The female menstrual cycles effect on strength and power performance in high-level female team athletes

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Abstract

Background: The number of female athletes competing in high level sport is substantial. However, women are still largely underrepresented in the scientific literature. The female menstrual cycle is characterized by changes in concentration of circulating hormone levels, possibly influencing performance. This study investigated the menstrual cycles effect on strength and power performance parameters in highly trained female team athletes.

Methods: Fifty-five participants were recruited for the study and tested throughout a six-week period. Participants were recruited from the team sports soccer, handball and volleyball, competing at the highest national level or the second division. Twelve players were currently representing their national team. The testing protocol consisted of maximal voluntary isometric grip strength, 20-meter sprint, counter movement jump and leg-press. Based on self-reported use of hormonal contraceptives, participants were divided into a non-hormonal contraceptive group and hormonal contraceptive group, the latter working as a control group. Menstrual cycle phase in the non-hormonal group was confirmed by serum hormonal levels.

Results: There was no difference between the groups during the menstrual cycle for any of the outcome variables, with an alpha level of 0.05. High interindividual variability was present in both groups, suggesting that time of testing or competition can yield different results.

Conclusion: For high level female team athletes, there is no difference in performance based on hormonal contraceptive status, suggesting the menstrual cycle does not alter strength and power performance.

Keywords: Menstrual cycle, performance, female athletes, Strength, power, hormones

Sammendrag

Bakgrunn: Antall kvinnelige utøvere som konkurrerer på høyt sportslig nivå er betydelig. Kvinner er likevel høyst underrepresentert innen forskning. Den kvinnelige menstruasjonssyklusen er karakterisert av forandringer i konsentrasjonen av sirkulerende hormon nivå, noe som kan påvirke prestasjonsevne. Denne studien undersøkte menstruasjonssyklusens påvirkning på styrke og kraft prestasjonsparametre i godt trente kvinnelige lagidrettsutøvere.

Metode: femtifem Deltagere ble rekruttert til studien og testet igjennom en seksukers periode. Deltagerne ble rekruttert fra lagidrettene fotball, håndball, volleyball og konkurrerte på høyeste nasjonale nivå eller i andredivisjon. Tolv spillere representerte for tiden landslag. Test protokollen besto av maksimal isometrisk grepstyrke, 20-meter sprint, svikthopp og beinpress. Basert på selvrapportert bruk av hormonell prevensjon, ble deltagerne delt inn i en ikke-hormonell prevensjonsgruppe og en hormonell prevensjonsgruppe, hvor sistnevnte fungerte som en kontroll. Menstruasjonssyklusfase i den ikke-hormonelle prevensjonsgruppen ble bekreftet av serum hormonnivå.

Resultat: Det var ingen forskjell imellom gruppene igjennom menstruasjonssyklusen for noen av utfallsvariablene, med et alfanivå på 0.05. Høy interindividuell variabilitet var til stede i begge gruppene, som antyder at tidspunkt for testing og konkurranse kan forårsake forskjell i resultat.

Konklusjon: I godt trente kvinnelige lagidrettsutøvere er det ingen forskjell i prestasjon basert på hormonell prevensjonsstatus. Dette kan bety at menstruasjonssyklusen ikke påvirker prestasjon relatert til styrke og kraft parametre.

Nøkkelord: Menstruasjonssyklus, kvinnelige utøvere, prestasjon, kraft, styrke, hormoner

Forord

De siste to årene som masterstudent har vært utrolig lærerike. Jeg har utviklet meg både personlig og faglig, noe jeg føler gjenspeiler seg i denne oppgaven.

Jeg ønsker å takke min utrolig dyktige veileder, førsteamanuensis Inger Haukenes ved Universitetet i Bergen (UIB) for god veiledning og hjelp igjennom hele prosessen. Jeg ønsker også takke høgskolelektor og P.hd kandidat Morten Kristoffersen ved fysiologisk laboratorium, høgskolen på Vestlandet (HVL) som gjorde dette prosjektet mulig, sammen med høgskolelærer Lars Peder Vatshelle Bovim fra Sim Arena, HVL. Videre vil jeg takke professor emeritus Rolf Moe-Nilssen for god veiledning og samtaler, spesielt rettet mot statistikk og metode. En takk rettes også til Førsteamanuensis Elisabeth Ersvær, for hjelpen i planlegging og godkjenning av prosjektet.

Jeg ønsker å trekke fram det gode tverrfaglige samarbeidet ved HVL, som også inkluderer førsteamanuensis Silje Mæland og professor Lise Bjørkhaug Gundersen. Måten studenter fra ulike profesjonsretninger har fått delta i utforming og gjennomføringen av prosjektet er et eksempel til etterfølgelse. Jeg ønsker også takke professor Anette Harris ved UIB for bidraget inn i prosjektet. Videre har samarbeidet mellom UIB og HVL vært godt, noe som forhåpentligvis kan videreutvikles i framtiden. En stor takk rettes også til alle deltagerne i prosjektet og studentene som bidro med testing og analyser. Uten dere ville dette aldri vært mulig.

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''Education is not the learning of facts, it's rather the training of the mind to think'' *Albert Einstein*

Abbreviations

MC – Menstrual cycle

HC – Hormonal contraceptive

LH – Luteinizing hormone

FSH – Follicle stimulating hormone

GnRH – Gonadotropin releasing hormone

MVIGS – Maximal voluntary isometric grip strength

RPP – Relative peak power

CMJ – Countermovement jump

HCG – Hormonal contraceptive group

NHCG – Non-hormonal contraceptive group

HRT – Hormonal replacement therapy

FP – Follicular phase

LP – Luteal phase

OC – Oral contraceptive

HUS – Haukeland university hospital

FWE – Family wise error

EBA – Evidence based approach

RTP – Return to play

N- Sample size

Definitions

Definitions explained here is used in the context of this study.

Amenorrhea – The absence of menstruation for three months or more

Oligomenorrhea – Abnormally infrequent or scantly menstruation flow

Eumenorrheic – Normal or regular menstruation

Menarche – The first occurrence of menstruation in females

Acute performance testing – Performance testing without specific training or preparation for the upcoming task or test

Periodized training – Training interventions strategically implementing specific phases to maximize performance for a given parameter

Endogenous – Growing or originating from within an organism

Exogenous – Growing or originating from outside an organism

Bibliography

List of figures

List of tables

1 Introduction

Female participation in high-level sport is substantial, accounting for 45% of the athlete participation during the Olympics (IOC, 2016). However, women are highly underrepresented in the scientific literature. The physiology of men and women differs, in that females, approximately between the age of thirteen and fifty experience monthly changes in serum hormone levels, termed the menstrual cycle (MC). Thus, studies investigating male performance don't necessarily translate to females. Therefore, further understanding of the MC's effect on performance is necessary. The hormonal fluctuations seen throughout the MC may have implications for periodization, training and competition structure in female athletes.

Testing of physiological parameters is a crucial tool in modern sports performance. In recent years the physiology laboratory at the Western Norway university of applied science (HVL) has conducted physical performance tests on multiple elite athletes, across several domains. Experience over time implies that the results of female athletes don't necessarily correlate with the capacity being shown in competition and training, for some individuals. These experiences, along with previously conducted research, implies that the time of testing may influence results and that the female MC may cause alterations to performance.

The sex hormone estrogen have been proposed to induce anabolic and muscle building processes in females (Lowe, Baltgalvis, & Greising, 2010). It has also been shown to attenuate muscle damage during phases of the MC with elevated circulating estrogen concentrations (Carter, Dobridge, & Hackney, 2001). Indeed, the effect of hormone replacement therapy (HRT) using exogenous estrogen, has been shown to attenuate loss of muscle strength in peri and post-menopausal women. This is also supported by rodent studies, showing that estrogen is beneficial in relation to muscle strength. This effect is not necessarily accomplished by increased muscle size through hypertrophy, but rather by effecting the intrinsic quality of skeletal muscle, enabling muscle fibres to generate greater force (Lowe et al., 2010). Progesterone, another female sex hormone which plays an important role in the MC, have been associated with protein catabolism, conceivably attenuating muscle strength (Oosthuyse & Bosch, 2010). These findings together with empirical evidence, would suggest

that muscle strength and power can be ameliorated in periods were circulating estrogen levels are elevated, thereby affecting athletic performance in female athletes. Despite the positive evidence shown in the literature regarding estrogen´s effect on muscle strength, there is contradictory evidence to this. For example, Constantini et al. (2005), Lebrun et al. (1995) and Jansen de Jonge (2003) found no significant differences in muscle strength through the MC (N. W. Constantini, Dubnov, & Lebrun, 2005; de Jonge, 2003; C. M. Lebrun, McKenzie, Prior, & Taunton, 1995). Further, Greeves et al. (1997) reported that patients undergoing *in vitro fertilization* with supraphysiological levels of estrogen did not change the strength of the first dorsal interosseus muscle, contraindicating the hypothesis that estrogen ameliorates muscle strength and power. Even though there were individual differences, these above findings suggest that hormonal fluctuations do not affect the muscles ability to generate force, thereby athletic performance (Greeves, Cable, Luckas, Reilly, & Biljan, 1997)

The field of research regarding the MC's effect on performance, is hampered with inconsistent findings. These findings are coupled with several studies showing methodological inaccuracies. The major underlying fault of these studies is failure to adequately identifying the MC phase of participants (C. M. Lebrun, 2008, p. 43). Hormonal contraceptives (HC), and its effect on performance is still not fully understood. Despite this, the majority of evidence points to it not affecting physiological parameters (Muscle strength, muscle endurance, anaerobic capacity). However, potential altering characteristics of HC agents on performance cannot be excluded (Myllyaho et al., 2018; Nichols, Hetzler, Villanueva, Stickley, & Kimura, 2008; Ruzic, Matkovic, & Leko, 2003).

1.1 Hormones and their effect in females

Hormones are signalling molecules secreted into the blood stream to act on targeting cells and their receptors, thereby influencing metabolic processes in the human body. Three general classes of hormones exist, which are Proteins and polypeptides, steroids and nuclei acid hormones (J. E. Hall, 2016, pp. 925-926).

The female body is unique, in that hormonal fluctuations occur throughout the MC. There are several endogenous hormones active during the MC, affecting the body in various ways depending on the phase of the MC. These phases are broadly termed the follicular phase (FP) and the luteal phase (LP) and are separated by ovulation approximately in the middle of the MC (Mihm, Gangooly, & Muttukrishna, 2011; Oosthuyse & Bosch, 2010). The female hormonal system consists of three hierarchies of hormones, which are a hypothalamic releasing hormone, *gonadotropin releasing hormone* (GnHR), the anterior pituitary sex hormones, *follicular-stimulating hormone (*FSH), *luteinizing hormone* (LH) and the ovarian hormones *estrogen* and *progesterone (J. E. Hall, 2016, p. 1039).*

1.1.1 Gonadotropic hormones

GnHR is released from the hypothalamus and causes the release of the two gonadotropic hormones FSH and LH. While the gonadotropic hormones FSH and LH together with the ovarian hormones estradiol (estrogen) and progesterone are secreted at different rates during the monthly menstrual cycle, the amount of GnHR is consistant and is secreted in bursts averaging every ninety minutes. FSH and LH secreted by the anterior pituitary gland, stimulate ovarian target cells by combining with specific receptors in the ovarian target cell membrane. These activated receptors cause growth and proliferation of the aforementioned cells. The gonadotropic hormones are also key in stimulating sex hormone synthesis. This is done through activation of the cyclic adenosine monophosphate second messenger system in the cell cytoplasm, causing formation of protein kinase and multiple key enzymes (J. E. Hall, 2016, pp. 1039-1041)

1.1.2 Sex hormones

The main female sex hormones are the ovarian hormones estrogens and progestins. Sex hormones are steroids, synthesized in the ovaries mainly from cholesterol derived from the blood, but also from acetyl coenzyme A. Indeed, androgens, are also a female sex steroid, but at significant lower concentration than the estrogen and progesterone (J. E. Hall, 2016, pp. 1042-1046). Various forms of female sex hormones (endogenous and exogenous) exerts myriads of diverse and complex effects on physiological parameters, potentially influencing athletic performance (N. Constantini & Hackney, 2013, p. 285)

Estrogens

Of the estrogens, estradiol (β-estradiol) is the most important, with estrone and estriol also being present in the plasma of the female body (J. E. Hall, 2016, p. 1041). Endogenous estrogen is secreted mainly by the ovaries, but also by the adrenals. Following menopause, the ovaries cease the production of estrogen, and circulating levels decline. Exogenous forms of estrogen includes ethinyl, estradiol and mestranol. Estrogens signals through two nuclear receptors. Estrogen receptor alpha (ERα) and estrogen receptor beta (ERβ). (Simpson, 2003; Wend, Wend, & Krum, 2012; Zallone, 2006).

Estrogen plays a function in almost all cells and tissues in the body. Albeit, at lower levels than those found in the reproductive tissues. Many of these functions have been observed in menopausal women, were estrogen production is decreased and include brain, cardiovascular, immune and musculoskeletal function. Thus, the role of estrogen in the human body is still not fully understood. (Wend et al., 2012). Estrogens primarily promote proliferation and growth of specific cells in the body that are responsible for the development of female sexual characteristics (J. E. Hall, 2016, pp. 1044-1046).

Progestins

Progesterone is the most important progestin hormone, and in large the only important progestin related to hormonal function. In normal non-pregnant females, progesterone is secreted in significant amounts during the latter half of each ovarian cycle, from the corpus luteum. The main function of progesterone is to promote secretory changes in the uterine endometrium, thus preparing the uterus for implantation of the fertilized ovum (J. E. Hall, 2016, pp. 1046-1047).

Beside its primary function, progesterone, also possesses other physiological properties. Progesterone have been shown to ameliorate basal body temperature, increasing it by 0.3-0.5 ° C (N. Constantini & Hackney, 2013, p. 282; Horvath & Drinkwater, 1982; Kelly, 2006; Marshall, 1963; Stachenfeld, Silva, & Keefe, 2000; L. A. Stephenson & Kolka, 1993). However, the regulated body temperature in women is at its lowest during the FP coincident with the cyclic estrogen surge of the menstrual cycle (Lou A. Stephenson & Kolka, 1999). Indeed, both progesterone and estrogen has been reported to alter metabolic responses. The individual hormones can have antagonistic, synergetic or additive effects, eliciting different physiological responses (Campbell & Febbraio, 2001, 2002; N. Constantini & Hackney, 2013, p. 282; D'Eon et al., 2002; Hatta, Atomi, Shinohara, Yamamoto, & Yamada, 1988; Oosthuyse & Bosch, 2010).

Androgens

The major androgens affecting women are, dehydroepiandrosteronesulphate, dehydroepiandrosterone, androtenedione, testosterone and dihydrotestosterone, the first three being considered pro-androgens, as they require conversion to testosterone to express their androgenic effects. The most potent and important androgen is testosterone. Daily production rate in females is between 0.1-0.4 mg, with circulating levels in the range of 0.2-0.7 ng/ml (Burger, 2002). Testosterone rates is at its lowest concentration in the early FP, rises to a mid-cycle peak and the LP concentrations are higher than those in the early FP (Abraham, 1974). Indeed, the concentrations of androgens are highly ameliorated in men compared to women. Nonetheless, androgens play an important role in several physiological processes in women, including development of reproductive functions and hormonal homeostasis. They also present the immediate precursors for the biosynthesis of estrogens (Bachmann et al., 2002).

1.2 The female menstrual cycle

The normal reproductive years of the female are characterized by monthly rhythmical changes in the rates of secretion of the female hormones and corresponding physical changes in the ovaries and other sexual organs. This pattern is known as the MC (J. E. Hall, 2016, p. 1039).

The female MC consists of three hierarchies of hormones. This cascade of hormones influencing each other consist of hypothalamic, anterior pituitary and ovarian hormones (J. E. Hall, 2016, p. 1039). Between menarche and menopause, non-hormonal contraceptive users typically have a monthly menstrual cycle, with cyclical rise and falls in hormone concentrations (D. Martin, Sale, Cooper, & Elliott-Sale, 2018; Stricker et al., 2006).

The FP, which is the first half of the monthly MC is predominated by the anterior pituitary hormones, FHS and LH, with low levels of progesterone and continuously rising estrogen levels. Preceding ovulation (late FP), there is a peak in estrogen levels and a change in the

negative feedback to the hypothalamus. The second half of the cycle, the LP, is characterized by an increase in the ovarian steroid hormone progesterone and partly estrogen, due to high secretion from the corpus luteum. If conception and implantation do not occur, falling levels of hormones cause the lining of the uterus to be shed as menstrual blood flow and the process starts over (Dawson & Reilly, 2009; C. M. Lebrun, 2008, p. 39).

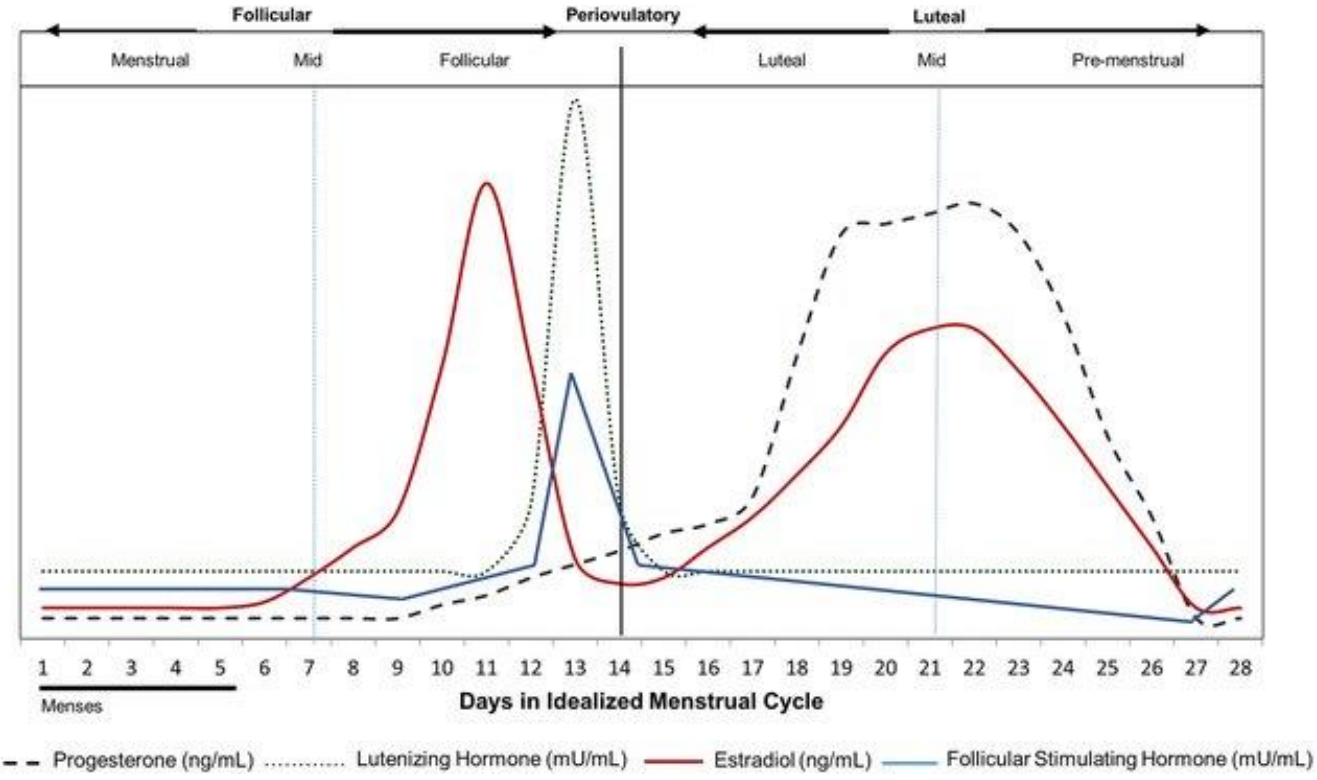

Figure 1.Illustration of the female menstrual cycle, showing serum hormonal levels according to menstrual cycle phases. Used with permission from author (Draper et al., 2018).

During the course of the MC, the collective LP increase in both estrogen and progesterone can cause a variety of symptoms, collectively termed molimina. These include fluid retention, breast tenderness, appetite and mood changes. In most healthy women, these hormone - induced changes are not accompanied by any marked affective aberrations. However, in some women these symptoms are burdensome and are termed Premenstrual syndrome. The onset of menstruation usually leads to relief of these symptoms. Strenuous physical training and extensive energy deficit may also lead to the onset of amenorrhea. This is characterized by

loss of menstruation and may increase risk for diseases such as osteoporosis. The female athlete triad refers to a constellation of menstrual dysfunction, low energy availability and decreased bone mineral density. In women competing in sports which emphasize aesthetics, the prevalence of amenorrhea can be as high as 69% compared to 2-5 % in the normal population (C. M. Lebrun, 2008, p. 43; Nazem & Ackerman, 2012).

1.3 Exogenous hormones and contraceptives

HC´s provide steady levels of exogenous estrogen and progestin, acting primarily by disrupting the normal hypothalamic-pituitary-ovarian (HPO) axis to supress ovulation. It also affects the cervical mocus, making up its primary inhibitory effects. (N. Constantini & Hackney, 2013, p. 282; Rivera, Yacobson, & Grimes, 1999).

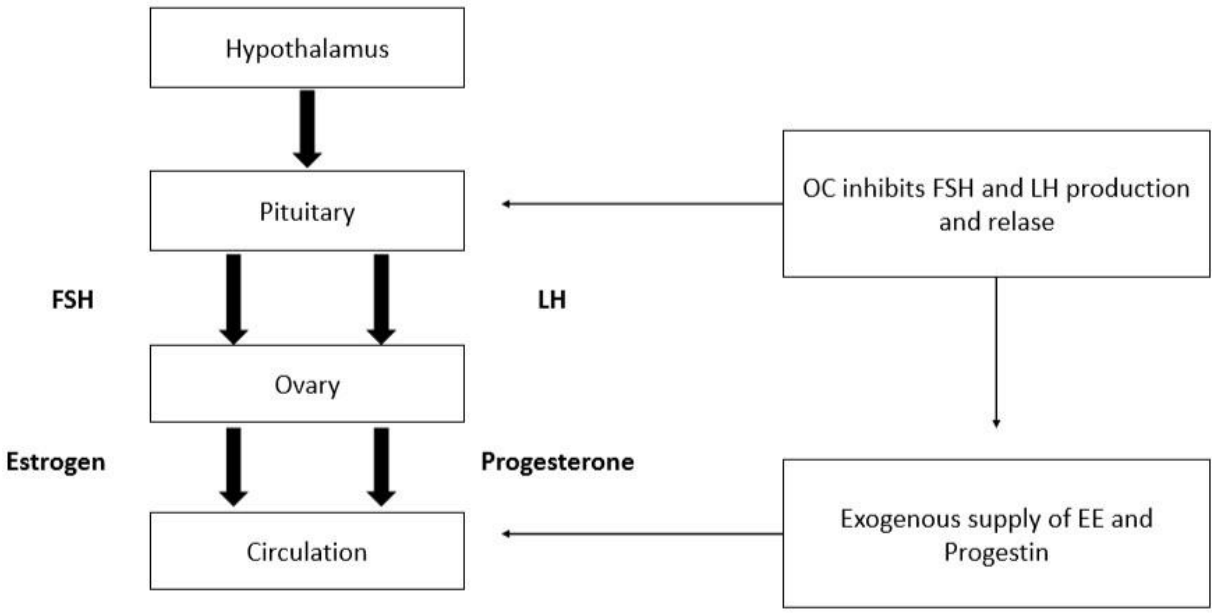

Figure 2. Suppression of endogenous female sex steroid hormones in the HPO axis caused by combined oral contraceptives (OC). FSH= Follicle stimulating hormone; LH= luteinizing hormone; EE= Ethinyl estradiol. Adapted from (N. Constantini & Hackney, 2013, p. 284)

The oral contraceptive pill (OC) is the most popular hormonal birth control option, and it's estimated that the prevalence of use in athletic populations matches that within the general community (Bennell, White, & Crossley, 1999; K. S. Hall, White, Reame, & Westhoff, 2010; Torstveit & Sundgot-Borgen, 2005). Several types of OC´s exist and contemporary low dose combinations pills have a three to fourfold decrease in estrogen content and a tenfold decrease in progestin, compared to earlier generations of OC´s (Petitti, 2003). In monophasic preparations, estrogen and progesterone concentrations are pre-established over the entire pill cycle, while in biphasic and triphasic formulations, amounts vary to imitate the normal cyclical patterns of the MC. Most OC´s incorporate synthetic estrogen in the form of ethinyl estradiol. Synthetic progestins, are found in the forms of norethindrone, norethindrone actate, norethynodrel, ethynodiol diacetate, levonorgestrel and norgestrel (N. Constantini & Hackney, 2013, p. 284; Sitruk-Ware, 2008). For women, unable to abide the combined use of these hormones or those with medical contraindications to estrogen use, there are progestin-only preparations, such as progestin mini-pills (Kaunitz, 1994). Increased and irregular menstrual bleeding with use of progestin–only preparations may be disruptive for athletic training and competition. Limited information exists on these contraceptive methods in athletes and their associated effect on exercise and athletic performance (N. Constantini & Hackney, 2013, p. 285; Mestad, Kenerson, & Peipert, 2009).

In addition to OC's, women have a variety of contraceptive options. Barrier methods, such as the condom have few side effects and does not alter performance, however, its use require consistent administration with intercourse. Long - acting reversible contraceptives (LARCs), which comprise intrauterine devices, progestin – only – implants and progestin injections. LARCs have the lowest failure rate of reversible contraceptive methods (Cea-Soriano, García Rodríguez, Machlitt, & Wallander, 2014; Grimes, 2009).

Further, HC users have been proposed as a suitable control group for investigation of the MC's effect in eumenorrheic females, due to the consistent concentration of circulating sex hormones (Sims & Heather, 2018).

1.4 Hormones and athletic performance

The various female sex hormones (endogenous and exogenous) exerts several diverse and complex effects on multiple physiologic parameters, with the potential to alter performance. Therefore, considerations for exercise performance in women differ significantly from those of men (Charkoudian & Joyner, 2004; N. Constantini & Hackney, 2013, p. 285).

Female sex hormones and their effect on athletic performance is a complex field, with little consensus. Studies investigating the MC's effect on athletic performance are not in agreement, which is also the case regarding HC agents, albeit, the latter seems to have supporting evidence of little effect on performance (Myllyaho et al., 2018; Nichols et al., 2008). There are several reasons for these inconsistent findings. Population and training status of subjects being studied, leading to insufficient statistical power together with individual differences can skew results. Contradictory findings may also be explained by diversity in testing protocols, training intensity, training volume, timing of testing, nutritional considerations and sleep. Much of the early research is also filled with methodological inaccuracies, mainly inconsistent definitions and documentation of MC phases (N. Constantini & Hackney, 2013, p. 290).

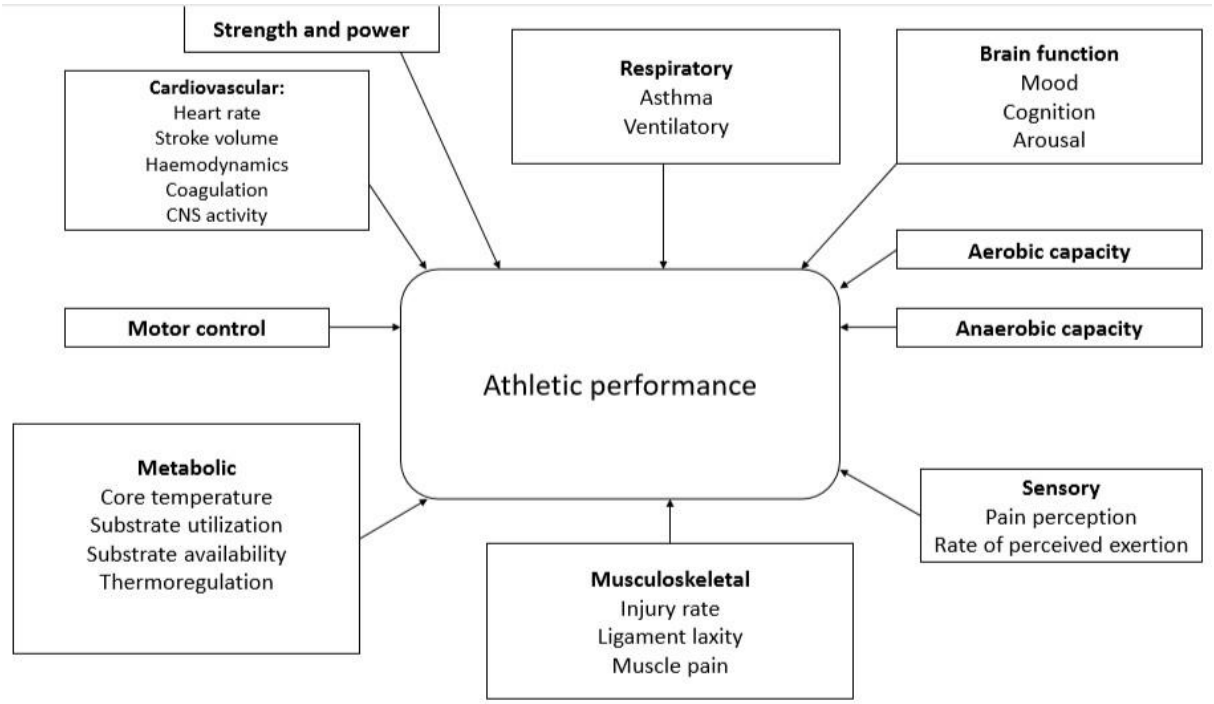

Figure 3. Components of physical performance that may be affected by endogenous hormones during the menstrual cycle. Adapted from (N. Constantini & Hackney, 2013, p. 294).

1.5 Hormonal contraceptives effect on strength and power

Studies have shown that HRT, using exogenous estrogen have positive effects on muscle strength and attenuate muscle loss in peri and post-menopausal women (Lowe et al., 2010; S. K. Phillips, Rook, Siddle, Bruce, & Woledge, 1993). This is also one of the explanations to why estrogen may ameliorate muscle strength in women (Dawson & Reilly, 2009). However, most research does not report any increased physiological effect on muscle performance or strength in women following HC usage (Elliott, Cable, & Reilly, 2005; Myllyaho et al., 2018; Nichols et al., 2008). For example, Nichols et al. (2008) reported that significant increases in strength and torque production was observed regardless of contraceptive status over a twelve-week intervention period. Further they concluded that OC's did not provide any benefit beyond the stimulus of the training intervention (Nichols et al., 2008). For women with

menstrual dysfunction or with the need for contraception, HC´s may provide a balanced hormonal milieu for training and competition, and predictable onset of menstruation. In this regard, HC's may possibly provide performance enhancement by attenuation of premenstrual symptoms (Bennell et al., 1999). Potential side effects caused by HC's can be minimized with lower doze triphasic pills and newer progestins (N. Constantini & Hackney, 2013, p. 207). Overall, further understanding of the different types of HC agents and their effect on athletic performance seems to be needed.

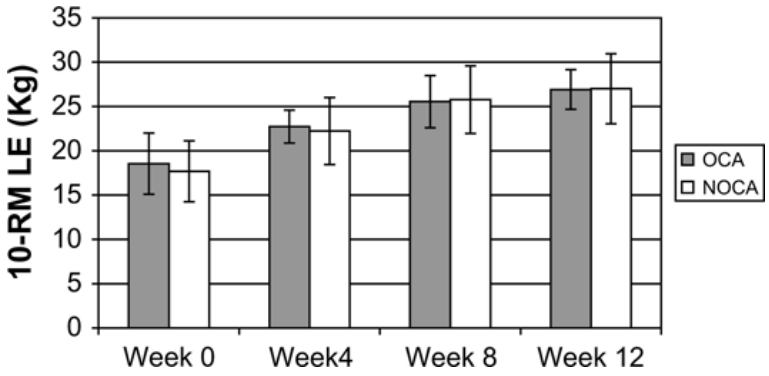

Figure 4. Illustrates Mean knee extension peak torque production during a 12-week training period for oral contraceptive agents (OCA) and non-OCA (NOCA) users. Used with permission from authors (Nichols et al., 2008).

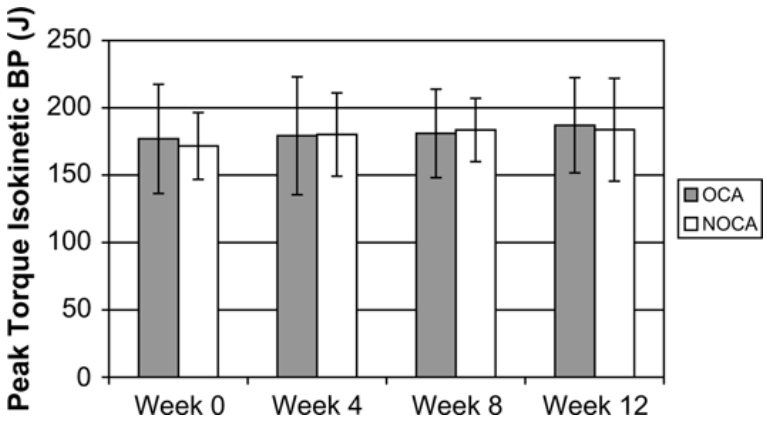

Figure 5. Illustrates Mean isokinetic bench press peak torque productions during a 12-week training period for oral contraceptive agents (OCA) and non-OCA (NOCA) users. Used with permission from authors (Nichols et al., 2008).

1.6 The menstrual cycles effect on strength and power

There are several studies reporting increased muscular strength and performance based on hormonal fluctuations through the MC, including during or just before menstruation (Bambaeichi, Reilly, Cable, & Giacomoni, 2004; S. K. Phillips et al., 1993; Sarwar, Niclos, & Rutherford, 1996; Wearing, Yuhosz, Campbell, & Love, 1972), and during the LP (Birch & Reilly, 2002; Dawson & Reilly, 2009). Indeed, muscular strength have also been shown to be ameliorated during the FP (Dawson & Reilly, 2009; S K Phillips, Sanderson, Birch, Bruce, & Woledge, 1996). In the study by Phillips et al. (1996) there was no direct correlation with estrogen levels and increase in strength, but the authors hypothesise that if the mechanism is working through the steroid receptors rather than the membrane receptor, it is possible that estrogen has a delayed onset of action. Thus, increases in strength caused by estrogen, may be imminent when circulating estrogen levels begins to decline (C. M. Lebrun, 2008, p. 48; S K Phillips et al., 1996). Several studies have also shown increased strength gains and lean body mass through FP periodized training (Reis, Frick, & Schmidtbleicher, 1995; Sung et al., 2014; Wikström-Frisén, Boraxbekk, & Henriksson-Larsén, 2017). It has been suggested that there is a link between the estrogen peak observed in the FP and an increase in muscle strength reported (Dawson & Reilly, 2009; Greeves et al., 1997; S K Phillips et al., 1996). Further, it is proposed that the increase in muscular strength is due to estrogen improving the intrinsic quality of skeletal muscle, thus increasing its ability to generate force (Lowe et al., 2010). However, there are contradictory evidence showing no effect of the MC on strength and power related performance (Abt et al., 2007; Bushman, Masterson, & Nelsen, 2006; Davies, Elford, & Jamieson, 1991; Dawson & Reilly, 2009; DiBrezzo, Fort, & Brown, 1991; Higgs & Robertson, 1981; Jonge, Boot, Thom, Ruell, & Thompson, 2001; Constance M. Lebrun, Joyce, & Constantini, 2013; C. M. Lebrun et al., 1995). For example Dawson & Reilly (2009) concluded that if there is an influence of the MC on strength, the increase in late FP and at ovulation is likely to be mediated, either directly or indirectly, by the gonadotrophin

hormones (Dawson & Reilly, 2009). Testosterone also has the potential to play a role in the variation in strength during the MC because testosterone receptors are present in muscle, with blood levels of this hormone being elevated at the time of ovulation (C. M. Lebrun, 2008, p. 48).

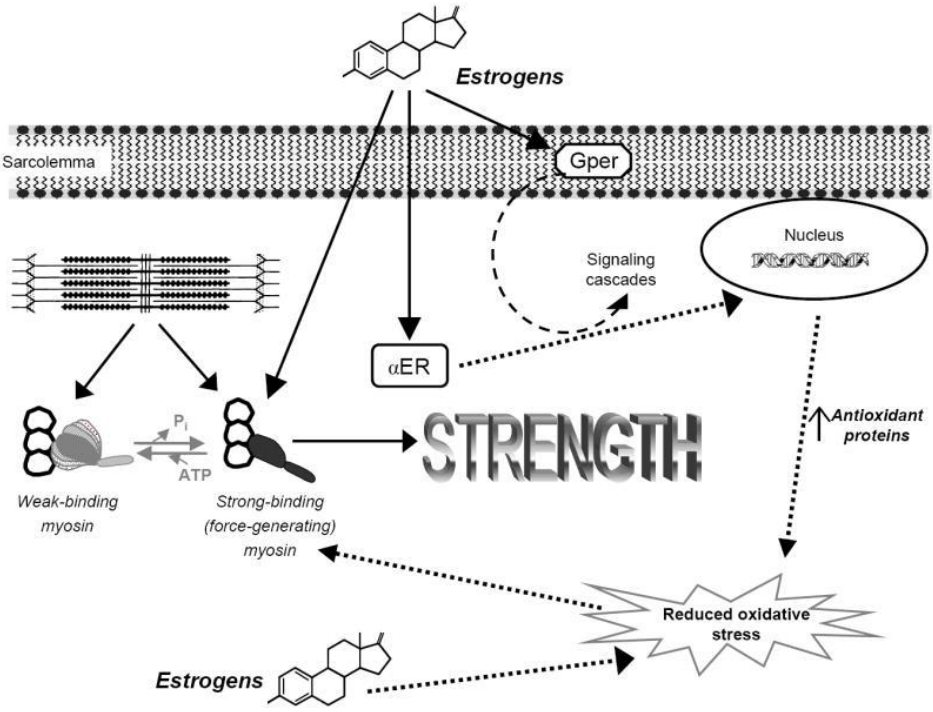

Figure 6. Illustration of how estrogen may influence muscle strength. Lowe et al. (2010) hypothesize that estrogen receptor (ER) content in muscle is responsive to circulating estrogen. Therefore, ER's initiate signalling cascades and/or regulate genes that result in a reduction in oxidative stress fibres. Estrogen may also have direct antioxidative effects. The authors speculate that a reduction in oxidative stress would preserve myosin structure function, conferring a beneficial effect on strength. ***Solid*** *arrows represent experimental evidence confirmed in the literature.* ***Dashed*** *arrows represent hypothesized mechanisms of estrogen action in skeletal muscle by the authors. Gper= G protein-coupled receptor; ATP = Adenosine triphosphate; P_i = Inorganic phosphate. The model is used with permission from the authors (Lowe et al., 2010).*

In summary, the evidence regarding the MC's effect on muscular strength and performance is inconclusive. This is partly due to methodological inaccuracies, making a comparison of findings difficult. Extrapolation of these findings to high level athletic populations is also difficult, as most studies are done in general populations. Genetic endowment has been shown to significantly influence sport performance and potential. Thus, athletes competing at high levels may react differently to hormonal fluctuations during the MC (Eynon et al., 2011). There also appears to be individual variability in response to different performance

parameters, further complicating our understanding of the MC's effect on performance in females (N. Constantini & Hackney, 2013, p. 307).

1.7 Purpose of the study

This study investigated the MC's effect on strength and power parameters over a six-week period. The main objective was to examine if there were any alterations to performance caused by the MC, in high level female team athletes by comparing a non-hormonal contraceptive group (NHCG) with a hormonal contraceptive group (HCG). Significant Differences in results were taught to be caused by the hormonal fluctuations, as the HCG would have steady circulating levels of exogenous hormones during the MC, being the only known difference between groups. Altering effects on performance caused by the MC may have implications for testing, training and competition, in that different phases of the MC could elicit different performance outcomes depending on the time of execution.

1.8 Hypothesis and research question

Research question:

Is there a difference in performance between the HCG and NHCG throughout the MC?

Hypothesis:

H0: There is no significant difference in performance between the HCG and NHCG throughout the MC.

HA: There is a significant difference in performance between the HCG and NHCG throughout the MC.

2 Material and Methods

2.1 Study Design

The design was a prospective cohort, comparing an HCG (N=23) and NHCG (N=12) once a week throughout a six-week period. Weekly outcome measures were then compared between the HCG and NHCG. This class of design is a quasi-experiment, testing descriptive causal hypotheses about manipulative causes (Shadish, 2002, pp. 13-14). This design was chosen, as it allows for similar testing of participants with HC status being the only known difference between the groups. Participants were tested at the same time and day (± 2 days) throughout the intervention period, as far as this was possible (6 week-follow up). A maximum of four participants were permitted to arrive the testing facility simultaneously, with thirty minutes between intervals. The whole testing regiment lasted between forty-five and sixty minutes. Based on self-reported use of HC's, subjects were divided into an HCG and NHCG post hoc. Thus, testers were not aware of participants HC status, minimizing the change of testing bias. MC phases were later confirmed through serum hormone levels in the NHCG and four weeks representing the FP/LP (two measurements in each phase) were selected for statistical analysis. For the HCG four weeks were chosen based on self-reported onset of menstruation. Confirmation of MC phase in females using HC is not possible, due to a steady flow of exogenous hormones throughout the MC (Allen et al., 2016). Thus, self-reported measure was used, being the only alternative within the frames of the study. However, participants in the HCG also provided blood samples for purposes beyond the scope of this master thesis. Participants were introduced to the physical tests before participation, although a separate familiarization session was not possible due to the participants competition schedule. Therefore, to minimize a learning effect, participants conducted two test attempts for each exercise before recording started.

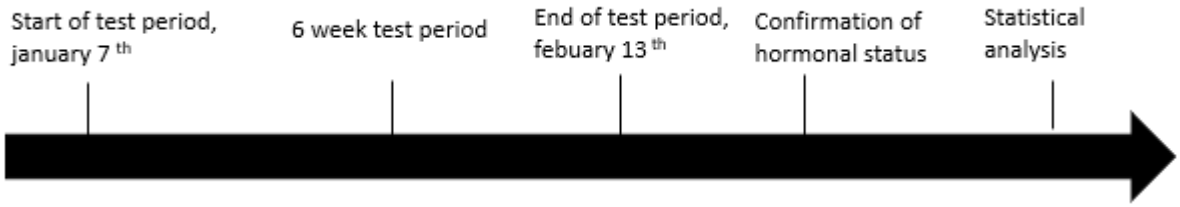

Figure 7. Timeline illustrating important timepoints during the study.

2.2 Setting

The study was part of a collaboration between HVL, the university of Bergen (UIB) with funding from *idrettscampus* Bergen. In addition, The Olympic federation of western Norway served as consulting partner. Official planning and organization of the study began in august 2018 and inclusion criteria were decided. The final study protocol, outcome variables and logistical framework was completed in November 2018. Questionnaires and testing procedures was completed in in early December 2018, and the start of the study was set to January 7th. All testing during the six-week period was performed at HVL, Hordaland, Bergen. Outcome variable testing was conducted at the faculty of sport science, while biological material was handled and stored at the faculty of biochemistry, before being transferred to Haukeland university hospital, Bergen, Norway (HUS) following the end of testing. Recruitment was conducted between august and December 2018, with data collection/testing between January 7th and February 13th. Unless indicated by blood samples, baseline questionnaire or requested by participants, no further follow up was provided following the aforementioned dates.

2.2.1 Recruitment

After finalizing the inclusion criteria's, clubs were formally contacted via official channels and informed about the project. Due to the substantial commitment of testing once a week

over a six-week period, clubs invited were restricted to the city of Bergen, Hordaland. This limited the access to players meeting the inclusion criteria of competing at the highest level in a team sport acknowledged by the Norwegian sports federation. After consulting with the Olympic center of western Norway, it was decided that one handball team (N=12) from the second highest division would be invited, to increase the sample size. This was done as they matched the elite teams in terms of training volume and fulfilled the additional criteria. Proceeding the initial information provided, teams were further instructed about the study design and its involvements. One team chose to decline the invitation for participation, due to the excessive burden the study could appose to their season. Further, teams that accepted the invitation gave players the option of declining for personal reasons, hence, not all players from the included teams were present. December 13th there was an official meeting for participants accepting the invitation, providing further information and outlining logistical instructions. Baseline questionnaire was issued to participants following acceptance of participation. As team's response rate varied, reception of the answered baseline questionnaires differed, lasting from medio December 2018 to medio January 2019. Both baseline and weekly questionnaires were administered electronically via *Survey xact*. Response time was set to 2 weeks from receiving the baseline questionnaire, with digital inquiries being issued every week after this.

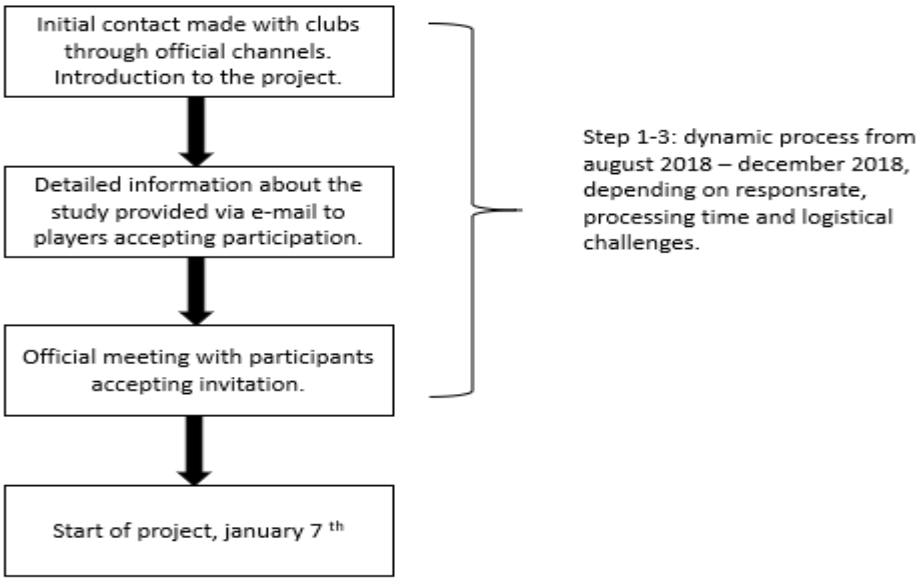

Figure 8. Illustrates the recruitment process during the initial planning phase of the study august 2018 – January 2019.

2.3 Participants

Fifty-five female athletes from the county of Hordaland, Norway were included and received the baseline questionnaire. Participants were recruited from the team sports soccer, handball and volleyball, competing at the highest national level or the second division (national division). Twelve players Were currently representing their national team. Inclusion criteria included participants being free of any injury or disease that would prohibit testing, eighteen years of age and competing at a national level in their respective team sport. Amenorrhoeic participants were excluded from the study. This was done post hoc, as response rate and handling of baseline questionnaires providing the information was not completed before January 7th.

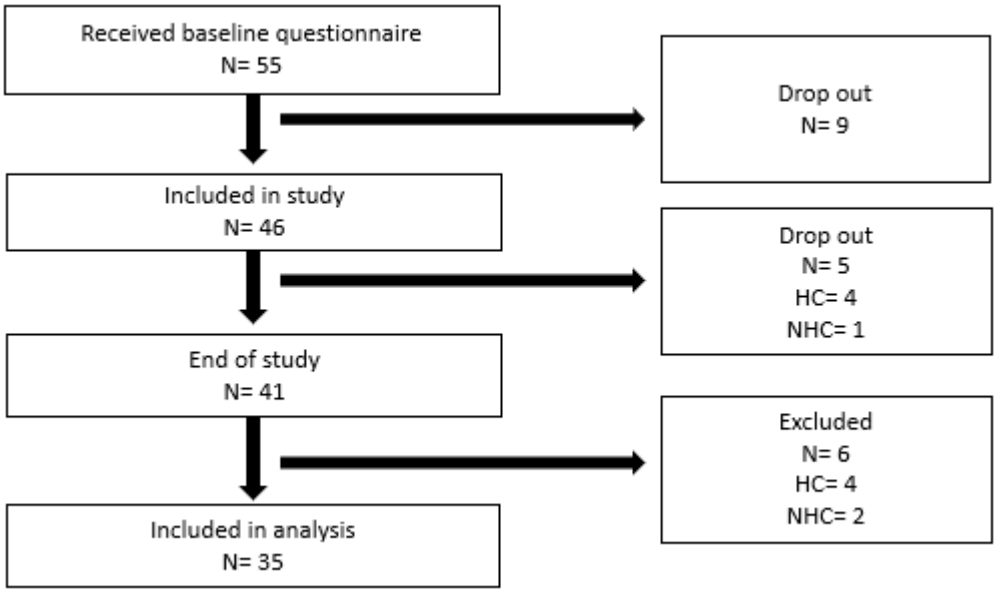

Figure 9. Flow chart displaying the drop-out rate and exclusion during the study. N= Number of participants; HC= Hormonal contraceptive user; NHC= Non-hormonal contraceptive user. HC status for drop out prior to the start of the study is not included, due to incomplete questionnaires from some of the participants.

Table 1. Hormonal contraceptives used by participants in study (HCG).

Name of product	Type	Ingridients
Mikrogynon	Monophasic combination pill	Levonorgestrel, Ethinylestradiol
Loette 28	Monophasic combination pill	Levonorgestrel, Ethinylestradiol
Orcalon	Monophasic combination pill	Levonorgestrel, Ethinylestradiol
Carazette	Progestogen-only pill	Desogestrel
Yasmin	Monophasic combination pill	Drospirenone, Ethinylestradiol
Almina	Monophasic combination pill	Levonorgestrel, Ethinylestradiol
Diane	Monophasic combination pill	Cyproterone, Ethinylestradiol
Nexplanon	Implant	Etonogestrel
Jaydess	Interauterine system	Levonorgestrel

Table 2. Showing participant working status. Full time professional= no other obligations outside of sport. Full time job= working full time in addition to training and competition in sport. Part time job= part time % job in addition to training and competition in sport. Student= Full or part time study in addition to training and competition in sport.

Status	Number of participants
Full time professional	1
Full time job	1
Part time job	7
Student	37

2.4 Procedures

The testing protocol consisted of maximum voluntary isometric grip strength (MVIG), 20-meter sprint, counter movement jump (CMJ) and leg-press. At the start of every visit, participants answered a questionnaire and body composition was measured electronically using In-body 720 (Biospace, Tokyo, Japan). Blood samples were collected before initiating the physical testing, to avoid any alterations in serum blood levels following physical strain. They were also instructed to avoid any caffeine consumption 12 hours preceding every visit, as caffeine is shown to have ergogenic effects related to sport performance (Goldstein et al., 2010; Grgic, Grgic, et al., 2019). On the day of testing, participants were encouraged to eat

approximately the same type and amount of food and liquid over the six-week period. Testing was conducted on the same day (± 2 days) and time (± 2 hours) to control for circadian variations. The ambient conditions were always held between 18-20 ° C.

2.5 Outcome measures

At every visit prior to testing, with the exception of MVIG, participants followed a standard 15-minute warm-up on a stationary bicycle ergometer (wattbike Ltd, Nottingham, UK) holding approximately 100 watts (W).

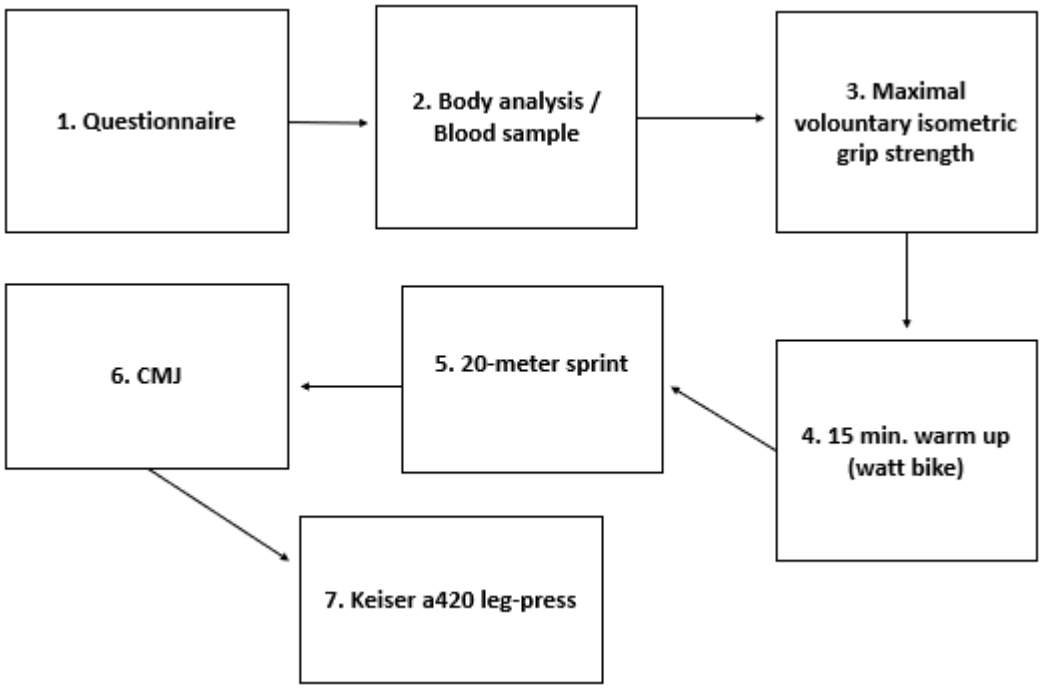

Figure 10. Process illustrating the chronological order of measurements during testing.

2.5.1 Questionnaires

After initial recruitment, subjects completed the LEAF-1 questionnaire at baseline, detailing demographic characteristics, menstrual information, HC status, training status, previous injuries and general health. The LEAF-Q questionnaire has been validated for use to identify female endurance athletes at risk for developing the female athlete triad, but can also be used for female athletes in general (Melin et al., 2014). Further, at every visit, participants were asked to complete a questionnaire designed specifically for this study, including self-reported onset of menstruation, nutritional intake, subjective expectation for testing, alcohol consumption, injury/pain information and sleep hygiene. For self-reported onset of menstruation, time since last bleeding was used to identify MC phase. An alternative for participants currently menstruating were also provided. During testing, a staff member was always present to assist participants with the questionnaire if needed. The questionnaires can be found in appendix two and three.

2.5.2 Anthropometric measurement

Anthropometric measurement was completed at approximately the same time throughout the six-weeks. Participants were asked to use the same type of sportswear at every visit and was measured with clothes, subtracting 1 kilogram (KG) from their bodyweight through a built-in mechanism in the machine. The monitor displaying results was covered during measurement to avoid influencing the participants. Height (cm), weight (KG) and fat free mass (FFM) (kg) were recorded and can be found in table five.

2.5.3 Maximal voluntary isometric grip strength

Maximal voluntary isometric grip strength (MVIGS). Isometric grip strength of the dominant hand was measured using a digital pinch/grip analyser (MIE, medical research Ltd, Leeds, UK). The evidence regarding validity and reliability of handheld dynamometry is extensive

(Stark, Walker, Phillips, Fejer, & Beck, 2011). During the test, participants were seated with a slight forward bend of the trunk, their elbow resting on the thigh with 90 ° elbow flexion. Participants were instructed to exert maximal force for 3-5 seconds. If they deviated from the instructions given, their attempt was disallowed. Two attempts were completed for each subject. If the force produced in the last attempt exceeded the previous with >5%, a new attempt was performed. The monitor displaying results was turned away from the participants, so that they would not be affected by their score. There was a 30 second break between attempts. Standard instructions were given to participants before and during the test. The best recording was used for data analysis.

2.5.4 20-meter sprint

20-meter sprint. Sprint performance was measured over a 20-meter track which is illustrated in figure eleven. The track was made out from a portable non-slipping surface (Hitashita international, ON, Canada) and was fixed to the ground. Times were recorded at 5, 10 and 20-meters using single beam photocells (Brower timing systems, Utah, USA), a reliable and commonly used measurement tool for evaluating sprint performance and running speed (T. A. Haugen, Tonnessen, & Seiler, 2012; Shalfawi, Enoksen, Tønnesen, & Ingebrigtsen, 2012). Photocells were fixed at 120 cm, except for the first pair of photocells, which were fixed at 10 cm. All sprints were performed from a standing start, with the dominant foot behind and a slight forward bend of the trunk. Each participant carried out two attempts separated by two minutes of rest. If the second attempt was > 5% faster than the previous one, a new attempt was performed. Participants decided themselves when to start each test from the starting position, with recording being initiated by interception of the photocell beam. The best 20-meter total time was used for data analysis.

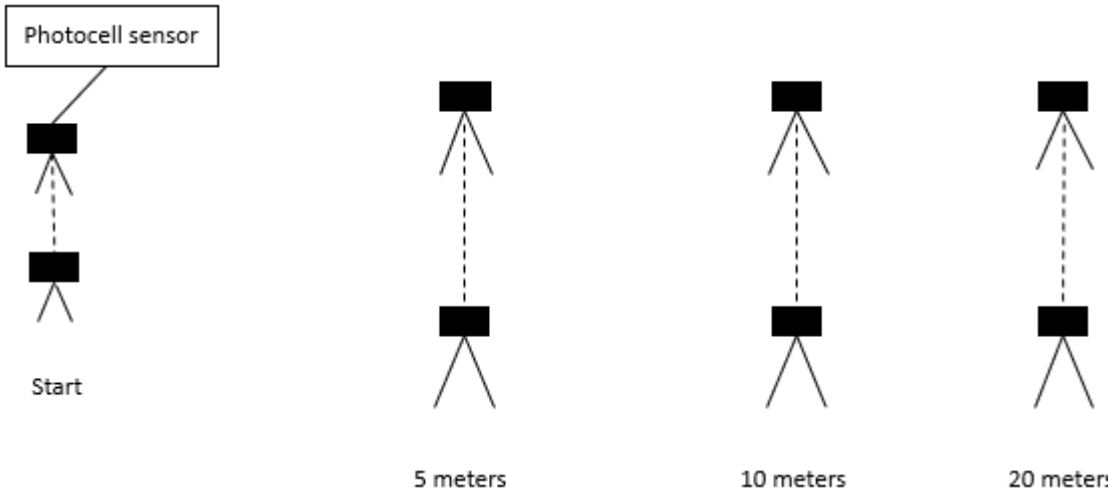

Figure 11. Illustration of the 20-meter sprint track and photocell sensor set-up. The first photocell sensors were set up at calf height, to initiate the timing sequence at first movement of the feet.

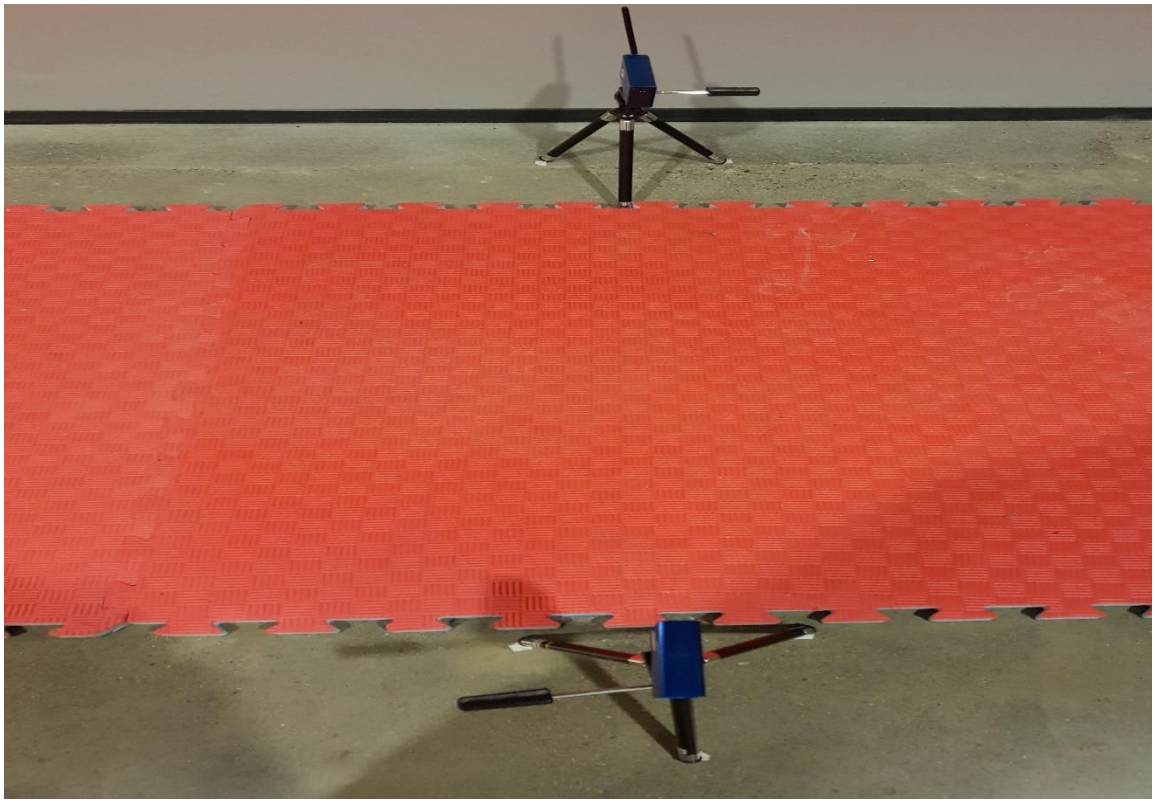

Figure 12. Picture of the first photocells sensors (start) from the 20-meter sprint course. Sensors at 5,10 and 20 meters were elevated at waist height to prohibit early interception of the laser beam.

2.5.5 Countermovement jump

Countermovement jump (CMJ). CMJ performance was measured using a force plate (Kistler instruments, Hampshire, UK). When utilizing force plates, the vertical velocity of the center of mass is calculated by using the vertical force trace. An equation of uniform acceleration can then be used to calculate jump height of an individual (Moir, 2008). This measuring device is highly prevalent in sports science and a valid measure of jump height (Buckthorpe, Morris, & Folland, 2012). Participants were instructed to lower their legs as far as they wanted, with a countermovement following the eccentric phase of the motion. The hands were held at the hips throughout the entire movement to avoid excessive momentum. If they deviated from the standard instructions given, the attempt was disallowed. Participants performed two attempts, with a minimum of 30 second rest between efforts. If the last attempt exceeded the previous with > 1 cm, a new attempt was made. The best result was used for data analysis.

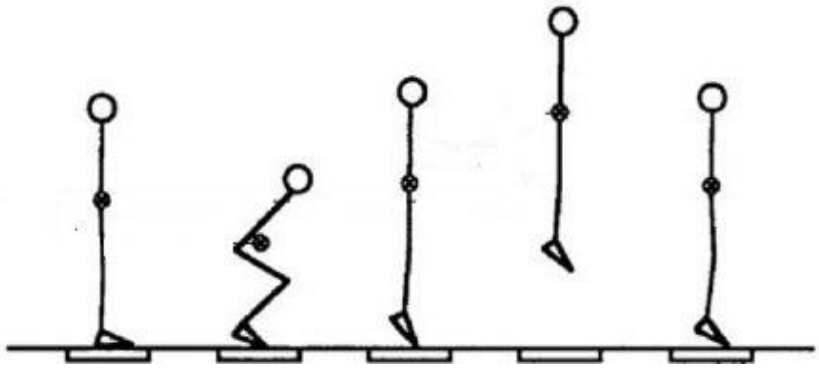

Figure 13. Illustration of the CMJ performed on the force platform, adopted with permission from the author (Mandic, Knezevic, Mirkov, & Jaric, 2016). Changes were made to the illustration to fit the intended purpose.

2.5.6 Leg press (Relative peak power)

Leg press. Maximal concentric strength and power was measured using computer interfaced Keiser a420 pneumatic leg press (Keiser, Fresno, CA), developed for research purposes. Participants completed a 10-repetition pre-established test developed by Keiser for the a420

machine. The test requires an estimated 1-repetition maximum weight, which were chosen based on participants self-reported training history, bodyweight, sport and previous internal testing of comparable demographics. The software then adjusted the resistance for each repetition and prompted the user for time to do each repetition. Resistance gradually increased in pre-established steps until participants reached failure. Rest time between repetitions increased together with the resistance to avoid fatigue. Verbal encouragement was given during the test to facilitate maximal effort. The test started with the femur in 90° angle vertical to the ground, and the feet placed in a pre-established position. Participants were instructed to keep their hands on the handles of the machine and to exert maximal effort for every repetition. Results are reported as Relative peak power (RPP) KG/watt (W).

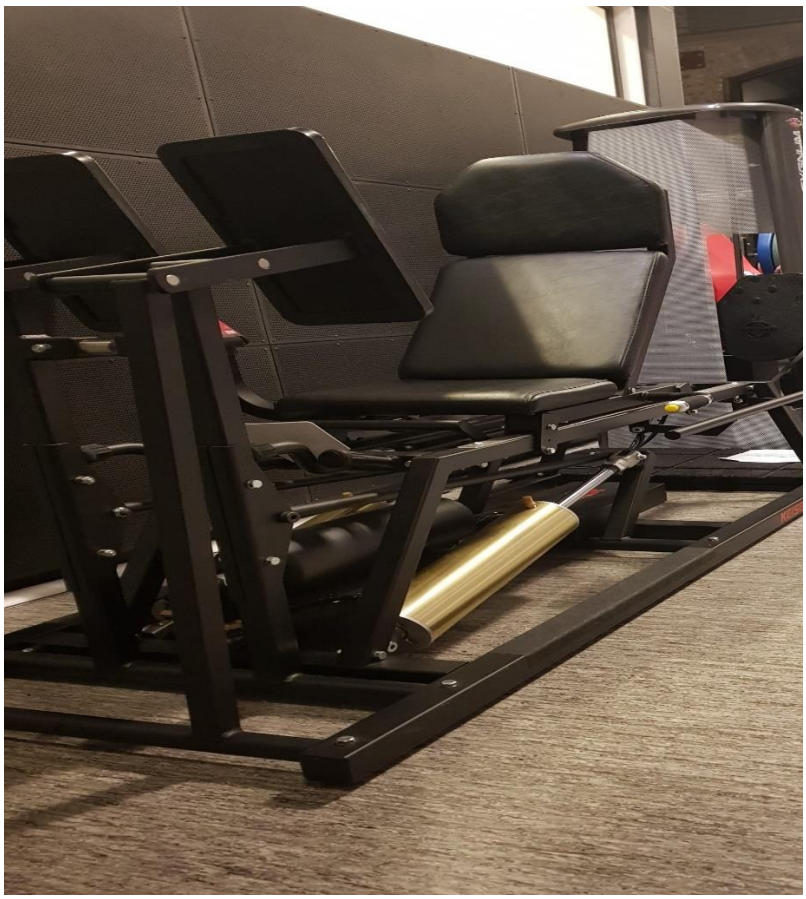

Figure 14. Picture of the Keiser A420 being used in the project.

2.5.7 Blood analysis

Venous blood samples were collected from participants at the start of every visit, after completing the questionnaire and anthropometric measurement. Blood was collected using eclipse blood collection needles (BD Vacutainer, Franklin Lakes, NJ) by qualified biomedical laboratory scientists (BLS). Blood samples were then centrifuged at 2000 RPM for ten minutes (Thermo scientific SL1R centrifuge, Thermo Fisher, Waltham, MA, USA) and stored at -80° C, until analysis. All blood sample analysis was conducted at HUS and followed official international guidelines. The blood samples were transferred to HUS at the end of the intervention period, February 13th.

Blood samples were analysed for progesterone, estrogen (estradiol), FSH, LH and SHGB. Analysis of progesterone and estrogen levels were performed using liquid chromatography-mass spectrometry. FSH, LH and SHGB was analysed using chemical luminescence methods (Immunlite 2000 XPi, siemens, Erlangen, Germany).

2.6 Determination of menstrual cycle phase

For confirmation of MC phase, serum hormone levels were compared to reference levels for FSH, LH, estrogen and progesterone provided by HUS. The results were analysed by licenced medical staff, according to their guidelines which is updated regularly. For estrogen, reference values are 143-1615 pmol/L for the whole MC (Haukeland, 2019d). Reference values for FSH, LH and progesterone are presented in table three and four.

Table 3.Reference values for luteinizing hormone (LH) and Follicle-stimulating hormone (FSH) in the follicular phase (FP), ovulation and luteal phase (LP) stated as IE/L (Haukeland, 2019a, 2019b).

	LH (IE/L)	FSH (IE/L)
Follicularphase	1,1-11,6	2,8-11,3
Ovulation	17,7-77	5,8-21
Lutealphase	< 14,7	1,2-9

Table 4. Reference values for progesterone, stated as nmol/L. The table illustrates the serum hormonal ranges used to determine the day of the MC (Haukeland, 2019c).

Day of MC	Progesterone (nmol/L)
0-11	< 3,6
12-15	1,5-5,5
16-19	5,5-8,5
20-22	19-76
23-35	4,9-13

2.7 Ethical considerations

All individuals participated on a voluntary basis, receiving detailed information about the study protocol, before accepting the invitation. They were informed about the possibility to drop out at any time and had to sign an informed consent form (appendix x).

The project was supervised and administered by licensed medical professionals, thus, it was admitted under the patient injury act. A licenced attorney and medical doctor were made available for participants if needed. Potential irregularities in blood samples analysed were followed up by a licenced physician. All blood samples were stored in a biological blood bank at HVL and personal information handled without the possibility of identification in accordance with the privacy act. The study protocol was approved by the regional ethical

committee of western Norway (REK) and by the ethical committee of HVL (appendix 4). No adverse events were reported due to the intervention of the study.

2.8 Statistical analysis

Statistical analysis was performed using IBM SPSS 25 (IBM, Armonk, NY, USA). One-way repeated measures analysis of variance (ANOVA) were used to analyse for group differences throughout the MC. Bonferroni correction post-hoc were conducted to reduce the chance of type one error with significant ANOVAS. Significance level were set at $p < 0.05$ a priori. Participants with incomplete data were excluded from the statistical analysis. Descriptive statistics were used to provide participant characteristics. All values are reported as mean ± standard deviation (SD) unless specified. Between group comparison is based on the interaction between hormonal status*menstrual cycle (time).

2.8.1 Repeated measures ANOVA

Repeated measures design (GLM) is a statistical model were the same individuals participate in all conditions of the study. By using this model, we can better control for individual differences related to the variables we are investigating. We can achieve this by testing the same participants at different time points, as done in this prospective study design (Field, 2009, p. 458).

The accuracy of the ANOVA depends on the assumption that scores are independent, this assumption is violated when repeated measures are conducted. Consequently, the traditional F-test will lack accuracy. To avoid false positives (type 1 error) the assumption of sphericity must be met for repeated measures designs, when using within-subjects effect. Sphericity refers to the condition where variances of the differences between all repetitions are equal. Mauchly's test of sphericity tests the hypothesis of equality between variances of differences.

A significant test of $P < 0.05$ concludes that there are significant differences and that assumption of sphericity has been violated (Field, 2009, pp. 459-460). If we are faced with a significant Mauchly's test, the use of a multivariate test of repeated measures is possible, as the assumption of sphericity is not needed for this test. We can also look at the interaction between independent variables, in this case, allowing us to compare the effect between the HCG and NHCG (Field, 2009, pp. 476-477, 585; O'Brien & Kaiser, 1985). When using GLM designs, participants missing one or several measurement points will be excluded from the analysis. Hence, for this study, the number of participants for each outcome variable will vary, as some participants did not complete every test on all measuring timepoints. Using mixed model designs, we could have avoided this, as this method will account for missing variables. However, with GLM being highly prevalent in the existing related literature and the number of missing variables proving quite low overall, it was decided to stick with GLM for this master thesis.

2.8.2 Post hoc analysis

Post hoc tests, refers to the analysing of experimental results. They are generally based on the family wise error rate (FWE), which is the probability of at least one false positive in a data set. Post hoc tests use pairwise comparisons, comparing all different outcomes of the treatment groups, based on the t-test. The Bonferroni correction divides the alpha level by number of comparisons, ensuring that the cumulative type 1 error is below 0.05 (Field, 2009, pp. 372-374) In this experiment, Bonferroni correction was made post hoc in the event of significant ANOVAS, ensuring that FWE did not occur. The Bonferroni correction suffers from loss of power, as it is quite stringent, increasing the risk of type 2 error.

3 Results

3.1 Descriptive statistics

Table 5. Descriptive data showing mean value ± standard deviation (SD) of participant characteristics in hormonal contraceptive group (HCG) and non-hormonal contraceptive group (NHCG). N=number of participants

	HCG	NHCG
N	23	12
Age	20,5 ± 2,6	22,5 ± 4,2
Height (cm)	171 ± 7,6	169 ± 8,6
Weight (kg)	66,9 ± 8,3	63,3 ± 8,9
Fat free mass (FFM)	53,4 ± 6,7	51,4 ± 7,6
Training volume (Monthly hours)	52,7 ± 14,8	52,9 ± 13,7

Table 6. Comparison of self-reported onset of menstruation and onset of menstruation based on serum hormonal levels in the NHCG.

Individual	Self reported onset of menstruation (week)	Hormonal confirmation of menstruation (week)
1	4	4
2	5	5
3	3/4	4
4	2/5	2/5
5	No reported onset of menstruation	No test values during menstruation
6	3	3
7	2/6	2/6
8	No onset of menstruation	Between week 2 and 3
9	2	2
10	1/6	1/6
11	2	2
12	No onset of menstruation	Between week 1 and 2

3.2 Maximal voluntary isometric grip strength

There was no statistically significant difference between groups for MVIGS. Repeated measures ANOVA showed that the result was not significantly affected by HC status, F (3,29)

=0,362, P > 0.05. Overall, both groups demonstrated weekly variations in MVIGS, week four (week of the menstrual cycle) displaying the highest values for both groups with a mean of 31,3 ± 5,6 kg in the HCG and 29,3 ± 4,9 kg in the NHCG respectively (Table 7). Results are illustrated in figure fifteen.

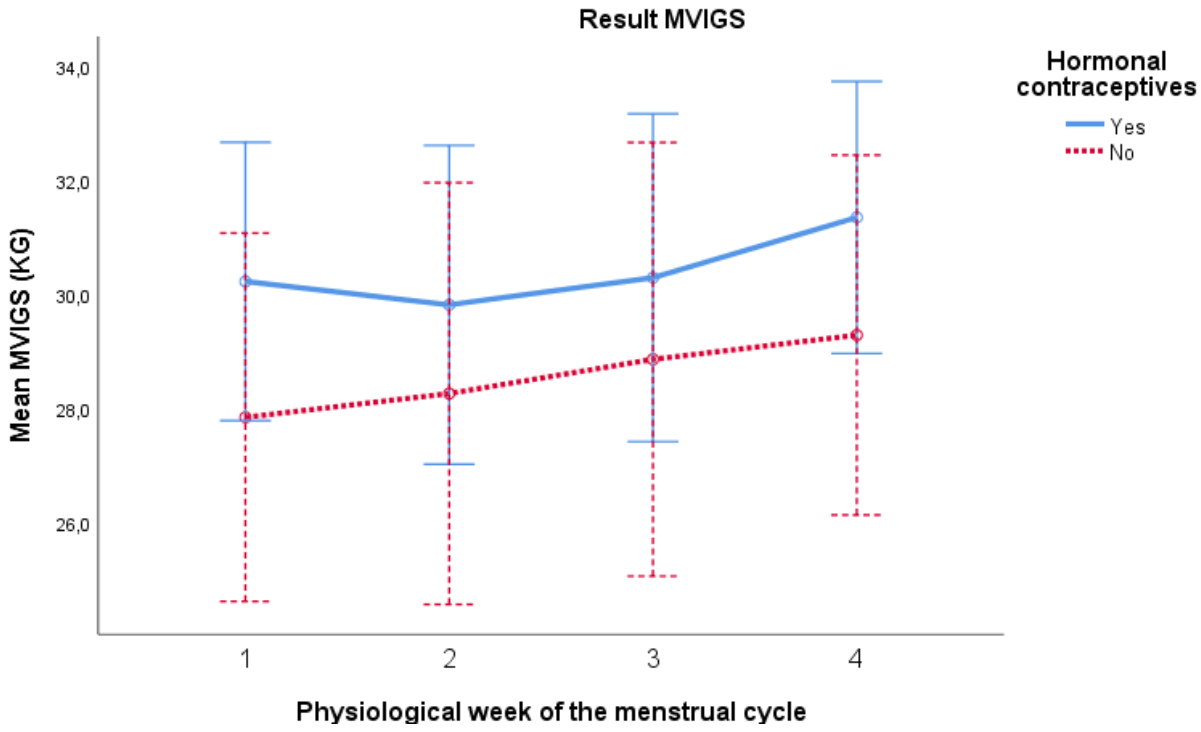

Figure 15. Mean values through the menstrual cycle with 95% confidence intervals. MVIGS = Maximal voluntary isometric grip strength. Week 1-4 represents the physiological weeks of the menstrual cycle based on serum hormone levels in the Non-hormonal contraceptive group (NHCG) and self-reported onset of menstruation in the hormonal contraceptive group (HCG).

Table 7. Descriptive statistics showing between group comparison of maximal voluntary isometric grip strength (MVIGS) through the menstrual cycle. HCG= Hormonal contraceptive group, NHCG= Non-hormonal contraceptive group.

	HCG			NHCG		
MC	N	Mean (kg)	SD ±	N	Mean (kg)	SD ±
Week 1	21	30,2	5,8	12	27,8	4,9
week 2	21	29,8	6,7	12	28,3	5,5
week 3	21	30,3	7,1	12	28,9	5,1
week 4	21	31,3	5,6	12	29,3	4,9

3.3 Sprint

There was no statistically significant difference between groups for 20-meter sprint performance. Repeated measures ANOVA showed the result was not significantly affected by HC status, F (3,24) =0,710, P > 0.05. Both groups showed consistent measures through the four weeks of the MC with results ranging from 3,209 ± 0,123 seconds to 3,194 ± 0,132 seconds in the HCG and 3,232 ± 0,114 seconds to 3,203 ± 0,114 seconds in the NHCG respectively (table 8). The results are illustrated in figure sixteen.

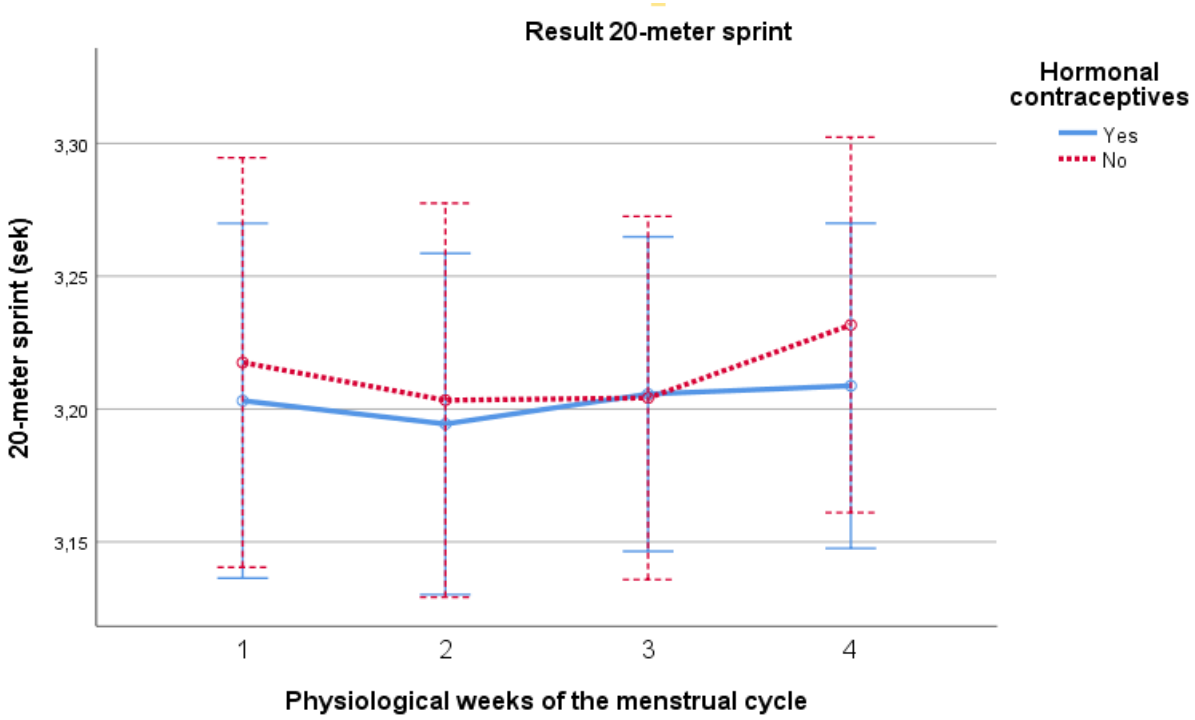

Figure 16. Mean values through the menstrual cycle with 95% confidence intervals for 20-meter sprint.

Table 8. Between group comparison through the menstrual cycle for 20-meter sprint. (s)= seconds

	HCG			NHCG		
MC	N	Mean (s)	SD ±	N	Mean (s)	SD ±
Week 1	16	3,203	0,134	12	3,218	0,123
week 2	16	3,193	0,132	12	3,203	0,114
week 3	16	3,206	0,125	12	3,204	0,100
week 4	16	3,209	0,123	12	3,232	0,114

3.4 Counter movement jump

There was no statistically significant difference between groups for the CMJ. Repeated measures ANOVA showed the result was not significantly affected by HC status, F (3, 26) = 2,361, P > 0.05. The CMJ displayed a greater between group difference compared to the other tests, with means ranging from 0,330 ± 0,063 meters to 0,307 ± 0,043 meters in the HCG and 0,306 ± 0,030 meters to 0,301 ± 0,025 meters in the NHCG respectively (table 9). The results are illustrated in figure seventeen.

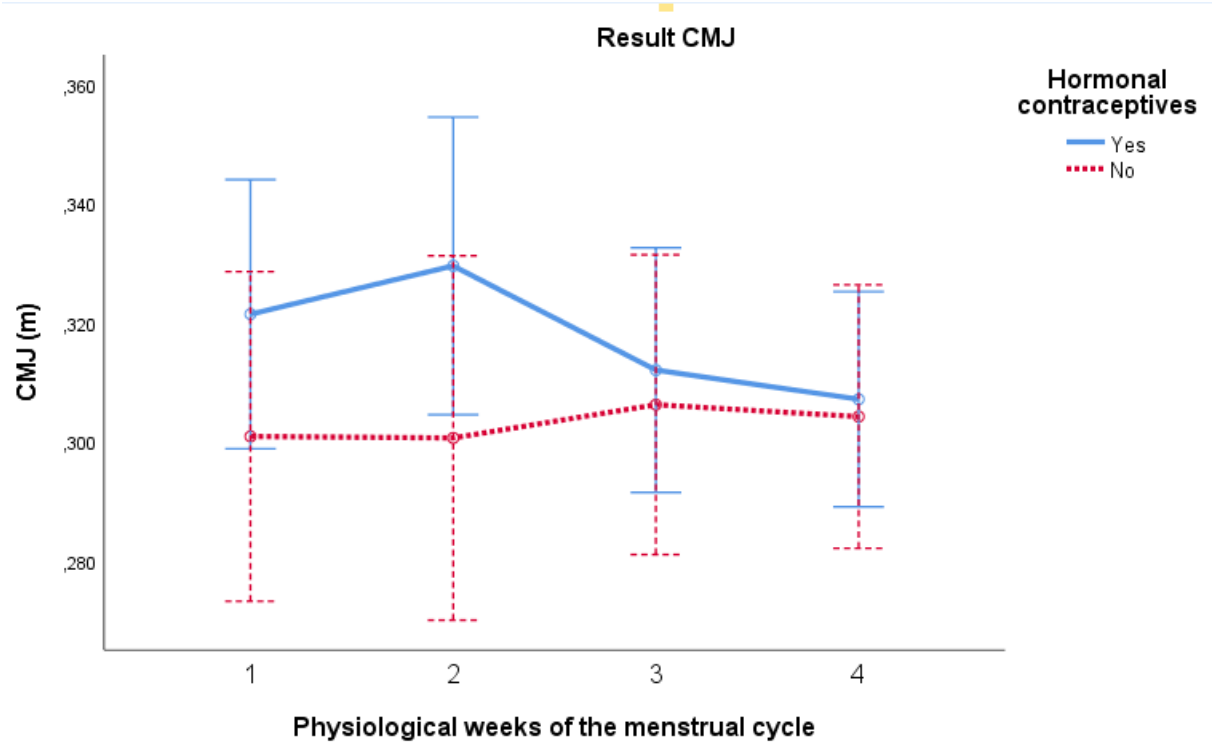

Figure 17. Mean values for the physiological weeks of the menstrual cycle for HCG and NHCG with 95% confidence intervals.

Table 9. Between group comparison of CMJ through the menstrual cycle. CMJ = Counter movement jump, (m)= meters.

	HCG			NHCG		
MC	N	Mean (m)	SD ±	N	Mean (m)	SD ±
Week 1	18	0,321	0,056	12	0,301	0,025
week 2	18	0,330	0,063	12	0,301	0,027
week 3	18	0,312	0,049	12	0,306	0,030
week 4	18	0,307	0,043	12	0,304	0,026

3.5 Leg press

Relative peak power (RPP) was selected for statistical analysis of the leg press. This parameter reflected the overall performance from the leg press, adjusting for the individual difference in bodyweight. There was no statistically significant difference between groups in RPP (peak power/bodyweight). Repeated measures ANOVA showed the result was not significantly affected by HC status, $F(3, 26) = 1{,}746$, $P > 0.05$. The NHCG displayed greater variation between weeks compared to the HCG with means ranging from 24,1 ± 2,5 W to 22,9 ± 2,6 W and 23,6 ± 2,6 W to 23,2 ± 2,8 W (table 10) respectively. The results are illustrated in figure eighteen.

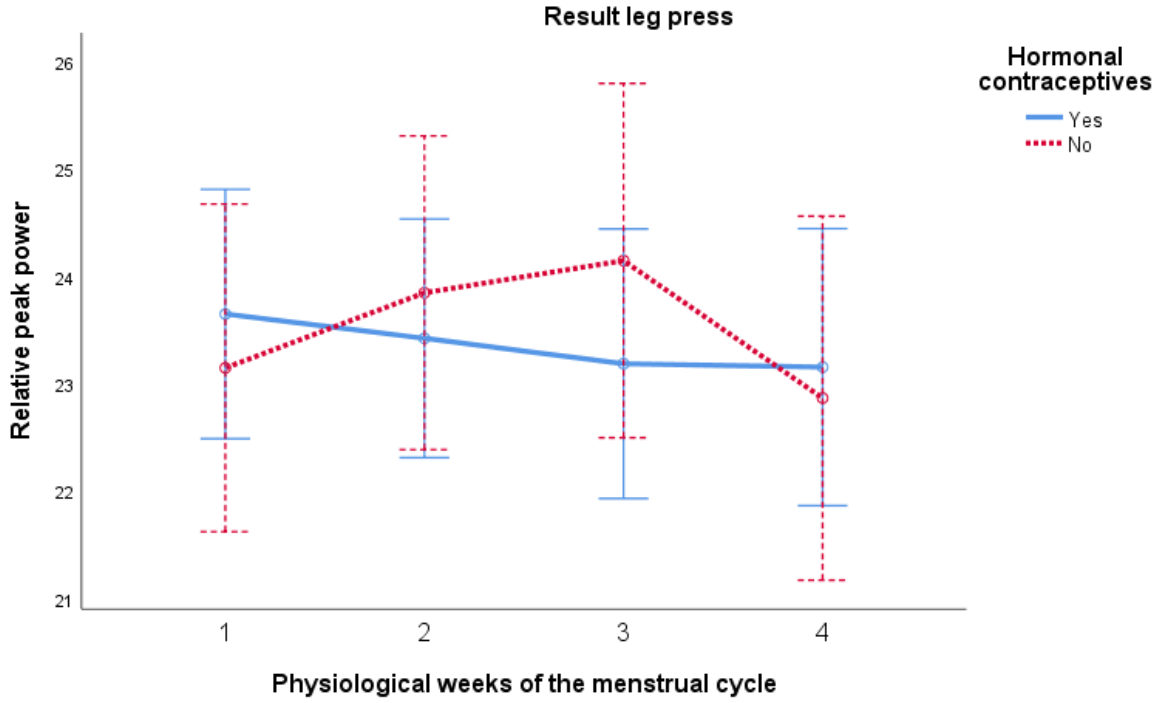

Figure 18. Mean values for the weeks of the menstrual cycle with 95% confidence intervals.

Table 10. Between group comparison through the menstrual cycle. (W)= Watt

	HCG			NHCG		
MC	N	Mean (w)	SD ±	N	Mean (W)	SD ±
Week 1	19	23,6	2,6	11	23,1	2,3
week 2	19	23,4	2,4	11	23,8	2,4
week 3	19	23,2	2,8	11	24,1	2,5
week 4	19	23,2	2,8	11	22,9	2,6

3.6 Comparison of outcome measures

*Table 11. Between group comparison of Wilks' Lambda (multivariate test) for MVIGS, 20-meter sprint, CMJ and relative peak power based on the interaction HC status*Menstrual cycle.*

	Value	F	Hypothesis df	Error df	Sig.
MVIGS	0,964	,361[b]	3,000	29,000	0,782
20-meter sprint	0,918	,710[b]	3,000	24,000	0,556
CMJ	0,786	2,361[b]	3,000	26,000	0,094
Relative peak power	0,832	1,746[b]	3,000	26,000	0,182

4 Discussion

4.1 Main results

This study sought out to investigate the MC's effect on strength and power performance parameters in high level female team athletes. Methodologically, this was done by comparing the two groups NHCG and HCG, the latter working as a control group. The main findings of the study show that there is no statistically significant difference between the groups, suggesting that HC status does not alter strength and power performance in high level female team athletes, when tested in a controlled environment. This is in line with several studies, reporting no difference in performance through the different phases of the MC, including (C. M. Lebrun et al., 1995) (Abt et al., 2007; Bushman et al., 2006; Jonge et al., 2001). Indeed, these studies are performed in different demographics. However, this current study indicates that highly trained female team athletes does not differ in their response to hormonal fluctuations occurring during the MC.

4.2 Outcome variables

The participants of the study were highly trained athletes, competing in sports were strength and power is significantly related to performance. Thus, the hypothesis that the MC could alter performance through its different phases is highly relevant, as it could have major implications for competition and testing. Previous studies demonstrating altering effects on strength and power performance, elicited by hormonal fluctuations are in large intervention studies, utilizing periodized training protocols (Reis et al., 1995; Sung et al., 2014; Wikström-Frisén et al., 2017). For example, Sung et al. (2014) reported increased muscle strength in participants following a FP periodized training regimen compared to the LP over three MC's. This, in contrast to acute performance as investigated in our study, could better utilize the proposed mechanism behind estrogens ameliorating effects on muscle strength (Lowe et al.,

2010). This could be one explanation to why none of the outcome variables in our study showed statistically significant results, with CMJ demonstrating greatest between group difference P = 0.094. Indeed, Phillips et al. (1996) reported up to 10% increase in Maximal voluntary force of the adductor pollicis during the FP of the menstrual cycle (S K Phillips et al., 1996). The authors argue that this test requires no learning, reducing the influence of psychological factors on the results. This is contradictory to our results using MVIGS with a handheld dynamometer, which is a comparable test.

Multiple variables were tested during the six-week testing period for this study including MVIGS, CMJ, 20-meter sprint and leg press (RPP). The outcome variables were chosen as they are highly prevalent in sports science literature and related to the nature of team sports. The exception being MVIGS, chosen due to its simplicity and validity. Indeed, this measurement is also prevalent in the literature. None of the dependent variables investigated approached the a priori set significant level of $p < 0.05$, indicating that strength and power performance does not significantly vary depending on serum hormone levels. Further, several dependent variables increase the chance of FWE, hence, the need for post hoc correction given significant results (Knudson, 2009). However, despite the increased likelihood of results approaching significance of $p < 0.05$ due to FWE, none of the dependent variables did. This reinforces the non-significant findings, showing no difference between groups, indicating no effect of the MC on strength and power performance on a group level.

MVIGS displayed small weekly variations in performance for both groups with the greatest variations between week one and four for both groups, with a mean difference of 1,12 kg for the HCG and 1,4 kg for the NHCG respectively. Week four produced the greatest overall score for both groups. This cannot be contributed to learning effect, as the weeks are based on self-reported menstruation and hormonal confirmation, representing the physiological week of the menstrual cycle, not the chronological order of completed measurements. As both groups displayed variations in their score, it is difficult to determine if the MC impacted these variations in the NHCG as suggested by the alternative hypothesis.

CMJ also displayed slight weekly variations in both groups, however, the HCG showed the greatest deviation with a mean difference of 1,7 cm between week one and four, which is a 5% increase. This could be viewed as a substantial difference, considering the marginal differences in high level sports. A different pattern manifested itself in the HCG for CMJ, with a FP peak before declining in the LP. If estrogen indeed does ameliorate muscle strength and power, this FP increase in performance would be plausible. However, with HC agents providing a steady supply of exogenous hormones throughout the MC, minimizing the hormonal fluctuations, this weekly difference in the HCG was unexpected. As highlighted by Myllyaho et al. (2018), different HC agents may exert varying effects on performance. This, However, is not yet thoroughly investigated in the literature and therefore cannot be determined.

RPP showed weekly variations, with the NHCG displaying the greatest change between week one and three, with a mean difference of 1 W/kg. Also, for this variable, the greatest value manifests itself in the LP of the MC, coinciding with MVIGS. This is in line with Birch & Reilly. (2002) who reported increased maximal voluntary force in the LP (Birch & Reilly, 2002). However, MC phase was confirmed via temperature and moliminal symptoms, demonstrating low accuracy of measurement (Allen et al., 2016).

20-meter sprint displayed the least variation in both groups, with quite stable result across the intervention period. Evaluating this outcome variable, it is possible that 20-meter is too short of a distance to accurately measure any significant variations between weeks, as results were very marginal. As highlighted in the systematic review by Haugen & Buchheit (2016), the uncertainty of detecting small changes is substantial. The aforementioned authors also discourage the use of single beam photocells for 10 and 20-meter sprints, due to large absolute errors (T. Haugen & Buchheit, 2016). Double beam photocells could have added increased the accuracy, but these were not available within the budget of the study. Initially a 40-meter sprint was planned for the study protocol, however, due to limited space in the research facility a 20-meter track was used. Thus, the sensitivity of this variable is questionable.

Although no significant findings between groups were found, several of the variables do show significantly weekly variations in performance for both the HCG and NHCG. As small changes in performance may impact the results in high level sport, our findings indicate that the time of testing could yield different results for high level female team athletes in general. However, based on our results and hypotheses, these variations cannot be directly contributed to hormonal fluctuations occurring during the MC, as no difference between groups were observed. A plausible explanation for the weekly variations observed during this study is individual physiological load. Andersson et al. (2008) showed that full match recovery in female soccer players can last up to seventy-two hours. The recovery time from handball is also shown to be substantial (Andersson et al., 2008; Ronglan, Raastad, & Børgesen, 2006). Thus, Individual training and competition strain could be one explanation to weekly variations, as testing in a fatigued state could alter results. The aforementioned studies also indicate that player position and sport could affect the outcome variables, causing weekly variations in this study.

4.3 Estrogens effect on strength and power

As highlighted in the introduction, estrogen has been proposed as the main hormone contributing to increases in muscle strength and power in females. This study contraindicates the hypothesis that estrogen ameliorates muscle strength and power production. This supports the findings of Jonge et al. (2001), showing no correlation between estrogen serum hormonal levels and muscle strength, fatiguability and contractile properties (Jonge et al., 2001). For this study (master thesis), estrogen levels during the FP varied through the range from 143 pmol/l to 1615 pmol/l. As highlighted by Jonge et al. (2001), the large variations seen in serum estogen levels is partly caused by secretory pulses of these hormones. In addition, the MC is highly individual in their length, thus, the participants may have been in different stages of their respective cycle phases. We adjusted for this with the six- week design, providing us with two measurements in each phase. Regardless, no effect of estrogen on strength or power where observed on a group level. This is also in line with Greeves et al. (1997) who demonstrated that supraphysiological doses of estrogen did not significantly increase muscle strength, questioning the findings of Phillips et al. (1996) and Sarwar et al.

(1996) suggesting estrogen ameliorates muscular strength (Greeves et al., 1997; S K Phillips et al., 1996; Sarwar et al., 1996). Further, Sarwat et al. (1996) did not measure serum hormone levels, decreasing the validity of the study.

Several studies have shown estrogen HRT to be beneficial in decreasing attenuation of muscle strength in peri and post-menopausal women. It is hypothesised that estrogen alters myosin function during muscle contractions through ER's, in a typical steroid manner (Lowe et al., 2010). Although, several of the studies looking at the mechanism of estrogen in muscle function is done in rodents and more work is needed. Recently, Morton et al. (2018) found that increased muscle mass was not correlated with circulating testosterone, but rather testosterone receptor content in health young men (Morton et al., 2018). As hypothesised by lowe et al. (2010), skeletal muscle seems to be an estrogen responsive tissue, working through the ER's. This may help explain why studies adopting periodized training protocols, utilizing elevated circulating estrogen levels have produce increased muscle strength, as opposed to more modest results displayed when measuring acute performance tests. Exploiting a preferable hormonal milieu may promote strength gains over time, as adaptations in strength may needs time to manifest itself, even if estrogen improves intrinsic quality of contractile fibres. Individual ER content may therefore be partly indicative of muscle strength gains.

4.4 self-reported onset of menstruation

Four out of twelve participants showed deviating results when comparing self-reported menstruation to serum hormone levels (table 6). Two of the individuals reported no onset of menstruation through the six-week period. However, post hoc analysis of sermon hormone levels indicate that menstruation did occur. One individual had one week absent from testing and did not report any onset of menstruation. However, hormonal analysis of the remaining samples indicate that menstruation occurred during the week absent. As described by Allen et al. (2016), self-reported onset of menstruation has several limitations regarding reliability, including within woman variability, making cycle phase determination arduous (Allen et al.,

2016). Thus, serum hormonal levels were used for determination of cycle phase, regardless of self-reported menstruation in the NHCG.

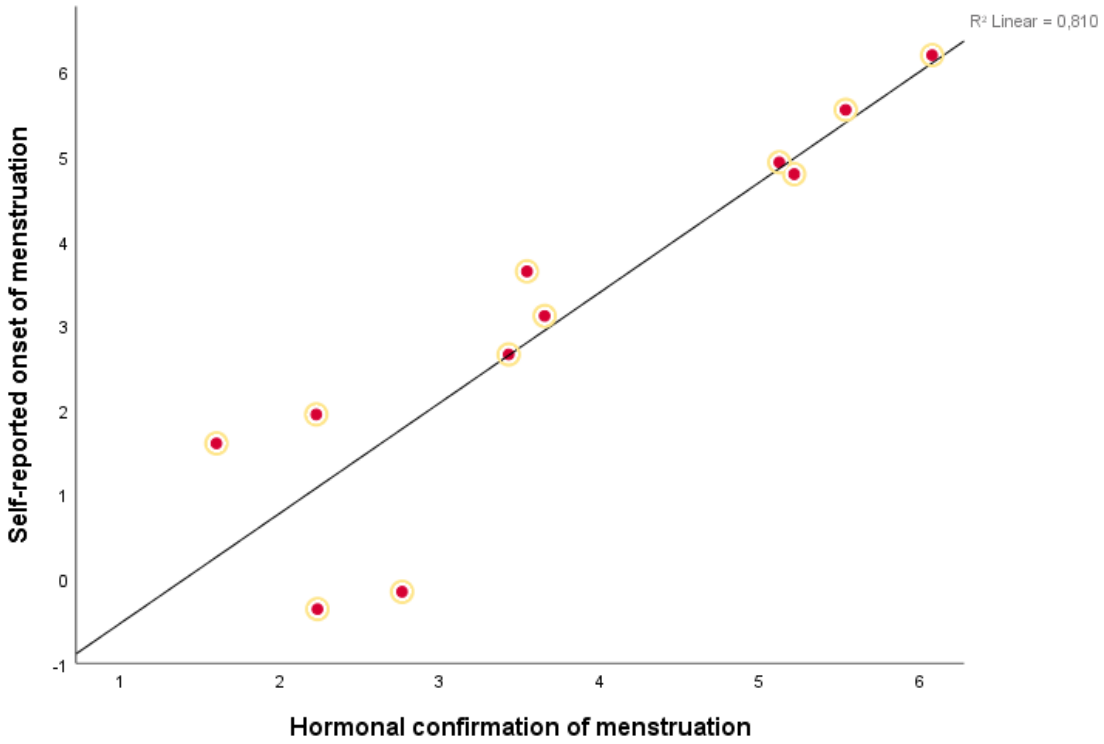

Figure 19. Jittered scatter plot (-0.5 - 0.5) of the correlation (Pearson correlation coefficient) between self-reported onset of menstruation and hormonal confirmation of menstruation, based on table six (result section). For participants reporting more than one menstruation, supported by hormonal confirmation, only one of the weeks are illustrated. One participant is excluded from the illustration due to being absent during the week of menstruation.

It is possible that several participants misinterpreted the weekly questionnaire for determination of menstrual phase, contributing to the reduced correlation between self-reported onset of menstruation and serum hormone levels. However, our findings indicate that caution should be made when using self-reported onset of menstruation for determination of cycle phase. As illustrated in figure nineteen, there are some substantial outliers reducing the correlation between self-reported onset of menstruation and hormonal confirmation of menstruation. Further, it was decided that N=12 in the NHCG group was too low to make any valid statistical conclusion or generalisation, Hence, no correlation statistics were included in the results. The questionnaire used in the study can be found in appendix three.

4.5 HCG vs NHCG

Although no statistical difference was observed between the two groups, some interindividual variability were present in both groups. With a relatively small sample size, it is possible that the results would approach being statistically significant with a larger population. As highlighted by Sims et al. (2018), HC users are proposed as a suitable controls when measuring the effects of the MC (Sims & Heather, 2018). However, several types of contraceptive agents, with different active ingredients and exogenous hormones were administered in the HCG (table 1). Thus, impact on performance may vary, depending on the type of contraceptive agent being used. To my knowledge, there is no study investigating the effect on different types of HC's related to performance. Future research should therefore investigate if different HC preparations elicit disparate responses on athletic performance. This could possibly affect the selection of HC agents used in athletic populations. However, caution should be taken when addressing this, as different HC agents can elicit individual responses in terms of side effects. Thus, hypothetical findings proving disparity between HC agents and performance could propose an ethical dilemma for coaches and athletes.

Future research investigating the MC's effect on performance parameters could also consider using male participants as a control group. As males does not display the same hormonal fluctuations elicited by the MC, potential alterations in performance would likely not be induced by cyclical changes in circulating hormone levels.

4.6 Strength and limitations

To my knowledge this is the first study investigating the MC's effect on strength and power performance in high level female team athletes. Strengths of the present study include investigation of until now unknown effects of the female MC for this demographic, promoting

evidence for consideration regarding performance in female athletes. It also highlights the possibility of conducting prospective research in high level female athletes, which is often considered problematic due to training and competition schedule, paired with female physiology.

Drop out and exclusion:

Fifty-five individuals received and answered the baseline questionnaire for participation in the study (figure 9). Nine individuals dropped out prior to testing, due to personal reasons or injury. Five participants chose to drop out during the six-week testing period, due to personal reasons or injury. No adverse effects due to the testing protocol were reported. Four participants were excluded post hoc as they did not report onset of menstruation during the six-week period, failing to fulfil the inclusion criteria. Hence, determination of MC phase was not possible. Due to a misunderstanding with the hormonal laboratory at HUS, blood samples from two participants were not analysed in time for statistical analysis. Therefore, these participants were excluded from the analysis, as self-reported onset of menstruation alone is insufficient for determination of cycle phase.

Study design:

The study is not without limitations. We chose four weeks out of the six-week testing period, representing two measures in each part of the MC (Follicular and luteal), based on self-reported onset of menstruation and serum hormonal levels. Ideally, measurements over several MC's would have provided increased validity to our results, due to individual variations. However, with the population being high level athletes with busy training and competition schedules and the time frame permitted for the master thesis, this was not possible. The design provided us with relatively high internal validity, however, this in turn effects the external validity of the study. Therefore, caution should be made to extrapolate the findings of this study beyond the investigated population. Further, quasi-experimental designs lack random assignment. Thus, quasi-experimental groups may differ from the treatment condition in systemic ways, other than presence of the condition (HC status). These potential

differences could be alternative explanations for the observed effects of the study. Random assignment in the form of randomized controlled trial (RCT), prescribing HC agents or placebo would minimize these confounders on the observed effect (Shadish, 2002, p. 14). However, conducting random assignment when comparing HC usage would be considered unethical, leaving us with only speculative hypotheses regarding this question.

Testing:

Preliminary visits for familiarisation and determination of baseline 1 RM weight should have been conducted prior to the testing intervention. This would have helped decrease potential learning effects during initial trials. However, logistical reasons made this difficult, thus considerations were made during testing to account for this (see section methodology).

Testing was originally scheduled for the same day (± 2 days) and time (± 2 hours) throughout the six-week period to account for circadian variations. However, due to national team obligations and training/competition schedules being fluent in pre-season, some participants were tested outside these frames at one or several points. This could have impacted the results, as several studies have shown circadian rhythm to affect performance in strength and power endeavours. Indeed, Grgic et al. (2019) in their meta-analysis concluded that expression of strength is greater in the evening compared to the morning. This is in agreement with Martin et al. (1999) and Guette et al. (2005). However, the circadian effect on test results also depend on the athletes normal training schedule (Grgic, Lazinica, et al., 2019; Guette, Gondin, & Martin, 2005; A. Martin, Carpentier, Guissard, van Hoecke, & Duchateau, 1999). Thus, the impact of the incidents where the circadian schedule could not be obtained is not fully known.

interrater-reliability could have contributed to alterations in results. Standard instructions were given to participants prior to each test, to ensure similar testing conditions. Description of the test were placed at each station to aid the testers in enforcing the standardised conduction of the tests. However, these standards may have varied depending on the tester at hand. Interrater-reliability tests could have been done prior to the start of the study protocol,

ensuring agreement between testers. As with several other challenges highlighted during this project, time and resources did not permit this. This could have major implications for the final results and should be considered when interpreting the study.

Participants:

Participants consisted of athletes from the sports soccer, handball and volleyball. Logistically the season differs between the three sports, with handball and volleyball being in-season amid the testing period. This resulted in different physical volume and intensity during the six-week study. Cumulative fatigue and stress related to competition and pre-season training could potentially have affected the results. With the participants competing at a professional or semi-professional level, alterations to their schedule was not possible. These individual differences in volume and intensity however, could have impacted the results.

Female elite team athletes do not have the same financial opportunities as males, thus, most of the participants engaged in other activities to complement their athletic career (table 2). In fact, only one participant who completed the study reported being a full-time athlete. psychological demands related to non-athletic endeavours could add additional stress and effect the physical performance of athletes (Soligard et al., 2016). This possible confounder is not something we could control for but should still be addressed. This is also a challenge for future studies, utilizing high-level female athletes and one can only hope this gender discrepancy changes with time.

Another limitation of the study is the relatively low sample size. With the inclusion criteria's being rigid in terms of athletic performance level, increasing the sample size proved difficult. Drop-outs and exclusion also contributed to the low sample size. Together with high interindividual variability in test results, the study is prone to both false positives and negatives (Dumas-Mallet, Button, Boraud, Gonon, & Munafò, 2017). Indeed, low sample size is a common challenge in sport and exercise science, investigating populations competing at a high level. This challenge should be considered for future research regarding the MC's effect on performance.

A priori inclusion age was set at eighteen years for participants. This was specified in the consent form issued to participants before confirmation of participation in the study. During the follow-up period, a breach of this inclusion criterion was discovered through the baseline questionnaire for two participants. To overcome this challenge, alterations to the permitted age was changed and accepted by REK. This could have been avoided by issuing the baseline questionnaire earlier, as well as faster processing of the material when received from participants. It should be noted that several participants had delays in delivering the baseline questionnaire, although several inquiries were issued. Due to a slow recruitment process, baseline questionnaires were issued at different rates depending on confirmation of participation. Thus, some participants delivered this during the early weeks of the study. A clear cut off date for confirmation of participation could have been made to avoid this problem. However, this was not done as the sample size was limited.

4.7 Physiotherapeutic relevance

Physiotherapy as a profession is changing intact with society and development in our understandings of the human body. For example, in later years there have been increasing pressure on physiotherapists to adopt an evidence-based approach (EBA), integrating best research along with clinical reasoning (Scurlock-Evans, Upton, & Upton, 2014). Indeed, the scope of practice is also evolving, including physiotherapies roles in sport. Several high-level sport teams incorporate physiotherapists as an integral part of determining total training load, return to play (RTP) and physical preparation. Further, physiotherapy represented the largest single professional discipline during the London Olympics, manifesting its place in sports medicine (Grant et al., 2014). In adopting tasks expanding beyond the clinic, understanding of physiological, psychological and biological processes are integral and should be part of an EBA. When dealing with female populations, understanding of the MC is necessary. This study provides insight into the MC's role on acute performance, supplementing already

existing literature on the subject. Potential alterations caused by the MC should be taken into consideration when evaluating tests, RTP or training schedules, as well as prescribing training load and intensity. Understanding the MC's role in females may also be helpful in clinical work with non-athletic populations. Although findings from this study may not be directly comparable to other populations, it can help give insight into similar challenges for females in general.

5 Conclusion and practical application

Testing of strength and power performance parameters in high level team athletes was not significantly affected by HC status when comparing the HCG and NHCG during the MC. These findings suggest that MC phase should not be a major consideration for athletic testing or competition, emphasising strength and power performance. However, interindividual variability in results and hormonal levels, together with a small sample size makes firm conclusions or guidelines arduous. Communication between coach and athlete, regarding the individual response to the MC should therefore be emphasised in relation to testing and competition. Based on our study and the current literature, possible alterations to strength and power performance elicited by the menstrual cycle are more likely to be seen during cycle dependent periodized training interventions, rather than acute testing of performance. Our findings should be interpreted with caution, due to the limitations of the study.

Although small, both the HCG and NHCG displayed small weekly variations throughout the menstrual cycle. As different HC agents may elicit disparate physiological responses affecting performance, male controls could be considered instead of HC users to better highlight if performance changes are caused by the MC. This emphasises the need for better understanding of different types of HC's effect on performance. Future studies should also consider greater control of training and competition intensity and volume in participants. Although this may prove difficult, including players exclusively from one sport can provide a more homogenous seasonal pattern. This could help minimize the diversity in individual load for the participants. Further, this study supports existing literature regarding the validity of self-reported onset of menstruation. Therefore, caution should be made when conducting or interpreting studies utilizing this method.

This study has proved that prospective research on high level female athletes, indeed, is possible. Thus, the argument that female physiology, paired with hectic schedules makes this ambitious is not necessarily valid. Future research should also strive to include high level female athletes, to gain further insight into how physiological processes influence performance.

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List of appendices

Appendix 1.

Forespørsel om deltakelse i forskningsprosjektet

Hvordan påvirker menstruasjonssyklusen muskulære prestasjonsparameter hos kvinnelige elite- utøvere

Dette er et spørsmål til deg om å delta i et forskningsprosjekt for å se på sammenhengen mellom menstruasjonssyklusen og muskulære prestasjonsparametre. Testing av fysiologisk prestasjon er vanlig innen idrett for å evaluere utøvere, samt måle framgang. Kvinners menstruasjonssyklus gjør at det hormonelle miljøet i kroppen endres i løpet av menstruasjonssyklusen, avhengig av hvilken fase man befinner seg i. Det er begrenset med forskning som ser på menstruasjonssyklusen og hvordan denne påvirker muskulære prestasjonsparametre og vi ønsker derfor se nærmere på dette. Det er også ønskelig å undersøke den direkte effekten av søvn på idrettsprestasjon, og samtidig den direkte effekten av menstruasjonssyklus på søvn hos unge toppidrettsutøvere.

Studiet er en del av et masterprosjekt innen fysioterapivitenskap, samtidig som det vil inkludere Bachelorprosjekter fra Bioingeniør, Fysioterapi og Idrett ved Høgskolen på Vestlandet. Vi henvender oss til deg da du oppfyller kravene som toppidrettsutøver jf. Olympiatoppen i Norge sin definisjon av dette.

Hva innebærer PROSJEKTET?

Kvinnelige utøvere på elite nivå skal gjennomføre en fysisk og fysiologisk testprotokoll over en 6 ukers periode for å se om endringer i det hormonelle miljøet påvirker disse testene. Utøverne vil måtte møte opp 1 gang i uken gjennom 6 uker for å gjennomføre protokollen. Dette for å sikre testing i de forskjellige fasene av menstruasjonssyklusen hvor man forventer en endring i det hormonelle miljøet. Det vil ved hver test gang også bli utført blodprøvetaking for å følge menstruasjonsfasen med biomarkører (som hormoner, blodceller og signalmolekyler).

Deltagerne vil ved oppmøte ta en blodprøve, besvare et spørreskjema og deretter gjennomgå de fysiske testene: beinpress, maksimalt vertikalhopp, håndholdt Dynamometer og 40 meter sprint. Man vil også bli bedt om å oppgi hvor utmattende testene var på en skala. Deltagernes vekt og høyde vil også bli målt ved oppmøte. Hele prosessen vil ta ca. 60 minutter. Deltagerne vil også bli bedt om å loggføre inntak av mat og drikke samt unngå inntak av koffein 12 timer før testene skal utføres for å unngå at prestasjonen eller biomarkører blir påvirket av dette. Det ønskes også at siste måltid tatt før fysisk testing ikke er kortere enn 2 timer, for å unngå påvirkning på hormonverdier.
Deltagerne skal fylle ut søvndagbok daglig i alle seks ukene og bære en digital søvnmonitor (aktigraf) i alle 6 ukene. En aktigraf er på størrelse med en liten klokke som deltagerne bærer på armen hele døgnet. Deltagerne blir instruert til å trykke på en knapp på aktigrafen når de skal legge seg og når de står opp.

I prosjektet vil vi innhente og registrere opplysninger om deg. Opplysninger som vil bli registrert er kontaktinformasjon (navn, alder, bosted), søvnvaner, treningsregime og idrettsprestasjonsevne, helseopplysninger (bla. p-pille bruk og eventuell omfang av smerter, skader eller sykdom) og biologiske opplysninger (biomarkører i blodprøver).

Mulige fordeler og ulemper

Risiko ved deltagelse i dette prosjektet er regnet som svært liten. Skulle det oppstå smerte, ubehag eller komplikasjoner ved blodprøvetaking vil denne bli avbrutt. Studien skjer under veiledning og vurdering av autorisert helsepersonell eller helsepersonell og vil være omfattet av pasientskadelovens dekningsområde. I forhold til de fysiske testene som utføres er skaderisiko svært liten og restitusjonstiden lav.

Frivillig deltakelse og mulighet for å trekke sitt samtykke

Det er frivillig å delta i prosjektet. Dersom du ønsker å delta, undertegner du samtykkeerklæringen på siste side. Du kan når som helst og uten å oppgi noen grunn trekke ditt samtykke. Dersom du trekker deg fra prosjektet, kan du kreve å få slettet innsamlede prøver og opplysninger, med mindre opplysningene allerede er inngått i analyser eller brukt i vitenskapelige publikasjoner. Dersom du senere ønsker å trekke deg eller har spørsmål til prosjektet, kan du kontakte **Elisabeth Ersvær, elisabeth.ersver@hvl.no, tlf: 55 58 76 05 eller Marcus Småvik Dasa, Marcus.smavik.dasa@gmail.com, tlf: 92855997**

Hva skjer med OPPLYSNINGENE om deg?

Opplysningene som registreres om deg skal kun brukes slik som beskrevet i hensikten med prosjektet. Du har rett til innsyn i hvilke opplysninger som er registrert om deg og rett til å få korrigert eventuelle feil i de opplysningene som er registrert. Du har også rett til å få innsyn i sikkerhetstiltakene ved behandling av opplysningene.

Alle opplysningene vil bli behandlet uten navn og fødselsnummer eller andre direkte gjenkjennende opplysninger. En kode knytter deg til dine opplysninger gjennom en navneliste.

Det er kun Elisabeth Ersvær, elisabeth.ersver@hvl.no, tlf: 55587605 som har tilgang til denne listen etter 6-ukers testperiode.

Opplysningene om deg vil bli anonymisert eller slettet senest fem år etter prosjektslutt.

Hva skjer med prøver som blir tatt av deg?

Prøvene som tas av deg skal oppbevares i en forskningsbiobank (REK 2018/1529) tilknyttet prosjektet, samt i en generell biobank (REK 2016/787). Oppbevaring i generell biobank gjør at prøvene kan brukes i fremtidige prosjekter, som dekkes av formålet, etter godkjenning fra REK (se eget samtykkeskjema). Det er blodserum eller blodplasma som skal lagres i Biobank for «Idrett, Helse og Funksjon: Biomarkører» lokalisert på Høgskolen på Vestlandet – avdeling Kronstad med ansvarshavende **Elisabeth Ersvær, elisabeth.ersver@hvl.no, tlf: 55587605**

Forsikring

Prosjektet utføres under veiledning av helsefagarbeidere og vil derved falle inn under pasientskadeloven.

Godkjenning

Regional komité for medisinsk og helsefaglig forskningsetikk har vurdert prosjektet, og har gitt forhåndsgodkjenning (2018/1529)

Etter ny personopplysningslov har behandlingsansvarlig Høgskolen på Vestlandet og prosjektleder Elisabeth Ersvær et selvstendig ansvar for å sikre at behandlingen av dine opplysninger har et lovlig grunnlag. Dette prosjektet har rettslig grunnlag i EUs personvernforordning artikkel.

Du har rett til å klage på behandlingen av dine opplysninger til Datatilsynet.

kONTAKTOPPLYSNINGER

Dersom du har spørsmål til prosjektet kan du ta kontakt med **Elisabeth Ersvær, elisabeth.ersver@hvl.no, tlf: 55587605**. Dersom deltagerne har spørsmål knyttet til søvn kan de i prosjektperioden kontakte søvnforskere ved Universitetet i Bergen (professor Anette Harris, 55583219 eller professor Ståle Pallesen, 55588842).

Du kan ta kontakt med institusjonens personvernombud dersom du har spørsmål om behandlingen av dine personopplysninger i prosjektet. **Personvernombud ved HVL Advokat Halfdan Mellbye**, personvernombod@hvl.no, tlf: 55301031.

Jeg samtykker til å delta i prosjektet og til at mine personopplysninger og mitt biologiske materiale brukes slik det er

Sted og dato	Deltakers signatur
	Deltakers navn med trykte bokstaver

Appendix 2.

Spørreundersøkelse i forbindelse med prosjektet ´´Hvordan påvirker menstruasjonssyklus muskulære parametre hos kvinnelige toppidrettsutøvere?´´.
Følgende spørsmål skal besvares med å krysse av på det alternativet(ne) som best beskriver din situasjon. Hvis du skal svare skriftlig på noen spørsmål vil dette være spesifisert.

Fødselsdato

_____

Navn

_____

LEAF-Q Spørreskjema

1. Nåværende idrett/idrettsgren

_____

2. Hvilke andre idretter/idrettsgrener har du eventuelt drevet aktivt med tidligere?

_____

3. Hvor gammel var du da du begynte spesialisere deg i nåværende idrett?

_____

4. På hvilket nivå konkurrerer du? (evt spesifiser hvilket landslagsnivå Sr, jr e.l.)

Klubb _____

Landslag _____

Profesjonelt _____

Annet _____

5. Er du idrettsutøver på heltid?

(1) ❑ ja

(2) ❑ nei

6. Hvis nei, hva driver du med ved siden av idretten?

(1) ❑ Heltidsjobb

(2) ❑ Deltidsjobb

(3) ❑ Studier/skolegang

(4) ❑ Annet

7. Eventuelt hvilken utdannelse har du?

_____

8. Hva er ditt høyeste maksimale oksygenopptak (vo2max) de siste 12 månedene? (et svar alternativ)

Ml/kg/min _____

L/min _____

vet ikke/har ikke målt _____

9. Hva er din beste plassering i Norgesmesterskap (NM)/enkeltkonkurranse i norgescup?

(1) ❑ 1-3 plass

(2) ❑ 4-6 plass

(3) ❑ 7-10 plass

(4) ❑ 11 plass eller dårligere

(5) ❑ Ikke deltatt i NM eller norgescup

(6) ❑ Husker ikke

10. Hva er din normale treningsmengde i forberedelses- og oppbyggingsfasen (Ikke konkurransefasen) i gjennomsnitt per måned? (antall timer)

_____

11. Hvor gammel er du?

_____

12. Hvor høy er du? (cm)

_____

13. Hva er din nåværende vekt? (kg)

_____

14. Hva er din høyeste vekt med nåværende høyde? (bortsett fra evt. graviditet)

_____

15. Hva er din laveste vekt med nåværende høyde?

_____

16. Hva anser du som din konkurranse/´´match vekt´´?

_____

17. Hva er din fettprosent % ? (dersom du har målt denne)

_____

18. Har du kronisk sykdom?

(1) ❑ ja

(2) ❑ Nei

19. Dersom ja, hvilken/hvilke sykdom(mer)/plager?

_____

20. Har du vært skadet i løpet av det siste året og dermed hatt fravær fra eller vært markant begrenset i forhold til din trenings/konkurranseevne?

(1) ❑ Nei, slett ikke

(2) ❑ Ja, 1-2 ganger

(3) ❑ Ja, 3-4 ganger

(4) ❑ Ja, 5 ganger eller fler

21. hvis ja, hvor mange ganger i løpet av det siste året har du ikke trent eller deltatt i konkurranse som planlagt på grunn av skader?

(1) ❑ 1-7 dager

(2) ❑ 8-14 dager

(3) ❑ 15-21 dager

(4) ❑ 22 dager eller flere

22. Hvis ja, hvilke typer skader har du hatt i løpet av det siste året?

_____

23. Evt. kommentar eller utdypning angående skader

_____

24. Føler du deg oppblåst eller oppsvulmet i magen, også når du ikke har menstruasjon?

(1) ❑ Ja, flere ganger/dag
(3) ❑ Ja, flere ganger/uke
(2) ❑ Ja, 1-2 ganger/uke eller skjeldnere
(4) ❑ Skjeldent eller aldri

25. Har du kramper og/eller magesmerter, som ikke kan relateres til din menstruasjon?

(1) ❑ Ja, flere ganger/dag
(3) ❑ Ja, flere ganger/uke
(2) ❑ Ja, 1-2 ganger/uke eller skjeldnere
(4) ❑ Skjeldent eller aldri

26. I gjennomsnitt, hvor ofte har du avføring?

(1) ❑ Flere ganger/dag
(2) ❑ 1 gang/dag
(3) ❑ Hver 2. dag
(4) ❑ 2 ganger/uke
(5) ❑ 1 gang/uke eller skjeldnere

27. Hvordan pleier din avføring å være?

(1) ❑ Normal (fast eller bløt)
(2) ❑ Meget tynn, som diare
(3) ❑ Hard og tørr

28. Evt. kommentar eller utdypning angående magefunksjon

_____

29. Bruker du P-piller?

(1) ❑ Ja

(2) ❑ Nei

30. Hvis ja, hvilken type P-piller bruker du? (hvis du vet dette)

_____

31. Hvis ja, hvorfor bruker du P-piller?

(1) ❑ Prevensjonsmiddel

(2) ❑ Redusere menstruasjonssmerter

(3) ❑ Redusere blødningsmengden

(4) ❑ For å regulere menstruasjonssyklus i forbindelse med konkurranser etc.

(5) ❑ Hvis ikke, uteblir mensen

(6) ❑ Annet

32. Hvis nei, har du brukt P-piller tidligere?

(1) ❑ Ja

(2) ❑ Nei

33. hvis du har brukt p-piller tidligere, når og hvor lenge?

_____

34. Bruker du noen annen form for hormonell prevensjon? (f.eks. p-stav, hormonspiral)

(1) ❑ Ja

(2) ❑ Nei

35. Hvis ja, hvilken type?

(1) ❑ P-plaster

(2) ❑ P-stav

(3) ❑ Hormonspiral

(4) ❑ Annet

36. Hvor gammel var du da du fikk din første menstruasjon?

(1) ❑ 11 år eller yngre

(2) ❑ 12-14 år

(3) ❑ 15 år eller eldre

(4) ❑ Husker ikke

(5) ❑ Har aldri hatt mensen (hvis du svarer her, kan du hoppe til spørsmål nr 51)

37. kom din første menstruasjon naturlig? (av seg selv)

(1) ❑ Ja

(2) ❑ Nei

(3) ❑ Husker ikke

38. Hvis nei, hva ble gjort for å igangsette din menstruasjon?

(1) ❑ Hormonbehandling

(2) ❑ Vektøkning

(3) ❑ Redusere treningsmengde

(4) ❑ Annet

39. Har du normal menstruasjon?

(1) ❑ Ja

(2) ❑ Nei (gå til spørsmål 47)

(3) ❑ vet ikke (gå til spørsmål 47)

40. Hvis ja, når hadde du sist menstruasjon?

(1) ❑ 0-4 uker siden

(2) ❑ 1-2 måneder siden

(3) ❑ 3-4 måneder siden

(4) ❑ 5 måneder eller lenger

41. Hvis ja, har du regelmessig menstruasjon? (Hver 28.-34. dag)

(1) ❑ Ja, som regel

(2) ❑ Nei, som regel ikke

42. Hvis ja, hvor mange dager pleier du ha blødning?

(1) ❑ 1-2 dager

(2) ❑ 3-4 dager

(3) ❑ 5-6 dager

(4) ❑ 7-8 dager

(5) ❑ 9 dager eller mer

43. Hvis ja, har du noen ganger problemer med kraftig menstruasjonsblødning?

(1) ❑ Ja

(2) ❑ Nei

44. Hvis ja, hvor mange menstruasjonsblødninger har du hatt i løpet av det siste året?

(1) ❑ 12 eller flere

(2) ❑ 9-11

(3) ❑ 6-8

(4) ❑ 3-5

(5) ❑ 0-2

45. Hvis nei eller husker ikke, hvor lenge er det siden du hadde sist menstruasjon?

(1) ❑ 2-3 måneder

(2) ❑ 4-5 måneder

(3) ❑ Mer enn 6 måneder siden

(4) ❑ Jeg er gravid og har derfor ikke menstruasjon

(5) ❑ Jeg bruker minipiller og har derfor ikke menstruasjon

46. Har din menstruasjon uteblitt helt i 3 måneder eller lenger uten uten at det skyldes graviditet eller minipille?

(1) ❑ Nei, det har aldri skjedd

(2) ❑ Ja, det har skjedd tidligere

(3) ❑ Ja, jeg opplever det nå

47. Opplever du at din menstruasjon endrer seg ved økt treningsintensitet, frekvens og/eller varighet?

(1) ❑ Ja

(2) ❑ Nei

48. Hvis ja, hvordan? (sett ett eller flere kryss)

(1) ❑ Jeg blør mindre

(2) ❑ Jeg blør færre dager

(3) ❑ Min menstruasjon uteblir

(4) ❑ Jeg har kraftigere blødning

(5) ❑ Jeg blør i flere dager

Appendix 3.

Følgende spørsmål skal besvares før du gjennomfører testene i dag. Les spørsmålene godt og velg det alternativet/ne som passer deg best. Om spørsmål skal besvares i skrift er dette presisert.

ID nummer

_____

Hvordan føler du formen din er i dag?

(1) ❑ Svært dårlig

(2) ❑ dårlig

(3) ❑ Middels

(4) ❑ Bra

(5) ❑ Svært bra

Hva var siste måltid du spiste før testen?

_____

Har du vært beruset de siste 4 dagene?

(1) ❑ Ja

(2) ❑ Nei

Har du skader/plager som hindrer deg i å delta på en eller flere tester i dag?

(1) ❑ Ja

(2) ❑ Nei

Hvis ja, beskriv skaden/plagen(e)

_____

Når hadde du sist menstruasjon? (antall dager siden sist blødning)

_____

Hvis du har menstruasjon nå kryss her

(1) ❑ Kryss her

Hvordan tror du menstruasjon vil påvirke ditt resultat på dagens test?

(1) ❑ Noe positiv påvirkning

(2) ❑ Positiv påvirkning
(3) ❑ Ingen påvirknig
(4) ❑ Noe negativ påvirkning
(5) ❑ Negativ påvirkning

Føler du menstruasjon har påvirket din prestasjon i kamp eller treningssituasjon den siste uken?

(1) ❑ Ikke i det hele tatt
(6) ❑ I svært liten grad
(4) ❑ I liten grad
(5) ❑ I høy grad
(3) ❑ I svært høy grad

Hva er din subjektive mening om hvordan du vil prestere på testene i dag?

	Svært dårlig	dårlig	Middels	Bra	Svært bra
Spenst test	(1) ❑	(2) ❑	(3) ❑	(4) ❑	(5) ❑
20-meter sprint	(1) ❑	(2) ❑	(3) ❑	(4) ❑	(5) ❑
Beinpress (keiser)	(1) ❑	(2) ❑	(3) ❑	(4) ❑	(5) ❑
Dynamometer (håndklype)	(1) ❑	(2) ❑	(3) ❑	(4) ❑	(5) ❑

Hvordan begrunner du svaret over?

(1) ❑ Menstruasjon påvirker
(2) ❑ Søvn påvirker
(3) ❑ Andre faktorer

Hvor plaget har du vært av følgende den siste uken?

	Ikke plaget	Litt plaget	En del plaget	Alvorlig plaget
Hodepine	(1) ❑	(2) ❑	(3) ❑	(4) ❑
smerte i kjeve	(1) ❑	(2) ❑	(3) ❑	(4) ❑
Nakke/skuldersmerter	(1) ❑	(2) ❑	(3) ❑	(4) ❑
Smerte i øvre del av ryggen	(1) ❑	(2) ❑	(3) ❑	(4) ❑

	Ikke plaget	Litt plaget	En del plaget	Alvorlig plaget
Smerte i nedre del av ryggen/sete	(1) ❑	(2) ❑	(3) ❑	(4) ❑
Smerte i brystkassen	(1) ❑	(2) ❑	(3) ❑	(4) ❑
Magesmerter	(1) ❑	(2) ❑	(3) ❑	(4) ❑
Smerter i armer	(1) ❑	(2) ❑	(3) ❑	(4) ❑
Smerte i bein	(1) ❑	(2) ❑	(3) ❑	(4) ❑

Hvis du var plaget av en/flere av disse lidelsene, hvor lang tid var du plaget?

_____

Appendix 4.

REK
REGIONALE KOMITEER FOR MEDISINSK OG HELSEFAGLIG FORSKNINGSETIKK

Region:	Saksbehandler:	Telefon:	Vår dato:	Vår referanse:
REK vest	Jessica Svärd	55978497	20.09.2018	2018/1529/REK vest
			Deres dato:	Deres referanse:
			14.08.2018	

Vår referanse må oppgis ved alle henvendelser

Elisabeth Ersvær
Bergen - AIØ/IBK/Bioingeniørutdanningen

2018/1529 Menstruasjonssyklusen sin påvirkning på kontraktile parametre hos kvinnelige toppidrettsutøvere

Forskningsansvarlig: Høgskulen på Vestlandet
Prosjektleder: Elisabeth Ersvær

Vi viser til søknad om forhåndsgodkjenning av ovennevnte forskningsprosjekt. Søknaden ble behandlet av Regional komité for medisinsk og helsefaglig forskningsetikk (REK vest) i møtet 05.09.2018. Vurderingen er gjort med hjemmel i helseforskningsloven (hforsknl) § 10.

Prosjektomtale
Innen toppidrett kan små endringer i prestasjon ha betydning for sluttresultatet. Erfaring over tid tyder på at fysiologiske testresultatene til kvinnelige idrettsutøvere ikke alltid representerer den kapasiteten man ser de innehar i trening og konkurranse. Menstruasjonssyklus kan bidra til variasjon i test resultater. Kjønnshormoner påvirker også utvikling og aktivitet til immunforsvaret. På bakgrunn av dette ønsker vi å undersøke om menstruasjonssyklusen til kvinnelige toppidrettsutøvere påvirker muskulære prestasjonsparametre, når de testes under kontrollerte forhold. Vi ønsker også å undersøke hematologiske og immunologiske mediatorer sin eventuelle endring gjennom menstruasjonssyklusen.

Vurdering

Søknadsplikt?
Prosjektet skal undersøke om fysisk prestasjon hos kvinner varierer gjennom menstruasjonssyklus. Komiteen vurderer det slik at formålet med prosjektet er ny kunnskap om helse og sykdom og at det faller innenfor helseforskningslovens virkeområde.

Forsvarlighet
Komiteen finner at prosjektet har liten risiko og er forsvarlig å gjennomføre.

Biobank
Prosjektet skal lagre prøver i den generelle biobanken «Idrett, Helse og Funksjon: Biomarkører» med referansenummer 2016/787. Elisabeth Ersvær er ansvarshavende. REK vest har ingen innvendinger til dette.

Studiepopulasjon
Toppidrettsutøvere på lagene Arna-Bjørnar, Fana, Tertnes, Sandviken og Viking (N=100).

Tester
Man vil gjennomføre fysiske og fysiologiske tester av studiepopulasjonen: en gang per uke over 6 uker per

individ. Fysiske tester: Beinpress, Maksimalt vertikalhopp, Maksimal håndstyrke, 40 meter sprint, 6 sekunder sprint på sykkelergometer.

Venøse blodprøver, fysiologiske tester
Prøvetaking av akkreditert bioingeniør. Hormonanalyser gjøres ved hormonlaboratoriet, HUS. For full blodcellestatus og CRP benyttes automaiserte hemocytometer. For immuncellesubset analyse benyttes flow cytometry og antistoff. For immunmediator analyser forøvrige vil kommersielle EIA/ELISA benyttes. Serum/plasma vil oppbevares i generell biobank. Fullblod og celler vil analyseres samme dag som tapping og deretter destrueres. REK vest har ingen innvendinger til dette.

Spørreundersøkelse
Digitalt spørreskjema ved bruk av SurveyXact skal brukes. Spørreskjema er preliminært og ferdigutformet spørreskjema må derfor ettersendes REK vest på post@helseforskning.etikkom.no.

Samtykkeskriv
Et informasjons- og samtykkeskriv for prosjektet og et separat informasjons- og samtykkeskriv for den generelle biobanken. Komiteen mener at det bør forklares i informasjons- og samtykkeskriv for prosjektet at lagring i en generell biobank innebærer at prøvene kan brukes i fremtidige prosjekter som dekkes av formålet etter godkjenning av REK.

Informasjonsskrivet må også revideres i tråd med ny mal på REKs nettsider, slik at informasjonen som gis til deltakerne er forenlig med ny personopplysningslov. Komiteen ber om at revidert informasjonsskriv ettersendes REK vest sammen med øvrig tilbakemelding.

REK har utarbeide ny mal for informasjonsskriv i henhold til de nye reglene som trådte i kraft 20. juli med ny personopplysningslov og EUs personvernforordning (GDPR). Endringene i malen er små men viktige. Endringene kan få betydning for framtidig bruk av data som innhentes med samtykke som grunnlag. REK vest setter derfor som vilkår at et nytt informasjonsskriv som er revidert i henhold til den nye malene sendes inn på epost.

I informasjons- og samtykkeskriv for den generelle biobanken står «Materiale og opplysningene om deg lagres permanent og vil analyseres i forbindelse med spesifiserte forskningsprosjekter.». Komiteen gjør oppmerksom på at en biobank kun kan lagre metadata for å identifisere prøvene, ikke helseopplysninger.

Lagring av data og koblingsnøkkel
Under test-perioden vil det foreligge en koblingsnøkkel i papirform (en loggbok) som vil oppbevares i en låst skuff på idrettlaboratoriet på HVL. Etter testperioden vil koblingsnøkkel lagres på HVL sin forskningsserver.

REK vest går ut fra at alle data lagres på forskningsserver etter testperioden, ikke bare koblingsnøkkelen.

For de digitale spørreskjema vil det samles inn personopplysninger. Etter endt test-periode på 6-uker vil all datamateriale være kodet etter samme koblingsnøkkel som benyttet på de fysiske testene/blodprøvetaking. Direkte identifiserbare med 11-sifret personnummer eller navn, adresse og/eller fødselsdato under i hele prosjektperioden.

Det er uklart for komiteen hvorfor det ikke er mulig å bruke kodede opplysninger i prosjektperioden istedenfor for å bruke personnummer.

Vilkår

Revidert informasjons- og samtykkeskriv må sendes til REK vest til post@helseforskning.etikkom.no

Vi gjør oppmerksom på at det kreves et juridisk grunnlag for å behandle personopplysninger. Nytt av 20. juli 2018 er at REKs godkjenning ikke lenger gir et juridisk grunnlag for å behandle personopplysninger. Nå må denne behandlingen også oppfylle krav i personvernforordningen. Fortsatt skal alle forskningsprosjekter

som omfattes av helseforskningsloven forhåndsgodkjennes av REK, men egen institusjon har ansvar for at behandlingen av personopplysninger er i henhold til personvernforordningen.

Vedtak
REK har gjort en helhetlig forskningsetisk vurdering av alle prosjektets sider. Prosjektet godkjennes med hjemmel i helseforskningsloven §§ 10 og 33 på betingelse av at ovennevnte vilkår tas til følge.

Sluttmelding og søknad om prosjektendring
Prosjektleder skal sende sluttmelding til REK vest på eget skjema senest 31.03.2021, jf. hfl. § 12. Prosjektleder skal sende søknad om prosjektendring til REK vest dersom det skal gjøres vesentlige endringer i forhold til de opplysninger som er gitt i søknaden, jf. hfl. § 11.

Klageadgang
Du kan klage på komiteens vedtak, jf. forvaltningsloven § 28 flg. Klagen sendes til REK vest. Klagefristen er tre uker fra du mottar dette brevet. Dersom vedtaket opprettholdes av REK vest, sendes klagen videre til Den nasjonale forskningsetiske komité for medisin og helsefag for endelig vurdering.

Med vennlig hilsen

Marit Grønning
prof. dr.med.
Komiteleder

Jessica Svärd
rådgiver

Kopi til: Eirin.Fausa.Pettersen@hvl.no