Game demands and fatigue profiles in elite football – an individual approach

Game demands and fatigue profiles in elite football – an individual approach

Implications for training and recovery strategies

Dan Fransson



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Abstract

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The physical activities performed during a football game are of intermittent prolonged character, including explosive actions and running at different speeds. The prolonged intermittent activities are conjoined with periods where physical intensity is markedly increased. The intense periods and prolonged activities affect the physiological and metabolic systems which provoke fatigue both temporarily throughout the game as well as towards the end of a game. Therefore, physical training in football should aim to reach physiological and metabolic adaptations to be able to resist fatigue in order to perform optimally throughout the game. Furthermore, post-game recovery and restoration of performance seems to be a slow process. Physical game demands, training responses and recovery can vary largely between players and needs to be studied with individual emphasis.

The aim of the thesis is to improve the understanding of physical game demands, fatigue profiles in male elite football players with an emphasis on individual differences and implications for fitness training strategies. Running distance and in-game fatigue profiles were investigated through an analysis of game activity data from top-class football players (n = 473). Post-game fatigue and recovery profiles were examined using maximum voluntary contraction in various muscle groups after a simulated football model in competitive players (n = 12). Inter-individual relations between physical game demands and physical response in different small-sided game formats were investigated with global positioning system techniques on professional players (n = 45). Finally, muscular adaptations and physical performance responses of two different

training protocols (four weeks of small-sided games or speed endurance training) were examined by means of pre- and post-intervention muscle biopsies and performance tests on 39 competitive football players.

The results demonstrated that all playing positions indicate temporary fatigue after intense periods during a football game. However, after shorter intense periods central defenders were the only position that did not show a decline in running performance. A large inter-player variation in running performance between and within playing positions was found. Post-game fatigue showed large inter-player differences between various muscle groups and between players. Muscle performance in all investigated groups had recovered within 24 hours post-game except trunk-muscles, which was back to baseline values within 48 hours post-game. The physical response in small-sided game formats differed from game demands on an individual level. High intensity training was more potent in up-regulating muscle oxidative capacity and physical performance compared to small-sided games.

In conclusion, individual differences in game demands and fatigue profiles are large and need to be considered when planning training. Small-sided games seem not to be the most appropriate training method to meet the individual game demands of all individual players. Thus, in order to increase exercise performance and associated physiological adaptations, additional high-intensity training should be considered for some individual football players.

Sammanfattning

Matchspel i fotboll karakteriseras av intermittent långvarigt arbete bestående av explosiva aktioner och löpning i olika hastigheter. Det intermittenta långvariga arbetet blandas med perioder där den fysiska intensiteten höjs markant. Dessa korta intensiva perioder och det långvariga intermittenta arbete påverkar fysiologiska och metabola processer vilka framkallar trötthet tillfälligt under och i slutet av matcher. Därför bör fotbollsspelares fysiska tränings syfta till att erhålla fysiologiska och metabola anpassningar för att kunna motstå trötthet och prestera under hela matchen. Återställande av fysiologiska parametrar och prestationsförmåga efter en fotbollsmatch, s.k. återhämtning, är en långsam process. Fysiska matchkrav, återhämtning och träningsrespons kan variera till stor del mellan spelare och behöver studeras med en individuell inriktning.

Syftet med denna avhandling är att öka förståelsen av fysiska matchkrav, trötthet och återhämtningsprofiler hos manliga elitfotbollsspelare med betoning på individuella skillnader som underlag för träningsstrategier. Fysiska matchkrav och prestationsnedsättningar undersöktes genom analys av löpdistans hos spelare på högsta nivå i Europa (n = 473). Vidare undersöktes trötthet och återhämtning i olika muskelgrupper genom maximal frivillig muskelkontraktion hos spelare på tävlingsnivå (n = 12) efter simulerad fotbollsmatch. Interindividuella samband mellan fysiska match-krav och olika format av smålagsspel, studerades med global positioneringsteknik hos spelare på professionell nivå (n = 45). Slutligen undersöktes skillnader i muskulära förändringar, fysisk prestation mellan individuellt genomförd högintensiv träning och smålagsspel med hjälp av muskelbiopsiteknik och flera prestationstester på 39 spelare på hög tävlingsnivå.

Resultaten visade att alla spelpositioner indikerade temporär trötthet efter intensiva perioder under match. Emellertid, efter kortare intensiva perioder var centrala försvarare den enda positionen som inte visade en nedsättning i löpdistans. Variationen i löpdistans under matcher var stor mellan olika positioner samt inom olika positioner. Prestationsnedsättningar efter simulerade fotbollsmatcher visade stora skillnader mellan enskilda muskelgrupper och mellan spelare. Alla muskelgrupper återhämtades inom 24 timmar efter spel utom mag-musklerna som återvände till utgångspunkten inom 48 timmar efter matchen. Fysisk prestation under smålagsspel skiljde sig i stor utsträckning från match-krav på en individuell nivå. Gruppen som genomförde

individuell högintensiv träning var den enda som visade uppreglering av muskulär oxidativ kapacitet och hade större ökning i fysisk prestation än smålagsspel.

Sammanfattningsvis är individuella skillnader i spelkrav och återhämtningsprofiler i olika muskelgrupper stora. Smålagsspel kanske inte är den lämpligaste träningsmetoden för att möta de individuella matchkraven. Adderad träning som utförs som individuell högintensiv träning kan vara en lämplig metod för att säkerställa fysiologiska anpassningar och ökad fysisk prestation hos fotbollsspelare.

List of original papers

This Thesis is based on the following papers.

Fransson D, Krustrup P and Mohr M. Running intensity Ι fluctuations indicate temporary performance decrement in topclass football. Science and Medicine in Football. 2017;1(1):10-17 Π Fransson D, Larsen JV, Fatouros IG, Krustrup P and Mohr M. Fatigue responses in various muscle groups in well-trained competitive male players after a simulated soccer game. Journal of Human Kinetics. 2018;(61):85-97 III Fransson D, Borjesson M, Bradley PS, Krustrup P and Mohr M. Characteristics of small-sided games vs. full game in elite soccer players. (Submitted for publication) IV Fransson D, Nielsen TS, Olsson K, Christensson T, Bradley PS, Fatouros IG, Krustrup P, Nordsborg NB and Mohr M. Skeletal muscle and performance adaptations to high-intensity training in elite male soccer players - speed endurance runs vs. small-sided game training. Journal of Applied Physiology. 2018;(118):111-121

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Abbreviations

AAT	Arrowhead agility test
ACC	Acceleration
ANOVA	Analysis of variance
ATP	Adenosine triphosphate
AT	Attacker
CD	Central defender
СМ	Central midfielder
CMJ	Counter movement jump
CK	Creatine kinase
СР	Creatine phosphate
CS	Citrate synthase
CST	Copenhagen soccer test
CV	Coefficient of variation
d	Distance
DEC	Deceleration
e	Effort
FB	Fullback
FR	Fast running (>17 km ⁻ h ⁻¹)
FSG	Full-sized game
GLUT-4	Glucose transporter type 4
GS	Glycogen synthase
GPS	Global positioning system
HAD	Beta-hydroxyacyl-CoA-dehydrogenase enzyme
HR	Heart rate
HIR	High-intensity running (>14 km ⁻¹)
HSR	High-speed running (>21 km·h ⁻¹)
IA	Intense acceleration (>3 $m s^{-2}$)
ID	Intense deceleration ($<-3 \text{ m} \cdot \text{s}^{-2}$)
ICC	Intra-class correlation coefficient
K^+	Potassium
MCT	Monocarboxylate transporter
MIA	Medium intense acceleration (>2 m \cdot s ⁻²)
MID	Medium intense deceleration (<-2 m [·] s ⁻²)
MRV	Maximum running velocity

MVC	Maximal voluntary contraction strength test
Na ⁺	Sodium
NHE	Na ⁺ /H ⁺ exchanger
PFK	Phosphofructokinase
RST	Repeated sprint ability test
SEM	Speed endurance maintenance training
SET	Speed endurance production training
SOD	Superoxide dismutase
SSG	Small-sided games
TD	Total distance
WM	Wide midfielder
Yo-Yo IR2	Yo-Yo Intermittent recovery test level 2

Definitions

Competitive football players	Football players that are not professional, but still compete in the top three divisions
Elite football players	Players that are included in top-class, professional and competitive player categories and have a minimum of four training sessions a week
Fatigue	Failure to maintain the required or expected power output
Inter-player	Between players
Intra-player	Within the same player
Micro movement	Small football movements such as acceleration and deceleration
Peak intense period	A period of 1, 2 or 5-minutes were the player covers the greatest distance or the maximum number of efforts of a certain variable compared to similar time-periods during a football game
Physical capacity	The ability for a player to maintain physical activities on a given exercise intensity
Physical game demands	The characteristics of football players activities during match-play; examples can be distance covered and efforts in different speeds and distance or efforts when the player is accelerating or decelerating
Physiological response	Changes in one or more of the body's systems in response to a physical stimuli
Small-sided game	Training drill where football is played by a reduced number of players on a smaller area than the regular official pitch size

Professional football players	Players that play football for their living and also compete in the top 2 divisions
Top-class players	Football players competing in the five best leagues in Europe or participating in international games and tournaments

Abstract

SAMMANFATTNING

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Introduction

Football is the most popular sport in the world and is played by millions men, women and children in different age groups and different levels of play around the world [1]. The performance in football is multi-factorial and complex and is dependent on tactical, technical, physical and psychological parameters. In football it is of importance to be able to make quick and accurate decisions and have a high level of playing skill, but performance also requires high ability levels of physical capacity [2]. This thesis will focus on the physical and physiological aspects of the game of football with an individual approach and its relations to fatigue and training in elite football.

Physical demands in elite football

Football match-play has an intermittent character and elite men's football players cover a total distance (TD) of 9-14 km during a game [3-5]. The physical activities that occur during a football game are very complex. The major part (~85%) of a game consist of low intense activities, like standing, walking and jogging [3]. The remainder consists of physically demanding activities such as high-intensity running (HIR) defined as activities above ~14 km h⁻¹, high speed running (HSR) defined as activities above ~20 km h⁻¹ and sprinting defined as activities above ~25 km h⁻¹ [5-11]. Football players complete ~1500-3100 m in HIR, ~300-1100 m in high speed running and 153-360 m in sprinting during a game [11, 12]. In addition, distance of intense acceleration (IA) (>3m/s²) and deceleration (ID) (<-3 m s⁻²) during football games on a professional level has been reported to be ~180 and ~188 m respectively [13].

Studies have shown that there can be large differences between activity profiles of different playing positions in a football game. Midfield players cover the longest TD compared to all other positions, while central defenders (CD) cover the least [14]. Furthermore, Di Salvo and colleagues demonstrated that wide midfielders (WM) cover the longest distance in HIR (~11535 m) and CD the least (~9885 m) [15]. CD and central midfielders (CM) (~204 and ~152 m respectively) also performed the least sprint distance compared to fullbacks

(FB) (~287 m), WM (~346 m) and attackers (AT) (~264 m) [15]. Similar findings have been observed in several studies [4, 7, 9].

It has been found that physical demands increase markedly during periods of a football game [16]. One study has described these intense periods in detail (using the English Premier League), by analysing the greatest high speed running (>19.8 km \cdot h⁻¹) distance during a 5-min period. The number of bouts increased with 125% in peak 5-min periods compared to average, and the work:rest ratio increased from an average of 1:12 to 1:2 in peak 5-min periods [16]. Furthermore, WM covered 9-22% more, with CD 19-27% less distance in high speed than all other positions in the most intense 5-min period [16].

The physical aspects of the game have changed over the years. In the English Premiere League the distance covered and number in HIR efforts increased by $\sim 30\%$ and $\sim 50\%$ from the 2006/2007 season to the 2012/2013 season respectively [17]. Moreover, sprint distance and number of sprints increased by ~ 35 and $\sim 85\%$ respectively, with FB displaying the greatest increase in both HIR and sprint distance across the seven seasons [18]. Another example of increased game intensity is that Danish professional football players spent 37% more time in sprinting in a study from 2003 [5] than in a previous study with the same match analysis method in 1991 [3]. Finally, a recent study found that physical game demands increase when playing against stronger opponents [19]. Together, these results indicate that the intensity of a football game, at least on elite level, has increased markedly during the last decades. The results also points to the importance of preparing players for intense periods during a football game and to cope with the increased physical game demands of modern football.

Physiological response during a football game

Activities during football game-play seems to cause severe stress on both the aerobic and anaerobic energy systems. The HIR, sprinting, accelerating and decelerating in a game causes an internal load on the physiological and metabolic systems. Numerous studies have found that the aerobic energy systems are highly taxed during a football game, with a mean and peak heart rate (HR) of around 85% and 98% respectively of maximum values [1, 20]. Furthermore, football players' HR is seldom below 65% of maximum during a game [21], which means that the blood flow to the working muscle is

continuously higher than at rest. This indicates that the aerobic system is the main energy system for football players.

The main substrates which provide energy for muscle contractions during a football game are carbohydrates stored as glycogen in skeletal muscle and liver cells [22]. Glycogen levels in the thigh muscle have been found to be depleted at half time when players started the game with low levels (~200 mmol·kg⁻¹ d.w.) [23]. It has also been demonstrated that ~50% of the individual muscle fibres of both type I and II are depleted or almost depleted after a football game [24]. Moreover, blood levels of free fatty acids (FFA) increases throughout a football game (before game = $555 \pm 74 \,\mu$ M; after game = $1365 \pm 111 \,\mu$ M) [24]. Similar result has been found in an earlier study [1], indicating a shift in substrate utilisation towards fat oxidation which is probably related to the gradually reduced muscle glycogen stores towards the end of a football game.

During a football game the anaerobic glycolysis system contributes as an energy source to regenerate ATP [25, 26]. Mean blood lactate has been observed to be 2-10 mmol·l⁻¹ with individual values above 12 mmol·l⁻¹ during a football game [22, 27], while mean muscle lactate can be 16-17 mmol·kg⁻¹ with peak values reaching 25 mmol·kg⁻¹ [24]. Furthermore, after intense periods during a football game blood lactate levels can rise up to 16.9 mmol·l⁻¹ and muscle pH declines from pre-match values with ~3%, while hydrogen ion levels are elevated by ~60% compared to prematch values [24]. Thus, the increased lactate values and muscle acidosis indicates that the glycolysis activity is high during periods of a football game.

Fatigue during a football game

When skeletal muscles are used intensively, a progressive decline in performance occurs. This is defined in the context of match-play as fatigue [28]. The decline in performance can be visible as soon as the second repetition in maximal activation, and fatigue can also be detected after a longer time as a failure to maintain the original intensity in sub-maximal activation [29]. Fatigue has been divided into central and peripheral fatigue. Central fatigue has been defined as limiting processes inside the spinal cord and above, while peripheral fatigue has been defined as limiting processes in the peripheral nerve, neuromuscular junction and muscle [28]. Studies suggest that a small degree of central failure of activation often occurs during maximal activation of muscles [30], but it is also clear that much of exercise induced fatigue arises in specific

muscles and can therefore be studied in isolated muscles (for review see Allen, 2008) [28].

Temporary fatigue during a football game was first described by Mohr et al (2003). The researchers observed that after a peak intense 5-minute period where the greatest distance of HIR occurred, running performance was reduced in the following 5-minute period by $\sim 12\%$ compared to an average 5-min period of the game [5]. Peak intense 5-min periods have been examined and running performance decrements of 6-17%, directly after these periods, have been found in several other studies investigating elite male [4, 7, 31, 32] and elite female players [33]. While investigating English Premier League players, a decline in performance was also detected after peak intense periods of IA and ID distance (10.4-11-4%) [34]. In addition, one study found a decrease in repeated sprint ability (RSA) directly after an intense period in the first half of football match-play, but at the end of the first half the RSA had recovered [24]. Altogether, the above findings indicate that physical performance is inhibited after peak intense periods during a football game and potentially leads to temporary fatigue, although physical performance fluctuation during a football game may be affected by tactical formations, score line and pacing strategies [7]. For example, one study investigating 11 central midfielders over 35 games from the first league in France, found a small increase (3%) in HIR distance following peak 5-min periods compared to the average 5-min period [35]. Another study investigating HSR (>19.8 km·h-1) distance in peak 5-min periods observed a 15% decrease in running performance in the 5-min period following a peak 5min period compared to average for all players together, though only CM (-33%) and AT (-26%) had a decline in HSR performance when playing positions were taken into account [16]. Thus, not all playing positions necessarily lead to performance decrements after peak periods, and this may be related to different tactical roles and player types. This calls for a positional or individual approach to peak period game demands. Detailed knowledge concerning the most physically demanding period of the game can therefore be of high practical importance.

The methodology most commonly used when investigating the physical game demands of football are video [5, 33] or a multi-camera approaches with predefined 5-min periods [4, 7, 16, 31, 32]. Using predefined periods in research increases the risk of omitting the real peak intense periods during a football game, occurring within two predefined periods. Varley et al (2012) observed a $\sim 25\%$ and $\sim 50\%$ difference in HIR distance between predefined and real peak

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periods and performance decrements, respectively [36]. Furthermore, previous studies have used the manufacturers' software, which often include algorithms, filters and predefined dwell-times that may affect the data [37]. Exporting and analysing the raw data from games will exclude the filters and therefore the potential performance decrements after the real peak periods can be analysed more accurately. Another limitation in the previous literature may be that the 5-minute peak intensity period is a relatively long time during a football game and may include a large part of low intense activities, like standing, walking and jogging (un-published observations). Therefore, an analysis of even shorter peak periods is warranted in the scientific literature.

Besides the fact that periods of decreased performance occur temporarily during a football game, studies have demonstrated that the total HIR and sprinting distance is lower in the second half than in the first half in a football game [4-7, 31, 32, 38]. The amount of HIR also seems to be lower (20-45%) in the last 15-minutes compared to the first 15-minutes of the game [4, 5, 35]. In a study by Mohr et al (2003), substitutes covered 15% more HIR distance during the last 15-min period of the game than players completing the full game [5]. This phenomenon has been found in a more recent study investigating English Premiere League players [7]. Finally, it has been demonstrated that intermittent high intensity running, jumping and sprinting performance declines directly after, compared to before, a football game [27]. Altogether, the above findings indicate that physical performance seems to be effected negatively towards the end of a football game.

Physiological mechanisms of fatigue in football

The cause of a temporary decline in performance after intense periods during a football game appears to be complex, with several contributors such as pacing strategies and tactical circumstances [7], as well as limitations of the physiological and metabolic systems. Focusing on the physiological mechanisms, it has been suggested that temporary fatigue is associated by increased levels of muscle lactate or accumulation of hydrogen ions leading to lower muscle pH [22]. In contradiction, one study found that muscle pH is only moderately reduced and muscle lactate moderately increased during a football game, indicating that temporary fatigue is caused by other physiological limitations [24]. Furthermore, it has been proposed that temporary fatigue can be initiated by a decline in creatine phosphate (CP) levels as CP levels have been

shown to be almost fully depleted in individual muscle fibres at the point of fatigue [24]. Nonetheless, CP levels in the final part of Yo-Yo IR2, including a large anaerobic component, was not different from baseline levels, which argues against the hypothesis that lower levels of CP are a main cause to temporary fatigue during [36]. Extracellular accumulation of potassium (K⁺) has been suggested as a potential mechanism involved in the development of temporary fatigue during a football game [37-39]. At the point of exhaustion after intense exercise lasting 3-5 min, the concentration of K⁺ can reach levels of around 12 mmol·l⁻¹ [39]. These levels are enough to depolarise the muscle membrane potential and reduce the ability of force development [40]. Moreover, a high work rate in peak intense periods during a football game has been associated with a higher level of skeletal muscle Na⁺-K⁺ pump subunits [41] and anaerobic capacity in intermittent high intensity tests [42]. In order to be able to maintain a high exercise intensity and delay muscle fatigue, the Na⁺-K⁺ pump subunits may be an important shuttle of accumulated extracellular K⁺.

The underlying physiological mechanisms of performance decrements towards the end of a football game have also been studied. The decrease of blood lactate and the increase of fatty acids at the later end of a football game is likely to be a result of the low levels or depletion of the glycogen stores, and may affect the exercise performance [1]. This is confirmed by several studies taking muscle biopsies after a game [24, 43, 44]. Moreover, glycogen depletion has been observed in multiple muscle cell locations after a soccer game [44], which is likely to affect muscle function [45]. Furthermore, Beta-hydroxyacyl-CoA-dehydrogenase enzyme (HAD), a common marker involved in the oxidation of fatty acids, seems to be the strongest muscular predictor of football endurance [41]. In fact, a significant correlation was found between skeletal muscle maximal HAD activity and TD covered (r = 0.66) during a football game. The authors also found a correlation (r = 0.55) between HAD activity and distance covered at high intensity during the last 15-minutes of a football game [41]. Collectively, the above findings are pointing out HAD activity as an important marker for fatigue resistance in a glycogen depleted state, as has been demonstrated at the end of a football game [24].

Post-game fatigue and recovery in football

The physical performance seems to decline and fatigue occurs in the end stages of a football game, with the recovery process of getting back to the same

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performance level being relatively slow, taking several days [46]. In today's elite football there is a high number of games including league, different cup and national team games during a normal season. The schedule is often congested with 2-3 games in a given week, with only 3-4 days to recover.

The physical and physiological recovery process after a football game has been defined as when the value of a specific marker has returned to or above pre-game values [47]. As mentioned in the introduction above, high glycogen levels seem to be important for the maintenance of intensity throughout a football game. Studies have concluded that the time required for the restoration of glycogen to baseline levels after a football game is ~24-48 hours [43], but for type II fibres it may take up to 72 hours to completely restore muscle glycogen [44]. It thus seems important for football players to restore glycogen levels after a football game, in order to retain the ability to perform high-intensity activities. In addition to lowered glycogen levels, a number of performance and physiological markers have been studied during the recovery period after a football game.

The physical strain during a football game cause muscle damage and is defined as a mechanical disruption of the muscle fibre which includes membrane damage, myofibrillar disruption characterised by myofilament disorganization and loss of Z-disk integrity [48]. Muscle damage is linked to a temporary decrease in muscle function, increased muscle soreness and an increase of intracellular proteins leaking into the blood circulation [49]. The elevation of muscle damage blood markers has been strongly correlated with the number of sprints during a football game (r = 0.88 and r = 0.75 for creatine kinase (CK) and myoglobin, respectively) [50]. The time course of these different blood markers between studies differ markedly and are back to baseline within 48-72 hours after a football game.

The most common tests used to investigate fatigue and recovery in physical performance after a football game are single sprints, repeated sprints, jumping ability and maximal voluntary contraction strength tests (MVC). Neuromuscular performance has a large variation between studies and complete recovery occurs within 5 to 96 hours post-game [46, 51-55]. Studies have found that MVC ability in knee flexors can be effected for up to 72 hours after a football game [52, 55]. However, as football is a sport with complex movements it would be rational to assume that muscle groups other than knee flexors and extensors are effected during and after a football game. Individual muscles or muscle groups may be loaded differently, depending on the

individual playing style, training status of the individual muscle group, muscular imbalances and motor skills. Yet, no study has investigated the time-course of fatigue and recovery kinetics for multiple muscle groups in the lower limb and trunk muscles of competitive football players. Furthermore, previous studies have described post-game fatigue and recovery kinetics on group level. Thus, the scientific literature is lacking information of inter-player variation in recovery kinetics on a given physical load after a football game, and fatigue and recovery in all individual muscle groups effected during game-play.

The large variation in fatigue and recovery markers after a game can be explained by the multitude of different football game protocols applied in different studies. Some studies used real football game settings or a naturalistic design [46, 51, 52]. The large individual and positional variations in physical characteristics [65] which have been found between football players, makes it rather complex to accurately decide when the players have recovered. An individual focus and the application of simulated football models with standardised physical and physiological responses would be preferable to study inter-individual differences in fatigue development and recovery during and after a football game. A few studies have used simulated protocols performed on treadmills in a laboratory environment [55, 56], which has been reported to be less demanding for the aerobic system than a real football game [55]. One simulated football model performed on a football field has shown strong validity in physical and physiological response and fatigue development towards the end of a real football game [57], but post-game recovery kinetics are yet to be explored.

Training to resist fatigue in football

It is of great importance for football players to have a high endurance capacity and also to be able to perform maximum or near maximum repeated exercise in intense periods and throughout the game in order to maintain performance and reduce the risk of injury [1, 5, 58, 59]. For example, elite football players with high aerobic intermittent capacity have reduced risk of injury when they are exposed to a rapid increase in workload during the season [58, 59]. It has also been found that team sport players with high anaerobic capacities have lower risk of injury than players with low anaerobic capacities, when exposed to a given physical load [60]. Furthermore, Mohr et al (2016) found a very strong correlation between physical capacity and performance during a football game

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on a competitive level [41]. Similar results have been demonstrated in other studies [54, 61]. These findings, along with previous mentioned correlations between physical game-variables and muscular proteins and enzyme activity, indicates that physical training methods for football players should aim to resist fatigue temporarily, throughout and after a football game, in order to maintain performance and decrease the risk of injury during the football season.

Aerobic training in football

Aerobic training in football is important for endurance capacity and to maintain a high intensity in the later stages of a football game [62]. It is well known that aerobic high intensity interval training in HR zones of 90-95% of maximum HR improves aerobic performance in athletes [63, 64]. It has been shown that adding aerobic high intensity training intervals led to improvement in VO2 maximum, ventilatory and lactate threshold, as well as the ability to oxidise fat relative to carbohydrates [65]. Furthermore, studies have found that fat oxidation is positively affected by submaximal aerobic training [66] and high intensity interval training [67]. Moreover, positive changes in muscular oxidative capacity [68] and an up-regulation in muscular antioxidative capacity [69] has been demonstrated after a period of aerobic training. However, most studies have investigated inactive subjects or amateur players, and studies investigating elite football players are warranted [70].

Small-sided games (SSG) are one of the most common training drills, as it has been shown to develop both technical and tactical skills while imposing a significant physiological load on the players [71, 72]. It has also been shown that SSG can increase aerobic capacity in football athletes [73, 74]. There are a number of variables affecting the training intensity of SSG formats. For example, increasing the pitch-area during SSG led to elevated HR, lactate levels and rate of perceived exertion [75, 76].

As described above, a number of studies have examined the physical and physiological responses during different SSG regimes, but information on muscular enzymes and protein content responses to football SSG play is lacking in the scientific literature. Moreover, as the physical demands of a football game seems to differ between positions and between individual players, training for specific game demands would be preferable.

During the last decade, a few studies have compared the physical and physiological demands of different SSG formats and game-play in various variables [77, 78]. Dellal and colleagues (2012) found that playing SSG with 4 players against 4 players (4v4) included more HIR and sprinting distance per minute played for all different playing positions when compared to a friendly game [79]. Another study compared physical friendly game demands with different SSG formats (3v3, 5v5 and 7v7) with a constant area per player (210 m²) [77]. Contradictory to the previous study, distance covered and time spent in HSR and sprint per hour played, was greater in friendly games than in SSG, while overall workload and distance covered was greater in SSG [77]. Similar results were found in a more recent study [80]. Previous studies compared means between physical variables of SSG and game-play, thought the interindividual variation in physical game demands seems to be large. It is therefore of more interest to examine the associations between physical responses in different SSG and game-play with an inter-individual approach. Furthermore, comparing physical variables of SSG and the whole game does not give information regarding how different SSG regimes relate to physical demands of peak intense periods occurring during the game.

Anaerobic training in football

Except for the high aerobic demands of football game-play, short intense periods during game-play have been shown to markedly increase HIR and sprinting, and recovery time is decreased between high intensity efforts [16]. As discussed earlier, it has been shown that immediately after these intense periods players seem to experience temporary fatigue [4, 5, 7, 62]. In these periods the anaerobic systems are highly taxed [24] and the importance of training the anaerobic energy pathways for players to be able to resist fatigue and perform during these periods is highlighted.

Speed endurance training

During the last ten years intensified training has been studied in a football environment. The most studied intensified training regimes on football players have been speed endurance training which is subcategorised to speed endurance production training (SET) and speed endurance maintenance training (SEM).

SET requires a maximum all-out effort with an exercise time of 10-40 seconds and a recovery period 5 times as long as the working time [26]. The purpose of SET is to be able to perform near maximum for a short period of time [64]. Studies have found that SET performed as running drills elicited

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higher intensity and physiological response than SET performed as 1v1 SSG [81] and is also of a higher intensity than SEM [82]. Furthermore, additional SET training has been shown to improve intermittent high intensity and repeated sprint performance in male competitive [82-84], junior [85] and professional football players [86]. Moreover, Thomasson et al (2009) reported an elevation of fatigue resisting markers such as Na⁺-K⁺ ATPase subunits [87] when lowering the training volume (\sim -30%) for 2 weeks, while adding SET to elite football players' training. Contradictory to the study by Thomasson and colleagues, another study reported decreased or unchanged Na⁺-K⁺ ATPase subunits after five weeks of SET training once a week [84]. In addition, SET has been shown to improve VO2 max with 3-7% and an increase in VO2 max running speed in moderately trained subjects after two weeks of training [83]. SET may also improve the oxidative capacity of football players, which is essential for optimal recovery from intense exercise in a game, since several studies have demonstrated an up-regulation of mitochondrial function after SET [83, 88, 89]. However, this has not been studies in competitive male football players.

Differing from SET, SEM training has an exercise time of 20-90 seconds with 50-100% of maximum effort. Recovery time is the same as exercise time and the aim of SEM is an increased ability to sustain high intensity [64]. In contrast to SET, SEM is well suited to SSG with a lower number of players (1v1, 2v2, 3v3) [26, 81, 82]. This type of training has been shown to increase the ability to perform high intensity exercise [26, 82, 90, 91].

There is scientific evidence indicating that different types of speed endurance training is beneficial for football players' physical performance, but some of the studies show conflicting results in muscular response and protein expression, and more research is warranted before conclusions can be drawn. Furthermore, most speed endurance studies have compared physiological responses to SET with inactive subjects or to physical responses SEM. Information about how physical and muscular responses differ between speed endurance training and other training regimes in elite football players is warranted. Thus, as SSG is a commonly used physical training method in football environments, it would be of high relevance to investigate differences in physical and muscular adaptations between speed endurance training with SSG in well-trained football players.

Aims

By the application of quantitative methods, the overall purpose of the present thesis was to study physical demands, fatigue and recovery profiles in male elite football players with an emphasis on individual variations between players and implications for fitness training strategies. The thesis is founded on four studies. The specific aims of the four studies were:

- Study 1: To examine different types of short-lasting peak-intensity periods in top-class football as well as the variability of these periods in relation to playing position and individual game demands.
- Study 2: To study the individual fatigue and recovery responses of multiple muscle groups after a standardised workload resembling a competitive football game using a simulated football model.
- Study 3: To investigate the inter-player relationships between physical game demands in full sized football games and those found in conventional small-sided game formats.
- Study 4: To examine performance responses and muscular adaptations in individual speed- endurance production training compared to small-sided games in elite male football players.

Methods

Overview

In all four studies, a quantitative approach was chosen. Study 1 is a descriptive study, focusing on intense periods and using individual multi-camera game activity data from one male English Premiere League team and their opponents over the course of three seasons. In study 2 a within-subject design was chosen, and a cohort of well-trained competitive football players from the second and third division in Sweden was included in the study. Fatigue and recovery data from different muscle groups was collected measuring isometric voluntary contraction and blood markers before, during and after two simulated football games. In study 3 male professional players competing in the first and second division in Sweden were included and a correlational design was applied to verify the association between physical metrics, using global positioning system techniques, during various small-sided game formats and full sized games. Finally, a randomised controlled design was applied in study 4 and competitive players from two teams in the third division in Sweden participated. To measure physical and physiological differences between speed endurance training and SSG, muscle biopsy techniques and different physical testing protocols were applied. Parametric statistics were used in all four studies.

Ethical considerations

In Studies 1 and 3 institutional approval was given before starting and Studies 2 and 4 were approved by the local ethics committee in Gothenburg (Dnr: 351-15 and Dnr: 687-15, respectively). In all studies except Study 1, for logistical reasons, all participants were informed in writing as well as being verbally informed about the potential risks and discomforts and all gave their written consent before taking part in the study. All studies were conducted in accordance with the declaration of Helsinki (2008). To ensure confidentiality of the participating players in the studies, all data was anonymised before analysis and the computer was stored and locked in a cabinet between analyses. In Study 4, muscle samples using the Bergström needle muscle biopsy technique was

conducted. The complication rate of this specific biopsy technique has been found to be very low (<1%) with skin infection as the most common complication (0.06%) [92, 93]. Soreness, swelling, pain and discomfort have been reported by subjects after undergoing muscle biopsies, though are very rare (\sim 2 out of 16 000 samples) and seem to be fully resolved within 7 days [94]. Thus, it seems that the muscle biopsy technique used in Study 4 is of low risk of participants and is a time efficient method for the collection of muscle tissue for research.

Study 1

Participants

In Study 1, altogether 1105 individual game observations from 473 top-class players belonging to top teams in the English Premiere League were collected. The participants who played full time represented five different playing positions: CD (n = 100), FB (n = 72), CM (n = 74), WM (n = 56) and AT (n = 58). Furthermore, substitutes playing only in the second half and at least the last 15-min of a game, were examined in the same playing positions: CD (n = 12), CM (n = 24), WM (n = 30) and AT (n = 32).

Data collection

In Study 1, a multi-camera system (Amisco. Pro, version 1.0.2, Nice, France) was used to capture game activity during games included in the study. The system measures at a frequency at 10 Hz and the signals and angles are converted into digital raw data and transported to computers for further analysis. Distances were divided into the following speed categories: Total distance (TD) was defined as >0 km·h⁻¹, Jogging as >11 km·h⁻¹, high intensity running (HIR) as >14 km·h⁻¹, fast running (FR) as >17 km·h⁻¹, high speed running (HSR) as >21 km·h⁻¹ and sprint >24 km·h⁻¹. These speed categories have been used in previous studies [32, 41]. Distance data in the different categories was analysed in 1, 2, 5 and 15-minute moving average periods. Individual data from 62 games involving 24 different teams was analysed over three seasons in the English Premier League.

To investigate in-game fatigue patterns distance covered in the previous described speed categories were analysed in 1, 2, 5 and 15-min intervals using a moving average in a macro in Microsoft Excel (version 2013). Furthermore, we

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used raw data from the multi-camera system to avoid unnecessary filters and algorithms that can affect the data analysis using manufactures' software.

Statistical analysis

The Shapiro-Wilks test was used to test the data for normality. The results are presented as the mean and standard deviation (SD). Differences in distance covered in different speed categories between first and second half was analysed using a student's paired T-test. Differences in distance covered in different speed categories between the five playing positions were determined using one-way analysis of variance (ANOVA). Differences between periods of 1, 2, 5 and 15-min were analysed using one-way ANOVA with repeated measures. Two-way ANOVA with repeated measures and one-way ANOVA were used to analyse differences between full-time players and substitutes. When significant differences were detected Tukey's post-hoc test was used to identify specific differences between the means of playing positions and periods during the game. Statistical significance was set to p < 0.05. Pearson's regression test was used to determine and test the correlation coefficient. The statistical testing was conducted using the statistical package for the social science (SPSS version 23) (IBM, New York, USA).

Methodological considerations and limitations

The multi-camera approach has previously been used in studies [4, 95] and has been proven to be a valid (ICC > 0.95) [96] and reliable (CV < 2.4%) [4] measure of distances in different speed categories in football. Furthermore, the multi-camera system has been shown to be a sensitive method to detect running fluctuations during a football game [11]. One limitation with multi-camera systems is that they do not measure ACC and DEC variables with high accuracy [97]. Therefore, in Study 1 we only investigated running distance in different speed categories and excluded micro movements.

We also used moving average time periods. A moving average is a calculation used to analyse data points by creating a series of averages in parts of the full data set. Using moving averages of 1, 2, 5 and 15-min intervals it is possible to analyse the real peak period of the game compared to pre-defined time intervals used in previous studies [98].

Study 2

Participants

Twelve competitive male football players from the Swedish second and third divisions were used in the study. The mean \pm SD data is as follows: age: 23 \pm 4 years; body mass: 75 \pm 6 kg; height: 180 \pm 8 cm; VO_{2max}: 61 \pm 3 mlO₂·min⁻¹·kg⁻¹; Yo-Yo IR2 performance: 927 \pm 124 m. All included participants had to perform at a minimum of 800 m in the Yo-Yo IR2 test and be free from injury at least six weeks prior to the start of each study. Further, all players had at least five years' experience of training and game-play on competitive level or higher. All participants were encouraged to maintain normal eating and sleeping habits during the data collecting period and avoiding drinks high in caffeine and alcohol.

Data collection

The timeline and procedures of the data collection in Study 2 can be viewed in Figure 1. One week before the start of the data collection, the subjects conducted a VO_{2max} treadmill test and a Yo-Yo IR2, with three days in between. One hour after the Yo-Yo IR2 the participants also completed one familiarisation session of the simulated football model and MVC measurements, to determine individual configurations. Two simulated football games were then performed, separated by 72 hours of recovery. Immediately after the simulated football model, the participant walked back to the laboratory (~400m) for MVC testing. Blood samples were taken before warm-up, immediately after the game and every 24 hours during the recovery, with subjects seated.
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Figure 1. Timeline and procedures including pre-experimental testing and familiarisation. The thin vertical arrows point out the time point were the maximum voluntary contraction tests (MVC) and blood sampling took place. CST1 = the first Copenhagen soccer test and CST2 = the second Copenhagen soccer test.

Simulated football model

The Copenhagen soccer test (CST) was used in Study 2 and is a simulated football model performed individually on a football field (Figure 2). A repeated sprint test (RST) was performed after a warm-up consisting of five 2 x 20-m shuttle sprints with 30 s of rest in between and times were recorded using Muscle Lab V8 (Bosco System, Rome, Italy) photocells with precision of 0.001 s. The CST consist of 2 x 45-min halves with a 15-min break between them. CST is divided into 18 periods of approximately 5-min each, varying in intensity between low (L), medium (M), and high (H). Blood samples from the fingertips were collected at rest, after the warm-up, before the first half, after 15, 30, and 45-min of the first half, before the second half, and after 15, 30, and 45-min of the second half. In Figure 2 the specific activities can be visualized and a more detailed description of the CST protocol can be read in paper 2.



Figure 2. Schematic presentation of the Copenhagen soccer test, with all movement in various directions. BW = backwards running, LS = low speed running, MS = moderate speed running, Slide = backward slide. (The illustration is adopted from Bendiksen et al, 2012.)

Physical and physiological response during CST

To asses physical activities during CST, 10-Hz S5 GPS devices (Catapult Innovations, Melbourne, Australia) placed between the players' shoulder blades were used. Distance covered between 11-14, 14-17, 17-21, 21-24 and 24-40 km·h⁻¹ were analysed during the simulated football model, and medium intense accelerations (MIA) and medium intense decelerations (MID) were collected and defined as efforts >2 m·s⁻² and -<2 m·s⁻², respektively. Polar chest-strap monitors were used to asses HR and was measured in 5 s intervals, and every 15-min during the simulated game a blood sample from the fingertip was taken for lactate analysis.

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Maximum isometric voluntary contraction

Before and after the CST, the subjects conducted MVC in different muscle groups (knee flexors, knee extensors, ankle extensors, hip adductors, hip lumbar/thoracic abductors. flexors, lumbar/thoracic extensors, and lumbar/thoracic rotators) using the David system F300 (David, Outokumpu, Finland) except for ankle extensors (Isomed 2000, D&R Ferstl, Hernau, Germany). Before each MVC on CST and on recovery days subjects conducted a standardised warm-up routine consisting of a 10 min jog on a treadmill at a speed of 10 km·h⁻¹ as well as 4 repetitions of concentric lumbar/thoracic rotator contractions on each side, with 30 kg resistance. The test was supervised by experienced personnel and they also supported the participants with verbal encouragement during the test. (For more details see paper 2)

VO2max test

In a laboratory environment a VO2 max treadmill (RL2500E, Rodby, Sweden) protocol was performed. The test included a 3 minute warm up at 10 km \cdot h⁻¹ and 1° elevation followed by a gradual increase in velocity and elevation until volitional exhaustion. Heart rate was continuously monitored in order to determine the individual HR max during the CST. Pulmonary oxygen uptake was measured by open-circuit spirometry (every 30 s) using an automated online pulmonary gas exchange system via breath-by-breath analysis (Jaeger Oxycon Pro, Erich Jaeger, Viasys Healthcare, Germany). The system was calibrated before each trial with two gases of known concentrations.

Yo-Yo intermittent recovery test level 2

In the week before the first CST a Yo-Yo Intermittent recovery test level 2 (Yo-Yo IR2) was used to determine the intermittent anaerobic running capacity of the participants. The test consisted of repeating two 20-m runs at a progressively increased speed controlled by audio bleeps from an audio recorder. Between each running bout the participants had a 10-s rest period. The participants were asked to maximise their effort and were verbally encouraged throughout the test. When the participants failed to reach the finishing line in time twice, the distance covered was recorded and represented the test result. The test was performed on artificial turf on a 2-m-wide and 20-m-long running lane marked by cones.

Blood analysis

Blood samples were taken from an antecubital vein in the right arm using flexible Venflon cannulas with participants seated. Venous blood was drawn in vacutainer EDTA tubes and serum separation tubes. EDTA tubes were centrifuged immediately at 4°C and plasma stored at -80°C until analysis. Blood in serum separation tubes was allowed to coagulate at room temperature, centrifuged and serum stored at -80°C until analysis. Blood samples from a fingertip were also taken during the CST to measure capillary blood lactate using a Biosen analyzer (Biosen C-line, EKF-diagnostic GmbH, Magdeburg, Germany).

Statistical analysis

The data was tested for normality using the Shapiro-Wilk test. Data is presented as mean and SD. The mean of the MVC as well as CST sprint times, lactate and blood levels were analysed using one-way ANOVA with repeated measures. If a significant interaction was found, Tukey's post-hoc test was used to identify the point of difference. Significance levels were set to p < 0.05. Correlation was investigated between physical capacity, GPS, blood and the percentage change in MVC performance 0 hour post CST, using a Pearson product moment correlation. SigmaStat for windows version 11.0 (Systat Software, San Jose, USA) was used for all statistical analysis.

Methodological considerations and limitations

The Copenhagen soccer test (CST) is based on a study conducted on Italian professional football players [5], and football specific movements are included. The CST has been found to elicit similar physical and physiological responses as well as fatigue development as a real game on elite level and has been found to be of high reproducibility [57].

In Study 2 we used maximum isometric voluntary contraction (The David system F300, David, Outokumpu, Finland) to measure potential muscular performance changes in different muscle groups after simulated football models. This specific system has been tested for reliability with ICC values exceeding 0.75 [99]. The systems used have been shown to have ICC and CV of 0.80-0.88 and 5.4-7.3% respectively, for isometric hamstring measurements in elite football players [100]. One limitation in Study 2 may be a lack of information on test-retest values of other muscle groups such as hip abductors

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and hip adductors. Moreover, most reliability studies on MVC have investigated students or elderly and test-retest values is warranted on well-trained athletes to be able to draw large conclusions.

The inclusion criteria in Study 2 of a Yo-Yo IR2 performance result of >800 m were determined based on a study by Krustrup et al (2008), where all international elite players displayed a result of over 800 m [36]. The test has been strongly related to distance covered in intense periods during a football game and muscle variables of importance to anaerobic capacity [41]. The Yo-Yo IR2 test has been shown to be of reasonable reproducibility (CV = 9.6%) [61]. In Study 2, one potential limitation of the Yo-Yo IR2 test result may be that we did not use HR monitors and although the participants were asked to maximise their effort and encouraged verbally, they may have finished before they reached maximum effort.

Study 3

Participants

In Study 3, forty-five professional male football players from two teams in the Swedish first division and one team in the second division participated. The players play in the following positions: CD (n = 9), FB (n = 9), CM (n = 8), WM (n = 9) and AT (n = 10). The characteristics of the players were as follows: age: 24 ± 5 years; body mass: 77 ± 5 kg; height: 181 ± 5 cm; and Yo-Yo IR 2 performance: 1077 ± 171 m. All players had a minimum of 5 years of training and competing on elite level.

Data collection

A Yo-Yo IR2 test was performed by the participants two weeks before the start of the study for descriptive data. During the study period of six weeks during preseason the participants performed three types of SSG with goalkeepers, as well as six full-sized games (FSG). The participating players included in the study played three FSG each and played at least the first half (45-min), and the average of these was analysed. Pitch size during the FSG was 105 m (length) x 65 m (width), which gives a relative pitch area per player of 620 m². Data from the first half of the FSG was analysed as it has been shown to be more intense than the second half [4, 5]. The players were assessed once in each SSG as this training regime has a high reproducibility (ICC = 0.99) [101].

GAME DEMANDS AND FATIGUE PROFILES IN ELITE FOOTBALL

The SSG formats were 4v4, 6v6 and 8v8 and one of the SSG formats was completed 72-96 hour before each FSG. Participants were divided into teams by the researchers according to their position during FSG. The total playing time during all different SSG was 18 min divided in 6×3 min (recovery 1 min), 2×9 min (recovery 2 min) and 1×18 min for 4v4, 6v6 and 8v8 respectively. The pitch sizes were chosen from the team's normal training: 30×40 (240m² per player), 50×40 (286m² per player) and 70×60 m (467m² per player), respectively. GPS devices (10-Hz S5, Catapult Innovations, Melbourne, Australia) were used to estimate physical activity during SSG and FSG and the same GPS devices were placed on the same players in all data collections.

Physical variables and speed categories

Activities during SSG and FSG were divided into the following definitions: maximum running velocity (MRV); total distance (TD) as >0 km·h^{-1;} distance (d) and efforts (e) in high-intensity running (HIR) was defined as >14 km·h^{-1;} fast running (FR) as >17 km·h⁻¹; and high speed running (HSR) was defined as >21 km·h⁻¹. GPS was also used to estimate total acceleration (ACC) and deceleration (DEC) distance and efforts. Intense acceleration (IA) and intense deceleration (ID) distance and efforts were also analysed during FSG and SSG and defined as changes of velocity >3 m·s⁻² [34].

Peak periods were defined as the greatest distance or the maximum number of effort in the physical variables in 1-min, 2-min and 5-min periods during the first half of the FSG. The time-periods were predefined and independent of each other. Data was transferred to the manufacturer's software (Catapult Sprint version 1.5.4). Dwell-time for minimum effort duration was set to >0.4 seconds. Data sets were verified for number of satellites connected (mean >8) and horizontal dilution of precision (mean < 1.2) before being included in the analysis [102].

Statistical analysis

Data is presented as mean and standard deviation. The Pearson productmoment correlation coefficient was used to analyse associations between the physical variables during each SSG and mean of first half of FSG. To determine the magnitude of correlations, Hopkins (2009) thresholds were used and defined as weak (>0.1), medium (>0.3), strong (>0.5), very strong (>0.7) and extremely strong (>0.9) [103]. Coefficient of variation (CV) was used to analyse

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inter-individual variation of the physical variables in the different SSGs and the first half of FSG. Pearson's regression test was used to analyse the causal relationship between physical variables during each SSG and the mean of the first half of FSG. Significance level was set to p < 0.05. The data was exported to Microsoft Excel for analysis and further exported to SPSS for statistical testing.

Methodological considerations and limitations

GPS is a system, connected to a number of satellites, providing the unit with position and time. GPS technology has improved in terms of accuracy and precision in the past years [104], even though the technology has a number of limitations. For example, high inter-unit errors between different models has been found [105] and other sources of error could include satellite availability [106], algorithms and filters in hardware or software [107]. GPS units with 10 Hz measuring frequency from the brand used in Study 2, 3 and 4 has been found to be able to validly measure distances during linear running and simulated circuits in team sport in different speeds and distances (CV = 1.9-10.5%) [104]. Furthermore, the validity of GPS based acceleration data from the units used in Study 2, 3 and 4 is CV = 3.6-5.9% compared to laser technology [108]. The same study displayed inter-unit CV values for acceleration of 1.9-4.3% [108]. The data collection took place on open space football-fields witch made it possible to have high satellite availability.

Study 4

Participants

Thirty-nine competitive male football players from two teams in the third division in Sweden agreed to take part in the study. Their data is as follows: age: 21 ± 2 years; height: 184 ± 7 cm; body mass: 78 ± 8 kg; Yo-Yo IR2 performance of 573 ± 142 m. The participants represented all outfield positions: CD (n = 7), FB (n =8), CM (n = 6), WM (n = 10), AT (n = 8). The study started two weeks into the pre-season (January 2016) and the participants had four training sessions a week and did not play any games during the intervention period. All participants had at least five years of experience of football on a competitive level were free from injury at least 6 weeks prior to the data collection.

Data collection

Based on playing positions in their respective teams, the participants were randomised for a speed endurance training group (SET; n = 21) or a SSG training group (SSG; n = 18). The two groups performed two different types of training which were added to the players' normal training programs three times a week for 4 weeks in total. The normal training lasted for ~60 min and included ~15-min warm-up, ~15-min technical training and ~30 min tactical training. The SET drill was individually performed in 30-s intervals separated by 150 s of passive recovery (Figure 3). The participants continued the drill for 30 seconds regardless of whether they reached the finish-line before that time. The number of exercise intervals was six during the first intervention week, eight during the second and third weeks, and ten during the fourth week. The participants were asked to run with maximum effort during the entire drill and were continuously given verbal encouragement.

The SSG group performed a 6v6 football game on a pitch 40 m long and 32 m wide. The training was performed in intervals recommended for moderate intense training for SSG [109] lasting 2 x 7 min in the first week, 2 x 8 min in the second and third weeks, and 2 x 9 min in the fourth week. The participants had a passive recovery interval of 2 min between exercises. The 6v6 SSG were played with normal rules and players were instructed to keep high intensity and were verbally encouraged.



Figure 3. Speed endurance production training drill.

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Physical and physiological training response

A GPS (10 Hz, S5, Catapult Innovations, Melbourne, Australia) was used to asses physical activities during the intervention training. Total distance (TD) was defined as >0 km·h⁻¹, high intensity running distance (HIR) as >14 km·h⁻¹ and high-speed running distance as >21 km·h⁻¹, and were chosen to analyse running intensity during the intervention. Moreover, intense accelerations (IA) and decelerations (ID) were defined as efforts >3 m·s⁻² and <-3 m·s⁻² respectively. To get an indication of difference in the glycolytic loading between the two intervention groups, a blood sample from the fingertip was taken during one of the training sessions in the third week of the intervention and analysed for lactate levels. The baseline sample was taken 5-min before the normal training and in the SET group the second and third sample was taken after 4 and 8 sets respectively. In the SSG group the second and third sample was taken between the first and second interval.

Assessment of performance effects of intervention

To assess the physical effects of each intervention training method, different physical test protocols were performed 1 week before and after intervention. The participants conducted a Yo-Yo IR2 (see the description on method section of Study 2) outside (~3-8°C) on artificial grass. A 5 x 30 m repeated sprint test (RST) with 25 seconds of passive recovery (light jogging back to starting line) between sprints was also performed. A mean sprint time over the five sprints was calculated as well as a fatigue index as the percentage difference between the first and last sprint [110].

An arrowhead agility test (AAT) was also performed before and after the intervention. Cones were placed in an arrowhead shape with two cones representing the starting line. The test consisted of two trials to the left and two to the right and the fastest time in each direction was analysed [111]. Both RST and AAT was performed indoors (~20°C) on a wooden floor and sprint times were recorded with Muscle Lab V8 (Bosco System, Rome, Italy). All participants were familiar with the tests as they were included in the team's testing battery.

Muscle biopsies and analysis of protein expression

One week before the start of the intervention and three days after the last training, 27 participants had a muscle biopsy taken from the m. vastus lateralis

of the dominant leg (~ 70–120 mg wet weight). A modified Bergström needle biopsy technique with suction was used to collect muscle samples [94]. The muscle tissue was taken in resting conditions with the subjects lying in a supine position on a portable bed. The muscle tissue was immediately frozen in liquid nitrogen and stored at -80 °C. The frozen sample was weighed after freezedrying as well as 1 hour later to correct for the water content. After freezedrying, the muscle samples were dissected free of blood, fat, and connective tissue. Next, 1-2 mg dry weight muscle tissue was extracted in 1 M HCl, hydrolysed at 100 °C for 3 hours, and the glycogen content determined using the hexokinase method. Maximal CS, 3-hydroxyacyl-CoA-dehydrogenase phosphofructokinase (PFK) activities were determined (HAD), and fluorometrically in triplicate for each biopsy on a separate piece of muscle from the biopsy, as described by Lowry in 2012 [112].

The protein expression was determined through the Western Blotting technique as described by Thomassen et al (2010), where ~2 mg muscle tissue was split and analysed in duplicates for each sample [87]. Muscle markers for ion transportation Na⁺-K⁺ ATPase $\alpha 1$, $\alpha 2$, $\beta 1$ and phospholemman protein 1 (FXYD1) were analysed, as well as monocarboxylate transporter 4 (MCT4) and Na⁺/H⁺ exchanger 1 (NHE1) for muscle lactate regulation. Glucose transporter type 4 (GLUT-4) and glycogen synthase (GS) were analysed and are markers for the ability to transport glucose into the muscle fibre and the glycogen storage capacity in the muscle. Finally, superoxide dismutase 1 and 2 (SOD 1 and 2) and catalase (CAT) were examined for antioxidant adaptations.

Statistical analysis

Change scores in physical test performance and muscular adaptations before and after the intervention between SET and SSG as well as within group differences were determined using the two-way ANOVA test. Physical training responses and lactate levels were compared between groups using two-way ANOVA with repeated measures. The Newman-Keuls post-hoc test was used when a significant interaction was detected, to identify the point of difference.

Methodological considerations and limitations

The repeated sprint ability (RST) is defined as a short duration sprint less than 10 seconds with below 30 seconds of recovery-time in between, and is repeated 3-5 times [113]. This type of activity has been linked to a football game at

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professional level [114]. Moreover, in a study by Mohr et al, a very large correlation (r = -0.76) between total sprint time during the last 15-minutes of a football game and total time in RST was found [41]. Finally, sprint time during RST performance seems to be of high reproducibility (CV = 0.8%) in professional football players [115].

Research has been conducted investigating the importance of agility skills in football [116-120]. These suggestions are based on the fact that football players change their activity every fourth second [12, 121] and that in a single football game there is about 700 changes of direction [121]. The test used in Study 4 is called the Arrowhead Agility Test (AAT) which has been used in previous studies [111, 122] and has been shown to have a high reproducibility with a CV value of <1% [110].

To reduce variability in the fluorometrical muscle analysis we used a triple sample technique with the mean of the three muscle samples used. In a previous study CV values for duplicate muscle samples in maximum enzyme activity has been shown to be 5-12% and 12-25% for protein expression using the western blotting technique [123]. CV values for maximal enzyme activity in Study 4 were shown to be between 4-7%.

Results

Study 1

Game demands and characteristics of peak periods

Distance covered in total during games and distance covered in peak periods is visualised in Table 1. Less distance was covered during the last 15-min in total, >11, >14 km \cdot h⁻¹ and >17 km \cdot h⁻¹ compared to the first four 15-min periods of the game (p < 0.05). The drop in distance covered (p < 0.05) from the mean of the first five 15-min intervals compared to the last 15-min interval were 7 ± 1 , 15 ± 1 , 19 ± 0 , 32 ± 4 and $48 \pm 6\%$ for TD covered, and distance in speed categories >11, >14, >17 and >21 km \cdot h⁻¹, respectively. Further, 22-40% of the 2 and 5-min peak periods were present in the first 15-min of the games in speed categories >11 >14, >17 km \cdot h⁻¹ while they were more evenly distributed throughout the game in speed categories >21 and >24 km \cdot h⁻¹. The peak 1 min distance at speed >14 km·h⁻¹ was 8-12% longer (p < 0.05) in the first 15-min period in each half of the game than the last two 15-min period of the game. Furthermore, peak 2 min distance in >14 km \cdot h⁻¹ was 6-15% greater in the first 15-min period of each half than in the last 15-min period of each half. Finally, the distance covered in peak 5-min >14 km \cdot h⁻¹ in the first 15-min period was 10-22% greater than in all other 15-min periods of the game.

The players had a decline in the 5-min periods after the peak-distance 1, 2 and 5-min periods, (6-20%, p < 0.05) in running distance at >14 km·h⁻¹ of 21, 18 and 17 m, respectively, compared to the average distance covered in a 5-min interval in a game. Declines in distance covered directly after peak 1, 2 and 5min distance were also present in speed categories >17 and >21 km·h⁻¹, but not in speed zone 24 km·h⁻¹ compared to mean values.

Substitutes, only playing the last part of the game covered similar TD in the last 15-min as full time players, but in speed categories >11-> 24 km·h-1, substitutes covered 18-39 % greater (p < 0.05) distances than players playing the full game. Substitutes covered similar distances in peak periods and had similar performance decrements in the 5-min period following peak distance

periods of 1, 2 and 5-min, in speed zone >14 km \cdot h⁻¹, compared to average 5-min, as full time players.

	Total game (m)	1 min peak (m)	2 min peak (m)	5-min peak (m)
>14 km·h⁻¹	2367 ± 35	98 ± 5	135 ± 8	235 ± 13
>17 km·h ⁻¹	1300 ± 70	75 ± 4	96 ± 5	157 ± 8
>21 km·h⁻¹	505 ± 11	51 ± 3	61 ± 3	88±5
>24 km·h⁻¹	225 ± 7	40 ± 2	45 ± 2	58 ± 3

Table 1. Distance covered in total game and in peak periods.

Mean \pm SD of distance covered (m) in speed categories >14, 17, 21 and 24 km·h⁻¹ in total as well as in peak 1, 2 and 5-min periods during game.

Peak periods and playing position

Figure 4 displays positional differences in distance covered at speed zone >14 km·h⁻¹ in peak 1 min (4C), peak 2 min (4B) and peak 5-min (4A) data as well as distance covered in 5-min after each peak period, and the average distance covered for all 5-min periods throughout the game. It can be stated that CM and WM covered a greater distance in peak 1 min and peak 5-min than all other positions (p < 0.05). All positions covered greater distance in peak 1 and 5-min periods (p < 0.05) than CD and greater distance in peak 2 min periods (p < 0.05) than CD and greater distance in peak 2 min periods (p < 0.05) than CD and greater distance in peak 2 min periods (p < 0.05) than CD with an exception of AT, at speed zone >14 km·h⁻¹ (Figure 4A, C). In the 5-min intervals following the peak 5-min, less (p < 0.05) distance was covered (>14 km·h⁻¹) for all playing positions compared to the average distance in a 5-min period of a game (Figure 4A). Moreover, the same pattern was apparent in the 5-min intervals following peak 1 and 2-min periods for all playing positions except CD.

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Figure 4. Distance covered at speeds >14 km·h⁻¹ during peak 5 (A), 2 (B) and 1-min (C), as well as in the next 5-min periods immediately after peak 5, 2 and 1 min and in 5-min periods on average for central defender (CD, n = 100), full-back (FB, n = 72), central midfielder (CM, n = 74), wide midfielder (WM, n = 56) and attacker (AT, n = 58). Data is mean \pm SD. * denotes significant difference from CD. # denotes significant difference from next 5-min. \$ denotes significant difference from FB and AT. Significance level is p < 0.05.

Inter-player variation and individual examples

A large range in distance covered in total and in peak periods was present in all speed categories with absolute values of 856-4161, 314-2557 and 104-1222 m in speed categories >14, >17 and >21 km ·h¹, respectively. Ranges in peak 1, 2 and 5-min distance were 84-159, 139-200 and 259-378 at >14, 67-121, 103-162 and 160-261 at > 17 and 21-115, 22-132 and 28-188 at >21 km ·h⁻¹ in absolute values. Total HIR distance (>14 km ·h⁻¹) and peak 1, 2 and 5-min periods showed significant correlations (r = 0.69–0.90; p < 0.05) with peak 5-min displaying the strongest and peak 1-min the weakest inter-individual relationship.

In Figure 5 the inter-individual differences in game demands in HIR distance in total and in peak 1-min periods between positions and within positions is visualised. A large inter-individual variation can also be seen within the different

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playing positions. For example, one CM covered ~4000 m in total and ~160 m in peak 1-min distances while another CM covered ~1500 m in total and ~80 m in peak 1-min distances at >14 km·h⁻¹ during a game. Another example of difference is that one CD covered ~3000 m in total and ~300 m in peak 5-min distances while another CD covered ~1000 m in total and ~100 m in peak 5-min distances at >14 km·h⁻¹ during a game. Similar ranges can be seen within all playing positions during a complete game and peak 1, peak 2 and peak 5-min distances covered >14 km·h⁻¹.



Figure 5. Inter-individual relationships between total distance covered in high-intensity running (>14 km·h⁻¹) and distance covered in high intensity running (>14 km·h⁻¹) during peak 1-min periods (n = 360). Green plot = Central defenders (CD), orange plot = Fullbacks (FB), red plot = Wide midfielders (WM), yellow plot = Central midfielders (CM), blue plot = Attackers (AT) and black line around the plot = total.

Study 2

Physical and physiological response to Copenhagen Soccer Test

Mean and peak heart rate (HR) responses during the CST were 82 ± 2 and 97 ± 2 of HR_{max} and the distance in different speed categories as well as number

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of medium intense accelerations (MIA) and decelerations (MID) can be seen in Table 2. Test-retest values of distance in each speed categories between the first and second CST were CV = 0.7, 1.1, 11.5, 16.2, 7.1, 22.4 and 66.0% in speed categories of 0-11, 11-14, 14-17, 17-21, 21-24 and 24-40 km ·h⁻¹ respectively. CV values of MIA and MID were 29% and 28% respectively. The last sprint (7.604 \pm 0.564 s) of the CST was significantly slower (p < 0.05) than the first sprint of the CST (7.335 \pm 0.238 s).

In overall time points, blood lactate was higher (p < 0.05) in the first half (7.7 ± 0.3 mmol·l⁻¹) than in the second half (6.3 ± 0.4) of the game. Blood lactate was higher than baseline (2.1 ± 0.9 mmol·l⁻¹) at all time points throughout the CST with the highest value (8.7 ± 2.4 mmol·l⁻¹) 15-min into the game, and was lowered to 6.9 ± 2.5 mmol·l⁻¹after 90 minutes of the game (p < 0.05).

	Mean ± SD
Total distance (m)	11283 ± 400
Distance 0-11 km·h ⁻¹ (m)	7655 ± 233
Distance 11-14 km·h ⁻¹ (m)	834 ± 260
Distance 14-17 km·h ⁻¹ (m)	745 ± 193
Distance 17-21 km·h ⁻¹ (m)	1457 ± 168
Distance 21-24 km·h ⁻¹ (m)	531 ± 222
Distance 24-40 km·h ⁻¹ (m)	62 ± 79
Acceleration Efforts >2 m/s ²	49 ± 14
Deceleration Efforts <-2 m/s ²	38 ± 11

Table 2. Mean \pm SD of physical activities during two Copenhagen soccer tests (n = 12).

Muscle-specific performance

A decrease in all individual muscle groups was found in MVC torque 0 hours after a simulated football game (p < 0.05), and performance in all muscle groups had returned to baseline values within 24 hours after CST (Figure 6A). Knee flexors displayed the largest drop in muscle performance with 14% (198 ± 36 vs. 171 ± 42 Nm) 0 post CST. Lumbar/thoracic extensors performance dropped with 12% (353 ± 51 vs. 308 ± 42 Nm), lumbar/thoracic flexors with 10% (212 ± 31 vs. 191 ± 34 Nm), lumbar/thoracic rotators with 9% (203 ± 43 vs. 184 ± 47 Nm), hip adductors with 9% (437 ± 129 vs. 405 ± 145 Nm), ankle extensors with 9% (191 ± 37 vs. 175 ± 37 Nm) and knee extensors with 8% (240 ± 51 vs. 219 ± 49 Nm) 0 hours after CST. Hip abductors displayed the smallest decline in muscle performance with 6% (315 ± 51 vs. 296 ± 49 Nm) 0 hours after CST.

When individual muscle groups were put together, two different combined muscle groups (knee joint muscles = knee flexors and extensors; trunk muscles = combined lumbar/thoracic extensors, rotators, and flexors), showed the greatest drop in performance with 11% by an absolute change of 376 ± 48 vs. 349 ± 52 and 258 ± 80 vs. 229 ± 70 Nm, respectively 0 hours after CST (p < 0.05; Figures 6B and 6D). Moreover, a decline of 4% in strength performance remained 24 hours post-game for the trunk muscles (258 ± 80 vs. 248 ± 77 Nm, (p < 0.05)) and all other muscle groups recovered. Finally, there was a tendency of an inverse correlation between percentage drop in sprint-time during the last sprint of the CST and the percentage decline in maximal hamstrings performance 0 hours after CST (r = -0.63, p = 0.052).



Figure 6. Post-game MVC results in individual muscle groups (A), pooled in specific muscle categories (B). Data is presented as mean \pm SEM values. C shows individual results in individual muscle groups and D shows individual results in pooled specific muscle categories. * denotes significant differences from baseline at p < 0.05.

Markers of muscle damage and inflammation

There was a significant (p < 0.05) elevation of plasma CK activity 0 hours after CST (~112%) and 24 hours after CST (~190%) compared to the baseline, and the CK activity returned to baseline values within 48 hours after CST. Myoglobin concentration increased with ~559% 0 hours after CST and was back to baseline within 24 hours after CST. No significant difference was observed for concentrations of CRP, which remained unchanged at all time points throughout the recovery period. A significant positive correlation was found between change in CK activity values 24 hours post-game and distance covered during the Yo-Yo IR2 test (r = -0.70; p = 0.02).

Inter-player variation and individual examples

The individual differences in fatigue and recovery responses in the various muscle groups was large between players and between muscle groups (Figure 6C). For example, one of the players had a decline in core muscles pooled together of ~40% 0 hours post-game and it remained below 30% of baseline values 48 hours post-game. The same player had a drop in muscle performance in knee joint muscles of ~8% and returned to baseline values at 24 hours post-game. Another player decreased the performance in knee joint muscles 0 hours by ~40% and remained decreased by ~20% at 48 hours post-game, while the same player's performance in core muscles had declined by ~20% 0 hours post-game and was returned to baseline values at 24 hours post-game and was returned to baseline values at 24 hours post-game and was returned to baseline values at 24 hours post-game.

Study 3

Associations between small-sided games and fullsized games

Overall, 38% of the investigated physical variables showed significant correlations (p < 0.05) between the different SSG formats and first half of FSG. In the 8v8 SSG format, 50% of the physical variables correlated with the FSG, followed by 38% in 6v6 SSG and 19% in the 4v4 SSG format. An extremely strong correlation was found in IDe between the 8v8 SSG format and FSG (r = 0.90, p < 0.001). Very strong correlations were found in IDe and DECd between 6v6 and FSG (r = 0.87, p < 0.001 and 6v6, r = 0.87, p = 0.000, respectively). In 8v8 SSG, four speed categories (FRd, HSRd, FRe and HSRe) displayed strong correlations (r = 0.55-0.65, p < 0.03) with FSG, while no correlations were found in 4v4 and 6v6 in any of the speed categories compared to FSG, except for TD that displayed strong to very strong correlations (4v4, r = 0.61, p < 0.03; 6v6, r = 0.79, p < 0.002) with FSG.

Associations between small-sided games and peak periods during full-sized games

Overall, 22-33% of the physical variables displayed significant correlations (p < 0.05) between SSG and peak intense periods during FSG (Table 3A-C). A very strong correlation was found between 6v6 SSG and peak 5-min period (TD, r = 0.82, p = 0.007) and between 8v8 SSG and peak 2-min period (HSRd r =

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0.82, p = 0.001). The 4v4 SSG format correlated with 11% (DECe, r = 0.64, p = 0.008), 11% (ACCe, r = -0.66. p = 0.005) and 22% (TD, r = 0.67, p = 0.005; and DECe r = 0.57, p = 0.016) of the physical variables in peak 1, 2 and 5-min during FSG, respectively.

In 6v6 SSG format 11, 22 and 33% of the physical variables were significantly correlated with peak 1 (TD, r = 0.73, p = 0.007), peak 2 (r = 0.63, p = 0.027; ACCe, r = 0.67, p = 0.004) and peak 5-min period (TD, r = 0.82, p = 0.001; ACCe, r = 0.81, p = 0.002 and DECe, r = 0.62, p = 0.02) respectively. The 8v8 SSG had the most significant correlations between peak intense periods of the FSG of the different SSG formats in the investigated physical variables.

Peak 1-min periods correlated with 44% (FRd, r = 0.63. p = 0.021; HSRd, r = 0.67, p = 0.012; HSRe, r = 0.60, p = 0.03 and DECe, r = 0.55, p = 0.05), peak 2-min periods with 67% (HIRd,r = 0.61, p = 0.028; FRe, r = 0.59, p = 0.034; HSRe, r = 0.64, p = 0.018; ACCe r = 0.60. p = 0.03; FRd, r = 0.72, p = 0.005 and HSRd,r = 0.82, p = 0.001) and peak 5-min periods with 44% (FRd, r = 0.66. p = 0.019; HSRd, r = 0.74. p = 0.007; HSRe, 0.60. p = 0.02 and DECe, r = 0.58. p = 0.04) of the variables in the 8v8 SSG.

Table 3 A-C. Correlation (r) between different physical variables of players in the 4v4 (n =16), 6v6 (n =12) and 8v8 (n =14) small sided games and the peak 1-min period (A), peak 2-min period (B) and peak 5 min period (C) of full sized games.

	4v4	6v6	8v8
TD	0.42	0.73*	-0.04
HIRd	0.27	0.41	0.51
FRd	0.21	0.42	0.63*
HSRd	-0.18	0.41	0.67*
HIRe	0.41	0.17	0.13
FRe	0.33	0.18	0.46
HSRe	0.00	0.40	0.60*
ACCe	-0.15	0.41	-0.38
DECe	0.64*	0.40	0.55*

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3			
	4v4	6v6	8v8
TD	0.47	0.63*	0.25
HIRd	0.33	0.40	0.61*
FRd	0.25	0.37	0.72*
HSRd	-0.05	0.42	0.82*
HIRe	0.41	0.07	0.32
FRe	0.39	0.14	0.59*
HSRe	-0.21	0.47	0.64*
ACCe	-0.66*	0.67*	0.60*
DECe	0.43	0.39	0.51
	4v4	6v6	8v8
TD	0.67*	0.82*	0.15
HIRd	0.35	0.49	0.33
FRd	0.05	0.14	0.66*
HSRd	0.03	0.21	0.74*
HIRe	-0.25	0.34	0.50
FRe	0.15	0.09	0.45
HSRe	-0.18	0.12	0.60*
ACCe	-0.29	0.81*	0.36
DECe	0.57*	0.62*	0.58*

Table 3 A-C. TD= total distance, HIR = high intensity running (>14 km·h⁻¹), FR = fast running (>17 km·h⁻¹), HSR = high-speed running (>21 km·h⁻¹), ACC = acceleration (>0 m·s⁻²), DEC = deceleration (->0 m·s⁻²). The abbreviation for distance is d and effort is e. * denotes significant correlation (p < 0.05) with full-sized games.

Inter-individual relationships and individual examples

The inter-player variation was large in all variables during all types of SSG (CV = 6-67%) and FSG (CV = 7-48%), as well as peak 1, 2 and 5-min periods during the FSG (CV = 6-45%). In figure 7A, one example of a strong inter-individual relationship (R^2 = 0.55) in one speed category (HSRd), between one SSG format (8v8) and peak intense period (peak 5-min) during FSG is displayed. Figure 7B

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shows another example in one speed category (HSRd) between one SSG format (4v4) and FSG, but where no inter-individual relationship was found ($R^2 = 0.06$).



Figure 7. (A) Inter-individual relationship (R²) between peak 5-min high-speed running distance (>21 km·h⁻¹) during full-sized games (FSG) and high-speed running distance during 8v8 small-sided games (SSG). (B) Inter-individual relationship (R²) between total high-speed running distance full-sized games (FSG) and high-speed running distance during 4v4 small-sided games (SSG).

Study 4

Physical and physiological training response

TD covered during the intervention period was higher (p < 0.05) in the SSG group (1683 ± 348 m, n = 13) than in the SET group (1364 ± 84, n = 17). Distance covered in the other speed categories were higher in the SET (>14 km · h⁻¹, 826 ± 102 vs 180 ± 133 m; >21 km · h⁻¹, 239 ± 53 vs 14 ± 15m) than in the SSG group. The SET group also had a higher (p < 0.05) number of IA (38

 \pm 9 vs 27 \pm 14) and ID (50 \pm 8 vs 19 \pm 11) than the SSG group. There were no significant group differences (p < 0.05) between the SET and SSG groups (3.4 \pm 1.7 vs 2.6 \pm 1.9 mmol·l⁻¹) in blood lactate concentrations at baseline, but the second (11.8 \pm 2.8 vs 4.7 \pm 2.0 mmol·l⁻¹) and third (13.7 \pm 3.4 vs 4.8 \pm 2.3 mmol·l⁻¹) intervals showed more than double the amount of lactate in the SET group compared to the SSG group.

Performance effects of intervention

The changes in physical performance between baseline and post intervention can be seen in Figure 8. The distance covered by the SET and the SSG group in Yo-Yo IR2 was 569 \pm 147 and 563 \pm 145 respectively, with no between group differences at baseline (p < 0.05). Both intervention SET and SSG groups increased Yo-Yo IR2 performance (p < 0.05) with 323 \pm 125 and 222 \pm 113 m, respectively and a ~39% longer distance was observed in the SET group (p <0.05). The fatigue index in RST improved in both groups (p < 0.001), and no change nor difference between groups in mean RST time nor AAT was detected.



Figure 8. Relative change from before to after intervention in distance covered in the Yo-Yo intermittent recovery test, level 2 (Yo-Yo IR2), repeated sprint test (RST), RST fatigue index (RST F.I) and Arrowhead Agility Test (AAT) performance in SET (n =21; solid bars) and SSG (n =18; open bars). # denotes significant between-group differences in change score. * denotes significant within-group difference from before to after intervention. Significance level p < 0.05.

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Maximal enzyme activity and protein expression

In Figure 9, changes in maximal activity in the investigated muscle enzyme is shown. There were an elevation in the SET group only in skeletal muscle CS maximal activity (p < 0.05) over the intervention period from 25.5 ± 3.1 to $30.0 \pm 3.1 \,\mu\text{mol}\cdot\text{g}^{-1}\cdot\text{min}^{-1}$ (n = 15), with a larger (p < 0.05) change score in SET compared to SSG (n = 11; Fig. 9). Muscle HAD maximal activity was increased in both groups (p < 0.05) after intervention (SET, 15.3 ± 1.9 to $18.5 \pm 4.0 \,\mu\text{mol}\cdot\text{g}^{-1}\cdot\text{min}^{-1}$; SSG, 15.7 ± 2.8 to $19.5 \pm 3.0 \,\mu\text{mol}\cdot\text{g}^{-1}\cdot\text{min}^{-1}$) with no differences between groups. No changes nor between group differences were observed in muscle PFK maximal activity after intervention.



Figure 9. Relative change from before to after intervention in citrate synthase (CS), 3-Hydroxyacyl-CoA dehydrogenase (HAD), and phosphofructokinase (PFK) maximal enzyme activity determined in muscle tissue from vastus lateralis muscle in SET (n =15; solid bars) and SSG (n =11; open bars). # denotes significant between-group differences in change score. * denotes significant within-group difference from before to after intervention. Significance level *p* <0.05.

In Figure 10, relative changes in the investigated muscle proteins are displayed. There was an up-regulation of protein expression for $\alpha 1 \text{ Na+-K+}$ ATPase subunit in the SET (19 ± 26%; n = 15; p < 0.05) and SSG group (37 ± 41%; n = 12; p < 0.01) where the change in the SSG group tended to be higher than in the SET group (p = 0.07). After intervention values of protein expression for FXYD1 and $\alpha 2$, $\beta 1 \text{ Na+-K+}$ ATPase subunits were not significantly different from baseline in the two groups and no between group differences were

observed. MCT4 protein expression was increased in SET group (n = 15) with $30 \pm 41\%$ (p < 0.05) as well as the SSG group (n = 12) with $61 \pm 49\%$ (p < 0.01) and no between group difference was detected. NEH1 displayed no within or between group differences after intervention while muscle buffer capacity showed a significant within group difference (p < 0.05) in SSG group only but no between group difference.

In markers for substrate levels, GLUT-4 protein expression increased in the SSG group (n = 12) only with 40 \pm 54% (p < 0.05), while protein expression for GS decreased in the SET group (n = 15) only, with 22 \pm 30% (p < 0.05). SOD2 protein expression was elevated in both intervention groups (SET, 28 \pm 32% and SSG, 37 \pm 29%, p < 0.05) while protein expression for SOD1 and CAT where unchanged after intervention.



Figure 10. Relative change from before to after intervention in Na⁺-K⁺ATPase α 1, α 2, β 1 and FXYD1, MCT4 and NHE1 protein expression as well as buffer capacity in muscle tissue from vastus lateralis muscle in SET (n =15; solid bars) and SSG (n =11; open bars). # denotes significant between-group differences in change score. * denotes significant within-group differences from before to after intervention. Significance level *p* <0.05.

Inter-player variation and individual examples

Inter-player variations were large in the physical variables investigated during training in the two intervention groups. The ranges in TD covered during the intervention training were larger in the SSG group (1151-2336 m) compared to

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the SET group (1226-1542 m). Furthermore, the two intervention groups had large ranges in high intensity running (>14 km \cdot h⁻¹) distance covered (SSG, 36-462 vs SET, 405-921 m) and high-speed running (>21 km \cdot h⁻¹) distance covered (SSG, 0-102 vs SET, 125-323 m).

In Figure 11, inter-player variation of changes in Yo-Yo IR2 high is displayed in absolute values for both intervention groups (in m). Ranges in Yo-Yo IR2 improvement were larger in the SET group (200-680 m) than in the SSG group (80-360). No significant correlations were found between physical variables during the training and Yo-Yo IR2 performance. However, the three players who had the greatest performance improvements in Yo-Yo IR2 were found in the SET group and the six players who had the lowest performance improvements belonged to the SSG group.



Figure 11. Absolute change (m) from before to after intervention in Yo-Yo IR2 performance (m) on an individual level. Circle = SSG group (n= 18) triangle = SET group (n=21).

Discussion

This thesis is the first to describe short intense peak periods of top-class male football players in detail using a large sample, analysed with a moving average of raw data. The major findings of Study 1 were that CM and WM covered the greatest distance and CD the least distance in short-term peak periods. Moreover, all playing positions covered less high-intensity distance in the 5-min period following peak 5-min periods and all playing positions except CD covered less distance in the 5-min period following peak 1 and 2-min periods compared to the average 5-min period. In addition, it was shown that substitutes only playing the last 15 min covered greater distance in highintensity than player playing the full game, and substitutes show similar declines in high-intensity distance after short term intense periods. Finally, large individual variations were found in high-intensity running distance in total and in peak intense periods of a top-class football game.

This thesis is also the first to investigate post-game fatigue and recovery kinetics in multiple muscle groups following a simulated football model in competitive football players. The major findings of Study 2 were that fatigue occurs in all investigated muscle groups 0 hours post-game and trunk displays the longest recovery time and knee joint muscle displays the largest decline in muscular performance. A large individual variation in fatigue and recovery kinetics was observed between players.

The thesis also provides novel data on inter-individual physical responses from commonly used small-sided game formats which was compared with fullsized professional game demands in total and in short term peak periods. The principal finding of Study 3 was that physical responses from the small-sided games differ markedly from full sized game demands in total and in peak intense periods.

Finally, this thesis examined performance and muscular adaptations of individual speed endurance training in comparison to small-sided games in competitive football players. The findings displayed a clear difference in training responses where the SET group showed a greater increase in high intensity performance and muscular oxidative enzyme activity, while the SSG group tended to induce a larger increase of ion transport protein expression and glucose transporter as well as a higher level of muscle glycogen capacity.

Physical game demands

In Study 1, we investigated peak intense periods of top-class players in high detail. High-intensity running distance in peak 5-min periods has been described in previous literature in professional football with similar speed-zones, and our result is in line with these results (~230 m) [4, 5, 31]. Moreover, in the classic study by Mohr et al (2003) top-class players from the Italian first league performed a mean of around 220 m in high intensity running distance in their peak 5-min periods [5], which is similar to our findings (235 m) in Study 1 (Table 1). These results indicate that the game demands in peak 5-min periods has not increased in the past decade in top-class football, although the high intensity running distance of the game in total on professional level increased with $\sim 30\%$ during the same time period [17]. However, in the study by Mohr et al (2003) the researchers used manual video based analysis [5] while semi-automated multi-camera analysis was used in the present study. The two methods have been proven to be of good reliability, but absolute values may differ markedly between the two methods, especially within the high intensity category [11]. This makes it hard to draw large conclusions about the evolution of peak period game demands.

The majority of peak intense periods were present early in the first half of the game in the speed categories >14 and 17 km \cdot h⁻¹ (50-59%). These findings and the fact that distance covered >14 km \cdot h⁻¹ in peak 1, 2 and 5-min is greater in the first 15-min than later parts in the game indicates that the initial 15-min may be the most physically demanding period of the game. On the other hand, peak periods in speed categories >21 and >24 km \cdot h⁻¹ were more evenly spread throughout the game. Since distance covered >14 km \cdot h⁻¹ has been shown to be of a higher volume than, for example, sprinting, [6, 32] and may also pick up some of the short intense runs, this speed category may provide a better and more precise measure of the most metabolically demanding game-periods. Distance covered >14 km \cdot h⁻¹ may therefore be a more precise variable than higher speed categories for physiological performance for football players. However, recent studies have found that individualised speed-categories based on maximum aerobic speed and maximum velocity seems to be even more related to individual metabolic demands than general speed-categories [124,

DISCUSSION

125], yet this approach is more problematic with a very large sample of topclass players used in the present study.

Study 1 found a decline in running distance ranging between 6-20% in the 5-min period following peak 1, 2 and 5-min periods compared to average 5-min periods in speed categories >14, 17 and >21 km \cdot h⁻¹. Similar results have been found in several studies investigating peak 5-min periods [4, 5, 16, 31, 32], but Study 1 is the first study to report the same performance deficit in the 5-min period after even shorter peak intense periods of 1 and 2-min. These findings point out that after a short intense period of only 1 min a player's ability to perform optimally may be deteriorated for as long as 5 min thereafter. Thus, it is likely to have a negative effect on the ability to maintain the tactical plan according to individual roles, and may impair high-intensity exercise performance in important runs.

Finally, we observed that substitutes only playing the last 15 min of a game had similar performance decrements after a short intense period as did players playing the entire game, which strengthens the fact that temporary declines in performance may reflect physiologically mediated fatigue.

Total running distance in the game was around 2400, in speed category >14 km·h⁻¹. High intensity running distance (>14 km·h⁻¹) in the present study is slightly greater than what has been found in previous literature from professional football (~2089 m) analysed with the same method [4]. This may be a result of slightly different speed categories (>14 vs >15 km·h⁻¹), but it has been found in some studies that top-class players competing on international level spent relative more time than other professional football players in high intensity running (8.7+0.5 vs 6.6+0.4% of total time) [5]. Distance covered in the last 15-min of most speed categories declined with 7-48% compared to the mean of the first 15-min periods. Furthermore, we also found that substitutes playing the last 15-min of the game cover 18-39% greater in total, in all speed categories except for TD in comparison to full game players. Similar results have been reported in previous research [4, 5, 9] and this supports the fact that running performance is impaired towards the end of a football game.

Playing position and peak periods

The present study found differences between playing positions in high-intensity running in peak periods. Our finding show that CM and WM covered the greatest HIR distance in peak intense periods of 1 and 5-min and CD the least of all positions (Figure 4A, C). This finding is consistent with the literature [4, 5, 7]. In line with our findings one previous study found that WM cover a greater distance in high speed (>19.8 km \cdot h⁻¹) in peak 5-min periods than all other positions on a professional level [16], but the same study found that CM covered less distance in high speed than all other positions except for CD, where no differences were found. The differences for CM in our study of running distance may be related to playing formation and the tactical role of the position, which may have a large effect on running performance during football games [31].

The results of Study 1 demonstrates that all positions had a temporary decline in performance in the 5-min period after peak periods, except CD after peak 1 and 2-min in speed zone >14 km \cdot h⁻¹ (Figure 4A-C). Thus, the tactical limitations of the CD position, may result in less opportunity to engage in high intensity exercise, which may result in lesser physiological taxations during the very short peak period in comparison to the other out-filed positions. Based on these findings, it collectively seems that an intense period of 1 or 2-min can affect running performance up to 5 minutes afterwards in most tactical roles, however, it seems that CD do not experience temporary fatigue after the shortest (1-2 min) peak periods. In fact, CD cover 16-20% and 15-25% less high intensity running distance than all other positions in peak 1 and 2-min periods respectively, which support the statement made above.

Fatigue during a football game

Study 1 shows for the first time that temporary performance decrements occurs after really short intense periods, lasting 1 and 2-min in a football game, and that players can be effected for up to 5-min thereafter. Previous studies have observed that very fast running distance covered during peak 5-min periods have been shown to be associated to Yo-Yo IR2 performance (r = 0.51) [24, 41], which is a test having a large anaerobic component [36]. Furthermore, studies have also found that during intense periods the anaerobic energy pathways is highly taxed and that RST performance is inhibited directly after an intense game period even early in a game [24]. Moreover, a recent study found a large correlation between high intensity distance covered during peak 5-min periods and protein expression of several Na⁺–K⁺ ATPase subunits in the quadriceps muscle [41]. In addition, other muscle proteins, for example NEH1 and FXYD1, also correlated with in game temporary fatigue index (r = -0.53)

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and peak 5-min fast running distance (r = 0.54), respectively. As the Na⁺–K⁺ ATPase pump and other sarcolemma transporters have been shown to contribute to maintaining muscle cell homeostasis during exercise [126]. These findings strongly indicate that the temporary performance decrements can be partly caused by physiological fatigue and may be caused by a high taxation of the anaerobic systems or disturbances in the muscle ion transporters. Thus, the study opens up the possibility for further and detailed studies of the physiological cause of temporary fatigue in competitive football and similar team sports, such has ice hockey, basketball and team handball.

In Study 1, the running distance in various speed categories was decreased in the last 15 minutes of each half and especially in the last 15 minutes of a game. It has also been found in previous studies that neuromuscular and intermittent anaerobic performance declines after a football game [44, 127, 128]. The performance decrements towards the end and after football games indicates development of fatigue caused by prolonged intermittent activity. Fatigue towards the end of a football game has been associated with lowered or depleted glycogen levels in all fibre types [24] and subcellular locations [129], which is likely to affect Ca²⁺ handling and impair high intensity running performance during the last running period of a game.

Inter-player variation

Study 1 clearly demonstrates large individual variations between players in all speed categories in the total game, as well as in peak intensity periods (Figure 5). This is a common finding in previous game analysis research [4, 6, 7]. The inter-player variability in distance covered in peak periods appears to increase gradually when the peak period duration is shorter. For example, the range in peak 1-min distance at speed zone >14 km h⁻¹ between players was 84-159 m, meaning that one player covers only 84 m and another player 159 m of HIR in the most intense 1-min period of the game (Figure 5). This specifies that the game demands of peak periods of the individual player is highly specific. Furthermore, correlation between full game HIR distance covered in peak periods was strong. Peak 1-min distance correlation was weaker ($\mathbf{r} = 0.69$) than peak 5-min distance ($\mathbf{r} = 0.90$), compared to full game high intensity distance. More outliers are present in the peak 1-min correlation and this indicates that the shorter peak periods, the more specific game demands may be for the individual player. Moreover, HIR in the total game may not

always be a precise measure of the demands in the most demanding game sequences. In addition, the intense Yo-Yo IR2 test relates to the peak intensity periods during football games (r = 0.50-0.56), and Yo-Yo IR1 which is of a more aerobic character does not [41]. This indicates the importance of investigating the peak periods in a practical environment, which may differ from the full game demands.

Finally, Figure 5 also reveals that a large variation is observed within the different playing positions in peak periods, which has been shown in a previous study, investigating peak 5-min periods [16]. For example, one CM covering a double amount of peak 1-min game distance more than another CM, in HIR (160 vs 80 m) or one CD covering a triple amount of distance in the same speed zone as another CD (300 vs 100 m) in peak 5-min periods (figure 5). Collectively, these findings highlight that physical demands in peak intense periods is very explicit for the individual players in top-class football. Even though significant differences between positions are clear, differences within positions can be large and specific on an individual level. It would be of importance for coaches to analyse peak intensity periods in detail in order to plan and conduct training regimes to prepare football players for the individual game demands. This may be especially important during high intensity training.

Muscle specific post-game fatigue

The physical activities performed during a football game are characterised as demanding concentric and eccentric muscular work, causing muscle damage and decreased voluntary activation, which can lead to a decrease in physical performance [130]. Study 2 is the first attempt to investigate fatigue and recovery kinetics in multiple muscle groups after a simulated football game. Maximal muscular performance decrements were present in all individual muscle groups 0 hours after CST, with knee flexors displaying the largest decrease (14%) (Figure 6A). The large decrease in knee flexor performance may be a cause of the forceful knee extensions when running and kicking, where the eccentric contraction of the hamstrings at high speeds occur, to counteract anterior shear forces and decelerate the forward movement and internal rotation of the tibia [131]. Former studies have examined fatigue responses in knee flexors and extensors and found similar declines in MVC (~9-15%) directly after a football game [44, 52, 127] and a simulated protocol [55]. In our study a drop in muscle performance in ankle extensors was present 0 hours

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after game (9%), which is in contradiction to two earlier studies showing no decline in MVC performance in the same muscle group [132, 133]. Furthermore, the relatively large decline in muscular performance in lumbar/thoracic extensors (12%), may be a result of the difficulty in isolating this muscle group from hip extensors in MVC testing. With this notion in mind, the decrease in muscle performance may be caused by the hip extensors working eccentrically and concentrically during sprinting and high speed running.

When the muscle groups were pooled together, trunk muscles had not returned to baseline 24 hours after CST (4%), in contradiction to all other muscle groups. Trunk muscles are involved in many of the activities occurring during games, producing force when serving as a stable base for the moving limbs [134]. It seems that these forces can cause fatigue in the trunk muscles lasting more than 24 hours after games. With an exception of the trunk muscles, all individual and pooled muscle groups were back to baseline values 24 hours after CST, and this is in accordance to what Silva et al (2013) found. There are, however, conflicting results in MVC recovery after a football game. Two studies found that MVC performance was affected negatively 48 hours post-game [44, 135] and even recovery times up to 72 hours post-game has been observed [52, 100, 127]. The prolonged recovery times in MVC in some of the studies can be explained by the daily practice conducted by the participants in the studies. For example, Draginidis et al (2015) observed a decrease of MVC performance in a control group only participating in training sessions [127]. In our study, the participating players did not conduct any physical exercise on recovery days, which may explain the relatively fast recovery in muscle performance. Another explanation may be an effect of adaptation due to multiple testing despite the fact that the players were familiarised with the testing procedures on one occasion. With this notion, future studies may consider a higher number of familiarisation sessions in order to minimise the risk of training effect during the recovery period.

Inter-player variation

The inter-player variation was large between players of fatigue and recovery responses in the different muscle groups. For example, one of the players had a decline in core muscles of $\sim 40\%$ 0 hours post-game and it remained below $\sim 30\%$ of baseline values 48 hours post-game (Figure 6C-D). The same player

had a drop in muscle performance in knee joint muscles of ~8% and returned to baseline values at 24 hours post-game. These findings indicate that some players may have extended time for recovery in specific muscle groups, being more sensitive than other muscle groups. Information about recovery profiles in specific muscle groups for the individual player is of importance to plan training and recovery strategies within a competitive team setting. Monitoring individual muscle groups may be especially important in congested periods as some players play several games in a week, which has been linked to a higher risk of injury that may be caused by accumulated fatigue [136].

Simulated football model

The Copenhagen soccer test is proven to give comparable physical and physiological responses to a normal game on elite level [57]. However, in Study 2 the total sprinting distance was markedly lower than in games on elite level (~60 vs 150-250 m) [4, 6, 10]. Furthermore, the number of MIA and MID was also ~50% lower than those found in an elite football game [137]. Possible explanations for the disparate results are related to the GPS technology, including different time resolution (5 vs. 10 Hz), software algorithms and filters used [138]. Nevertheless, future research should aim to include greater sprints distances and higher number of MIA and MID into simulated football models in order to mimic physical and physiological responses even closer to the modern game.

The CST caused a similar response in muscle damage as previously detected in competitive football [46, 139], as plasma CK activity values increased 0 hours and 24 hours after the game, and returned to baseline 48 hours post-game. CK values have been associated with intense activities such as HIR, sprinting and change of directions during football games in earlier studies [140]. Moreover, the fact that the change score for CK activity 24 hours after CST correlated with Yo-Yo IR2 performance, points out the importance of a high intermittent anaerobic capacity to increase the ability to recover after football games. Altogether, these findings strengthen the fact that CK activity in the blood may be used as a marker for muscle damage in football. Finally, the tendency of a correlation between the decline in sprint time during the final sprint during the CST and decline in maximal hamstrings performance, shows that fatigue occurring during football may be related to isometric maximal muscular performance.
Fatigue resistance training in football

Study 1 shows that fatigue is likely to occur after short-term intense periods as well as towards the end in top-class football games. The study also shows a large inter-individual variability in game demands in total and during intense periods. It may therefore be of importance that training strategies are at least partly conducted using an individual approach. Using an individual approach may be a better way to prepare players for individual game demands and ensure optimal physiological adaptations.

Associations between small-sided games and fullsized games

SSGs are a very common training strategy of fitness training in various levels of football as it has been proven to increase physical, technical and tactical abilities [141]. Study 3 is the first to compare important physical parameters in full games as well as intense game periods, with common SSG formats. In Study 3, large individual variations were found in all variables (CV = 6-67%) in the different SSG formats and FSG, which is in accordance to a recently published study on medium sized SSG [142]. The majority of the investigated variables showed no correlations (62%) between the FSG and the SSG formats. As 8v8 SSG correlated with FSG in 50% of the parameters compared to 6v6 (38%) and 4v4 (19%), indicates that 8v8 SSG is the best of these formats for mimicking the individual physical game demands. Furthermore, strong correlations were found in several variables for acceleration and deceleration and the very strong correlations (IDe, r = 0.9) in 8v8 (DECd, r = 0.87; IDe,r = 0.87) in 6v6, indicates that 8v8 and 6v6 SSG may be formats of training that meet the physical demands of decelerations in games for the individual player.

With an exception of TD, none of the variables for distances and efforts in the various speed categories during 4v4 and 6v6 SSG were correlated to the responses during FSG. This indicates that these two formats of SSG may not be player-specific training protocols to prepare players for their physical game demands. For example, HIRd has been shown to be an important variable for aerobic performance towards the end of football games [41] and is linked to performance during intense periods [5, 41] and temporary fatigue [5]. None of the investigated SSG formats displayed significant correlations with FSG in this important variable. However, in the present study we used pitch-sizes that are commonly used by the teams participating in the study. When calculating the area per player in the different formats the 8v8 SSG was much greater (467m² per player) than 6v6 (286m² per player) and 4v4 (240m² per player), which is normal in practical training. It has been shown in previous studies that increasing the area per player increases the intensity of the SSG and may therefore increase the physical response [143]. As the area per player during FSG is ~620 m², the area per player in the 8v8 SSG is closer to the real game and this may explain the higher number of correlations compared to 6v6 and 4v4. However, for tactical and technical reasons, it might be unrealistic to play 4v4 on a similar area per player as 8v8 SSG.

Associations between small-sided games and peak intense periods

Physical performance in peak 1, 2 and 5-min periods correlated with the different SSG formats in 22, 33 and 33% of the investigated variables (Table 3A-C). No correlation was found between 4v4 nor 6v6 SSG and none of the peak periods in the variables for running distance and efforts, except TD. The fact that 4v4 and 6v6 SSG do not correlate with any of the peak period in important markers of HIRd and HSRd, indicates that the training formats investigated in Study 3 may not give the optimal physiological stimulus needed to adapt to the player-specific physical demands of peak intense periods. On the other hand, 8v8 SSG was strongly related to HSRd during the three peak periods investigated (r = 0.67-82). In Figure 7A it can be seen that 8v8 SSG can explain ~55% of the variance of peak 5-min distance in high speed running. This is strong evidence that players with the greatest physical game demands in peak 5-min periods can also get the physical load in 8v8 SSG necessary to meet these demands. On the contrary, inter-individual associations between 4v4 SSG and FSG in HSRd showed no correlation. In figure 6B it can be seen that two players display similar game demands (~500 m), but completely different physical responses in HSRd in 4v4 SSG (5 vs 60 m).

Nevertheless, it is of high importance that football coaches are very careful when deciding which format of SSG and pitch sizes to use in order to target the physical aim of the training session for the individual player. Some players may need additional individual training to target the physical and physiological aim of the session, in order to adapt to their physical game demands.

Physical response of speed endurance and small-sided games

In study 4, we compared physical and muscle physiological adaptations between individual speed endurance productions training with 6v6 SSG. The results of the physical parameter measured during the two training regimes, showed very large difference in HIRd where the SET group performed a four-fold greater distance (SET: ~826 vs SSG: ~180m), despite the SSG group covering more ground in total (SET: ~1364 vs SSG: ~1683m). The SET group also stimulated the glycolytic system to a higher extend than the SSG group. Indeed, blood lactate was ~65% higher during SET than SSG. Moreover, around 40% greater improvement in Yo-Yo IR2 performance was present in the SET group compared to the SSG group after intervention. Similar performance improvements have been found in previous studies investigating the effect of speed endurance production training on physical performance [82, 83, 88, 123]. However, the Yo-Yo IR2 performance on the time of baseline was relatively low with a mean of the two groups of ~566 m, compared to other studies on male university football players (~680m) [82], moderate elite players (~771m) and top-class players ($\sim 1059m$) [36]. This may be explained by the fact that the start of the intervention was in the beginning of the preseason, and the players had been off for 5-6 weeks with very little or even no training. It would be relevant to investigate physical performance effects of the two training methods in full season for players with Yo-Yo IR2 baseline test score values of higher standards.

The SET group exhibited a nearly 40% larger Yo-Yo IR2 performance upgrade on group level than the SSG group. However, the individual results of the performance improvements (Figure 11) reveals that for some individuals 6v6 SSG seems to provide the necessary workload to increase Yo-Yo IR2 performance to a similar extent as SET. The lactate levels seem a lot lower in the SSG group, but as the 7-9 min duration of each set in the 6v6 play is relatively long, individual players may have had high physical intensity and thus high lactate levels for short periods in the beginning of the 7-9 min period, and then the levels of lactate were lower at the end of the period and during the blood sample collection. This may be one explanation for the fact that some player in the SSG group experienced similar Yo-Yo test improvements and physiological adaptations as SET group. On the other hand, it can also be seen in Figure 11 that the three players who had the greatest improvement in Yo-Yo IR2 performance belonged to the SET group and the five players who had the lowest improvements belonged to the SSG group. Collectively, these findings indicate that an individual approach to training in football may be required as a supplement to conventional training approaches such as SSG for some players in order to achieve a higher physical adaptation.

Muscular adaptations to small-sided games and speed endurance

In contrast to the SSG group, the SET group displayed an up-regulation of muscle CS maximal activity, which is a common marker for oxidative capacity in skeletal muscle [144, 145] (Figure 9). Previous studies have found similar results in untrained subjects [144, 146] and well-trained athletes [147, 148]. In addition, an up-regulation in CS has been found when adding SET training to the normal training in endurance trained cyclists [88]. Finally, a recent study found that intermittent high-intensity training was superior in muscle mitochondrial adaptations to continuous moderate training, although both groups performed equal amounts of work and session durations [149]. Collectively, the above results are in line with present findings and indicates that it may be more beneficial for elite football players to conduct SET than SSG in order to improve muscular oxidative capacity, which is highly important to recovering rapidly after intense actions in a game.

In Study 1 it was demonstrated that running performance in high intensity declines towards the end of football games. Previous studies have found depletion of muscle glycogen towards the end of football games [24] and HIRd in the last 15 min has been linked to HAD maximum activity. With this in mind, and the fact that both the SET and SSG groups displayed elevation in HAD to similar extents, indicates that both training regimes provides adequate stimulus to enhance the ability to utilise more fat as a substrate. In addition, the SSG group showed a tendency to larger glycogen storage than the SET group, with the highest muscle adaptations in GLUT-4 activity. These findings indicate that SSG may be a better training regime in order to increase muscle glucose transport capacity and muscle glycogen storage. This is contradictory to previous studies showing an up-regulation in GLUT-4 muscle content [150] and muscle glycogen storage capacity [89]. However, these studies were not investigating trained athletes.

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It was recently found that protein expression for muscle Na^+-K^+ ATPase subunits explained ~50% of the variance in HIRd covered in peak 5-min periods during a football game [41]. This specific muscle marker has been found in previous studies to be important to resist fatigue during high intensity exercise [151]. However, in Study 4, both groups had an up-regulation in α_1 Na⁺-K⁺ ATPase subunits, with a tendency to a larger up-regulation in the SSG group (Figure 10). Therefore, the greater improvement in Yo-Yo IR2 test performance may be unrelated to the increase in Na⁺-K⁺ ATPase subunit protein expression. This finding is contradictory to the findings by Mohr et al (2016), where a strong correlation was found between Yo-Yo IR2 performance and protein expression for Na⁺-K⁺ ATPase subunits [41]. On the other hand, it was recently found that training adaptations in muscle markers related to fatigue resistance can differ between muscle groups [152]. The results from Study 2 clearly demonstrate that multiple muscle groups, for example knee flexors, are fatigued after football specific activity, but in Study 4 we investigated muscle samples taken only from vastus lateralis. Other muscle groups that are highly active during football training may have adapted differently.

In Study 4, no change was found in SOD1 in any of the intervention groups. However, an elevation of SOD2 was detected in both groups. One explanation for these findings may be that SOD1 is located in the cytosol of the muscle fibre while SOD2 is located in the mitochondrial intermembrane space. It may be an interplay between antioxidant reserves and mitochondrial adaptation to training. An association between an up-regulation in SOD2 and oxygen consumption has been found in previous literature [153] and this is partly in line with our findings, displaying an increase in SOD2 protein expression and an elevation in CS enzyme activity in the SET group. However, no change was detected in CS enzyme activity in the SSG group, despite an increase of $\sim 40\%$ in SOD2, which points out that other muscle signalling mechanisms may be important for mitochondrial biogenesis.

Conclusions

The present thesis demonstrates that performance decrements during a football game occur after short intense peak periods lasting 1-5-min. The thesis also shows that temporary performance decrements are present for all outfield positions after peak periods except for central defenders after 1 and 2-min peak periods. Furthermore, a large inter-player variation in short-term peak periods was detected, where peak 1-min periods displayed the largest variation. Collectively these results calls for an individual approach when planning training.

The current thesis revealed that fatigue occurs in multiple muscle groups and that knee flexors displayed the largest decrease in muscle performance, and trunk muscles had the slowest recovery after simulated football games. Fatigue responses also vary largely between individual muscle groups as well as between individual players. A strong inverse correlation was found between training status and degree of muscle damage after simulated football games, which highlights the importance of a high physical capacity in order to recover after football games.

The results of the present thesis found that physical responses during different small-sided game formats did not correlate to a large degree, but that the 8v8 format seems to be the best able to meet the individual game demands in general and in intense peak periods of full-sized football games. As most of the small-sided game formats seem not to be sufficient for meeting individual game demands, some players may need additional training to increase physical performance.

Finally, adding individual speed endurance training to the normal training improves intermittent high intensity exercise performance, to a greater degree than adding 6v6 SSG in competitive football. Furthermore, the added speed endurance training resulted in an up-regulation in muscle oxidative capacity.

Practical implications

Short-term peak intense periods seem to cause temporary fatigue in all playing positions as well as substitutes only taking part in the second half, in top-class football. This indicates that short-term peak periods are really intense and may be the worst case scenario for physical performance during a football game. Therefore, physical training should partly be based on peak intense periods. The large inter-player variations in short-term peak intense periods, points out the importance of an individual approach to physical training in elite football.

Fatigue occurs in multiple muscle groups after a football game and the recovery of these groups seem to be very individual for different players. In a practical football environment, football coaches and sport scientists should monitor several muscle groups in order to get a better understanding of the recovery kinetics of the individual player after a football game.

The majority of the variables investigated in Study 3 showed no relationship between the different small-sided game formats and full-sized games. This indicates that some individual players might not get the physical or physiological stimulus in small-sided games that is required in their full-sized games. Coaches should monitor football players during small-sided games to detect players that may not be physically challenged compared to their full-sized game demands.

Finally, coaches should add speed endurance training into their normal training in order for the players to increase high intensity exercise performance and muscular oxidative capacity.

Future directions

The present thesis has focused on physical demands in elite football and fatigue patterns during football games and how to prepare players to resist fatigue. The results from the present thesis indicate that fatigue occurs towards the end as well as temporarily during games, as the distance covered in high intensity is declined. However, we do not know how the decline in physical performance affects the tactical abilities during games in critical periods. Therefore, future research should consider an integrated approach in order to get a more holistic view and combine the physical and tactical variables of a football game. Furthermore, to get a deeper understanding of the physiological response in and after peak intense periods, it would be preferable to create simulated protocols based on peak period game data and measure physiological response. Moreover, as physical game demands in total and in peak intense periods differ markedly between playing positions, future simulated protocols should be considered to be position specific.

The different small-sided game formats in Study 3 showed large inter-player differences in important physical variables compared to full-sized games. This can be an effect of the lower area per player in the small-sided game formats than in the full-sized game. Future studies should investigate the inter-player relationships between different small-sided games formats with a standardised area per player that is similar to the one in real match-play. Moreover, it would be of interest how inter-player variations during different small-sided games are related to match-play in tactical and physical performance for a more integrated approach. Finally, researchers should consider investigating muscle adaptations in different muscle groups to get a deeper understanding of training adaptations of different regimes.

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