Muscle glycogen utilisation during Rugby match play: Effects of pre-game carbohydrate

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## Abstract

Objectives: Although the physical demands of Rugby League (RL) match-play are wellknown, the fuel sources supporting energy-production are poorly understood. We therefore assessed muscle glycogen utilisation and plasma metabolite responses to RL match-play after a relatively high (HCHO) or relatively low CHO (LCHO) diet. Design: Sixteen (mean  $\pm$  SD age; 18  $\pm$  1 years, body-mass; 88  $\pm$  12 kg, height 180  $\pm$  8 cm) professional play-ers completed a RL match after 36-h consuming a non-isocaloric high carbohydrate (n = 8; 6 g kg day–1) or low carbohydrate (n = 8; 3 g kg day–1) diet. Methods: Muscle biopsies and blood samples were obtained pre- and post-match, alongside external and internal loads quantified using Global Positioning System technology and heart rate, respectively. Data were analysed using effects sizes  $\pm$ 90% CI and magnitude-based inferences. Results: Differences in pre-match muscle glycogen between high and low carbohydrate conditions (449  $\pm$  51 and 444  $\pm$  81 mmol kg–1 d.w.) were unclear. High (243  $\pm$  43 mmol kg–1 d.w.) and low carbo-hydrate groups (298  $\pm$  130 mmol kg–1 d.w.) were most and very likely reduced post-match, respectively. For both groups, differences in pre-match NEFA and glycerol were unclear, with a most likely increase in NEFA and glycerol post-match. NEFA was likely lower in the high compared with low carbohydrate group post-match (0.95  $\pm$  0.39 mmol l–1 and 1.45  $\pm$  0.51 mmol l–1, respectively), whereas differences between the 2 groups for glycerol were unclear (98.1  $\pm$  33.6 mmol l–1 and 123.1  $\pm$  39.6 mmol l–1) in the high and low carbohydrate groups, respectively. Conclusions: Professional RL players can utilise ~40% of their muscle glycogen during a competitive match regardless of their carbohydrate consumption in the preceding 36-h.

# Introduction

Rugby league (RL) is a high-intensity intermittent team sport played over 80 min. Players are required to perform repeated bouts of relatively short, high-intensity efforts such as sprints, high-impact collisions, and sudden changes of direction, interspersed with low-intensity activities such as standing, walking, and jogging.1–3Playing position dictates the predominance of activ-ity performed during a game, with forwards (props, 2nd row and loose forward) primarily carrying the ball forward into collisions to make positive ground, and the adjustables (scrum-half, stand-off,hooker) and outside backs (fullback, wingers and centres) travelling greater distances, running more into open spaces and support-ing offensive plays.3Total distance covered relative to match time (m min–1) is similar between positions,4,5~90–95 m min–1.All players experience physical collisions regularly throughout agame25with forwards being involved in one collision per minute, which is greater than both the adjustables (~0.6 collisions per the match demands of professional RL

have been well described 3,6,7 the sources of energy fuelling such workloads are poorly understood.8 Several studies in professional soccer have reported large changes in players' muscle glycogen concentrations immediately after a competitive fixture.8,9 Many players finished a match with less than 200 mmol kg-1 dry weight (d.w.) of muscle glycogen and some showed almost complete depletion in the whole muscle (<50 mmol kg-1 d.w.)10 and individual fibres.8 Blood metabolites have also indicated mobilisation of lipids after a football match, with increases in blood NEFA and glycerol concentrations.8,11 These metabolic changes have been used to describe fatigue in soccer players, characterised by reductions in high-intensity running and sprinting.12 Moreover, high-intensity running is also impaired in players starting a game with low muscle glycogen.12 Metabolic and match demands data have helped to devise physi-ological training programmes and nutritional strategies to enhance performance and/or delay fatigue in soccer.13 Such studies have also formed the basis of nutritional position stands,14 which have then been translated for use in RL. However, there are distinct dif-ferences in the game characteristics between RL and soccer, most notably the greater distances covered by soccer players 11,15 and the multiple physical collisions observed in rugby.16 Therefore, the suitability of using such studies to inform nutritional practices of rugby is questionable.17,18 Despite limited empirical evidence, tra-ditional nutritional practice in rugby has been to load with CHO in the days leading up to a game, 14 including doses between 6 and 10 g kg-1 body mass. Many professional rugby players might not strictly adhere to this advice17,18 possibly because their large body mass makes such large CHO intakes difficult to consume (poten-tially >1 kg of CHO per day for some larger players). Accordingly, the effects of an acute CHO intake on muscle glycogen concentration and performance of professional RL players warrants investigation. The aim of the present study was to therefore quantify the phys-iological and metabolic demands of RL match-play after 36-h on a relatively high (HCHO) or a relatively

low (LCHO) carbohydrate diet. To this end, we studied professional male academy players dur-ing a competitive RL match having followed 36-h of either 6 g kg–1 (HCHO) or 3 g kg–1 (LCHO) carbohydrate diet. We hypothesised that (1) a competitive RL match would result in muscle glycogen deple-tion in players similar to that seen after competitive soccer games and (2) players consuming 6 g kg–1 CHO would perform better in the second half compared with those players that followed the 3 g kg–1 CHO diet. 2.

## Methods

Sixteen professional rugby players (mean  $\pm$  SD age 18.2  $\pm$  0.8 years, body-mass 88.4  $\pm$  12.4 kg, height  $180 \pm 8.1$  cm) from a Super League rugby club academy volunteered to take part in this study. All of the players were full time professionals with  $\sim$ 70% of the players appearing in the first team squad at some point during the season. Using a random number generator, players were split in to one of two dietary groups. All players competed on the same team and completed a calendared 80-min competitive RL match with a 10 min halftime period against a team from the same league, with 16 and 14 players giving a muscle biopsy and a venous blood sample pre- and post-match, respectively. Two players declined to give a post-match muscle biopsy and were withdrawn from this aspect of the study. Therefore all muscle biopsy and blood data are presented as n = 14 whereas GPS and HR data are presented on n = 16. Ethics approval for the study was granted by the ethics committee of Liverpool John Moores University and all participants provided written informed consent before starting the study. Players began a standardised five-day lead in to a competitive rugby match as prescribed by the club (Table 1a). Players followed an individualised diet plan beginning 36-h before kick off, with habitual diet ingested before the intervention. In a standardised order, players provided a muscle biopsy followed by a venous blood sample before and after the match, alongside urine samples and countermovement jumps. Only water was ingested between pre-and post-match samples. Relative distances covered, low intensity

activity, high intensity running and repeated high-intensity efforts (RHIE) performed by all players during the match were assessed using GPS technology (Optimeye S5, Catapult Innovation, Melbourne, Australia) as previously described.17,18 Heart rate was also monitored using a coded transmitter unit (Polar, Oy, Finland) strapped to the chest with data transmitted and recorded to GPS units for later download and analysis. Individualised diets were designed and provided to all participants. Player body mass was recorded and grouped to the closest 10 kg (70, 80, 90, 100 or 110 kg) which was used to prescribe the CHO content of the individual's diet. In a randomised design, players were initially divided into forwards and backs, ranked according to body mass, and then block randomised to one of two diet groups comprising either relatively high CHO ( $\sim$ 6 g kg day–1 CHO,  $\sim$ 1.8 g kg day–1 protein and 0.7 g kg day-1 fat) or a non-isocaloric relatively low CHO ( $\sim$ 3 g kg day-1 CHO,  $\sim$ 1.8 g kg day-1 protein and 0.7 g kg day-1 fat). Although it could be argued that 6 g kg day-1 might not appear a particularly 'high CHO diet' and likewise 3 g kg day-1 is not a 'low CHO diet', these intakes are typically ingested by rugby players during the training week and leading up to competition, respectively.17,18 Moreover, given the large muscle mass of rugby players, 6 g kg day-1 results in almost 600 g of total CHO, which could be considered high. It must, how-ever, be stressed that although 'high' and 'low' terminology is used throughout this manuscript, the absolute amount of CHO consumed by the players should be considered when comparing to the previous literature. CHO was taken in the form of both solids and fluids, with groups fluid matched ( $\sim$ 3.5 L). Foods were selected, purchased, and distributed by the clubs nutritionist and delivered to the players accompanied by a meal plan. All players were given strict instructions to follow the diet beginning the day before the match, not to consume any foods or liquids other than what was provided and to finish all meals provided. All players self-reported that they had strictly adhered to the diets prescribed to them. Muscle samples were obtained using a biopsy gun (Monopty 12g, BARD, Brighton, UK) from the middle of the vastus lateralis muscle on the right leg as previously described.19 Although the increase in inflammatory markers after a muscle biopsy have been well documented, 20 given our key measure was glycogen, it was decided that post-match biopsies would be taken from a new incision close to the original site ( $\sim 2$  cm proximal) after the same procedures.19 All muscle samples were immediately snap frozen in liquid nitrogen and stored at -80 °C for later analysis. Blood samples (5 ml) were drawn from a superficial vein in the anticu-bital fossa of the forearm using standard venipuncture techniques (Vacutainer Systems, Becton, Dickinson). Samples were collected into three vacutainers (Serum Separating Tube, EDTA and Lithium Heparin tubes, Nu-Care Products, UK), and were stored on ice (apart from serum) until centrifugation at 1500 RCF for 15 min at 4 °C. All post-match muscle and blood samples were taken within 40-min of the match finishing, which was achieved by having four qualified researchers performing the biopsies simultaneously. Muscle glycogen concentration was determined as previously described19 and analysed in duplicate with the mean of the two measures recorded. Intra-assay coefficients of variation (CV) was <5%. Blood was analysed spectrophotometrically for glycerol, NEFA and glucose concentrations using commercially available kits (Randox, Laboratories, Antrim, UK) with intra-assay CV of <5% for all assays. Countermovement jump (CMJ) height was estimated from an individual's flight time using a contact timing mat (Just Jump, Pro-biotics Inc, Alabama) as previously described.21 Urine samples were collected in a 30 ml container (Sterilin universal, Sterilin, UK) upon arriving at the club and were analysed for osmolality measured in mOsm kg-1 using a handheld osmometer (Osmocheck, Perform- Better, UK) which has previously been validated.22 Magnitude-based inferential statistics were employed to pro-vide information on the size of the differences allowing a more practical and meaningful explanation of the data. Differences in (i) movement characteristics between the first and second half, and (ii) for urine osmolality,

CMJ height, muscle glycogen con-centration and blood metabolites before and after the game were analysed for HCHO and LCHO groups using Cohen's effect size (ES) statistic ± 90% confidence limits (CL) and magnitude-based infer-ences.23 Threshold probabilities for a meaningful effect based on the 90% confidence limits (CL) were: <0.5% most unlikely, 0.5–5% very unlikely, 5–25% unlikely, 25–75% possibly, 75–95% likely, 95–99.5% very likely, >99.5% most likely. Effects with confidence limits across a likely small positive or negative change were classi-fied as unclear.24 All analyses were completed using a predesigned spreadsheet.16 3.

#### Results

Muscle glycogen data are presented in Fig. 1. Differences in muscle glycogen between HCHO and LCHO were unclear during the first (ES  $\pm$  90% CL: 0.05  $\pm$  1.31) and second halves (0.65  $\pm$  1.93). Muscle glycogen most likely reduced from pre- to post-match by 45  $\pm$  9.5% (4.96  $\pm$  1.43) and very likely reduced by 38.2  $\pm$  17.5% (-2.08  $\pm$  1.21) for HCHO and LCHO, respectively.

Differences in plasma glucose between HCHO and LCHO were unclear pre-  $(-1.86 \pm 0.96)$ and post-match ( $-0.06 \pm 0.77$ ). Changes in plasma glucose from pre- to post-match were unclear for both HCHO and LCHO groups with a  $-1 \pm 18.8\%$  reduction ( $-0.15 \pm 3.02$ ) and  $9.3 \pm 18.2\%$  increase ( $1.11 \pm 2.07$ ), respectively. Differences in plasma glycerol between HCHO and LCHO were unclear pre- ( $0.4 \pm 0.79$ ) and post-match ( $0.66 \pm 0.91$ ). Plasma glycerol most likely increased from pre- to post-match by  $52 \pm 24.4\%$  ( $2.94 \pm 1.38$ ) and by  $122 \pm$ 68% ( $1.70 \pm 0.64$ ) for HCHO and LCHO, respectively. Differences in NEFA between HCHO and LCHO were unclear pre-match ( $0.27 \pm 0.74$ ) and likely higher for LCHO post-match ( $0.95 \pm 0.93$ ). NEFA concentration most likely increased from pre-to post-match by  $172.6 \pm$  97.3%  $(1.21 \pm 0.42)$  and 226.7  $\pm$  175.9%  $(1.89 \pm 0.82)$  for HCHO and LCHO, respectively. Differences in overall total distance relative to playing time  $(0.22 \pm 0.63)$ , low intensity activity  $(-0.13 \pm 0.80)$  and high inten-sity running  $(-0.31 \pm 0.58)$  were unclear between HCHO and LCHO. Total distance relative to playing time from the first to the sec-ond half possibly decreased in HCHO ( $-0.30 \pm 0.45$ ) but was unclear for LCHO ( $-0.23 \pm 0.68$ ). A change in low intensity activity from the first and second half was unclear in HCHO ( $-0.08 \pm$ 0.65) and LCHO (0.04  $\pm$  0.60). Conversely, high intensity running in the sec-ond half was likely lower for HCHO ( $-0.55 \pm 0.36$ ) and very likely lower for LCHO ( $-1.42 \pm 0.69$ ). Differences in percentage of heart rate peak (%HRpeak) between HCHO and LCHO were unclear dur-ing the first (0.74  $\pm$  0.67) and second halves (0.01  $\pm$  0.82). %HRpeak possibly reduced from first to second half ( $-0.23 \pm 0.15$ ) and likely reduced ( $-0.48 \pm 0.63$ ) for HCHO and LCHO, respectively. Differ-ences in RHIE's between HCHO and LCHO were unclear during the first  $(0.32 \pm 0.42)$  and second halves  $(0.39 \pm 0.43)$ . A change in RHIE's from the first and second half were unclear in HCHO ( $0.19 \pm 1.24$ ) and LCHO ( $0.53 \pm 0.87$ ). First and second half move-ment characteristics for HCHO and LCHO are shown in Table 1b. Average urine osmolality was  $354.4 \pm 226.3$  and  $593.8 \pm 299$  mOsmol l-1 for HCHO and LCHO, respectively. HCHO were likely more hydrated than LCHO ( $0.74 \pm 0.67$ ). Average jump height was  $50.4 \pm 5.7$  cf.  $53.3 \pm 8.9$  cm pre-match,  $49.4 \pm 6.4$  cf.  $53.3 \pm 8$  cm post-match, and  $42.6 \pm 5.1$  cf.  $46.6 \pm 6.7$  cm 24-h post-match for HCHO and LCHO, respectively. Differences in CMJ between HCHO and LCHO were unclear pre-  $(0.39 \pm 1.05)$  post-  $(0.5 \pm 0.86)$  and 24h post-match (0.62  $\pm$  0.86). CMJ possibly reduced from pre- to post-match (1.01  $\pm$  0.54) and increases were likely trivial  $(0.37 \pm 0.57)$  for HCHO and LCHO, respectively. CMJ most likely reduced 24-h post-match ( $-1 \pm 0.26$ ) and ( $-0.75 \pm 0.16$ ) for HCHO and LCHO, respectively. 4. Discussion We provide novel data demonstrating that RL match-play can induce an approximate 40% glycogen depletion and moreover, manipulation of energy in the

form of CHO did not appear to affect resting glycogen availability, glycogen utilisation, or mark-ers of match-play work load. Analysis of players' internal and external loads revealed these data were consistent with com-petitive Super League match-play intensities.5 with mean total distance covered relative to match time of ~85-94 m min-1 and heart rates 78-83% HRpeak. Interestingly, no differences were found in total, high and low intensity running distance, RHIEs, or %HRpeak between the HCHO and LCHO conditions during the first or sec-ond half. In the second half, there were likely and very likely reductions in high intensity running distance, for the HCHO and LCHO groups, respectively. Our GPS data therefore suggest no major advantage of consuming more energy in form of CHO to enhance the high intensity running capabilities of professional rugby league players. However, tactical changes and the abil-ity of players to control locomotive rate means future studies using simulated match-play should be employed to confirm these findings. Despite major differences in the match day demands between soccer and rugby, similar pre- and post-match muscle glycogen concentrations were reported.8 The relatively high muscle glyco-gen concentrations observed in the present study were achieved after CHO intakes typically ingested by players during the training week (3 g kg-1; 240-330 g CHO) and leading up to competition17,18 (6 g kg-1; 480-660 g CHO) which could be described as low or rel-atively high CHO diets, respectively. Although no dietary analysis was performed before the study with the view of reducing player burden and maintaining ecological validity, we speculate that the training and nutrition of the players earlier in the week resulted in a reasonable muscle glycogen concentration before the 36-h CHO load leading into the game. This is pertinent given that the magni-tude of muscle glycogen re-synthesis is heavily influenced by the starting muscle glycogen concentration.25–27 Indeed, it is typical in rugby for training to taper towards match day whilst CHO intake gradually increases18 and therefore it is feasible that some of the players started the one day load with adequate muscle glycogen concentrations.

Moreover, 3–6 g kg–1 CHO in athletes with a high muscle mass, i.e. rugby players28 results in a total CHO intake of  $\sim$  300–600 g and might not be considered a low CHO intake for team-sport athletes. Given that the studies used to formulate CHO intake guidelines have recruited athletes with a much lower body mass, 19,29 we suggest absolute rather than relative amounts might be more appropriate when prescribing CHO recommendations for rugby players. However, this suggestion requires further investiga-tion. Interestingly, analysis of individual players suggests that the group receiving ~600 g of CHO for 36-h pre-match demonstrated a more homogenous pre-match glycogen concentration. We therefore speculate that  $\sim 600$  g of CHO be recommended for 36-h pre-match for competitive rugby players although future studies would be required to assess the efficacy of this suggestion as well as assessing glycogen utilisation in single fibres.8 A limitation of the present study was that post-match biopsies were collected over a 40-min period rather than at precisely the same time due Future studies might wish to take measures from fewer playersbut over a series of matches to facilitate a more rapid removal offissue as well as consider a half-time muscle biopsy. Analysis of the CMJ data suggested that there was no signifi-cant difference between the 2 groups in terms of recovery from the game. All players presented with significantly reduced CMJ theday after the game, which is likely due to muscle damage causing areduction in maximal force-generating capacity.30,31This decrease in CMJ was not affected by manipulating energy in the form of CHOin the days leading into the game, confirming previous suggestions hat pre-exercise CHO manipulation does not affect recovery fromdamaging exercise.32Increased lipid mobilisation was evidenced by large increasesin NEFA and glycerol concentrations after the match for both theHCHO and LCHO group. Considering the intermittent nature of aRL match, elevated lipid oxidation, despite what would appearsufficient muscle glycogen concentrations even at the end of thematch, might reflect periods of low intensity activity (walking orjogging) and when the ball was out of

play. Interestingly, pre- and post-match NEFA concentrations were similar to those observedin football8despite differences in match-play activity. Therefore, combined with the similar reductions in post-match muscle glyco-gen, our data suggest that RL and football might possess similarmetabolic demands.5. ConclusionWe have for the first time attempted to quantify the metabolic demands of professional RL match-play whilst manipulating energy in the form of CHO. We report that a competitive RL matchcan result in ~40% muscle glycogen depletion and that match-day performance variables did not differ between the 6 g kg-1or3 g kg-1CHO conditions. However, further analysis suggested thatthe higher CHO intake results in a more homogenous pre-matchglycogen concentration between the players. Therefore, despite nodifferences in movement characteristics between the low and thehigh CHO groups, we postulate that an absolute amount of ~600 gCHO 36-h pre-match is recommended strategy for rugby leagueplayers. Future studies might wish to further titrate these CHO rec-ommendations as well as assessing the effects of a rugby match onmuscle glycogen in individual muscle fibres.Practical implications•The present study has demonstrated that using the microbiopsytechnique, muscle biopsies are possible before and after a com-petitive rugby league match proving a framework for futureresearch in this sport.•Approximately 40% of muscle glycogen appears to be used dur-ing a competitive rugby league match, although the effects onindividual fibres remain unresolved. There were no noticeable differences in high intensity runningcapability between the two dietary groups, suggesting that thelower carbohydrate condition had no gross effects on performance.•Notwithstanding the limitations of interpreting movement char-acteristics of a single match, we propose that a diet comprising~600 g CHO 36 h before a match could be recommended to ensureall players commence the match with appropriate muscle glyco-gen concentrations.•It would appear that carbohydrate consumption in the week lead-ing into a rugby match is the major contributor to pre-matchmuscle glycogen concentration, rather than the carbohydrate be monitored by coaches and players. Acknowledgements The authors would like to thank Dan Owens, Kelly Hammond, Rob Allen, Carl Langan-Evans and Robert Naughton for their assis-tance with the data collection and all of the professional players for partaking in this study. The authors would also like to thank Professor Robert Cooper for his training in the collection of muscle biopsies. No financial assistance was provided for the preparation of the manuscript.

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