1. Introduction

During soccer games, players run 10 to 12 km, with a mean intensity of approximately 80-90% of maximal heart rate (HRmax) (Stolen et al., 2005). They often walk, jog, run, change of directions and jump repeatedly for 90 min (Mohr et al., 2003). Soccer players must play one to two games in a week during the season. Aerobic and anaerobic fitness is necessary for high performance during games. An earlier study has demonstrated that distance covered over high-intensity running during games associates with not only the score of Yo-Yo intermittent recovery test level 1 but also sprint and jump performances in youth soccer players (Castagna et al., 2010). Furthermore, Buchheit et al. (2010) have demonstrated that physical fitness is related to match running performance in youth soccer players aged from 13 to 18 years, when statistically controlling for age and total playing time, indicating that relationship between physical fitness and match running performance may be less affected by age.

There are seasonal variations in the aerobic and anaerobic fitness of soccer players (Fessi et al., 2016; Magal et al., 2009). Earlier studies have revealed that training load derived from heart rate (HR), defined as one of the indices of internal loads (Buchheit, 2014), affects the relative change in aerobic fitness through a preseason (Castagna et al., 2011; Manzi et al., 2013). For example, internal load is positively related to the relative changes in the score of the Yo-Yo intermittent recovery test level 1 and velocity at lactate threshold.
and onset of blood lactate accumulation (OBLA) during preseason in professional soccer players (Manzi et al., 2013).

Short distance (10 m and 30 m) sprint time and height of countermovement jumps in professional soccer players improve during a preseason (Fessi et al., 2016). However, Arcos et al. (2015) demonstrated that a negative relationships between internal load derived from the rate of perceive effort (RPE) and relative changes in anaerobic fitness exists during the time from preseason to in-season in young professional male soccer players. This implies that an excess of training load would lead to a decrease in anaerobic fitness (Arcos et al., 2015). Less information is known about the relationships between training load derived from a GPS and the relative changes in anaerobic fitness in soccer players.

Body composition such as fat mass (FM) and fat-free soft tissue mass (FFSTM), which are highly associated with whole body and appendicular skeletal muscle masses (Kim et al., 2002), are considered to be good measures of musculoskeletal injury risk and anaerobic fitness (Bilsborough et al., 2015; Kaplan et al., 1995; Nye et al., 2014). In fact, increase of FM may be a factor in the increase of musculoskeletal injury risk in lower extremities in athletes (Kaplan et al., 1995; Nye et al., 2014). Furthermore, FFSTM is positively related to squat jump performance (Bilsborough et al., 2015). Fat mass (FM) decreases and fat-free soft tissue mass (FFSTM) increases during preseason, but the FM and FFSTM do not change during the in-season (Milanese et al., 2015). For soccer players, however, it is unknown whether body composition is related to training load during preseason. For muscle hypertrophy, high training load is needed (Mitchell et al., 2012). High acceleration may be required for achieving high velocity during running, implying that players need to exert force against the ground. If a player runs a long distance with high velocity during a training session, high training load may occur. During preseason, therefore, long distance covered at high velocity would lead to an increase of FFSTM.

There are many previous studies concerning the relationship between training load and physical fitness changes in professional soccer players (Castagna et al., 2013; Manzi et al., 2013). One study demonstrated that in small-sided games, distance covered over high velocity is less in amateur soccer players compared to professional ones (Dellal et al., 2011). Collegiate soccer players are amateurs, yet the schedule of collegiate soccer is similar to that of professional one. In Japan, there are many amateur soccer leagues. In collegiate soccer, many of collegiate soccer players competing in intercollegiate competition try to become professional soccer players in domestic and international teams. Elucidating the relationships between training load and physical fitness changes will be useful for coaches and prospective soccer players. Therefore, this study aims to examine the relationships between changes in body composition, physical fitness, and internal and external load during preseason in collegiate male soccer players.

2. Methods
2.1. Subjects

Eight collegiate male soccer players (20.4 ± 0.7 years, 177.0 ± 5.2 cm, 72.3 ± 5.8 kg) voluntarily participated in this study. They were part of the same team competing in a national university league in Japan, and had been playing competitive soccer for >9 years. They had participated in regular training for more than five days (>1.5 hours/day) per week. Goalkeepers were excluded from data analysis. Participants were free of cardiovascular, metabolic, and immunologic disorders and/or orthopedic abnormalities, and were not using any medications that affected muscular function and size. All players provided informed written consent. This study was approved by the institutional review ethics committee and follows the recommendations of the Declaration of Helsinki.

2.2. Experimental design

The experimental period was from the middle of January to the end of March (79 days). Physical fitness was measured by sprinting, jumping and body composition before and after the preseason. The tests were conducted on indoor artificial grass at 1st week and the end week of preseason. To quantify external load, distance covered during every training session and informal games was measured with a GPS. To determine internal load, HR was measured with a HR monitor. Data collection for internal and external loads was completed outdoors on natural grass.

The regular training program consisted of warm-up
with and without balls (<20 min), ball possession (15-20 min), small-sided games (e.g. 2 vs. 2 and 4 vs. 4) (15-20 min), and large-sided games (e.g. 8 vs. 8 and 11 vs. 11) (15-20 min), and cooling down (10 min). Furthermore, there were one or two informal matches during the weeks throughout the preseason.

2.3. External load during training sessions and games

As an index of external load, distance covered during every training session and informal games were determined. Players wore a fitted vest with a pocket for the GPS unit (SPI-HPU, GPSports Systems, Canberra, Australia) during training sessions and games. GPS data collection was conducted with at least 8 satellites. The data of x- and y-coordinates were stored in the unit at a frequency of 5 Hz for each player. The GPS is capable of recording the coordinates of players during ball games, and is with high reliability in outdoor fields (Johnston et al., 2012). The validity and reliability of the GPS system have been previously reported (Coutts & Duffield, 2010; Koklu et al., 2015), and documented for use with soccer players (Torreno et al., 2016). The data were entered into a software (Team AMS R1 2016.7, GPSports System s), and the coordinates were exported. After the data were corrected for latitude and longitude and origin of the coordinate axis was set to (0, 0) on a corner flag of the pitch, the location of each player was relatively expressed in a pitch. All analyses were conducted with Matlab (Matlab 2017b, Mathworks). Total distance covered and the distance covered in six velocity zones (<6, <11, <13, <18, <24, and over 24 km/h) were calculated from the coordinates’ data, based on research from a previous study (Kai et al., 2018). We defined the distance covered over 18 km/h and over 24 km/h as high-intensity running (DHR) and sprinting (DSprinting), respectively.

2.4. Internal load during training sessions and games

HR was recorded using a telemetry system (Polar RC3, Finland) during training sessions and games. HR was averaged for every training session or game through the preseason period. HRmax was obtained from the Yo-Yo intermittent recovery test level 2 (YYIR2) as follows. The HRs during training sessions and games were normalized to individual HRmax, and expressed as relative value (%HRmax). The exercise intensity zone was assigned to five zones (50-59, 60-69, 70-79, 80-89, and 90-100% of %HRmax). Time spent in each zone was also determined. TRIMP was calculated from Edward’s equation (Edwards, 1993);

\[
\text{TRIMP} = \text{duration in zone 1} \times 1 + \text{duration in zone 2} \times 2 + \text{duration in zone 3} \times 3 + \text{duration in zone 4} \times 4 + \text{duration in zone 5} \times 5
\]

where zone 1 = 50-59%HRm ax, zone 2 = 60-69%HRm ax, zone 3 = 70-79%HRm ax, zone 4 = 80-89%HRm ax, and zone 5 = 90-100%HRm ax (Scott et al., 2013).

2.5. Body composition

Height and body mass were measured using a stadiometer and a leg-to-leg bioelectrical impedance analyzer with a computer-programmed athletic mode (DC-320, TANITA, Japan) to the nearest 0.1 cm and 0.1 kg, respectively. Participants were instructed to refrain from alcohol intake for 24 h prior to the experiment and from having a meal 2 h prior to the measurement. Before and after the preseason, body composition was measured after a rest day.

Whole body percent fat mass (%FM), fat (FM) and fat-free soft tissue masses (FFSTM) were determined with a dual-energy X-ray absorptiometry (DXA) scanner (Hologic Delphi A-QDR, USA). Participants lay supine on a bed with arms and legs straight. Room temperature was usually kept at 22°C. DXA-derived body composition has been shown to have good accuracy and reliability in team sport athletes (Bilsborough et al., 2014). The repeatability of body composition measurement was confirmed in our previous study (Takai et al., 2018).

2.6. 30-m sprint test

Participants performed a 30-m sprint test twice with maximal effort. Time of the 30 m test was measured to the nearest 0.01 s using a photocell electronic system (TC Timing System, Brower Timing System). Each player started from a stationary standing position 0.5 m behind start line at which the 1st gate was set. The test was conducted with a rest interval of >1 min between trials. Running velocity
was used for data analysis. The highest value between trials was used.

2.7. Pro-agility test

To assess agility and quickness capacity, participants performed a pro-agility test twice. Each player stood at the middle line (start line) and sprinted in one direction for 5 m. The player changed direction and sprinted back for 10 m to another line, and turned around and sprinted to the start line (5 m). Time was measured by a photocell electronic system (TC Timing System, Brower Timing System) to the 0.01 s. The test was conducted with a rest interval of >1 min between trials. Agility time was converted to velocity by dividing running distance (20m) by time. The highest value between trials was used.

2.8. Countermovement jump test

Participants were in a standing position, and performed countermovement jumps (CMJ) with arm swings as high as possible. They were instructed to keep the same position when taking off and landing on a mat. Flight time (s) was measured by a Matswitch system (Multi Jum p Tester, DKH, Japan). Height of CMJ was calculated from the equation; height (cm) = g × flight time squared × 8\(^{-1}\), where g is acceleration due to gravity (9.81 m/s\(^2\)). Two trials were completed with a rest interval of >1 min between the trials. The highest value between trials was used.

2.9. Yo-Yo intermittent recovery test 2 (YYIR2)

As described in the earlier studies (Krustrup et al., 2015; Krustrup et al., 2006), participants performed YYIR2, which consisted of repeated two 20-m runs at a progressively increased speed, controlled by audio bleeps from a tape recorder. In the interval of each running bout, participants performed 5 m × 2 jogging during 10-s rest. When the participants failed to reach the finishing line in time twice, the distance covered was recorded and adopted as the representative score. The test was performed on 2-m-wide and 20-m-long running lane marked by color markers. We adapted the test in this study, because some players could complete YYIR 1 test in regular physical tests conducted in a season. In our preliminary study, HRmax obtained from the YYIR test did not change in a season for collegiate male soccer players, and no significant difference in the HRmax was found. So, we used the HRmax obtained at baseline to estimate %HRmax and TRIMP in this study.

2.10. Statistical analysis

Descriptive data are presented as mean ± SD. The independent variables were body composition, physical fitness, internal and external loads (%FM, FM, FFSTM, mean velocities of 30-m sprint and proagility tests, height of CMJ, and a score of YYIR2). To estimate intra-individual variation in each of the independent variables for each player, the relative change (%Δ) was calculated from the following equation; (Post-Pre)/Pre × 100. To test significant changes in the independent variables between before and after a preseason, a paired t-test was conducted. Effect size (Δ) was calculated by dividing the difference between the mean values of the independent variables before and after the preseason by the SD at baseline (Cohen, 1988). Pearson’s product-moment correlation (r) analysis was conducted to examine the relationships among the independent variables throughout the preseason. If relative change in each of the independent variables was significantly related to their value at baseline, a partial correlation coefficient was also calculated to examine the relationship between %Δ in the corresponding variable and training load while controlling the initial value of the variable. Correlation coefficients were interpreted as being trivial (r<0.1), small (0.1<r<0.3), moderate (0.3<r<0.5), and large (0.5<r<0.7), very large (0.7<r<0.9), nearly perfect (r>0.9) or perfect (r = 1) according to Hopkins (2010). The probability level for all statistical analysis was set at p<0.05. All statistical analyses were conducted using a statistical software program (SPSS statistics 25.0, IBM Co., New York, USA).

3. Results

The number of days analyzed in this study was 47 ± 4 days for HR and 53 ± 2 days for GPS, respectively. The discrepancy between the preseason period (79 days) and the number of days was due to some rest days and unavailable data by technical error by the GPS. Figure 1 presents day to day TRIMP and total distance covered in a preseason period. The mean value of external and internal loads per day through
During the preseason period, FM and %FM significantly decreased by 1.2% (p = 0.014, ES = -0.28) and 9.6% (p = 0.002, ES = -0.37), and FFSTM increased by 2.6% (p = 0.030, ES = 0.43) (Table 1). There were no significant changes in other measured variables before and after the period (Table 1).

Table 2 provides correlation coefficients between the %Δ in the measured variables and each internal and external load. The %Δ in FFSTM was largely related to DHIR (r = 0.76, p = 0.030) and DSprinting (r = 0.88, p = 0.004) during preseason (Figure 2). The %Δ in velocity of a pro-agility test tended to be positively related to DSprinting, but the corresponding relationship did not reach a significant level (r = -0.69, p = 0.082, Table 2). There were no significant relationships between other measured variables and external load (Table 2). The %Δ in the score of YYIR2 was largely related to each of DHIR and DSprinting, and the score of YYIR2 was associated with %HRmax and TRIMP during preseason. These results indicate that the intra-individual variation in soccer-related aerobic fitness may depend on internal load (%HRmax and TRIMP), and the increase of FFSTM may be due to external load (DHIR and DSprinting). However, the intra-individual variation in the score of a pro-agility test was negatively related to internal load, indicating that high internal load during preseason may lead to attenuation of agility in collegiate soccer players.

Compared to earlier findings (Malone et al., 2015), total distance covered (8373 ± 1863 m) in this study is slightly higher than that of professional soccer players in a preseason period. Furthermore, %HRmax (71 ± 5%) is almost the same as that (approximately 70% of HRmax) of professional soccer players (Manzi et al., 2013). These findings imply that internal and external loads of the soccer players examined here may be the same or greater to those in professional soccer players.
Table 1  Changes in physical fitness through preseason

<table>
<thead>
<tr>
<th></th>
<th>Before</th>
<th>After</th>
<th>Dif (%)</th>
<th>Effect Size (△)</th>
</tr>
</thead>
<tbody>
<tr>
<td>BM, kg</td>
<td>70.6 ± 5.6</td>
<td>71.3 ± 4.8</td>
<td>1.2</td>
<td>0.13</td>
</tr>
<tr>
<td>%FM, %</td>
<td>12.5 ± 3.4</td>
<td>11.3 ± 3.1*</td>
<td>-9.6</td>
<td>-0.37</td>
</tr>
<tr>
<td>FM, kg</td>
<td>9.0 ± 3.1</td>
<td>8.2 ± 2.7*</td>
<td>-8.5</td>
<td>-0.28</td>
</tr>
<tr>
<td>FFSTM, kg</td>
<td>58.9 ± 3.6</td>
<td>60.4 ± 3.4*</td>
<td>2.6</td>
<td>0.43</td>
</tr>
<tr>
<td>YYIR2, m</td>
<td>975 ± 321</td>
<td>1035 ± 251</td>
<td>12.2</td>
<td>0.21</td>
</tr>
<tr>
<td>V30, m/s</td>
<td>7.15 ± 0.21</td>
<td>7.09 ± 0.16</td>
<td>-0.8</td>
<td>0.32</td>
</tr>
<tr>
<td>VCOD, m/s</td>
<td>4.15 ± 0.08</td>
<td>4.08 ± 0.15</td>
<td>-1.6</td>
<td>0.58</td>
</tr>
<tr>
<td>CMJ, cm</td>
<td>51.2 ± 7.8</td>
<td>51.9 ± 5.0</td>
<td>5.3</td>
<td>0.11</td>
</tr>
<tr>
<td>HRmax, bpm</td>
<td>189 ± 8</td>
<td>191 ± 6</td>
<td>1.4</td>
<td>0.28</td>
</tr>
</tbody>
</table>

Values are means and SDs.
* indicates significant changes between before and after a preseason.
BM, body mass; %FM, percentage of fat mass to body mass; FM, fat mass;
FFSTM, fat-free soft tissue mass; YYIR2, Yo-Yo intermittent recovery test level 2;
V30, mean velocity of a 30-m sprint; VCOD, mean velocity of a proagility test;
CMJ, countermovement jump; HRmax, maximal heart rate

Effect size (△) was calculated by dividing the difference between the mean values of the independent variables before- and after a preseason into SD at baseline.

Table 2  Correlation coefficients between the relative changes in the measured variables and each of internal and external load

<table>
<thead>
<tr>
<th></th>
<th>%FM</th>
<th>FM</th>
<th>FFSTM</th>
<th>YYIR2</th>
<th>V30</th>
<th>VCOD</th>
<th>CMJ</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total distance covered</td>
<td>-0.643</td>
<td>-0.459</td>
<td>0.390</td>
<td>-0.072</td>
<td>-0.053</td>
<td>0.089</td>
<td>0.094</td>
</tr>
<tr>
<td>Distance covered at HIR</td>
<td>-0.033</td>
<td>0.231</td>
<td>0.757*</td>
<td>-0.352</td>
<td>-0.529</td>
<td>0.651</td>
<td>-0.240</td>
</tr>
<tr>
<td>Distance covered at Sprinting</td>
<td>0.270</td>
<td>0.548</td>
<td>0.882*</td>
<td>-0.396</td>
<td>-0.535</td>
<td>0.694</td>
<td>-0.458</td>
</tr>
<tr>
<td>%HRmax</td>
<td>0.260</td>
<td>0.137</td>
<td>-0.211</td>
<td>0.845*</td>
<td>0.161</td>
<td>-0.943*</td>
<td>0.038</td>
</tr>
<tr>
<td>TRIMP</td>
<td>0.072</td>
<td>-0.006</td>
<td>-0.106</td>
<td>0.811*</td>
<td>0.137</td>
<td>-0.871*</td>
<td>-0.009</td>
</tr>
</tbody>
</table>

HIR, running over 5.0 m/s; Sprinting, running over 6.67 m/s;
%HRmax, percentage of heart rate during training sessions to maximal heart rate; TRIMP, training impulse
%FM, percentage of fat mass to body mass; FM, fat mass; FFSTM, fat-free soft tissue mass;
YYIR2, Yo-Yo intermittent recovery test level 2; V30, mean velocity of a 30-m sprint; VCOD, mean velocity of a proagility test;
CMJ, countermovement jump
during a preseason period.

The %HRmax and TRIMP for the preseason were positively associated with the intra-individual variation in the score of YYIR2. This supports earlier findings of the relationships between the intra-individual variation in aerobic fitness and HR-based training load for professional soccer players (Castagna et al., 2011; Manzi et al., 2013). Manzi et al. (2013) suggests the internal load to change aerobic fitness, which is calculated from the regression equation with relative change in aerobic fitness and internal load. Namely, the value is defined as intersection point of x-axis. As seen in Figure 3, the points were 68.4% for %HRmax and 283.2 a.u. for TRIMP. In other words, loading internally during preseason may be necessary to change aerobic fitness.

There are some possibilities that no significant change in the score of YYIR2 was found in this study. Firstly, the current finding may be affected by the initial value of YYIR2. In fact, the relative change in the score of YYIR2 tended to be negatively related to the initial value of YYIR2 (r = -0.680). Secondly, inter-individual variation in internal load may result in no change in the score. In this study, the relationship between the relative change in YYIR2 and internal load was significant, when controlling the score of YYIR2 at baseline. Therefore, we speculate that inter-individual variation in internal load may influence on

Figure 2  Relationships between intra-individual variation in fat-free soft tissue mass (FFSTM) and external load indices during preseason
A: distance covered at >18 km/h (D_HIR), B: distance covered at >24 km/h (D_Sprinting).

Figure 3  Relationships between intra-individual variation in Yo-Yo intermittent recovery test 2 (YYIR2) and internal load indices during preseason
A: percentage of heart rate during training to maximal heart rate (%HRmax), B: training impulse (TRIMP).
change in aerobic fitness for a preseason period.

In this study, %FM and FM decreased by 1.2\% and 8.5\%, and FFSTM increased by 2.6\% through the preseason. The current results are consistent with earlier findings for professional soccer players (%FM, -1.3\%; FM, -11.9\%; FFSTM: 1.3\%) (Milanese et al., 2015). Furthermore, the intra-individual variation in FFSTM was associated with D_{HIR} and D_{Sprinting}, but not with any variables of internal load. This indicates that the change in FFSTM may partially depend on external load through a preseason period in collegiate soccer players. Locomotion at HIR during soccer games is similar to straight running when measuring with GPS (Kai et al., 2018). To achieve relative high velocity, high acceleration may be required, implying that players need to exert force against the ground. Hence, it is possible that high training load might occur with players running long distances with high-intensity during a preseason.

Another interpretation of the relationship between intra-individual variation in FFSTM and external load, might be that, for collegiate soccer players, the reduced FFSTM might be regained by running the distance covered over HIR during preseason. A 5-week detraining from end of in-season to start of preseason (off-season period) decreases fat-free mass in elite male soccer players (Suarez-Arrones et al., 2019). Therefore, the intersections of the regression line between the intra-individual variation in FFSTM and D_{HIR} or D_{Sprinting} (Figure 2) might mean that the minimum value at which the reduced FFSTM in off-season period is regained in preseason period, 345 m for D_{HIR} and 41 m for D_{Sprinting}.

No significant changes in the ability of sprinting and jumping were found. Caldwell and Peters (2009) have observed significant improvements of jump, sprint and agility performances in a preseason period for semiprofessional soccer players. This discrepancy may be due to a difference in the prescribed training programs. Training programs reported in the prior study consist of not only soccer-specific training but also aerobic and anaerobic training (Caldwell & Peters, 2009). The prescribed training program in this study mainly consisted of soccer training alone. Adding regular soccer training to physical training (e.g. strength and plyometric training) leads to enhancement of anaerobic fitness such as jumping, sprinting and agility, while anaerobic capacities are not improved by a prescription of soccer specific training alone (Ronnestad et al., 2008). However, intra-individual variations in physical fitness were found in this study. Furthermore, the variation was related to the internal or external training load. Therefore, current findings indicate that even though soccer specific training alone is prescribed through a preseason, the player performing the training with high training load might be able to strengthen anaerobic and aerobic fitness.

Another reason for no significant changes in sprint and jump performance might be the magnitude of internal loads in a preseason period. The intra-individual variation in velocity of a pro-agility test was negatively related to %HRmax and TRIMP. Gabbett and Domrow (2007) have demonstrated that the greater internal load derived from the rating of perceived exertion (RPE) results in a decrease of agility performance in rugby players during the early-competition training phase. Additional data are needed to clarify the mechanisms of the negative associations.

There are some limitations in this study. Firstly, Manzi et al. (2009) have demonstrated that the TRIMP derived from the individual weight coefficient of the exponential relationship between lactate concentration and HR reserve in each player (Banister’s TRIMP) is suitable for determining the relationship between training load and running performance in recreational long distance runners. We adopted Edward’s TRIMP in this study. In this study, however, the relationship between the intra-individual variation in the score of YYIR2 and weekly TRIMP in the preseason period was significant with a very large effect size (r = 0.81). Second, we could not examine position-related differences in the relationships between the relative changes in physical fitness and internal or external load through the preseason because of the small sample size. An earlier study indicates that no significant differences in high-speed distance covered and %HRmax exist among positions in a preseason period (Malone et al., 2015). Third, change in body composition may be affected by nutrition status (Bilsborough et al., 2017). Further investigation is needed to clarify these issues.

The current results can be useful for soccer coaches and players to design a training regime in a preseason period. For collegiate soccer players, intra-individual variation in aerobic fitness may depend on internal load, and anaerobic fitness may be due to not only internal load but also external load during preseason. The earlier study demonstrates that soccer specific
training alone cannot enhance physical fitness in soccer players (Ronnestad et al., 2008). However, the current findings indicate that even though soccer specific training alone is prescribed through a preseason, the player performing the training with high training load might be able to strengthen anaerobic and aerobic fitness. The current findings also indicate that excessive internal load in soccer players during soccer specific training might attenuate anaerobic fitness during preseason.

5. Conclusion

For collegiate male soccer players, intra-individual variations in anaerobic fitness such as running in different directions and fat-free soft tissue mass may partially depend on individual external training load, and aerobic fitness may be affected by individual internal training load during preseason.

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