

RAUL RÄMSON

Adaptation of selected blood
biochemical stress and energy turnover
markers to different training regimens
in highly trained male rowers



TARTU UNIVERSITY PRESS

Institute of Sport Pedagogy and Coaching Science, University of Tartu, Estonia

Dissertation is accepted for the commencement of the Degree of Doctor of Philosophy in Exercise and Sport Sciences on 26 May 2011 by the Council of the Faculty of Exercise and Sport Sciences, University of Tartu, Tartu, Estonia.

Supervisors: Researcher Jarek Mäestu, PhD, University of Tartu, Tartu, Estonia
 Professor Jaak Jürimäe, PhD, University of Tartu, Tartu, Estonia

Opponent: Assistant Professor Daniela A Rubin, PhD, California State University Fullerton, Fullerton, USA

Commencement: Jakobi 5–203, Tartu on 26 August 2011 at 4 p.m.

Publications of this dissertation were granted by the University of Tartu and ESF grant 6671

ISSN 1406–1058
ISBN 978–9949–19–802–3 (trükis)
ISBN 978–9949–19–803–0 (PDF)

Autoriõigus Raul Rämson, 2011

Tartu Ülikooli Kirjastus
www.tyk.ee
Tellimus nr 451

CONTENTS

LIST OF ORIGINAL ARTICLES	7
1. INTRODUCTION	8
2. REVIEW OF LITERATURE	9
2.1. Rowing training	9
2.2. Training monitoring in rowing	12
2.2.1. Psychological monitoring of rowing training	12
2.2.2. Blood biochemical monitoring of rowing training	13
3. THE AIM OF THE STUDY	17
4. METHODS	18
4.1. Four week high volume low intensity training study	18
4.1.1. Participants	18
4.1.2. Study design	18
4.1.3. Testing schedule	19
4.1.4. Body composition	19
4.1.5. Control of energy intake and energy expenditure	20
4.1.6. The perceived recovery stress-state	20
4.1.7. Anaerobic threshold measurement	21
4.1.8. Long distance rowing test	21
4.1.9. Venous blood sampling	22
4.1.10. Statistical analysis	22
4.2. Cytokines and performance improvement study	23
4.2.1. Participants	23
4.2.2. Study design	23
4.2.3. Body composition	24
4.2.4. Maximal 6000 meter test	24
4.2.5. Venous blood sampling	24
4.2.6. Statistical analysis	25
5. RESULTS	26
5.1. Four week high volume low intensity training study	26
5.1.1. The perceived recovery-stress state	27
5.1.2. Energy expenditure and energy intake	28
5.1.3. Changes in the fasting blood biochemical values	29
5.1.4. Changes in the exercise induced blood biochemical values	29
5.2. Cytokines and performance improvement study	30
6. DISCUSSION	32
6.1. Four week high volume low intensity training study	32
6.2. Cytokines and performance improvement study	37
7. CONCLUSIONS	39

8. REFERENCES	40
SUMMARY IN ESTONIAN	
Valitud vere biokeemiliste stressi ja energia tasakaalu markerite adaptatsioon erinevatele treeningutele hästi treenitud sõudjatel	48
ACKNOWLEDGEMENTS	50
PUBLICATIONS	51
CURRICULUM VITAE	81

LIST OF ORIGINAL ARTICLES

PAPER I

Rämson R, Jürimäe J, Jürimäe T, Mäestu J. **The influence of increased training volume on cytokines and ghrelin concentration in college level male rowers.** *European Journal of Applied Physiology*. 2008;104(5):839–46.

PAPER II

Rämson R, Jürimäe J, Jürimäe T, Mäestu J. **Behavior of testosterone and cortisol during an intensity-controlled high-volume training period measured by a training task-specific test in men rowers.** *Journal of Strength and Conditioning Research*. 2009;23(2):645–51.

PAPER III

Mäestu J, Jürimäe J, Purge P, Rämson R, Jürimäe T. **Performance improvement is associated with higher postexercise responses in interleukin-6 and tumor necrosis factor α concentrations.** *Journal of Sports Medicine and Physical Fitness*. 2010;50(4):524–9.

In Papers I and II, Raul Rämson had primary responsibility for protocol development, subject's enrollment, performing measurements, data analysis, and writing the manuscripts. In the third Paper, Raul Rämson had primary responsibility for subject's enrollment, performing measurements, data analysis and also writing the first version of the manuscript.

I. INTRODUCTION

In rowing the annual training volume is relatively high compared to the actual racing time and may be even up to 1200 hours per year. This means that in rowing the planning and designing the annual training plan is very complex because of different capacities a rower has to develop (strength, endurance, flexibility). The described high training volume also needs carefully planned recovery cycles. The balance between training and overtraining is very delicate. Many athletes associate high training volumes with limited recovery periods. This may disrupt the fragile balance and the accumulation of exercise stress and this can often result in overreaching. If the imbalance between training and recovery persists, the result may be a long-term decrement in performance capacity, in which restoration of performance capacity may take several weeks or months. Therefore, it is very important to design effective training plan and use systematic training monitoring.

The evaluation of the clinical state of an athlete is already among the most complicated tasks in sports medicine and carefully planned training monitoring process is a critical component in the athlete's preparation for competition. To improve in physical performance, athletes should use periods of higher physical stress followed by reduction of the stress level to achieve adaptations at the cellular level. There is still lack of valid diagnostic tools that would reliably help to monitor the training process of the athletes and to prevent excessive training stress that can lead athletes to overtraining. Although competitive rowing efforts last only 5 to 8 minutes, the elite rower generally trains twice a day, with each training session usually lasting during the preparatory period up to 120 minutes or even more. Total training time thus seems extraordinary, considering the duration of the races. Furthermore, the increase in low intensity endurance rowing has been reported to increase during the last decades in elite rowers (Fiskerstrand et al. 2004) and it has also been demonstrated that for rowers the trained kilometers are positively related to the success in championships (Steinacker 1993). However, those types of high volume low intensity training increase the risk of overtraining with daily amount of training volume and particular monotonic training (Fry et al. 1998; Raglin et al. 1999). Furthermore those types of trainings are related to very high energy expenditure that may further stress the organism. Therefore, the aim of the current thesis is to investigate the adaptation of the selected stress and energy turnover markers during different training regimens in highly trained male rowers.

2. REVIEW OF LITERATURE

2.1. Rowing training

Although competitive rowing efforts last only for about 5 to 8 minutes, the elite rower generally trains twice a day, with each training session usually lasting from 60 to 120 minutes or even more. Total training time thus seems extraordinary, considering the duration of the races (Hagerman 2000). During rowing competition, anaerobic alactic and lactic as well as aerobic capacities are stressed to their maximum (Steinacker 1993). If a rower is to be successful, he or she will need to train in every aspect of his abilities that is needed: good technique, flexibility, mobility, strength, power, endurance and speed, and it is important that he or she has a thorough understanding of the role that each of these abilities is important in creating a good rower. There are certain principles that apply to all types of training. These are overload, recovery, specificity, reversibility and evaluation (McArthur 1997).

The two main objectives of rowing training are: 1) to improve a rowers' ability to compete at a greater percentage of maximal oxygen consumption without producing significant lactate accumulation; 2) and to improve the rowers' ability to tolerate and clear lactate. The type of training that most effectively addresses the first objective is training at or above anaerobic threshold. Interval training at high intensities with sufficient rest periods to remove most accumulated lactate improves the athletes ability to tolerate accumulated lactate (Hagerman 2000). It has also been found that the inability to train for one week results in a significant decline in aerobic capacity (Steinacker, 1993).

A training stimulus, which is referred as a single training unit of exercise or other tasks, will generate a response of the trained system if the stimulus is high enough. Repetition of the same training stimulus will result in a smaller response and after repeated training stimulus responses will reach to the limit. By increasing the intensity and duration of a training unit or by increasing the number of training units, further increases in performance can be reached. Increases in training load on the other side will increase the need for recovery and regeneration. Therefore, training has to be organized in phases to induce a training response and to allow recovery (Steinacker et al. 1998).

Rowing training is mainly focused on improving aerobic capacity with the proper relationship of anaerobic and strength training (Mäestu et al. 2005). During competition, a rower depends mainly on his or her aerobic metabolism because energy stores and glycolysis are limited to cover the energy demand only for approximately 1.5–2.0 minutes (Steinacker 1993). Therefore, endurance training, training below anaerobic threshold or blood lactate concentration below $4 \text{ mmol}\cdot\text{l}^{-1}$, is the mainstay of success in rowing (Steinacker 1993; Hagerman 2000; Mäestu et al. 2005). During the last three decades annual training volume in highly trained rowers has increased about 20%, with most of the increase occurring during the winter period and at the intensities

below or around aerobic threshold (Fiskerstrand et al. 2004). Large increases in basic endurance training at intensities clearly below the anaerobic threshold have been utilized. Training at high intensities at or above race pace has been de-emphasized compared to 1970s. Greater emphasis has been placed on training at intensities requiring 90–95% of maximal oxygen consumption, most often in the form of long interval bouts lasting 4–8 minutes (Fiskerstrand et al. 2004).

The average annual training volume among Norwegian international rowers increased from 924 hours during the 1970s, to 966 hours in the 1980s to 1128 hours in the 1990s (Fiskerstrand et al. 2004). The training volume may be divided into two nearly equal halves, the preparatory period and the competition period. International Norwegian medal winners in rowing currently train between 1100 and 1200 hours a year (Fiskerstrand et al. 2004). However, this may probably reflect the upper sustainable/optimal training load for rowers. Low-intensity long-distance training volume has been increased every decade among Norwegian rowers as can be seen in Figure 1.

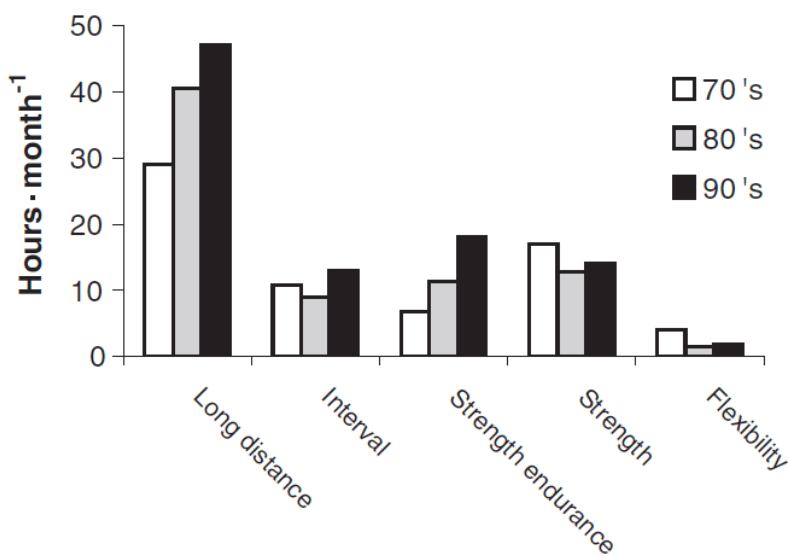


Figure 1. Changes in distribution of training volume among training types from 1970's to 1990's in successful Norwegian international rowers (Fiskerstrand et al. 2004).

Earlier studies suggested that endurance training for rowing is effective only if it is done at a blood lactate concentration between 2.5 mmol·l⁻¹ and 3.5 mmol·l⁻¹ (Steinacker 1988; Urhausen 1988). However, it is very difficult for rowers to exercise for 45 minutes at an intensity corresponding to a lactate concentration of 3.5 mmol·l⁻¹ (Hartmann 1990). It is suggested that endurance training for rowing lasts about 60–120 minutes, however it is very complicated to do it at so

high blood lactate concentrations. Endurance training in rowing at lactate levels $3.5 \text{ mmol}\cdot\text{l}^{-1}$ or higher is not advisable. To improve oxygen use in the muscle, continuous long distance training should be done at a heart rate 130–160 beats per minute and clearly below the anaerobic threshold and at lactate levels around $2 \text{ mmol}\cdot\text{l}^{-1}$ (Hagerman 2000).

Most of the trainings of a rower are at relatively low intensity (blood lactate values around $2 \text{ mmol}\cdot\text{l}^{-1}$). During preparatory period low intensity trainings up to the intensity of aerobic threshold are about 90–94% of the total training load. Only 5–8% of trainings are at intensities between aerobic and anaerobic thresholds. Just before the competitive period the amount of low intensity training is decreased to 86–88%. Workload between the aerobic and anaerobic thresholds remains almost unchanged. There is a slight increase 1% to 4% training intensities where the blood lactate level is up to $8 \text{ mmol}\cdot\text{l}^{-1}$. During the competitive period most of the trainings 70–77% is performed still at low intensities (up to $2 \text{ mmol}\cdot\text{l}^{-1}$), but there has been great increase intensities between aerobic and anaerobic thresholds (up to 22%). Intensities above the anaerobic threshold do not vary much, and remain from 1% to 8%. Training at low intensities influences mostly slow twitch fibers that are critical for rower ensuring high power during the 2000 meter competition. That kind of training also will increase capillary-fiber ratio in working muscles. In successful rowers high capillary-fiber ratio has been found (Hasart 1988). At least 80% of muscle fibers have slow twitch properties, the proportion of slow twitch fibers being greater in the more successful competitors (Steinacker 1993). Accordingly, to specific muscle fiber characteristics the amount of low intensity training is important. Therefore, it appears that not only the determination of anaerobic threshold, but also the determination of aerobic threshold is important in the monitoring process of rowers.

It has been demonstrated that for rowers the trained kilometers are positively related to the success in championships, but the risk of overtraining increases with daily amount of training volume and particular monotonic training (Fry et al. 1998). Use of several training methods, concentration on different training stimuli, variation of training intensity and volume by short time overloading and following recovery, may further increase performance (Steinacker 1993; Altenburg 1997). Therefore, the training plan must contain also recovery cycles. Overtraining is a state of overexertion which may be attained when training load and the associated disturbances in homeostasis are not matched by recovery. The first phase of overtraining is quickly reversible and is referred as overreaching. This refers to a state of training in which normal training load is increased with increasing signs of strain and overstrain and incomplete recovery (Steinacker 1993)

The major categories of training intensities are anaerobic, transition anaerobic threshold, and utilization (Hagerman 2000). Anaerobic training emphasizes the phosphagen and glycolytic energy systems, whereas transition training is designed to provide a maximal stimulus to the cardiovascular and

respiratory systems. The anaerobic-threshold training represents a transition stimulus between aerobic and anaerobic training, and finally utilization training is designed to improve the transport of oxygen to skeletal muscle and its consumption by the muscle fibers (Hagerman 2000).

Over the year, the percentage of specific on water rowing training is about 52–55% for the 18 year old, 55–60% for the 21 year old and up to 65% for the older athlete. Resistance training is in the range 20% at the 18 year old and 16% at the adult athlete, and general athletic training is in the range from 26–23% respectively (Altenburg 1997). It is important to increase specific rowing training with increased training experience (Steinacker et al. 1998). Rowers should attempt to maintain higher aerobic capacity during the off-season so that there is less decrement to overcome as the season begins. It appears that during the off-season, rowers should deemphasize resistance training at low velocities and emphasize power development at higher velocities, train more specifically for the types and velocities of movements used in the rowing technique and at speeds necessary to mimic the competitive pace (Hagerman 2000). It appears that elite rowers would benefit more from performing only simulated or actual rowing training during off-season rather than including resistance training during this period. Also it appears that task-specificity training has the greatest influence on important physiologic responses of elite rowers (Hagerman 2000).

2.2. Training monitoring in rowing

2.2.1. Psychological monitoring of rowing training

Mood state and the level of physiologically related stress and recovery seem to reflect well the clinical state of athletes and are closely related to actual performance (Secher 1993; Steinacker et al. 2000). The most frequently used psychometric questionnaires used in assessing training stress are: 1) the Borg ratio scale (Borg 1998), which was developed to subjectively measure the intensity of the exercise; 2) the Profile of Mood States (POMS) that was initially developed as an economical method of identifying and assessing transient, fluctuating affective state (McNair 1992); and 3) the Recovery-Stress Questionnaire for Athletes (RESTQ-Sport) (Kellmann et al. 2001). It has been argued that the Borg ratio scale and the POMS are not the best instruments to monitor trainings in athletes, because recovery aspect is only vaguely reflected (Kellmann et al. 2001). Furthermore, these instruments do not allow us to know what the reason of the mood change is. Restricting the analysis to the stress dimension alone is insufficient, especially in high performance areas, since the management of training intensity and volume is tightly linked to outstanding performance (Steinacker et al. 2000). The RESTQ-Sport measures the extent to which persons are physically and/or mentally stressed, whether or not they are capable of using individual strategies for recovery as well as which strategies are used (Kellmann et al. 2000; Kellmann et al. 2001). The RESTQ-Sport

consists of 77 items, 19 scales (7 general stress, 5 general recovery, 3 sport-specific stress, 4 sport-specific recovery) with four items each plus one warm-up item. Participants answer retrospectively and a Likert-type scale is used with values ranging from 0 to 6 indicating how often the respondent participated in various activities during the past 3 days/nights (Kellmann 2010). The RESTQ-Sport has been used in various sports to monitor athletes and the impact of training during the preparation camp for World Championships and Olympic Games (Kellmann et al. 2000; Kellmann et al. 2001; Mäestu et al. 2006; Coutts et al. 2007). In rowing, it has been found that increases in training volume were reflected in elevated stress and reduced recovery scores measured by the RESTQ-Sport for athletes (Mäestu et al. 2006). Contrary, the recovery period, in general, caused significant increases in Recovery scales and decreases in Stress scales (Kellmann et al. 2000; Kellmann et al. 2001; Mäestu et al. 2006). The resulting RESTQ-index, the difference between Stress and Recovery scales, has been found to be in dose-response relationship with training volume and cortisol concentration in blood (Mäestu et al. 2006).

2.2.2. Blood biochemical monitoring of rowing training

The acute responses in the endocrine system during stressful trainings are related to the intensity and duration of the specific exercise stimulus and also to the condition of the athlete (Simsch et al. 2002). Hormones influence the regeneration phase through the modulation of anabolic and catabolic processes after training and exercise (Jürimäe et al. 2001; Mastorakos et al. 2005). Hormonal mechanisms most assuredly help mediate both short-term homeostatic control and long-term cellular adaptations to any type of stress. Different fasting or exercise-induced hormonal responses were often proposed for monitoring overreaching and overtraining situations and also for the recovery period with conflicting results (Mackinnon et al. 1997; Steinacker et al. 2000; Simsch et al. 2002; Urhausen et al. 2002; Mäestu et al. 2003; Meeusen et al. 2004; Mäestu et al. 2005). An advantage of an exercise induced hormone changes instead of fasting hormone has been proposed, because exercise induced blood biochemical values may be more sensitive to training load (Urhausen et al. 1995; Mäestu et al. 2005; Jürimäe 2008). However, studies using a design with an acute exercise test before and after periods of altered training load are scarce, and the hormonal data from these studies are somewhat conflicting. Some studies have found different exercise induced hormonal responses after stressful training periods (Mäestu et al. 2003; Meeusen et al. 2004). In contrast, doubling the training load does not alter the stress hormone responses to maximal exercise test was demonstrated (Ronsen et al. 2001). Similarly, no differences were found in acute cortisol response to a 20 km bicycle time trial following 28 days of intensified training (Ndon et al. 1992).

It can be argued that the maximal exercise test may not be the best solution to monitor the blood biochemical response after high volume, low intensity

trainings. It is well established that an intensity threshold exists for stress hormone responses to exercise. For example, exercise intensity higher than 60% aerobic power is sufficient for the increase in cortisol (Vervorm et al. 1991) and about 50% to increase in testosterone (Maresh et al. 1988) and at maximal intensities the increase is higher. Therefore, the high exercise intensity itself may have the greater influence to the observed change in the biochemical parameter than the impact of the low intensity training. Furthermore, the athlete spends only about 20% to 30% of the total time during the graded exercise test in the range of 2 mmol·L⁻¹ and lower blood lactate concentration. Therefore, if the training period is aimed to stress the low intensity energy systems the validity of the measurement of biochemical parameters after high intensity exercise may not be high enough. It can be stated that the more direct is the relationship between a parameter and a specific performance, the more value the testing has.

Testosterone and cortisol values are often used to monitor training load of rowers (Steinacker et al. 2000; Jürimae et al. 2001; Simsch et al. 2002; Mäestu et al. 2003), because changes in the concentrations of those hormones are known to affect the recovery rate and the duration of the recovery after exercise (Kuipers et al. 1988). Studies have found that high load endurance trainings caused an increase in fasting cortisol and a decrease in fasting testosterone concentrations (Urhausen et al. 2002), while other studies have found different results (Steinacker 1999; Mäestu et al. 2003). Those results indicate that the fasting stress hormone responses are not specific and do not mirror exactly the amount of physical stress during different training cycles in athletes (Urhausen et al. 1995; Steinacker et al. 2000; Mäestu et al. 2003; Meeusen et al. 2004; Kraemer 2006).

In response to increased training stress, some athletes are unable to maintain sufficient intake of calories, thus suffering negative energy balance causing further stress. Peripheral feedback to the hypothalamus is supplied by different peptide hormones and several cytokines. However, there is now evidence that several cytokines and peptide hormones (including interleukin-6 [IL-6], tumor necrosis factor- α [TNF- α], leptin and ghrelin) have receptors in the hypothalamus so they can act directly on the hypothalamic neuronal network (Gaillard et al. 2000) and contribute to the responses to nutrition, fasting, and exercise (Steinacker et al. 2004). Such peripheral signals instead of stress hormones have been proposed for monitoring possible overtraining (Smith 2000).

To date, leptin is probably the most studied cytokine when describing the energy homeostasis and excessive training stress (Noland et al. 2001; Simsch et al. 2002; Jürimae et al. 2003; Mäestu et al. 2003; Desgorces et al. 2004). It is well established that leptin is an adipocyte-derived hormone that acts directly on the hypothalamus that regulates a large number of molecules and represents several physiologic functions including regulation of body mass and energy balance (Shintani et al. 2001; Kraemer et al. 2002; Bouassida et al. 2010). The most important function of leptin is its influence on energy balance. Leptin

decreases when energy intake is restricted, causing energy conservation and decreased thermogenesis (Ahima et al. 1996; McMurray et al. 2005). Leptin can be considered an interface between energy intake and energy expenditure in athletes (Jürimäe et al. 2009). Leptin may act as a metabolic hormone and may also have effects on hypothalamic regulation during training in athletes (Steinacker et al. 2004). Some studies suggest that reduction of body fat mass is necessary to reduce leptin concentration as a result of exercise training (Hickey et al. 1996; Perusse et al. 1997; Kraemer et al. 2002), but an independent effect of exercise and increased energy expenditure without changes of body fat reduction on leptin has also been demonstrated in athletes (Simsch et al. 2002; Mäestu et al. 2003; Jürimäe et al. 2007). Studies in rowers have highlighted that during high load endurance training fasting levels of leptin decrease and an increase can be observed when training load is decreased. Short-term, high intensity exercise-induced changes in leptin have been suggested as the first sign of overreaching (Jürimäe et al. 2003).

Ghrelin is secreted by endocrine cells in the gastrointestinal tract and has been found to regulate energy balance and body mass by modulating expression levels of orexigenic peptides in the hypothalamus (Wren et al. 2001; Kojima et al. 1999) and coordinate energy balance by increasing appetite and food intake (Wren et al. 2000; Cummings 2006). The main known receptor for ghrelin is the growth hormone (GH) secretagogue receptor and it is thought that ghrelin targets areas in the hypothalamus and in the forebrain (Meier et al. 2004). Ghrelin is present in two major forms including acyl ghrelin and des-acyl ghrelin (Chen et al. 2009). Acyl ghrelin has a potent effect on eating behavior, causing an increase in hunger (Ariyasu et al. 2001; Korbonits et al. 2001) and plays a key role in the central regulation of feeding and energy balance (St Pierre et al. 2003). Interestingly, total plasma ghrelin levels are significantly changed during acute and chronic alteration of nutritional status with low levels in simple obesity but higher levels after weight loss (Hansen et al. 2002; Espelund et al. 2005). It has reported that 6 weeks of exercise training at a low to moderate exercise intensity resulted in reduced total ghrelin levels in rat plasma and soleus muscle (Ghanbari-Niaki et al. 2009). Moreover, there are some data to suggest that under certain conditions, exercise may suppress circulating ghrelin levels which could decrease feeding behavior in humans (Kraemer et al. 2004; Kraemer et al. 2007). Long-term exercise intervention studies have demonstrated that ghrelin levels increase in response to exercise induced weight loss not the food restriction *per se*, acting via negative feedback loop that regulates body weight (Foster-Schubert et al. 2005). Unfortunately, data on the influence on training is only available on overweight, mostly female subjects, while no data is available on male athletes. It can be hypothesized that if a caloric restriction is apparent during periods of training ghrelin levels should increase also in athletes to stimulate appetite and therefore provoke higher caloric intake. Exercise is a potent stimulus for secretion of many stress hormones (Steinacker et al. 2000), and acute exercise mediated negative energy

balance may contribute to the regulation of plasma leptin (Hickey et al. 1996; Desgorces et al. 2004) and ghrelin (Kojima et al. 1999; Wren et al. 2000) concentrations.

Excessive musculoskeletal stress associated with insufficient rest and recovery may induce a local acute inflammatory response that may evolve into chronic inflammation and produce systemic inflammation. It is suggested that this might be the cause of overtraining and further the overtraining syndrome (Smith 2000). Despite that, numerous studies on leptin describe the training status of the athlete however, less evidence exist in the scientific literature of the responses of IL-6 and TNF- α to the adaptation/response to the training period. A 12 week training program had no effect on fasting concentrations of IL-6 and TNF- α with physically active and inactive subjects (Stewart et al. 2007). An increase in IL-6 concentration after acute intense interval training period with an increase in fatigue and general malaise has been found (Robson-Ansley et al. 2007). Numerous studies on humans have shown that IL-6 and TNF- α increase during endurance exercise, as an acute inflammatory response (Pedersen et al. 1998; Ostrowski et al. 1999). TNF- α is also secreted at the onset of an inflammatory cascade and act locally at the site of injury/infection (Dinarello 1997). Although, IL-6 is synthesized by fat tissue (Fantuzzi 2005) its rise after exercise is fully accounted from the working skeletal muscles (Steensberg et al. 2000). The post-exercise rise in IL-6 levels is also dependent on the glycogen content (Steensberg et al. 2001) which further depends on the proper energy intake and energy balance. Therefore, negative energy balance may have an impact of higher IL-6 concentrations after exercise due to higher muscular damage caused by lowered energy reserves. To our best knowledge there are no studies that could describe the influence of low intensity high volume training period on IL-6 and TNF- α concentration in highly trained athletes.

In conclusion, there is still a lack of valid diagnostic tools that would help us to prevent overtraining. Different hormonal responses were often proposed for monitoring overreaching and overtraining situations and also for the recovery period (Steinacker et al. 1998; Mäestu et al. 2003). However, as training is rather complex in its nature – there is probably a need to study specific markers for specific training conditions, not necessarily the performance itself.

3. THE AIM OF THE STUDY

The main aim of the current thesis was to investigate the adaptation effects of male rowers to low intensity high volume training period.

According to the main aim the specific aims were:

1. To investigate changes in the concentrations of fasting state stress hormones (testosterone and cortisol) during a high volume, low intensity aerobic endurance training period in male rowers.
2. To investigate changes in the concentrations of exercise-induced stress hormones (testosterone and cortisol) after a task specific test during high volume, low intensity endurance training period in male rowers.
3. To investigate changes in the concentrations of fasting state cytokines (IL-6, TNF- α , leptin) and ghrelin during a high volume endurance training period in male rowers.
4. To investigate changes in the concentrations of exercise-induced concentrations of IL-6, TNF- α , leptin and ghrelin after a task specific test during a high volume, low intensity endurance training period in male rowers.
5. To investigate the responses in the concentrations of cytokines (IL-6 and TNF- α) to maximal rowing ergometer test in the conditions of increased rowing performance.

4. METHODS

The current dissertation is combined of two different studies. Study I consisted of the four week high volume, low intensity training cycle, while study II was designed to study long term adaptation and was one year long.

4.1. Four week high volume low intensity training study

4.1.1. Participants

Eight highly trained male rowers participated in the study. The subjects were national level medalists with two subjects belonging to the National Under-23 year's team, who won the gold medal at the U23 World Championships. Subjects' main characteristics are presented in Table 1. None of the subjects had an unsatisfactory medical history or were taking any medication. The subjects were free to stop participating in the study if they liked. All subjects were informed about the study procedures, possible risks and the purpose of the investigation before they signed a written consent as approved by the Medical Ethics Committee of the University of Tartu.

Table 1. Descriptive data (Mean±SD) of the participants (n=8) in the Study I.

Variable	Mean±SD	Minimum	Maximum
Age (years)	20.2±1.6	17	22
Height (cm)	183.9±4.6	178	191
Body mass (kg)	81.0±5.4	74.7	92.6
BMI (kg/m ²)	23.9±1.0	22.5	25.4
Experience (years)	6.5±2.2	3	10

BMI – Body mass index

4.1.2. Study design

The study was conducted during the preparatory period i.e. from the end of October to mid-November. During this period rowers train at low intensity and high volume to build up the aerobic capacity (Mäestu et al. 2005). Training volume during the first week was (Week 1) 587.3 minutes, second week (Week 2) 865.7 minutes, third week (Week 3) 1059.1 minutes and fourth week (Week 4) 578 minutes. Eighty percent of training volume was low-intensity endurance rowing training on single sculls (session time from 75 to 150 minutes; lactate values lower than 2 mmol·L⁻¹), 10 percent was low-intensity running or cycling at the similar intensity and 10 percent was strength endurance training in the

gym that consisted of 40–50% of 1 repetition maximum intensity for 30–50 repetitions.

The whole study period lasted for six weeks, which included the four week training period (Week 1, Week 2, Week 3, Week 4) and one week before and after the four week training period (Figure 2). The week before the training period was a moderate standardized training for athletes to execute the pre training baseline measurements. During this week the training load was about 10 hours. During Week 1 training load was about 10 hours. Training volume was increased about 50% during Week 2 and again 10–15% during Week 3 individually for each subject. The athletes trained six days a week and one day (Monday) was meant for recovery. Training volume was decreased after the experimental period to about the same volume as it was before the four week training period. The training schedule was the same each week. Training sessions were supervised by experienced coaches, who were fully instructed of the study design and the expectable outcome.

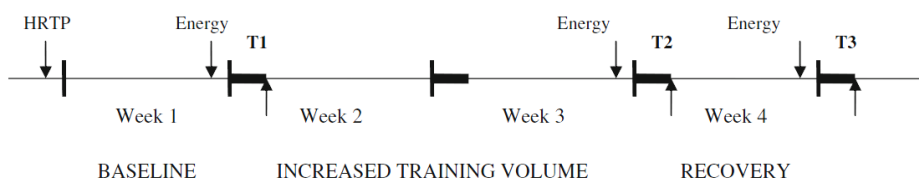


Figure 2. The schematic view of the study I. HRTP – heart rate turn point test; Energy – food diary and energy expenditure calculations ; T1, T2, T3 (indicated by upward arrows) – testing battery complexes that include 24h resting day with DXA, Fasting state blood in the morning (8.00 am), 2 hour rowing test (LDT) (starting between 2.30 and 3.00 pm) and RESTQ-Sport questionnaire (filled on the previous day). The bold black line represents the resting day.

4.1.3. Testing schedule

The fasting state blood, a 10-ml sample from an antecubital vein with the participant in the upright position was drawn in the morning before the long distance rowing test (LDT). Fasting blood samples were obtained at 08.00 hours after the day of full rest (at the beginning of Week 2, at the beginning of Week 4, and after Week 4, T1, T2 and T3, respectively). Additionally, 10-ml venous blood samples were obtained before (PRE), immediately after (POST) and 30 minutes after (POST30') the LDT.

4.1.4. Body composition

The height (Martin metal anthropometer) and body mass (A&D Instruments Ltd, UK) of the participants were measured to the nearest 0.1 cm and 0.05 kg respectively. Body composition measurements were done with the Lunar

DPX-L total body scanner (Lunar Corporation, Madison, WI) which was operated in the medium scan mode (~20 min). The calibration of the machine was done daily as suggested by the manufacturer. The subjects were measured while wearing underwear only, with the arms at the both sides. Body composition was measured during the resting day.

4.1.5. Control of energy intake and energy expenditure

The participants were asked to record all foods and drinks consumed during the last two weekdays (i.e., Thursday and Friday) and one weekend day (Saturday) preceding three testing sessions (Figure 2). The special diets were not prepared to the subjects. They were allowed to eat as much as they liked/felt during the study period, but were asked to maintain their usual dietary habits and everyday activities before trials. The daily energy intake was calculated as the average of the 3 days (Mäestu et al. 2008). The subjects were also instructed by an experienced dietician, and their daily nutritional intake consisted of a high-carbohydrate diet with the composition remaining stable throughout the training season (Jürimäe et al. 2006). Daily energy expenditure during the same days was calculated according to the method of Bouchard et al. (Bouchard et al. 1983). Briefly, a day was divided into 96 periods of 15 min each, and the subject had to fill each period with an activity and an intensity scale from 1 to 9. The scale and the corresponding activities were explained to the subjects before completing the energy expenditure questionnaire.

4.1.6. The perceived recovery stress-state

The perceived recovery stress-state was assessed using the RESTQ-Sport (Kellmann et al. 2001), which has been adapted to Estonian language previously (Jürimäe et al. 2002). Filling out the questionnaire was done in the morning of the day before T1, T2 and T3. The standardized scores for Stress and Recovery were calculated as follows. The scores of stress-related scales (scales 1 to 7, 13, 14 and 15) and the recovery-oriented scales (scales 8 to 12, 16, 17, 18, 19) were summed and divided by the number of scales and were then converted to standardized values by subtracting the global sample mean and dividing the difference by the standard deviation. Thus, a standardized recovery and the stress score could be obtained on a common scale, which allowed computing a difference between stress and recovery (standardized RESTQ-index) (Kellmann 1999).

4.1.7. Anaerobic threshold measurement

Stepwise incremental rowing ergometer test was performed on wind resistance-braked Concept II rowing ergometer (Morrisville, USA) before Week 1 to determine target HR values for prolonged exercise test and for the four week training cycle. The rowers were fully familiarized with the use of the apparatus. Resistance was set at level 5 to 6 of maximal 10 according to the practical experience of the coach. Rowers were equipped with instruments and sat quietly for 2 minutes on the ergometer before starting to exercise at 40 W. Workload was increased by 20 W every minute until voluntary exhaustion. Power and stroke rate were recorded continuously on the computer display of the rowing ergometer. The test was designed to elicit maximal power output at approximately 15 min in each subject (Hofmann et al. 2007). Subjects were strongly encouraged to achieve maximal performance. The subjects were asked not to participate in any physical activity exercises during the previous 24 hours. Heart rate (HR) was measured continuously throughout the test using a commercially available HR monitor (Polar S 725X, Polar Electro, Finland). The room temperature during the testing was in the range of 20 to 22°C and humidity around 50 to 60%. Anaerobic threshold (AnT) determination was performed using linear regression turn point analysis (Hofmann et al. 2007). HR turn point (HRTP) was calculated as described previously (Hofmann et al. 1997). The HRTP was defined as the deflection of the HR performance curve at approximately 90% of maximal HR. Two regression lines were calculated, and the intersection point between both optimized regression lines was termed the HRTP (Hofmann et al. 1997; Hofmann et al. 2007).

4.1.8. Long distance rowing test

Long distance rowing test (LDT) was performed on single sculls for approximately 2 hours (Jürimäe et al. 2001). The length of the test was chosen as one of the typical training session of rowers during the preparatory period. LDT was performed on the day after resting day (i.e. on Tuesday). Testing time (PRE test measurements at 14.30 to 15.00) was held constant for each subject to avoid diurnal changes in biochemical parameters. Target heart rate was set at the level obtained during the incremental test using a practical set ± 3 beats·min⁻¹ of 80% HRTP. Similar HR values were used also for 90% of the trainings during the four week training period. The participants were not allowed to eat two hours before the test and they were asked to visit the toilet before weighing. The participants were not allowed to drink during the test (Jürimäe et al. 2001). The temperature was around 18–20°C and there was no notable wind. Blood samples in the amount of 20µl were collected from fingertip for lactate analysis and were measured using the photo enzymatic method (Lange, Germany). Fingertip blood samples were obtained before the LDT, during the LDT (after 60 minutes) and 5 minutes after the LDT.

4.1.9. Venous blood sampling

A 10-ml blood sample was obtained from an antecubital vein with the subject in the upright position. Fasting blood samples were obtained at 8.00 after the resting day before (T1) and after (T2) the high training volume period and after the recovery period (T3). The plasma was separated and frozen at -20°C for later analysis. Venous blood samples were also drawn during LDT (before, immediately after the test and 30 minutes after). Samples from one individual were run in the same assays. Leptin was determined in duplicate by radioimmunoassay (Mediagnost GMBH, Germany). This assay has a detection limit of $0.01\text{ ng}\cdot\text{ml}^{-1}$, and the intra-assay and inter-assay coefficients of variation (CV) were $<5\%$ and $<7.5\%$ respectively (Mediagnost GMBH, Germany). Ghrelin were determined using a commercially available radioimmunoassay (RIA) kits (Linco Research, USA). The sensitivity of this kit was $93\text{ pg}\cdot\text{l}^{-1}$, and the intra- and inter-assay CV were $<10\%$ and 14.7% respectively. Insulin was determined utilizing IMMULITE 2000 (DPC, Los Angeles, CA, USA). The intra- and inter-assay CV were 4.5% and 12.2% respectively. IL-6 and TNF- α were measured with high-sensitivity ELISA kits (Quantikine HS, R&D Systems Europe, Oxon, UK). For IL-6 the intra-assay and inter assay CV were $<7.8\%$ and 9.6% , respectively and for TNF- α $<8.5\%$ and $<10\%$. Glucose was measured by means of the hexokinase/glucose 6-phosphate-dehydrogenase method via commercially available kit (Boehringer, Mannheim, Germany). Cortisol and testosterone were analyzed in duplicate on IMMULITE 2000 (DPC, Los Angeles, USA). Samples from one individual were run in the same assays. The inter- and intra-assay coefficient of variations was less than 5% . Post-exercise changes in blood, cellular and plasma volumes were calculated using the formulae of Dill and Costill (Dill et al. 1974) and all exercise induced changes in biochemical parameters were corrected to those changes.

4.1.10. Statistical analysis

Statistical analyses were performed using SPSS for Windows, version 13.0 (SPSS, Chicago, IL, USA). Means and standard deviation ($\pm\text{SD}$) were determined. Friedman analyses of variance by ranks were used to examine changes, as the data were not normally distributed. The Wilcoxon matched pairs signed-ranks test was also used to assess the differences between the measured variables. The level of significance was set at $P < 0.05$.

4.2. Cytokines and performance improvement study

4.2.1. Participants

Nine highly trained male rowers (age 19.7 ± 1.0 ; height 190.67 ± 4.24 cm; weight 91.07 ± 6.24 kg) volunteered to participate in the study (Table 2). The participants had no history of immune or cardiovascular disease had no infection or illness at least one month before each testing and during the testing. They have all been medal winners in international world regattas either at junior or under-23 level. In both years, the measurements took place at the beginning of the preparatory period after the period of relative rest. The rowers were fully familiarized with the procedures before providing their written consent to participate at the experiment that was in accordance with the Declaration of Helsinki and approved by the Medical Ethics Committee of the University of Tartu.

Table 2. Anthropometrical and body compositional parameters of the participants (n=9) in study II

Variable	Test 1	Test 2
Height (cm)	190.67 ± 4.2	191.23 ± 4.6
Body mass (kg)	91.07 ± 6.2	91.48 ± 6.8
Fat free mass (kg)	76.30 ± 4.1	76.93 ± 4.9
Fat mass (kg)	10.09 ± 2.6	10.14 ± 2.6
Body fat %	11.51 ± 2.6	11.70 ± 2.6

4.2.2 Study design

During both years, the measurements took place at the beginning of the preparatory period after the period of relative rest. The participants had trained on the average of four times per week during the four weeks preceding the study. The main goal of training during that period was to recover from previous competition season and to prepare for the coming training season. Mean weekly training volume during this period was about 6–8 hours/week. Each subject completed body composition measurements and a maximal 6000 meter rowing ergometer test to determine cortisol, IL-6 and TNF- α responses to a relatively short-term high intensity exercise protocol. The same protocol was used one year later. Rowers did not participate in any physical activity in 24 h before 6000 meter test. A 10-ml venous blood samples were obtained before (PRE), immediately after (POST) and 30 minutes after (POST30') the both 6000 meter tests. Body composition was measured using dual-energy X-ray absorptiometry before the 6000 meter test on previous day.

4.2.3. Body composition

The height (Martin metal anthropometer) and body mass (A&D Instruments Ltd, UK) of the participants were measured to the nearest 0.1 cm and 0.05 kg respectively. Body composition measurements were done with the Lunar DPX-L total body scanner (Lunar Corporation, Madison, WI) which was operated in the medium scan mode (~20 min). The calibration of the machine was done daily as suggested by the manufacturer. The subjects were measured while wearing underwear only, with the arms at the both sides. Body composition was measured during the resting day.

4.2.4. Maximal 6000 meter test

Maximal 6000 meter ergometer performance was assessed in the afternoon on a wind-resistance-braked rowing ergometer (Concept II, Morrisville, VT, USA). All participants were in a post-absorptive condition, having eaten a meal about 2 h before the test (Karila et al. 2008). The participants were instructed by an experienced dietician previously and their daily food intake during the study period consisted of a high-carbohydrate diet with the composition remaining relatively stable. During the second testing (Test2, one year later) the subjects were given the diet protocols that they have been eating during previous testing for breakfast. However, the pre-exercise diet and the fluid intake were not measured (Jürimäe et al. 2005). During the rowing performance test, athletes were asked to cover a distance of 6000 meter in the least time possible. This testing session was the first qualifying test to current season National Team selection. The athletes were fully familiarized with the use of this apparatus and the 6000 meter test, as it was a regular test during their preparatory period. Power and stroke frequency were monitored and recorded continuously via computer display of the rowing ergometer.

4.2.5. Venous blood sampling

A 10-ml fasting blood sample was obtained from an antecubital vein with the participant in the upright position at 10.00 to 11.00 am. During Test 2 the blood sampling was done at same time for each subject. The plasma was separated and frozen at -20°C for later analysis. Venous blood samples were also drawn post-test and 30 minutes post-test. Samples from one individual were run in the same assays IL-6 and TNF- α were measured with high-sensitivity ELISA kits (Quantikine HS, R&D Systems Europe, Oxon, UK). For IL-6 the intra-assay CV was <6.9% and for TNF- α <6.6%, respectively. These samples were corrected to the shifts in the plasma volume according to Dill and Costill (Dill et al. 1974).

4.2.6. Statistical analysis

Means and standard deviation (SD) were determined. Friedman analyses of variance by ranks were used to examine changes, as the data were not normally distributed. The Wilcoxon matched-pairs signed-ranks test was also used to assess the differences between the measured variables. Pearson correlation coefficients were calculated. The level of significance was set at $P < 0.05$

5. RESULTS

5.1. Four week high volume low intensity training study

Training load was increased significantly during Weeks 2 and 3 compared to Week 1 (Figure 3). Significant increase ($P < 0.05$) in training load was also between Week 2 and 3. During the recovery week (Week 4) training load was significantly decreased but was not significantly different from Week 1 ($P > 0.05$).

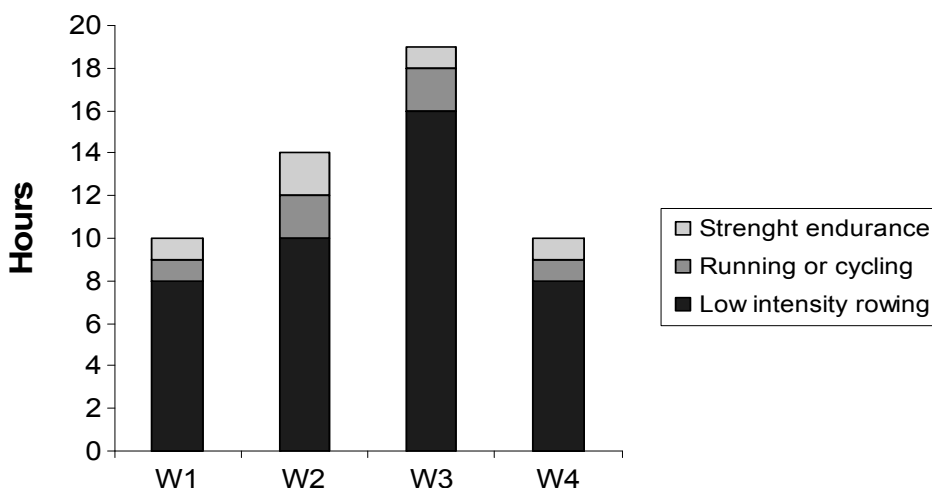


Figure 3. Mean training volume during the 4-week training period in study I. W1 – W4 represent the corresponding weeks.

The average HRTP during the stepwise incremental test was 166.0 ± 6.7 ($\text{beats} \cdot \text{min}^{-1}$) and it corresponded to 86.8 ± 2.8 % of the maximal HR. The value of the HRTP was taken as the parameter from which the calculation of the intensity (80% HRTP) of the LDT was done with the practical application of the 6 beat range. During all LDT the athletes had to row at 132.8 ± 5.4 bpm. During the LDT subjects rowed on the average of 21 km (Table 3). There were no significant differences between the covered distances during the three different LDT tests (Table 2). However, the average HR during LDT was significantly lower during LDT2 ($P < 0.05$).

There were no significant differences in the blood lactate values during each long distance test, however when comparing different testing sessions blood lactate was lower during LDT2 compared to LDT1 and LDT3 ($P < 0.05$). The results showed that the HRTP based method yielded the subjects to work at the

intensities that were close to (slightly lower) than the 2 mmol·L⁻¹ blood lactate concentration (Table 3), which is suggested as an intensity for extensive rowing training during the preparation period (Steinacker 1993; Halson et al. 2002; Mäestu et al. 2005).

Table 3. The parameters of three long-distance tests (LDT's) during the Study I.

Variable	LDT 1	LDT 2	LDT 3
Distance (km)	20.5 ± 1.5	21.0 ± 1.5	21.1 ± 2.1
Time (min)	122.9 ± 2.3	120.8 ± 3.5	123.7 ± 3.1
Heart rate (bpm)	133.0 ± 4.4	131.1 ± 5.3 ^{1,3}	132.8 ± 5.3 ²
Pretest body mass (kg)	81.0 ± 5.4	79.6 ± 5.7 ¹	80.1 ± 5.7
Posttest body mass (kg)	79.7 ± 5.4 [*]	78.5 ± 5.7 ^{*1}	78.8 ± 5.3 [*]
Lactate pre (mmol·L ⁻¹)	1.8 ± 0.4	1.8 ± 0.4	1.8 ± 0.2
Lactate 60' (mmol·L ⁻¹)	1.8 ± 0.4	1.8 ± 0.5	2.0 ± 0.2
Lactate post (mmol·L ⁻¹)	1.7 ± 0.1	1.5 ± 0.2 ^{1,3}	1.8 ± 0.5 ²

Numbers indicate significant difference from pointed week. *– Significantly different from pre test value (P < 0.05).

Body mass of the subjects decreased significantly during each LDT. Moreover, the pre and posttest body mass was significantly lower during LDT2 compared to LDT1 (P < 0.05). Average heart rate was significantly lower during LDT2 compared to LDT1 and LDT3 (P < 0.05). Post exercise blood lactate values were also found to be significantly lower during LDT2 (P < 0.05). No other significant differences were found in blood lactate values during three LDT.

5.1.1. The perceived recovery-stress state

In perceived recovery-stress state, significant increases were found in *Social Stress* and *Fatigue* after high training volume period (T1 vs. T2) (P < 0.05). The *Success* score has also increased after the recovery period compared to the baseline measurements (T1 vs. T3) (P < 0.05). The score of *Physical Complaints* decreased significantly after the recovery period compared to the high volume period (T2 vs. T3) (P < 0.05). No other significant differences were found in the scores of the RESTQ-Sport. RESTQ index was significantly decreased after high volume trainings and was returned to base level after the recovery week (P < 0.05) (Figure 4).

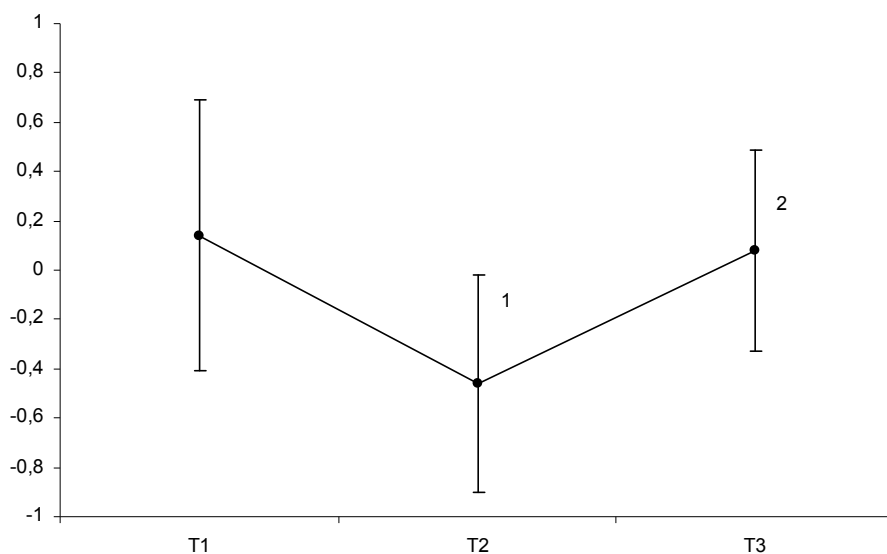


Figure 4. Changes in RESTQ-index during the three testing sessions. Numbers indicate the significant differences from the pointed session ($P < 0.05$). T1 – before high volume training period; T2 – after high volume training period; T3 – after recovery period.

5.1.2. Energy expenditure and energy intake

There was a significant increase in the energy expenditure after high volume trainings at T2 compared to T1 ($P < 0.05$) (Table 4), while no difference was observed between the baseline T1 and the measurements after the recovery week T3 ($P > 0.05$). The corresponding caloric intake also increased significantly from T1 to T2 but resulted in negative energy balance of the average of 455 kcal/day.

Table 4. Energy expenditure and the corresponding energy intake during the 3 testing sessions.

	T1	T2	T3
Energy expenditure ($\text{kcal} \cdot \text{day}^{-1}$)	4576.0 \pm 1009.0	5329.1 \pm 854.1 ¹	4473.0 \pm 1206.9
Caloric intake ($\text{kcal} \cdot \text{day}^{-1}$)	4487.6 \pm 832.4	4879.9 \pm 820.7*	4158.1 \pm 1206.9

¹ – Significantly different from T1 and T3 ($P < 0.05$)

* – Significantly different from the corresponding energy expenditure ($P < 0.05$)

T1 – before high volume training period; T2 – after high volume training period; T3 – after recovery period.

5.1.3. Changes in the fasting blood biochemical values

There were no significant changes in fasting state cortisol, testosterone, insulin, ghrelin, TNF- α and IL-6 values during the four week study period. There was a significant 29% decrease in fasting leptin concentration at T2 compared to T1 (from 1.3 ± 0.5 to 0.9 ± 0.3 ; $P < 0.05$) and leptin tended to increase at T3 compared to T2 (from 0.9 ± 0.3 to 1.2 ± 0.6 , respectively; $P < 0.1$). TNF- α concentration tended to increase (from 1.3 ± 0.5 $\text{pg}\cdot\text{ml}^{-1}$ to 1.5 ± 0.2 $\text{pg}\cdot\text{ml}^{-1}$; $P < 0.1$) at T3 compared to T1.

5.1.4. Changes in the exercise induced blood biochemical values

There were no significant changes in cortisol and testosterone concentrations during the three LDTs. However, testosterone concentration was significantly decreased at POST 30 compared to POST value during LDT2, which was held after the two week high training volume period (Table 5). During LDT2 POST 30' values of cortisol tended to be decreased compared to post-test values ($P = 0.063$). Leptin concentration was significantly decreased at POST test compared to PRE test value and also POST 30' test compared to PRE test value during LDT2. At LDT2 POST 30' leptin concentration was also significantly decreased compared to LDT1. At LDT3 IL-6 concentration was significantly increased compared to LDT2 level. IL-6 concentrations increased significantly at POST and POST 30' test compared to PRE test values during all the exercise tests (LDT1, LDT2 and LDT3). POST 30 level of IL-6 was also significantly higher than POST value during LDT1. TNF- α concentration increased significantly at POST test compared to PRE test and then decreased significantly at POST 30' test compared to POST test during LDT2. There was a tendency of the increased POST 30 ghrelin concentration compared to PRE values ($P = 0.063$). Ghrelin concentration increased significantly at POST 30 test compared to PRE test value during LDT3. Insulin concentrations increased significantly at POST and POST 30' test compared to PRE test values during all the tests (LDT1, LDT2 and LDT3). Due to the changes in body mass during the LDT, all the changes in hormone concentrations were corrected to the changes in plasma volume.

Table 5. Concentrations of leptin, IL-6, TNF- α , ghrelin, insulin, cortisol and testosterone during the three long distance tests.

Parameter	LDT1	LDT2	LDT3
Leptin (ng·ml⁻¹)			
PRE	1.1±0.3	1.1±0.2	1.0±0.4
POST	1.0±0.2	0.8±0.2 ^{1*}	1.0±0.4
POST 30'	1.0±0.3	0.8±0.2 ^{1#}	0.9±0.3
IL-6 (pg·ml⁻¹)			
PRE	0.8±0.4	0.7±0.4	1.1±0.3 ²
POST	3.5±1.6*	3.7±1.9*	3.6±1.5*
POST 30'	4.4±2.0 [#]	3.8±2.5 [#]	3.9±1.6 [#]
TNF-α (pg·ml⁻¹)			
PRE	1.4±0.4	1.3±0.5	1.0±0.4
POST	1.5±0.5	2.1±1.1*	1.0±0.4
POST 30'	1.4±0.7	1.4±0.5 [#]	0.9±0.3
Ghrelin (pg·ml⁻¹)			
PRE	780.5±221.7	819.4±194.6	755.6±140.9 ²
POST	842.1±216.5	822.9±215.5	851.3±229.1
POST 30'	876.3±221.6	841.8±194.0	884.4±172.6 [#]
Insulin (μIU·ml⁻¹)			
PRE	17.2±9.4	11.3±5.1	11.0±4.0
POST	4.0±1.7*	5.2±1.3*	4.5±2.6*
POST 30'	4.1±2.3 [#]	5.0±2.2 [#]	4.7±1.6 [#]
Cortisol (ng·ml⁻¹)			
PRE	288.7±82.8	250.0±91.7	308.3±127.1
POST	311.9±90.4	316.7±136.3	300.2±84.2
POST 30'	275.5±99.3	247.2±72.1	285.2±69.5
Testosterone (ng·ml⁻¹)			
PRE	14.9±4.9	16.0±4.5	14.8±3.0
POST	15.3±3.6	16.7±5.1	16.2±1.4
POST 30'	14.8±5.6	15.6±5.3 [#]	16.5±2.1

LDT1 after baseline, LDT2 after high-volume training. LDT3 after recovery period

* Significantly different from the pre test value ($P < 0.05$)

Significantly different from the post test value ($P < 0.05$)

Number indicates the difference from the pointed testing ($P < 0.05$)

5.2. Cytokines and performance improvement study

There were no significant changes in the anthropometrical and body compositional parameters of the subjects during the study period (Table 6). The maximal 6000 meter rowing ergometer performance was significantly improved during one year period from Test1 to Test2 (330.3±21.9 W to 349.2±20.3 W, respectively, $P < 0.05$), with only one subject failed to improve and for two

subjects the improvement was less than 10 W. PRE test plasma IL-6 concentrations were similarly low at PRE test for two different testing (Table 6) indicating no severe infections at the beginning of the testing. Cortisol and IL-6 were significantly increased during both testing sessions, while TNF- α was only increased after Test 2 when compared to pre-test values. POST-exercise and Post 30 values of TNF- α were significantly higher at Test 2 compared to Test 1, while only POST 30 values of IL-6 were significantly higher at Test 2 compared to Test 1.

Table 6. Changes in IL-6, TNF- α and cortisol during 6000 meter all-out rowing ergometer race in one year from Test1 to Test2 in study II

Parameter	Test 1	Test 2
IL-6 (pg.ml ⁻¹)		
PRE	0.73±0.29	0.65±0.37
POST	2.60±1.47*	3.61±2.54*
POST 30	3.61±2.54*#	6.56±2.58*# ¹
TNF-α (pg.ml ⁻¹)		
PRE	1.62±0.34	1.59±0.30
POST	1.80±0.23	1.98±0.25 ¹
POST 30	1.77±0.22	2.12±0.23 ¹
Cortisol (ng.ml. ⁻¹)		
PRE	397.6±120.8	431.7±186.3
POST	510.9±123.1*	574.1±119.6*
POST 30	591.7±86.2*	683.8±106.4*

* – significantly different from the PRE test value; # – significantly different from the POST test value; 1 – significantly different from the corresponding value at Test 1. (P < 0.05).

There were no significant relationships between post exercise IL-6, TNF- α and cortisol concentrations and the 6000 meter rowing ergometer performance ($r = -0.101 - -0.658$; $P > 0.05$), but post-exercise changes in IL-6 concentration were significantly related to changes in performance ($r = -0.667$ and $r = -0.865$ for POST and POST 30, respectively; $P < 0.05$). There were also no relationships between the measured body compositional and blood biochemical parameters (data not shown).

6. DISCUSSION

6.1. Four week high volume low intensity training study

In this study, high volume training period of male rowers was investigated. This training period consisted of specific rowing training. Furthermore, training intensities were determined and the calculated intensity corresponding to aerobic threshold was used 90% of the total training volume. However, the total amount of training stress was somewhat smaller than in previous studies (Steinacker et al. 2000; Simsch et al. 2002; Mäestu et al. 2003). The main result of this study is that during high training volume phases fasting leptin values decrease significantly and 30 minutes post exercise induced testosterone concentration is decreased compared with low training volume. Moreover, a tendency ($P = 0.063$) of a decreased cortisol concentration at 30 minutes post exercise was observed after high training volume phase. We also found significant decreases in exercise induced concentrations of leptin and TNF- α after high volume training period. Contrary, there was no significant change in ghrelin or IL-6 concentrations after high volume training period, while significant increases were found at baseline and after the recovery week. Hormone concentrations should be corrected by changes in the plasma volume, as a decrease in plasma volume could increase the value (Hackney et al. 1995). Therefore, all exercise induced hormonal concentrations here are corrected for exercise-induced plasma volume alterations.

Anabolic and catabolic stress hormones are often used to monitor training load of rowers (Steinacker et al. 2000; Jürimäe et al. 2001; Simsch et al. 2002; Urhausen et al. 2002; Mäestu et al. 2003), because changes in the concentrations of those hormones are known to affect the recovery rate and the duration of the recovery after exercise (Kuipers et al. 1988), however with controversy results (Mackinnon et al. 1997; Urhausen et al. 2002; Mäestu et al. 2003). In this study we did not find any changes in the concentrations of the fasting testosterone and cortisol levels during high volume endurance training cycle. Moreover, the individual responses to high training volume showed that high training volume period caused both, increases and decreases in fasting hormone values. Therefore, our results expand the data on fasting levels of testosterone and cortisol, indicating that the response of these hormones during high training stress is not uniform and are dependent on several factors (i.e. training intensity, volume, sport discipline, condition of the athlete, etc).

It can be argued that if we stress some of the organ/energy systems in the body during trainings and we want to determine whether this amount of stress can be measured, then we have to test the organism in the same conditions as during the stress conditions. Therefore, instead of fasting hormones, the measurement of exercise induced hormones will have the advantage to study the adaptive state of athletes (Urhausen et al. 1995; Fry et al. 1998). Results of

previous studies, using exercise induced hormone values in the prediction and/or determination of overtraining are controversial. For example, there were no changes in post-exercise cortisol and testosterone concentrations after high load training cycle in high level cyclists (Hoogeveen et al. 1996). However, it has to be mentioned that in previous studies exercise induced changes in hormonal parameters have been investigated in a standardized tests i.e. stepwise incremental tests, maximal performance tests, which may not be sensitive enough to represent the impact of low intensity training cycle. Therefore, in this study a specific exercise test (two hours of low intensity on-water rowing) similar to the trainings of the high volume training cycle was used. We found a significant decrease in 30 minutes post exercise testosterone concentration compared to post exercise value after the period of high training volume (i.e. during LDT2), while no significant changes were observed during LDT1 and LDT3 indicating lower anabolic activity. Furthermore, the 30 min post exercise concentrations of cortisol showed a tendency to decrease ($P = 0.063$) compared to post exercise values at LDT2. It is known from the literature that cortisol increases during exercise, when the exercise intensity is at 60% or higher of individual maximal oxygen consumption, while most of the changes and perhaps effects of this hormone occur after exercise during the early recovery period (Hackney et al. 1999; Daly et al. 2005). It has been proposed that cortisol response becomes inversed in acute fatigued state (Vervoorn et al. 1991). Post exercise increases in cortisol are essential because of its catabolic action on the proteins that were damaged during exercise, thus leaving a “pool” to the synthesis of new amino acids (Virus 1995). The roles of hormones in the recovery phase and their effects on the receptor and extracellular levels remain to be better established. For example, Kraemer et al (Kraemer et al. 2006) have recently shown that muscular androgen receptor content was increased following heavy resistance exercise that may stimulate cellular uptake of testosterone and therefore decrease circulating testosterone concentrations. In contrast, the androgen receptor content was not changed or even decreased after resistance training with significantly shorter duration (Ratamess et al. 2005). However, there is very limited data of the impact endurance exercise on androgen receptor content. In contrast, studies on testosterone concentration after low intensity exercise have shown no significant differences in testosterone or cortisol during two hours of rowing at 80% of the 4 mmol·L⁻¹ anaerobic threshold, which are similar to our data at baseline and after recovery period, but not after high volume period. Therefore, decreased concentration of testosterone may be probably the result of a suppression of the hypothalamus-pituitary-gonadal-axis and may indicate the decreased adaptivity of aerobic energy systems. Future studies are needed to test whether this state is the sign of excessive training stress and may lead to overreaching or even overtraining if the training stress would have been continued.

There was also a significant decrease in the pretest body mass before LDT 2. The loss of body mass has been considered also as one sign of overtraining (i.e.

excessive training stress) and during the stressful training periods a loss of body mass must not be overlooked (Lehmann et al. 1993), although in many studies the body mass has been unchanged (Simsch et al. 2002; Jürimäe et al. 2003). Another sign of the different training status of the athletes was the significant decrease in average HR during LDT 2 when compared to LDT1 and LDT3 (Table 3), although some consideration must be taken before interpreting those results. The decrease of the average HR probably reflects the subjectivity of the subjects to choose the lower intensities of the pre-determined six beat range as can be seen by decreased post exercise lactate values during LDT2. Different studies (Hooper et al. 1995; Steinacker et al. 1998; Halson et al. 2002) have also found decreased (maximal) blood lactate values in overreached/overtrained athletes. However, the interpretation of change in lactate values can only be done if the work intensity is reliably controlled. Our study design the HR range of 6 beats during LDT2 would not allow us to draw such a conclusion.

Significant decreases in leptin concentration at LDT at T2 are intriguing. Previous studies on exercise induced effects on leptin have found significantly decreased leptin concentration immediately after prolonged exercise with an estimated energy expenditure of at least 2800 kcal (Zaccaria et al. 2002) or that the leptin levels experience a delayed (after 9 hours) decrease after acute exercise (estimated energy expenditure at least 850 kcal) (Nindl et al. 2002). In contrast, significantly decreased leptin concentration after 6.5 min maximal rowing ergometer exercise after stressful training period was found (Jürimäe et al. 2003). The energy expenditure during the low intensity two hour on-water rowing is approximately 1200 – 1500 kcal, thus significantly lower than the suggested 2800 kcal that are needed to observe the immediate decrease in leptin concentration. This was also evident during LDT1 and LDT3 where leptin concentrations remained unchanged. However, at T2 the diary analysis of the subjects revealed that they were in the negative energy balance for approximately 455 kcal/day (Table 4), despite that they were allowed to eat as much as they felt during the high volume training period to individualize their subjective energetic demands. However, it is known that participants tend to underreport the caloric intake. In contrast, at T1 and T3 the energy intake and expenditure were not significantly different (Table 4). Therefore, our results support the finding that in the conditions of excessive training stress and decreased energy availability the condition of the athlete can be monitored with the exercise induced leptin values (Jürimäe et al. 2003). However, there is also indication, that leptin changes during prolonged exercise, if energy expenditure is higher than 1000 kcal, are always close to the statistical significance level, demonstrating sometimes significance and sometimes not and should therefore be treated with special care to make definitive conclusions.

It was also interesting to find that fasting ghrelin concentrations did not increase after the extended volume trainings. We hypothesized that if caloric restriction is prevalent an increase in ghrelin concentration would occur to provoke energy intake. Previous studies have highlighted that ghrelin con-

centrations increase after training period (Foster-Schubert et al. 2005) and/or during the period of caloric restriction (Hansen et al. 2002). However, those results are not completely comparable with our study since those studies have used overweight, female subjects in contrast to highly trained lean male athletes used in this study. In contrast, no significant exercise induced increase was found in ghrelin concentration at T2 while significant increase was observed in T3 ($P < 0.05$) and a tendency at T1 ($P = 0.051$). Different studies have indicated that if the energy expenditure during the exercise is high enough an increase in ghrelin concentration may occur (Christ et al. 2006; Jürimäe et al. 2007). Therefore, the behavior of ghrelin concentration at T2 is difficult to explain. However, as the pre exercise measurement was at 2 hour postprandial state (Kallio et al. 2001; Jürimäe et al. 2007) compared to fasting state measured in the morning of the same day, it is possible that nutrition may have had a possible exercise induced effect. As the average negative energy balance was different in three testing conditions (Table 4) the postprandial decrease in ghrelin concentration may have been different i.e. at lower energy restriction the decrease is higher as supported by significantly decreased pre-exercise ghrelin concentrations at LDT3 compared to LDT2 (Table 5). Previous research has also shown that ghrelin concentrations reach nadir approximately one hour after meal and changes in ghrelin concentration are smaller after a meal than after overnight fast in healthy subjects (Ariyasu et al. 2001). In our study increases in ghrelin concentration were 12 and 17% at LDT1 and LDT3, respectively but only 2 % at LDT3. It can be speculated that during a specific metabolic conditions of previous high volume trainings (high energy expenditure), negative energy balance or the temporarily restricted caloric conditions as fasting state and probably relatively low body energy reserves (low body fat%) the further exercise-induced effect of energy expenditure on ghrelin concentration is down-regulated. This can be supported by some previous studies. For example, stable ghrelin concentrations during and after high intensity 27 minutes running at fasting state was found (Kraemer et al. 2004). In contrast, post-exercise concentration of ghrelin nearly reached significance ($P = 0.051$) after 30 minutes rowing slightly above anaerobic threshold in 2 hour postprandial state (Jürimäe et al. 2007) and the behavior of ghrelin was similar in our study at LDT1 and LDT3 (Table 5). A recent study indicated that during caloric restriction and training competitive male bodybuilders showed an increase in ghrelin after 6 weeks but after 12 weeks there was no further increase in ghrelin concentration (Mäestu et al. 2008). Therefore, the mechanism of how the pretest metabolic condition i.e. negative energy balance influences the behavior of post-test concentrations of ghrelin needs further research.

There is proposed that the condition of an athlete during training may be monitored by several cytokines such as IL-1, IL-6 and TNF- α (Smith 2000). This study failed to detect any significant changes in fasting concentrations in IL-6 and TNF- α however, post exercise TNF- α concentration was significantly increased after high training volume period. Several studies have shown that IL-

6 and TNF- α increase during exercise as in the acute inflammatory response (Pedersen et al. 1998; Ostrowski et al. 1999). It is known that the IL-6 response depends on the intensity and duration of the exercise and the amount of muscle mass involved. The exercise induced increase in IL-6 concentration can be attributed mostly to muscle produced IL-6 (Steensberg et al. 2000). The currently used exercise intensity protocol (2 hours of rowing at approximately aerobic threshold) could have been of too less intensive to the working muscles that the additional metabolic strain at LDT2 was not sufficient to the further post exercise IL-6 concentration. Carbohydrate ingestion during the exercise may attenuate the increase in plasma IL-6 (Starkie et al. 2001) and cytokine levels are lower with carbohydrate supplementation compared with no carbohydrate supplementation (Nehlsen-Cannarella et al. 1997). However, only about 5 times increase in IL-6 was detected during each rowing trial which is however, quite small since a 100 fold increase in IL-6 can be found after a marathon race (Ostrowski et al. 1999) Similarly, no increases in IL-6 and TNF- α were found after a 2-h rowing either in placebo or carbohydrate conditions (Henson et al. 2000). In contrast, plasma levels of TNF- α were significantly increased post exercise after increased training period during LDT2. However, the source of TNF- α during exercise is not completely revealed. Adipocytes within the muscle may be an additional source of TNF- α (Coppack 2001). TNF- α is known to stimulate lipolysis in adipocytes and also inhibit insulin action on glucose transport (Steinacker et al. 2004). Therefore, the increase in TNF- α at LDT2 may indicate higher stress to lipid metabolism in higher energy deficit conditions (Table 5).

It would have been advantageous to use also biochemical values from a maximal test in order to compare better the differences with the training specific test. However, it would have been difficult to design a study like that, because of the timetable as two performance tests might interfere each other if a correct recovery period between them is avoided. This was tried to overcome by slightly smaller training stress than previously used in rowers (Steinacker et al. 2000; Simsch et al. 2002; Jürimäe et al. 2003) to study the sensitivity of a training specific test. The changes in above mentioned biochemical parameters and the decreased RESTQ-index after stressful trainings may indicate that the subjects in this study might be in the condition of overreaching. Decreased RESTQ-index has been found on the mountain biker during overtraining (Kellmann et al. 2001) and dose response relationship between training stress and RESTQ-index has been found in high level male rowers (Mäestu et al. 2006).

6.2. Cytokines and performance improvement study

The main finding of this study was that the improvement in performance over 6000 meter simulated rowing race results in higher post-exercise concentrations of plasma IL-6 and TNF- α , while the concentration of cortisol remained unchanged and that changes in post-exercise IL-6 concentrations were related to changes in 6000 meter performance. It has been found that regular exercise appears to lower significantly inflammatory cytokine concentrations and that physically active individuals have lower plasma concentrations of IL-6 and TNF- α when compared to age- and gender-matched inactive groups (Reuben et al. 2003; Stewart et al. 2007). No changes in pre exercise concentrations of cortisol, IL-6 and TNF- α were observed.

IL-6 response to exercise is the most well characterized of all the cytokines. IL-6 is well known to contribute to general sickness behavior in mice (Bluthe et al. 2000) and it has been reported that exogenous administration of IL-6 prior exercise impairs performance and increases subjective feelings of fatigue (Robson-Ansley et al. 2004). The baseline concentrations of IL-6 in our study were similar to other studies, none of the subjects had values higher than 4 pg·ml⁻¹. Similarly, about 5–10 fold increases have been found by other authors using similar intensities (Cox et al. 2007). The absence of IL-6 response after 2 hours of rowing may indicate a lower degree of muscle damage was reported (Henson et al. 2000). The post exercise concentrations of IL-6 are also related to the muscle mass involved (Starkie et al. 2001). In rowing, it is known that up 70 % of the whole body muscles are involved. This can explain relatively high post exercise values of IL-6 after 20 minutes of high intensity workout. However, this does not explain significantly higher IL-6 concentrations during the second testing. It is known that the acute IL-6 response to prolonged exercise may be due to mechanisms independent of muscle damage, such as glycogen depletion (Starkie et al. 2001) or hypoglycemia (Gleeson et al. 2000). However, since both trials were controlled for pretest exercise and food intake those possibilities may be ruled out. Henson et al. (Henson et al. 2000) suggested that no changes in these inflammatory cytokines occur when exercise intensity is not sufficient to induce significant stress hormone response. Accordingly, no changes in fasting cortisol concentrations as a result of prolonged endurance training period were observed (see Table 6) similarly to our previous investigation (Jürimäe et al. 2001). However, it is well known that IL-6 is released from muscle tissue during exercise in relation to glycogen levels (Steensberg et al. 2001). It has been argued that adiponectin may affect glucose metabolism (Hulver et al. 2002). We have previously hypothesized that the increased levels of IL-6 and TNF- α as a result of negative energy balance are the stimuli for the change in adiponectin concentration to trigger the organism for metabolic demands in stressful situations (Jürimäe et al. 2006). We have found post-exercise IL-6 to be related to post-exercise adiponectin concentrations ($r =$

0.643; $P < 0.05$) (unpublished results). However, adiponectin was not measured during this study not allowing to draw the final conclusions.

The reason for increased responses in cytokines during the second testing in the present study remains unclear. It was hypothesized that greater cortisol responses to exercise would have blunted IL-6 responses (Edwards et al. 2006). Neither our, nor Edwards data supported that hypothesis, since cortisol response to both time trials was similar. Furthermore, in our study there were no relationships between the studied cytokines and fat mass and fat free mass (data not shown), therefore significantly higher post-exercise concentrations of IL-6 and TNF- α are not the result of changes in fat mass or fat free mass values (Table 6).

The most likely explanation for the increase in IL-6 and TNF- α concentrations during second testing can be the improved self-mobilization of “going further” beyond the limits. This may account for higher resistance to fatigue and higher activation of additional muscle fibers at the end of the race. Ronsen et al (Ronsen et al. 2002) speculated that the increased IL-6 concentration may reflect the energy crisis within the working muscle, as a result of increased activation of faster motor units that use more glycogen for fuel compared to slow motor units. Furthermore, it has been proposed that exercise induced injury of skeletal muscle fibers triggers the local production on IL-6, possibly through stimulation by TNF- α and also IL-1 β (Ostrowski et al. 1998). Therefore, higher IL-6 concentrations may be the result of higher concentrations TNF- α . This, speculative although, supports the finding that in conditions of higher acute negative energy balance, increases in TNF- α occur. Similar findings were observed in our first study where TNF- α was increased after 2 h low intensity rowing only after high volume training period, when there was negative energy balance in the organism.

7. CONCLUSIONS

Based on the results of the current dissertation the following conclusions were made:

1. There is no change in fasting concentrations of testosterone and cortisol after four week high volume low intensity endurance training period in male rowers.
2. There is a significant decrease in testosterone concentration after low intensity 2 hour rowing test performed after four week high volume low intensity endurance training period, while a tendency to decrease was found in cortisol concentration.
3. The four week high volume low intensity endurance training period induces decreases in fasting leptin concentration.
4. The four week high volume low intensity endurance training period induces decreases in post exercise concentrations of leptin and increases in ghrelin and TNF- α , with no effect on IL-6 concentration.
5. Long term training adaptation and increases in performance induces higher responses in IL-6 and TNF- α to maximal rowing ergometer test.

8. REFERENCES

1. Ahima, R. S., D. Prabakaran, C. Mantzoros, D. Qu, B. Lowell, E. Maratos-Flier and J. S. Flier (1996). Role of leptin in the neuroendocrine response to fasting. *Nature* 382(6588): 250–252.
2. Altenburg, D. (1997). The german talent-identification and talent-development program. H. Perry, Dieterle, I. Lousanne.
3. Ariyasu, H., K. Takaya, T. Tagami, Y. Ogawa, K. Hosoda, T. Akamizu, M. Suda, T. Koh, K. Natsui, S. Toyooka, G. Shirakami, T. Usui, A. Shimatsu, K. Doi, H. Hosoda, M. Kojima, K. Kangawa and K. Nakao (2001). Stomach is a major source of circulating ghrelin, and feeding state determines plasma ghrelin-like immunoreactivity levels in humans. *J Clin Endocr Metab* 86(10): 4753–4758.
4. Bluthé, R. M., B. Michaud, V. Poli and R. Dantzer (2000). Role of IL-6 in cytokine-induced sickness behavior: a study with IL-6 deficient mice. *Physiol Behav* 70(3–4): 367–373.
5. Borg, G. (1998). Perceived Exertion and Pain Rating Scales. *Human Kinetics*. Champaign.
6. Bouassida, A., K. Chamari, M. Zaouali, Y. Feki, A. Zbidi and Z. Tabka (2010). Review on leptin and adiponectin responses and adaptations to acute and chronic exercise. *Br J Sports Med* 44(9): 620–630.
7. Bouchard, C., A. Tremblay, C. Leblanc, G. Lortie, R. Savard and G. Theriault (1983). A method to assess energy expenditure in children and adults. *Am J Clin Nutr* 37(3): 461–467.
8. Chen, C. Y., A. Asakawa, M. Fujimiya, S. D. Lee and A. Inui (2009). Ghrelin gene products and the regulation of food intake and gut motility. *Pharmacol Rev* 61(4): 430–481.
9. Christ, E. R., M. Zehnder, C. Boesch, R. Trepp, P. E. Mullis, P. Diem and J. Decombaz (2006). The effect of increased lipid intake on hormonal responses during aerobic exercise in endurance-trained men. *Eur J Endocrinol* 154(3): 397–403.
10. Coutts, A. J., L. K. Wallace and K. M. Slattery (2007). Monitoring changes in performance, physiology, biochemistry, and psychology during overreaching and recovery in triathletes. *Int J Sports Med* 28(2): 125–134.
11. Cox, A. J., D. B. Pyne, P. U. Saunders, R. Callister and M. Gleeson (2007). Cytokine responses to treadmill running in healthy and illness-prone athletes. *Med Sci Sport Exer* 39(11): 1918–1926.
12. Cummings, D. E. (2006). Ghrelin and the short- and long-term regulation of appetite and body weight. *Physiology & Behavior* 89(1): 71–84.
13. Daly, W., C. A. Seegers, D. A. Rubin, J. D. Dobridge and A. C. Hackney (2005). Relationship between stress hormones and testosterone with prolonged endurance exercise. *Eur J Appl Physiol* 93(4): 375–380.
14. Desgorces, F. D., M. Chennaoui, D. Gomez-Merino, C. Drogou and C. Y. Guezennec (2004). Leptin response to acute prolonged exercise after training in rowers. *Eur J Appl Physiol* 91(5–6): 677–681.
15. Dill, D. B. and D. L. Costill (1974). Calculation of percentage changes in volumes of blood, plasma, and red cells in dehydration. *J Appl Physiol* 37(2): 247–248.
16. Dinarello, C. A. (1997). Proinflammatory and anti-inflammatory cytokines as mediators in the pathogenesis of septic shock. *Chest* 112(6 Suppl): 321S–329S.

17. Edwards, K. M., V. E. Burns, C. Ring and D. Carroll (2006). Individual differences in the interleukin-6 response to maximal and submaximal exercise tasks. *J Sport Sci* 24(8): 855–862.
18. Espelund, U., T. K. Hansen, K. Hojlund, H. Beck-Nielsen, J. T. Clausen, B. S. Hansen, H. Orskov, J. O. L. Jorgensen and J. Frystyk (2005). Fasting unmasks a strong inverse association between ghrelin and cortisol in serum: Studies in obese and normal-weight subjects. *J Clin Endocr Metab* 90(2): 741–746.
19. Fantuzzi, G. (2005). Adipose tissue, adipokines, and inflammation. *J Allergy Clin Immunol* 115(5): 911–919; quiz 920.
20. Fiskerstrand, A. and K. S. Seiler (2004). Training and performance characteristics among Norwegian International Rowers 1970–2001. *Scand J Med Sci Spor* 14(5): 303–310.
21. Foster-Schubert, K. E., A. McTiernan, R. S. Frayo, R. S. Schwartz, K. B. Rajan, Y. Yasui, S. S. Tworoger and D. E. Cummings (2005). Human plasma ghrelin levels increase during a one-year exercise program. *J Clin Endocrinol Metab* 90(2): 820–825.
22. Fry, A. C., W. J. Kraemer and L. T. Ramsey (1998). Pituitary-adrenal-gonadal responses to high-intensity resistance exercise overtraining. *Journal of Applied Physiology* 85(6): 2352–2359.
23. Gaillard, R. C., E. Spinedi, T. Chautard and F. P. Pralong (2000). Cytokines, leptin, and the hypothalamo-pituitary-adrenal axis. *Ann N Y Acad Sci* 917: 647–657.
24. Ghanbari-Niaki, A., H. Abednazari, S. M. Tayebi, A. Hossaini-Kakhak and R. R. Kraemer (2009). Treadmill training enhances rat agouti-related protein in plasma and reduces ghrelin levels in plasma and soleus muscle. *Metabolism-Clinical and Experimental* 58(12): 1747–1752.
25. Gleeson, M. and N. C. Bishop (2000). Special feature for the Olympics: effects of exercise on the immune system: modification of immune responses to exercise by carbohydrate, glutamine and anti-oxidant supplements. *Immunol Cell Biol* 78(5): 554–561.
26. Hackney, A. C., M. C. Premo and R. G. McMurray (1995). Influence of aerobic versus anaerobic exercise on the relationship between reproductive hormones in men. *J Sports Sci* 13(4): 305–311.
27. Hackney, A. C. and A. Viru (1999). Twenty-four-hour cortisol response to multiple daily exercise sessions of moderate and high intensity. *Clin Physiol* 19(2): 178–182.
28. Hagerman, F. C. (2000). *Physiology of Competitive Rowing*. Exercise and Sport Science. W. K. Garrett, D. T. Lippincott Williams & Wilkins: 834–873.
29. Halson, S. L., M. W. Bridge, R. Meeusen, B. Busschaert, M. Gleeson, D. A. Jones and A. E. Jeukendrup (2002). Time course of performance changes and fatigue markers during intensified training in trained cyclists. *J Appl Physiol* 93(3): 947–956.
30. Hansen, T. K., R. Dall, H. Hosoda, M. Kojima, K. Kangawa, J. S. Christiansen and J. O. L. Jorgensen (2002). Weight loss increases circulating levels of ghrelin in human obesity. *Clin Endocrinol* 56(2): 203–206.
31. Hartmann, U., Mader, A., Hollmann, W. (1990). Heart rate and lactate during endurance training programs in rowing and its relation the duration of exercise by top flight rowers. *FISA Coach*(1): 1–4.

32. Hasart, E., Gabriel, B., Grabs, D. (1988). Enzymaktivitäten energieliefernder Stoffwechselsysteme in der Muskulatur von Sporttreibenden zyklischer Sportarten. *Med Sport* 28: 195–201.
33. Henson, D. A., D. C. Nieman, S. L. Nehlsen-Cannarella, O. R. Fagoaga, M. Shannon, M. R. Bolton, J. M. Davis, C. T. Gaffney, W. J. Kelln, M. D. Austin, J. M. E. Hjertman and B. K. Schilling (2000). Influence of carbohydrate on cytokine and phagocytic responses to 2 h of rowing. *Med Sci Sport Exer* 32(8): 1384–1389.
34. Hickey, M. S., R. V. Considine, R. G. Israel, T. L. Mahar, M. R. McCammon, G. L. Tyndall, J. A. Houmard and J. F. Caro (1996). Leptin is related to body fat content in male distance runners. *Am J Physiol-Endoc M* 34(5): E938–E940.
35. Hickey, M. S., R. V. Considine, R. G. Israel, T. L. Mahar, M. R. McCammon, G. L. Tyndall, J. A. Houmard and J. F. Caro (1996). Leptin is related to body fat content in male distance runners. *Am J Physiol* 271(5 Pt 1): E938–940.
36. Hofmann, P., T. Jürimäe, J. Jürimäe, P. Purge, J. Mäestu, M. Wonisch, R. Pokan and S. P. von Duvillard (2007). H RTP, prolonged ergometer exercise, and single sculling. *Int J Sports Med* 28(11): 964–969.
37. Hofmann, P., R. Pokan, S. P. von Duvillard, F. J. Seibert, R. Zweiker and P. Schmid (1997). Heart rate performance curve during incremental cycle ergometer exercise in healthy young male subjects. *Med Sci Sports Exerc* 29(6): 762–768.
38. Hoogeveen, A. R. and M. L. Zonderland (1996). Relationships between testosterone, cortisol and performance in professional cyclists. *Int J Sports Med* 17(6): 423–428.
39. Hooper, S. L. and L. T. Mackinnon (1995). Monitoring Overtraining in Athletes – Recommendations. *Sports Med* 20(5): 321–327.
40. Hulver, M. W., D. H. Zheng, C. J. Tanner, J. A. Houmard, W. E. Kraus, C. A. Slentz, M. K. Sinha, W. J. Pories, K. G. MacDonald and G. L. Dohm (2002). Adiponectin is not altered with exercise training despite enhanced insulin action. *Am J Physiol-Endoc M* 283(4): E861–E865.
41. Jürimäe, J. (2008). Methods for monitoring training status and their effects on performance in rowing. *Int Sportmed J* 9(1): 11–21.
42. Jürimäe, J., P. Hofmann, T. Jürimäe, J. Mäestu, P. Purge, M. Wonisch, R. Pokan and S. P. von Duvillard (2006). Plasma adiponectin response to sculling exercise at individual anaerobic threshold in college level male rowers. *Int J Sports Med* 27(4): 272–277.
43. Jürimäe, J., P. Hofmann, T. Jürimäe, R. Palm, J. Mäestu, P. Purge, K. Sudi, K. Rom and S. P. von Duvillard (2007). Plasma ghrelin responses to acute sculling exercises in elite male rowers. *Eur J Appl Physiol* 99(5): 467–474.
44. Jürimäe, J., T. Jürimäe and P. Purge (2001). Plasma testosterone and cortisol responses to prolonged sculling in male competitive rowers. *J Sports Sci* 19(11): 893–898.
45. Jürimäe, J., E. Lätt, K. Haljaste, P. Purge, A. Cicchella and T. Jürimäe (2009). Influence of puberty on ghrelin and BMD in athletes. *Int J Sports Med* 30(6): 403–407.
46. Jürimäe, J., J. Mäestu and T. Jürimäe (2003). Leptin as a marker of training stress in highly trained male rowers? *Eur J Appl Physiol* 90(5–6): 533–538.
47. Jürimäe, J., J. Mäestu, P. Purge, T. Jürimäe and T. Sööt (2002). Relations among heavy training stress, mood state, and performance for male junior rowers. *Percept Mot Skills* 95(2): 520–526.

48. Jürimäe, J., P. Purge and T. Jürimäe (2005). Adiponectin is altered after maximal exercise in highly trained male rowers. *Eur J Appl Physiol* 93(4): 502–505.
49. Jürimäe, J., P. Purge and T. Jürimäe (2006). Adiponectin and stress hormone responses to maximal sculling after volume-extended training season in elite rowers. *Metabolism* 55(1): 13–19.
50. Jürimäe, J., P. Purge and T. Jürimäe (2007). Effect of prolonged training period on plasma adiponectin in elite male rowers. *Horm Metab Res* 39(7): 519–523.
51. Jürimäe, J., R. Rämson, J. Mäestu, P. Purge, T. Jürimäe, P. J. Arciero and S. P. von Duvillard (2009). Plasma visfatin and ghrelin response to prolonged sculling in competitive male rowers. *Med Sci Sports Exerc* 41(1): 137–143.
52. Kallio, J., U. Pesonen, M. K. Karvonen, M. Kojima, H. Hosoda, K. Kangawa and M. Koulu (2001). Enhanced exercise-induced GH secretion in subjects with Pro7 substitution in the prepro-NPY. *J Clin Endocrinol Metab* 86(11): 5348–5352.
53. Karila, T. A., P. Sarkkinen, M. Marttinen, T. Seppala, A. Mero and K. Tallroth (2008). Rapid weight loss decreases serum testosterone. *Int J Sports Med* 29(11): 872–877.
54. Kellmann, M. (2010). Preventing overtraining in athletes in high-intensity sports and stress/recovery monitoring. *Scand J Med Sci Spor* 20: 95–102.
55. Kellmann, M., D. Altenburg, W. Lormes and J. M. Steinacker (2001). Assessing stress and recovery during preparation for the world championships in rowing. *Sport Psychol* 15(2): 151–167.
56. Kellmann, M. and K. D. Gunther (2000). Changes in stress and recovery in elite rowers during preparation for the Olympic Games. *Med Sci Sports Exerc* 32(3): 676–683.
57. Kellmann, M. and K. W. Kallus (2001). Recovery-stress questionnaire for athletes : user manual. Champaign, IL, Human Kinetics.
58. Kellmann, M., Kallus, K. W. (1999). Mood, Recovery-Stress State and Regeneration. Overload, Performance Incompetence, and Regeneration in Sport. M. Lehmann. New York, Kluwer/Plenum Publishers: 101–117.
59. Kojima, M., H. Hosoda, Y. Date, M. Nakazato, H. Matsuo and K. Kangawa (1999). Ghrelin is a growth-hormone-releasing acylated peptide from stomach. *Nature* 402(6762): 656–660.
60. Korbonits, M., S. A. Bustin, M. Kojima, S. Jordan, E. F. Adams, D. G. Lowe, K. Kangawa and A. B. Grossman (2001). The expression of the growth hormone secretagogue receptor ligand ghrelin in normal and abnormal human pituitary and other neuroendocrine tumors. *J Clin Endocrinol Metab* 86(2): 881–887.
61. Kraemer, R. R. and V. D. Castracane (2007). Exercise and humoral mediators of peripheral energy balance: Ghrelin and adiponectin. *Exp Biol Med* 232(2): 184–194.
62. Kraemer, R. R., H. Chu and V. D. Castracane (2002). Leptin and exercise. *Exp Biol Med (Maywood)* 227(9): 701–708.
63. Kraemer, R. R., R. J. Durand, E. O. Acevedo, L. G. Johnson, G. R. Kraemer, E. P. Hebert and V. D. Castracane (2004). Rigorous running increases growth hormone and insulin-like growth factor-I without altering ghrelin. *Exp Biol Med* 229(3): 240–246.
64. Kraemer, R. R., R. J. Durand, D. B. Hollander, J. L. Tryniecki, E. P. Hebert and V. D. Castracane (2004). Ghrelin and other glucoregulatory hormone responses to eccentric and concentric muscle contractions. *Endocrine* 24(1): 93–98.

65. Kraemer, W. J., B. A. Spiering, J. S. Volek, N. A. Ratamess, M. J. Sharman, M. R. Rubin, D. N. French, R. Silvestre, D. L. Hatfield, J. L. Van Heest, J. L. Vingren, D. A. Judelson, M. R. Deschenes and C. M. Maresh (2006). Androgenic responses to resistance exercise: Effects of feeding and L-carnitine. *Med Sci Sport Exer* 38(7): 1288–1296.
66. Kuipers, H. and H. A. Keizer (1988). Overtraining in Elite Athletes - Review and Directions for the Future. *Sports Medicine* 6(2): 79–92.
67. Lehmann, M., C. Foster and J. Keul (1993). Overtraining in Endurance Athletes – a Brief Review. *Med Sci Sport Exer* 25(7): 854–862.
68. Mackinnon, L. T., S. L. Hooper, S. Jones, R. D. Gordon and A. W. Bachmann (1997). Hormonal, immunological, and hematological responses to intensified training in elite swimmers. *Med Sci Sport Exer* 29(12): 1637–1645.
69. Mäestu, J., J. Jürimäe and T. Jürimäe (2003). Effect of heavy increase in training stress on the plasma leptin concentration in highly trained male rowers. *Horm Res* 59(2): 91–94.
70. Mäestu, J., J. Jürimäe and T. Jürimäe (2003). Hormonal reactions during heavy training stress and following tapering in highly trained male rowers. *Horm Metab Res* 35(2): 109–113.
71. Mäestu, J., J. Jürimäe and T. Jürimäe (2005). Monitoring of performance and training in rowing. *Sports Medicine* 35(7): 597–617.
72. Mäestu, J., J. Jürimäe and K. Kreegipuu (2006). Changes in perceived stress and recovery during heavy training in highly trained male rowers. *Sport Psychol* 20(1): 24–39.
73. Mäestu, J., J. Jürimäe, I. Valter and T. Jürimäe (2008). Increases in ghrelin and decreases in leptin without altering adiponectin during extreme weight loss in male competitive bodybuilders. *Metabolism* 57(2): 221–225.
74. Maresh, C. M., M. R. Cook, H. D. Cohen, C. Graham and W. S. Gunn (1988). Exercise testing in the evaluation of human responses to powerline frequency fields. *Aviat Space Environ Med* 59(12): 1139–1145.
75. Mastorakos, G. and M. Pavlatou (2005). Exercise as a stress model and the interplay between the hypothalamus-pituitary-adrenal and the hypothalamus-pituitary-thyroid axes. *Horm Metab Res* 37(9): 577–584.
76. McArthur, J. (1997). High performance rowing. Wiltshire, The Crowood Press Ltd.
77. McMurray, R. G. and A. C. Hackney (2005). Interactions of metabolic hormones, adipose tissue and exercise. *Sports Medicine* 35(5): 393–412.
78. McNair, D. N., Lorr, M., Droppelman, L. F. (1992). Profile of Mood States. Manual. San Diego.
79. Meeusen, R., M. F. Piacentini, B. Busschaert, L. Buyse, G. De Schutter and J. Stray-Gundersen (2004). Hormonal responses in athletes: the use of a two bout exercise protocol to detect subtle differences in (over)training status. *Eur J Appl Physiol* 91(2–3): 140–146.
80. Meier, U. and A. M. Gressner (2004). Endocrine regulation of energy metabolism: Review of pathobiochemical and clinical chemical aspects of leptin, ghrelin, adiponectin, and resistin. *Clin Chem* 50(9): 1511–1525.
81. Ndon, J. A., A. C. Snyder, C. Foster and W. B. Wehrenberg (1992). Effects of chronic intense exercise training on the leukocyte response to acute exercise. *Int J Sports Med* 13(2): 176–182.

82. NehlsenCannarella, S. L., O. R. Fagoaga, D. C. Nieman, D. A. Henson, D. E. Butterworth, R. L. Schmitt, E. M. Bailey, B. J. Warren, A. Utter and J. M. Davis (1997). Carbohydrate and the cytokine response to 2.5 h of running. *Journal of Applied Physiology* 82(5): 1662–1667.
83. Nindl, B. C., W. J. Kraemer, P. J. Arciero, N. Samatallee, C. D. Leone, M. F. Mayo and D. L. Hafeman (2002). Leptin concentrations experience a delayed reduction after resistance exercise in men. *Med Sci Sport Exer* 34(4): 608–613.
84. Noland, R. C., J. T. Baker, S. R. Boudreau, R. W. Kobe, C. J. Tanner, R. C. Hickner, M. R. McCammon and J. A. Houmard (2001). Effect of intense training on plasma leptin in male and female swimmers. *Med Sci Sport Exer* 33(2): 227–231.
85. Ostrowski, K., C. Hermann, A. Bangash, P. Schjerling, J. N. Nielsen and B. K. Pedersen (1998). A trauma-like elevation of plasma cytokines in humans in response to treadmill running. *J Physiol-London* 513(3): 889–894.
86. Ostrowski, K., T. Rohde, S. Asp, P. Schjerling and B. K. Pedersen (1999). Pro- and anti-inflammatory cytokine balance in strenuous exercise in humans. *J Physiol* 515 (Pt 1): 287–291.
87. Pedersen, B. K., K. Ostrowski, T. Rohde and H. Bruunsgaard (1998). The cytokine response to strenuous exercise. *Can J Physiol Pharmacol* 76(5): 505–511.
88. Perusse, L., G. Collier, J. Gagnon, A. S. Leon, D. C. Rao, J. S. Skinner, J. H. Wilmore, A. Nadeau, P. Z. Zimmet and C. Bouchard (1997). Acute and chronic effects of exercise on leptin levels in humans. *J Appl Physiol* 83(1): 5–10.
89. Raglin, J. and A. Barzdukas (1999). Overtraining in athletes: The challenge of prevention. A consensus statement. *Acsms Health Fit J* 3(2): 27–31.
90. Ratamess, N. A., W. J. Kraemer, J. S. Volek, C. M. Maresh, J. L. VanHeest, M. J. Sharman, M. R. Rubin, D. N. French, J. D. Vescovi, R. Silvestre, D. L. Hatfield, S. J. Fleck and M. R. Deschenes (2005). Androgen receptor content following heavy resistance exercise in men. *J Steroid Biochem* 93(1): 35–42.
91. Reuben, D. B., L. Judd-Hamilton, T. B. Harris and T. E. Seeman (2003). The associations between physical activity and inflammatory markers in high-functioning older persons: MacArthur studies of successful aging. *J Am Geriatr Soc* 51(8): 1125–1130.
92. Robson-Ansley, P. J., A. Blannin and M. Gleeson (2007). Elevated plasma interleukin-6 levels in trained male triathletes following an acute period of intense interval training. *Eur J Appl Physiol* 99(4): 353–360.
93. Robson-Ansley, P. J., L. de Milander, M. Collins and T. D. Noakes (2004). Acute interleukin-6 administration impairs athletic performance in healthy, trained male runners. *Can J Appl Physiol* 29(4): 411–418.
94. Ronsen, O., K. Holm, H. Staff, P. K. Opstad, B. K. Pedersen and R. Bahr (2001). No effect of seasonal variation in training load on immuno-endocrine responses to acute exhaustive exercise. *Scand J Med Sci Spor* 11(3): 141–148.
95. Ronsen, O., T. Lea, R. Bahr and B. K. Pedersen (2002). Enhanced plasma IL-6 and IL-1ra responses to repeated vs. single bouts of prolonged cycling in elite athletes. *Journal of Applied Physiology* 92(6): 2547–2553.
96. Secher, N. H. (1993). Physiological and Biomechanical Aspects of Rowing – Implications for Training. *Sports Medicine* 15(1): 24–42.
97. Shintani, M., Y. Ogawa, K. Ebihara, M. Aizawa-Abe, F. Miyayama, K. Takaya, T. Hayashi, G. Inoue, K. Hosoda, M. Kojima, K. Kangawa and K. Nakao (2001). Ghrelin, an endogenous growth hormone secretagogue, is a novel orexigenic

- peptide that antagonizes leptin action through the activation of hypothalamic neuropeptide Y/Y1 receptor pathway. *Diabetes* 50(2): 227–232.
98. Simsch, C., W. Lormes, K. G. Petersen, S. Baur, Y. Liu, A. C. Hackney, M. Lehmann and J. M. Steinacker (2002). Training intensity influences leptin and thyroid hormones in highly trained rowers. *International Journal of Sports Medicine* 23(6): 422–427.
 99. Smith, L. L. (2000). Cytokine hypothesis of overtraining: a physiological adaptation to excessive stress? *Med Sci Sport Exer* 32(2): 317–331.
 100. St Pierre, D. H., L. X. Wang and Y. Tache (2003). Ghrelin: A novel player in the gut-brain regulation of growth hormone and energy balance. *News Physiol Sci* 18: 242–246.
 101. Starkie, R. L., M. J. Arkinstall, I. Koukoulas, J. A. Hawley and M. A. Febbraio (2001). Carbohydrate ingestion attenuates the increase in plasma interleukin-6, but not skeletal muscle interleukin-6 mRNA, during exercise in humans. *J Physiol-London* 533(2): 585–591.
 102. Steensberg, A., M. A. Febbraio, T. Osada, P. Schjerling, G. van Hall, B. Saltin and B. K. Pedersen (2001). Interleukin-6 production in contracting human skeletal muscle is influenced by pre-exercise muscle glycogen content. *J Physiol-London* 537(2): 633–639.
 103. Steensberg, A., G. van Hall, T. Osada, M. Sacchetti, B. Saltin and B. K. Pedersen (2000). Production of interleukin-6 in contracting human skeletal muscles can account for the exercise-induced increase in plasma interleukin-6. *J Physiol-London* 529(1): 237–242.
 104. Steinacker, J. M. (1988). *Methoden für die Leistungsdiagnostik und Trainingssteuerung im Rudern und ihre Anwendung*. Rudern Berlin. J. Steinacker, Springer: 39–54.
 105. Steinacker, J. M. (1993). Physiological-Aspects of Training in Rowing. *International Journal of Sports Medicine* 14: S3–S10.
 106. Steinacker, J. M., Kellmann, M., Böhm, B.O., Liu, Y., Opitz-Greiss, A., Kallus, K.W., Lehmann, M., Altenburg, D, and Lormes, W (1999). Clinical findings and parameters of stress and regeneration in rowers before world championships. *Overload, Performance Incompetence, and Regeneration in Sport*. M.Lehmann. New York, Kluwer Academic/Plenum Publishers.
 107. Steinacker, J. M., W. Lormes, M. Kellmann, Y. Liu, S. Reissnecker, A. Opitz-Gress, B. Baller, K. Gunther, K. G. Petersen, K. W. Kallus, M. Lehmann and D. Altenburg (2000). Training of junior rowers before world championships. Effects on performance, mood state and selected hormonal and metabolic responses. *J Sport Med Phys Fit* 40(4): 327–335.
 108. Steinacker, J. M., W. Lormes, M. Lehmann and D. Altenburg (1998). Training of rowers before world championships. *Med Sci Sport Exer* 30(7): 1158–1163.
 109. Steinacker, J. M., W. Lormes, S. Reissnecker and Y. Liu (2004). New aspects of the hormone and cytokine response to training. *Eur J Appl Physiol* 91(4): 382–391.
 110. Stewart, L. K., M. G. Flynn, W. W. Campbell, B. A. Craig, J. P. Robinson, K. L. Timmerman, B. K. McFarlin, P. M. Coen and E. Talbert (2007). The influence of exercise training on inflammatory cytokines and C-reactive protein. *Med Sci Sports Exerc* 39(10): 1714–1719.
 111. Zaccaria, M., A. Ermolao, G. S. Roi, P. Englaro, G. Tegon and M. Varnier (2002). Leptin reduction after endurance races differing in duration and energy expenditure. *Eur J Appl Physiol* 87(2): 108–111.

112. Urhausen, A., H. Gabriel and W. Kindermann (1995). Blood Hormones as Markers of Training Stress and Overtraining. *Sports Medicine* 20(4): 251–276.
113. Urhausen, A. and W. Kindermann (2002). Diagnosis of overtraining – What tools do we have? *Sports Medicine* 32(2): 95–102.
114. Urhausen, A., Mueller, M., Foerester, H. J., Weiler, B., Kindermann, W. (1988). Trainingssteuerung im Rudern, *Dtsch Z Sportmed*
115. Vervoorn, C., A. M. Quist, L. J. Vermulst, W. B. Erich, W. R. de Vries and J. H. Thijssen (1991). The behaviour of the plasma free testosterone/cortisol ratio during a season of elite rowing training. *Int J Sports Med* 12(3): 257–263.
116. Viru, A. (1995). Hormonal Functions and Exercise. *Current Therapy in Sports Medicine* (3rd ed.). J. S. Torg, Shepard, R. J.
117. Wren, A. M., L. J. Seal, M. A. Cohen, A. E. Brynes, G. S. Frost, K. G. Murphy, W. S. Dhillon, M. A. Ghatei and S. R. Bloom (2001). Ghrelin enhances appetite and increases food intake in humans. *J Clin Endocrinol Metab* 86(12): 5992.
118. Wren, A. M., C. J. Small, H. L. Ward, K. G. Murphy, C. L. Dakin, S. Taheri, A. R. Kennedy, G. H. Roberts, D. G. Morgan, M. A. Ghatei and S. R. Bloom (2000). The novel hypothalamic peptide ghrelin stimulates food intake and growth hormone secretion. *Endocrinology* 141(11): 4325–4328.

SUMMARY IN ESTONIAN

Valitud vere biokeemiliste stressi ja energia tasakaalu markerite adaptatsioon erinevatele treeningutele hästi treenitud sõudjatel

Sõudmises on aastane treeningmaht suhteliselt suur võrreldes võistlusdistanti läbimiseks kuluva ajaga, ulatudes kuni 1200 tunnini aastas. Samas, tasakaal treeningu ja ületreeningu vahel on suhteliselt habras. Seetõttu on väga oluline õige treeningu- ja puhkeperioodide planeerimine ning süstemaatiline treeningu monitooring. Treeningu monitooring koosneb peamiselt psühholoogilisest ja füsioloogilisest monitooringust ning keha koostise ja saavutusvõimega seotud parameetrite monitooringust. Meeleolu seisund, füsioloogiline stress ja taastumine peegeldavad hästi sportlase kliinilist seisundit ning on tihedalt seotud sportlase tegeliku saavutusvõimega. Kirjanduses on välja pakutud erinevaid puhkeoleku ja treeningu järgseid stresshormoonide reaktsioone, ennetamaks ületreeningut, aga kahjuks on erinevate uuringute tulemused vastuolulised ja päris kindlat arengusuunda veel pole. Samuti ei suuda mõned sportlased suurenenud treeningstressi puhul säilitada piisavat kalorige tarbimist, mistõttu võib tekkida organismis energia puudujääk, mis omakorda suurendab väsimuse kuhjumist veelgi. Seetõttu on välja pakutud treeningu monitooringuks erinevaid peptiidhormoone ja tsütokiine ehk signaalmolekule. Käesoleva töö peamine eesmärk oli uurida sõudjate adaptatsiooni suuremahulise madala intensiivsusega treeninguperioodile. Vastavalt peamisele eesmärgile püstitasime järgmised ülesanded:

1. Uurida puhkeoleku seisundi stresshormoonide (testosteroon ja kortisool) tasemete muutust 4 nädalase suuremahulise treeninguperioodi mõjul.
2. Uurida stresshormoonide koormusjärgseid muutusi 4 nädalase suuremahulise treeninguperioodi mõjul.
3. Uurida puhkeoleku seisundi IL-6, TNF- α , leptiini ja greliini tasemete muutust 4 nädalase suuremahulise treeninguperioodi mõjul.
4. Uurida IL-6, TNF- α , greliini ja leptiini koormusjärgseid muutusi 4 nädalase suuremahulise treeninguperioodi mõjul.
5. Uurida IL-6, TNF- α muutusi maksimaalsel sõudeergomeetritel paranenud saavutusvõime tingimustes.

Käesolev töö põhineb kahel erineval uuringul. Esimene uuring koosnes nelja-nädalasest madala intensiivsusega suuremahulisest treeninguperioodist. Teine uuring kestis aasta ja oli mõeldud uurimaks pikaajalist treeningu adaptatsiooni.

Esimeses uuringus osales 8 hästi treenitud meessõudjat. Uuring koosnes nelja-nädalasest treeninguperioodist, millele eelneval nädalal viidi läbi astmeliste koormustega test. Esimesel treeningunädalal oli treeningute maht 10 tundi, teisel nädalal 15 tundi, kolmandal nädalal ligikaudu 20 tundi, neljas nädal oli

mõeldud taastumiseks ja treeningu maht oli 10 tundi. Uuringu käigus oli kolm testimise sessiooni: 1. pärast esimest nädalat, 2. pärast kolmandat nädalat ja 3. pärast neljandat nädalat. Testimise sessioonile eelnev päev oli puhkepäev ja treeninguid ei toimunud. Iga testimise sessioon koosnes hommikusest puhkeoleku vereproovist, RESTQ-Sport küsimustiku täitmisest, ning 2-tunnisest madala intensiivsusega sõudmise testist. Samuti määrati kolmel korral vaatlusaluste energia tarbimine ning -kulu. Kahetunnise sõudmise testi käigus määrati vaatlusalustel südamelöögisagedus, läbitud distants, vere laktaadisisaldus ning võeti veenivere proovid. Veeniverest määrati järgmised markerid: testosteroon, kortisool, insuliin, IL-6, TNF- α , greliin ja leptiin.

Teises uuringus osales 9 kõrgelt treenitud sõudjat, kõik uuritavad olid rahvusvahelistel võistlustel medalivõitjad. Ettevalmistava perioodi alguses sooritasid vaatlusalused 6000-meetri sõudeergomeetritesti ja täpselt aasta pärast sooritasid nad sama testi uuesti. Vaatlusalustelt võeti veenivere proovid enne testi, kohe pärast testi ja 30 minutit pärast testi lõpetamist. Veeniverest määrati IL-6, TNF- α ja kortisooli väärtused. Testile eelneval päeval määrati ka kehakoostise parameetrid.

Vastavalt töö tulemustele tehti järgmised järeldused:

1. Neljanädalane suuremahuline madala intensiivsusega treeninguperiood ei mõjuta puhkeoleku testosterooni ja kortisooli taset.
2. Testosterooni tase langeb oluliselt pärast kahetunnist sõudmise testi neljanädalase madala intensiivsusega treeninguperioodi mõjul, samas on jälgitav ka kortisooli taseme languse tendents.
3. Neljanädalase madala intensiivsusega treeninguperioodi mõjul langeb puhkeoleku leptiini sisaldus veres.
4. Neljanädalane madala intensiivsusega treeninguperiood alandab koormusjärgset leptiini kontsentratsiooni ning suurendab greliini ja TNF- α kontsentratsiooni veres, samas ei ole muutusi IL-6 kontsentratsioonis.
5. Pikaajaline treeningu adaptatsiooni ja saavutusvõime kasvuga kaasneb suurenenud IL-6 ja TNF- α kontsentratsioon veres pärast maksimaalset ergomeetritesti.

ACKNOWLEDGEMENTS

Especially I would like to thank:

- My academic supervisors researcher Jarek Mäestu and Professor Jaak Jürimäe, for all the advice and support during preparation of the thesis until finishing;
- Professor Toivo Jürimäe for good advice;
- Dr. Kersti Sulg for conducting the medical procedures;
- All the participants, who went through this demanding study;
- My family, understanding and supporting me during my PhD studies.

PUBLICATIONS

CURRICULUM VITAE

Raul Rämson

Date of birth: 3 January 1984

Citizenship: Estonian

Email: rtype@ut.ee

Position held

2004 – ... Academic Sport Club of Tartu University, rowing coach

Education

2007–2011 University of Tartu, PhD

2005–2007 University of Tartu, MSc

2002–2005 University of Tartu, BSc

1990–2002 Tartu Kivilinna Gymnasium

Special Courses

2007 International Summer School for PhD students, Pühajärve, Estonia

2010 Studies at California State University Fullerton, California, USA

Main Research Interests

Coaching sciences – Overtraining – Endocrinology

CURRICULUM VITAE

Raul Rämson

Sünniaeg: 3. jaanuar 1984

Kodakondsus: Eesti

E-post: rtype@ut.ee

Töökogemus

2004 – ... Tartu Ülikooli Akadeemiline Spordiklubi, sõudmise treener

Haridustee

2007–2011 Tartu Ülikool, PhD

2005–2007 Tartu Ülikool, MSc

2002–2005 Tartu Ülikool, BSc

1990–2002 Tartu Kivilinna Gümnaasium

Erialane enesetäiendus

2007 Rahvusvaheline doktorantide suvekool, Pühajärve, Eesti

2010 Öpingud California State University Fullerton, Kalifornias, USA

Peamised uurimisvaldkonnad

Treeninguõpetus – Ületreening – Endokrinoloogia

DISSERTATIONES KINESIOLOGIAE UNIVERSITATIS TARTUENSIS

1. **Lennart Raudsepp.** Physical activity, somatic characteristics, fitness and motor skill development in prepubertal children. Tartu, 1996, 138 p.
2. **Vello Hein.** Joint mobility in trunk forward flexion: methods and evaluation. Tartu, 1998, 107 p.
3. **Leila Oja.** Physical development and school readiness of children in transition from preschool to school. Tartu, 2002, 147 p.
4. **Helena Gapeyeva.** Knee extensor muscle function after arthroscopic partial meniscectomy. Tartu, 2002, 113 p.
5. **Roomet Viira.** Physical activity, ecological system model determinants and physical self-perception profile in early adolescence. Tartu, 2003, 167 p.
6. **Ando Pehme.** Effect of mechanical loading and ageing on myosin heavy chain turnover rate in fast-twitch skeletal muscle. Tartu, 2004, 121 p.
7. **Priit Kaasik.** Composition and turnover of myofibrillar proteins in volume — overtrained and glucocorticoid caused myopathic skeletal muscle. Tartu, 2004, 123 p.
8. **Jarek Mäestu.** The perceived recovery-stress state and selected hormonal markers of training stress in highly trained male rowers. Tartu, 2004, 109 p.
9. **Karin Alev.** Difference between myosin light and heavy chain isoforms patterns in fast- and slow-twitch skeletal muscle: effect of endurance training. Tartu, 2005, 117 p.
10. **Kristjan Kais.** Precompetitive state anxiety, self-confidence and athletic performance in volleyball and basketball players. Tartu, 2005, 99 p.
11. **Aire Leppik.** Changes in anthropometry, somatotype and body composition during puberty: a longitudinal study. Tartu, 2005, 161 p.
12. **Jaan Ereline.** Contractile properties of human skeletal muscles: Association with sports training, fatigue and posttetanic potentiation. Tartu, 2006, 133 p.
13. **Andre Koka.** The role of perceived teacher feedback and perceived learning environment on intrinsic motivation in physical education. Tartu, 2006, 137 p.
14. **Priit Purge.** Performance, mood state and selected hormonal parameters during the rowing season in elite male rowers. Tartu, 2006, 101 p.
15. **Saima Kuu.** Age-related contractile changes in plantarflexor muscles in women: associations with postactivation potentiation and recreational physical activity. Tartu, 2006, 101 p.
16. **Raivo Puhke.** Adaptive changes of myosin isoforms in response to long-term strength training in skeletal muscle of middle-aged persons. Tartu, 2006, 99 p.

17. **Eva-Maria Riso.** The effect of glucocorticoid myopathy, unloading and reloading on the skeletal muscle contractile apparatus and extracellular matrix. Tartu, 2007, 114 p.
18. **Terje Sööt.** Bone mineral values in young females with different physical activity patterns: association with body composition, leg strength and selected hormonal parameters. Tartu, 2007, 94 p.
19. **Karin Tammik.** Neuromuscular function in children with spastic diplegic cerebral palsy. Tartu, 2007, 102 p.
20. **Meeli Saar.** The relationships between anthropometry, physical activity and motor ability in 10–17-year-olds. Tartu, 2008, 96 p.
21. **Triin Pomerants.** Ghrelin concentration in boys at different pubertal stages: relationships with growth factors, bone mineral density and physical activity. Tartu, 2008, 80 p.
22. **Tatjana Kums.** Musculo-skeletal function in young gymnasts: association with training loads and low-back pain. Tartu, 2008, 128 p.
23. **Maret Pihu.** The components of social-cognitive models of motivation in predicting physical activity behaviour among school students. Tartu, 2009, 116 p.
24. **Peep Päll.** Physical activity and motor skill development in children. Tartu, 2009, 102 p.
25. **Milvi Visnapuu.** Relationships of anthropometrical characteristics with basic and specific motor abilities in young handball players. Tartu, 2009, 114 p.
26. **Rita Gruodytė.** Relationships between bone parameters, jumping height and hormonal indices in adolescent female athletes. Tartu, 2010, 82 p.
27. **Ragnar Viir.** The effect of different body positions and of water immersion on the mechanical characteristics of passive skeletal muscle. Tartu, 2010, 142 p.
28. **Iti Määrsepp.** Sensorimotor and social functioning in children with developmental speech and language disorders. Tartu, 2011, 90 p.
29. **Ege Johanson.** Back extensor muscle fatigability and postural control in people with low back pain. Tartu, 2011, 106 p.
30. **Evelin Lätt.** Selected anthropometrical, physiological and biomechanical parameters as predictors of swimming performance in young swimmers. Tartu, 2011, 90 p.