

Research article

Time-of-day effects on EMG parameters during the Wingate test in boys

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Abstract

In boys, muscle power and strength fluctuate with time-of-day with morning nadirs and afternoon maximum values. However, the exact underlying mechanisms of this daily variation are not studied yet. Thus, the purpose of this study was to examine the time-of-day effects on electromyographic (EMG) parameters changes during a Wingate test in boys. Twenty-two boys performed a 30-s Wingate test (measurement of muscle power and fatigue) at 07:00 and 17:00-h on separate days. Surface EMG activity was recorded in the Vastus lateralis, rectus femoris and vastus medialis muscles throughout the test and analyzed over a 5-s span. The root-mean-square (RMS) and mean-power-frequency (MPF) were calculated. Neuromuscular efficiency (NME) was estimated from the ratio of power to RMS. Muscle power (8.22 ± 0.92 vs. 8.75 ± 0.99 W·kg⁻¹ for peak power and 6.96 ± 0.72 vs. 7.31 ± 0.77 W·kg⁻¹ for mean power, $p < 0.001$) and fatigue (30.27 ± 7.98 vs. 34.5 ± 10.15 %, $p < 0.05$) during the Wingate test increased significantly from morning to evening. Likewise, MPF (102.14 ± 18.15 vs. 92.38 ± 12.39 Hz during the first 5-s, $p < 0.001$) and NME (4.78 ± 1.7 vs. 3.88 ± 0.79 W·mV⁻¹ during the first 5-s, $p < 0.001$) were higher in the evening than the morning; but no significant time-of-day effect was noticed for RMS. Taken together, these results suggest that peripheral mechanisms are more likely the cause of the child's diurnal variations of muscle power and fatigue during the Wingate test.

Key words: Diurnal variation, muscle power, muscle fatigue, electromyography, pre-pubertal.

Introduction

Previous studies in adults have reported that performances of short duration present the common characteristic to be better in the afternoon (e.g., between 16:00 and 20:00 hours) than in the morning (e.g., between 06:00 and 10:00 hours) (Chtourou et al., 2011b; 2012a; 2012b; 2012c; 2012d; 2012e; Racinais, 2010). To the best of our knowledge, there appears to be only one study examining the diurnal variation of short-term maximal performances in children (Souissi et al., 2010) in which we found that performances for strength and power (grip strength, Squat-Jump, Five-jump and cycle Wingate tests) improved significantly from morning to afternoon (Souissi et al., 2010). Indeed, during the Wingate test we showed that peak (PP) and mean (MP) powers improved from morning to afternoon with amplitude of 5.5 % and 6.3 % respectively (Souissi et al., 2010). However, the exact

mechanisms responsible for these time-of-day effects are still under discussion (Chtourou and Souissi, 2012; Hayes et al., 2010; Racinais, 2010).

It is generally accepted that the diurnal increase in central body temperature could act as a passive warm-up that may increase nerve conduction velocity, joint suppleness, and muscle strength (for review see Hayes et al., 2010). Therefore, increased core temperature due to circadian patterns would facilitate enhancement of the neuromuscular and metabolic systems (Racinais, 2010). In addition, some authors suggest that the diurnal improvement in short-term maximal performance is not due to a modification in neural drive but rather due to an improvement of the muscle contractile properties at the end of the day (Chtourou et al., 2011b; Guette et al., 2005; Martin et al., 1999). In fact, the maximal voluntary force has been observed to be higher in the evening without modification of the electrical activity (recorded by surface electromyography (EMG) and quantified by root mean square activity (RMS)) of the biceps brachialis (Guette et al., 2005) or the vastus lateralis (Racinais et al., 2005). These findings have been evidenced by a higher mechanical response to an electrical stimulation of the motor nerve (i.e., Pt) in the evening than in the morning (Martin et al., 1999) as well as a higher RMS/force ratio during a maximal voluntary contraction (MVC) in the evening than in the morning (Gauthier et al., 1996; Racinais et al., 2005). However, other studies found a higher EMG activity in the evening and suggest that both central (neural input to the muscles) and peripheral (contractile state of the muscle) mechanisms may be altered across a day (Callard et al., 2000; Castaingts et al., 2004).

In addition, during the Wingate test, performance is also dependent on the ability of the subjects to produce and maintain a high level of force and/or power output throughout the test (Bar-Or, 1987). This ability is limited by "muscle fatigue" which can be defined as the reduction in the force-generating capacity of the neuromuscular system that occurs during sustained activity (Bigland-Ritchie et al., 1983). To date, the only study that has investigated the time-of-day effect on performances during the Wingate test showed that the fatigue index (FI) was higher in the evening than in the morning with amplitude of 10.2 % (Souissi et al., 2010). Although muscle fatigue has been extensively studied in adults, a limited number of studies concerning children's neuromuscular response to fatigue during intense fatiguing contractions are only

available (Armatas et al., 2010). Comparisons between children and adults have shown that children are more fatigue resistant during the Wingate test (Hebestreit et al., 1993). Indeed, Hebestreit et al. (1993) showed that the decrease in short-term muscle power was lower in 10-year-old boys than in 22-year-old men (-9.4 % vs. -41.2 %, respectively). During short repeated sprints cycling, a common finding in studies involving children is that, in contrast with the adults, there is little or no drop in PP or MP (Ratel et al. 2004). The possible mechanisms of this decreased fatigue in children could include factors such as muscle mass, muscle morphology, energy metabolism, and neuromuscular activation (Ratel et al. 2006).

To the authors' knowledge, no previous study has investigated the central (e.g., central nervous command, arousal, motivation) and/or peripheral (contractility, metabolism, and muscle fibres morphology) factors which can explain the time-of-day effects on performances (i.e., muscle power and fatigue) during a complex (i.e., pluri-axial, pluri-articular and requiring several muscular chains based on a free pedal rate) dynamic movement such as the Wingate test. Thus, the aim of this work was to explore the effect of time-of-day on the time course of EMG parameters changes during a Wingate test in boys. To this end, the collection of surface EMG can provide interesting information to quantify the neural drive and muscle fatigue. First, EMG amplitude could be used as an index to quantify the neural drive associated to power output, and thus to identify the putative role of the central mechanism in the diurnal variation on Wingate performance (Hug and Dorel, 2009; Hug, 2011). Also, as previously reviewed by Hug and Dorel (2009) and Hug (2011), the frequency content of the EMG signal could be used to assess muscle fatigue (Hug and Dorel, 2009; Hug, 2011).

Methods

Participants

Twenty two healthy boys (age = 11.02 ± 0.52 years [mean \pm SD], body height = 1.53 ± 0.07 m, body mass = 41.11 ± 7.36 kg), recruited from a public school in the city of Sfax which represents one of the biggest and most extended cities in Tunisia, volunteered to participate in this study. All boys were classified as prepubertal (stage 1) by a pediatrician according to Tanner criteria (Tanner, 1962). The study was conducted in accordance with the Declaration of Helsinki and was approved by the Clinical Research Ethics Committee of the National Centre of Medicine and Science of Sports of Tunis (CNMSS). A written informed consent and assent was obtained from the children's parents, and children respectively.

Experimental design

Participants visited the laboratory on four separate occasions that included two initial familiarisation sessions in order to minimize the learning effect (Chtourou et al., 2012f). The remaining 2 sessions were completed during the course of the subsequent week at two different times of day (i.e., 07:00 and 17:00 hours) in random order on non-consecutive days (i.e., approximately 24 hours separated each test day). Over each test session, subjects com-

pleted a 30 s Wingate test. At the beginning of each test session oral temperature and body mass were measured. Oral temperature was recorded with a calibrated digital clinical thermometer (Omron[®], Paris, France; accuracy: 0.05 °C) inserted sublingually for at least 3 min with the subjects in a seated resting position for at least 15-min. Digital scales were used to determine body mass (Tanita, Tokyo, Japan; precision: 100 g).

Before the morning test session, subjects were instructed to wake up at 06:00 h, as recommended by Bougard et al. (2009). They were fasting and allowed to drink only one glass of water. Before the evening test session, subjects were requested to ingest their last meal at least 5 h before performing the test. All subjects had the same standard isocaloric meal before testing. They were asked to abstain from vigorous exercise activity for 48 h before each session. Throughout the experimental protocol, the mean ambient temperature and relative humidity of the laboratory were kept stable (21.2 ± 1.0 °C and 45.4 ± 7.9 %, respectively).

Wingate test

The Wingate test was conducted on a friction-loaded cycle ergometer (Monark 894^E, Stockholm, Sweden) interfaced with a microcomputer. This test consisted of a 30-s maximal sprint against a constant braking resistance dependent on the subjects' body mass ($0.07 \text{ kg} \cdot \text{kg}^{-1}$ body mass) according to the optimization tables of Bar-Or (1987). The test began from a rolling start, at 60 rpm against minimal resistance (weight basket supported). When a constant pedal rate of 60 rpm was achieved, a countdown of "3-2-1-go!" was given by the experimenter. Then, subjects were instructed to pedal as fast as they could during 30-s. During the test, they were strongly and vigorously exhorted to sprint maximally throughout the 30 s. The Peak power (PP) over 1-s and the Mean power (MP) over the 30-s period were recorded. The percentage of decrease in power or fatigue index (FI), is the difference between the instantaneously 1-s highest and lowest powers divided by the highest power. In addition, as recommended by Lericollais et al. (2009), to study the fatigue phenomenon inherent to the Wingate test, the decline in power output values (i.e., power output over each 5 s span) throughout the 30-s of the exercise, were also calculated (Chtourou et al., 2011a).

EMG measurement and analysis

EMG signals were obtained using differential bipolar surface electrodes (model DE-2.1, Delsys[®] Inc., Boston, USA). Surface electrodes were fixed longitudinally over the muscle belly and parallel to the muscle fibers direction of the vastus lateralis (VL), rectus femoris (RF) and vastus medialis (VM) muscles of the right leg in accordance with the European Recommendations for Surface Electromyography (Hermens et al., 2000). The electrode placements on the VL were at $\approx 2/3$ distance between the anterior superior iliac spine and lateral aspect of the patella. For the VM, the electrodes were placed at $\approx 20\%$ of the distance between the medial gap of the knee joint and the anterior superior iliac spine. A reference electrode was placed on a bony prominence on the patella of the other

leg. The signal was amplified, filtered (band-pass second order Butterworth filter 20–450 Hz, gain = 1000), recorded at 1000 Hz (Bagnoli-4 EMG System, DelSys® Inc., Boston, USA) and stored in a personal computer for subsequent analysis by the EMGworks 3.0 Delsys Analysis software. An elastic bandage was used to prevent cable movement during cycling. To ensure reliable electrode replacement throughout the experiment, electrodes sites were carefully marked with a waterproof felt-tip pen. Prior to electrode placement, the skin was shaved and cleaned with an alcohol-ether-acetone solution.

The EMG data were recorded between the onset and the end of the 30 s. EMG recording was initiated by a digital trigger coincident with the start of the test and data collection stopped by a digital signal at the end of the 30 s. The recorded root mean square (RMS) and mean power frequency (MPF) were averaged over 5 s spans, giving six values for each signal (0-5, 5-10, 10-15, 15-20, 20-25, and 25-30 seconds). The RMS values were normalized to the highest value recorded during the test as recommended by Rana (2006). Neuromuscular efficiency (NME) was estimated from the ratio of power to non-normalized RMS and used as an index of neuromuscular fatigue as previously reported in the literature (Hug and Dorel, 2009). MPF, NME and the normalized RMS values of the VL, RF and VM muscles were summed and averaged together to be more representative of the quadriceps muscle group.

Statistical analyses

Statistical tests were processed using STATISTICA Software (StatSoft, France). Results are reported as the mean \pm SD (standard deviation). A paired Student's t-test was used to investigate differences between morning and evening for the Wingate test (PP, MP and FI) and oral temperature. EMG data (normalized RMS, MPF and NME) and power output over 5 s spans were analyzed using a two-way analysis of variance (ANOVA) with repeated-measures (6 [segments] \times 2 [time-of-day]). When appropriate, significant differences between means were assessed using the Tukey's post-hoc test. Effect sizes were calculated as partial eta-squared η_p^2 to assess the practical significance of our findings. Test-retest reliability was assessed by intra-class correlation coefficients (ICCs) and standard error of measurement (SEM). Statistical significance was established at $p < 0.05$.

Results

Temperature and performances during the Wingate test

The morning and evening data for temperature and Wingate test are displayed in Table 1. Oral temperature was higher in the evening than in the morning ($p < 0.001$) with amplitude of $0.7 \pm 0.4^\circ\text{C}$.

ICC and SEM for PP, MP, and FI showed high reliability (ICC higher than 0.87, 0.83, and 0.81 respectively and absolute SEM lower than $0.4 \text{ W}\cdot\text{kg}^{-1}$, $0.3 \text{ W}\cdot\text{kg}^{-1}$, and 3% respectively). PP, MP, and FI during the Wingate test were significantly lower at 07:00 than 17:00 h ($p < 0.001$ for PP and MP and $p < 0.05$ for FI) [amplitude (peak-to-trough): 5.9 ± 4.9 , 4.5 ± 4.0 % and 8.9 ± 24.0 %, respectively].

Table 1. Time-of-day effects on oral temperature, peak power (PP), mean power (MP) and fatigue index (FI) ($n = 22$). All values are expressed as mean (\pm SD).

	Morning	Evening
Temperature ($^\circ\text{C}$)	36.09 (.32)	36.88 (.30) ***
PP ($\text{W}\cdot\text{kg}^{-1}$)	8.22 (.92)	8.75 (.99) ***
MP ($\text{W}\cdot\text{kg}^{-1}$)	6.96 (.72)	7.31 (.77) ***
FI (%)	30.27 (7.98)	34.5 (10.15) *

* $p < 0.05$; *** $p < 0.001$

Time course of power output and surface EMG changes

Time course changes in power output and in EMG parameters are displayed in Figure 1 and Table 2 respectively.

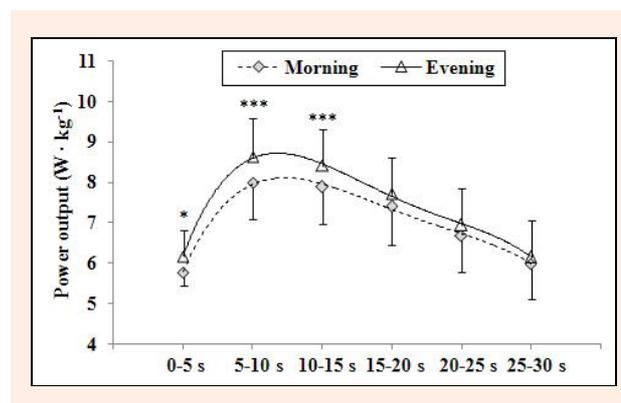


Figure 1. Time-of-day effects on the power output ($n = 22$) over a 5-s span during the Wingate test. Values are expressed as mean (\pm SD). *, ***: indicates statistical significant difference at the levels of $p < 0.05$ and $p < 0.001$ between 07:00 and 17:00 h.

There was a significant main effect of segments for power output ($F_{(5,105)} = 129.9$, $p < 0.001$, $\eta_p^2 = 0.81$), MPF ($F_{(5,105)} = 144.5$, $p < 0.001$, $\eta_p^2 = 0.83$), NME ($F_{(5,105)} = 34.6$, $p < 0.001$, $\eta_p^2 = 0.68$), and RMS ($F_{(5,105)} = 18.9$, $p < 0.001$, $\eta_p^2 = 0.62$), the post-hoc test revealed that MPF and RMS decreased throughout the 30 s of the exercise; however power output and NME increased during the first 15-s of the test and then decreased during the last 15-s. In addition, the time-of-day effect of power output ($F_{(1,21)} = 19.3$, $p < 0.001$, $\eta_p^2 = 0.63$), MPF ($F_{(1,21)} = 7.4$, $p < 0.05$, $\eta_p^2 = 0.6$), and NME ($F_{(1,21)} = 12.7$, $p < 0.01$, $\eta_p^2 = 0.69$) were significant indicating that MPF and NME were higher in the evening than the morning over the Wingate test (Table 2). However, power outputs values were higher in the evening only during the first 15 s of the exercise (Figure 1). The time-of-day effect of RMS was not significant ($F_{(1,21)} = 1.1$, $p > 0.05$, $\eta_p^2 = 0.09$).

Furthermore, a significant interaction of time-of-day and segments was demonstrated only for power output ($F_{(5,105)} = 2.8$, $p < 0.05$, $\eta_p^2 = 0.51$) indicating that the significant diurnal variation observed throughout the first 15 s of the exercise was blunted during the end of the Wingate test (i.e., last 15 s) (Figure 1). However, the time-of-day \times segments interaction for MPF ($F_{(5,105)} = 2.2$, $p > 0.05$, $\eta_p^2 = 0.07$), NME ($F_{(5,105)} = 34.6$, $p < 0.001$, $\eta_p^2 = 0.74$), and RMS ($F_{(5,105)} = 0.6$, $p > 0.05$, $\eta_p^2 = 0.06$) were not significant.

Table 2. Root mean square (RMS), mean power frequency (MPF) and neuromuscular efficiency (NME) over the Wingate test in the morning and the evening (n = 22). All values are expressed as mean (\pm SD).

		0-5 s	5-10 s	10-15 s	15-20 s	20-25 s	25-30 s
Morning	RMS (%)	79.14 (5.42)	83.14 (4.88)	80.9 (5.39)	78.48 (5.18)	74.95 (3.57)	70.93 (9.49)
	MPF (Hz)	92.38 (12.39)	91.79 (12.25)	87.43 (12.03)	84.98 (10.81)	83.53 (10.87)	81.27 (10.87)
	NME ($W \cdot mV^{-1}$)	3.88 (.79)	5.08 (.98)	5.15 (1.04)	5.0 (1.1)	4.72 (1.01)	4.49 (1.04)
Evening	RMS (%)	78.02 (11.55)	80.88 (9.08)	78.62 (8.28)	74.74 (7.17)	72.57 (9.22)	70.83 (12.85)
	MPF (Hz)	102.14 (18.15) ^a	99.49 (17.00) ^a	94.81 (17.37) ^a	92.53 (17.02) ^a	90.72 (17.41) ^a	89.65 (17.54) ^a
	NME ($W \cdot mV^{-1}$)	4.78 (1.7) ^a	6.36 (1.97) ^a	6.31 (1.82) ^a	6.07 (1.85) ^a	5.62 (1.61) ^a	5.22 (1.81) ^a

^a Significant differences between the Wingate test time points at the level of $p < 0.001$.

Discussion

The aim of this study was to determine time-of-day effects on Wingate performances and associated surface EMG parameters in boys. Our results confirm the diurnal variations of PP, MP and FI during the Wingate test with an acrophase in the evening. Results also showed a diurnal variation of time course changes in EMG parameters (i.e., MPF, NME).

Time-of-day effect on performances during the Wingate test

Our data indicates that the child's short-term performances during the Wingate test were better in the evening than in the morning. These results in agreement with our previous results (Souissi et al., 2010; Chtourou et al., 2012a; 2012b) that showed a significant diurnal variation of PP, MP and FI during the same test. In addition, our findings are consistent with those of previous studies on adults (Souissi et al., 2002; 2004; 2007; 2010; Chtourou et al., 2011b; 2012c; 2012d).

Among the factors frequently presented in the literature to explain the diurnal variation of the muscle power, some authors have postulated the hypothesis of a causal link between core temperature and muscular performances fluctuation (Bernard et al., 1998). In agreement, we showed a significant increase of core temperature at the end of the day (i.e., 17:00 h). This increase of temperature might increase muscle glycogenolysis, glycolysis, and high-energy phosphate degradation during exercise and then increase the rate of ATP turnover associated with the exercise and/or changes in the anaerobic/aerobic contribution to ATP resynthesis (Febbraio et al., 1996). Although rectal temperature is usually preferred as a marker of the body clock, the monitoring of rectal temperature in the present study presented problems of social acceptability (Guette et al., 2005). Therefore, for ethical reasons, only the oral temperature method was acceptable to the subjects. Moreover, recent findings suggested that Wingate test performances' daily variations are mainly due to a higher aerobic contribution in energy production (Souissi et al., 2007), faster VO_2 kinetics, and better net efficiency (work performed/energy expended above that at rest) in the evening compared to the morning (Brisswalter et al., 2007). However, the exact mechanisms explaining such time-of-day effect on anaerobic performance in boys are not known yet.

Time-of-day effect on EMG parameters

Our results indicates that RMS values was unaffected by the time-of-day of testing. The absence of a significant

diurnal variation of this parameter suggests that central motor command failure did not play a key role in the diurnal variation observed on muscular power during the Wingate test. These findings are in agreement with previous studies (Gauthier et al., 1996; Guette et al., 2005; Martin et al., 1999; Nicolas et al., 2005) that showed the higher muscle performance in the evening was observed without modification of the electrical activity. Guette et al., (2005) investigated the possibility that central command is modified during the day using twitch interpolation and EMG activities normalized to the M-wave. The authors observed that the RMS/M ratio is not affected by time-of-day. These results indicate that participants' capacity to activate the musculature is not time of-day dependent, and, therefore, central mechanisms cannot explain diurnal variance in short-term maximal performances. These findings have been evidenced by a higher mechanical response to an electrical stimulation of the motor nerve (i.e., Pt) in the evening than in the morning (Martin et al., 1999) as well as a higher RMS/force ratio during maximal voluntary contraction (MVC) in the evening than in the morning (Gauthier et al., 1996; Sedliak et al., 2008). These results suggest that changes in contractile properties of muscle tissue (i.e., peripheral mechanisms) could be the main source of the diurnal variation during short-term anaerobic performances rather than alternation in central nervous command. In contrast, Callard et al. (2000) found higher EMG activities in the evening, and attributed part of the diurnal variation in MVC to changes in the central nervous system and subsequent failure to stimulate the musculature in the morning. However, in this study, the EMG normalization procedure (RMS/M ratio) was not employed.

Our results showed also that muscle fatigue as well as MPF and NME were higher in the evening than in the morning. As indicated in the introduction, the diurnal increase in body temperature could act as a passive warm-up that may increase nerve conduction velocity, joint suppleness, and muscle strength via the enhancement of Ca^{2+} released by the sarcoplasmic reticulum. Therefore, the diurnal changes in core temperature could explain, in part, the changes in MPF. A change in MPF and NME during a fatiguing task is mainly due to a decrease in action potential muscle conduction velocity (Basmajian and De Luca, 1985). Thus, the higher initial MPF and NME values in the evening (0-15 s) may have been the reflect of an increased activation of 'fatigue-sensitive' type II motor units following the onset of sprint cycling. Indeed, a greater decrease of MPF has been often reported during fatigue for subjects or muscles with a higher per-

centage of Type II fibers (Halin et al., 2003). The spectral shift toward lower frequencies during fatigue is principally attributed to muscle fiber conduction velocity slowing, which are induced by the accumulation of metabolic and ionic by-products (Halin et al., 2003; Hug et al., 2006). Therefore, the higher initial MPF in the evening result in a more pronounced slowing down of the muscle fiber conduction velocity (Hunter et al., 2003) compared to the morning.

The power decrease observed during this high-intensity cycling exercise could be mainly attributable to peripheral fatigue (Hug and Dorel, 2009; Hug, 2011; MacIntosh and Shahi, 2011). Indeed, the peripheral fatigue probably involved several sites, but intracellular processes associated with excitation-contraction coupling are one of the sites mostly involved (MacIntosh and Shahi, 2011). In this context, metabolic and ionic changes (e.g., Ca^{2+} released by the sarcoplasmic reticulum, Ca^{2+} sensitivity of the contractile proteins, myosin ATPase activity), that were higher in the evening than the morning (Chtourou et al., 2012a), are mainly responsible for the inhibition of the excitation-contraction processes and thus are at the origin of the developed peripheral fatigue (MacIntosh and Shahi, 2011). These peripheral changes, which occur during the day, are a series of phenomena that could be an explanation of the diurnal variation of muscle power and fatigue observed in this experiment.

Conclusion

In conclusion, the present experiment has shown the existence of a diurnal fluctuation in short-term exercise performances (i.e., muscle power and fatigue) during the Wingate in boys. These time-of-day effects are paralleled with the diurnal variation of the MPF and NME; however, RMS was unchanged. This suggests that the diurnal improvement in muscle power and fatigue is not due to a modification in neural drive but rather due to an improvement of the muscle contractile properties in the evening.

From a practical point of view, sports scientists, clinicians, and coaches should consider the effect of time of day in studies, training programs, and competitive events involving boy's short-term performances.

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Key points

- In boys, performances during the Wingate test fluctuate with the time-of-day.
- MPF and NME are higher in the evening during the Wingate cycling test.
- RMS is unaffected by the time-of-day.
- The evening improvement in muscle power and fatigue is due to an enhancement of the muscle contractile properties.

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