Paper : Nutrition

Recommendation for Higher Energy and Protein Intake for Match Days Compared with Training Days in Japanese Male Senior High School Soccer Players

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This study aimed to recommend the energy and protein requirements on general training and match days for Japanese male senior high school soccer players. Seven players were surveyed and measured during two consecutive days of training (training group). Nine players were surveyed and measured during two consecutive days of expeditions (match group). The recommended energy intake (EI) was estimated using total energy expenditure (TEE) measured by Misfit Shine, a triaxial acceleration sensor-mounted activity meter. The recommended protein intake was estimated using measured values obtained by the nitrogen balance (NBAL) method. Results suggested that EI should be 48–52 kcal/kg/day on training days and 49–54 kcal/kg/day on match days. Results also suggested that protein intake should be 0.8–1.5 g/kg/day on training days and 1.4–2.4 g/kg/day on match days. Moreover, we believe that the recommended energy and protein intake for Japanese male senior high school soccer players on match days should be higher than on training days.

Keywords: energy, protein, male senior high school soccer players, triaxial acceleration sensor-mounted activity meter, nitrogen balance method

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1. Introduction

Appropriate energy intake (EI) is the cornerstone of an athlete’s diet as it supports optimal body function, determines the macro and micronutrient intake capacity, and influences body composition (Thomas et al., 2016). The International Society of Sports Nutrition (ISSN) recommends 40–70 kcal/kg/day energy requirement for moderate-to-high intensity athletes (Kerksick et al., 2018). When considering nutritional requirements for athletes, it is necessary to first understand the daily total energy expenditure (TEE) of the athletes including training. At present, the gold standard for measuring TEE in field studies involving humans is the doubly labeled water (DLW) method. Previous studies have suggested TEE of professional soccer players in Japan, Korea, and Britain using this method and reported a range of approximately 45–55 kcal/kg/day (Ebine et al., 2002; Kim et al., 2003; Anderson et al., 2017). However, the DLW method is expensive. Furthermore, although the stable isotopes of hydrogen and oxygen used in the DLW method are completely different in nature from the radioactive isotopes, the word “isotope” is often associated with radioactivity in Japan; this negative connotation of this term is associated with the country’s history of nuclear explosion (Ebine et al., 2002). The acceleration sensor-mounted activity meter has a lower introduction cost than the DLW method, places less burden on the subject than the breath gas analysis method, and has a higher validity and reliability than the pedometer and questionnaire methods (Sasai et al., 2015). At present, the triaxial acceleration sensor-mounted activity meter is an inexpensive and feasible method for estimating daily physical activity (Fokkema et al., 2016). Most of these activity meters can measure acceleration in three directions and be used to estimate the type of movement, count steps, calculate TEE and energy intensity, and estimate sleep patterns, among other functions (Henriksen et al., 2018). It has been found that the steps, distance, TEE, and sleep parameters of some consumer-based devices included in the triaxial acceleration sensor-mounted activity meter are reliable (Evenson et al., 2015). In short, the triaxial acceleration sensor-mounted activity meter has more practical applications than the DLW method; however, but studies using these activity meters are limited to be able to recommend the energy requirements for athletes.

Athletes require more protein to maintain a positive muscle protein balance because the oxidation of amino acids and degradation of muscle protein increase with increased TEE and intense activity (Thomas et al., 2016). Previous reports that have suggested the optimal protein intake to be 1.2–2.0 g/kg/day should be considered (Phillips et al., 2016; Witard et al., 2016; Jager et al., 2017; Tipton and Witard, 2007). In Japan, protein requirements of 2.00 g/kg/day for power sports, 1.75 g/kg/day for ball sports, and 1.50 g/kg/day for endurance sports have been shown based on previous studies in Europe and the United States (Lemon et al., 1984; Brouns et al., 1989; Lemon, 1991; Lemon, 1994; Lemon, 1996; Lemon, 1998). The nitrogen balance (NBAL) method is the gold standard method to assess protein requirements and metabolism in a free-living state (Matsuda et al., 2018). The protein requirements of adolescent soccer players in France were suggested based on the NBAL method (Boisseau et al., 2002; Boisseau et al., 2007). However, similar studies using Japanese soccer players are limited.

This study aimed to recommend the energy and protein requirements for Japanese male senior high school soccer players on general training and match days.

2. Methods

2.1. Participants and study design

All participants and their parents were informed about the study procedures and any associated potential risk before signing the written assent and consent documents, respectively. This study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the research ethics board of the Kawasaki University of Medical Welfare (No. 17–025).

In this study, we evaluated the body composition, nutrient intake status, TEE, and total urinary nitrogen of 35 Japanese male senior high school soccer players who submitted the consent forms. Of the 35 participants, 16 were free of illness or injury and able to complete all investigations and measurements. However, the remaining 19 participants were excluded from the analysis because they were unable to complete urine collection. Out of the 16 players, 9 were surveyed, and their energy and protein requirements were measured during two consecutive days of match in September 2017 to simulate a match period (match group). The remaining 7 players were surveyed, and their energy and protein requirements were measured during two consecutive days of training in September 2018 to simulate a training period (training group).

2.2. Protocol

Figure 1 shows an overview of the study protocol. In the training group, the training lasted 2 h (9:00 am–11:00 am). The first day comprised a 30-min warm-up, followed by seven sets of 10-min intrasquad matches, and finally, seven types of strength training (i.e., reverse lunge, deadlift, pull-up, bench press, sit-up, side plank, and Russian twist) for 3–5 sets with a load of 8–12 repetition maximum (RM). The second day comprised a 30-min warm-up, followed by two sets of 30-min intrasquad matches, and finally, seven types of strength training (i.e., back squat, Romanian deadlift, bent over row, overhead press, front bridge, side vent, and pall of press) for 3–5 sets with a load of 8–12 RM. The subjects in the training group primarily consumed meals cooked by their parents at home and their own food purchased at convenience stores on the first and second days. In the match group, practice matches (70-min/match) with soccer clubs from other high schools were played. On the first day, they played one and a half practice match from 1:00 pm–3:30 pm after a 30-min warm-up session. On the second day, they played one practice match from 1:00 pm–2:30 pm after a 30-min warm-up session. The subjects in the match group primarily consumed meals cooked by their parents at home and their own food purchased at convenience stores for

Subjects	First day	Second day	Third day
Training group (n=7)	30-min warm-up; seven sets of 10-min intrasquad matches; 7 types of strength training	30-min warm-up; two sets of 30-min intrasquad matches; 7 types of strength training	
Match group (n=9)	30-min warm-up; One and a half practice match (70- min/match)	30-min warm-up; One practice match (70-min/match)	

Survey and measurement	First day	Second day	Third day
Body composition	Body height: medical examination at the high school. Body mass and body composition: measured before breakfast.		
Nutrient intake	Training group: data on the names and weights of all foods consumed by the subjects during the two consecutive days were collected from the subjects or their guardians. Match group: data on the names and weights of meals cooked by parents at home and foods purchased by the subjects at convenience stores were collected from subjects or their parents. a dietitian weighed food prepared by the accommodation and team staff.		
TEE	Misfit Shine (which is equipped with a triaxial acceleration sensor) was attached to each subject's non-dominant hand using the accessory armband.		
Urine collected	The second urine output on the first day to the first urine output on the second day.	The second urine output on the second day to the first urine output on the third day.	

Figure 1 Overview of the study protocol

breakfast and lunch on the first day and dinner on the second day. Meanwhile, meals prepared by the accommodation and team staff were consumed for dinner on the first day and breakfast and lunch on the second day.

The height used was the value obtained from the medical examination at the high school to which a subject belongs. Body mass and composition (e.g., skeletal muscle mass, body fat mass, skeletal muscle ratio, body fat ratio, and lean body mass) were measured before breakfast on the first day in the training and match groups. Body mass was measured using a innerScanDual RD-902 (TANITA, Co., Ltd.) . Body composition were measured using a physion MD (Nippon Shooter, Co., Ltd.) with the bioelectric impedance (BI) method. Nutrient intake was assessed by the analysis of daily food records. In the training group, data on the names and weights of all foods consumed by the subjects during the two consecutive days were collected from the subjects or their guardians. Photographs of the foods were also taken before consumption. In the match group, data on the names and weights of meals cooked by parents at home and foods purchased by the subjects at convenience stores were collected from subjects or their parents. Photographs of the foods were taken before consumption. A dietitian weighed food prepared by the accommodation and team staff. Nutritional value was calculated by averaging the two days survey values using Excel EIYOUKUN Ver. 8.0 (KENPAKUSYA, Co., Ltd.) (Takahashi, 2003), and daily nutrient (i.e., energy, proteins, lipids, carbohydrates, dietary fiber, vitamin B_1, vitamin C, calcium, and iron) intakes were calculated. The energy and energy-producing nutrient (i.e., proteins, lipids, and carbohydrates) requirements were also calculated per body mass. TEE was measured using the Misfit Shine (MISFIT, Inc.), which is equipped with a triaxial acceleration sensor. The Misfit Shine measures 27.5 mm in width, 3.3 mm in depth, and 27.5 mm in height; it weighs 9.4 g and is waterproof, so it does not need to be removed when taking a bath. Most TEE values are calculated from the basal metabolic rate (BMR). BMR is estimated by the prediction formula of Mifflin-St Jeor (Frankenfield et al., 2005). A previous study has shown the accuracy of measuring TEE compared with the metabolic chamber and DLW methods (Murakami et al., 2016). Furthermore, the measurement accuracy in the free-living condition has also been verified (Brooke et al., 2017). The device was attached to each subject's non-dominant hand using the accessory armband.

In addition, to prevent the equipment from falling off during training or matches, the wearing site was covered with a cotton wristband or tape. After confirming the measurement data with smartphone-application data associated with the device, the average of the measured values for the two days was calculated to determine TEE per day, TEE per body mass, EE during exercise, and EE during exercise per body mass. Diet-induced thermogenesis, which was estimated to be 10% of EI, was added to the calculation of TEE. Urinemate P (Sumitomo Bakelite, Ltd.) was distributed to the subjects for urine collection. The second urine output on the first day to the first urine output on the second day and the second urine output on the second day to the first urine on the third day were collected. Urinemate P is a two-layer structure comprising an upper and a lower chamber; a 1/50 volume of urine excreted in the upper chamber can flow into the lower chamber by opening and closing the cock to accurately reflect the component ratio in urine for 24 h (Yasutake et al., 2014; Sakuma et al., 2017). The collected urine sample was dispensed into a storage container to measure the amount of urine; it was then frozen and stored at ≤−18°C. Thereafter, the sample was thawed in a laboratory at a university, and the total nitrogen content in the urine was measured by the micro-Kjeldahl method (Shepard and Jacobs, 1951). For practical reason, the amounts of fecal and transdermal nitrogen excretions were not measured. In the present study, an estimated value of 15.0 mg/kg was used (FAO/OMS/UNU, 1986). The nitrogen balance was calculated by subtracting the excreted nitrogen (total urine, fecal, and transcutaneous nitrogen excretion) from the nitrogen intake. Nitrogen intake was calculated by dividing protein intake by the nitrogen protein conversion factor (6.25) (Ministry of education culture sports science and technology, 2016). Then, the necessary amount for nitrogen equilibrium maintenance (protein intake per body mass when the nitrogen balance value is 0 g/day) was determined from the nitrogen balance value and protein intake per body mass. The utilization efficiency (85%) of the daily dietary mixed protein was taken into consideration in calculating the necessary amount for nitrogen equilibrium maintenance (Ministry of Health Labour and Welfare, 2020). Our study subjects were in a growing period, and they needed more protein depositions accumulated with growth. The estimated protein requirement was obtained by adding protein depositions accumulated with growth to the protein required (0.035 g/kg) to maintain nitrogen equilibrium (Ministry of Health Labour and Welfare, 2020). The recommended amount of protein was obtained by multiplying the estimated protein requirement and recommended dietary allowance coefficient of 1.25 (Ministry of Health Labour and Welfare, 2020).

2.3. Statistical analysis

Statistical analysis software SPSS Ver. 26.0 (IBM Japan, Ltd.) was used for statistical processing. Normality of the data was confirmed by the Kolmogorov-Smirnov test. To compare the parameters between the training and match groups, an unpaired *t*-test was used for items with normally distributed data and a Mann-Whitney U test was used for items with non-normally distributed data. To compare energy balance values within the training and match groups, a paired *t*-test was used for items with normally distributed data. The 95% confidence intervals (CI) for TEE per body mass and EI per body mass were determined by multiplying the standard error by 1.96. The recommended protein intake was calculated using Pearson's product-moment correlation coefficient between the nitrogen balance value and protein intake per body mass in the training and match groups. The 95% CI for the required protein intake for maintaining nitrogen equilibrium was determined by multiplying the standard error of the regression line by 1.96. The significance level was set at <5% in all cases.

3. Results

3.1. Body composition

Table 1 shows the body composition of the training and match groups. There was no significant difference in body composition between the two groups. The individual's body heights and body masses fell between the 3rd and 97th percentiles on the growth curve (Nato et al., 2004).

3.2. Nutrient intake status

Table 2 shows the nutrient intake status of the training and match groups. Dietary fiber, vitamin

Table 1 Body composition of the training and match groups

	training group (n=7)			match group (n=9)			Significant difference*
Body height, cm	170	±	4	171	±	6	p=0.597
Body mass, kg	59.7	±	2.6	64.4	±	7.5	p=0.111
Skeletal muscle mass, kg	26.6	±	1.7	27.4	±	2.7	p=0.474
Skeletal muscle rate, %	44.5	±	3.1	42.7	±	2.0	p=0.178
Body fat mass, kg	4.6	±	1.6	7.2	±	3.8	p=0.110
Body fat rate, %	7.6	±	2.5	10.8	±	4.8	p=0.138
Lean body mass, kg	55.2	±	1.9	57.2	±	4.1	p=0.248

Data are represented as mean ± standard deviation.

* To compare the parameters between the training and match groups, an unpaired *t*-test was used for items with normally distributed data and a Mann-Whitney U test was used for items with non-normally distributed data.

C, calcium intakes in the training group were significantly higher than in the match group (dietary fiber: $p < 0.001$, vitamin C: $p = 0.004$, and calcium: $p = 0.033$). However, there were no significant differences in other nutrient intakes between the two groups.

3.3. TEE and energy balance

Table 3 shows TEE of the training and match groups. TEE, EE during exercise, and EE during exercise per body mass in the match group were significantly higher than those in the training group (TEE: $p = 0.009$, EE during exercise: $p = 0.005$, and EE during exercise per body mass: $p = 0.008$). Conversely, there was no significant difference between groups for TEE per body mass and BMR.

Figure 2 shows the energy balance per body mass of the training and match groups. TEE per body mass values were 50 ± 2 kcal/kg/day (CI: 48–52 kcal/kg/day) and 52 ± 4 kcal/kg/day (CI: 49–54 kcal/kg/day) for the training and match groups, respectively. EIs per body mass were 52 ± 5 kcal/kg/day (CI: 48–55 kcal/kg/day) and 53 ± 6 kcal/kg/day (CI: 49–57 kcal/kg/day) for the training and match groups, respectively. There was no significant difference for energy balance in either group.

3.4. Nitrogen balance and protein requirements

Table 4 shows the total urinary nitrogen content and nitrogen balance of the training and match groups. The total urinary and excreted nitrogen levels were significantly higher in the match group than in the training group ($p < 0.001$). There was no significant difference between the groups in the nitrogen intake. The nitrogen balance value was significantly higher in the training group than in the match group ($p = 0.003$).

Figure 3 shows the nitrogen balance and protein intake per body mass of the training and match groups. There were significant positive correlations between the nitrogen balance and protein intake per body mass in both the training ($y = 10.960x - 13.048$, $r = 0.745$, $p = 0.021$) and match ($y = 10.118x - 7.388$, $r = 0.786$, $p = 0.036$) groups. The required protein intake for maintaining nitrogen equilibrium was 0.73 g/kg/day (CI: 0.5–1.0 g/kg/day) in the training group and 1.19 g/kg/day (CI: 0.9–1.5 g/kg/day) in the match group. The recommended protein intake was 0.8–1.5

Table 2 Nutrient intake status of the training and match groups

	training group (n=7)			match group (n=9)			Significant difference*
Energy, kcal/day	**3085**	**±**	**375**	**3389**	**±**	**298**	**p=0.091**
Energy per body mass, kcal/kg/day	**52**	**±**	**5**	**53**	**±**	**6**	**p=0.616**
Protein, g/day	**110**	**±**	**11**	**113**	**±**	**13**	**p=0.635**
Protein per body mass, g/kg/day	**1.8**	**±**	**0.2**	**1.8**	**±**	**0.2**	**p=0.449**
Fat, g/day	**89**	**±**	**10**	**88**	**±**	**10**	**p=0.863**
Fat per body mass, g/kg/day	**1.5**	**±**	**0.2**	**1.4**	**±**	**0.2**	**p=0.325**
Carbohydrate, g/day	**443**	**±**	**83**	**511**	**±**	**53**	**p=0.068**
Carbohydrate per body mass, g/kg/day	**7.4**	**±**	**1.2**	**8.0**	**±**	**1.0**	**p=0.114**
Dietary fiber, g/day	**17**	**±**	**2**	**12**	**±**	**2**	**p<0.001**
Vitamin B_1, mg/day	**1.5**	**±**	**0.3**	**1.2**	**±**	**0.1**	**p=0.067**
Vitamin C, mg/day	**136**	**±**	**53**	**55**	**±**	**23**	**p=0.004**
Calcium, mg/day	**734**	**±**	**172**	**538**	**±**	**159**	**p=0.033**
Iron, mg/day	**9.5**	**±**	**1.6**	**8.1**	**±**	**0.7**	**p=0.051**

Data are represented as mean ± standard deviation.

* To compare the parameters between the training and match groups, an unpaired *t*-test was used for items with normally distributed data and a Mann-Whitney U test was used for items with non-normally distributed data.

g/kg/day for the training group and 1.4–2.4 g/kg/day for the match group.

4. Discussion

This study is one of the first to clarify the energy and protein requirements for Japanese male senior high school soccer players on general training and match days by field research. The recommended EI was estimated from TEE measured by the triaxial acceleration sensor-mounted activity meter. We found that recommended EIs were 48–52 kcal/kg/day and 49–54 kcal/kg/day on the training and match days, respectively. The required proteins for maintaining nitrogen equilibrium were calculated at 0.73 g/kg/day and 1.19 g/kg/day on the training and match days, respectively. Besides, the recommended protein intakes were 0.8–1.5 g/kg/day and 1.4–2.4 g/kg/day on the training and match days, respectively.

In this study, the recommended EIs in the training and match groups were within the range of recommended energy requirements for moderate-to-high intensity athletes by ISSN. In addition, it was similar to TEE range of professional soccer players estimated by the DLW method. The DLW method is the gold standard for measuring TEE in human field research. In this study, we used an activity

Table 3 TEE of the training and match groups

	training group (n=7)			match group (n=9)			Significant difference*
TEE , kcal/day	2986	±	163	3318	±	253	p=0.009
TEE per body mass, kcal/kg/day	50	±	2	52	±	4	p=0.264
EE during exercise, kcal/day	468	±	124	764	±	191	p=0.005
EE during exercise per body mass, kcal/kg/day	8	±	2	12	±	3	p=0.008
BMR, kcal/day	1581	±	48	1635	±	104	p=0.193
BMR per body mass, kcal/kg/day	26	±	2	26	±	0	p=0.105

Data are represented as mean ± standard deviation. TEE: total energy expenditure. EE: energy expenditure. BMR: basal metabolic rate (estimated from the prediction equations Mifflin-St Jeor).

* To compare the parameters between the training and match groups, an unpaired *t*-test was used for items with normally distributed data and a Mann-Whitney U test was used for items with non-normally distributed data.

meter called Misfit Shine, which is equipped with a triaxial acceleration sensor in consideration of practicality. However, the use of activity meters does have limitations. Isometric or resistance exercises may burn a substantial of calories, but activity meters may perceive it as sedentary time because of the lack of motion (Bai et al., 2016). It has been reported that TEE measured by most activity meters was underestimated compared with TEE measured by the DLW method (Murakami et al., 2016). Hence, we believe that the actual TEE of the subjects in this study could be higher than the value obtained from the activity meter.

Previous studies on British male adolescent academy-level soccer players have revealed that match days had higher TEE than moderate training and rest days and had the largest EI deficit (Briggs et al., 2015). In this study, no difference in physique was observed between the training and match groups. Also, the training lasted 2 h in the training group, and the one or one and half practice match lasted after 30-min warm-up in the match group; therefore, there is no difference in exercise times between the groups. However, EE during exercise per body mass was higher in the match group than that in the training group, which was consistent with the findings of the previous study (Briggs et al., 2015). In the match group, the exercise intensity was considered higher than that in the training group, EE during exercise was higher, and as a result, TEE was also higher. Therefore, we thought that the match days required more EI than the training days.

Carbohydrate provides a key fuel for the brain and central nervous system, and it is a versatile substrate for muscular work where it can support exercise over a large range of intensities due to its utilization by both anaerobic and oxidative pathways (Thomas et al., 2016). Carbohydrate is stored as glycogen in the liver and muscle. Muscle glycogen is a predominant energy source for soccer players (Anderson et al., 2016). For the endurance program (1–3 h/day, mod-high-intensity exercise), 6–10 g/kg/day of carbohydrate was recommended (Thomas et al., 2016). Before an important soccer match, soccer players should have a high carbohydrate intake with at least 8 g/kg/day-carbohydrate diet and avoid eating a <3 g/kg/day-carbohydrate diet before the match (Souglis et al., 2013). According to a latest meta-analysis (Steffl et al., 2019), carbohydrate intake in adult or junior soccer players is below the recommended carbohydrate

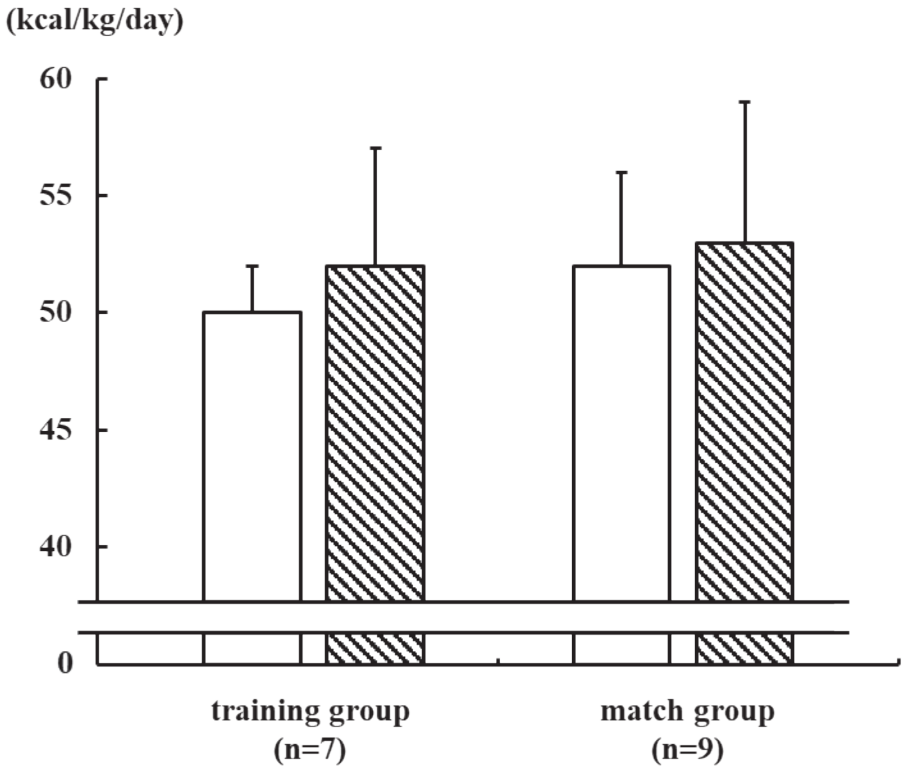

Figure 2 Energy balance per body mass of the training and match groups

Bar charts are represented as mean values and error bars are represented as standard deviations. TEE: totle energy expenditure. EI: energy intake.
To compare energy balance values within the training and match groups, a paired *t*-test was used for items with normally distributed data (training group: $p = 0.493$; match group: $p = 0.367$).

intake for endurance programs and important soccer matches (adult players: 4.3 g/kg/day, CI 3.2–5.4 g/kg/day; junior players: 5.7g/kg/day, CI 5.4–5.9 g/kg/day). In this study, training and match group's carbohydrate per body mass was higher than that of junior players and met the recommended carbohydrate intake for endurance programs and important soccer matches (training group: 7.4 ± 1.2 g/kg/day; match group: 8.0 ± 1.0 g/kg/day).

Resistance exercise increases muscle protein synthesis, but it also increases muscle protein degradation (Phillips et al., 1997). Dietary protein supplementation augments change in muscle mass and strength during prolonged resistance exercise training. It is more effective in improving lean body mass in young or resistance-trained individuals than in older or untrained individuals (Morton et al., 2018). According to the Academy of Nutrition and Dietetics, Dietitians of Canada, and American College of Sports Medicine, the importance of protein intake at the right time for all athletes, even if muscle hypertrophy is not the primary purpose of training, has been emphasized (Thomas et al., 2016). The dietary protein intake necessary to support metabolic adaptation and for protein turnover (repair and remodeling) should be 1.2–2.0 g/kg/day (Thomas et al., 2016). Previous studies have stated that a protein intake of 1.4–1.7 g/kg/day should be adequate for soccer players (Lemon, 1994). In this study, the recommended protein intakes were 0.8–1.5 g/kg/day and 1.4–2.4 g/kg/day for the training and match groups, respectively. Amino acid (units that make up protein) can provide

Table 4 Total urinary nitrogen content and nitrogen balance of the training and match groups

	training group (n=7)			match group (n=9)			Significant difference*
Total urinary nitrogen, g/day	**5.3**	**±**	**0.9**	**10.8**	**±**	**2.2**	**p<0.001**
Nitrogen excreted, g/day	**6.2**	**±**	**0.8**	**11.8**	**±**	**2.3**	**p<0.001**
Nitrogen intake, g/day	**17.7**	**±**	**1.8**	**18.1**	**±**	**2.0**	**p=0.635**
Nitrogen balance, g/day	**11.4**	**±**	**2.4**	**6.4**	**±**	**3.1**	**p=0.003**

Data are represented as mean ± standard deviation.

* To compare the parameters between the training and match groups, an unpaired *t*-test was used for items with normally distributed data and a Mann-Whitney U test was used for items with non-normally distributed data.

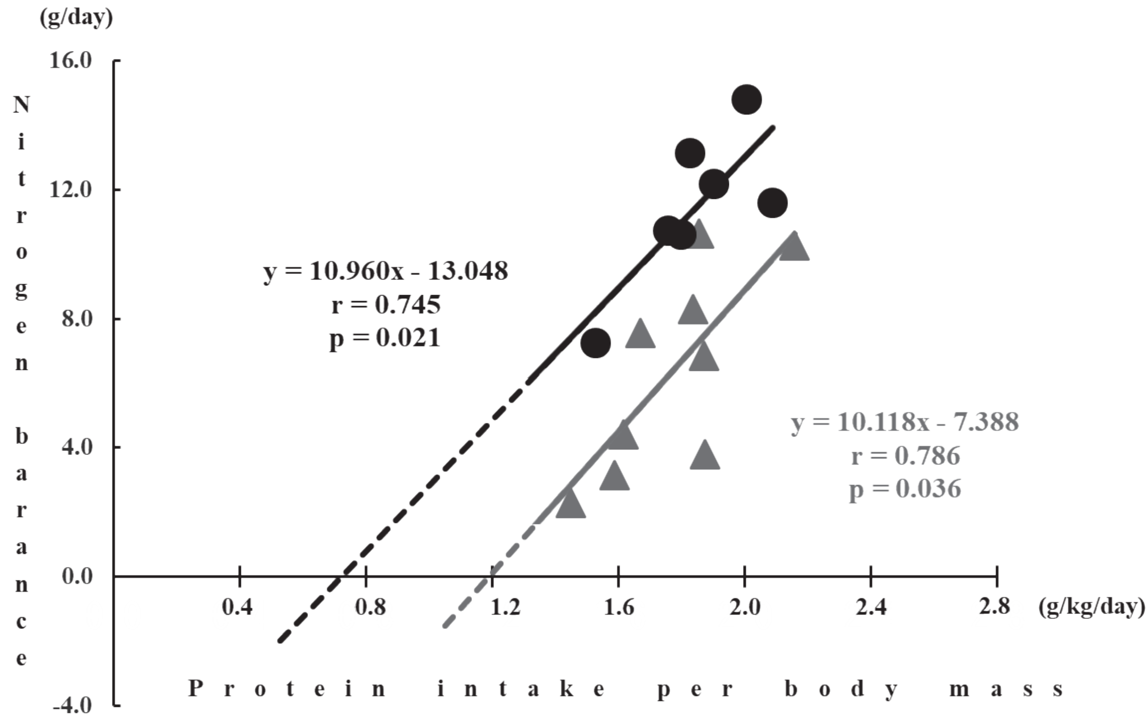

Figure 3 Nitrogen balance and protein intake per body mass of the training and match groups

● Training ▲ Match

The recommended protein intake was calculated using Pearson's product-moment correlation coefficient between the nitrogen balance value and protein intake per body mass in the training and match groups.

up to 4–10% of energy based on the state of intake of other nutrients and intensity of activity (Lemon and Mullin, 1980). Furthermore, carbohydrate and protein combinations are a traditional strategy employed by endurance as well as strength-power athletes to increase exercise performance, promote glycogen repletion, minimize muscle damage, and promote a positive nitrogen balance (Kerksick et al., 2017). If carbohydrate ingestion is low (≤0.8 g/kg/h), it appears that 0.3–0.4 g/kg/h of protein should be coingested to maximize muscle glycogen resynthesis during short-term recovery (Alghannam et al., 2018). Although these considerations focus on maximizing the response to the stimulus of exercise training, adolescents have additional protein requirements to support general growth and development (Aerenhouts et al., 2013). According to the latest meta-analysis (Steffl et al., 2019), adult or junior soccer players have relatively high protein intake (adult players: 1.9 g/kg/day, CI 1.4–2.4 g/kg/day; junior players: 1.8g/kg/day, CI 1.7–1.9 g/kg/day). In this study, the subjects also had relatively high protein intake (training group: 1.8 ± 0.2 g/kg/day; match group: 1.8 ± 0.2 g/kg/day). However, whether the protein requirement required by the NBAL method is the amount required to maximize the performance and training effect of athletes is not clear (Phillips, 2012). The indicator amino acid oxidation (IAAO) method defines protein requirement as the protein intake required for the maximization of whole-body protein synthesis, which could be consistent with the protein intake for the maximization of performance and training effect of athletes measured by the NBAL method (Humayun et al., 2007). The recommended protein intake suggested by the IAAO method was >31–53% of previously recommended levels for populations on endurance training based on the NBAL method (Kato et al., 2016). Evidence of a high protein diet causing harm in athletes has not been confirmed (Antonio et al., 2014). Owing to these considerations, we believe that the recommended protein intake for Japanese male senior high school soccer players on match days should be higher than that on training days.

Limitations

This study is based on field research and has several limitations. It is not a randomized controlled trial model, so there are not enough subjects to determine more accurate recommended energy and protein intakes. Furthermore, fecal and transdermal nitrogen excretions were not measured for practical reasons and estimated values were used. Whereas, in a previous study involving animal models of experimental renal failure and patients with chronic renal failure, feeding dietary fiber increased fecal nitrogen excretion and decreased urine nitrogen excretion (Younes et al., 2004). In this study, high dietary fiber intake in the training group might have resulted in the transfer of urinary nitrogen into fecal nitrogen; the urine nitrogen excretion in the training group might have been calculated low. The actual nitrogen balance on the training days may be low and recommended protein intake may be high. Therefore, further studies with a greater number of subjects are required, and the studies should be conducted under dietary controlled conditions.

There are limitations on estimating TEE using the activity meter. Different devices have different algorithms, and the calories of isometric or resistance exercises are underestimated. The NBAL method is still the gold standard method for assessing protein requirements and metabolism in a free-living state. However, the protein requirement for the populations suggested by this method may be underestimated compared with the IAAO method. Therefore, limitations do need to be acknowledged when adopting such methods.

5. Conclusion

In conclusion, the recommended EIs were 48–52 kcal/kg/day and 49–54 kcal/kg/day on the training and match days, respectively. In addition, the recommended protein intakes were 0.8–1.5 g/kg/day and 1.4–2.4 g/kg/day on the training and match days, respectively. Moreover, we believe that the recommended energy and protein intakes for Japanese male senior high school soccer players on match days should be higher than on those on training days.

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References

Aerenhouts, D., Cauwenberg, J.V., Poortmans, J.R., Hauspie, R., and Clarys, P. (2013). Influence of growth rate on nitrogen balance in adolescent sprint athletes. Int. J. Sport Nutr. Exerc. Metab., 23:409-417.

Alghannam, A.F., Gonzalez, J.T., and Betts, J.A. (2018). Restoration of muscle glycogen and functional capacity: Role of post-exercise carbohydrate and protein co-ingestion. Nutrients., 10: 253.

Anderson, L., Orme, P., DiMichele, R., Close, G.L., Morgans, R., Drust, B., and Morton, J.P. (2016). Quantification of training load during one-, two- and three-game week schedules in professional soccer players from the English Premier League: Implications for carbohydrate periodization. J. Sports Sci. 2016, 34, 1250-1259.

Anderson, L., Orme, P., Naughton, R.J., Close, G.L., Milsom, J., Rydings, D., O'Boyle, A., Di Michele, R., Louis, J., Hambley, C., Speakman, J.R., Morgans, R., Drust, B., and Morton, J. (2017). Energy intake and expenditure of professional soccer players of the English Premier League: Evidence of carbohydrate periodization. Int. J. Sport Nutr. Exerc. Metab., 27:128-138.

Antonio, J., Peacock, C.A., Ellerbroek, A., Fromhoff, B., and Silver, T. (2014). The effects of consuming a high protein diet (4.4 g/kg/d) on body composition in resistance-trained individuals. J. Int. Soc. Sports Nutr., 11:19.

Bai, Y., Welk, G.J., Nam, Y.H., Lee J.A., Lee J-M., Kim, Y., Meier, N.F., and Dixon, P.M. (2016). Comparison of consumer and research monitors under semistructured settings. Med. Sci. Sports Exerc., 48:151-158.

Boisseau, N., Creff, C.L., Loyens, M., and Poortmans, J.R. (2002). Protein intake and nitrogen balance in male non-active adolescents and soccer players. Eur. J. Appl. Physiol., 88:288-293.

Boisseau, N., Vermorel, M., Rance, M., Duché, P., and Mirand, P.P. (2007). Protein requirements in male adolescent soccer players. Eur. J. Appl. Physiol., 100:27-33.

Briggs, M.A., Cockburn, E., Rumbold, P.L.S., Rae, G., Stevenson, E.J., and Russell, M. (2015). Assessment of energy intake and energy expenditure of male adolescent academy-level soccer players during a competitive week. Nutrients., 7:8392-8401.

Brooke, S., An, H-S., Kang, S-K., Noble, J., Berg, K., and Lee, J-M. (2017). Concurrent validity of wearable activity trackers under free-living conditions. J. Strength Cond. Res., 31:1097-1106.

Brouns, F., Saris, W.H., Stoecken, J., Beckers, E., Thijssen, R., Rehrer, N.J., and ten Hoor, F. (1989). Eating, drinking, and cycling. A controlled Tour de France simulation study, Part II. Effect of diet manipulation. Int. J. Sports Med., 10:41-48.

Ebine, N., Rafamantanantsoa, H.H., Nayuki, Y., Yamanaka, K., Tashima, K., Ono, T., Saitoh, S., and Jones, P.J. (2002). Measurement of total energy expenditure by the double labeled water method in professional soccer players. J. Sports Sci., 20:391-397.

Evenson, K.R., Goto, M.M., and Furberg, R.D. (2015). Systematic review of the validity and reliability of consumer-wearable activity trackers. Int. J. Behav. Nutr. Phys. Act., 12:1-22.

FAO/OMS/UNU. (1986). Nutritional and protein requirements. In FAO/OMS/UNU experts report 724 (pp. 226) Genève: OMS.

Fokkema, T., Kooiman, T., Krijnen, W.P., Van Der Schans, C., and DE Groot, M. (2016). Reliability and validity of ten consumer activity trackers depend on walking speed. Med. Sci. Sports Exerc., 49:793-800.

Frankenfield, D., Roth-Yousey, L., and Compher, C. (2005). Comparison of predictive equations for resting metabolic rate in healthy nonobese and obese adults: A systematic review. J. Am. Diet. Assoc., 105:775-789.

Henriksen, A., Mikalsen, M.H., Woldaregay, A.Z., Muzny, M., Hartvigsen, G., Hopstock, L.A., and Grimsgaard, S. (2018). Using fitness trackers and smartwatches to measure physical activity in research: analysis of consumer wrist-worn wearables. J. Med. Internet. Res., 20: e110.

Humayun, M.A., Elango, R., Ball, R.O., and Pencharz, P.B. (2007). Reevaluation of the protein requirement in young men with the indicator amino acid oxidation technique. Am. J. Clin. Nutr., 86:995-1002.

Jager, R., Kerksick, C.M., Campbell, B.I., Cribb, P.J., Wells, S.D., Skwiat, T.M., Purpura, M., Ziegenfuss, T.N., Ferrando, A.A., Arent, S.M., Smith-Ryan, A.E., Stout, J.R., Arciero, P.J., Ormsbee, M.J., Taylor, L.W., Wilborn, C.D., Kalman, D.S., Kreider, R.B., Willoughby, D.S., Hoffman, J.R., Krzykowski, J.L., and Antonio, J. (2017). International society of sports nutrition position stand: protein and exercise. J. Int. Soc. Sports Nutr., 14:20.

Kato, H., Suzuki, K., Bannai, M., and Moore, D.R. (2016). Protein requirements are elevated in endurance athletes after exercise as determined by the indicator amino acid oxidation method. PLOS ONE., 11: e0157406.

Kerksick, C.M., Arent, S., Schoenfeld, B.J., Stout, J.R., Campbell, B., Wilborn, C.D., Taylor, L., Kalman, D., Smith-Ryan, A.E., Kreider, R.B., Willoughby, D., Arciero, P.J., VanDusseldorp, T.A., Ormsbee, M.J., Wildman, R., Greenwood, M., Ziegenfuss, T.N., Aragon, A.A., and Antonio, J. (2017). International society of sports nutrition position stand: Nutrient timing. J. Int. Soc. Sports Nutr., 14:33.

Kerksick, C.M., Wilborn, C.D., Roberts, M.D., Smith-Ryan, A., Kleiner, S.M., Jäger, Collins, R., Cooke, M., Davis, J.N., Galvan, E., Greenwood, M., Lowery, L.M., Wildman, R., Antonio, J., and Kreider, R.B. (2018). ISSN exercise and sports nutrition review update: Research and recommendations. J. Int. Soc. Sports Nutr., 15: 38.

Kim, H.R., Lee, S.J., Park, J.H., Ebine, N., Yamanaka, K., Tashima, K., and Saitoh, S. (2003). Measurement of total energy expenditure by the doubly labeled water method during a competitive season in Korea professional football league players. Jpn. J. Phys. Educ. Hlth. Sport. Sci., 48:717-723. (in Japanese).

Lemon, P.W.R. (1991). Protein and amino acid needs of the strength athlete. Int. J. Sport Nutr., 1:127-145.

Lemon, P.W.R. (1994). Protein requirements of soccer. J. Sports. Sci., 12:17-22.

Lemon, P.W.R. (1996). Is increased dietary protein necessary or beneficial for individuals with a physically active lifestyle? Nutr. Rev., 54:169-175.

Lemon, P.W.R. (1998). Effect of exercise on dietary protein requirements. Int. J. Sport Nutr., 8:426-447.

Lemon, P.W.R., and Mullin, J.P. (1980). Effect of initial muscle glycogen levels on protein catabolism during exercise. J. Appl. Physiol., 48:624-629.

Lemon, P.W.R., Yarasheki, K.E., and Dolny, D.G. (1984). The importance of protein for athletes. Sports Med., 1:474-484.

Matsuda, T., Kato, H., Suzuki, H., Mizugaki, A., Ezaki, T., and Ogita, F. (2018). Within-day amino acid intakes and nitrogen balance in male collegiate swimmers during the general preparation phase. Nutrients., 10:1809.

Ministry of Education Culture Sports Science and Technology. (2016). Standard tables of food composition in Japan 2017. Ishiyaku Publishers. Inc., Tokyo. (in Japanese).

Ministry of Health Labour and Welfare. (2020). Dietary reference intakes for Japanese 2020. Dai-ichi Shuppan Co. Ltd., Tokyo. (in Japanese).

Morton, R.W., Murphy, K.T., McKellar, S.R., Schoenfeld, B.J., Henselmans, M., Helms, E., Aragon, A.A., Devries, M.C., Banfield, L., Krieger, J.W., and Phillips, S.M. (2018). A systematic review, meta-analysis and meta-regression of the effect of protein supplementation on resistance training-induced gains in muscle mass and strength in healthy adults. Br. J. Sports Med., 52:376-384.

Murakami, H., Kawakami, R., Nakae, S., Nakata, Y., Ishikawa-Tanaka, K., Tanaka, S. and Miyachi, M. (2016). Accuracy of wearable devices for estimating total energy expenditure: Comparison with metabolic chamber and doubly labeled water method. JAMA. Intern. Med., 176:702-703.

Nato, N., Murata, M., Kawano, M., Taniguchi, T., and Ohtake, T. (2004). Growth standard for children from 0 up to 18 years of age. J. Child. Health., 63:345-348.

Phillips, S.M., Tipton, K.D., Aarsland, A., Wolf, S.E., and Wolfe, R.R. (1997). Mixed muscle protein synthesis and breakdown after resistance exercise in humans. Am. J. Physiol., 273:99-107.

Phillips, S.M. (2012). Dietary protein requirements and adaptive advantages in athletes. Br. J. Nutr., 108:158-167.

Phillips, S.M., Chevalier, S., and Leidy, H.J. (2016). Protein “requirements” beyond the rda: Implications for optimizing health. Appl. Physiol. Nutr. Metab., 41:565-572.

Sakuma, M., Ohta, H., and Arai, H. (2017). Assessment of dietary phosphorus intake using 24-hour urine collection and effects of dietary pattern on phosphorus intake in young adults. Jpn. J. Nutr. Diet., 75:131-140. (in Japanese).

Sasai, H., Hikihara, Y., Okazaki, K., Nakata, Y., and Ohkawara, K. (2015). Overview of physical activity assessments by accelerometer-based activity monitors and their potential application for physical activity promotion. Res. Exer. Epidemiol., 17:6-18. (in Japanese).

Shepard, D.L., and Jacobs, M.B. (1951). Modified Micro-Kjeldahl apparatus. J. Am. Pharm. Assoc., 40:154-155.

Souglis, A.G., Chryssanthopoulos, C.I., Travlos, A.K., Zorzou, A.E., Gissis, I.T., Papadopoulos, C.N., and Sotiropoulos, A.A. (2013). The effect of high vs. low carbohydrate diets on distances covered in soccer. J. Strength Cond. Res., 27:2235-2247.

Steffl, M., Kinkorova, I., Kokstejn, J., and Petr, M. (2019). Macronutrient intake in soccer players–a meta-analysis. Nutrients., 11:1305.

Takahashi, K. (2003). Food Frequency Questionnaire based on food groups for estimating individual nutrient intake. Jpn. J. Nutr. Diet., 61:161-169. (in Japanese).

Thomas, D.T., Erdman, K.A., and Burke, L.M. (2016). Position of the academy of nutrition and dietetics, dietitians of Canada, and the American College of Sports Medicine: Nutrition and athletic performance. J. Acad. Nutr. Diet., 116:501-528.

Tipton, K.D., and Witard, O.C. (2007). Protein requirements and recommendations for athletes: Relevance of ivory tower arguments for practical recommendations. Clin. Sports Med., 26:17-36.

Witard, O.C., Wardle, S.L., Macnaughton, L.S., Hodgson, A.B., and Tipton, K.D. (2016). Protein considerations for optimising skeletal muscle mass in healthy young and older adults. Nutrients, 8:181.

Yasutake, K., Horita, N., Sawano, K., and Tsuchihashi, T. (2014). Salt intake among children estimated by nutrition survey and 24-h urine collection. J. Jpn. Diet. Associ., 57:46-53. (in Japanese).

Younes, H., Alphonse, J.C., and Deteix, R. (2004). Role of dietary fibers in the nutritional management of chronic renal failure. Nephrologie., 25:283-285.

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- Yotsumoto, H., Kataoka, K., Tanabe, H., Taki, A., Kajii, R., Ono, A., and Matsueda, S. (2019) Hemoglobin concentration and nutrient intake of male senior high school soccer players. Kawasaki Medical Welfare Journal, 28: 423-432. (in Japanese).

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