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CONTENTS

LIST OF ABBREVIATIONS

AP	active oral contraceptive pill phase
AT	anaerobic threshold
BMI	body mass index
CHO EE	carbohydrate energy expenditure
CCA	competitive cyclic athletes
EE rate	energy expenditure rate
E/P ratio	estrogen to progesterone ratio
FP	follicular phase
HR	heart rate
HR_{AT}	heart rate at anaerobic threshold
HR_{max}	maximal heart rate
La	blood lactate
Lipid EE	lipid energy expenditure
LP	luteal phase
NAP	not active oral contraceptive pill phase
NC	normally menstruating rowers
OC	oral contraceptive
Pa_{AT}	power at anaerobic threshold
Pa_{max}	maximal power
RCA	recreationally trained cyclic athletes
RER	respiratory exchange ratio
RER_{AT}	respiratory exchange ratio at anaerobic threshold
ROC	recreationally trained athletes taking oral contraceptive pills
Total EE	total energy expenditure
V_E	minute ventilation
$V_{E/AT}$	minute ventilation at anaerobic threshold
V_E/VO_2	ventilatory equivalents for oxygen
V_E/VCO_2	ventilatory equivalents for carbon dioxide
V_E/VO_{2AT}	ventilatory equivalents for oxygen at anaerobic threshold
V_E/VCO_{2AT}	ventilatory equivalents for carbon dioxide at anaerobic threshold
VO_2	oxygen consumption
VO_{2AT}	oxygen consumption at anaerobic threshold
VO_2max	maximal oxygen consumption
Δ La	net increase of blood lactate

LIST OF ORIGINAL PUBLICATIONS

I. **Vaiksaar S**., Jürimäe J., Mäestu J., Purge P., Kalytka S., Shakhlina L., Jürimäe T. No effect of menstrual cycle phase and oral contraceptive use on endurance performance in rowers. *Journal of Strength and Conditioning Research*, 2011; 25(6): 1571–1578.

II. **Vaiksaar S**., Jürimäe J., Mäestu J., Purge P., Kalytka S., Shakhlina L, Jürimäe T. No effect of menstrual cycle phase on fuel oxidation during exercise in rowers. *European Journal of Applied Physiology*, 2011; 111(6): 1027–1034.

III. **Vaiksaar S**., Jürimäe J., Mäestu J., Purge P., Kalytka S., Shakhlina L., Jürimäe T. Monophasic oral contraceptive cycle phase and endurance capacity of rowers. *Perceptual and Motor Skills*, 2011; 113(3): 764–72.

Paper I, II and III, Sille Vaiksaar had primary responsibility for protocol development, subjects' enrollment, performing measurements, preliminary and final data analyses, and writing of the manuscripts.

1. INTRODUCTION

The female hormones estrogen and progesterone fluctuate predictably across the menstrual cycle in normally cycling eumenorrheic women. In addition to the reproductive function, these hormones have been reported to influence other physiological systems, including their action during endurance exercise that may have implications for endurance performance (Oosthuyse and Bosch, 2010, 2012). Some studies have found variations in endurance performance between menstrual cycle phases, showing better performance in the follicular phase (FP) (Campbell et al., 2001), while others have found a marked improvement (Jurkowski et al., 1981) or a trend for improvement (Nicklas et al., 1989; Oosthuyse et al., 2005) during the luteal phase (LP). In contrast, there are studies that have observed no effect of menstrual cycle phase on endurance performance parameters (Bailey et al., 2000; Dombovy et al., 1987; McLay et al., 2007; Shaharudin et al., 2011). The lack of consensus in different menstrual cycle phase studies may in part be due to a small subject number in different studies and numerous methodological issues in determining specific menstrual cycle phase, nutritional status, performance test selection and performance level of studied women (Constantini et al., 2005; Lebrun, 1994; Oosthuyse and Bosch, 2010; Redman and Weatherby, 2004). Furthermore, the findings of menstrual phase studies may be confounded by the high variability in the concentrations of ovarian hormones between subjects and from day to day variation within subjects during any particular menstrual cycle phase (Oosthuyse and Bosch, 2010; Rechichi et al., 2009). Female athletes, however, train and compete in their specific sporting events at all stages of the menstrual cycle. All above-mentioned factors make it difficult to draw definite conclusions on the possible effects of normal menstrual cycle phases on training and performance parameters in female athletes in sport-specific conditions.

The use of oral contraceptives (OC) may provide a more stable environment to evaluate the possible effect of reproductive hormones on physiological variables and exercise performance in women (Rechichi et al., 2009). In addition, the use of OC pills may be advantageous for female athletes who are negatively affected by their menstrual cycle, as OC pills may provide a stable yet controllable hormonal milieau for training and performance (Constantini et al., 2005). Accordingly, it is apparent that OC use is becoming more prevalent in female athletes (Rechichi and Dawson, 2009; Rechichi et al., 2008). However, up to date, OC users have not been included in the majority of menstrual cycle research and limited information is available regarding the influence of the OC use on athletic performance in female athletes (Lebrun et al., 2003; Rechichi et al., 2009). Furthermore, most of the OC research has compared differences between OC use and non-use, while few researchers have studied the within-cycle effects of the OC use on athletic performance (Rechichi et al., 2009). There are some studies to suggest that endurance performance is not affected by changes from active pill (AP) to non-active pill (NAP) phase of the OC cycle

(Lynch et al., 2001; Rechichi et al., 2008), while there is also a potential for variation in endurance performance throughout an OC cycle (Rechichi et al., 2009). The exogenous hormones contained in AP phase during OC cycle suppress endogenous estrogen and progesterone levels (Fotherby, 1996), which in turn may influence substrate metabolism during endurance performance in athletes (Lebrun et al., 2003; Rechichi et al., 2008). Accordingly, it is important to study training and performance parameters in female athletes during different phases of the OC cycle in sport-specific conditions.

Rowing is a strength-endurance type of sport and during a rowing race aerobic capacities of the athletes are stressed to their maximum (Steinacker, 1993). Accordingly, submaximal (anaerobic threshold [AT]) and maximal (maximal oxygen consumption [VO_2max]) aerobic metabolism values are the most important physiological determinants of rowing performance over 2000 metre distance (Jürimäe, 2008; Mäestu et al., 2005). Rowers utilize relatively large body mass where all extremities and trunk muscles are involved compared to other endurance sports, which results in higher energy consumption during exercise (Jürimäe, 2008; Mäestu et al., 2005). In addition, female rowers present higher body fat values in comparison with athletes in other endurance events (Mäestu et al., 2005). Large amounts of rowing training are performed as low-intensity long-distance exercise sessions (Mäestu et al., 2005) and rowing exercise at the intensity of 70 % VO_2max closely approximates a rower´s typical training session (Jürimäe et al., 2001, 2009). To our best knowledge, there are no studies performed, which have investigated the effect of menstrual cycle phase and OC use on different aerobic performance parameters and also on substrate oxidation and lactate concentration during low-intensity long-distance exercise in sport specific conditions in female rowers. To date, only one study has investigated the effect of menstrual cycle phase on AT (Forsyth and Reilly, 2005), and VO_2max and 2000 metre time trial (Forsyth and Reilly, 2008) parameters in female rowers. Accordingly, the purpose of the present dissertation was to investigate whether different endurance performance and training parameters are affected by acute hormonal fluctuations throughout the normal menstrual cycle and the synthetic menstrual cycle of the OC users in well trained and recreationally trained rowers. The obtained results could assist female rowers to make more informed decisions about the possible effects of menstrual cycle phase and OC use on sport-specific endurance performance and everyday training parameters.

2. REVIEW OF LITERATURE

2.1. Studies investigating physiological responses to exercise in normally menstruating women

Women experience a circamensal rhythm termed the menstrual cycle, where the ovarian hormones fluctuate during the menstrual cycle (Oosthyse and Bosh, 2010; Reilly, 2000). An average length of an ovulatory menstrual cycle is 28 days and the menstrual cycle is typically divided into two phases: the follicular (proliferative) phase (FP; days 1–14) and the luteal (secretory) phase (LP; days 15–28) (Harber, 2004; Loucks and Horvath, 1985). These major phases are separated by the event of ovulation. Normal menstrual cycle length in eumenorrheic women is between 25 to 38 days (Harber, 2004; Oosthyse and Bosh, 2010) and if the menstrual cycle interval becomes shorter or longer, the shift in length occurs in the FP of the menstrual cycle (Harber, 2004; Loucks and Horvath, 1985). The female hormones estrogen and progesterone are primarily secreted from the ovaries and to a lesser extent from the adrenal glands in women (Lebrun, 1994) and fluctuate predictably across the menstrual cycle in normally cycling eumenorrheic women (Harber, 2004; Oosthyse and Bosh, 2010). These hormone concentrations are relatively low during the FP of the menstrual cycle. While at the end of the FP of the menstrual cycle, just before ovulation occurs, a sharp increase in estrogen occurs for a short period of time and at the beginning of the LP estrogen decreases to the previous level. A gradual increase followed by a gradual decrease in estrogen and progresterone is seen during the LP of the menstrual cycle (Harber, 2004; Oosthyse and Bosh, 2010). Although estrogen and progesterone primarily function to support reproduction, they have also been reported to influence other physiological systems of the female organism (Lebrun, 1994; Oosthyse and Bosh, 2010). However, it has to be taken into account that women train and compete in different sporting events during all stages of the menstrual cycle (Harber, 2004; Oosthyse and Bosh, 2010).

There are different studies that have been conducted to assess physiological responses of different exercises with different duration in various levels of female athletes (Brun et al., 2011; Jürimäe and Jürimäe, 2004; Kendall et al., 2012; Roepstorff et al., 2002). In these studies, women have been investigated mostly only once during early stages of their menstrual cycle, when the circulating estrogen and progesterone concentrations are considered to be lowest (Jürimäe and Jürimäe, 2004; Oosthyse and Bosh, 2010). While the menstrual cycle has broadly been divided into two distinct phases (FP and LP), most of the researchers to date have endeavoured to compare physiological responses to different exercises with various intensities in women between these identified phases of the menstrual cycle (Bishop, 1997; Campbell et al., 2001; Devries et al., 2006; Dombovy et al., 1987; Middleton and Wenger, 2006; Shaharudin et al., 2011; Sunderland and Nevill, 2003). Accordingly, majority of previous studies evaluating the influence of menstrual cycle phase on exercise performance parameters have usually made comparisons between the early or mid-follicular phase (when estrogen and progesterone levels are low) and the mid-luteal phase (when estrogen and progesterone levels are elevated) (Dean et al., 2003; De Souza et al., 1990; Forsyth & Reilly, 2005,2008; Giacomoni et al., 2000; Lebrun et al, 1995; Nicklas et al., 1989; Shaharudin et al., 2011; Zderic et al., 2001). However, there are also few studies that have included the period just before ovulation (i.e., the late FP), when estrogen is its highest and progesterone concentration is low to observe possible differences in physiological parameters in response to exercise performance during the menstrual cycle (Bemben et al., 1995; Oosthuyse et al., 2005; Tsampoukos et al., 2010**).** It has been suggested that the combination of the low progesterone and very high estrogen that characterise this late FP of the menstrual cycle could result in estrogen promoting performance without the anti-estrogenic effects of progesterone as may occur during the LP of the menstrual cycle (Oosthuyse et al., 2005). In addition, the estrogen to progesterone ratio (E/P ratio) during the LP may be an important determinant of the overall impact of the ovarian hormone milieu on exercise performance during this menstrual cycle phase (Bailey et al., 2000; D`Eon et al., 2002; Jurkowski et al., 1981; Nicklas et al., 1989; Oosthuyse et al., 2005). It has been demonstrated that studies reporting a better exercise performance during the LP had also higher E/P ratio, while the studies that found no change in performance parameters had a lower E/P ratio (Oosthyse and Bosh, 2010).

In summary, to date, most studies have used only two distinct phases of the menstrual cycle (FP and LP) to investigate possible menstrual phase-associated differences in exercise performance. At present, the findings of menstrual phase investigations may be confounded by the high variability in the circulating estrogen and progesterone concentrations between exercising females and from day to day within subjects during any particular phase of the menstrual cycle (Oosthyse and Bosh, 2010).

2.2. Effect of the menstrual cycle phase on aerobic performance and exercise metabolism in normally menstruating women

Although the potential impact of the menstrual cycle phase on aerobic performance has been the focus of several investigations (Campbell et al., 2001; De Souza et al., 1990; Dean et al., 2003; Forsyth and Reilly, 2005; Galliven et al., 1997; Ives et al., 2011; Lebrun et al., 1995; Oosthuyse et al., 2005; Redman et al., 2003; Shaharudin et al., 2011; Smekal et al., 2007; Zderic et al., 2001), many findings remain largely inconclusive probably due to the small subject numbers, subject selection, high intra- and inter-individual variability in estrogen and progesterone concentrations, and the pulsatile secretion of these hormones (Rechichi et al., 2009). Furthermore, not all studies have verified specific menstrual cycle phase by the concomitant analysis of ovarian hormones and discrepancies in the results could also be caused by the inappropriate determination of the menstrual cycle phase (Forsyth and Reilly, 2005).

Maximal oxygen consumption determined in different test protocols on a treadmill (Bemben et al., 1995; Lebrun et al., 1995), on a cycle ergometer (Dean et al., 2003; Shaharudin et al., 2011; Smekal et al., 2007; Zderic et al., 2001) and also on a rowing ergometer (Forsyth and Reilly, 2005, 2008) have produced only limited evidence of the influence of different menstrual cycle phases on VO_2max values in exercising women. Forsyth and Reilly (2008) found that VO_2max during a continuous, 3 min incremental test to exhaustion on a rowing ergometer was significantly higher in the LP than in the FP of the menstrual cycle in recreationally active female rowers (2.64±0.61 vs 2.92± 0.57 l/min; $p<0.05$). In contrast, Lebrun et al. (1995) reported that VO_2max on a treadmill was slightly but significantly ($p<0.05$) lower in LP versus FP of the menstrual cycle when using the absolute VO_2max values (3.19±0.09 vs 3.13± 0.08 l/min) for comparison in a group of recreationally active female athletes. When converting VO_2max to body mass, this significant difference between the menstrual cycle phases disappeared (FP: 53.7±0.9 ml/min/kg; LP: 52.8± 0.8 ml/min/kg; $p>0.05$) (Lebrun et al., 1995). However, it has repeatedly been shown that VO_2max and time to exhaustion in maximal ramp tests were mostly unchanged by the menstrual cycle phase in women of various performance levels (Bemben et al., 1995; Beidleman et al., 1999; Casazza et al., 2002; Dean et al., 2003; De Souza et al., 1990; Dombovy et al., 1987; Jurkowski et al., 1981; Lebrun et al., 1995; Redman et al., 2003; Smekal et al., 2007).

Similarly, the research literature appears to be fairly consistent from different curve-fitting methods that the exercise intensity that induces the point of inflection corresponding to the AT remains unchanged by the menstrual cycle phase in exercising women (Dean et al., 2003; Dombovy et al., 1987; Schoene at al., 1981; Smekal et al., 2007). However, Forsyth and Reilly (2005) found that menstrual cycle phase appeared to affect the intensity that corresponded to 4 mmol/l AT on a rowing ergometer. In the LP of the menstrual cycle, AT values corresponding to the intensity of 4 mmol/l occurred at a significantly higher exercise intensity compared to the FP of the menstrual cycle in female rowers (Forsyth and Reilly, 2005). However, when data were re-examined using a log-log transformation method to determine AT, no significant differences were observed for any of the measured variables at the intensity of AT (power output, heart rate, VO_2) between FP and LP phases of the menstrual cycle (Forsyth and Reilly, 2005). In another study, Bemben et al. (1995) found that AT occurs at a significantly higher percentage of VO_2max in the early FP compared with the late FP and LP of the menstrual cycle on a treadmill exercise.

The relative utilization of carbohydrates and lipids as fuel sources during aerobic exercise can be influenced by factors such as exercise intensity (Goedecke et al., 2000; Venables et al., 2005), training status (Jeukendrup et al., 1997), diet (Casazza et al., 2004; Goedecke et al., 2000) and also a relative hormonal mileau during exercise (Casazza et al., 2004; Devries et al., 2006; Horton et al., 1998). Some studies have demonstrated that females, compared to males, utilize less carbohydrate to fuel endurance exercise, as evidenced by a lower respiratory exchange ratio (RER) (Carter et al., 2001; Horton et al., 1998; Tarnapolsky et al., 1995; Venables et al., 2005), while other studies have shown a similar relative utilization of carbohydrates and lipids in females and males exercising at the same relative intensity (Marliss et al., 2000; Roepstorff et al., 2002; Romijn et al., 2000). It has been suggested that possible gender differences in the relative fuel utilization during endurance exercise are due to differences in circulating estrogen (Bunt, 1990; Devries et al., 2006).

Estrogen may promote aerobic exercise metabolism by altering carbohydrate and lipid oxidation with progesterone often appearing to act antagonistically (Oosthuyse and Bosch, 2010; 2012). Specifically, Oosthuyse and Bosch (2010) recently suggested that the magnitude of the increase in the ovarian hormones between menstrual cycle phases and the E/P ratio could be an important factor determining the effect on substrate metabolism. It has been argued that relatively high progesterone concentration during the LP of the menstrual cycle may have countered the benefits of an elevated estrogen concentration during endurance exercise (McLay et al., 2007; Oosthuyse et al., 2005). Accordingly, several studies report that there appears to be only small difference in substrate metabolism during prolonged exercise due to the endogenous ovarian hormone fluctuations across the normal menstrual cycle (Campbell et al., 2001; Casazza et al., 2004; Devries et al., 2006; McLay et al., 2007; Oosthuyse et al., 2005; Suh et al., 2002). There are reports of no significant differences in RER (De Souza et al., 1990; Horton et al., 2002; Suh et al., 2002), blood glucose (Devries et al., 2006; Horton et al., 2002; Suh et al., 2002) or blood lactate (De Souza et al., 1990; Devries et al., 2006; Horton et al., 2002; Nicklas et al., 1989) concentrations due to the ovarian hormone variations during the menstrual cycle. In contrast, certain data suggest that there may be a greater carbohydrate oxidation and a lower lipid oxidation during submaximal exercise (<70% of VO_2max) in the FP compared to the LP of the menstrual cycle (Campbell et al., 2001; Hackney, 1999; Hackney et al., 1991, 1994; Wenz et al., 1997; Zderic et al., 2001). However, most of the studies have been performed in women with high habitual physical activity (Casazza et al., 2004; Devries et al., 2006; Hackney, 1999; Hackney et al., 1991,1994; Horton et al., 2002; Suh et al., 2002; Zderic et al., 2001) and only very few with highly trained women (Campbell et al., 2001). Accordingly, further clarification of the effect of the menstrual cycle on substrate utilization during exercise in relation to exercise intensity and training status in exercising women is needed.

In summary, most of the research performed on the possible effects of different menstrual cycle phases on aerobic performance and exercise metabolism in normally menstruating women has been done with habitually physically active women and very few with athletes. To our best of knowledge, only Forsyth and Reilly (2005, 2008) have used recreationally active rowers. Taken together, the menstrual cycle phase has only occasionally been found to influence aerobic performance and substrate oxidation in normally menstruating women. However, further studies are needed before any conclusions can be drawn.

2.3. Effect of the oral contraceptive use on aerobic performance and exercise metabolism

As previously stated, a number of physiological variables during aerobic performance have been reported to vary with fluctuations in female hormones observed throughout a normal menstrual cycle (Campbell et al., 2001; Forsyth and Reilly, 2008; Lebrun, 1993; Smekal et al., 2007). However, the wide inter- and intra-individual variation in circulating estrogen and progesterone concentrations within a natural menstrual cycle and varying lengths of the menstrual cycle between exercising women may make valid interpretations of some investigations difficult (Janse De Jonge, 2003; Redman and Weatherby, 2004; Rechichi et al., 2009). For this reason, the OC cycle may provide a more stable environment to assess the effect of ovarian hormones on physiological variables in women (Rechichi, 2009). Although OC pills are mainly used for birth control in normally menstruating young women, these pills have also frequently used for medical treatment like eating disorders, menstrual cycle regulation and also amenorrhea in female athletes (Lebrun et al., 2003; Miller and Klibanski 1999; Rickenlund et al., 2004). Accordingly, it is apparent that OC use is becoming more prevalent in athletes. For example, only 5 to 12% of athletes were using an OC in the early 1980s, while nowadays one survey reported that about 83% of elite level female athletes were taking OC pills (98% monophasic and 2% triphasic) (Rechichi et al., 2009).

Oral contraceptive pills suppress endogeneous production of estrogen and progesterone, which prevents the midcycle surge of gonadotrophins, inhibiting ovulation and subsequent pregnancy (Fotherby, 1996; Krug et al., 1994; Rechichi et al., 2009; Redman and Weatherby, 2004). Monophasic OC use involves a constant dose of synthethic estrogen and progesterone over 21 days (AP phase, days 1–21) during which endogenous ovarian hormones are suppressed and general hormonal milieu is stable, followed by seven day of placebo pills [NAP phase, days 22–28) (Rechichi et al., 2009). However, during the first days of NAP phase both endogenous estrogen and progesterone may continue to be suppressed (Rechichi et al., 2008). It has been reported that synthetic estrogen is detectable for up to two days after discontinuation, while some progestogens are detectable for up to five days (Rechichi et al., 2009). While a triphasic OC pills contain a changing dosage of both estrogen and progesterone throughout three phases of the 21-day AP phase followed by seven day NAP phase (Redman and Weatherby, 2004).

Despite the widespread use of OC pills among female athletes, OC users have not been included in the majority of menstrual cycle research and only limited information is available regarding the influence of OC use on athletic performance in female athletes (Burrows and Peters, 2007; Bushman et al., 2006; Lynch et al., 2001; Rechichi et al., 2007, 2008; Redman and Weatherby, 2004). Furthermore, most of the OC research has compared differences between OC use and non-use (Casazza et al., 2004, Lebrun et al., 2003; Rickenlund et al., 2004), while few researchers have studied the within-cycle effects of the OC use on athletic performance (Rechichi et al., 2009). Therefore, only few studies have used monophasic OC pills investigating the within-cycle effects of OC use on athletic performance (Bushman et al., 2006; Giacomoni and Falgairette, 2000; Lynch et al., 2001; Rechichi et al., 2007, 2008; Rechichi and Dawson, 2012; Sunderland et al., 2011). Studies that have focused on the differences between non-OC use followed by the OC use have mainly focused on evaluating body composition during OC treatment showing no significant change in body mass or body fat mass (Loyd et al., 2002; Reubinoff et al., 1995; Rickelund et al., 2004). There are also few studies that have examined the effects of OC treatment on different indicators of athletic performance like VO_2max, intermittent exercise performance and anaerobic power values (Bryner et al., 1996; Bushman et al., 2006; Casazza et al., 2002; Lebrun et al., 2003; Lynch and Nimmo, 1998; Lynch et al., 2001; Tegnalia et al., 1999).

Most studies that have investigated aerobic exercise parameters within an OC cycle, have found no variation as a result of acute hormonal change in untrained (Bryner et al., 1996; De Bruyn-Prevost et al., 1984; Lynch et al., 2001; Sunderland et al., 2011) and trained (Rechichi et al., 2008; Rechichi and Dawson, 2012; Reilly and Whitley, 1994) women on OC pills. For example, 13 female athletes performed a 1-h maximal cycle ergometer test at three time points of OC cycle (Rechichi et al., 2008). Testing times were during the AP phase, early NAP phase and late NAP phase (Rechichi et al., 2008). No significant differences were present between the testing times for mean aerobic performance parameters, while significantly increased ventilatory response was seen during the AP phase compared to two NAP phase measurement times (Rechichi et al., 2008). It has to be taken into account that the results of some investigations are limited (Bryner et al., 1996; Lynch et al., 2001), because the testing was conducted at both times during the AP pill phase of the OC cycle, when progestogen intake was similar (Rechichi et al., 2009). In contrast, Giacomoni and Falgairette (2000) demonstrated significant variation in VO_2 across three submaximal treadmill exercise bouts throughout an OC cycle. Specifically, submaximal VO_2 was 3.0 to 5.8% lower in the AP phase of the OC cycle compared to NAP phase in untrained women. In addition, running economy (ml O_2/kg/km) was significantly improved when women were on late AP phase compared to NAP phase regardless of the treadmill running test intensity stage. Taken together, the results are not clear and future studies are warranted to determine whether OC cycle has some effects on aerobic performance parameters during maximal and submaximal tests in women with different aerobic performance levels.

While some studies have reported significant variations in blood lactate concentrations (Rechichi et al., 2008; Rechichi and Dawson, 2012; Redman and Weatherby, 2004), other investigations have found no differences in circulating blood lactate as a result of exercise throughout an OC cycle (Bernandes and Radomski, 1998; Bonen et al., 1991). For example, Rechichi et al. (2008) found that blood lactate concentration was significantly higher after 1-h cycling time trial test in AP phase (6.2±2.7 mmol/l) compared to early NAP phase (5.1± 1.9 mmol/l) in trained female athletes. However, it was suggested that as there was no corresponding difference in blood glucose concentrations, it was difficult to attribute the variation in the lactate result to shifts in substrate metabolism (Rechichi et al., 2008). It has been argued that endogeneous sex hormones may have secondary effects on substrate metabolism as estrogen has been linked to increased fat and reduced carbohydrate oxidation during prolonged exercise, while progesterone may oppose the lipolytic effects of estrogen (D'Eon, et al., 2002). Accordingly, OC use has been reported to increase fat oxidation during exercise (Bemben et al., 1992; Casazza et al., 2004), while other studies have demonstrated no difference in substrate metabolism (Suh et al., 2003). However, it has to be taken into account that in addition to OC use, differences in test protocols (e.g., exercise mode, duration and intensity) may also influence the relative oxidation of carbohydrates and fats as fuel sources during aerobic exercise (Goedecke et al., 2000; Venables et al., 2005).

In summary, OC use is becoming more prevalent in female athletes. However, to date, most of the research has compared differences between OC use and non-use, while very few studies have examined within-cycle effects of the OC use. Future studies are needed to specify the effects of the different exogeneous and endogeneous hormonal profiles on athletic performance.

3. AIMS OF THE STUDY

There are investigations to suggest variations in physiological responses and athletic performance during testing and training for female athletes at specific phases of the menstrual cycle. However, to our best knowledge, there are no studies performed, which have investigated the effect of menstrual cycle and oral contraceptive use on aerobic performance and training parameters in sport-specific conditions in female rowers. Accordingly, the general aim of the present study was to investigate the effects of the normal menstrual cycle and the synthetic menstrual cycle of the oral contraceptive users on aerobic performance and training parameters in female rowers.

The specific aims of the present study were to:

1. investigate the effects of menstrual cycle phases on submaximal and maximal aerobic performance characteristics in normally menstruating rowers and in rowers taking oral contraceptive pills (Paper I);
2. investigate the effects of menstrual cycle phases on substrate oxidation and lactate concentration during prolonged aerobic exercise in normally menstruating rowers (Paper II);
3. investigate the effects of menstrual cycle phases on substrate oxidation and lactate concentration during prolonged aerobic exercise in rowers taking oral contraceptive pills (Paper III).

4. MATERIAL AND METHODS

4.1. Participants

In total, 24 eumenorrheic female rowers voluntarily took part in this study. All participants were trained rowers or used to be trained rowers, now being only recreationally active rowers. Only women who reported a regular menstrual cycle during the last six months and women taking OC pills minimum three months preceding the tests were recruited in the study. In the first study, where rowers were asked to perform incremental exercise test on a rowing ergometer at different phases of the menstrual cycle, they were distinguished on the basis of both menstrual status and athleticism. Eight competitive cyclic athletes (CCA), seven recreationally trained cyclic athletes (RCA) and nine recreationally trained athletes taking OC pills participated in the first study (Paper I). The athletes in the CCA group were defined as athletes who competed successfully at the national and international level, whereas recreationally trained athletes exercised regularly but not at this competitive level (Thong et al., 2000). In the other studies, normally cycling (n=11) (Paper II) and OC using (n=8) (Paper III) rowers completed 1-h rowing ergometer exercise at 70% of VO_2max during different phases of the menstrual cycle (Figure 1).

All participants were free of injuries and diseases, and were not taking any medication as determined by health history questionnaire. The cyclic athletes without OC pills (NC) were required to have menstrual cycle duration of 24–35 days, with at least six months of documented menstrual cycles, and were not using the OC pills for at least six months preceding the study (Casazza et al., 2004; Dean et al., 2003; Smekal et al., 2007). Participants whose menstrual cycle occurred later than 35 days were excluded (Thong et al., 2000). The rowers in the OC group were required to be taking OC pills for at least three months preceding the study (Lebrun et al., 2003; Rechichi, et al., 2008). A monophasic OC pill cycle consisted of 21 days of AP phase containing a constant concentration of estrogen and progesterone, and seven days of NAP phase containing of placebo pills. Rowers used a common low-dose monophasic OC pills that contained of 20 μg ethinylestradiol and 75 μg gestodene.

The study was conducted during the preparatory period for the competitive rowing season. The main goal of training during the preparatory period was to increase aerobic base through aerobic extensive endurance training sessions. The training intensity was below AT for approximately 90% of the entire training time (Jürimäe et al., 2000, 2006; Mäestu et al., 2005). The study design, purpose, and possible risks were explained to the participants and written informed consent was obtained from the participants before the investigation. The study protocol was approved by the Medical Ethics Committee of the University of Tartu.

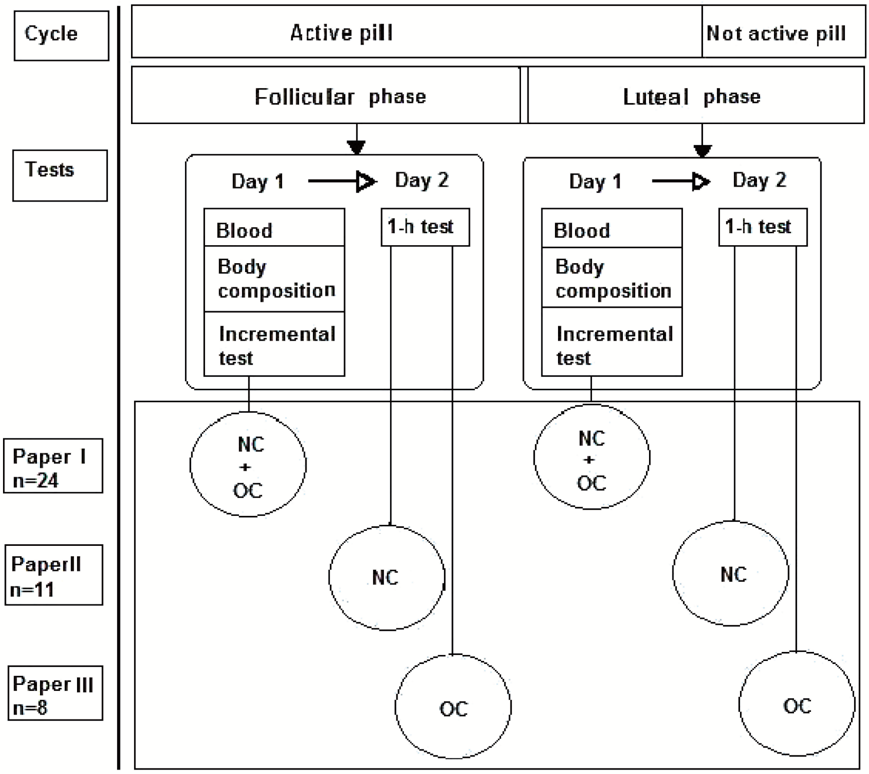


Figure 1. The schematic view of the study and Papers I, II and III. NC- normally menstruating rowers, OC- rowers using oral contraceptives, Body composition- anthropometric and body composition measurements; Blood- fasting blood samples.

4.2. Test procedures

Rowers completed two experimental testing sessions during the FP (determined as days 7–11 from the onset of menstruation) and during the LP (determined as days 18–22 from the onset of menstruation) of the menstrual cycle (Casazza et al., 2004; Dombovy et al., 1987; Horton et al., 2002; Ives et al., 2011; Smekal et al., 2007; Suh et al., 2002; Timmons et al., 2005). For OC users, two experimental testing sessions were carried out during the AP phase (days 7–11) and during the NAP phase (days 22–24) of the OC pill cycle (Casazza et al., 2004; Devries et al., 2006; Timmons et al., 2005) (Figure 1). Test order was balanced with the respect to the cycle phase and test time was standardized between 4.00 and 6.00 p.m. On the day before exercise tests, no physical activities were allowed (Jürimäe et al., 2000; Smekal et al., 2007). Two identical testing sessions were conducted in both menstrual cycle and OC cycle phases. The first testing session included incremental rowing ergometer test that was followed by body composition measurements (Paper I). One hour endurance rowing ergometer session was conducted on the following day after the incremental rowing ergometer test (Papers II and III) (Figure 1). The participants were in a post-absorptive state having eaten a meal for about 2 h or more before each test (Jürimäe et al., 2000). Over the testing period, participants were asked to maintain a regular and constant volume and intensity of training and were asked to standardize their dietary intake within the 24 h prior to each test (Smekal et al., 2007), but there were no dictations for menus. Participants recorded the meal before the first test and they ate the similar meal before the second test to reach nearly identical nutritional intake (Smekal et al., 2007).

The information about previous menstrual cycles was used to identify the phases of the menstrual cycle (Dean et al., 2003; Smekal et al., 2007). The length of the menstrual cycle was calculated from the first day of menses to the day preceding the next menses (Dean et al., 2003; Smekal et al., 2007). Menstrual cycle phases were later confirmed by estradiol and progesterone concentrations from the fasting blood samples (Hackney, 1999; Nicklas et al., 1989; Rickenlund et al., 2004; Suh et al., 2002). The accepted concentration ranges for the ovarian hormones during both menstrual cycle phases for normally menstruating rowers were: 85–220 pmol/l for estradiol and < 3 nmol/l for progesterone during the FP, and 230–750 pmol/l for estradiol and > 16 nmol/l for progesterone during the LP. Therefore, the resting level of progesterone higher than 16 nmol/l was required to confirm LP in normally menstruating rowers (Landgren et al., 1980). As it has been suggested that estradiol must differ at least by 2-fold between menstrual cycle phases in order to produce a significant impact on substrate metabolism during endurance exercise (D´Eon et al., 2002), a 2-fold increase in estradiol concentration was also required for the inclusion of the participant´s data in the analysis (Oosthuyse et al., 2005). Estradiol and progesterone concentrations were determined in duplicate on Immulite 2000 (DPC, Los Angeles, CA, USA). The intra- and interassay coefficients of variation (CVs) for estradiol were 5.3% and 6.5%, and for progesterone 5.4% and 3.4%, respectively.

4.3. Anthropometric and body composition measurements

The height (Martin Metal Anthropometer) and body mass (A&D Instruments Ltd, Oxfordshire, UK) of the participants were measured to the nearest 0.1 cm and 0.05 kg, respectively. Body composition was measured using dual-energy X-ray absorptiometry. Scans of the whole body were performed on each of the subjects using a Lunar DPX-IQ densitometer (Lunar Corporation, Madison, WI, USA) and analysed for fat (FM) and fat free (FFM) masses. During the scanning, participants were lying on their back with their arms at their sides wearing light clothes only. The CVs for body composition measurements were less than 2%.

4.4. Incremental rowing ergometer test

A stepwise rowing ergometer test was performed on a wind resistance-braked rowing ergometer (Concept II; Morrisville, VT; USA) to determine maximal and submaximal aerobic performance parameters, and also target heart rate (HR) values for 1-h endurance rowing ergometer test. The rowers were fully familiarized with the use of the apparatus. Participants were equipped with instruments to measure HR and respiratory gas exchange variables and sat quietly for 1 min on the ergometer before starting to exercise at 40 W. Load was increased by 15 W every minute until maximal voluntary exhaustion was reached. Power and stroke rate were recorded continuously on the computer display of the rowing ergometer. The test was designed to reach the maximum in approximately 15 min in each participant (Hofmann et al., 2007). Subjects were strongly encouraged to achieve maximal performance. Capillary blood samples for enzymatic determination of lactate (Lange, Germany) were collected before and 3, 5 and 7 min post-exercise. The net increase of blood lactate (Δ La) was obtained by subtracting the pre-trial value from the peak value attained during the recovery phase.

Heart rate (HR) was recorded at every 5 s during the test using Sporttester Polar 725X (Polar Electro Oy, Kempele, Finland). Respiratory gas exchange variables were measured throughout the test in a breath-by-breath mode using a portable open circuit spirometry system (MetaMax 3B, Cortex Biophysic GmbH, Germany) and data were stored in 10 s intervals. Oxygen consumption (VO_2), carbon dioxide production (VCO_2) and minute ventilation (V_E) were continuously measured. The mean respiratory exchange ratio (RER), ventilatory equivalents of O_2 (V_E/VO_2) and CO_2 (V_E/VCO_2) were calculated from the recorded VO_2, VCO_2 and V_E measurements. The analyzer was calibrated before each test with gases of known concentrations according to the manufacturer´s guidelines. All data were processed by means of computer analysis using standard software (MetaSoft, Cortex Biophysic GmbH, Germany) and the system for HR analysis. To establish that VO_2max was reached, the attainment of a plateau in VO_2 with increasing work rate was used as a criterion. When this plateau in VO_2 was not observed, a RER exceeding 1.1 and theoretical maximal cardiac frequency were used as a criterion (Dean et al., 2003; Zderic et al., 2001).

Anaerobic threshold determination was performed using linear regression turn point analysis (Hofmann et al., 2007). Turn points in HR, V_E, V_E/VO_2 and V_E/VCO_2 were calculated as described previously (Hofmann et al., 2007). The HR turn point was defined as the deflection of the HR performance curve at approximately 90% of maximal HR (Hofmann et al., 2007). Two regression lines were calculated and the intersection point between both optimized regression lines was termed as the HR turn point and was used in the AT analysis (Hofmann et al., 2007). The corresponding power at AT (Pa_{AT}) was determined using the extrapolation method. The suggested method has been found to be reliable in determining the individual intensity for aerobic-anaerobic transition in rowers (Hofmann et al., 2007).

4.5. Endurance exercise protocol

The exercise test consisted of rowing on a rowing ergometer for 1-h at the intensity of 70% VO_2max. Target HR was set at the level obtained from the incremental test using a practical set ±2 bpm of 70% VO_2max (Jürimäe et al., 2001, 2009). Rowers were asked to increase exercise intensity smoothly and the target HR was achieved after the first 5 min. The participants were instructed to maintain the target HR steady state for the entire exercise session and to reduce exercise intensity to accommodate the required HR steady state as needed (Jürimäe et al., 2001, 2009). Respiratory gas exchange variables were measured throughout the test in a breath-by-breath mode using a portable open circuit spirometry system (MetaMax I, Cortex, Germany) for 5 min at rest and during the 1-h exercise session as described above. One hour exercise session was stopped for 1 min blood lactate measurements after each 15 min of exercise. Respiratory gas exchange measurements were computed at rest and for 10 min of each 15 min of exercise period after the first 5 min of stabilisation period to exclude the impact of 1 min stop (Goedecke et al., 2000). Fat and carbohydrate oxidation and total energy expenditure were estimated from the RER using stoichiometric equations (Frayn, 1983), with the assumption that urinary nitrogen excretion rate was neglible (Venables et al., 2005). These equations have previously been used in females to assess submaximal exercise substrate oxidation, during which the RER was < 1 (Devries et al., 2006; Roepstorff et al., 2002; Suh et al., 2002; Venables et al., 2005). Capillary blood samples for enzymatic determination of lactate (Lange, Germany) were collected before, and after 15, 30, 45 and 60 min of the start of exercise.

4.6. Statistical analyses

Means and standard deviations (±SD) were determined. Analyses of variance and LSD *post hoc* tests, or paired t-tests were used to evaluate differences between measured variables. The level of significance was set at $p<0.05$.

5. RESULTS

5.1. Submaximal and maximal aerobic performance characteristics in normally menstruating rowers and in rowers taking oral contraceptive pills (Paper I)

Mean (±SD) characteristics of different study groups are presented in Table 1. There were no significant differences ($p>0.05$) in age at menarche, measured body composition and training experience values between studied groups. However, athletes in ROC group were older ($p<0.05$) in comparison with other groups. Athletes in CCA group had significantly higher weekly training volume in comparison with RCA and ROC groups (Table 1). In ROC group, mean estradiol values were significantly different ($p<0.05$) between FP (176.8±51.9 pmol/l) and LP (481.4±131.0 pmol/l) of the menstrual cycle. Similarly, mean progesterone values were significantly different between FP and LP of the menstrual cycle (1.9±0.5 vs 38.9±11.0 nmol/l; $p<0.05$) in CCA rowers. Mean values for female sex hormones in RCA group were 163.4±98.2 pmol/l for estradiol and 1.3±0.7 nmol/l for progesterone in FP, and 517.7±21.7 pmol/l for estradiol and 30.2±5.6 nmol/l for progesterone in LP. These hormone values were also significantly different ($p<0.05$) between the menstrual cycle phases in RCA group. Estradiol concentrations were significantly lower ($p<0.05$) in ROC compared with rowers in CCA and RCA groups, but they were not different ($p>0.05$) across menstrual cycle in ROC group (FP: 84.7±26.7 pmol/; LP: 51.4±28.7 pmol/l). Progesterone values were similar in all groups in FP. Progesterone levels were significantly higher in LP than FP in CCA and RCA groups. Consequently, progesterone concentrations in LP in ROC were lower ($p<0.05$) than in LP in CCA and RCA groups. Progesterone values were not different between menstrual cycle phases in ROC group (1.5±0.4 vs 1.0±0.3 nmol/l). Therefore, values for estradiol and progesterone were within normal reference ranges during both the FP and LP in both groups of normally menstruating athletes (Landgren et al., 1980). No significant differences ($p>0.05$) in measured body composition parameters were found between FP and LP in all groups (data not shown).

Table 1. Mean (±SD) study characteristics of competitive cyclic athletes (CCA), recreationally trained cyclic athletes (RCA) and recreationally trained athletes taking oral contraceptives (ROC).

Variable	CCA (n=8)	RCA (n=7)	ROC (n=9)
Age (yrs)	18.8±2.1	18.0±0.9	21.0±2.6*#
Age at menarche (yrs)	12.8±1.1	12.4±1.0	12.6±1.0
Height (cm)	174.0±4.2	169.8±5.2	172.9±4.3
Body mass (kg)	69.0±10.7	65.7±6.5	69.6±12.7
BMI (kg/m^2)	22.6±2.8	22.7±1.7	23.2±3.5
Body fat%	27.4±6.2	29.6±3.3	27.3±2.8
Fat mass (kg)	18.4±6.5	18.9±3.4	16.7±2.3
Fat free mass (kg)	48.1±7.0	45.1±3.9	46.2±2.5
Training experience (yrs)	5.8±2.0	4.4±1.4	6.3±2.2
Training volume (h/week)	9.8±2.3	4.5±1.0*	6.0±3.5*

BMI-body mass index.
* Significantly different from CCA; p<0.05.
# Significantly different from RCA; p<0.05.

No significant differences (p>0.05) in Pa_{max}, VO_2max, V_E, the ventilatory equivalents for oxygen (V_E/VO_2) and carbon dioxide (V_E/VCO_2), RER, HR_{max}, peak La and Δ La values measured in FP versus LP were found in CCA, RCA or ROC groups (Table 2). Only V_E/VCO_2 was significantly higher (p<0.05) in LP compared with FP in ROC group. In addition, V_E/VCO_2 value in LP was also significantly higher in athletes in ROC group in comparison with RCA women. Power output values in CCA group in both menstrual cycle phases were significantly higher than the corresponding values in RCA and ROC groups. Other measured parameters in FP were not different between studied groups, while V_E value in LP was significantly lower in RCA women compared to women in CCA group. Similarly, VO_2max, and HR_{max} values in LP were significantly lower in ROC in comparison with CCA women.

Table 2. Maximal ventilatory and metabolic exercise responses of competitive cyclic athletes (CCA), recreationally trained cyclic athletes (RCA) and recreationally trained athletes taking oral contraceptives (ROC).

Variable	CCA (n=8)		RCA (n=7)		ROC (n=9)	
	FP	LP	FP	LP	FP	LP
Pa_{max} (W)	256.1±38.7	261.3±41.8	224.3±23.9#	225.4±24.0#	231.1±28.5#	226.6±27.2#
VO_2max (l/min)	3.35±0.51	3.46±0.64	2.98±0.58	3.00±0.29	3.18±0.51	3.03±0.32
VO_2max (ml/min/kg)	49.0±6.6	50.6±7.1	45.2±9.4	45.4±4.1	45.2±5.7	44.5±5.2#
V_E (l/min)	123.5±12.3	124.6±17.8	112.5±24.4	105.0±19.1#	120.1±16.1	122.0±13.3
V_E/VO_2	35.9±6.1	35.7±6.2	36.7±5.0	36.8±3.8	36.9±4.8	39.3±3.2
V_E/VCO_2	28.4±3.0	28.7±3.1	27.7±1.9	27.4±1.5	27.2±2.6	29.9±1.9*†
RER	1.24±0.10	1.27±0.11	1.32±0.12	1.31±0.11	1.35±0.16	1.32±0.10
HR_{max} (bpm)	190.0±3.2	191.9±3.1	189.4±7.5	190.0±8.8	186.4±3.6	184.5±6.1#
Peak La (mmol/l)	13.0±2.4	13.4±2.6	14.3±3.5	13.8±2.5	12.3±2.7	11.7±1.1
Δ La (mmol/l)	11.2±2.4	11.2±3.1	12.5±3.5	11.7±3.1	10.5±2.8	10.2±1.0

FP – follicular phase; LP – luteal phase; Pa_{max} – maximal power; VO_{2max}– maximal oxygen consumption; V_E– minute ventilation; V_E/VO_2 – ventilatory equivalents for oxygen; V_E/VCO_2 – ventilatory equivalents for carbon dioxide; RER – respiratory exchange ratio; HR_{max} – maximal heart rate; Peak La – peak blood lactate; Δ La – net increase of blood lactate.
* Significantly different from follicular phase; $p<0.05$;
# Significantly different from the corresponding value in CCA group; $p<0.05$;
† Significantly different from the corresponding value in RCA group; $p<0.05$.

No significant differences ($p>0.05$) in Pa_{AT}, VO_2, $\%VO_2max$, V_E, the ventilatory equivalents for oxygen (V_E/VO_2) and carbon dioxide (V_E/VCO_2), RER, HR and $\%HR_{max}$ values measured in FP versus LP were observed in all three groups (Table 3). Only V_E/VCO_2 was significantly higher in LP compared with FP in ROC group. No significant differences ($p>0.05$) in measured parameters between three different groups were found in FP. RER was significantly higher ($p<0.05$) in RCA and ROC groups in comparison with athletes in CCA group. In addition, P_{AT} and V_E in LP were significantly lower ($p<0.05$) in RCA in comparison with CCA.

Table 3. Ventilatory and metabolic exercise responses at the level of anaerobic threshold in competitive cyclic athletes (CCA), recreationally trained cyclic athletes (RCA) and recreationally trained athletes taking oral contraceptives (ROC).

Variable	CCA (n=8)		RCA (n=7)		ROC (n=9)	
	FP	LP	FP	LP	FP	LP
Pa_{AT} (W)	185.0±30.1	186.1±30.8	162.1±25.1	160.6±17.3#	172.2±26.9	174.9±26.8
VO_{2AT} (l/min)	2.77±0.46	2.87±0.36	2.48±0.41	2.48±0.33	2.56±0.58	2.58±0.45
$\%VO_2max$	82.9±7.7	84.0±9.0	83.9±5.9	83.2±10.4	80.1±11.8	85.0±8.8
VO_{2AT} (ml/min/kg)	40.5±5.3	42.5±5.8	37.8±6.9	37.6±6.7	37.1±6.7	37.4±5.6
$V_{E/AT}$ (l/min)	77.6±17.1	76.8±7.1	69.9±12.1	67.8±7.4#	70.8±16.5	75.4±13.6
V_E/VO_{2AT}	25.7±1.7	25.8±2.1	26.8±3.8	26.8±1.7	26.7±3.8	28.3±3.2
V_E/VCO_{2AT}	25.2±1.8	26.2±1.6	25.0±1.6	24.7±1.5	24.3±2.4	25.9±2.0*
RER_{AT}	1.03±0.06	1.01±0.06	1.07±0.13	1.10±0.05#	1.10±0.13	1.08±0.09#
HR_{AT} (bpm)	172.5±4.4	172.9±4.2	171.7±8.8	172.3±9.2	167.9±7.1	168.1±6.1
%HRmax	91.5±1.1	90.0±1.6	90.5±3.6	90.8±1.3	90.0±3.2	91.1±0.9

FP – follicular phase; LP – luteal phase; Pa_{AT} – power at anaerobic threshold; $\%VO_{2max}$ – percent of maximal oxygen consumption; VO_{2AT} – oxygen consumption at anaerobic threshold; $V_{E/AT}$ – minute ventilation at anaerobic threshold; V_E/VO_{2AT} – ventilatory equivalents for oxygen at anaerobic threshold; V_E/VCO_{2AT} – ventilatory equivalents for carbon dioxide at anaerobic threshold; RER_{AT} – respiratory exchange ratio at anaerobic threshold; HR_{AT} – heart rate at anaerobic threshold.
* Significantly different from follicular phase; $p<0.05$.
# Significantly different from the corresponding value in CCA group; $p<0.05$.

5.2. Substrate oxidation and blood lactate concentration during prolonged aerobic exercise in normally menstruating rowers (Paper II)

There were no significant differences ($p>0.05$) in the energy intake (FP: 2723±397 vs 2683±383 kcal/d) or energy expenditure (2887±467 vs 2814± 565 kcal/d) over the days preceding the FP and LP of the menstrual cycle. Rowers were weight stable throughout the study period, with no significant changes in body composition or VO_2max values between the menstrual cycle phases (Table 4). Circulating estradiol (FP: 110.0±31.3 pmol/l; LP: 460.9± 108.0 pmol/l) and progesterone (FP: 1.5±0.6 nmol/l; LP: 25.8±10.2 nmol/l) concentrations confirmed the menstrual cycle phases, with a 4-fold increase in estradiol ($p<0.05$) and a 17-fold increase in progesterone ($p<0.05$) in the LP compared with the FP. Estradiol to progesterone ratio was also significantly increased ($p<0.05$) from FP (85.2±36.4) to LP (20.3±9.7).

Table 4. Mean (±SD) subject characteristics in normally menstruating athletes (n=11).

Variable	FP	LP
Body mass (kg)	67.2±8.4	69.3±8.3
Body fat%	27.7±4.8	27.9±4.6
Fat mass (kg)	18.0±3.9	18.2±3.8
Fat free mass (kg)	47.7±6.0	48.0±5.9
VO_2max (l/min)	3.18±0.52	3.15±0.42
VO_2max (ml/min/kg)	47.5±8.3	45.2±5.7
VO_{2AT} (l/min)	2.69±0.44	2.68±0.38
AT (%VO_2max)	84.9±6.6	85.3±9.5

FP – follicular phase; LP – luteal phase; VO_{2max} – maximal oxygen consumption; VO_{2AT} – oxygen consumption at anaerobic threshold; AT – anaerobic threshold.

Energy expenditure, VO_2 and HR during 1-h rowing ergometer exercise were not significantly different ($p>0.05$) among menstrual cycle phases (Table 5). On the average, athletes rowed at 69.3% and 69.8% of VO_2max during the FP and LP, respectively. HR was not significantly different at any point between the trials (data not shown).

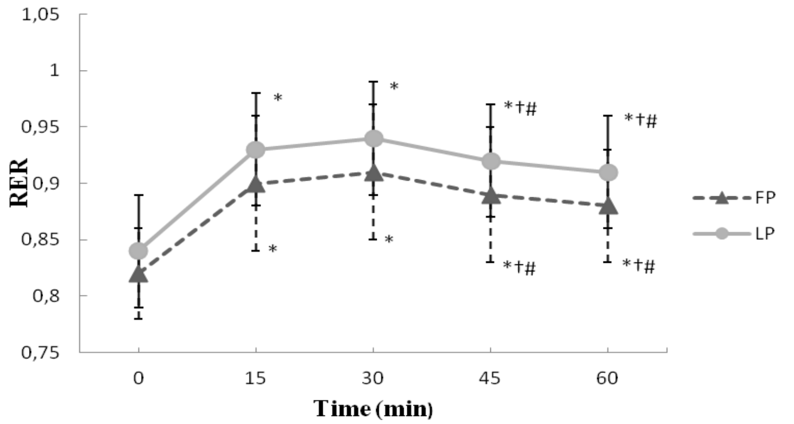
Table 5. Mean (±SD) measured physiological and energy expenditure values during 1-h submaximal exercise in normally menstruating athletes (n=11).

Menstrual phase	HR (bpm)	VO_2 (l/min)	% VO_2max	RER	EE rate (kcal/min)	Total EE (kcal/60 min)
FP	152.8±5.6	2.2±0.4	69.2±2.2	0.89±0.06	10.4±1.3	627.8±89.6
LP	153.1±6.7	2.2±0.5	69.8±2.3	0.93±0.05	10.8±1.6	649.7±100.2

FP – follicular phase; LP – luteal phase; HR – heart rate; VO_2 – oxygen consumption; %VO_2max – percent of maximal oxygen consumption; RER – respiratory exchange ratio; EE rate – energy expenditure rate; Total EE – total energy expenditure.

Resting RER and RER during the entire 1-h exercise period were not significantly different between menstrual cycle phases (Figure 2). However, there was a significant increase ($p<0.05$) in RER during the transition between rest and exercise at both menstrual cycle phases. In addition, a further increase ($p<0.05$) in RER occurred after the first 30 min of exercise at both menstrual cycle phases. At rest, about 50% of the energy was derived from carbohydrate sources during both menstrual cycle phases, but no significant phase effect was observed (Table 6). During exercise, at least 70% of the energy used was derived from carbohydrate sources (FP: ≥7.4 kcal/min; LP: ≥7.6 kcal/min). The contribution of the amount of carbohydrate to energy expenditure decreased ($p<0.05$) after the first 30 min of exercise, while the contribution of the amount of lipid to energy expenditure was not significantly changed ($p>0.05$) throughout the 1-h exercise in both menstrual cycle phases. Blood lactate concentrations increased ($p<0.05$) during the transition between rest and exercise and remained relatively constant during the whole 1-h exercise in both menstrual cycle phases (Figure 2).Therefore, no significant differences in measured parameters during the 1-h exercise between different menstrual cycle phases were observed (Table 6).

a



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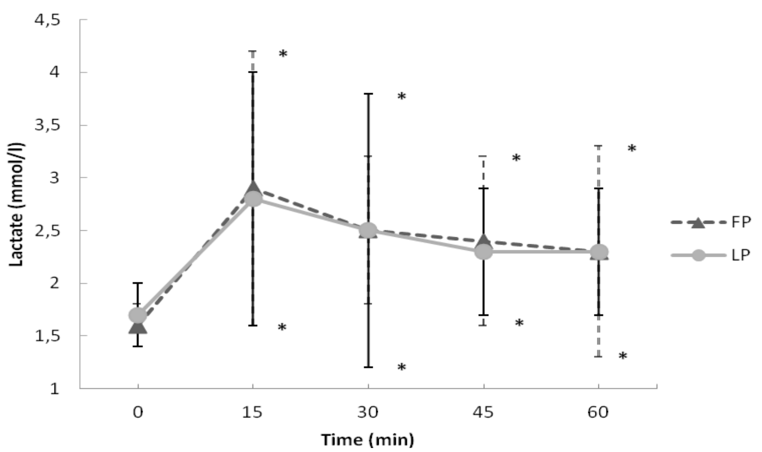

Figure 2. Respiratory exchange ratio (RER) (a) and blood lactate concentrations (b) at rest and during 1-h ergometer rowing at an intensity of 70% VO_2max at the follicular (FP) and luteal (LP) phases of the menstrual cycle in rowers (n=11).
* Significantly different from 0 min; $p<0.05$;
† Significantly different from 15 min; $p<0.05$;
# Significantly different from 30 min; $p<0.05$.

Table 6. Mean (±SD) energy expenditure values during rest and 1-h submaximal rowing ergometer exercise in normally menstruating athletes (n=11).

Variable	Menst-rual phase	Rest	Exercise			
			0–15 min	16–30 min	31–45 min	46–60 min
CHO EE (kcal/min)	FP	0.8±0.4	8.2±2.1*	7.9±2.4*	7.4±2.3*†#	7.4±2.4*†#
	LP	0.8±0.5	8.4±2.2*	8.1±2.0*	7.7±2.1*†#	7.6±2.1*†#
Lipid EE (kcal/min)	FP	0.7±0.5	2.4±1.8*	2.6±2.0*	2.9±1.9*	2.9±2.0*
	LP	0.8±0.4	2.5±1.8*	2.7±1.8*	2.9±1.8*	3.0±1.9*
EE rate (kcal/min)	FP	1.5±0.4	10.6±0.4*	10.5±1.3*	10.3±1.4*†	10.3±1.4*†
	LP	1.6±0.5	10.9±1.5*	10.8±1.5*	10.6±1.3*†	10.6±1.3*†

FP – follicular phase; LP – luteal phase; CHO EE – carbohydrate energy expenditure; Lipid EE – lipid energy expenditure; EE rate – energy expenditure rate.
* Significantly different from resting conditions; $p<0.05$;
† Significantly different from 0–15 min; $p<0.05$;
# Significantly different from 16–30 min; $p<0.05$.

5.3. Substrate oxidation and blood lactate concentration during prolonged aerobic exercise in rowers taking oral contraceptive pills (Paper III)

Rowers were weight stable (AP: 69.6±13.6 kg; NAP: 69.8±13.3 kg), with no significant changes in body composition (data not shown) or VO_2max (AP: 45.9±5.7 ml/min/kg; NAP: 44.3±5.5 ml/min/kg) between the OC cycle phases ($p>0.05$). There were no significant differences in estradiol (AP: 84.7± 26.7 pmol/l; NAP: 51.4±28.7 pmol/l) and in progesterone (AP: 1.5±0.4 nmol/l; NAP: 1.1±0.2 nmol/l) concentrations between OC cycle phases ($p>0.05$). Mean EE, Pa, V_E, VO_2, HR and blood lactate during 1-h rowing ergometer exercise were not significantly different ($p>0.05$) between the AP and NAP of the OC cycle phases (Table 7). In addition, no significant differences between OC phases were observed ($p>0.05$) for mean EE rate, carbohydrate EE and lipid EE during the 1-h endurance test (Table 7). During exercise, at least 70% of the energy was obtained from carbohydrate oxidation (AP: ≥7.1 kcal/min; NAP: ≥8.0 kcal/min) (Figure 3). There was a tendency for a decreased carbohydrate oxidation and increased lipid oxidation during the AP phase in comparison with the NAP phase.

Table 7. Mean (±SD) for physiological and energy expenditure values during 1-h submaximal rowing ergometer exercise in athletes taking oral contraceptive pills (n=8).

	AP	NAP
Pa (W)	104.4±20.5	104.4±22.4
V_E (l/min)	56.1±8.7	55.6±12.6
VO_2 (l/min)	2.0±0.3	2.0±0.4
% of VO_{2max}	64.5±4.6	63.6±7.7
HR (bpm)	147.7±7.1	148.4±7.0
La (mmol/l)	2.1±0.8	2.3±1.0
RER	0.93±0.04	0.95±0.03
EE rate (kcal/min)	9.2±1.6	9.8±1.4
CHO EE (kcal/min)	7.1±2.1	8.0±1.4
Lipid EE (kcal/min)	2.1±0.9	1.7±1.3
Total EE (kcal/60 min)	551.7±95.6	587.6±88.5
% Carbohydrate of total EE	75.7±14.2	82.6±11.1
% Lipid of total EE	24.2±14.2	17.4±11.1

AP – active oral contraceptive pill phase; NAP – not active oral contraceptive pill phase; Pa – power; V_E – minute ventilation; VO_2 – oxygen consumption; % of VO_{2max} – percent of maximal oxygen consumption; HR – heart rate; La – blood lactate; RER – respiratory exchange ratio; EE rate – energy expenditure rate; CHO EE – carbohydrate energy expenditure; Lipid EE – lipid energy expenditure; Total EE – total energy expenditure.

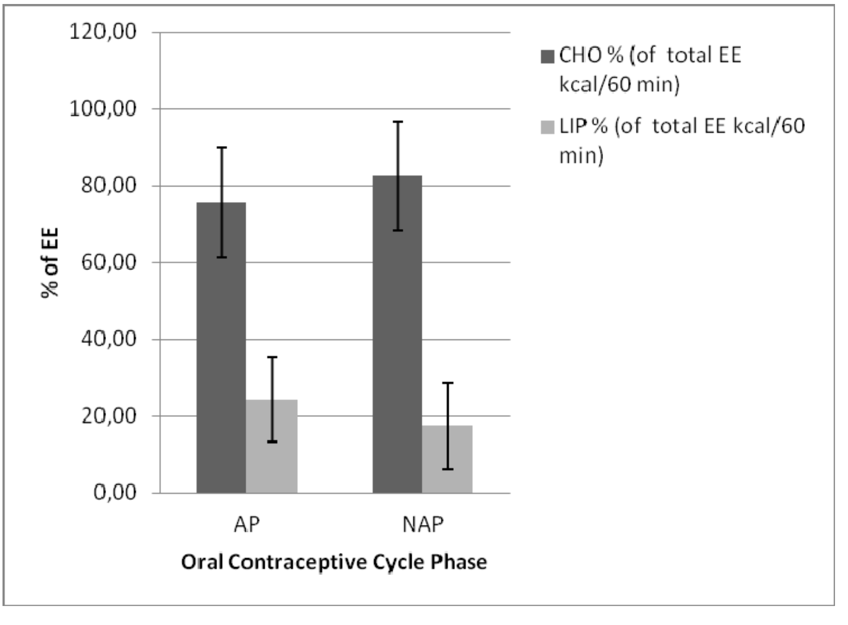

Figure 3. Mean (±SD) lipid (LIP) and carbohydrate (CHO) oxidation values during 1-h erometer rowing exercises at the intensity of 70% VO_2max at active pill (AP) and non-active pill (NAP) phases of the oral contraceptive cycle in athletes taking oral contraceptive pills (n=8).

6. DISCUSSION

6.1 Effects of the menstrual cycle phase and oral contraceptive use on aerobic endurance performance parameters

This study investigated possible changes in maximal and submaximal aerobic performance characteristics between the early and late stages of the menstrual cycle in healthy rowers, and evaluated whether endurance performance parameters across menstrual cycle phase differed between normally cycling women and OC users. In the current study, the menstrual cycle phases in normally menstruating rowers were characterized by different estrogen and progesterone concentrations: in FP, low estrogen and low progesterone; and in LP, elevated estrogen and elevated progesterone (Casazza et al., 2004; Oosthuyse and Bosch, 2010; Forsyth and Reilly, 2005, 2008; Zderic et al., 2001). In rowers taking OC pills, both estrogen and progesterone values continued to be suppressed in early withdrawal phase of LP (NAP phase) as seen in FP (AP phase) similarly to other studies with athletes (Ives et al., 2011; Rechichi and Dawson, 2009; Rechichi et al., 2008). The incremental rowing ergometer tests conducted at two different phases of the menstrual cycle in this study provided a valid reflection of sport-specific rowing performance in studied rowers (Forsyth and Reilly, 2005; Jürimäe et al, 2006), as opposed to time to exhaustion (Dean et al., 2003; Lebrun et al., 2003) and 1-h maximal performance (Rechichi and Dawson, 2009) tests, which may be more valid for the assessment of endurance capacity (Bishop, 1997). Exercise intensities at VO_2max and AT are important determinants of rowing performance over 2000 metre distance (Ingham et al., 2002; Jürimäe et al., 2000; Womack et al., 1996). Similarly to our study, incremental ergometer tests have also been used in other menstrual cycle studies with physically active women (De Souza et al., 1990; Smekal et al., 2007). The current study observed no significant difference in either maximal aerobic (i.e., Pa_{max}, VO_2max) or aerobic-anaerobic transition (i.e., P_{AT}, VO_{2AT}) performance characteristics across the menstrual cycle in normally cycling athletes with different performance levels (CCA and RCA groups) and also in rowers taking OC pills. These results demonstrated that sport-specific endurance performance was not influenced by the phase of the normal menstrual cycle and the synthetic menstrual cycle of the OC users in studied rowers.

The results of the present investigation are in agreement with other studies, which have also found no effect of the menstrual cycle phase on Pa_{max}, VO_2max and HR_{max} values (see Table 2) in physically active women with normal menstrual cycle (De Souza et al.,1990; Dean et al., 2003; Forsyth and Reilly, 2005; Smekal et al., 2007). However, we cannot confirm the results of Lebrun et al. (2003) study, which found significantly lower values for VO_2max in LP vs FP of the menstrual cycle in normally cycling athletes. The present observation of no menstrual cycle differences in peak La concentration is also similar to what has also been found in other studies (De Souza et al., 1990; Dean et al., 2003; Forsyth and Reilly, 2005; Smekal et al., 2007). In contrast, Jurkowski et al. (1981) observed lower blood La concentrations during the LP compared to the FP after exercise at 66% and 90% Pa_{max} in exercising women. This was accompanied by a twofold increase in time to exhaustion at these exercise tests. It was suggested that the primary factor related to less blood La accumulation during the LP is a decreased production of La (Jurkowski et al. 1981). However, other investigators have suggested that the menstrual cycle does not alter the metabolic response to exercise at different intensities for various durations and that substrate metabolism should be comparable during FP and LP of the menstrual cycle (Dean et al., 2003; Lamont et al., 1986). Taken together, the results of current study and that of others (De Souza et al., 1990; Dean et al., 2003; Forsyth and Reilly, 2005; Smekal et al., 2007) suggest that normal cyclic variations in estrogen and progesterone are without significant effects on any measure of maximal endurance performance in trained females.

Results of Pa_{AT}, VO_{2AT} and HR_{AT} measures at aerobic-anaerobic transition intensity (see Table 3) in our studied rowers are comparable with those of others that have reported no significant changes in endurance performance at the level of AT between FP and LP of the menstrual cycle in normally cycling athletes (De Souza et al., 1990; Dean et al., 2003; Smekal et al., 2007). In contrast, the only other study with rowers found that AT at fixed blood La concentration of 4 mmol/l occurred at a significantly higher Pa_{AT}, VO_{2AT} and HR_{AT} values in LP compared to FP of the menstrual cycle (Forsyth and Reilly, 2005). However, the VO_2max values (2.75±0.63 l/min) in rowers in Forsyth and Reilly (2005) study were lower in comparison with normally cycling rowers in the current study (CCA: 3.35±0.51 l/min and RCA: 2.98±0.58 l/min). Thus, the performance level of athletes could explain different results of our study and that of Forsyth and Reilly (2005). It has been suggested that using fixed values of blood La in physiologic assessment, consideration should be given to the menstrual cycle phase in which the test is carried out in athletes (Forsyth and Reilly, 2005). However, the exercise intensity at 4 mmol/l may overestimate the aerobic-anaerobic transition intensity and therefore is not recommended to use in rowing training (Jürimäe and Jürimäe, 2009; Mäestu et al., 2005). Similarly to the results of our investigation (see Table 3), when the AT data in Forsyth and Reilly (2005) study were examined using a log-log transformation method, a curve-fitting procedure (Dmax), a visual breakpoint method, and a ventilatory threshold method, no differences in Pa_{AT}, VO_2 and HR values were observed between FP and LP of the menstrual cycle. These results together suggest that although the glycogen-sparing effect of estrogen in the LP may cause a decrease in La production in muscle during exercise (Forsyth and Reilly, 2005), menstrual cycle phase does not affect aerobic-anaerobic transition intensity at least in more trained athletes.

The research to date addressing energy supply during exercise at different phases of menstrual cycle is inconsistent. For example, the premise of a greater use of lipids as an energy source is supported by the findings of a lower RER in the LP of the menstrual cycle (Dombovy et al., 1987; Redman et al., 2003), while other studies have reported unchanged RER between different menstrual cycle phases (Dean et al., 2003; Smekal et al., 2007). In our study, the assessed energy supply variables (RER, V_E/VO_2, V_E/VCO_2) were not significantly different between the FP and LP of the menstrual cycle at maximal and aerobic-anaerobic transition intensities (see Tables 2 and 3). Accordingly, our data do not support the assumption of the change in energy supply during exercise throughout the time course of the menstrual cycle in normally cycling athletes.

Most previous researchers who have investigated aerobic exercise parameters during AP (FP) and NAP (LP) phase within an OC cycle have found no variation as a result of acute hormonal change (Bryner et al., 1996; Lynch et al., 2001; Rechichi and Dawson, 2009). Furthermore, Rechichi and Dawson (2009) measured aerobic exercise parameters during two separate withdrawal phases in LP (i.e., early in withdrawal phase, 2–3 days post active pill cessation and late in withdrawal phase, 6–7 days post active pill cessation) and also found no differences in 1-h maximal cycling performance test in trained female cyclists and triathletes. The early withdrawal phase in LP in that study (Rechichi and Dawson, 2009) corresponds to our measurement time in LP (NAP phase) of the OC cycle in rowers. In addition, subjects used low-dose monophasic OC pills similarly to the female athletes in Rechichi et al. (2009) study. Performance parameters (Pa_{AT}, VO_2, HR) measured at maximal and aerobic-anaerobic transition intensities (see Tables 2 and 3) were also not different between FP (AP phase) and LP (NAP phase) among our rowers taking OC pills. However, there was a tendency toward increased ventilatory response during the ingestion of NAP phase as demonstrated by higher levels of V_E, V_E/VO_2 and V_E/VCO_2 at maximal and also aerobic-anaerobic transition intensities, while V_E/VCO_2 value reached statistical significance in both intensities. These results were unexpected as previous findings have implicated progesterone as a factor causing an increase in respiratory drive (Rechichi et al., 2008). However, in LP (NAP phase) both estradiol and progesterone values continued to be suppressed similarly to other studies with athletes (Rechichi and Dawson, 2009; Rechichi et al., 2008). Furthermore, Rechichi et al. (2009) observed reduced endogenous progesterone values also in later phases of the LP (NAP phase) in athletes taking OC pills. Despite some differences in ventilatory response, the results of present study support the notion that endurance performance is not affected by OC phase in rowers. However, further studies with more frequent measurement times throughout OC cycle in athletes are needed before any conclusions can be drawn.

In summary, the results of the incremental rowing ergometer tests conducted during the two different phases of the menstrual cycle (FP and LP) demonstrated that there was no change in the measured endurance parameters in normally menstruating rowers with different performance levels and also in rowers taking OC pills. Therefore, female rowers taking or not taking OC pills should not be concerned about the timing of their menstrual cycle in regard to optimized maximal and submaximal aerobic rowing performance.

6.2. Effect of the menstrual cycle phase on substrate oxidation and blood lactate concentration during prolonged aerobic exercise

The present study investigated the effect of the normal menstrual cycle on resting and exercise substrate oxidation. This study was unique in that it used rowing exercise, where all extremities and trunk muscles are involved, which produces higher energy expenditure during exercise and rowers present relatively large body mass values in comparison with other endurance athletes (Jürimäe, 2008; Mäestu et al., 2005). To our best knowledge, no studies before have compared the response of moderate-intensity long-duration rowing exercise across different phases of the menstrual cycle. No significant differences were observed in the whole body carbohydrate and lipid oxidation at rest and during 1-hour rowing exercise at 70% of VO_2max in FP and LP of the menstrual cycle in rowers. In addition, there were no significant differences in blood La concentrations during exercise at both menstrual cycle phases.

The findings of the present investigation that resting RER and energy expenditure values were not significantly different between FP and LP are in line with other studies (Bailey et al., 2000; Horton et al., 2002; Suh et al., 2002). In addition, about 50% of the energy was derived from carbohydrate sources at rest at both menstrual cycle phases without any phase effect similarly to other studies (Bailey et al., 2000; Horton et al., 2002; Suh et al., 2002). The present data also agree with previous findings of no significant differences in resting blood La concentrations at different phases of the menstrual cycle (Devries et al., 2006; Horton et al., 2002; Suh et al., 2002). These results together suggest that normal cyclic variations in ovarian hormones are without significant effects on resting substrate metabolism in female athletes.

The main finding of the present investigation was that no significant differences in the whole body carbohydrate and lipid oxidation during moderate-intensity long-duration exercise at different phases of the menstrual cycle were observed in rowers. This is in agreement with other studies, which have measured exercise RER at the intensity of 45% (Suh et al., 2002), 50% (Horton et al., 2002), 65% (Devries et al., 2006; Suh et al., 2002; Zderic et al., 2001), 70% (Bailey et al., 2000), 75% (Hackney et al., 1994) and 80% (De Souza et al., 1990) of VO_2max in physically active healthy women. In contrast, other studies have found greater lipid oxidation and lower carbohydrate oxidation in the LP compared to FP of the menstrual cycle with exercise intensity higher than 50% of VO_2max (Campbell et al., 2001; Hackney, 1999; Zderic et al., 2001). However, Hackney et al. (1994) reported greater LP lipid oxidation only when exercise was performed at low (35% of VO_2max) and moderate (60% of

VO_2max) intensities, but not at the intensity of 75% of VO_2max in healthy eumenorrheic women. In that study, women performed a 30-min treadmill run in which the exercise intensity was increased in every 10 min (35%, 60% and 75%) (Hackney et al., 1994). In another study, Wenz et al. (1997) also found greater LP lipid oxidation during cycle ergometer exercise at the intensities of 30% and 50% of VO_2max but not during cycle ergometer exercise at the intensity of 70% of VO_2max in healthy eumenorrheic women. The reasons for those inconsistencies with the respect to menstrual cycle phase effects on exercise fuel oxidation could be the discrepancies in the exercise mode and protocol, the RER measurements during the exercise, pre-trial diet and exercise control.

It could be argued that the differences in the findings of substrate oxidation at different phases of the menstrual cycle between current study and the other study utilizing endurance-trained athletes (Campbell et al., 2001) could be explained in part by the differences in exercise duration and mode as both studies used exercise intensity of 70% of VO_2max. Athletes in Campbell et al. (2001) study cycled for 2 h, while athletes in our study rowed for 1 h. In cycling, only lower limbs are exercising, while whole body muscles are involved in rowing causing higher exercise energy expenditure (Jürimäe, 2008; Mäestu et al., 2005). Recently, the effect of exercise mode on substrate oxidation has also been demonstrated (Cheneviere et al., 2010). Another explanation could be the fact that athletes in Campbell et al. (2001) study exercised in a fasted state, while rowers in the present study were in a post-absorptive state having eaten a meal for about 2 h before the test to replicate the usual everyday conditions during training sessions (D'Eon et al., 2002; Jürimäe et al., 2006; Karila et al., 2008). Indeed, when endurance-trained athletes in Campbell et al. (2001) study were adequately supplied with carbohydrate throughout the exercise, the circulating levels of estrogen and progesterone during menstrual cycle had only a minimal effect on substrate oxidation during exercise. In addition, it has been found that estrogen supplementation has no effect on exercise RER (Carter et al., 2001). Accordingly, it could be suggested that changes in estrogen and progesterone across the normal menstrual cycle do not appear to be of sufficient magnitude to affect exercise substrate oxidation in post-absorptive state athletes as observed in the current study and by others (Campbell et al., 2001). However, additional studies with highly trained athletes are warranted before any conclusions can be drawn.

The current observation of no menstrual cycle phase differences in blood La concentrations during prolonged exercise (see Figure 2) is similar to our incremental tests results (see Table 2) and also to the results of other studies (Campbell et al., 2001; De Souza et al., 1990; Devries et al., 2006; Nicklas et al., 1989; Suh et al., 2002). It has been proposed that variations in blood La concentrations in menstrual cycle phases during exercise may occur only in a glycogen-depleted state (Lavoie et al., 1987) and/or in untrained females (Lynch and Nimmo, 1998). The subjects in the present study were trained and performed a steady state exercise in a fed state. However, measurement of substrate turnover rates should be performed to accurately estimate the relative contribution of glucose kinetics during exercise at different phases of the menstrual cycle in order to appropriately answer this question in rowers.

In summary, the results of present investigation demonstrated no significant differences in substrate oxidation and blood La concentration during 1-h rowing exercise at 70% of VO_2max in different phases of menstrual cycle in endurance-trained eumenorrheic athletes.

6.3 Effect of the monophasic oral contraceptive cycle phase on substrate oxidation and blood lactate concentration during prolonged aerobic exercise

The present study examined substrate oxidation during submaximal aerobic exercise at different phases of the monophasic OC cycle. The 1-h submaximal rowing exercise performed slightly below 70% of VO_2max in this study provided a valid reflection of sport-specific endurance capacity (Jürimäe, 2008; Mäestu et al., 2005), as opposed to incremental and time trial endurance performance tests (Forsyth and Reilly, 2005). No significant differences were found in the whole body carbohydrate and lipid oxidation during 1-h submaximal rowing exercise test at AP and NAP phases in rowers. In addition, there were no significant differences in post exercise blood La levels at both OC cycle phases.

Most previous research that have studied aerobic exercise parameters during different points within an OC cycle have found no significant variation as a result of acute hormonal change (Bryner et al., 1996; De Bruyn-Prevost et al., 1984; Rechichi et al., 2009; Reilly 1994). The results of present investigation also demonstrated that mean HR, Pa, V_E and VO_2 of the 1-h submaximal aerobic endurance test slightly below 70% of VO_2max were not substantially influenced by the phase of the OC cycle (see Table 7). Giacomoni and Falgairette (2000) also found no evidence of HR, V_E or substrate oxidation differences throughout the OC cycle in untrained females, who performed three sets of 4-min submaximal exercise bouts on a treadmill. However, submaximal VO_2 was significantly lower in the AP phase compared with the NAP phase of the OC cycle (Giacomoni and Falgairette, 2000). In another study, Rechichi et al. (2008) assessed the effect of 1-h cycle endurance test three times throughout a monophasic OC cycle in 13 well-trained cyclists, and found that the mean V_E was higher ($\approx 7\%$; $p<0.05$) during AP phase compared to two withdrawal phases despite no differences in performance and VO_2 parameters. It is possible that the variation in physiological parameters obtained in some studies between the different OC phases could be related to the differences in exercise mode, duration, intensity and the performance level of studied participants. In addition, variations in aerobic exercise parameters across different studies may also be related to the different types of monophasic OC agents used. Specifically, OC agents containing third generation progestogens (i.e., desogestrel and gesto-dene) might not have the same effect on aerobic exercise parameters compared to natural progesterone or the earlier progestogens (Rechichi, et al., 2009).

It has been suggested that endurance capacity would be optimal during AP phase of the OC cycle, as a result of a shift in substrate utilization toward increased lipid oxidation and decreased carbohydrate oxidation (Rechichi et al., 2008). In contrast to previous findings with OC use (Casazza et al., 2004), no significant differences ($p>0.05$) in substrate metabolism between AP and NAP phases of the OC cycle was found in the present study (see Table 7). However, there was a tendency towards increased lipid oxidation and decreased carbo-hydrate oxidation during AP phase compared to NAP phase of the OC cycle (see Figure 3). There may be some reduction in carbohydrate metabolism during submaximal aerobic exercise with OC use, mainly because of a decreased dependence on glycogen and altered secretion of sex hormones (D'Eon et al., 2002; Rechichi, et al., 2009). In addition, similarly to Rechichi et al. (2008) study, no significant differences were observed in corresponding RER values between different OC cycle phases (see Table 7). However, Rechichi et al. (2008) measured RER during two separate withdrawal phases of the OC cycle (i.e., early withdrawal phase, 2–3 days post AP cessation and late in withdrawal phase, 6–7 days post AP cessation). The early withdrawal phase in Rechichi et al. (2008) study corresponds to our measurement time in NAP phase of the OC cycle in rowers. It has to be considered that estrogen and pro-gesterone values continued to be suppressed in NAP phase of the OC cycle in our rowers. It is interesting to note that Rechichi et al. (2008) observed reduced endogeneous progesterone values also at later withdrawal phase of the OC cycle.

Similarly to other studies (Bemben et al., 1992; Bernandes and Radomski, 1998), no differences in post exercise blood La concentrations were observed at different phases of the OC cycle in studied rowers (see Table 7). In contrast, Rechichi et al. (2008) found significantly higher post-exercise blood La values in AP phase compared to early withdrawal phase of the OC cycle in trained athletes. However, the authors argued that despite significant difference, the physiological significance of 1 mmol/l blood lactate change (between 5 and 6 mmol/l) is likely to be relatively small and given that there was no correspon-ding difference in RER or blood glucose values, it is difficult to attribute the variation in blood La results to shifts in substrate metabolism (Rechichi et al., 2008). Taking together, the results of these studies do not support the direct link between OC cycle phases and post-exercise blood La concentrations.

In summary, the results of current study found no significant effect of OC cycle phase on substrate oxidation and blood La concentration during 1-h rowing exercise at slightly lower than 70% of VO_2max in endurance-trained rowers.

7. CONCLUSIONS

Sport-specific aerobic endurance performance and substrate oxidation during everyday training were not influenced by the specific phase of the normal menstrual cycle and the synthetic menstrual cycle of the oral contraceptive users in studied rowers. However, some cyclical variations in ventilatory response values should be considered when interpreting physiological test results in oral contraceptive users. Overall, normally menstruating female rowers and female rowers taking oral contraceptive pills should not consider the phase of the menstrual cycle for optimizing their aerobic performance during testing and everyday training.

The specific conclusions of the present investigation are:

1. the aerobic endurance performance parameters at aerobic-anaerobic transition intensity and at maximal intensity were not affected by the menstrual cycle and oral contraceptive phases in female rowers;
2. the whole body carbohydrate and lipid oxidation, and blood lactate concentration during one hour rowing exercise at about 70% of maximal oxygen consumption were not significantly different in follicular and luteal phases of the menstrual cycle in normally menstruating female rowers;
3. the whole body carbohydrate and lipid oxidation, and blood lactate concentration during one hour rowing exercise at about 70% of maximal oxygen consumption were not significantly different in active and not active pill phases of the oral contraceptive cycle in female rowers.

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SUMMARY IN ESTONIAN

Erinevate menstruaaltsükli faaside ja rasestumisvastaste vahendite kasutamise mõju naissõudjate valitud võistlusparameetritele

Naissportlased treenivad ja võistlevad menstruaaltsükli erinevates faasides. Naissuguhormooni estrogeeni ja progesterooni konsentratsioon organismis kõigub rütmiliselt kogu menstruaaltrükli jooksul. On leitud, et esrtogeen võib soodustada vastupidavuslikku sooritusvõimet läbi süsivesikute ja rasvade ainevahetuse muutmise. Samas on progesteroonile omistatud just vastupidine efekt. Menstruaaltsükli erienvate faaside mõju erinevatele kehalistele koormustele on uuritud palju ja erineva metoodikaga ning jõutud vastakatele järeldustele. Vähesed autorid on uurinud menstruaaltsükli erinevate faaside ja suukaudsete rasestumisvahendite kasutamise mõju sportlikule saavutusvõimele treenitud naissõudjatel. Järjest enam naissportlasi kasutab rasestumisvastaseid vahendeid. Kõige levinumad on suukaudsed monofaasilised rasestumisvastased vahendid. Sealjuures võib rasestumisvastaste vahendite tarbimine luua stabiisema hormonaalse keskkonna kehalise koormuse sooritamiseks kogu sünteetilise menstruaaltsükli jooksul. Kuna ka sünteetiline menstruaaltsükkel koosneb kahest erinevast hormonaalsest faasist, siis on oluline selgitada organismis toimuvad võimalikud muutused sportlikus sooritusvõimes sünteetilise menstruaaltsükli jooksul.

Klassikalise 2000 meetri sõudevõistluse jooksul saab organism 75–85% energiast aeroobsete energiatootmismehhanismide kaudu ja ülejäänud 15–25% anaeroobsete energiatootmismehanismide kaudu. Seetõttu treenivad sõudjad palju pikkadel, madala intensiivsusega koormustel, ning ettevalmistusperioodil jääb treeningute intesiivsus 80–85% ja võistlusperioodil 75% ulatuses alla anaeroobse läve intensiivsuse, peamiselt aeroobse läve intensiivsusel. Anaeroobne lävi (AnL) ja maksimaalne hapnikutarbimine (VO2max) on kõige olulisemad parameetrid, mis määravad 2000 meetri sõudmise võistlutulemust. Seevastu südmelöögisagedus ja vere lakraadi konsentratsionn on samas kõige olulisemad parameetrid treeningu intensiivsuse hindamisel.

Käesoleva uurimustöö peamiseks eesmärgiks oli määrata, kas erinevad aeroobse töövõime parameetrid on mõjutatud normaalse ja sünteetilise menstruaaltsükli jooksul toimuvatest naissuguhormoonide (estrogen ja progesteroon) konsentratsiooni muutustest nii treenitud kui ka vähem treenitud naissõudjatel.

Vastavalt peamisele eesmärgile püstitati järgmised konkreetsed ülesanded:

1. võrrelda, kas submaksimaalse ja maksimaalse aeroobse töövõime näitajad muutuvad menstruaalstükli erinevates faasides nii normaalse menstruaaltsükliga, kui ka rasestumisvastaste vahendite tsükliga naissõudjatel;

2. võrrelda, kas kestva aeroobse pingutuse aegne substraatide oksüdatsioon ja vere laktaadi kontsentratsioon erinevad menstruaalstükli erinevates faasides normaalse menstruatsiooni tsükliga naissõudjatel;
3. võrrelda, kas kestva aeroobse pingutuse aegne substraatide oksüdatsioon ja vere laktaadi konstentratsioon erinevad menstruaalstükli erinevates faasides rasestumisvastase vahendite tsükliga naissõudjatel

Käesolev uuring viidi läbi ettevalmistusperioodi jooksul. Uuringus osales 24 nii normaalse (n=15) kui ka sünteetilise menstrualtsükliga (n=9) erineva treenitus-astmega naissõudjat. Uuritavad läbisid kaks identset kahepäevast testimist kahes erinevas menstrualtsükli faasis: follikulaarses ja luteaalses faasis. Kogu testimisperioodil paluti uuritavatel säilitada stabiilset treeningkoormust ja päev enne testimist oli puhkepäev. Esimesel uuringupäeval võeti vaatlusalustel puhkeoleku vereproov estradiooli ja progesterooni kontsentratsioonide määramiseks, samuti määrati vaatlusalustel antropomeetrilised ja keha koostise näitajad DXA meetodil (keha rasva %, rasvamass ja ravavaba mass) ning sooritati astmeliselt tõusvate koormustega test sõudeergomeetril, kus määrati erinevad submaksimaalse ja maksimaalse aeroobse töövõime parameetrid. Järgmisel uuringupäeval sooritasid sõudjad 1-tunnise madalaintensiivsusega sõudeergomeetri testi, mille käigus määrati sõudjate vere laktaadi konsentratsioon ning hapnikutarbimise ja ventilatsiooni näitajad, mille alusel arvutati energiakulu.

Käesoleva uurimistöö tulemuste põhjal tehti järgmised konkreetsed järeldused:

1. Submaksimaalse ja maksimaalse aeroobse töövõime näitajad ei erine menstruaaltsükli ja suukaudse rasestumisvastase vahendite tsükli erinevates faasides uuritud naissõudjatel;
2. Süsivesikute ja rasvade oksüdatsioon ning vere laktaadi konsentratsioon 1-tunnise sõudeergomeetri testi jooksul intensiivsusega 70% maksimaalsest hapnikutarbimisest ei erinenud statistiliselt usutavalt menstruaaltsükli follikulaarses ja luteaalses faasis naissõudjatel.
3. Süsivesikute ja rasvade oksüdatsioon ning vere laktaadi konsentratsioon 1-tunnise sõudeergomeetri testi jooksul intensiivsusega 70% maksimaalsest hapnikutarbimisest ei erinenud statistiliselt usutavalt suukaudse rasestumisvastase vahendi aktiivse ja mitteaktivse tableti faasis naissõudjatel.

Kokkuvõttes näitasid antud uuringu tulemused, et naissõudjad ei pea muretsema sõudespetsiifiliste võistlustulemust mõjutavate näitajate muutuste pärast normaalse menstruaaltsükli või rasestumisvastaste vahendi tsükli erinevates faasides.

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