Muscle Metabolism and Fatigue during Simulated Ice Hockey Match-Play in Elite Players

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ABSTRACT

VIGH-LARSEN, J. F., G. ERMIDIS, V. RAGO, M. B. RANDERS, D. FRANSSON, J. L. NIELSEN, L. GLIEMANN, J. F. PIIL, N. B. MORRIS, F. V. DE PAOLI, K. OVERGAARD, T. B. ANDERSEN, L. NYBO, P. KRUSTRUP, AND M. MOHR. Muscle Metabolism and Fatigue during Simulated Ice Hockey Match-Play in Elite Players. *Med. Sci. Sports Exerc.*, Vol. 52, No. 10, pp. 2162–2171, 2020. **Purpose:** The present study investigated muscle metabolism and fatigue during simulated elite male ice hockey match-play. **Methods:** Thirty U20 male national team players completed an experimental game comprising three periods of 8 × 1-min shifts separated by 2-min recovery intervals. Two vastus lateralis biopsies were obtained either during the game ($n = 7$) or pregame and postgame ($n = 6$). Venous blood samples were drawn pregame and at the end of the first and last periods ($n = 14$). Activity pattern and physiological responses were continuously monitored using local positioning system and heart rate recordings. Further, repeated-sprint ability was tested pregame and after each period. **Results:** Total distance covered was 5980 ± 199 m with almost half the distance covered at high skating speeds (>17 km·h^{-1}). Average and peak on-ice heart rate was 84% ± 2% and 97% ± 2% of maximum heart rate, respectively. Muscle lactate increased ($P \leq 0.05$) more than fivefold and threefold, whereas muscle pH decreased ($P \leq 0.05$) from 7.31 ± 0.04 pregame to 6.99 ± 0.07 and 7.13 ± 0.11 during the first and last periods, respectively. Muscle glycogen decreased by 53% postgame ($P \leq 0.05$) with ~65% of fast- and slow-twitch fibers depleted of glycogen. Blood lactate increased sixfold ($P \leq 0.05$), whereas plasma free fatty acid levels increased 1.5-fold and threefold ($P \leq 0.05$) after the first and last periods. Repeated-sprint ability was impaired (~3%; $P \leq 0.05$) postgame concomitant with a ~10% decrease in the number of accelerations and decelerations during the second and last periods ($P \leq 0.05$). **Conclusions:** Our findings demonstrate that a simulated ice hockey match-play scenario encompasses a high on-ice heart rate response and glycolytic loading resulting in a marked degradation of muscle glycogen, particularly in specific sub-groups of fibers. This may be of importance both for fatigue in the final stages of a game and for subsequent recovery. **Key Words:** GLYCOGEN, PERFORMANCE, HIGH-INTENSITY, INTERMITTENT EXERCISE, TEAM SPORT, FIBER-TYPE

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Muscle metabolism varies markedly between high-intensity work executed as continuous or intermittent exercise. Accordingly, glycolytic flux and phosphocreatine (PCr) degradation decrease, whereas oxidative phosphorylation increases during consecutive bouts of intense exercise (1,2). Thus, aerobic metabolism becomes progressively more important with repeated exercise bouts due to the importance of oxygen availability for PCr resynthesis and as a compensatory mechanism for the decrease in anaerobic energy turnover (1,2). Metabolic responses likewise differ between continuous and intermittent all-out exercise scenarios. During

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a Wingate test or 3-min all-out test, for example, muscle lactate can reach levels of >100 mmol·kg^{-1}·d.w., and muscle creatine phosphate stores become as low as <20 mmol·kg^{-1}·d.w. (1,3). In contrast, during an intermittent exhaustive running protocol muscle lactate and creatine phosphate levels reach ~70 and ~30 mmol·kg^{-1}·d.w. (4). Furthermore, these changes are markedly reduced during peak intensity periods in team sports like soccer compared with continuous all-out exercise (5). Hence, muscle metabolism is highly complex during high-intensity intermittent exercise, especially in team and racquet sports characterized by an irregular movement pattern.

Ice hockey is a team sport with a proposed high anaerobic component (6,7). This relates to the fast-paced activity pattern with intense shifts lasting 30 to 80 s interspersed by longer duration (~2–3 min) recovery (6,7). These exercise bouts are repeated continuously in a competitive setting resulting in a total playing time of 15 to 25 min per player (6,7). Recently, match activities quantified during a top-class competitive ice hockey game demonstrated that players accomplish nearly half the total distance in high-intensity skating, resulting in indications of performance decrement in the later stages of a game (8). Thus, knowledge about muscle metabolism and possible fatiguing mechanisms during ice hockey match-play is warranted.

Exercise tolerance during intense exercise has historically been linked to lactic acid and muscle acidosis, but more recently, several other factors have been suggested as more likely contributors to fatigue (9). These include accumulation of inorganic phosphate, altered Ca^{2+} release or reuptake or factors related to muscle excitability, such as extracellular K^+ accumulation, Cl^- channel regulation and production of reactive oxygen species (9–11). However, when high-intensity exercise is repeated over prolonged periods, as in most team sports, other factors may additionally contribute to impaired exercise tolerance, such as hyperthermia, dehydration, muscle damage, and substrate availability (5,12). Accordingly, improved simulated ice hockey performance has been demonstrated after the ingestion of a sports drink containing electrolytes and carbohydrate as well as after administration of a carbohydrate-enriched diet (13,14). Likewise, soccer match-play has been demonstrated to induce a significant decline in levels of muscle glycogen, associated with reduced sprint performance at the end of a game. This included a large amount of individual muscle fibers almost totally depleted of glycogen, with an equal utilization pattern for fast- and slow-twitch fibers, which may indirectly provoke muscle fatigue (5). However, the activity pattern in ice hockey, characterized by more brief and intense intermittent bursts of exercise than soccer, may lead to a different muscle metabolic response, muscle fiber recruitment pattern, and utilization of muscle glycogen. Despite this, early investigations in ice hockey reported predominant utilization of muscle glycogen in type I fibers after match play, but with some discrepancy between studies (15,16). Moreover, because these results were obtained more than 40 yr ago and in nonelite players, this may not be the case presently in elite ice hockey.

Thus, the main aim of the present study was to examine muscle metabolism and fatigue development during simulated ice hockey match-play in elite male players. An additional aim was to study the fiber type-specific utilization of muscle glycogen during this exercise scenario. We hypothesized that the fast-paced activity pattern would result in a significant anaerobic energy turnover in addition to a high cardiovascular loading, and that this would induce a significant decline in muscle glycogen in fast-twitch as well as slow-twitch fibers concomitant with performance decrements during the game.

MATERIALS AND METHODS

Participants. Thirty male ice hockey players from the Danish U20 national team took part in the study (mean ± SD: age, 19 ± 1 yr; height, 184.3 ± 6.2 cm; weight, 83.4 ± 8.8 kg; muscle mass, 42.9 ± 5.2 kg; fat percentage, 10.3% ± 0.7%; countermovement jump height, 42.0 ± 5.2 cm; Yo-Yo Intermittent Recovery Test Level 1 Ice Hockey submaximal heart rate, 78.5 ± 6.5%HR_{max}). Inclusion criteria were no recent history of injury or illness and participation at the national team level. The sample consisted of 18 forwards and 12 defensemen. The study adhered to the code of ethics of the Declaration of Helsinki and was approved by the local ethics committee (application number 1-10-72-249-18). Written informed consent was obtained from all players before the study and no player younger than 18 yr participated in blood or muscle analyses.

Experimental design. The players took part in one experimental game which was modified to normalize the individual playing exposure and the work/rest ratio: each of three periods involved eight 1-min shifts per player separated by 2 min of recovery resulting in a total on-ice time of 24 min for each participant. This work/rest ratio is based on competitive elite ice hockey games for the players exposed to average or above average playing time and with recovery time in the lower range of average values, as for the most frequently used players (7,8). Hence, each team consisted of 15 players forming three lines playing 1 min alternately. The change of lines at 1-min intervals was signaled by a whistle followed by a rapid transition of players, with a new line from each team continuing to play instantly. The game was evenly matched and modified so that no penalties were awarded for rule infringements to avoid odd-man situations. Otherwise, the game was performed in compliance with official match standards consisting of three periods interspersed by ~18 min of recovery. Before the game players refrained from strenuous exercise for 48 h and from tobacco and caffeine for 12 h and were instructed to follow their habitual pregame diet. During the game no exogenous carbohydrate intake was permitted, whereas water ingestion was allowed *ad libitum*. Average water intake was 1.8 L, whereas the average net-loss of fluid was 0.7 L corresponding to less than 1% of total body mass. The activity pattern and associated physiological responses during exercise were continuously monitored using an indoor local positioning system (LPS) (n = 19) and heart rate measurements with accelerometer recordings embedded (n = 16). Muscle biopsies were performed before and after the game (n = 6) or instantly after a shift during the first and last periods

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(n = 7). In addition, blood sampling was performed pregame and after the first and last periods (n = 14), whereas repeated-sprint ability was measured pregame and after each period (n = 30).

Muscle and blood sampling. Blood was drawn from an antecubital vein with players placed in a seated position. All blood samples were drawn in lithium-heparin and serum-tubes and distributed in 2-mL tubes for different analyses. Lithium-heparin samples were kept cold and centrifuged after the session, whereas serum samples rested for 1 h in room temperature before centrifugation. Samples for lactate and glucose were distributed immediately after the blood was drawn from the serum-tubes. Afterwards all samples were transported on dry ice before being stored at −80°C. Blood samples were drawn before the game as well as at the end of the first and third periods (e.g., either after shift 6, 7, or 8). Blood samples at the end of the first and third periods were collected within 30 to 60 s after a shift with the players seated next to the rink. Muscle biopsies were obtained from m. vastus lateralis (70–120 mg w.w.) from the nondominant leg using the needle biopsy technique with suction. In preparation for the muscle biopsies, incisions were made under local anesthesia (10–20 mL Lidocaine) and afterward covered with sterile band aid strips and gauze. All biopsies were taken with players lying in the supine position on portable beds placed next to the rink. The delay in sampling time during the game was assessed and was ~30 s after the players came off the ice. For six players, this procedure was performed before the game and immediately after the game. Seven other players had biopsies taken during the first and third periods, one to two players at a time, immediately after shifts 2, 3, 4 or 5 at a fixed timepoint during each period.

Activity pattern and heart rate measurements. The activity pattern and associated physiological responses were continuously monitored. Heart rate and accelerometer data (200 Hz) were collected using a wearable system (Polar Team Pro, Polar, Kempele, Finland), attached on the front of their upper body using a chest strap. The data were subsequently transferred and analyzed on a laptop using the manufacturer software (Polar Team Pro software). In addition, player activity patterns were monitored using an indoor 20-Hz LPS tracking system from Catapult Sports (Catapult Clearsky T6; Catapult Sports, Australia). The validity of the system has been tested previously showing low errors of measurement (17). Anchor nodes were mounted around the rink, and the LPS system was spatially calibrated using a tachymeter (Leica Builder 509 Total Station; Leica Geosystems AG, Switzerland). Each player was equipped with a small mobile node (firmware version 1:40) measuring 40 × 52 × 14 mm positioned between the shoulder blades in a manufacturer-supplied harness. After the game, data were transferred to a laptop and processed and analyzed using the manufacturer's software system (OpenField software). Skating distances were coded into the following categories and speed thresholds as previously done in ice hockey (8): very slow speed skating (1.0–10.9 $km \cdot h^{-1}$), slow speed skating (11.0–13.9 $km \cdot h^{-1}$), moderate speed skating (14.0–16.9 $km \cdot h^{-1}$), fast speed skating (17.0–20.9 $km \cdot h^{-1}$), very fast speed skating (21.0–23.9 $km \cdot h^{-1}$), and sprint skating (>24.0 $km \cdot h^{-1}$). Total distance was given by the sum of all speed zones. In addition the number of explosive efforts (sum of high-intensity [>2.5 $m \cdot s^{-2}$] accelerations, decelerations, and change of directions) was captured, as well as all accelerations and decelerations >0.5 $m \cdot s^{-2}$ from the Polar units. Heart rate zones were coded as low (<80.0%HR_{max}), moderate (80.0–84.9%HR_{max}), high (85.0–89.9%HR_{max}), very high (90.0–94.9%HR_{max}), and maximal (>95.0%HR_{max}).

Repeated-sprint ability. After the pregame warm-up, a repeated-sprint test was performed consisting of 3 × 33.15-m all-out efforts from the goal line to the second blue line, interspersed by 25 s of active recovery (skating slowly back to the start). Each player initiated the sprint 50 cm behind the first timing gates (Witty Gate Wireless Training Timer; Microgate, Italy, with a precision of 0.001 s) and a dual beam setup was used to prevent interference from swinging limbs or the stick accidentally activating the gates prematurely. Verbal encouragement was provided to motivate the players to give a maximal effort each time. The repeated-sprint test was performed in the same manner approximately 5 min after the first, second, and third periods. The mean time of the three sprints were recorded as an indicator of repeated-sprint ability.

Muscle analyses. The muscle samples were frozen in liquid nitrogen instantly after sampling and subsequently stored at −80°C. The samples were weighed in frozen condition before and after freeze drying to determine water content. After freeze drying, muscle tissue was dissected free of blood, fat, and connective tissue. After extraction with $HClO_4$, neutralized muscle extracts were analyzed for lactate, adenosine triphosphate (ATP), PCr, and muscle creatine (Cr) as previously described (18). For the lactate analyses, the buffer solution used was analyzed for lactate in the same way as the muscle extract to estimate the total muscle lactate production while accounting for potential release to the buffer. Muscle glycogen content was determined spectrophotometrically (Beckman DU 650). Freeze-dried muscle tissue (1.5 mg) was boiled in 0.5 mL of 1 M HCl for 150 min before being quickly cooled, whirl mixed, and centrifuged at 3500g for 10 min at 4°C. Forty microliters of boiled muscle sample and 1 mL of reagent solution containing Tris buffer (1 M), distilled water, ATP (100 mM), $MgCl_2$ (1 M), $NADP^+$ (100 mM), and G-6-PDH were mixed before the process was initiated by adding 10 µL of diluted hexokinase. Absorbance was recorded for 60 min before the glycogen content was calculated. Muscle glycogen was expressed as millimoles per kilogram of dry weight. Muscle pH was measured by a small glass electrode (XC 161; Radiometer-analytical, Copenhagen, Denmark) after homogenizing a freeze-dried muscle sample of about 2 $mg \cdot d.w.^{-1}$ in a nonbuffering solution containing 145 mM KCl, 10 mM NaCl, and 5 mM iodoacetic acid.

A part of each muscle biopsy was mounted in an embedded medium (OCT Compound Tissue-Tek; Sakura Finetek, Zoeterwoude, the Netherlands) and frozen in isopentane precooled in liquid nitrogen and subsequently stored at −80°C.

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These samples were analyzed for fiber-type specific glycogen content by histochemical analyses. Serial 10-μm-thick sections were cut at −20°C. To evaluate the relative glycogen content of individual fibers one 10-μm-thick transverse section was first stained for glycogen by the periodic acid-Schiff reaction, after which primary (MHC Fast 1:1000, M4276; Sigma-Aldrich) and secondary antibodies (Alexa 488 1:1000, A28175; Thermo-Fischer) were applied to visualize type II myofibers. Under light microscopy, the staining intensity of the fibers was defined as full (755–100% density), partly full (505–75% density), partly empty (255–50% density), or empty (05–25% density). For every subject, ~100 randomly chosen fibers at each timepoint were analyzed for glycogen staining density. The relative glycogen content of each fiber type was categorized in a spectrum between the average of the 50 most full (100% full) and most empty (fully empty) fibers of the total pool of fibers (~1200 fibers) involving both premeasurements and postmeasurements. Mean optical density of a selected region outside of the muscle cells was used for background corrections, as done previously (19).

Blood analyses. Glucose was measured directly on HemoCue using the glucose dehydrogenase method and photometric detection. Lactate was kept in cold hemolyse buffer until the next day and measured on Biosen (EKF Diagnostics) by the enzymatic-amperometric method using chip-sensor technology. One drop (approximately 50 μL) serum was used for lactate and glucose cuvette analyses. Plasma free fatty acid (FFA) was analyzed on the Pentra 400 C using an enzymatic kit (WAKO Chemical, Germany). Plasma insulin was analyzed on the Cobas 8000, e602 (Roche) by a sandwich enzyme immunoassay and electrochemiluminescence using a two-point calibration curve.

Statistical analyses. Data are presented as means ± SD. To determine the sample size for glycogen and muscle metabolites, we assumed a minimum mean difference of 150 mmol·kg^{-1}·d.w. in muscle glycogen and a maximum SD of 100 mmol·kg^{-1}·d.w. (5). Our sample size calculation revealed that a minimum of 5 subjects were necessary to yield a power of 0.8 with an α-level of 0.05. For muscle metabolites during the game, larger variance was expected, and a minimum of six subjects was necessary to detect the smallest expected differences in muscle lactate based on expected average values of at least 15 mmol·kg^{-1}·d.w. from resting levels of 5 mmol·kg^{-1}·d.w. and a maximum SD of 12 mmol·kg^{-1}·d.w. (5). Additionally, for meaningful changes in repeated-sprint ability a minimum of 20 subjects were needed based on previous values obtained after a soccer game, with an expected decrease of at least 2% and with a maximum SD of 0.2 s (5).

Shapiro–Wilk test revealed that muscle and blood variables, as well as repeated-sprint ability were normally distributed ($P > 0.05$). Differences in muscle variables before and after the game as well as between the first and third periods were assessed by Student's paired *t* test. Differences between muscle variables before and after the game compared with measurements obtained during the first and third periods were assessed by Student's unpaired *t* test. Changes in repeated-sprint ability after each period were assessed by two-way ANOVA RM, with the two factors being timepoint (e.g., baseline and after each period) and sprint number (e.g., sprints 1, 2, and 3 during the three repeated sprints). Heart rate and accelerations and decelerations obtained by the Polar system, as well as blood variables obtained pregame and after the first and third periods, were evaluated using one-way ANOVA RM. However, the LPS system had a technical issue resulting in a pattern of missing data during the third period. We, therefore, applied a general linear mixed model to analyze differences in activity pattern throughout the game (20). In this case, the match period was set as fixed effect and individual subjects set as random effect, whereas LPS variables were dependent variables. When a significant interaction was found, multiple comparison procedures were applied using the Holm–Sidak method to detect where the significant difference existed. Significance level was set at $P \leq 0.05$. Finally, correlation coefficients were calculated between muscle variables and match activities using a Pearson Product Moment Correlation qualitatively interpreted as: $r \leq 0.1$ (trivial), 0.1–0.3 (small), 0.3–0.5 (moderate), 0.5–0.70 (large), 0.7–0.9 (very large), and ≥0.9 (almost perfect) (21). All statistical analyses were performed using the SigmaPlot statistical software (SigmaPlot for Windows version 14.0, Systat Software Inc., London, UK).

RESULTS

Muscle metabolites and pH. Table 1 shows changes in muscle metabolites and pH throughout the game. Muscle glycogen content was significantly lowered during the first and third periods and after the game ($P \leq 0.05$; Fig. 1D). Histochemical analyses based on staining density revealed that before the game, 58% and 51% of fast- and slow-twitch muscles were categorized as full or partly full of glycogen. After the

TABLE 1. Muscle metabolites before and after ($n = 6$) and during ($n = 7$) simulated elite ice hockey match-play.

	Baseline	During First Period	During Third Period	Post
Muscle glycogen (mmol·kg^{-1}·d.w.)	400 ± 22	302 ± 81*	207 ± 71**	188 ± 43**
Muscle ATP (mmol·kg^{-1}·d.w.)	22.8 ± 1.2	22.0 ± 3.3	21.6 ± 5.9	23.8 ± 1.6
Muscle PCr (mmol·kg^{-1}·d.w.)	79.1 ± 6.7	64.0 ± 16.5	70.9 ± 22.2	86.5 ± 11.7
Muscle lactate (mmol·kg^{-1}·d.w.)	6.9 ± 2.7	37.8 ± 20.1*	19.6 ± 12.0**	21.0 ± 15.9**
Muscle Cr (mmol·kg^{-1}·d.w.)	42.4 ± 5.7	55.9 ± 14.8	49.0 ± 18.5	47.1 ± 9.4
Muscle pH (–log H+)	7.31 ± 0.04	6.99 ± 0.07*	7.13 ± 0.11**	7.21 ± 0.14***

Data are presented as mean ± SD.
*Significant difference from baseline.
**Significant difference from baseline and first period ($P \leq 0.05$).
***Significant difference from first period.

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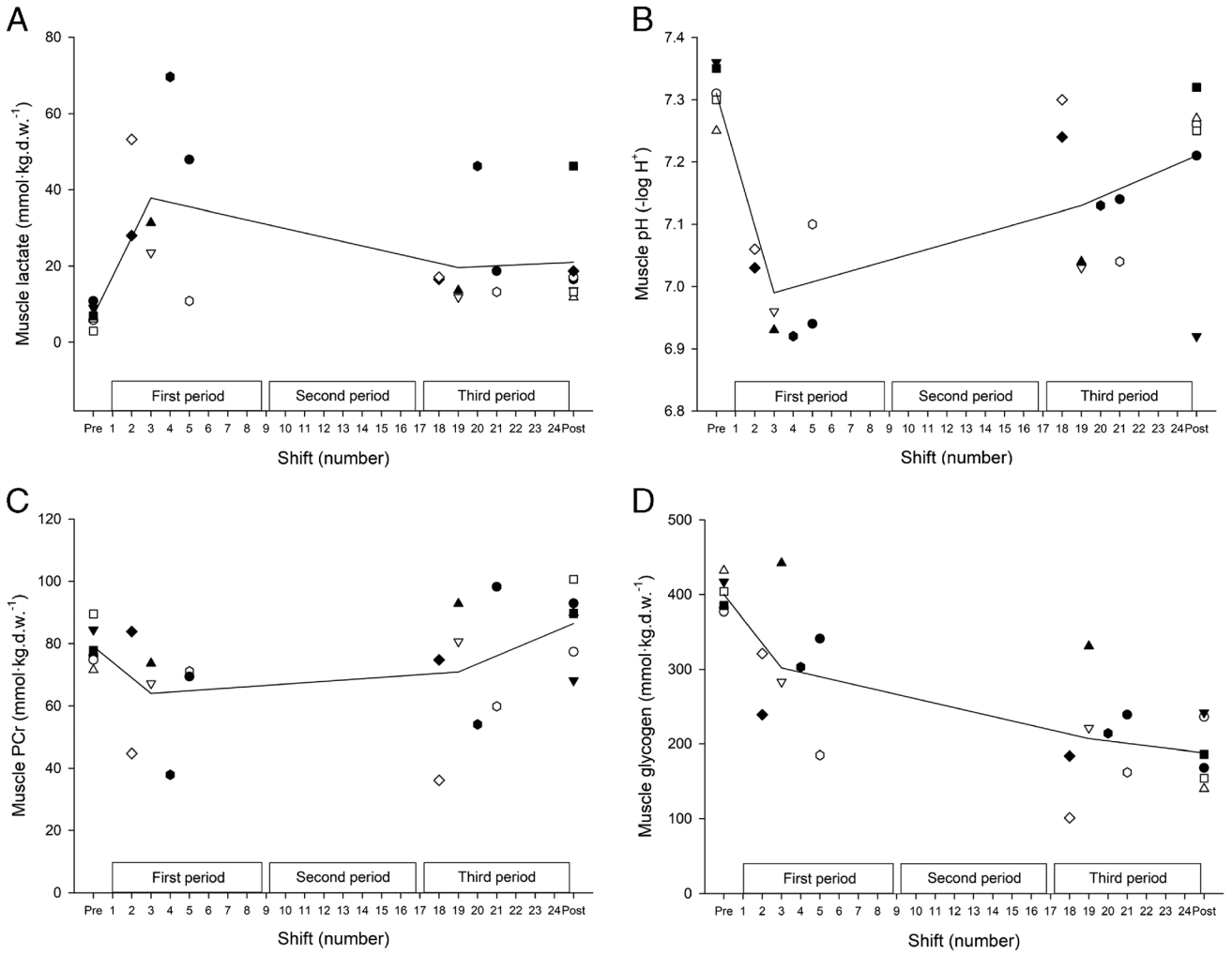

FIGURE 1—Muscle metabolites before and after (n = 6) and during simulated elite ice hockey match-play (n = 7). A, muscle lactate; B, pH; C, PCr; and D, glycogen. Individual and mean (*solid line*) values are presented.

game, only 6% and 5% of the fast- and slow-twitch fibers were categorized as full or partly full of glycogen, with 60% and 68% of fast- and slow-twitch fibers, respectively, categorized as depleted of glycogen (Fig. 2). Muscle lactate increased more than fivefold during the first period, and threefold during the third period ($P \leq 0.05$; Fig. 1A). Muscle ATP, PCr, and muscle Cr did not change significantly during the game, but the data were characterized by large interindividual differences (Fig. 1C). Muscle pH level decreased during the first and third periods ($P \leq 0.05$; Fig. 1B).

Blood metabolites and hormones. Changes in blood metabolites and hormones throughout the game are shown in Table 2. Blood lactate increased after the first period and remained high during the third period ($P \leq 0.05$; Fig. 3A). Blood glucose remained at similar levels throughout the game. Plasma FFA tended to increase after the first period ($P = 0.09$) and increased after the third period ($P \leq 0.05$). In contrast, plasma insulin levels decreased ($P \leq 0.05$) after the first and third periods.

Repeated-sprint ability. The mean sprint time for the repeated-sprint test before the game was 4.55 ± 0.19 s and increased ($P < 0.05$) by 2.9 ± 4.2% to 4.68 ± 0.15 s after the third period (Fig. 4). The mean repeated-sprint time after the third period also tended ($P = 0.06$) to be slower than after

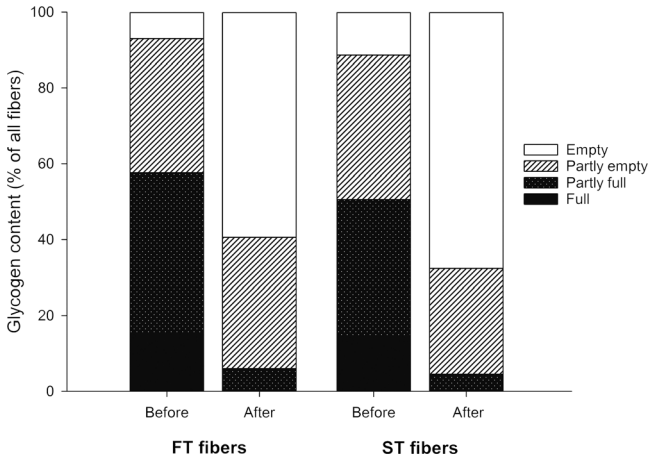

FIGURE 2—Fiber-type specific muscle glycogen content of fast- (FT) and slow-twitch (ST) fibers before and after simulated elite ice hockey match-play (n = 6).

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TABLE 2. Blood responses before, during, and after simulated elite ice hockey match-play (n = 14).

	Baseline	After First Period	After Third Period
Plasma lactate (mmol·L^{-1})	0.8 ± 0.3	4.7 ± 2.6*	4.9 ± 2.7*
Plasma glucose (mmol·L^{-1})	5.1 ± 1.1	5.4 ± 0.5	4.8 ± 0.6
Plasma FFA (μmol·L^{-1})	171 ± 59	288 ± 119*	459 ± 277**
Plasma insulin (pmol·L^{-1})	198 ± 146	95 ± 52*	44 ± 21**

Data are presented as mean ± SD.
*Significant difference from baseline.
**Significant difference from baseline and first period ($P \leq 0.05$).

the second period (4.60 ± 0.17 vs 4.68 ± 0.15 s). The first, second, and third sprints performed after the game increased ($P \leq 0.05$) from 4.47 ± 0.18, 4.57 ± 0.19, and 4.62 ± 0.19 s at baseline to 4.61 ± 0.15, 4.68 ± 0.19, and 4.74 ± 0.26 s, respectively, corresponding to ~1 m in the sprint test.

Match activities. Total distance covered during the game was 5980 ± 199 m, with no differences between the first, second, and third periods. Of this, 24%, 14%, and 17% was performed as very slow-speed, slow-speed, and moderate-speed skating (1407 ± 82, 833 ± 58, and 1038 ± 59 m), whereas almost half of the total distance was covered by high-intensity skating. This was distributed as 25%, 15%, and 5% of fast speed, very fast speed, and sprint skating, respectively (1468 ± 100, 911 ± 124, and 309 ± 75 m). In addition, the players performed 109 ± 14 explosive efforts with no difference between periods. In contrast, accelerations and decelerations above 0.5 m·s^{-2} obtained by the Polar units decreased by 9% and 10% during the second and third periods, respectively, compared to the first period (296 ± 31 vs 269 ± 15 and 266 ± 27 accelerations and decelerations, $P \leq 0.05$).

Heart rate measurements. Mean heart rate for the entire game was 76% ± 4% of individual maximum heart rate (%HR_{max}), whereas mean on-ice heart rate was 84 ± 2%HR_{max} and peak heart rate was 97 ± 2%HR_{max}. Mean on-ice heart rate was highest during the second period, whereas the cardiovascular strain during the third period was lower than during both the first and second periods (85 ± 3, 87 ± 3, and 82 ± 2%HR_{max} for the first, second, and third periods, respectively, $P \leq 0.05$). Accordingly, time in the highest heart rate zones was lower during the third period (time in zone 90–95%HR_{max}: 211 ± 75, 209 ± 5, and 170 ± 61 s and time in zone 95–100%HR_{max}: 87 ± 79, 96 ± 84, and 55 ± 68 s for the first, second, and third periods, respectively, $P \leq 0.05$). Accordingly, players spent more time below 80%HR_{max} during the last period ($P \leq 0.05$). In total, the players spent on average 22.5 ± 4.2 min in the heart rate zones above 85%HR_{max}, corresponding to almost one third of the total playing time and almost equal to the actual playing time on the ice.

Correlations. A strong significant correlation was observed between levels of blood lactate and number of explosive efforts per minute during the game (r = 0.71, $P \leq 0.05$), whereas the sum of fast, very fast and sprint skating distance covered tended to correlate with blood lactate levels (r = 0.46, P = 0.08). In contrast, no significant relationship was present between blood lactate levels and total distance covered.

DISCUSSION

The present study is the first to examine skeletal muscle metabolism and physiological responses in relation to exercise

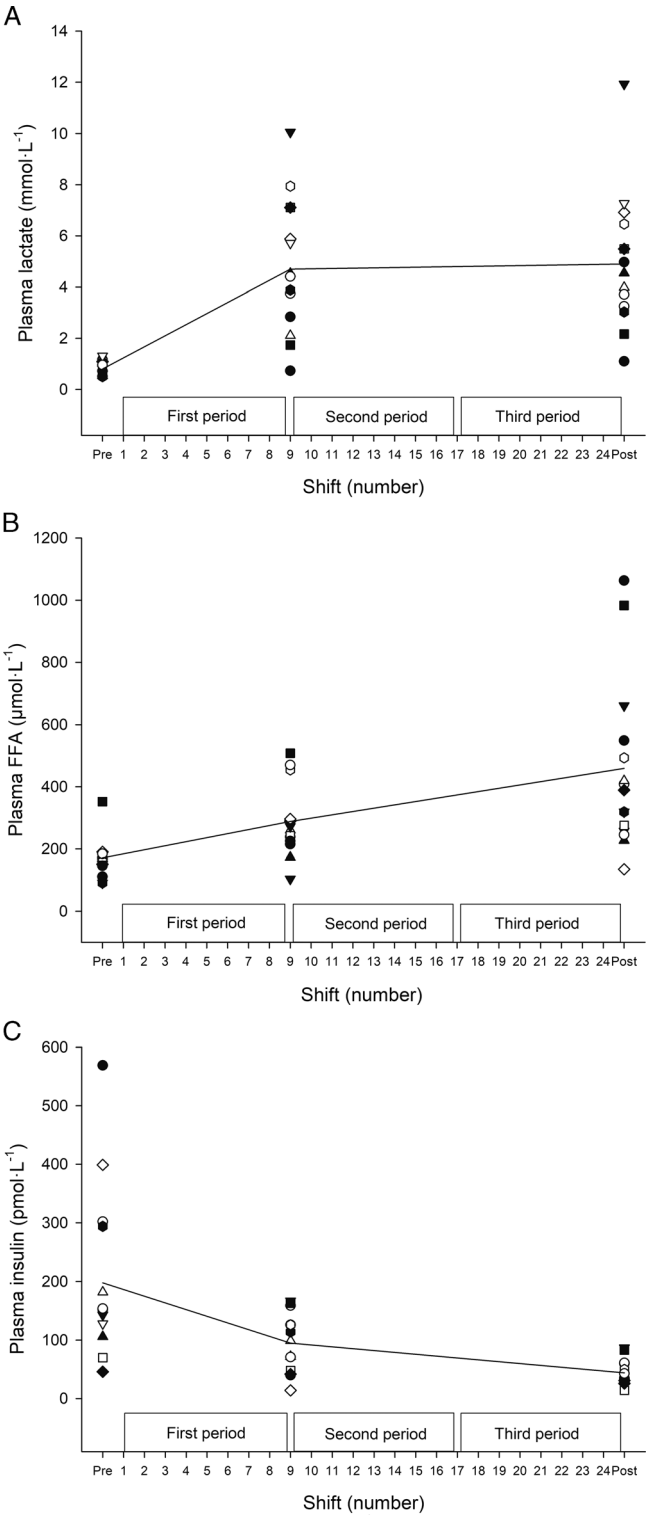

FIGURE 3—Blood metabolite and hormonal variables before simulated elite ice hockey match-play and after the first and third periods (n = 14). A, plasma lactate, (B) plasma FFA and (C) plasma insulin. Individual and mean (*solid line*) values are presented.

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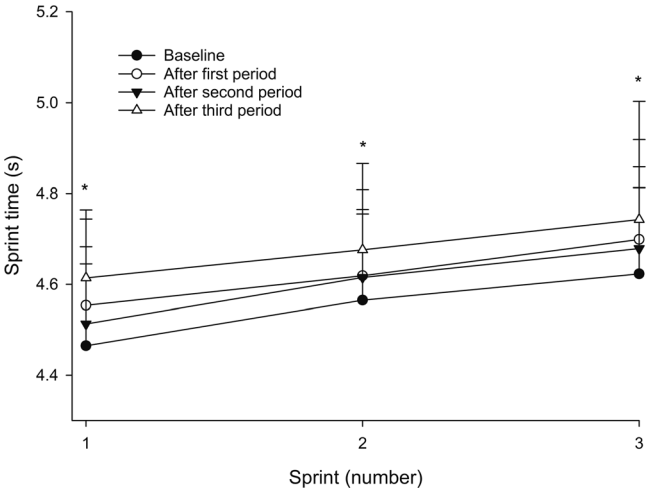

FIGURE 4—Repeated-sprint ability before simulated elite ice hockey match-play and after each period (n = 30). Data are presented as mean ± SD. *Significant difference from baseline ($P \leq 0.05$).

tolerance during simulated match-play in elite male ice hockey players. The glycogen utilization rate and elevated muscle lactate values demonstrate that thigh muscle anaerobic energy turnover was substantially elevated at peak timepoints during the game, in addition to an overall high aerobic loading. Hence, the players covered almost half the total distance at high intensities, resulting in deteriorated sprint performance and a lower number of accelerations and decelerations at the end of the game, coinciding with a large decline in muscle glycogen content. Interestingly, about two thirds of all fast- and slow-twitch fibers were depleted of glycogen, despite the relatively short effective time on the ice (~20 min). Although the muscle glycogen concentration was low, blood glucose levels remained stable. Furthermore, circulating FFA increased while muscle lactate levels and degree of acidosis were reduced during the last period, suggesting a lower glycolytic rate and potential for an increased reliance on beta-oxidation, although direct measurements are needed to confirm these findings.

Total distance covered during the game was ~6000 m, with nearly half the distance covered at high skating speeds (>17 $km \cdot h^{-1}$). Thus, it is likely that the high-intensity locomotion pattern could induce transient fatigue during the game, as has previously been demonstrated in other team sports (5). Accordingly, measurements of muscle metabolites throughout the game revealed a high anaerobic energy contribution reflected in individual muscle lactate concentrations as high as 70 $mmol \cdot kg^{-1} \cdot d.w.$ and low PCr values of around 35 $mmol \cdot kg^{-1} \cdot d.w.$ immediately after shifts, similar to values obtained at exhaustion after a high-intensity intermittent running protocol (4). Despite this, no main effect was observed for changes in PCr levels, probably reflecting the variation in sprint activities immediately before sampling and the time delay before obtaining the biopsy, allowing for a considerable resynthesis. Accordingly, estimated PCr values were as low as ~20 $mmol \cdot kg^{-1} \cdot d.w.$ immediately after the exercise bouts, using a previously reported resynthesis rate of 0.5 $mmol \cdot kg^{-1} \cdot d.w. \cdot s^{-1}$ in relation to our ~30-s muscle biopsy sampling delay (5). Nonetheless, muscle ATP levels remained stable, indicating that overall muscle energy status was maintained or that the restoration of homeostasis was obtained faster than our sampling ability. Muscle pH values were only moderately lowered and not in the range that would be expected to induce fatigue, suggesting that muscle acidosis is an unlikely contributor to performance impairment during this type of exercise (9). This is in accordance with previous findings demonstrating no relationship between reduced sprint performance during a soccer game and muscle lactate or pH (5). Instead, other factors, such as perturbations in ion concentrations across the membrane, accumulation of inorganic phosphate, impaired calcium regulation, and other mechanisms, have been proposed as more likely fatiguing agents as will be discussed later (9).

Average on-ice heart rate during the game was 82 to 87%HR_{max}, with peak values approaching maximal levels, which is comparable to previous findings in ice hockey (6,22) and soccer (5). Moreover, total time in the heart rate zones above 85%HR_{max} was almost 23 min, corresponding to about one third of the total game duration and very closely to the actual playing time. Thus, during an ice hockey game the aerobic energy system is significantly taxed despite the short duration of each shift and intermittent activity pattern (6). In support of this notion, the ice hockey players in the present study had an extraordinarily high muscle oxidative capacity (data not shown). This probably reflects the importance of oxidative phosphorylation for resynthesizing ATP and PCr between repeated bursts of high-intensity effort and allowing for a substantial contribution of the aerobic system to the energy provision during each shift, thus decreasing the anaerobic energy yield (1,2,23). Accordingly, Bishop et al. (24) demonstrated a close relationship between $\dot{V}O_{2peak}$ and repeated-sprint ability in a cohort of untrained subjects, though this association was only modest in competitive ice hockey players (25), suggesting that in a trained population factors other than $\dot{V}O_{2peak}$ are important for repeated-sprint performance.

The fast-paced skating profile and high cardiovascular and muscle metabolic loading resulted in a high rate of anaerobic glycolysis, as suggested by the elevated muscle lactate values and substantial muscle glycogen utilization. Thus, muscle glycogen content was reduced by more than 50%, from 400 to 188 $mmol \cdot kg^{-1} \cdot d.w.$ during the game. This is similar or even larger than previously reported utilization rates during a soccer game, where glycogen levels dropped from 449 to 255 $mmol \cdot kg^{-1} \cdot d.w.$ despite the ~fourfold to fivefold longer game duration (5). Moreover, we may even overestimate the postgame glycogen levels slightly, because our method for determining glycogen biochemically does not distinguish between glucose moieties from glycogen and glucose-6-phosphate, which is likely higher after exercise. Other investigations have reported comparable reductions in muscle glycogen of 60% after ice hockey match-play, or even 70% during a simulated intermittent skating protocol supporting our findings (6). In concert with this large drop in muscle glycogen, sprint performance

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was attenuated after the game, indicating fatigue development in accordance with previous findings in comparable team sports (5). Accordingly, fewer accelerations and decelerations and a lower exercise intensity, reflected by the decrease in cardiovascular loading in the last period, was demonstrated in the present study, which is in line with recent investigations in top-class competitive ice hockey reporting a reduction in high-intensity activities during the final stages of a game (8,26).

Thus, a reduced energetic state of the muscle may impair exercise tolerance when high-intensity exercise is repeated multiple times. Accordingly, muscle glycogen content in individual fibers was very low in ~60% to 70% of all fibers after the game, with a similar depletion pattern in each muscle phenotype. However, a significant number of fibers were already partly empty at the onset of exercise, contributing to this high degree of depletion. This may relate to the tight game and practice schedule in elite ice hockey combined with inadequate pregame carbohydrate loading, possibly not allowing players to reach optimal pregame levels. Moreover, muscle glycogen is stored heterogeneously within muscle fibers, and it is, therefore, expected that some fibers are partly depleted of glycogen already before exercise (27). The parallel depletion rate in both fast- and slow-twitch fibers is analogue to results obtained by Gollnick et al. (28) after high-intensity exercise. Hence, it was demonstrated that during low-intensity exercise, slow-twitch muscles are depleted initially before subsequent recruitment of fast-twitch fibers, whereas both fiber types are utilized simultaneously at higher exercise intensities. However, our results are in contrast with early studies of ice hockey match-play, as predominant utilization of type I fibers was reported, albeit both slow- and fast-twitch fibers were significantly taxed (15,16). This discrepancy may in part be explained by changes in the intensity of the game or by the fact that the present investigation was performed in an elite cohort, whereas these early studies involved sub-elite players. In addition, potential methodological differences may also affect these discrepancies. In accordance with our results, total or partial depletion of almost half of the individual muscle fibers has been shown after a soccer game, with a similar utilization pattern across fiber types (5). This marked drop in muscle glycogen, leading to depletion of many individual fibers, could potentially impair the ability to produce adequate muscle force required for maximal efforts and impact performance, despite whole muscle glycogen still being above critical levels. For example, an early study by Balsom et al. (29) demonstrated a drop in repeated-sprint performance on a cycling ergometer, when muscle glycogen levels were reduced to 180 $mmol \cdot kg^{-1} \cdot d.w.$, matching the end-game values in the present study. In addition, Bangsbo et al. (27) demonstrated an impaired glycolytic rate as muscle glycogen levels declined to around ~200 $mmol \cdot kg^{-1} \cdot d.w.$, which is supported by previous studies (30,31). Accordingly, muscle lactate levels and the degree of acidosis were lower in the third period in the present study, suggesting that the glycolytic rate may have been impaired. A decrease of more than 50% in glycogen content with a large fraction of fibers being very low on glycogen, may also have lowered one or several of the sub-cellular glycogen pools (32). As such, an association between the intramyofibrillar pool of muscle glycogen in close contact with key enzymes involved in excitation–contraction coupling and altered sarcoplasmic reticulum calcium release rate has been demonstrated (33) and a critical threshold of muscle glycogen content around 250 to 300 $mmol \cdot kg^{-1} \cdot d.w.$ suggested for maintenance of muscle function (34). Furthermore, it is possible that Na^+-K^+-ATPases responsible for maintaining muscle Na^+-K^+ gradients required for excitability could be affected by lowered levels of muscle glycogen, as these pumps preferentially utilize muscle glycogen for ATP resynthesis. Moreover, it has recently been demonstrated that inhibition of glycogenolysis prolongs the action potential repriming period, suggesting altered muscle excitability (35,36). Thus, several studies have shown depressed Na^+-K^+-ATPases activity after intense and prolonged exercise, but a clear link to muscle glycogen content remains to be established (11). In addition, chloride channels, likewise functioning as important regulators of muscle cell excitability, have been demonstrated to be sensitive to changes in muscle energetic state and are, therefore, likely to be prone to alterations in muscle glycogen content (10). Studies manipulating pregame glycogen content or administering carbohydrate supplements during ice hockey and other team sports have also pointed to an important role of muscle glycogen for exercise tolerance (13,14,22,37). Collectively, the large muscle glycogen utilization during this type of intense intermittent exercise is likely related to exercise tolerance and may have important implications for nutritional strategies before, during, and subsequent to games.

In association with the degradation in muscle glycogen, a large increase in plasma FFA concentration was observed at the end of the game concomitant with a reduction in plasma insulin. An increased availability of plasma FFA is associated with increased oxidation at rest and during low- to moderate-intensity exercise (38,39). However, during high-intensity exercise several regulatory steps besides fuel availability may be limiting for FFA oxidation, including transport across the muscle membrane and into the mitochondria, as well as overall mitochondria volume and capacity (38,39). Thus, the transfer of FFA may be inhibited by a high rate of glycolysis exceeding the rate of the tricarboxylic acid cycle. Therefore, increased availability of FFA in the present study may not necessarily reflect an increased rate of beta oxidation, but instead hormonal stimulated release into the bloodstream. However, if the rate of glycolysis is impaired at the later stages of the game, this may allow an increased reliance on fat utilization and act as a glycogen-sparing mechanism, though direct measurements need to confirm this hypothesis. In addition, despite not being a major factor, artificially increased levels of FFA have been shown to restore part of the oxidation rate of fatty acids during exercise at ~85%$\dot{V}O_{2max}$ (40). Accordingly, although speculative, fat oxidation may be of increasing importance in the final stages of a game, when carbohydrate stores are low.

The research design applied an experimental match setup with a fixed work/rest ratio and total playing time simulating

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a competitive scenario. However, unlike during a competitive game, time was not paused during stoppages to maintain a constant ratio between on-ice time and recovery. Thus, effective playing time was around 50 s per shift resulting in an effective individual playing time of ~20 min per player during the 24 min on the ice. The game took place during the preparation period before the U20 world championship, so all players were in peak condition and highly motivated to secure selection for the world championship squad. Accordingly, the skating distances (~6000 m) and intensity of the game correspond well with recent findings by Lignell et al. (8) from an official National Hockey League game, adjusting for differences in total playing time. Average total distance covered in other studies has ranged from ~4500 to 5500 m, but with less players exposed to the same playing time as in the present study (6,26). Average time on the ice for elite players is usually in the ranges of 15–25 min, with each shift lasting 30–80 s, separated by 2–3 min recovery or longer, and with some individual players, or several in case of overtime being exposed to more playing time (7,8). Thus, the present design represents the exercise scenario for players with average to above average playing time in an elite ice hockey game and with the minimum available recovery time between shifts. Hence, it is complex to characterize muscle metabolism during ice hockey match-play given the inherent variability in playing exposure and differing work/rest ratio between individual players and positional roles. It should, therefore, be noted that the results may differ for players with less playing time or longer rests between shifts.

In conclusion, oxidative and glycolytic energy systems are highly activated during ice hockey match-play as suggested by the substantial heart rate and muscle metabolite responses, resulting in fatigue development in the later stages of a game. This is likely associated with a markedly high utilization of muscle glycogen, resulting in depletion of ~65% of individual slow- and fast-twitch fibers.

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The results of this study are presented clearly, honestly, and without fabrication, falsification, or inappropriate data manipulation. The results of the present study do not constitute endorsement by ACSM. The authors report no conflicts of interest.

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