Muscle Glycogen Consumption and Recovery Kinetics Using ¹³C Magnetic Resonance Spectroscopy in Semi-Professional Soccer Players : A Case Study

Daisuke Nakamura^{*,**}, Mariko Nakamura^{***}, Yoko Tanabe^{****}, Keisuke Shiose^{*****}, Aya Ishibashi^{*****} and Hideyuki Takahashi^{****}

> *Rikkyo research institute of Wellness, Rikkyo University 3-34-1 Nishiikebukuro, Toshima-ku, Tokyo 171-8501 Japan **Weathernews Inc.
> Makuhari Techno Garden, Nakase 1-3 Mihama-ku, Chiba-shi, Chiba 261-0023 Japan daisuke.nakamura@ac.cyberhome.ne.jp
> ***Department of Sports Sciences, Japan Institute of Sports Sciences (JISS)
> ****Faculty of Health and Sport Sciences, University of Tsukuba
> ******Faculty of Education, University of Miyazaki
> *******Department of Life Sciences, The University of Tokyo

> > [Received April 2, 2021; Accepted June 28, 2021]

This study aimed to examine the relationship between thigh muscle glycogen consumption and recovery kinetics using ¹³C magnetic resonance spectroscopy (MRS) in semi-professional soccer players. Four male semi-professional soccer players participated in this study. Muscle glycogen levels were measured using ¹³C MRS before (Pre), within 2 h after the game (Post), 24 h after (A24h), and 48 h after (A48h) the Post. The rate of decrease in muscle glycogen levels tended to increase as high-intensity running distance (> 13 km \cdot h⁻¹; HIR) increased during the match. The recovery kinetics of muscle glycogen levels showed a strongly negative correlation with HIR (r=-1.00, p<0.05), and a moderately negative correlation with the total distance covered (TD) (r=-0.73, p>0.05) at A24h. Despite the small number of subjects, these results suggested that muscle glycogen consumption and recovery kinetics were affected by game performance, particularly HIR. Thus, these data could be beneficial as baseline data when planning an individualized recovery strategy after the match with taking into consideration of the match's external load.

Keywords: prolonged intermittent exercise, muscle glycogen, ¹³C magnetic resonance spectroscopy, soccer

[Football Science Vol.18, 43-50, 2021]

1. Introduction

Soccer is a prolonged intermittent exercise that involves various intensity exercises ranging from maximum intensity to low intensity running (Mohr et al., 2005). Given the activity duration (i.e., 80–120 min) and high-intensity intermittent profiles, muscle glycogen is considered the predominant energy substrate to support the metabolic demands during a match game. However, despite advances in the analysis of motor activity of soccer players (Randers et al., 2010; Varley et al., 2018), the relationship between muscle glycogen consumption and its recovery kinetics and each activity profile, such as distance covered in high intensity running, remains unclear.

The assessment of muscle glycogen was performed through invasive and non-practical muscle biopsies. However, ¹³C magnetic resonance spectroscopy (MRS) can measure muscle glycogen level noninvasively. Although some studies have reported a significant agreement between biopsy measurements and MRS methods at both high and low glycogen levels (Taylor et al., 1992), few studies have evaluated muscle glycogen level using ¹³C MRS in soccer players (Rico-Sanz et al., 1999a; Rico-Sanz et al., 1999b)

This study evaluated the game performance of semi-professional soccer players in an official match and investigated the relationship between thigh muscle glycogen consumption and recovery kinetics using ¹³C MRS. If these relationships are clarified, recovery strategies can be formulated according to each player's performance characteristics.

2. Methods

2.1. Subjects

Four male semi-professional soccer players from the Japan Football League (mean age, 23.3 ± 1.3 years; height, 174.5 ± 6.9 cm; body weight, 66.7 ± 3.9 kg; body fat, $8.5 \pm 0.8\%$) volunteered to participate in the study: two fullbacks (FB), one central midfielder (CM), and one forward (FW). Physical characteristics of each players are shown in **Table 1**.

2.2. Design

Participants stayed at the Japan Institute of Sports Sciences (JISS) for four days (the day before the game, day of the game, day after the game, and two days after the game). To avoid a large difference in total energy intake and excessive increase in body weight between the experimental period and before the experimental period, the participants consumed a prescribed diet formulated based on the results of a previous survey using the Excel Nutrition Youth Food Frequency Survey FFQg ver.3.5 (Kepakusha, Tokyo,

Table 1 Physical characteristics of each player

Japan, see **Table 2**). Bodyweight was measured every morning at the same time (6:45 am) and body fat was measured on the day of the game (Inbody 730, Inbody Japan, Tokyo, Japan). Muscle glycogen levels were measured after breakfast on the day of the game (Pre), within 2 h after the game (Post), 24 h after (A24h), and 48 h (A48h) after the Post. The participants did not perform any exercise, including recovery, for two days other than on the game day. The schematic representation of the experimental protocol was shown in **Figure 1**.

2.3. Muscle glycogen

All measurements were performed on the thigh muscle group of the right leg, and muscle glycogen concentration was measured using ¹³C MRS as described previously (Takahashi et al., 2015). To evaluate the decline and recovery of muscle glycogen level of each individual, the post and recovery of muscle glycogen level were evaluated based from the absolute value (mM) and relative value (%) which carunculate as follows; 100- {(absolute pre value-post absolute / pre value)*100 }.

2.4. Dietary control intake

The diet (total calorie intake) during the experiment period was calculated based on the daily intake of the participants. The caloric intake was determined,

	Age (yr)	Height (cm)	Body weight (kg)	Body fat (%)	LBM
Player A (FW)	25	167.3	61.6	7.5	57.0
Player B (FB)	23	171.8	65.5	8.2	60.1
Player C (FB)	23	175.1	69.7	8.9	63.5
Player D (CM)	22	183.6	69.8	9.2	63.4
Ave.	23	174.5	66.7	8.5	61.0

LBM; Lean body mass



 Table 2
 Planned and actual intake of total Energy and each nutrition intake during the experimental period

Figure 1 Schematic representation of the experimental protocol ↑; measurement MRS, ■; meals.

and the carbohydrate intake per body weight (BW) was 7 g/kg BW (Thomas et al., 2016). However, when the participants requested food or drinks other than those prescribed, they were permitted to consume a portion, and the nutritional contents were analyzed along with the content (e.g., amount, calorie intake) of the food and drinks provided at that time.

2.5. Game analysis

The in-game activities of the participants were evaluated using the 15 Hz global positioning system

the number of sprints over 22 km · h⁻¹ were also evaluated.
2.6. Statistics

The correlation coefficients between muscle glycogen level and total distance covered (TD) and

(SPI HPU, GPSport, Canberra, Australia). Based on

the report by Randers et al. (2010), activities were

categorized, and running distance with speeds of 13

km \cdot h⁻¹ or more was identified as HIR. Body impact (cut off value was < 5.0 g) (Abade et al., 2014) and

HIR performance were calculated using the Pearson's correlation coefficient at Post, A24h, and A48h. The significance level was set at < 5%. All statistical analyses were performed using TIBCO Spotfire[®] software.

2.7. Ethics

The study procedures were approved by the JISS Ethics in Human Research Committee, and all participants provided written informed consent before the start of the study. The study was conducted according to the Declaration of Helsinki.

3. Results

The carbohydrate intake for each subject post 24 h (Post -A24h) and 24–48 h (Post -A24h-A48) after the Post were shown in **Table 2**. TD and locomotor categories among individual players are given in

 Table 3
 TD and locomotor categories among individual players

Table 3. Figure 2 presents the absolute (Figure 2a) and relative changes (Figure 2b) in muscle glycogen levels before and after the match. Figure 3 shows the relationship between the decline in muscle glycogen levels and TD (Figure 3a) and HIR (Figure 3b). Figure 4 shows the relationship between the recovery rate of muscle glycogen levels and the TD (Figure 4a) and HIR (Figure 4b) at 24 h and 48 h after the game.

4. Discussion

This study showed that the thigh muscle glycogen level after the match decreased compared to the Pre value (**Figure 2**). This study also showed that as HIR increases, the rate of decrease in muscle glycogen levels tends to accelerate (**Figure 3b**). In addition, when we examined the recovery rate of muscle glycogen levels 24 hours after the match, we observed a strong negative correlation (r=-1.00, p<

	TD (km)	0-2 Km ∙h ⁻¹ (km)	2-7 Km∙h ⁻¹ (km)	7-9 Km∙h ⁻¹ (km)	9-13 Km•h ⁻¹ (km)	13-16 Km ∙h ⁻¹ (km)	16-22 km ∙h⁻¹ (km)	Sprint 22km∙h⁻¹< (km)	HIR >13km•h ⁻¹ (km)	Sprint (n)	Body impact (n)
Player A (FW)	11.2	0.1	3.9	1.2	2.8	1.3	1.4	0.6	3.3	30	1520
Player B (FB)	10.7	0.1	3.6	1.2	2.5	1.3	1.4	0.5	3.2	23	751
Player C (FB)	10.4	0.1	3.7	1.2	2.8	1.3	1.0	0.2	2.6	10	778
Player D (CM)	10.7	0.1	3.9	1.3	2.9	1.4	0.9	0.2	2.5	12	762

TD; total distance covered, HIR; high-intensity running distance ($> 13 \text{ km} \cdot \text{h}^{-1}$)







Figure 3 The relationship between the decline of muscle glycogen levels (%) and the TD (a) and HIR (b) TD; total distance covered, HIR; high-intensity running distance (speed > 13 km \cdot h⁻¹)



Figure 4 The relationship between the recovery rate of muscle glycogen levels and the total distance covered (a) and HIR (b) at 24 hours after the Post (left) and 48 hours after the Post (right)

The horizonal dotted line means the Pre value.

TD; total distance covered, HIR; high-intensity running distance (speed > 13 km \cdot h⁻¹)

0.05, Figure 4b) with HIR, and a moderate negative correlation with TD (r=-0.73, p>0.05, Figure 4a). These results suggest that the rate of decrease and recovery in muscle glycogen level may be largely influenced by high-intensity activity during a match.

Our results were similar to previous studies (Gunnarsson et al., 2013; Krustrup et al., 2011) using muscle biopsies in that thigh muscle glycogen levels after the match decreased when compared with the Pre values (Figures 2). The rate of decrease in muscle glycogen level after the match was 30.5±7.7%, which is slightly lower than that observed by Krustrup et al., who demonstrated a 43% decrease in vastus lateralis muscle glycogen levels before and after a soccer match (Krustrup et al., 2011). Furthermore, Gunnarsson et al. reported that muscle glycogen levels were reduced by approximately 54% after a simulated soccer match exercise (Gunnarsson et al., 2013). The difference in results between the present study and those of previous studies (Gunnarsson et al., 2013; Krustrup et al., 2011) may be due to differences in exercise intensity; the TD in Krustrup et al.'s study (Krustrup et al., 2011) and the TD in Gunnarsson et al.'s study (high intensity running over 18 km \cdot h⁻¹: 3.2 km) was higher than that in the present study (HIR: 2.9 km). However, these two studies evaluated the game performance not using GPS device, it is comparing to simply these two results with the present results.

Post-match carbohydrate intake affects the recovery of muscle glycogen levels (Burke et al., 2017; Costil, 1980). In this study, as American college of sports medicine guidelines indicate that athletes who performed a 90-minute exercise require an intake of 7-12 g / kg of carbohydrate per day, the post-match carbohydrate intake was set at 7g/kg BW. However, carbohydrate intake from post-match to 24 h postmatch and from 24 h to 48 h post-match was 8.1 g/kg BW and 7.9 g/kg BW on average, respectively (Table 2) due to allowing to intake other than the prescribed diet. This amount of carbohydrate intake was lower than that reported by Krustrup et al. which intakes 9.5 g of carbohydrates of BW per day (Krustrup et al., 2011). Although the carbohydrate intake after the match was lower in our study than in Krustrup et al.'s study, glycogen levels returned to baseline 48 h post-match in all participants. Meanwhile, when the recovery kinetics were examined individually, subjects A (FW) and B (FB) did not recover their muscle glycogen levels in A24h; in contrast, subjects

C (FB) and D (CM) recovered their muscle glycogen levels within A24h (**Figure 2b**). Interestingly, subjects A and B performed more sprints and covered a higher total distance in HIR compared to subjects C and D (**Table 3**).

Previous studies suggested that muscle damage could influence the resynthesis of muscle glycogen levels (Asp et al., 1996; Kiens and Richter, 1998; Krustrup et al., 2011; Zehnder et al., 2001). Muscle damage markers were not measured in the present study, but we believe that damage might have occurred. Some previous studies reported that highintensity activities were associated with increasing creatine kinase concentrations after the match (Russell et al., 2016; Thorpe and Sunderland, 2012). It has been shown that exercise transiently decreases the skeletal muscle glucose transporter GLUT4 protein content (Asp et al., 1995a) and impairs muscle glycogen resynthesis after exercise(Asp et al., 1995b). In the present study, Subjects A and C had similar reductions in muscle glycogen levels after the match (29% and 30%, respectively, Figure 2b), and the levels in subject C (FB) returned to normal after 24 h, but were unchanged in subject A (FW). In addition, subject A tended to cover more distance in HIR and had higher sprint numbers and TD than subject C. Moreover, as increasing in HIR or TD, there was a trend to the slower recovery of muscle glycogen (Figure 4a, Figure 4b). These performance-related data suggested that subject A (FW) might have had greater muscle damage than subject C (FB); thus, the time lag in recovery arises. From these data, although there was no data on muscle damage, muscle damage may explain the relationship between the recovery kinetics of muscle glycogen levels and the HIR and TD, particularly until 24 hours recovery after the match (Figure 4b, Figure 4a).

The results of this study have practical applications. After a competitive match, the recovery rate of muscle glycogen levels is affected by the running performance during a match. Given that muscle damage affects the recovery of muscle glycogen levels, fitness and conditioning coaches need to identify individualized recovery strategies involving cold water immersion which has an effect on muscle damage (e.g. creatine kinase) (Pooley et al., 2019), appropriate carbohydrate intake (Costill et al., 1990), and the promotion of recovery according to playing position with a lot of external loads (i.e. high intensity activities). Despite our small sample size, our results can be used as baseline data when planning an optimal nutrition/ recovery strategy for soccer players in consideration of their position and play characteristics.

This study also has some limitations. Fist, we did not measure muscle damage markers, which are thought to influence the recovery of muscle glycogen levels. However, as mentioned above, previous studies reported that the relationship between HIR and increase in creatine kinase concentrations after the match (Russell et al., 2016; Thorpe and Sunderland, 2012). The small sample size is another limitation of this study. This limitation likely to related to using ¹³C MRS methods to quantify the muscle glycogen level. Unlike muscle biopsy, this method is useful for evaluating muscle glycogen in soccer players in that it can evaluate a wider range of muscle glycogen kinetics in a non-invasive manner, but it is necessary to further analysis using more subjects. The present study only examined one condition of the carbohydrate intake, at 7 g/kg of BW, and this was likely to affect the muscle glycogen recovery kinetics. Future studies should investigate whether carbohydrate intake would exert a different effect on the recovery kinetics of muscle glycogen after a match.

5. Conclusion

In conclusion, the results of the present study provided evidence that muscle glycogen levels evaluated using ¹³C MRS immediately after a match decreased when compared with those before a match and that the recovery rate was related to the HIR during a match. Thus, these data could be beneficial as baseline data when planning an individualized recovery strategy after the match with taking into consideration of the match's external load.

References

- Abade, E. A., Goncalves, B. V., Leite, N. M., and Sampaio, J. E. (2014). Time-motion and physiological profile of football training sessions performed by under-15, under-17 and under-19 elite Portuguese players. Int. J. Sports Physiol. Perform., 9: 463-470.
- Asp, S., Daugaard, J. R., Kristiansen, S., Kiens, B., and Richter, E. A. (1996). Eccentric exercise decreases maximal insulin action in humans: Muscle and systemic effects. J. Physiol., 494 (Pt 3): 891-898.
- Asp, S., Daugaard, J. R., and Richter, E. A. (1995a). Eccentric exercise decreases glucose transporter GLUT4 protein in human skeletal muscle. J. Physiol., 482 (Pt 3): 705-712.

- Asp, S., Kristiansen, S., and Richter, E. A. (1995b). Eccentric muscle damage transiently decreases rat skeletal muscle GLUT-4 protein. J Appl Physiol (1985), 79: 1338-1345.
- Burke, L. M., van Loon, L. J. C., and Hawley, J. A. (2017). Postexercise muscle glycogen resynthesis in humans. J Appl Physiol (1985), 122: 1055-1067.
- Costil, D. M., JM. (1980). Nutrition for Endurance Sport: Carbohydrate and Fluid Balance. Int. J. Sports Medicine, 1: 2-14.
- Costill, D. L., Pascoe, D. D., Fink, W. J., Robergs, R. A., Barr, S. I., and Pearson, D. (1990). Impaired muscle glycogen resynthesis after eccentric exercise. J Appl Physiol (1985), 69: 46-50.
- Gunnarsson, T. P., Bendiksen, M., Bischoff, R., Christensen, P. M., Lesivig, B., Madsen, K., Stephens, F., Greenhaff, P., Krustrup, P., and Bangsbo, J. (2013). Effect of whey proteinand carbohydrate-enriched diet on glycogen resynthesis during the first 48 h after a soccer game. Scand. J. Med. Sci. Sports, 23: 508-515.
- Kiens, B. and Richter, E. A. (1998). Utilization of skeletal muscle triacylglycerol during postexercise recovery in humans. Am. J. Physiol., 275: E332-337.
- Krustrup, P., Ortenblad, N., Nielsen, J., Nybo, L., Gunnarsson, T. P., Iaia, F. M., Madsen, K., Stephens, F., Greenhaff, P., and Bangsbo, J. (2011). Maximal voluntary contraction force, SR function and glycogen resynthesis during the first 72 h after a high-level competitive soccer game. Eur. J. Appl. Physiol., 111: 2987-2995.
- Mohr, M., Krustrup, P., and Bangsbo, J. (2005). Fatigue in soccer: A brief review. J. Sports Sci., 23: 593-599.
- Pooley, S., Spendiff, O., Allen, M., and Moir, H.J. (2020). Comparative efficacy of active recovery and cold water immersion as post-match recovery interventions in elite youth soccer. J. Sports Sci., 38, 1423-1431. doi: 10.1080/02640414.2019.1660448.
- Randers, M. B., Mujika, I., Hewitt, A., Santisteban, J., Bischoff, R., Solano, R., Zubillaga, A., Peltola, E., Krustrup, P., and Mohr, M. (2010). Application of four different football match analysis systems: A comparative study. J. Sports Sci., 28: 171-182.
- Rico-Sanz, J., Zehnder, M., Buchli, R., Dambach, M., and Boutellier, U. (1999a). Muscle glycogen degradation during simulation of a fatiguing soccer match in elite soccer players examined noninvasively by ¹³C-MRS. Med. Sci. Sports Exerc., 31: 1587-1593.
- Rico-Sanz, J., Zehnder, M., Buchli, R., Kuhne, G., and Boutellier, U. (1999b). Noninvasive measurement of muscle high-energy phosphates and glycogen concentrations in elite soccer players by ³¹P- and ¹³C-MRS. Med. Sci. Sports Exerc., 31: 1580-1586.
- Russell, M., Sparkes, W., Northeast, J., Cook, C. J., Bracken, R. M., and Kilduff, L. P. (2016). Relationships between match activities and peak power output and Creatine Kinase responses to professional reserve team soccer match-play. Hum. Mov. Sci., 45: 96-101.
- Takahashi, H., Kamei, A., Osawa, T., Kawahara, T., Takizawa, O., and Maruyama, K. (2015). ¹³C MRS reveals a small diurnal variation in the glycogen content of human thigh muscle. NMR Biomed., 28: 650-655.
- Taylor, R., Price, T. B., Rothman, D. L., Shulman, R. G., and Shulman, G. I. (1992). Validation of ¹³C NMR measurement of human skeletal muscle glycogen by direct biochemical assay of needle biopsy samples. Magn. Reson. Med., 27: 13-20.
- Thomas, D. T., Erdman, K. A., and Burke, L. M. (2016). American College of Sports Medicine Joint Position Statement. Nutrition

and Athletic Performance. Med. Sci. Sports Exerc., 48: 543-568.

- Thorpe, R. and Sunderland, C. (2012). Muscle damage, endocrine, and immune marker response to a soccer match. J. Strength. Cond. Res., 26: 2783-2790.
- Varley, M. C., Di Salvo, V., Modonutti, M., Gregson, W., and Mendez-Villanueva, A. (2018). The influence of successive matches on match-running performance during an under-23 international soccer tournament: The necessity of individual analysis. J. Sports Sci., 36: 585-591.
- Zehnder, M., Rico-Sanz, J., Kuhne, G., and Boutellier, U. (2001). Resynthesis of muscle glycogen after soccer specific performance examined by ¹³C-magnetic resonance spectroscopy in elite players. Eur. J. Appl. Physiol., 84: 443-447.



Name: Daisuke Nakamura

Affiliation: *Rikkyo Research Institute of Wellness, Rikkyo University **Weathernews Inc.

Address:

*3-34-1 Nishiikebukuro, Toshima-ku, Tokyo 171-8501 Japan **Makuhari Techno Garden, Nakase 1-3 Mihama-ku, Chiba-shi, Chiba 261-0023 Japan

Brief Biographical History:

Daisuke Nakamura Ph.D.

2001-2012 Seikei University, Part-time lecturer.

2003-2018 Rikkyo University, Part-time lecturer.

2004-2010 Yokogawa Musashino football club, Coach.

2014-2019 Japan Institute of Sports Sciences, Researcher.

2019-2020 The Department of Sport and Wellness, Rikkyo University, Associate professor.

Main Works:

• Book;

- Nakamura, D., "Water intake", "Dehydration and performance", "Heat countermeasures in children", etc In: Heat countermeasures in sports activity. edited by Hiroshi Hasegawa and Daisuke Nakamura, NAP, 2021.
- Papers;

Nakamura, D.(2021). Heat countermeasures in soccer. The Journal of Japanese Society of Science and Football,16:10-26.

- Nakamura, D., Tanabe, Y., Arimitsu, T., Hasegawa, H., and Takahashi, H. (2020). Low caffeine dose improves intermittent sprint performance in hot and humid environments. J. Therm. Biol., 93:102698. doi 10.1016/j.jtherbio.2020.102698.
- Nakamura, D., Muraishi, K., Hasegawa, H., Yasumatsu, M., and Takahashi, H. (2020). Effect of a cooling strategy combining forearm water immersion and a low dose of ice slurry ingestion on physiological response and subsequent exercise performance in the heat. J. Therm. Biol., 89:102530. doi:10.1016/j.jtherbio.2020.102530.
- Nakamura, D., Suzuki, T., Yasumatsu, M., and Akimoto, T. (2012). Moderate running and plyometric training during offseason did not show significant difference on soccer related high-intensity performances compared with no-training controls. J. Strength Cond. Res., 26(12):3392-3397. doi: 10.1519/JSC.0b013e3182474356.

etc.

Membership in Learned Societies:

• Japanese Society of Science and Football