Glucose uptake is increased in trained vs. untrained muscle during heavy exercise

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Kristiansen, Soren, Jon Gade, Jørgen F. P. Wojtaszewski, Bente Kiens, and Erik A. Richter. Glucose uptake is increased in trained vs. untrained muscle during heavy exercise. J Appl Physiol 89: 1151–1158, 2000.—Endurance training increases muscle content of glucose transporter proteins (GLUT-4) but decreases glucose utilization during exercise at a given absolute submaximal intensity. We hypothesized that glucose uptake might be higher in trained vs. untrained muscle during heavy exercise in the glycogen-depleted state. Eight untrained subjects endurance trained one thigh for 3 wk using a knee-extensor ergometer. The subjects then performed two-legged glycolgen-depleting exercise and consumed a carbohydrate-free meal thereafter to keep muscle glycogen concentration low. The next morning, subjects performed dynamic knee extensions with both thighs simultaneously at 60, 80, and until exhaustion at 100% of each thigh’s peak workload. Glucose uptake was similar in both thighs during exercise at 60% of thigh peak workload. At the end of 80 and at 100% of peak workload, glucose uptake was on average 33 and 22% higher, respectively, in trained compared with untrained muscle (P < 0.05). Training increased the muscle content of GLUT-4 by 66% (P < 0.05). At exhaustion, glucose extraction correlated significantly (r = 0.61) with total muscle GLUT-4 protein. Thus, when working at a high load with low glycogen concentrations, muscle glucose uptake is significantly higher in trained than in untrained muscle. This may be due to the higher GLUT-4 protein content in trained muscle.

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their maximal pulmonary \( V_2 \) uptake, \( \text{VO}_2 \text{max} \), as measured during incremental cycling on a bicycle ergometer, was below 52 ml·min \(^{-1}\)·kg body wt \(^{-1}\). Average maximal \( V_2 \) uptake was 48 \pm 2 (SE) ml·min \(^{-1}\)·kg body wt \(^{-1}\).

**Experimental design.** Initially, a muscle biopsy was obtained from the vastus lateralis muscle in each leg, followed by a supervised endurance training program for 3 wk. One thigh was used for the training protocol (see Training) using the knee-extensor model (2), which allows dynamic exercise to be performed exclusively with the knee extensors. The other thigh served as an untrained control. Twenty-four hours after the last training session, the subjects performed an exercise bout on a bicycle ergometer. The work consisted of 20 min with continuous work at 75% of pulmonary \( V_2 \text{O}_2 \text{max} \), followed by a period of intermittent exercise, with a change of the workload every 1.5 min between 50 and 90% of \( V_2 \text{O}_2 \text{max} \) until the subject was unable to maintain a frequency at 75 rpm for a period longer than \( \sim 10 \) s. Thereafter, the high workload was diminished to 85% of \( V_2 \text{O}_2 \text{max} \) and so on until the workload changed between 50 and 60% of \( V_2 \text{O}_2 \text{max} \). The workload was terminated with 1.5 min of all-out sprint. The exercise test primarily activates the vastus lateralis and the rectus femoris muscles, and the purpose of the work was to empty the muscle glycogen in the whole spectrum of muscle fibers. After the exercise test (performed in the period 4–6 PM), the subjects were given a diet rich in fat to maintain the low muscle glycogen concentrations. The energy composition of the diet was 74 \pm 1% fat, 25 \pm 2% protein, and 1 \pm 0% carbohydrate, and the average energy intake was 10,412 \pm 526 kJ. The next morning, after having fasted overnight, the subjects arrived at the laboratory at 8:30 AM by bus, train, or car. After the subjects rested for 30 min in the supine position, Teflon catheters were placed in both femoral veins and one femoral artery under local anesthesia with use of aseptic techniques, and the tips of the catheters were advanced to \( \sim 2 \) cm below and above the inguinal ligament, respectively. For measuring venous blood temperature, a thermistor probe (Edslab T.D. probe 94-030-2.5-F, Baxter Healthcare) was inserted through each venous catheter and advanced 8 cm proximal to the catheter tip. After placement of the catheters, the subjects were taken to the experiment room, where they rested for \( \sim 1 \) h in the supine position. Then, resting blood samples were obtained from the three catheters simultaneously, and femoral venous blood flow was measured by the thermodilution method by use of bolus injections of 3-ml ice-cold sterile saline (2). Whenever blood was sampled or blood flow was measured, pneumatic cuffs below the knees were inflated to 230 mmHg to exclude circulation to the lower leg. Muscle biopsies were obtained from each vastus lateralis muscle under local anesthesia with lidocaine. Then, the subjects commenced two-legged dynamic knee extensions with a frequency of 60 extensions/min. Each leg was connected to a separate knee-extension ergometer, and each leg worked simultaneously at the same relative power output. Subjects were asked to rate perceived exertion in the two thighs according to the Borg scale, which uses a scale from 6 to 20 (4). The power output was adjusted slightly (by 2 W), if necessary, during exercise so that perceived exertion was rated equally in the two thighs to ensure exhaustion in both thighs at the same time. The power output was first set to 30 min at 60% of peak workload (PWL) for each thigh (27 \pm 1 and 34 \pm 1 W in UT and T, respectively), then to 80% of PWL (36 \pm 2 and 45 \pm 2 W in UT and T, respectively) for 20 min, and then to 100% of PWL (45 \pm 2 and 56 \pm 2 W in UT and T, respectively). At 100% of PWL, subjects exercised to complete exhaustion, which occurred at the same time in both legs (9 \pm 1 min). After exhaustion, muscle biopsies were immediately obtained from each thigh. Arterial and bilateral femoral venous blood was sampled simultaneously from the three catheters at rest, during exercise every 10 min, and at exhaustion. Bilateral femoral venous blood flow was measured during exercise immediately before blood sampling by use of constant infusion of ice-cold saline according to the thermodilution principle (2). Expiratory air was collected through a mouthpiece in Douglas bags at the end of every work period, and heart rate was measured continuously via chest electrodes.

**Training.** Subjects were accustomed to the one-legged dynamic knee-extensor apparatus with both legs before an incremental knee-extensor test was performed on each leg to determine the peak workload of the knee extensors. Pulmonary \( O_2 \) uptake was measured, and PWL for the knee extensors was defined as the workload when the initial linear relationship between workload and pulmonary \( O_2 \) uptake changed to an exponential one, indicating the recruitment of accessory muscles to stabilize the body at high workloads. Subjects were included in the study only if the PWL of the two knee extensors differed by \( \leq 5\% \). One subjects completed one training program of one thigh. Four subjects trained the dominant thigh and four subjects trained the nondominant thigh. The training program consisted of four sessions the first week, five sessions the second week, and six sessions the third week. Duration of exercise was gradually increased from 1 to 2 h per session by the end of the second week. Exercise workload was varied between 70 and 85% of pretraining PWL, and, for the last 5–10 min of every session, workload was increased to 100–110% of pretraining PWL to ensure recruitment of most of the muscle fibers (17). During the training period, subjects also practiced three times for 15 min, during which both thighs worked simultaneously at the same relative power output. In the beginning of the last exercise session, the subjects repeated the test for measuring the PWL on the trained leg to determine the relative workload in the final experimental exercise test. Our laboratory has recently shown that the PWL for the untrained leg is unchanged after an identical training period (39).

**Assays.** A small part of each muscle biopsy sample was quick frozen in liquid nitrogen and stored at \(-80^\circ\)C. Later, these muscle pieces were freeze-dried; dissected free of fat, blood, and connective tissues; and used for assay of GLUT-4 protein content through Western blotting. The GLUT-4 antibody was a goat polyclonal antibody produced against a synthetic peptide corresponding to the 13 COOH-terminal amino acids of GLUT-4. Antibody-antigen complexes were visualized within the linear response with an enhanced chemiluminescence detection kit (Amersham, Arlington Heights, IL). To quantify the signal, densitometric scanning was performed (Kem-En-Tec Software Systems, Copenhagen, Denmark). The GLUT-4 protein content per microgram protein was expressed in arbitrary units relative to a rat heart standard. Muscle samples were also analyzed for proglycogen, macroglycogen, and total muscle glycogen content with the method described by Adamo et al. (1). In short, a frozen muscle piece (10 mg) was immersed in 200 \( \mu \)l of ice-cooled 1.5 M perchloric acid (PCA) and was pressed against the plastic tubes with a glass rod to ensure that all the muscle was exposed to acid. The extraction continued on ice for 20 min. The samples were centrifuged at 3,000 rpm for 15 min, after which 100 \( \mu \)l of the PCA supernatant was separated for the determination of macroglycogen. The remaining PCA was discarded, and the pellet was kept for the determination of proglycogen. One milliliter of 1 M HCl was added to the macroglycogen and to the proglycogen samples,
which were boiled for 2 h. The samples were then neutralized with 2 M Tris base, vortexed, and centrifuged at 3,000 rpm for 5 min, and the supernatants were fluorometrically measured for glucosyl units (34). The rest of the muscle was also assayed for citrate synthase (CS), 3-hydroxyacyl-CoA dehydrogenase (HAD), glucose, and glucose 6-phosphate (G-6-P) by using standard enzymatic methods (34).

Blood glucose and lactate were measured with a Yellow Springs Instruments analyzer (Yellow Springs, OH). Hemoglobin concentration in blood and O₂ saturation of hemoglobin was measured with an OSM-3 hemoximeter (Radiometer, Copenhagen, Denmark). Expired air was analyzed for O₂ concentration by a Servomex paramagnetic analyzer and for CO₂ by a Beckman infrared CO₂ analyzer. Volume of expiratory air was measured with a Tissot-type spirometer.

Statistics. Values measured more than twice in each leg during exercise were compared with a two-way analysis of variance for repeated measures. The Student-Newman-Keuls test was used as a post hoc test. Values measured twice in each leg (before and after training) were compared with the paired Student's t-test. A significance level of 0.05 was chosen.

RESULTS

Muscle adaptations to training. The total muscle GLUT-4 protein content was identical in the two legs before training (Table 1). Training increased the GLUT-4 protein content by 66% in the T leg, whereas no significant change occurred in the UT leg (Table 1). Similarly, activities of CS and HAD were increased by training, whereas no change occurred in UT-muscle (Table 1). The PWL output during knee-extensor exercise in the T leg increased from 45 ± 2 W before training to 56 ± 2 W after training (P < 0.05). The pretraining values were 45 ± 2 W in the UT leg. According to a previous study from our laboratory, PWL does not change in the UT leg during training of the T leg (39).

Whole body response to two-legged kicking. Whole body O₂ uptake increased from 1.5 to 1.7 to 2.1 l/min going from 60 to 80 and to 100% of leg PWL. Average heart rate increased from 135 to 154 to 173 beats/min at 60, 80, and 100% of PWL, respectively. Perceived exertion was identical in the two legs, averaging 13 ± 0.3, 16 ± 0.3, and 20 ± 0.2 at 60, 80, and 100% of PWL, respectively.

Thigh and muscle response to exercise. Glucose uptake was similar in the two thighs at rest and increased similarly in the two thighs at 60% PWL (Fig. 1). When both thighs were considered, glucose uptake increased by 10.2 ± 3.3.

Table 1. GLUT-4 protein, citrate synthase and 3-hydroxy-acyl-CoA dehydrogenase

<table>
<thead>
<tr>
<th></th>
<th>Before Training</th>
<th>After Training</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>UT</td>
<td>T</td>
</tr>
<tr>
<td>TCM GLUT-4, relative units</td>
<td>0.32 ± 0.03</td>
<td>0.32 ± 0.03</td>
</tr>
<tr>
<td>CS, μmol·g dry wt⁻¹·min⁻¹</td>
<td>39 ± 2</td>
<td>40 ± 3</td>
</tr>
<tr>
<td>HAD, μmol·g dry wt⁻¹·min⁻¹</td>
<td>56 ± 4</td>
<td>56 ± 5</td>
</tr>
</tbody>
</table>

Values are means ± SE of 8 determinations. Total crude membrane (TCM) GLUT-4 protein content, citrate synthase (CS), and 3-hydroxyacyl-CoA dehydrogenase (HAD) activities in vastus lateralis muscle at rest in the untrained (UT) and trained (T) leg before and after 3 wk of 1-legged endurance training are presented. TCM GLUT-4 values are given per μg protein in units relative to a rat heart standard. *P < 0.05 compared with T values before training.

Fig. 1. Time course of glucose uptake in the trained leg and untrained leg while the subjects were at rest and when exercising at 60, 80, and 100% of the peak workload determined after 3 wk of training. Peak workload of 60, 80, and 100% corresponded to 27 ± 1, 36 ± 2 and 45 ± 2 W in the untrained thigh and 34 ± 1, 45 ± 2 and 56 ± 2 W in the trained thigh. Values are means ± SE for 8 subjects. *P < 0.05 compared with the untrained leg.
at 80% of PWL was not significantly higher than at 60%, but it increased significantly at 100% PWL compared with both 60 and 80% of PWL (Fig. 1). The glucose uptake was significantly higher in the T leg than in the UT leg after 20 min at 80% of PWL and also at 100% of PWL (Fig. 1). Although the intensity of exercise is at three different levels, it should also be noted that there is a time effect on the data. Glucose uptake is the product of arteriovenous difference (glucose extraction) and blood flow. Differences in glucose uptake between the T and UT legs could, therefore, be due to a difference in glucose extraction, in blood flow, or in both. Although clear tendencies toward increased extraction as well as blood flow in the T leg were observed, the differences did not reach statistical significance in either case (Table 2). Thigh blood flow generally increased with increasing workload (Table 2), as did thigh O\textsubscript{2} uptake (Table 2). Although O\textsubscript{2} uptake tended to be higher in the T compared with the UT leg, the differences did not reach significance (Table 2).

Lactate release was low at rest and during 60% PWL in both thighs (Fig. 2). At 80% PWL, the T thigh released some lactate, whereas this was not the case with the UT thigh. Moreover, lactate release increased sharply from the T leg at 100% PWL, whereas the UT leg neither released nor took up lactate (Fig. 2). Arterial blood lactate concentration was 0.5 ± 0.1 mmol/l at rest and increased to 0.7 ± 0.1 mmol/l at the end of 60% of PWL, to 1.5 ± 0.1 mmol/l at the end of 80% of PWL, and to 3.6 ± 0.4 mmol/l at the end of exercise.

Although subjects exercised to complete exhaustion in the afternoon and ingested a virtually carbohydrate-free diet in the evening before the actual experiment, the muscle glycogen concentration before the experiment was only moderately, but significantly, reduced compared with resting pretraining values in both legs (Table 3). Furthermore, the glycogen concentration be-

### Table 2. Glucose extraction (arteriovenous difference), thigh blood flow, and thigh O\textsubscript{2} uptake at rest and during 2-legged knee extensions in the untrained and trained thigh

<table>
<thead>
<tr>
<th>Workload</th>
<th>Rest</th>
<th>60% 10 min</th>
<th>20 min</th>
<th>30 min</th>
<th>40 min</th>
<th>50 min</th>
<th>59 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arteriovenous glucose, mmol/l</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UT</td>
<td>0.07 ± 0.02</td>
<td>0.11 ± 0.01</td>
<td>0.20 ± 0.03</td>
<td>0.19 ± 0.02</td>
<td>0.22 ± 0.02</td>
<td>0.21 ± 0.02</td>
<td>0.22 ± 0.03</td>
</tr>
<tr>
<td>T</td>
<td>0.07 ± 0.03</td>
<td>0.10 ± 0.02</td>
<td>0.18 ± 0.03</td>
<td>0.21 ± 0.03</td>
<td>0.25 ± 0.04</td>
<td>0.25 ± 0.03</td>
<td>0.26 ± 0.03</td>
</tr>
<tr>
<td>Blood flow, l/min</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UT</td>
<td>0.3 ± 0.1</td>
<td>4.5 ± 0.4</td>
<td>4.5 ± 0.3</td>
<td>4.4 ± 0.3</td>
<td>4.9 ± 0.4</td>
<td>4.7 ± 0.3</td>
<td>6.2 ± 0.6</td>
</tr>
<tr>
<td>T</td>
<td>0.3 ± 0.1</td>
<td>5.1 ± 0.3</td>
<td>4.7 ± 0.3</td>
<td>4.5 ± 0.3</td>
<td>5.2 ± 0.4</td>
<td>5.3 ± 0.4</td>
<td>6.4 ± 0.4</td>
</tr>
<tr>
<td>O\textsubscript{2} uptake, l/min</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UT</td>
<td>0.01 ± 0.00</td>
<td>0.52 ± 0.04</td>
<td>0.52 ± 0.04</td>
<td>0.52 ± 0.04</td>
<td>0.60 ± 0.05</td>
<td>0.59 ± 0.04</td>
<td>0.82 ± 0.09</td>
</tr>
<tr>
<td>T</td>
<td>0.01 ± 0.00</td>
<td>0.62 ± 0.04</td>
<td>0.58 ± 0.04</td>
<td>0.56 ± 0.04</td>
<td>0.66 ± 0.03</td>
<td>0.67 ± 0.05</td>
<td>0.89 ± 0.04</td>
</tr>
</tbody>
</table>

Values are means ± SE.

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Fig. 2. Time course of lactate flux in the trained leg and untrained leg while the subjects were at rest and when exercising at 60, 80, and 100% of the peak workload determined after 3 wk of training. Values are means ± SE for 8 subjects. *P < 0.05 compared with the untrained leg.
Therefore the experiment was higher \((P < 0.05)\) in the T muscle compared with the UT muscle. This was true for both pro- and macroglycogen and for the total muscle glycogen concentration. During exercise, glycogen breakdown was similar in UT and T muscle, and, at exhaustion, glycogen content was, consequently, significantly lower in UT than in T muscle (Table 3). In UT muscle, the content was, in fact, extremely low, and macroglycogen was virtually absent (Table 3). During exercise, glycogen content was, consequently, significantly lower in UT than in T muscle (Table 3). In UT muscle before training and after exercise.

**DISCUSSION**

In the present study, for the first time we demonstrate that glucose uptake is higher in T than in UT human skeletal muscle when working at the same high relative workload (same percentage of PWL) and with low glycogen concentrations. Because exercise was performed with the T and UT limb simultaneously, adaptations within the muscles are likely responsible for this increase in glucose utilization. Our results suggest that one such factor may be increased muscle GLUT-4 protein content.

Our laboratory (39) and others (3, 6, 15, 16) have previously shown that, when a given absolute submaximal power output is performed, endurance training leads to lower glucose uptake in muscle in the trained compared with the untrained state. We demonstrated that this reduction in glucose uptake in trained muscle was due to a smaller exercise-induced increase in sarcolemmal glucose transport capacity and sarcolemmal GLUT-4 protein content in trained compared with untrained muscle (39). Several other studies have compared trained and untrained individuals at the same relative submaximal exercise intensity, and found similar glucose uptake (22, 40), whereas some found a small decrease in the trained condition (7, 27, 41). Our study is the first to report that glucose uptake may actually be higher in trained muscle when working at the same relative exercise intensity at 80 and 100% of PWL and furthermore that glucose extraction during exercise at the highest workload correlated significantly with muscle GLUT-4 protein content. Thus our findings during exercise parallel observations showing a correlation between muscle GLUT-4 protein content and maximal insulin-stimulated muscle glucose uptake (9, 38). The reason that a relationship between muscle GLUT-4 and glucose uptake during exercise has not been described before may be due to the fact that only low-to-moderate, submaximal exercise intensities have been studied previously. In fact, a negative correlation between muscle GLUT-4 and whole body rate of disappearance of glucose has been described at the end of 40 min of bicycle ergometer cycling at 72% of \(\text{VO}_{2\text{max}}\) (35). At first glance, it is not readily apparent why our results and those of McConnell et al. (35) are so contradictory. However, in contrast to McConnell et al., in the present study, we investigated subjects during exercise that supposedly would result in maximal rates of glucose utilization. It is conceivable that the high muscle GLUT-4 protein content and the consequent potential for high glucose transport in T muscle is only utilized during such circumstances. Furthermore, in rat skeletal muscle, it has been shown that contraction-induced glucose transport is higher in red muscle with a high expression of GLUT-4 than in white muscle with a low GLUT-4 expression (21), thus supporting the view that muscle GLUT-4 protein content is positively correlated with maximal contraction-induced glucose transport.

The present study is also different from previous studies comparing trained and untrained subjects at the same relative exercise intensity because it excludes humoral differences between trained and untrained trials because the UT and T muscles were studied simultaneously. This has the advantage that the composition of the blood perfusing the T and UT muscles is

**Table 3. Proglycogen, macroglycogen, and total muscle glycogen content in resting untrained and trained muscle before and after training and after exhaustive 2-legged knee-extensor exercise**

<table>
<thead>
<tr>
<th></th>
<th>Before Training</th>
<th>Before Exercise</th>
<th>After Exercise</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Total glycogen, (\mu\text{mol/g dry wt})</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UT</td>
<td>485 ± 27</td>
<td>227 ± 17†</td>
<td>54 ± 12†</td>
</tr>
<tr>
<td>T</td>
<td>524 ± 21</td>
<td>347 ± 33†</td>
<td>190 ± 42†</td>
</tr>
<tr>
<td><strong>Proglycogen, (\mu\text{mol/g dry wt})</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UT</td>
<td>334 ± 19</td>
<td>179 ± 11†</td>
<td>45 ± 10†</td>
</tr>
<tr>
<td>T</td>
<td>327 ± 9</td>
<td>226 ± 14†</td>
<td>138 ± 26†</td>
</tr>
<tr>
<td><strong>Macroglycogen, (\mu\text{mol/g dry wt})</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UT</td>
<td>152 ± 12</td>
<td>49 ± 9†</td>
<td>8 ± 2†</td>
</tr>
<tr>
<td>T</td>
<td>197 ± 20</td>
<td>120 ± 20*†</td>
<td>52 ± 17*†</td>
</tr>
</tbody>
</table>

Values are means ± SE. *\(P < 0.05\) compared with UT values. †\(P < 0.05\) compared with values before training or before exercise.

**Table 4. Concentration of muscle glucose and glucose 6-phosphate in the untrained and trained leg at rest and at exhaustion**

<table>
<thead>
<tr>
<th></th>
<th>Rest Untrained</th>
<th>Rest Trained</th>
<th>Exhaustion Untrained</th>
<th>Exhaustion Trained</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Glucose, (\text{mmol/kg dry wt})</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>2.80 ± 0.28</td>
<td>2.67 ± 0.28</td>
<td>3.32 ± 0.54</td>
<td>7.34 ± 1.54†</td>
</tr>
<tr>
<td><strong>Glucose 6-phosphate, (\text{mmol/kg dry wt})</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.30 ± 0.06</td>
<td>0.31 ± 0.05</td>
<td>0.34 ± 0.06</td>
<td>0.81 ± 0.40*</td>
</tr>
</tbody>
</table>

*\(P < 0.05\) compared to the values in the untrained leg. †\(P < 0.05\) compared to resting values. 
identical, and, therefore, training-induced alterations in hormone responses and blood substrate levels can be excluded as confounders of the muscle response to exercise. Furthermore, we attempted to minimize differences in muscle glycogen levels between T and UT muscle by having the subjects perform a glycogen-depletion trial the day before the actual experiment. Although we were not completely successful in eliminating differences in muscle glycogen content between T and UT muscle, they were rather small (∼100 μmol/g dry wt before exercise). Still, muscle glycogen concentration was, at exhaustion, significantly higher in the T than in the UT leg, and, judging from lactate release, significant glycogenolysis was only occurring in the T leg at exhaustion (Fig. 2). This interpretation is supported by the higher glucose and G-6-P values in the T vs. UT muscle at exhaustion (Table 4). The latter values also suggest that glucose phosphorylation was limiting glucose uptake in the T and not in the UT muscle. This again leads to the speculation that glucose uptake might have been even higher in the T muscle had the glycogen levels been as low in the T as in the UT muscle at exhaustion. This interpretation is further supported by findings in perfused rat skeletal muscle, in which glucose uptake, glucose transport, and GLUT-4 translocation during contractions have been shown to increase less when muscle glycogen concentrations are high compared with when they are low (11, 23).

The increase in glucose uptake by exercise is, at least in part, mediated by translocation of GLUT-4-containing vesicles to sarcolemmal membranes (31, 39). The exercise-sensitive signaling pathway responsible for initiating the translocation of the GLUT-4 transporters is not well known. Recent reports (20, 26, 36) suggest that the 5′-activated AMP kinase (AMPK) is activated by muscle contractions and may be part of the pathway leading to GLUT-4 protein translocation. It should, however, also be noted that there is evidence against a significant role for AMPK activation in stimulating glucose transport in muscle (10). Whatever the molecular nature of the exercise-induced mechanism to increase muscle glucose transport is, it is probably sensitive to the energy status of the muscle cell (26). Because it has been shown that the decrease in muscle energy charge is dependent on the relative exercise intensity (25), it is reasonable to assume that, in the present study, energy status was similar in T and UT muscle during exercise of the same relative intensity. If so, the exercise-induced molecular signal to GLUT-4 translocation may also be similar in the two muscles. Combined with the larger GLUT-4 pool in T than in UT muscle, this might explain the larger glucose uptake in T vs. UT muscle during the strenuous exercise performed in the present study. In support of this notion is the significant correlation between glucose extraction and muscle GLUT-4 protein content (Fig. 3).

Biochemical and electron microscopic studies have suggested that glycogen exists in two forms in skeletal muscle (14, 32, 33). One form is macroglycogen, the mature glycogen particle (10,000 kDa). The other is proglycogen, a smaller intermediate in glycogen synthesis with a molecular mass traditionally said to be ∼400 kDa, although evidence has been presented that proglycogen molecular mass may, in fact, reach close to 1,000 kDa when muscle glycogen concentration is high (19). In resting muscle with normal glycogen content, only a minor fraction (∼20%) of the total glycogen pool is found in the macroglycogen form. In glycogen-supercompensated muscle, this macroglycogen fraction may increase to 50% or more, and it becomes the dominant form, whereas, in glycogen-depleted muscle, very little glycogen (∼10%) is in the macroglycogen form (19). In the present study, the higher glycogen content in the T leg was due to a higher pro- as well as macroglycogen content (Table 3). The present study shows that both pro- and macroglycogen are degraded during dynamic exercise but that UT muscle, on average, utilized more proglycogen (134 μmol/g dry wt) than T muscle (88 μmol/g dry wt), perhaps because the macroglycogen level was quite low in the UT muscle before exercise (49 ± 9 vs. 120 ± 20 μmol/g dry wt in UT vs. T muscle, respectively; P < 0.05).

In rat skeletal muscle, exercise training is, in some cases, paralleled by an increase in contraction-stimulated glucose transport (13, 28, 37). It has also been found that more tetanic contractions are required to maximally activate glucose transport in T than in UT rat skeletal muscle, possibly due to the training-induced increase in muscle glycogen levels (29). In accordance with these animal studies, the present study shows that higher glucose uptake can be reached in T

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Fig. 3. A: correlation between total muscle GLUT-4 protein content in untrained and trained muscle (○ and ●, respectively) and glucose arteriovenous (a-v) difference when subjects were exercising at 100% of the peak workload. B: correlation between total muscle GLUT-4 protein content in the untrained and trained muscle (○ and ●, respectively) and leg glucose uptake when subjects were exercising at 100% of the peak workload.
vs. UT human skeletal muscle when a protocol involving low glycogen levels and intense exercise is used.

In conclusion, glucose uptake is higher in T than in UT human skeletal muscle when working at the same high relative workload and with low glycogen concentrations. This difference may be related to the higher muscle GLUT-4 protein content in T vs. UT muscle. Such superior glucose utilization capacity in T muscle probably explains why trained subjects are able to utilize glucose at remarkably high rates when fed carbohydrates or infused with glucose during prolonged exercise when glycogen stores are low (5, 8).

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