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# Diurnal Variation in Maximal and Submaximal Strength, Power and Neural Activation of Leg Extensors in Men: Multiple Sampling Across Two Consecutive Days

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## Key words

- diurnal rhythms
- neuromuscular performance
- voluntary contractions
- sampling schedule

## Abstract

This study aimed to compare day-to-day repeatability of diurnal variation in strength and power. Thirty-two men were measured at four time points (07:00–08:00, 12:00–13:00, 17:00–18:00, and 20:30–21:30 h) throughout two consecutive days (day 1 and day 2). Power during loaded squat jumps, torque and EMG during maximal (MVC) and submaximal (MVC40) voluntary isometric knee extension contractions were measured. The EMG/torque ratio during MVC and MVC40 was calculated to evaluate neuromuscular efficiency. A significant time-of-day effect with repeatable diurnal patterns was found in power. In MVC, a significant time-of-

day effect was present on day 2, whereas day 1 showed a typical but nonsignificant diurnal pattern. EMG and antagonist co-activation during MVC remained statistically unaltered, whereas neuromuscular efficiency improved from day 1 to day 2. A similar trend was observed in MVC40 neuromuscular efficiency with significant time-of-day and day-to-day effects. Unaltered agonist and antagonist activity during MVC suggests that modification at the muscular level was the primary source for the diurnal variation in peak torque. A learning effect seemed to affect data in MVC40. In conclusion, the second consecutive test day showed typical diurnal variation in both maximum strength and power with no day-to-day effect of cumulative fatigue.

## Introduction

There are numerous biochemical, physiological and behavioral processes in humans oscillating approximately in a 24-hour period (>20 h to <28 h). Rhythms with such a period are generally termed as daily (nycthemeral). When a daily rhythm has proven an endogenous, self-sustaining origin, but it is still susceptible to modulation by 24-hour environmental cycles (e.g., the light-dark cycle, activity-rest cycle), it is called a circadian rhythm [23]. The term diurnal is commonly used to describe circadian or daily variation during the solar part of day.

Diurnal variation in neuromuscular performance with morning nadirs and afternoon maximum values is a common finding in various tests of the maximal voluntary strength of different muscles or muscle groups in dynamic or isometric conditions [5,9,17,33,40]. However, the exact mechanisms responsible for such an oscillation are poorly understood. When based on the surface EMG data during maximal voluntary and/or electrically induced contractions, Gauthier et al.

[15] and Castaingts et al. [4] reported that both central (neural input to the muscles) and peripheral (contractile state of the muscle) mechanisms may alter across a day. On the contrary, Martin et al. [33], Guette et al. [21] and Nicolas et al. [35] suggested agonist muscle tissue being the origin of diurnal changes in peak torque. A similar conclusion was made by Giacomoni et al. [18] reporting, however, a significant diurnal variation only when electrical twitches were superimposed during MVC in order to offset motivational confounds. They proposed that motivational component could have a masking effect on the diurnal variation in voluntary maximum strength of knee extensors.

Correspondingly to the contradictory views on the mechanisms, even the nature of diurnal variation in muscle strength itself has not been satisfactory resolved yet. Drust et al. [10] suggested “at least some input of endogenous mechanism”. Youngstedt and O'Connor [41] called for methodologically more sound experiments to be able to clearly distinguish between possible endoge-

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## Bibliography

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nous and exogenous factors responsible for its diurnal variation. Traditionally, there are two main laboratory based protocols used to assess whether a daily (diurnal) variation in a selected variable has a high endogenous component – constant routine (CR, [6]) and a forced desynchrony protocol (FD, [11]). However, both protocols have some drawbacks when used in physical performance testing. A CR requires usually from 24 to 40 hours of individual isolation in constant light and temperature conditions and, moreover, in recumbent posture which dramatically restricts the variety of performance tests. A FD protocol spreads well beyond several consecutive days of stay in a laboratory per subject. If a condition of a minimum 8-hour rest period between each of the test exertions must be fulfilled to remove any learning/fatigue effects [10], the FD protocol would require a big time commitment from a tested individual. A third laboratory based ultra short sleep/wake protocol has been recently used to show circadian rhythm in swim performance by Kline et al. [31]. Subjects were kept in the laboratory for 50–55 h under ultra short sleep/wake schedule, swim tests were separated by 9 h. In chronobiology of physical performance, a fourth protocol termed as “cyclic Latin square” is commonly used [14]. In this protocol, subjects perform the first session at a different time of day in a counter-balanced order. The consecutive tests are separated by at least eight hours due to the above stated reasons [10] and subjects are not usually required to stay in a laboratory between the tests. Thus a data representing one diurnal cycle are collected across several days and might be actually a mixture of both diurnal and interday [19] variability. In light of the above, multiple sampling across a day could partially discount from the time commitment (a FD protocol) and possible interday effect on diurnal fluctuation in maximum strength (CR, ultra short sleep/wake protocol). Despite a short duration of a single bout in the majority of maximum strength and power tests, great caution must be paid to possible effects of cumulative fatigue and/or other confounding factors. They could mask a true diurnal variation if such physical tests are performed repeatedly with rest periods of several hours only. However, this topic has not been satisfactorily covered by scientific research. It has been shown that diurnal rhythm in muscle torque can be observed by multiple testing within 24 hours [3,15]. Gauthier et al. [16] measured six times per diurnal part of one day with 3-hour rest intervals in between in subjects persisting the civil day time clues. They found no significant difference in the characteristics of the diurnal rhythm when compared to another design extending over 6 subsequent days with only one test session per day. However, it remains unknown whether repeating of multiple sampling per day across several consecutive days would alter diurnal patterns of neuromuscular performance rhythms due to cumulative fatigue, learning or other confounding factors. Therefore, the present study aimed to compare day-to-day repeatability of diurnal variation in maximal and submaximal strength, power and neural activity of knee extensor muscles when measured repeatedly across two consecutive days in civil day conditions.

## Materials and Methods

### Subjects

Thirty-two volunteers (32 ± 7 years, 82.6 ± 9.7 kg, 1.81 ± 0.05 m) with a similar background of health status and physical condition were recruited to the study. They had no experience in

strength training and no regular physical activity more than once a week during the past 3 years. Prior to the experiment, the subjects underwent 10 weeks of strength training. They were randomly assigned into two training groups – weight-stack device group and pneumatic device group. Both training groups performed an identical training program (in terms of training intensity, frequency and volume) carried out, on average, 2.2 times per week from 17:00 to 19:00 h. The training times were selected to closely mimic the time window of the diurnal peak occurrence, so the typical diurnal fluctuation, assumed to be present in our subjects, would not be disrupted in the case of possible time-specific adaptation [38]. The only training difference was in the device used for the development of explosive and maximum strength of lower extremities. Since there were no statistically significant differences between the two training groups after the intervention in two tests used during the experiment (isometric knee extension and loaded jump with 60% of one repetition maximum), both groups were merged together for the purpose of this report. In addition, the subjects were familiarized with the test procedures (except submaximal MVC) eleven weeks and one week prior to the actual experiment.

Background information regarding physical activity, motivational characteristics and medical history was collected by questionnaires. All subjects were considered healthy and had no medical contraindications that would affect the results of this study. No shift workers were accepted. The circadian type questionnaire [13] was applied to estimate morningness vs. eveningness, ability to overcome drowsiness and flexibility of sleeping habits. Additionally, self-reported stress load, length of sleep, daily activity and food intake information were collected throughout the two testing days and the day before. This study was approved by the Ethics Committee of the Central Hospital of Central Finland. An informed consent form was signed by subjects prior to the investigation.

### Experimental design

The subjects repeatedly performed the same set of strength measurements at four different time points across two consecutive days (day 1 & day 2), starting the first session always in the morning on day 1. The time points were as follows: morning (M) 07:00–08:00 h, noon (N) 12:00–13:00 h, afternoon (A) 17:00–18:00 h, and evening (E) 20:30–21:30 h. Each test session lasted 30 minutes per subject and consisted of 15 minutes of resting quietly in a supine position (oral temperature sampling) followed by approximately 15 minutes including warm-up and the actual test procedure. The test consisted of the recording of mechanical power output during the concentric phase of the squat jumps with the extra load of 60% of subject's repetition maximum (1RM) – (SJ60), maximal (MVC) and submaximal (MVC40) isometric knee extension force of the right leg at the knee angle of 120°. A three-minute rest period was allowed between the tests.

The subjects were scheduled to report at the laboratory in fixed 15-minute intervals (first one at 06:45 h, second at 07:00 h etc.) so four subjects could be measured within an hour. The ambient temperatures in the laboratory ranged from 20 to 22°C and the artificial light condition was constant across a day. The subjects were allowed to leave from the laboratory between the test sessions and requested to closely adhere to their habitual daily routine. However, several adjustments were applied to assure better control over important factors such as requesting the subjects to

refrain from alcohol, sexual and strenuous physical activity one day prior to and throughout the two test days.

#### Caffeine

We encouraged the subjects to avoid caffeine in any form, as well. However, strong habitual coffee consumers detected by the questionnaire were asked not to completely refrain but rather minimize their daily dose of caffeine drinks and not to ingest them within 3 hours prior to a measurement session. The reasoning for this was to avoid the “caffeine withdrawal syndrome” which may have a negative impact on vigilance and physical performance during the testing [32].

#### Diet

The diet during the two testing days was controlled and was identical for all subjects, designed according to the Finnish Nutrition Recommendations [34]. The overall daily energy intake goal was set at 10.5 MJ (2500 kcal) per day. All meals (breakfast, lunch, dinner and light supper) were served within 10 minutes after every measurement session was completed. The amount of food was identical for all subjects; however, they were allowed to consume more or less of basic foods (e.g., bread, pasta, rice and potatoes) and milk or fruit juice, within a limited range, to match their individual dietary needs. Water and non-caffeine beverages could be consumed ad libitum at meals and between meals. Since the subjects were after an overnight fast, approx. 250 ml of orange or apple juice was ingested 5 min before all morning test sessions to decrease the possible risk of relative or acute hypoglycemia during the measurements.

#### Sleep

The subjects spent the nights before and during the experiment at their homes. They were instructed to adhere as closely as possible to their usual length and time of sleep and not to switch the light on if awake during the night. This was done to prevent any disturbance in melatonin secretion [7] and its possible effect on selected variables measured during the following day.

#### Stress load

The subjects were asked to make a rating of perceived stress load during both testing days. On a scale from 1 (a stressless day) to 6 (maximum stress), levels 4 (rather heavy stress), 5 (very heavy stressful day), and 6 (unbearable stress) were considered harmful to the parameters collected.

#### Oral temperature

Sublingual oral temperature (temp) was collected by digital thermometer (Microlife, Heerbrugg, Switzerland) after 15 minutes of resting in a supine position to reduce the effect of prior activity. Two trials were required; if the values differed by more than 0.1 °C, additional trials were taken until two successive values with the difference of 0.1 °C or less were obtained. The highest value of all trials within a session was taken for further statistical analysis.

#### Warm-up

Ten technically performed squats to the 90° knee angle with the 25 kg load, one-minute stretching exercise for knee extensors, flexors and back extensor and two repetitions of unloaded squat and countermovement jumps served as a standardized warm-up for all participants.

#### SJ60

Maximal dynamic explosive strength of the lower limbs was measured using the loaded barbell squat jump with 60% of 1 repetition maximum (1 RM). 1-RM values were obtained 3 weeks prior to the experiment. The protocol consisted of three trials separated by a one-minute resting period. A barbell with weights, inbuilt in a Smith machine, was held on the shoulders. A sliding sensor arm based on infrared optics (ErgoPower, Ergo-test Technology A.S., Langesund, Norway) was placed on the end of the bar and mean power during the concentric phase of the jump was calculated instantly. The subjects started the trial from the standing position, squatting slowly down to the 90° knee angle which was announced by a sound signal. After reaching a stable squat position with no signs of obvious movement in any part of subject's body, an assistant gave a loud verbal command to jump. The subjects were requested to move the load as fast and as high as possible. A trial with the highest mean power output in the concentric phase of movement performed with the correct technique was taken for further analyses. In this variable, only data of sixteen subjects could be utilized for statistical analyses due to technical error affecting data of the second half of the participants.

#### MVC

Maximum voluntary isometric force of the knee extensors was tested using unilateral knee extensions of the right leg at 120° knee angle (180° knee fully flexed). The right leg was selected for the measurements exclusively in order to provide identical testing conditions. Laterality of the lower limb was not expected to have a significant impact on diurnal patterns of their performance, as has been recently showed by Guette et al. [21]. The subjects were secured to a sitting position in a knee extension device (Leg Ext/Curl Research, Hur Oy, Kokkola, Finland). The torso was fixed with two horizontal safety belts in a chest and waist area, the upper extremities were placed next to the body holding handgrips. Both thighs were strapped in a position with the cushioned straps placed about the knee joint. Interindividually similar positions of body segments were ensured by the adjustable back support and lever arm. A force transducer was attached to the lever arm and placed at the ankle level. The subjects were asked to produce maximal force rapidly and maintain it for three seconds. The force produced on the lever arm was amplified and the analog signal was collected by a biomonitor ME6000T8 (MEGA Electronics Ltd, Kuopio, Finland) and telemetrically transmitted and stored in the MegaWin software (MEGA). The sampling frequency was 1000 Hz. Two to three trials (if the second trial was more than 5% higher than the first one) were performed with a rest period of 1 minute. A trial with the highest peak force was saved for further analyses.

#### MVC40

After obtaining MVC, submaximal trials were measured. The subjects were asked to produce force of 40% from the MVC and maintain it as accurately as possible for 5 seconds. An instant visual feedback of produced and required force level was displayed on a screen placed in front of them. Three trials were performed separated by 5-second rest intervals in between. The data were recorded and processed identically as in MVC. The average torque and EMG activity were calculated from the second and third trial, the first trial was considered as preparatory.



**Table 1** Diurnal variation in resting oral temperature and performance characteristics during MVC and loaded squat jumps – absolute values

Time of day (h)	Variables (mean ± SD)		Power output SJ60 (W, n = 16)		Oral temperature (°C, n = 32)	
	Peak torque MVC (Nm, n = 32)		Day 1		Day 2	
	Day 1	Day 2	Day 1	Day 2	Day 1	Day 2
7:00	264.9 ± 41.9	257.4 ± 43.2	637.7 ± 124.2	649.4 ± 128.4	36.15 ± 0.3 <sup>§</sup>	36.05 ± 0.3 <sup>*</sup>
12:00	270.9 ± 40.3	276.8 ± 44.0	673.7 ± 145.7	681.3 ± 126.2	36.33 ± 0.3 <sup>§</sup>	36.34 ± 0.4 <sup>§</sup>
17:00	271.3 ± 45.6	279.9 ± 46.2	667.1 ± 132.9	674.5 ± 135.0	36.33 ± 0.3 <sup>§</sup>	36.43 ± 0.3 <sup>§</sup>
20:30	271.1 ± 41.9	274.0 ± 47.8	668.2 ± 120.5	676.9 ± 115.4	36.49 ± 0.2 <sup>*</sup>	36.56 ± 0.3

For statistics of MVC and loaded squat jump variables see the text; \*  $p < .001$ , morning temperature significantly lower than the rest of the day; <sup>§</sup>  $p < .01$ , temperature significantly lower in the morning than at noon and in the afternoon; <sup>§</sup>  $p < .05$ , temperature significantly lower at noon than in the evening; <sup>§</sup>  $p < .05$ , temperature significantly lower in the afternoon than in the evening; \*  $p < .001$ , temperature significantly higher in the evening than in the morning

## EMG

The surface bipolar electromyogram (EMG) was recorded during MVC and MVC40 actions from vastus lateralis (VL), rectus femoris (RF), vastus medialis (VM), and biceps femoris (BF) of the right leg, respectively. Silver/silver chloride pre-gelled electrode pairs and reference electrodes (Blue Sensor M, AMBU, Ballerup, Denmark) were placed according to the SENIAM recommendations [25]. The location of the electrodes was marked prior to the first test session with underskin ink dots [24] to ensure reliable positioning over the sessions. In addition, the same pairs of electrodes were kept attached on the skin during the entire test day and removed only after the E measurements sessions on both day 1 and day 2. EMG signal was recorded and processed in the identical way as the MVC force mentioned above.

## Data analysis

In MVC and MVC40, the force signal and EMG activity were analyzed by SPIKE2 software (Cambridge Electronic Design Ltd, Cambridge, England). The force signal was low-pass filtered and peak torque (MVC) and submaximal torque (MVC40) were calculated. Raw EMG signal was rectified, band-pass filtered (10–500 Hz) and expressed as root mean square (RMS) from the ± 100 ms time window around the peak torque and from 1-second period of the most constant torque signal during MVC and MVC40, respectively. Subsequently, RMS of VL, RF and VM during MVC were averaged in order to provide overall activity of surface knee extensor muscles (KE RMS – [21]). Antagonist muscle co-activation was obtained by the RMS of BF when it acted as an antagonist (MVC & MVC40) divided by the RMS of BF during isometric unilateral knee flexion at the knee angle 120° (MVC flex). MVC flex had been measured 14–10 days prior to the testing. Peak torque, power output and KE RMS of MVC were normalized with respect to the highest value found in each individual and expressed as percentage of this maximum value. The neuromuscular efficiency (NME<sub>max</sub> and NME<sub>submax</sub>) was calculated as RMS peak torque ratio from the normalized and absolute values in MVC and MVC40, respectively.

## Statistical analyses

Standard descriptive statistics (mean ± SD) were conducted on the physiological and neuromuscular performance variables. To evaluate diurnal variation in sublingual temperature, SJ60, MVC, EMG activity, and NME across two consecutive days, 2-factor (time of day – TOD, and DAY) general linear model (GLM) with repeated measures was performed across the eight selected time points. When repeated measures GLMs revealed significant F-ratios, pairwise comparisons with Ryan-Holm-Bonferroni adjustment were utilized to localize significant differences [1]. In addition,

hierarchical clustering was applied in trying to identify possible chronotype, sleep deprivation, or high stress subgroups in a circadian pattern of selected variables. Statistical significance was set at  $p < .05$ .

## Results



### Background variables

#### Chronotype

Twenty-one subjects scored as a neutral type in all three factors examined by the circadian type questionnaire [13]. Five subjects were identified as morning types, while four as evening types. Three subjects had high ability to overcome drowsiness, one subject scored low on this factor. Six subjects belonged to rigid sleeping types. Three subjects scored as extreme type in two factors (2 were evening types with rigid sleeping habits, 1 scored as a type with high ability to overcome drowsiness and with rigid sleeping habits), and one subject in all three factors (morning type with high ability to overcome drowsiness and rigid sleeping habits). There was no significant difference between extreme types and neutral types in diurnal pattern characteristics of any variable measured.

#### Sleep

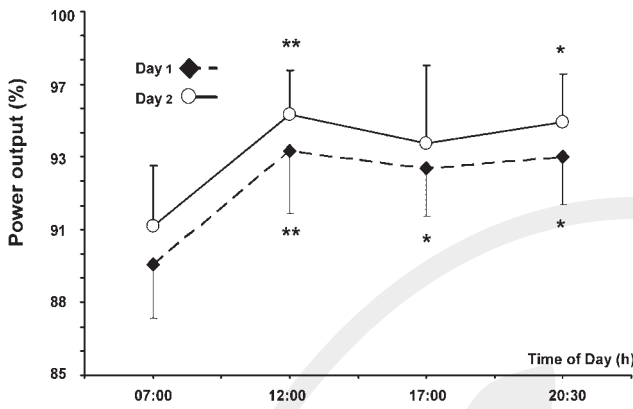
Information on sleep collected one night prior to and during the experiment showed that an average habitual length of sleep was 7.5 ± 0.9 h. The average sleeping time during the night prior to day 1 and the night between day 1 and day 2 was 6.8 ± 0.8 h, and 6.9 ± 0.7 h, respectively. There were five subjects who reported their sleep length prior to or during the experiment to be two or more hours shorter or longer compared to their individual average sleep length. These participants did not differ from the rest of the group in means of oral temperature nor the performance diurnal patterns.

#### Self-reported stress load

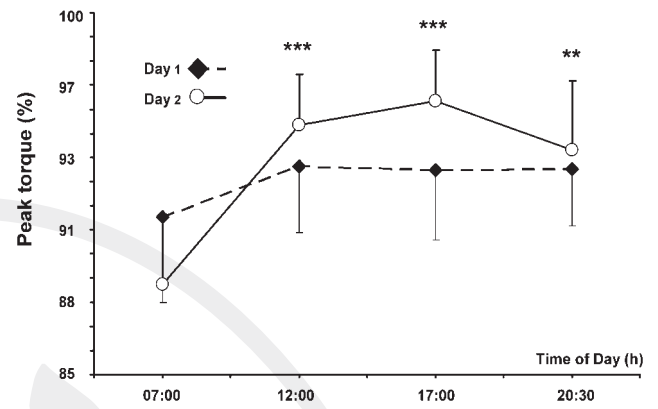
None of the subjects reported a stress level 5 or 6. Three subjects reported a stress level of 4 either on the first or second day. However, these three subjects were not identified as a significant subgroup in their variation of oral temperature and neuromuscular performance by hierarchical clustering and so did not differ from the rest of the group.

#### Oral temperature

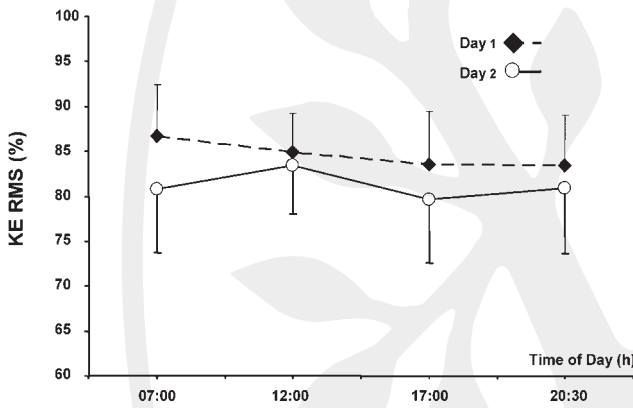
This variable exhibited a significant diurnal pattern with the lowest values observed at the 07:00 h session and the peaks were located at 20:30 h (TOD  $p < .001$ ). No effect of DAY was present,



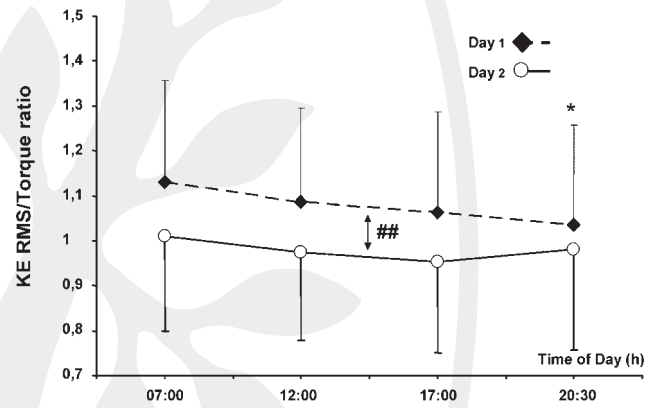
**Fig. 1** Diurnal variation in power output during the concentric phase of loaded squat jump with 60% of one repetition maximum (mean and SD) measured across two consecutive days (n = 16). Mean values expressed as a percentage of the maximum power output of that of each subject obtained during the two days, SD presented as half of the range. Statistical significance of pairwise comparisons is shown for both days separately. \* p < .05, \*\* p < .01, relative mean power output values significantly higher than at 07:00 h.



**Fig. 2** Diurnal variation in MVC peak torque of knee extensor muscles (mean and SD) measured across two consecutive days (n = 32). Mean values expressed as a percentage of the maximum torque of each subject obtained during the two days, SD presented as half of the range. Statistical significance of pair-wise comparisons is shown for both days separately. \*\* p < .01, \*\*\* p < .001, relative mean peak torque values significantly higher than at 07:00 h.



**Fig. 3** Relative EMG of knee extensors during MVC (mean and SD) measured across two consecutive days (n = 32). Mean values expressed in percentage of the maximum torque of each subject obtained during the two days, SD presented as half of the range. KE RMS, relative EMG activity calculated from the averaged root mean square values of RF, VL, and VM muscles.



**Fig. 4** EMG/torque ratio during submaximal unilateral isometric knee extensions at the level of 40% MVC measured across two consecutive days (n = 32). KE RMS – absolute EMG activity calculated from the averaged root mean square values of RF, VL, and VM muscles. SD presented as half of the range. ## p < .01, significant change in EMG/torque ratio from day 1 to day 2. \* p < .05, EMG/torque ratio significantly higher in the morning than in the evening on day 1.

although interaction of TOD and DAY were significant (p < .01) (Table 1). This was due to not significantly higher M temperature on day 1 as compared to M on day 2 and reversely lower A and E day 1 values in comparison to the same time points on day 2.

**Physical performance**

**SJ60**

A significant TOD main effect in mechanical power output was found in 2-factor GLM (TOD, p < .01) (Fig. 1). No significant main effect of DAY or interaction of TOD and DAY were present in this variable.

**MVC**

When examined by 2-factor GLM, the peak torque displayed a significant main effect of TOD factor (p < .001) and interaction of TOD and DAY (p < .01). Post hoc comparisons revealed that significant diurnal variation was present on day 2 only with M

values significantly lower when compared to the rest of day (p < .001) (Fig. 2).

EMG activity of the knee extensor during MVC expressed as KE RMS did not significantly alter between time points (Fig. 3). Day 1 mean KE RMS did not significantly differ from day 2 (DAY main effect p = .089). No significant main effects or interactions were found in BF co-activation.

TOD main effect was not significant in KE RMS<sub>norm</sub>/peak torque<sub>norm</sub> ratio during MVC (NME<sub>max</sub>). On the contrary, significant main effect of DAY was present (p < .05), with KE RMS<sub>norm</sub>/peak torque<sub>norm</sub> ratio being lower (NME<sub>max</sub> thus being higher) on day 2.

**MVC40**

KE RMS<sub>absol</sub>/MVC40 torque<sub>absol</sub> ratio values significantly decreased from day 1 to day 2 (DAY factor, p < .01) and there was a significant main effect of TOD factor present in neuromuscular efficiency during MVC40 (NME<sub>submax</sub> p < .05) (Fig. 4). BF co-ac-

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tivation did not alter in time course with no significant main effects or interactions.

Hierarchical clustering did not reveal any significant subgroups in a diurnal pattern of selected variables.

## Discussion

The main finding of this study was that diurnal variation in maximum strength and power was still evident in a protocol using repeated testing across two consecutive days. The diurnal fluctuations on the second day were comparable to the previous results obtained with different designs which controlled for cumulative fatigue and/or learning. The day-to-day repeatability of diurnal patterns was preserved in maximum power output. In MVC, however, diurnal variation during the first day was non-significant presumably due to imbalanced test design with day 1 morning being the first session for all subjects. Learning effect seemed to be present in submaximal strength actions to which the subjects were not accustomed during the preceding conditioning training and familiarization procedure.

In the present study, none of the possible intervening factors – length of sleep and its timing, self-reported stress level and chronotype of the subjects – had a significant effect on the data. Effect of nutritional status was not examined statistically. According to Youngstedt and O'Connor [41], the difference in timing of meal intake prior to tests may be one of the confounding factors affecting diurnal variation in neuromuscular performance. In the present study, the morning tests were taken after a 10-hour fast as compared to a 4-hour fast prior to the rest of tests. A glass of orange juice was served just before the 07:00 h warm-up to decrease the possible risk of hypoglycemia during the measurements. However, differences in nutritional status cannot be fully excluded as a potential confound in the present report. Sleep timing prior to measurements and extremes of chronotypes are two other intervening factors listed by Youngstedt and O'Connor [41]. Duration of sleep was, on average, 42 minutes shorter compared to the usual sleep length of the participants. Pilcher and Huffcutt [36] concluded that only less than 5 hours of sleep in a 24-h period can cause partial sleep deprivation and significantly decrease motor performance. In our study, none of the participants reported a sleep period shorter than 5 h prior to day 1 or day 2. Four subjects slept less than 6 hours before the day 1. However, their characteristics of diurnal variation in performance did not differ between day 1 and day 2 or the rest of the group. In addition, all subjects were required to wake up between 60 and 90 minutes before arriving to the lab. That should be sufficient time to prevent sleep inertia as its duration rarely goes beyond 30 minutes in non-sleep deprived individuals [39]. The opposite chronotypes of our subjects did not significantly differ from each other or from the majority of the neutral types in diurnal variation of the performance and EMG variables (tested separately for the three main factors of the circadian type questionnaire) [13]. However, Kerkhof and Dongen [30] reported that the difference of the circadian phases of body temperature rhythm between morning-types and evening-types of subjects is 2.1 h when measured in a constant routine protocol. Based on the assumption that there exists a parallelism between body temperature rhythm and diurnal variation in muscle strength (for review see [10]), we could expect relatively small differences in timing of the diurnal nadirs and peaks in performance indices. It is possible that the present study could not localize such a lit-

tle phase difference between chronotypes due to infrequent sampling and unequal distribution of the extreme chronotypes. Therefore, only an experiment with sufficient sampling frequency, equal amount of subjects in opposite extreme types, and controlled for masking effect of possible confounding factors (e.g., sleep) could properly address this issue.

Considering the above stated limitations, oral temperature was not statistically influenced by chronotype. It exhibited significant biorhythmic fluctuations under resting conditions on both days. We observed higher average morning temperatures at 07:00 h (36.2°C and 36.1°C on day 1 and 2, respectively) than usually reported for oral temperature – 35.6 or 35.8°C measured at 06:00 h in Gauthier et al. [15] and Nicolas et al. [35] respectively. A physiological increase from 06:00 h to 07:00 h due to normal circadian variation in body temperature could explain some but not all the difference (> 0.3°C). It resulted in approximately 50% smaller diurnal raise in temperature (0.34°C) compared to the values showed in the above-mentioned studies or some others [26,37]. It seems that effect of the measurement design could slightly alter diurnal rhythms of oral temperature, especially on day 1. We hypothesize that higher psychological arousal and/or motivation just before and during the first testing session (M day 1) might partly explain higher morning peak torque and EMG activity on day 1 as compared to day 2. However, a similar feature was not observed in SJ60 power output. The load percentage (60% of 1 RM) and the testing protocol in the SJ60 was based on previous studies [27,28] who found that power output in loaded squat jumps is maximized with loads of 60–70% 1 RM in the subject groups similar to ours. In the present study, the power output exhibited significant and comparable characteristics of diurnal variation (TOD  $p > .01$ ) on both days. There was no significant carry-over effect of cumulative fatigue from day 1 to day 2. The subjects performed, on average, 1.3% higher power output on day 2 at every session. Performance was significantly improved, on average, by 5% and 4.4% from morning as compared to the rest of the both day 1 and day 2. Reilly and Down [37] showed similar diurnal increase in explosive dynamic strength of the lower limbs. They reported an improvement of 5.1% in relative power output during the stair run and 6% in the standing broad jump length. It must be noted that the loaded squat action was part of the 10-week conditioning training in a half of the participants (correct data of eight subjects statistically analyzed). The other half (another eight participants) trained with a pneumatic leg press device (Hur Oy, Kokkola, Finland) instead but undertook twelve maximum familiarization trials eleven weeks (6 trials) and one week (6 trials) prior to the actual experiment. This difference did not cause a significant impact as tested by implementing group between-subject factor to 2-factor GLM. However, inspection examination of the plotted data showed somewhat (n.s.) lower average minimum-to-maximum differences (4.3% versus 8.2%) and a slight (n.s.) day-to-day difference in the diurnal pattern between the A and E sessions in subjects who trained with the pneumatic device. It implies possible influence of different specific skill level (technique) between these two groups. We hypothesize that improved technique may notably decrease, if not remove, its masking effects on the diurnal variation in power output. That would suggest that in unaccustomed individuals, technically demanding multi-joint dynamic actions may require short-term task-specific conditioning training in order to fully adapt to the test demands.

Interestingly, peak torque in MVC did not display as high repeatable diurnal pattern as power output in SJ60. Significant interac-



tions were found between day 1 and day 2. Day 2 data were in accordance with the previous studies [15,17,21] showing lower morning values and significantly improved performance during the rest of the day. The minimum-to-maximum difference between M and A was 8.7%. On the contrary, time of day effect was not present on day 1. The minimum-to-maximum MVC difference was rather low (2.4%) with significantly higher M values and significantly lower A performance as compared to the same time points on day 2. It is rather difficult to interpret these day-to-day differences. Partial explanation may stem from different (higher) psychological arousal and/or motivation just before and during the first morning test session as compared to the rest of experiment. Giacomoni et al. [18] and Bambaiechi et al. [2] reported that motivation could have a masking effect on diurnal variation in voluntary MVC in men and women, respectively. They found typical diurnal variation only when superimposed electrical twitches were added upon the MVC peak torque in order to offset motivational component. Counterbalanced order of the first session might diminish this drawback of the present study [10]. On the other hand, it would prolong recording from two to three subsequent days for most of the subjects. Since the aim was to gather data representing one diurnal period exclusively from a single day, morning day 1 was selected as a starting session for all participants. Thus, we were able to follow and compare real day-to-day differences.

Distinct from the SJ60 action, specific training status and familiarization process could not explain the above-mentioned day-to-day variation in MVC. Subjects were well-familiarized since they underwent from 6 to 7 maximum trials on three separate occasions (13, 11, and 1 week prior to the experiment). In addition, another 8 to 9 MVC knee extensions were performed at different knee angles (90° and 150°) at the same familiarization sessions. As possible confounding factors in our study, time-specificity to training and a cumulative fatigue effect must be taken into account. Regarding the first factor, Souissi et al. [38] reported that adaptation to strength training is greater at the time of day at which training was conducted than at other times. In the present study, all subjects trained between 17:00 and 19:00 h to prevent phase shifts in timing of the typical diurnal peak in strength and power. Therefore, the minimum-to-maximum difference of the diurnal pattern in both MVC and SJ60 may be partly affected (increased) by temporal specificity. However, the minimum-to-maximum difference in the present study (8.7% - day 2) was not larger as compared to the previously reported 12% in subjects without time-specific training history [21]. It suggests that the time-specificity plays, in this regard, a minor role in the present study. The later factor has been proposed to affect data collected with recovery periods shorter than 8 h [10]. Drust et al. suggested that a so called "cyclic Latin square" design [14], when subjects perform the first session at a different time of day in a counterbalanced order, removes the influence of any learning/fatigue effect. In our opinion, cumulative fatigue was not a significant factor in the present study. Diurnal variation on day 2 was comparable with the studies using the above-mentioned "cyclic Latin square" design [9,17,21]. In addition, Gauthier et al. [16] did not find significant differences in diurnal variation of peak torque between a design with multiple sampling within one day and a design extending over 6 subsequent days with only one test session per day. The frequency of sampling in the first design was even higher (every 3 h) in comparison to the present study.

Normalized and averaged myoelectrical activity of VL, RF, and VM muscles did not significantly differ between the time points and it was only nonsignificantly lower on day 2 ( $p = .089$ ) suggesting that neural input to agonist muscles was not statistically altered by time of day or DAY effect. This was in line with works of Nicolas et al. [35] and Guette et al. [21]. The latter study used twitch interpolation technique and EMG normalized to M-wave in order to better differentiate the influence of central and peripheral factors on EMG signal during isometric MVC of knee extensors. They concluded prevailing variation at the muscle tissue level rather than changes in the neural input to knee extensors muscles. On the contrary to our findings, Gauthier et al. [15] reported significantly increased EMG activity in the morning compared to the afternoon and attributed the diurnal variation in MVC torque both to changes in central nervous command and contractile state of the muscle. These conflicting results could be in part accounted for by differences in the test mode and muscle groups tested (upper versus lower extremities). Another important issue is the difference in the surface bipolar EMG data recording and analyzing procedures. Despite thorough control of the measurement procedures, it is always subject to various sources of errors [12,29] and the quantity of EMG as such, as well as its possible diurnal variation, should be treated with great caution.

Besides EMG activity of the knee extensors, the antagonist co-activation during MVC could be excluded from causing day-to-day and day 2 diurnal differences in the present study. BF co-activation remained unchanged and ranged from 8.1% to 9.8% of the BF EMG when acting as an agonist during knee flexion. The level of co-activation was comparable with the values reported previously; 9 - 13.5% [20] and 9.3% - 11.6% [21].

Neuromuscular efficiency ( $NME_{max}$  - inversely expressed as  $EMG_{norm}/peak\ torque_{norm}$  ratio) during MVC did not significantly differ across a day in spite of improvement from morning towards afternoon on both consecutive days. Similar results on MVC were obtained by Guette et al. [22] and Castaingts et al. [4] for the triceps surae. On the contrary, Nicolas et al. [35] and Gauthier et al. [15] found NME of knee extensor and elbow flexors to be significantly higher at 18:00 h compared to 06:00 h, respectively. As stated above, methodological issues related to EMG and muscle group dependence [22] might affect NME and its interpretation.

An interesting finding of the present study was the significant increase in MVC  $NME_{max}$  from day 1 to day 2 ( $p < .05$ ). This day-to-day variability in neuromuscular efficiency was even more pronounced during submaximum isometric contractions ( $p < .01$ ) altogether with significant TOD effect in  $NME_{submax}$ . Improved intra- and intermuscular coordination could be one possible explanation. Instant increase in  $NME_{submax}$  from session to session, especially on day 1, implies the presence of a learning effect. In that sense, significant TOD in MVC40 may actually result from a summing effect of both diurnal variation in NME (n.s. in the present study as suggested by  $NME_{max}$ ) and learning. In MVC, the peak-to-trough difference being rather low on day 1 and in line with the previous studies on day 2 suggests that learning had only a negligible effect on the diurnal variation in the peak torque.

To summarize, the repeatability of diurnal rhythm characteristics in maximum and submaximum strength and power across two consecutive days may depend on the level of familiarization with testing demands rather than other confounding factors like cumulative fatigue or chronotype. Taking the most extreme example, technically demanding and multi-joint SJ60 showed re-



peatable and significant diurnal variations in all, but slightly more pronounced in those subjects who performed this action during the conditioning training. On the other hand, significantly improved day-to-day performance ( $\uparrow$  neuromuscular efficiency) was found in submaximal knee extension despite its one-joint nature and low-difficulty technique. This test was a novel action for all participants with only one familiarization trial performed during the first test session.

In conclusion, repeating multiple sampling per diurnal part of a civil day over two consecutive days did not significantly impair normal diurnal variation in maximum strength and power. The second testing day exhibited fluctuations comparable to the previous studies with no day-to-day carryover influence of cumulative fatigue. Contractile properties of muscle tissue seemed to be the main source of diurnal variation in maximum isometric strength of the lower limbs, rather than alternation in neural drive and/or activation patterns. Further research is needed to address repeatability of diurnal variation in neuromuscular performance across three and more periods (days) and/or under partial sleep deprivation. That could justify appropriateness of forced desynchrony, constant routine or ultra short sleep/wake protocols in the quest for the origin and mechanism in daily variation of muscle strength.

## References

- Atkinson G. Analysis of repeated measurements in physical therapy research: multiple comparisons amongst level means and multi-factorial designs. *Phys Ther Sport* 2002; 3: 191–203
- Bambaeichi E, Reilly T, Cable NT, Giacomoni M. The isolated and combined effects of menstrual cycle phase and time-of-day on muscle strength of eumenorrheic females. *Chronobiol Int* 2004; 21: 645–660
- Callard D, Davenne D, Gauthier A, Lagarde D, Van Hoecke J. Circadian rhythms in human muscular efficiency: continuous physical exercise versus continuous rest. A crossover study. *Chronobiol Int* 2000; 17: 693–704
- Castaigne V, Martin A, Van Hoecke J, Pérot C. Neuromuscular efficiency of the triceps surae in induced and voluntary contractions: morning and evening evaluation. *Chronobiol Int* 2004; 21: 631–643
- Coldwells A, Atkinson G, Reilly T. Sources of variation in back and leg dynamometry. *Ergonomics* 1994; 37: 79–86
- Czeisler CA, Dumont M, Duffy JF, Steinberg JD, Richardson GS, Brown EN, Sánchez R, Rios CD, Ronda JM. Association of sleep-wake habits in older people with changes in output of circadian pacemaker. *Lancet* 1992; 340: 933–936
- Dauchy RT, Blask DE, Sauer LA, Barinard GC, Krause JA. Dim light during darkness stimulates tumor progression fatty acid uptake and metabolism. *Cancer Lett* 1999; 144: 131–136
- De Prins J, Hecquet B. Data processing in chronobiological studies. In: Touitou Y, Haus E (eds). *Biologic Rhythms in Clinical and Laboratory Medicine*. Berlin, Heidelberg, New York: Springer Verlag, 1994: 90–113
- Deschenes MR, Kraemer WJ, Bush JA, Doughty TA, Kim D, Mullen, KM, Ramsey K. Biorhythmic influences on functional capacity of human muscle and physiological responses. *Med Sci Sport Exerc* 1998; 30: 1399–1407
- Drust B, Waterhouse J, Atkinson G, Edwards B, Reilly T. Circadian rhythms in sports performance – an update. *Chronobiol Int* 2005; 22: 21–44
- Duffy JF, Dijk, D, Klerman EB, Czeisler CA. Later endogenous circadian temperature nadir relative to an earlier wake time in older people. *Am J Physiol* 1998; 275: 1478–1487
- Farina D, Merletti R, Enoka R. The extraction of neural strategies from the surface EMG. *J Appl Physiol* 2004; 96: 1486–1495
- Folkard S, Monk TH, Lobban MC. Towards a predictive test of adjustment to shift work. *Ergonomics* 1979; 22: 79–91
- Folkard S, Monk TH. Circadian rhythms in human memory. *Br J Psychol* 1980; 71: 295–307
- Gauthier A, Davenne D, Martin A, Cometti G, Van Hoecke J. Diurnal rhythm of the muscular performance of elbow flexor during isometric contractions. *Chronobiol Int* 1996; 13: 135–146
- Gauthier A, Davenne D, Gentil C, Van Hoecke J. Circadian rhythm in the torque developed by elbow flexors during isometric contraction. Effect of sampling schedules. *Chronobiol Int* 1997; 14: 287–294
- Gauthier A, Davenne D, Martin A, Van Hoecke J. Time of day effects on isometric and isokinetic torque developed during elbow flexion in humans. *Eur J Appl Physiol* 2001; 84: 249–252
- Giacomoni M, Edwards B, Bambaeichi E. Gender differences in the circadian variations in muscle strength assessed with and without superimposed electrical twitches. *Ergonomics* 2005; 48: 1473–1487
- Gleeson NP, Mercer TH. Reproducibility of isokinetic leg strength and endurance characteristics of adult men and women. *Eur J Appl Physiol* 1992; 65: 221–228
- Grabner MD, Campbell KR, Hawthorne DL, Hawkins DA. Electromyographic study of the anterior cruciate ligament-hamstrings synergy during isometric knee extension. *J Orthop Res* 1989; 7: 152–155
- Guette M, Gondin J, Martin A. Time-of-day effect on the torque and neuromuscular properties of dominant and non-dominant quadriceps femoris. *Chronobiol Int* 2005; 22: 1–18
- Guette M, Gondin J, Martin A, Pérot C, Van Hoecke J. Plantar flexion torque as a function of time of day. *Int J Sports Med* 2005; 26: 171–177
- Halberg F. Temporal coordination of physiologic function. *Cold Spr Harb Symp Quant Biol* 1960; 25: 289–310
- Häkkinen K, Komi PV. Electromyographic changes during strength training and detraining. *Med Sci Sports Exerc* 1983; 15: 455–460
- Hermens HJ, Freriks B, Merletti R, Hägg G, Stegeman DF, Blok J, Rau G, Disselhorst-Klug C. SENIAM 8: European Recommendations for Surface ElectroMyoGraphy. Netherlands: Roessingh Research and Development, 1999: 158
- Ilmarinen J, Ilmarinen R, Korhonen O, Nurminen M. Circadian variation of physiological functions related to physical work capacity. *Scand J Work Environ Health* 1980; 6: 112–122
- Izquierdo M, Ibanez J, Gorostiaga E, Garrues M, Zúñiga A, Antón A, Larrión JL, Häkkinen K. Maximal strength and power characteristics in isometric and dynamic actions of the upper and lower extremities in middle-aged and older men. *Acta Physiol Scand* 1999; 167: 57–68
- Izquierdo M, Häkkinen K, Gonzales-Badillo JJ, Ibanez J, Gorostiaga E. Effects of long-term training specificity on maximal strength and power of the upper and lower extremities in athletes from different sports. *Eur J Appl Physiol* 2002; 87: 264–271
- Keenan KG, Farina D, Maluf KS, Merletti R, Enoka R. Influence of amplitude cancellation on the simulated surface electromyogram. *J Appl Physiol* 2005; 98: 120–131
- Kerkhof GA, Van Dongen HPA. Morning-type and evening-type individuals differ in the phase position of their endogenous circadian oscillator. *Neurosci Lett* 1996; 218: 153–156
- Kline CE, Durstine JL, Davis JM, Moore TA, Devlin TM, Zielinski MR, Youngstedt SD. Circadian variation in swim performance. *J Appl Physiol* 2007; 102: 641–649
- Lane JD, Phillips-Bute BG. Caffeine deprivation affects vigilance performance and mood. *Physiol Behav* 1998; 65: 171–175
- Martin A, Carpentier A, Guissard N, Van Hoecke J, Duchateau J. Effect of time of day on force variation in a human muscle. *Muscle Nerve* 1999; 22: 1380–1387
- National Nutrition Council. Finnish Nutrition Recommendations. Ministry of Agriculture and Forestry. Committee report 1998 (7). Helsinki: Oy Edita Ab, 1999: 73
- Nicolas A, Gauthier A, Bessot N, Moussay S, Davenne D. Time-of-day effects on myoelectric and mechanical properties of muscle during maximal and prolonged isokinetic exercise. *Chronobiol Int* 2005; 22: 997–1011
- Pilcher JJ, Huffcutt AI. Effects of sleep deprivation on performance: a meta-analysis. *Sleep* 1996; 19: 318–326
- Reilly T, Down A. Investigation of circadian rhythms in anaerobic power and capacity of the legs. *J Sports Med Phys Fitness* 1992; 32: 346–347
- Souissi N, Gauthier A, Sesboue B, Larue J, Davenne D. Effects of regular training at the same time of day on diurnal fluctuations in muscular performance. *J Sports Sci* 2002; 11: 929–937
- Tassi P, Muzet A. Sleep inertia. *Sleep Med Rev* 2000; 4: 341–353
- Wyse JP, Mercer TH, Gleeson NP. Time-of-day dependence of isokinetic leg strength and associated interday variability. *Br J Sports Med* 1994; 28: 167–170
- Youngstedt SD, O'Connor PJ. The influence of air travel on athletic performance. *Sports Med* 1999; 28: 197–207