>CHO</td>
<td>24.6 ± 5.7</td>
<td>30.2 ± 6.5</td>
<td>32.0 ± 6.8</td>
<td>32.8 ± 7.0</td>
<td></td>
</tr>
<tr>
<td>Clearance rate, (\text{ml} \cdot \text{min}^{-1} \cdot \text{kg}^{-1})</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Con</td>
<td>2.5 ± 0.2</td>
<td>5.8 ± 0.7</td>
<td>6.7 ± 0.8</td>
<td>7.8 ± 1.0</td>
<td>8.2 ± 1.7</td>
</tr>
<tr>
<td>CHO</td>
<td>2.6 ± 0.1</td>
<td>6.3 ± 0.7</td>
<td>6.8 ± 0.5</td>
<td>8.4 ± 0.9</td>
<td>8.9 ± 1.0</td>
</tr>
</tbody>
</table>

Values are mean ± SE; \(n = 6\) (subjects involved in glucose kinetics facet of the study). Gut Ra, rate of gut glucose appearance; clearance rate, glucose clearance rate.
deed, it has been shown that carbohydrate ingestion cannot be fully overcome by feedback regulators. In-feed-forward regulation of liver glucose output that suppress EGP to resting levels during exercise. Our studies show that carbohydrate ingestion (19, 24) and infusion (16) at 50–70% \( V\dot{O}_2 \) max in humans have shown that infusion of glucose at a rate equal to EGP in a control trial was unable to fully suppress EGP (38). Studies at lower intensities (50–70% \( V\dot{O}_2 \) max) in humans have shown that carbohydrate ingestion (19, 24) and infusion (16) suppress EGP to resting levels during exercise. Our data lend some support to the theory (21, 22, 34) that, during intense exercise in humans, there is strong feed-forward regulation of liver glucose output that cannot be fully overcome by feedback regulators. Indeed, it has been shown that carbohydrate ingestion may have nonmetabolic effects that enhance exercise performance such as via alterations in central nervous system function (7). The studies that have reported improvements in exercise performance during exercise lasting \( \sim 1 \) h have used time trial-type protocols (2, 17). The present study utilized a time to exhaustion at a set workload protocol to enable meaningful comparisons between trials. It is possible that a time trial-type protocol may have picked up small differences in exercise performance between the CHO and Con trials because the coefficient of variation is much smaller with these protocols (\( \sim 3\% \)) than time-to-exhaustion protocols (18). However, it should be noted that the coefficient of variation for the time to exhaustion in the 13 subjects in the present study (12.1%) was much less than the 26.6% presented by Jeukendrup et al. (18) in subjects cycling at a similar intensity as those in the present study. Finally, it may be suggested that the invasive nature (e.g., muscle biopsies) of the present study may have affected the subjects’ performance such that any small benefit of carbohydrate ingestion may have been missed. However, carbohydrate ingestion did not increase time to exhaustion in the seven subjects who did not have muscle biopsies (CHO: 64.7 \( \pm \) 5.4 min, Con: 66.2 \( \pm \) 6.1 min).

In the present study, EGP increased to a similar extent during the first 17.5 min of exercise in both trials (Fig. 2B), despite significantly higher plasma glucose levels in the CHO trial early in exercise (Fig. 1A). At 22.5 min of exercise, in the face of even higher plasma glucose levels and increased plasma insulin levels in the CHO than the Con trial, EGP decreased in the CHO trial such that it was significantly lower than the Con trial but remained significantly elevated above the preexercise level throughout exercise (Fig. 2B). Similar findings have been observed in rats during exercise at \( \sim 85\% \) \( V\dot{O}_2 \) max during which infusion of glucose at a rate equal to EGP in a control trial was unable to fully suppress EGP (38). Studies at lower intensities (50–70% \( V\dot{O}_2 \) max) in humans have shown that carbohydrate ingestion (19, 24) and infusion (16) suppress EGP to resting levels during exercise. Our data lend some support to the theory (21, 22, 34) that, during intense exercise in humans, there is strong feed-forward regulation of liver glucose output that cannot be fully overcome by feedback regulators. Indeed, it has been shown that carbohydrate ingestion does not suppress EGP during exercise at 70% \( V\dot{O}_2 \) max in the heat, a manipulation that increases the relative intensity of the exercise (11).

It has been suggested that EGP is regulated during intense exercise in humans by catecholamines to a greater extent than by pancreatic hormones (21, 22, 34). In the present study, plasma epinephrine and norepinephrine concentrations during exercise were similar in the two trials (Table 2) and were much higher than what our laboratory previously observed during exercise at 70% \( V\dot{O}_2 \) max (24). Manzon et al. (21) found a close correlation between EGP and catecholamines when glucose was infused during exercise at \( >85\% \) \( V\dot{O}_2 \) max, and Sigal et al. (34) also found a close correlation between EGP and catecholamines during exercise at 87% \( V\dot{O}_2 \) max when an islet clamp was put in place. Although we suggest that our results indicate that feed-forward regulation of EGP is important during intense exercise, an alternative interpretation may be that EGP remained above resting levels throughout exercise in CHO because there was an insufficient rate of exogenous glucose absorption (Table 4). As was mentioned above, in the present study the rate of delivery of exogenous glucose was lower during exercise than what our laboratory observed at 70% \( V\dot{O}_2 \) max (Table 4; Ref. 24). During exercise at 70% \( V\dot{O}_2 \) max, we found that gut \( R_L \) in CHO was similar to the rate of EGP in the Con trial (24), but in the present study, gut \( R_L \) in the CHO trial (Table 4) was less than EGP in the Con trial (Fig. 2B). It is possible that this was the reason EGP in CHO was not suppressed to resting levels in the present study. Indeed, infusion of glucose at a rate sufficient to cause significant hyperglycemia and hyperinsulinemia by 10 min of exercise in humans exercising at 77% \( V\dot{O}_2 \) max prevents EGP rising above resting levels (15).

In conclusion, glucose ingestion during intense endurance exercise at 83% \( V\dot{O}_2 \) peak in trained men raised plasma glucose levels and increased glucose uptake. Glucose ingestion was able to only partially attenuate the increases in endogenous glucose production during exercise. Interestingly, only \( \sim 25\% \) (22 g) of the ingested glucose appeared in the peripheral blood. Glucose ingestion had no effect on carbohydrate oxidation, muscle metabolism, or exercise capacity. Because muscle PCr, IMP and lactate were unchanged from 32 min of exercise until exhaustion (\( \sim 70 \) min) in both trials, it
is likely that exhaustion occurred because of factors other than metabolic energy supply.

We thank the subjects who took part in this study for valiantly complying with a very tough protocol and Kathy McConell for dietary advice and analysis.

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REFERENCES


