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Low energy availability and risk factors in non-athletic females in New Zealand

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Abstract

Background: Low energy availability (LEA) has primarily been studied in athletic females with few studies examining the risk factors among healthy non-athletic females aged 18-30 years. Low energy availability can occur intentionally or unintentionally and may result from dietary manipulations or changes to exercise behaviours which can lead to long term negative health implications. Low energy availability has been shown to have a negative impact on bone health, menstrual function, psychological well-being, immune function, gastrointestinal function and metabolism. Therefore, understanding some of the risk factors of LEA in non-athletic females, inclusive of perceptions of eating and exercise, may allow for early identification of habits that contribute to LEA.

Method: The risk of LEA was determined using a validated screening tool; low energy availability in females questionnaire (LEAF-Q) which was completed through an online survey. Additional risk factors of LEA were further identified using the New Zealand Physical Activity Questionnaire - short form, Clinical Impairment Assessment (CIA) Questionnaire and Eating Disorder Questionnaire. Physical activity was further categorised across the week into low (<5 days of moderate and/or vigorous activity), moderate (≥5 and <10 days moderate and/or vigorous activity) and high (≥10 days moderate and/or vigorous activity) physical activity groups.

Participants: A total of 531 participants took part in the online questionnaire, of which 151 participants met the inclusion criteria for this study and were subsequently included in the analysis. Participants were predominantly New Zealand European (78.4%) with an average body mass index 23.8 ± 4.6 $kg{\cdot}m^{-2}$ and age 23 ± 3.2 years.

Results: Over half of the participants (51.0%) were classified as at risk of LEA based on the LEAF-Q scoring. The majority of participants (52.3%) used oral contraceptive pills. Of the females not using the oral contraceptive pill, 64.2% reported having normal menstruation and 29.1% reported they did not have normal menstruation. Although not significant, those who had irregular menstruation had higher sores across all three CIA components. Clinical impairment assessment global score on average was 10.3 ± 10.4, with 22.5% of all participants having a high level of impairment (≥48). Individuals with a high level of impairment had significantly higher LEAF-Q scores across all three CIA components; eating habits ($p<0.001$), exercise habits ($p=0.002$) and body shape ($p=0.002$). The study demonstrated 26.5% of participants were classified as having low physical activity levels, 42.4% as moderate physical activity levels and 31.1% as reaching high levels of physical activity. One-way ANOVA between physical activity groups (low/moderate/high) and LEAF-Q score demonstrated a significant positive difference ($p=0.025$). There was a significant positive relationship between the number of days of vigorous physical activity and LEAF-Q score ($p=0.038$).

Conclusion: This cross-sectional study adds to the current literature that investigates eating and exercise behaviours and the subsequent influence of LEA in healthy but non-athletic females in New Zealand. This study highlights the multi-directional relationships between exercise and eating behaviours and LEA risk. Non-athletic females in this current study presented with poor perceptions towards eating and body weight based on the CIA questionnaire which may place them at risk of LEA. Subsequently, non-athletic females may present with symptoms associated with chronic LEA. Early detection of LEA is needed among exercising females, given the consequences of LEA in the short and long term, early detection can enable early intervention and reduce the risk to long term health.

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Table of Contents

List of abbreviations

ANOVA	Analysis of variance
BMI	Body mass index
CIA	Clinical Impairment Assessment
EA	Energy availability
EDE-Q	Eating Disorder Questionnaire
EEE	Exercise energy expenditure
EI	Energy intake
FFM	Fat free mass
LEA	Low energy availability
LEAF-Q	Low Energy Availability Questionnaire
NZPAQ-SF	New Zealand Physical Activity Questionnaire - Short Form
RED-S	Relative energy deficiency syndrome
RDA	Recommended dietary allowance
SD	Standard deviation
T3	Triiodothyronine

List of figures

List of tables

Chapter 1: Introduction

Energy availability (EA) refers to the amount of energy remaining for normal body functions after exercise, and is determined as the difference between energy intake (EI) and exercise energy expenditure (EEE) expressed relative to the individual's fat free mass (FFM). This can mathematically be defined as EA = (EI - EEE) $\cdot kgFFM^{-1} \cdot day^{-1}$ (Reed et al., 2015; Areta et al., 2021). Low energy availability may be caused by reducing EI or by increasing exercise or a combination of both (Slater et al., 2016). Exercising females and especially athletes are at risk of LEA compared to sedentary females, largely due to insufficient EI to make up for the increased energy expenditure (Reed et al., 2015).

It is important to acknowledge the strong evidence for participation in sport and physical activity as health promoting, such as aiding in the prevention of non-communicable diseases such as type 2 diabetes and reducing the risk of heart disease, and strengthening bones, all of which have been outlined in longitudinal studies (Reiner et al., 2013). Such benefits may be achieved when adhering to the physical activity guidelines recommended in New Zealand (Dhankar et al., 2018). However, engaging in sport can also be a risk factor for unhealthy behaviours, due to high training loads, especially when an individual is in a state of LEA, in this instance further increases in exercise may have a negative impact on health (Slater et al., 2016; Brown et al., 2017).

There is often the idea that leaner is better, and it is viewed as advantageous for sporting performance. Athletes and other active individuals who perceive this idea, especially those who are already lean, but desire additional body fat loss may develop unhealthy eating behaviours and start inadequately fuelling their bodies as they increase their EEE, increasing their risk of LEA (Schofield et al., 2020). On the other hand, for individuals who are overweight or obese and seeking to lose additional body fat, weight loss may improve sport performance and other health outcomes such as improved body composition and lipid profile (Holowko et al., 2019).

Some sports, both at a competitive and non-competitive level, require individuals to be of a specific weight class, for example rowing or combat sports or where the athletes are aesthetically judged such as gymnastics and figure skating. In these cases, it can be difficult to achieve and maintain a low weight and performance goals over a season without the use of extreme weight control methods, which may include restrictive eating especially in the weeks leading up to the competition (Manore, 2015). Severe energy restriction combined with an intense training programme may lead to more negative consequences, resulting in slowed body fat loss and loss of lean muscle mass (Donnelly et al., 2009). These negative consequences are more likely to occur if energy restriction occurs over a short period of time and the restriction is more drastic. (Dirks & Leeuwenburgh, 2006).

Low energy availability has been identified as a health issue for both athletes and active individuals, with research focusing on the severe consequences observed in females (Slater et al., 2016). There have been reports of similar prevalence rates of LEA symptoms among female athletes and female sedentary controls (Hoch et al., 2009). Sub-optimal EA (>30 and ≤45 $kcal \cdot kgFFM^{-1} \cdot day^{-1}$) appears to be prevalent among 30% of female athletes and 35% of sedentary controls, while clinical LEA (≤30 $kcal \cdot kgFFM^{-1} \cdot day^{-1}$) is prevalent among 6% of female athletes and 4% of sedentary controls (Hoch et al., 2009). Given that LEA can occur in sedentary females, there is a need for further research in non-athletic females, as current research has primarily focused on athletes.

The way in which the body responds to acute and chronic LEA is multifactorial and includes altering reproductive hormones, and will likely result in menstrual dysfunction (Mountjoy et al., 2014). Among previously normally or regularly menstruating females LEA has been found to cause luteal phase defects (Williams et al., 2015), altered metabolic hormones (McCall & Ackerman, 2019) and reductions in fat mass (Koehler et al., 2017). Menstrual disorders are a result of a complex interplay of many factors, including weight loss, decreased body fat, abnormal eating attitudes and behaviours, exercise and stress (Amgain & Neupane, 2020). Recent evidence suggests that among exercising females 52% suffer from menstrual disturbances and 37% may have amenorrhea (Reed et al., 2015; Slater et al., 2016).

Previous experimental research suggests that menstrual disorders may occur on a continuum, with extreme disruptions in menstruation occurring with sharp increases in exercise stress (Chen & Brzyski, 1999). Increased EA through EI to support the demands of exercise training can often maintain normal menstrual function, even while females continue to exercise (Thein-Nissenbaum & Hammer, 2017). This may suggest that inadequate EI is the main contributor to LEA and menstrual dysfunction in exercising females (Reed et al., 2015). In addition to adequate EI, when moderate increases in training occur, at a slow enough rate for the reproductive system to adapt, less extreme disruptions may occur (Bedford et al., 2010). However, it should be noted that there is large individual variability in female menstruation that always needs to be considered when assessing menstrual health of the individual (Bullen et al., 1984).

The female athlete triad was first established in 1992 and involves the association between three components; disordered eating, amenorrhea and osteoporosis (Otis et al., 1997). The female athlete triad outlines the concerns associated with excessive exercising training and LEA resulting in amenorrhoea which leads to a reduction in oestrogen levels and in the long-term can lead to bone loss causing osteoporosis (Márquez & Molinero, 2013). The female athlete triad has been well documented although only includes clinical endpoints of each component and many athletic and recreational females may experience less severe manifestations but still suffer from related health implications (Slater et al., 2016). The clinical representation of the prevalence of the female athlete triad in female athletes compared to controls is still unclear and further research in recreational and non-athletic females needs to be carried out. A meta-analysis review among non-athletic and athletic females combined, found that approximately 16% had a prevalence of all three components of the female athlete triad, 27% had two components and 60% had at least one of the female athlete triad components (Gibbs et al., 2013).

Relative energy-deficiency in sport syndrome (RED-S) is a more recently (2014) developed model, and takes into consideration a multitude of physiological factors that exercising individuals may experience when in a chronic state of LEA (Mountjoy et al., 2014). The RED-S model identifies additional factors such as stress, depression, abnormal eating behaviours and eating disorders, cardiovascular health, growth, and metabolic and endocrine factors that all may be affected by a state of LEA (Mountjoy et al., 2014). Relative energy-deficiency in sport syndrome is diagnosed by exclusion of severe medical conditions and the frequency of RED-S symptoms which can be assessed using the RED-S clinical assessment tool (Mountjoy et al., 2014). Using this clinical assessment tool, health care professionals (i.e. physician, dietitian, physiologist, coach and athlete) can classify individuals as being at high, moderate or low risk of RED-S. This categorisation can then be used to determine when an individual is ready to return to sport.

It is recognised that the common aetiology of both RED-S and the female athlete triad is LEA. Low energy availability can occur intentionally or unintentionally and may result from dietary manipulations or changes to exercise behaviours. Therefore, effective treatment of LEA requires an integrative approach from health care professionals. This can be achieved when there is a common goal, of which the health and well-being of the individual needs to be addressed first (Schofield et al., 2020).

An increased awareness around the negative implications of LEA and the psychological and social risk factors for LEA is required. Raising awareness of these risk factors will enable active individuals to be informed about how their current attitudes and behaviours, for example, exercising and eating behaviours and body satisfaction, are impacting their long-term health (Ackerman et al., 2020).

1.1 Aims, objectives and hypothesis

The overall aim of the present study is to investigate the associations of eating and exercise behaviours on LEA risk in non-athletic females in New Zealand, along with the following objectives:

1. To assess physical activity, menstrual function and eating behaviours in non-athletic females in New Zealand through an online questionnaire.

2. To investigate relationships (multi-directional) between exercise, eating behaviours and LEA risk in non-athletic females in New Zealand.

We hypothesise that non-athletic females with high and/or frequent physical activity behaviours will be classified as 'at risk of LEA'. Those females at risk of LEA will have negative feelings towards exercise and eating compared to those not at risk of LEA.

Chapter 2: Review of the literature

2.1 Introduction

The aim of this literature review is to highlight the psychological and physiological benefits that can be achieved when the physical activity recommendations are met. Following this, current dietary habits, disordered eating behaviour and body dissatisfaction in regularly exercising females will be explored. Finally, the implications of inadequate EI and excessive exercise will be outlined, along with how this can result in LEA and its associated consequences.

2.2 Literature Search

We attempted to retrieve all research examining LEA among non-athletic females, while also aiming to avoid biased retrieval of searching only major journals. The literature search was completed between January 2019 to December 2020 and the following methods were used to accomplish a comprehensive search for relevant articles:

1. Key words (e.g., female, athletes, non-athletes, physical activity, LEA, female athlete triad, RED-S, eating, dietary habits, disordered eating, eating disorders) were used to conduct searches in the following databases: PubMed, Web of Science, Google Scholar, Massey Library Database (Discover).
2. The title of the article was reviewed to ensure its relevance, followed by the abstract. If the study was appropriate, the full article was reviewed and then included in the literature review presented within this chapter.

2.3 Benefits of a physically active lifestyle

Physical activity is recommended as part of a healthy balanced lifestyle. Currently the global recommendations on physical activity for health promote exercise and movement as primary prevention methods for non-communicable disease (World Health Organisation, 2020). This is largely a result of physical inactivity being identified as the fourth leading risk factor for global mortality (World Health Organisation, 2020). The recommendations of physical activity in New Zealand are set out by the Ministry of Health, with the current guidelines in New Zealand reflecting those of many other countries such as Canada, Australia, United Kingdom, United States (Tremblay et al., 2011; Australian Bureau of Statistics, 2013; Piercy et al., 2018).

2.3.1 Physical activity for health

The current physical activity recommendations in New Zealand are tailored to meet the needs of specific age and population groups, for example older adults and those who are breast-feeding or diabetic (Ministry of Health, 2020a). The current New Zealand physical activity guidelines for healthy adults aged 18 to 64 years is 150 minutes of moderate activity or 75 minutes of vigorous activity spread across the 7-day week (Ministry of Health, 2020a). For additional health benefits, the Ministry of Health recommends twice this amount of physical activity as well as muscle strengthening exercise (Ministry of Health, 2020a). Moderate intensity activities include brisk walking, bike riding, dancing and hiking at 50 to 85% maximum heart rate, while vigorous activities include running, swimming fast and team sports such as football and netball where an individual is exercising at a higher intensity (generally >85% of maximum heart rate) (Mann et al., 2014). The current physical activity recommendations aim to promote overall health and well-being and are associated with psychological and physiological benefits (Warburton et al., 2007). Most international guidelines recommend 150 minutes of moderate to vigorous activity a week, however some evidence suggests that although benefits are seen when these guidelines are met, benefits may also be seen with exercise volumes below these total minutes especially in overweight individuals that would benefit from simply becoming more active (Warburton & Bredin, 2017).

2.4 Psychological and physiological benefits of being physically active

2.4.1 Mental health

Regular physical activity in line with the current recommendations has been shown to have a positive correlation with self-esteem, cognitive functioning and psychological well-being and a negative correlation with anxiety and depression (Craft & Perna, 2004; Walsh, 2011). Associations between physical activity and mental health among adolescents and adults has gained interest in recent years given the increasing incidence and awareness of mental health issues in society such as depression, anxiety and bipolar disorder. In New Zealand, 5.0% of adults were reported to be suffering from mental health issues in 2012, with this statistic increasing to 8.8% in 2016, and to 11.8% in 2018 (Ministry of Health, 2019).

Mental health issues such as depression occurs throughout the general population and are especially prevalent among females, with incidence being nearly twice that reported in males (Ministry of Health, 2019). In 2019, 11.0% of males and 20.3% of females in New Zealand were diagnosed with depression (Ministry of Health, 2019). As such research investigating the effects and efficacy of exercise as a prevention or treatment option for depression and improving mental well-being has been prominent and flourishing area of research (Carter et al., 2015; Blumenthal et al., 2007; Tasci et al., 2019).

The psychological benefits of physical activity participation may help reduce depressive symptoms through additional parameters such as increased self-efficacy, positive thoughts, distraction from negative thoughts and enhanced self-concept (Blumenthal et al., 2007; Nabkasorn et al., 2006). Both aerobic and non-aerobic forms of physical activity have been effective in reducing depression, especially when physical activity is included on a regular basis (Bennie et al., 2019). Hoffman et al. (2011), found that among previously sedentary adults with major depressive disorder there was a linear increase in remission rates up until 150 minutes of physical activity per week, with the remission rates flattening off beyond this amount.

2.4.2 Physical activity and cardiovascular disease

In addition to psychological benefits, physical activity has a role to play in reducing the risk of cardiovascular disease and thus improving and supporting an individual's physiological health. Regular physical activity has been linked with reduced all-cause mortality and cardiovascular disease risk (Kraus et al., 2019). As such, females and males who met physical activity recommendations had 40% reduced risk of all-cause mortality and cardiovascular disease compared to those not meeting recommendations (O'Donovan et al., 2010). Physical activity has been shown to have favourable outcomes on cardiovascular risk factors by reducing triglyceride and low-density lipoprotein concentrations, increasing high-density lipoprotein concentration (Manco et al., 2007), and also by promoting fatty acid oxidation and decreasing glucose oxidation and resting blood pressure (Manco et al., 2007). Other risk factors for cardiovascular disease include being overweight and obese along with physical inactivity and a sedentary lifestyle (Li et al., 2006). Therefore, the implementation of regular physical activity is considered a viable and safe method for increasing energy expenditure and, in conjunction with dietary habits, is fundamental to helping individuals achieve weight control and improve physiological health outcomes (Osman & Abumanga, 2019; World Health Organisation, 2020).

2.4.3 The effects of physical activity on energy intake and body weight

Physical activity is considered an important component of energy balance that has the potential to influence body weight. Physical activity may increase hunger in some individuals and therefore increase EI, while for others physical activity may have the opposite effect and generate an energy deficit (King et al., 1997; Blundell et al., 2003). This has led to studies investigating the impact of different types (aerobic vs non-aerobic), durations and intensities of physical activity on appetite and EI (King et al., 1997). Overall, daily EI may also be influenced by an individual's metabolic rate, body composition, fat mass, gender and level of fitness (Hopkins et al., 2019). Some research suggests that appetite and subsequently EI may be reduced after an acute exercise session, a response often referred to as "exercise induced suppression of appetite" (King et al., 1997). In a study investigating the 24-hour response of exercise on appetite and EI among untrained obese females (mean BMI 32.5 $kg \cdot m^2$ and age 33 years) found that appetite and the desire to eat were significantly reduced following a high intensity exercise session (Khalaj & Mirzaei, 2020). In contrast, research among healthy, moderately active females reported that EI was increased one hour after brisk walking at 70% VO_{2peak} but was increased to a lesser extent following low intensity exercise (40% VO_{2peak}) despite exercising for a similar duration (Pomerleau et al., 2004). These results may suggest that low intensity exercise has the potential of inducing an acute negative energy balance in healthy active females (Pomerleau et al., 2004) placing them at a higher risk of LEA as they may consume inadequate energy following exercise, or assume that additional energy is not needed following low intensity exercise. However, a review by Thackray et al. (2016) reported that following acute exercise, EI remains unchanged in both healthy weight and overweight and obese females (Thackray et al., 2016). Whether this reduction in appetite and consequently EI persists over the ensuing days or week following regular exercise is yet to be determined. The short follow up period in many of these studies limits the ability to draw effective conclusions about the impact of exercise intensity and EI over a longer time period (i.e., months or years). One of the few studies to measure compensatory EI following moderate and high intensity exercise over a 14-day period found that calorie intake increased, and participants compensated for 56% of exercise induced energy deficits over the 14 days (Whybrow et al., 2008).

Appetite regulation and subsequently EI appears to vary across different studies and between males and females. In a study that randomly assigned males and females into an exercise group (30 minutes twice a day for 5 days) or a non-exercise control group, noted that females in the exercise group expended an additional 382 kcal per day through exercise, but did not increase their daily EI (Staten, 1991). In contrast, male participants increased their EI by 208 kcal per day following exercise for the 5-day exercise period (Staten, 1991). Due to the short duration of the study no changes in body weight were observed, although it was expected that if the caloric deficit persisted more than 5 days, then weight loss in female participants may have occurred (Staten, 1991).

Physical activity has been shown to increase the sensory attractiveness of food for females, but not for males, and thus physical activity does not suppress hunger the same way for females as for males (Bilski et al., 2009). There is high interindividual variability associated with hunger and EI which could help explain the different effects observed with EI following different exercise intensities, making individual responses to exercise and weight loss difficult to determine (Amaro-Gahete et al., 2019; Blundell et al., 2015).

There is overwhelming evidence that, particularly among overweight and obese individuals, modest weight reduction can improve long term health outcomes including reduced blood pressure, improved blood lipid status and reduced all-cause mortality risk (Poobalan et al., 2004). Although physical activity of 150 to 250 minutes per week can help to prevent weight gain and may also result in a modest weight loss (Donnelly et al., 2009) exercise alone without caloric restriction often results in only minimal weight loss (Franz et al., 2007). In fact, physical activity greater than 250 minutes per week may be needed to achieve significant weight loss (Donnelly et al., 2009). Importantly though, for individuals who are of a healthy weight and BMI, a further reduction in body weight is not necessarily correlated with the same health outcomes, and may in fact be detrimental . In a review, Harvie & Howell (2017) identified that intermittent energy restriction (>60% reduction) among people with overweight and obesity could improve eating behaviours and mood. However, the review raised the concern around the possible harm with intermittent energy restriction amongst normal weight individuals (Harvie & Howell, 2017). These concerns were based on evidence that large fluxes in hepatic and intramyocellular triglyceride stores may occur, and have been found to be three times greater after >60% energy restriction compared to a 12 hour overnight fast (Salgin et al., 2009). These fluxes have the potential to lead to skeletal muscle insulin resistance which may ultimately cause weight gain among normal weight individuals.

Among young females, four weeks of intermittent energy restriction compromised of four days with 70% energy restriction and three days ad libitum eating resulted in more negative eating behaviours (Laessle et al., 1996). At four weeks, there was an increase in feelings of hunger, worse mood, heightened irritability, difficulties concentrating, increased fatigue, eating-related thoughts and fear of loss of control (Laessle et al., 1996). These findings suggest that if an individual is going to restrict their EI, there is need for close supervision and caution for certain groups who may be more vulnerable to these behaviours.

To gain more insight into the impacts of EI following exercise, studies need to be carried out over a longer time period. For some individuals, exercise may increase EI, while for others it may decrease EI, and these behaviours may have to be monitored in order to help the individual with their overall health goal. Energy restriction and modest physical activity levels for those who are overweight and obese and presenting with adverse health outcomes may serve to aid in weight loss, improve eating and lifestyle behaviours. Within healthy weight individuals, maintaining a healthy diet to support daily living demands and exercise is usually advised.

2.5 Healthy diet to support physical activity

Along with regular physical activity, a healthy well-balanced diet plays a crucial role in overall health and reduces non-communicable disease risk. In addition, a well-balanced diet provides the body with essential vitamins, minerals and energy to carry out normal physiological functions (Ministry of Health, 2020b). Carbohydrates are an important macronutrient required to restore muscle and liver glycogen stores following exercise and aid in recovery (Burke et al., 2011; Hawley & Burke, 2010). In recent years training with low carbohydrate availability has be used as a dietary strategy to improve performance (Baar & McGee, 2008), with some evidence of enhanced metabolic adaptions to training (Burke et al., 2011). However, there is limited research demonstrating an improvement in exercise performance resulting from these metabolic adaptations (Burke et al., 2011), and individuals following this practice must increase their protein and fat intakes to maintain adequate total EI (Ministry of Health, 2014).

Most athletes ingest sufficient protein in their diet to maintain lean body mass (Tipton & Wolfe, 2004). Resistance trained female athletes were found to maximise anabolism and minimise protein oxidation by ingesting up to 2 $g \cdot kg^{-1} \cdot day^{-1}$ of protein 8 hours post resistance training (Malowany et al., 2019). Which is higher than the current recommended dietary allowance (RDA) of 0.8 $g \cdot kg^{-1} \cdot day^{-1}$ for the general population, indicating the importance of dietary protein for athletes and active individuals (Poortmans et al., 2012). If protein intakes are insufficient often when an individual is severely restricting their EI, this may affect the metabolic fuel used during exercise and at rest which has the potential to result in a catabolic state and muscle protein breakdown (McLoughlin et al., 1998).

While many females avoid dietary fat as a method for weight loss or maintaining a low body weight (Manore et al., 2017) fat intake less than 15% of total EI is considered too low to maintain body weight and may also compromise essential fatty acids and fat-soluble vitamins (vitamin A, D, E and K) intake (Jequier, 1999). Therefore, to maintain energy balance and optimise training benefits, exercising individuals must tailor their diet to ensure sufficient EI from all macronutrient groups and to match EI with total energy expenditure, effectively fuelling their daily workload (Cialdell-Kam & Manore, 2009; Wasserfurth et al., 2020).

2.6 Defining energy availability

Maintaining adequate EA is important for optimal health, however there are many challenges for individuals to achieve this. Reductions in EA may be unintentional through poor preparation, society pressures or work stress, or intentional for the purpose of weight loss. Regardless of intention, reduction in EI has the potential to result in a state of LEA for both acute and chronic periods of time. Energy availability is defined as, the available energy remaining from EI for normal and healthy physiological functioning (i.e., supporting growth, reproduction, cardiovascular functioning and muscle recovery) after accounting for EEE relative to FFM (Mountjoy et al., 2014). Mathematically, EA is defined as: EA = (EI- EEE)$\cdot$kgFFM$^{-1}\cdot$day^{-1} (Areta et al., 2021).

While there is still debate around the EA thresholds, there have been differences outlined for males and females. It has been concluded that an EA ≤30 kcal$\cdot$kgFFM$^{-1}\cdot$day^{-1} for females is inadequate and results in unfavourable physiological changes, and is termed *clinical LEA* (Melin et al., 2019). The majority of the research suggests that 30-45 kcal$\cdot$kgFFM$^{-1}\cdot$day^{-1} is sub-optimal EA for females and that optimal EA for adequate physiological functions should be at least >45 kcal$\cdot$kgFFM$^{-1}\cdot$day^{-1} for females and 40 kcal$\cdot$kgFFM$^{-1}\cdot$day^{-1} for males (Melin et al., 2019; Wasserfurth et al., 2020).

A state of LEA may last for short or long periods of time. In the short term, LEA leads to a negative energy balance and therefore a reduction in weight. However, even a short period (5 days) of clinical LEA (≤30 kcal·kgFFM^{-1}·day^{-1}) can cause severe endocrine and metabolic alterations (Loucks & Thuma, 2003). In the long term, females with LEA are at increased risk of menstrual dysfunction, gastrointestinal dysfunction, psychological disorders, cardiovascular impairment, poor bone health and metabolic issues, compared to individuals who regularly and consistently achieve adequate EA (Ackerman et al., 2020).

2.7 Low energy availability risk in female athletes and recreationally active females

There is a high prevalence of LEA amongst female athletes, with research reporting 20% to 60% of female athletes from various sporting backgrounds presenting with symptoms of LEA (Ackerman et al., 2018; Day et al., 2015; Melin et al., 2014; Melin et al., 2015). The increase in EEE, and training intensity and duration among female athletes competing at international or competitive level compared to recreationally active females places female athletes at a greater risk of LEA (76.7% vs 23.3%, respectively) (Logue et al., 2019). Females with LEA may have a 25% higher training duration (13.0 vs 9.7 hours) along with 37% higher EEE (1121 vs 705 kcal) than females with optimal EA (Melin et al., 2014; Melin et al., 2015). Other research has suggested that non-athletic females are at greater risk of LEA (69.2%) than elite athletes (60.4%) (Torstveit & Sundgot-Borgen, 2005).

The implications of LEA in non-athletic females are currently not well known. One study by Slater et al. (2016) found that 45% of recreationally active females in New Zealand had symptoms of LEA. A later study by Black et al. (2018) stated that among females currently meeting the physical activity guidelines in New Zealand, 63.2% were at risk of LEA based on the LEAF-Q scoring, where a score of ≥ 8 indicates risk of LEA. Further recent studies have found 23.3% to 35.0% of recreationally active females are at risk of LEA (Logue et al., 2019; Meng et al., 2020), which is lower than previously reported by Black et al. (2018). This may be attributed to differences in study design; Black et al. (2018) and Logue et al. (2019) included participants who were currently meeting the physical activity recommendations, as compared Meng et al. (2020) and Slater et al. (2016) who included exercising females, who may or may not have been meeting physical activity recommendations. Research among recreationally active females, showed that for every extra hour of exercise per week, the risk of LEA increased by 1.06 to 1.33 times (Logue et al., 2019; Slater et al., 2016).

The study by Black et al. (2018) further considered lipid and hormonal differences of recreationally active females between those at risk and not at risk of LEA based on the LEAF-Q scoring. Those who were at risk of LEA had lower calcium intakes (847 vs 1488 mg) and lower triiodothyronine (T3) concentrations (1.78 vs 2.01 noml/L) compared to females not at risk of LEA (Black et al., 2018). Low T3 concentrations can have further health implications on bone health given the role it plays in promoting bone formation (Ihle & Loucks, 2004). Inadequate calcium intakes can further impair bone formation especially if this occurs during adolescence, a time when peak bone mass is accrued (Goolsby & Boniquit, 2017). The negative impact LEA has on bone health can increase the risk of attaining a stress fracture injury and reduce bone strength, which has the potential to increase the risk of osteoporosis later in life (Papageorgiou et al., 2017). Low T3 concentrations may also indicate a reduction in basal metabolic rate which is the minimum amount of energy required to maintain normal physiological function at rest. A reduction in basal metabolic rate and a decrease in T3 was observed among normal weight females who were severely energy deficient (Koehler et al., 2017). A reduction in basal metabolic rate may result in individuals further decreasing their EI to achieve body weight reductions or goals and may further exacerbate the state of LEA.

Slater et al. (2016) considered other risk factors of LEA such as exercise behaviour, with 78% of participants having exercised at least once a month for greater than 60 minutes with the intention of losing or controlling their weight, and almost half (45.9%) of these participants were at risk of LEA (Slater et al., 2016). Of the recreational females involved in team and individual sports, 34.8% and 69.6%, respectively were at risk of LEA (Slater et al., 2016). This research highlights that LEA is not only an athlete problem but also has a profound impact on non-athletic recreational females within the community as outlined in Table 1.

Table 1. Estimated prevalence of LEA in recreation and general population females

Author	Sample size	Mean age ± SD (years)	Population	Participants with LEA <30 kcal·kg^{-1}·FFM^{-1}·day^{-1} (%)	Participants at risk of LEA (based on LEAF-Q score ≥8)
Meng et al. 2020	114	20 ± 2	Recreational athletes	N/A	35.1%
Logue et al. 2019	235	18-44	Recreationally active females	N/A	23.3%
Black et al. 2018	38	22.6 ± 5.6	Active females meeting or exceeding physical activity guidelines	N/A	63.2%
Slater et al. 2016	109	23.8 ± 6.9	Recreationally active females	N/A	45%
Reed et al. 2015	91	23.1 ± 0.5	Exercising females	31.9%	N/A
Reed et al. 2013	19	19.2 ± 0.3	Division I soccer players	26% pre-season, 33% during, 12% post-season	N/A

Abbreviations: SD = standard deviation LEA = low energy availability, FFM = fat free mass, LEAF-Q = low energy availability in female's questionnaire.

2.8 Methods to assess LEA and disordered eating behaviours

Early detection of LEA is important to allow early intervention. The risk of LEA in females is often assessed using the validated questionnaire, LEAF-Q (Logue et al., 2019), however, there are variations between studies in the methods used to assess LEA. In the study by Ackerman et al. (2018), LEA was determined by using participants' responses to eating disorder and disordered eating questionnaires. For the research by Ackerman et al. (2018) the brief-eating disorder in athletes' questionnaire, eating disorder screen for primary care and self-reported current or history of eating disordered or disordered eating were used. Low energy availability was determined by the individual's overall score to one or more of the questionnaires, indicating they were likely to have an eating disorder (Ackerman et al., 2018). It is worth noting that a female may be in a state of LEA with or without a clinical eating disorder and thus using disordered eating questionnaires may not always be appropriate when determining LEA risk. The calculation of EA, using the comparison of EI to EEE relative to FFM is often used as another method to determine LEA, particularly if disordered eating or an eating disorder is not observed in the participant cohort (Burke et al., 2018; Viner et al., 2015). However, this calculation is limited by error associated with some of the methods used to measure the components of EA (Gibbs et al., 2013). Therefore, in order to assess LEA risk based on presentation of symptoms associated with chronic LEA states, a standardised questionnaire such as LEAF-Q is frequently implemented by researchers and health care professionals.

2.9 Disordered eating behaviour and eating disorders in New Zealand

Disordered eating includes a wide range of eating behaviours and is experienced by both females and males. This condition may include the use of laxatives, binge eating, exercising for the purpose of burning calories, restriction of food and strict rules or behaviours around food (Hinton & Beck, 2005). The level of functionality and obsession around eating disorder thoughts and behaviours can help distinguish disordered eating patterns and a clinically diagnosed eating disorder, though it is worth noting that most eating disorders begin as disordered eating behavioural pattern. Needless to say, this does not imply that everyone who suffers from disordered eating will go on to develop an eating disorder such as anorexia nervosa, bulimia nervosa, binge eating disorder or eating disorders not otherwise specified (Reel et al., 2013).

Within New Zealand, approximately 1.7% of the general population will develop an eating disorder (Ministry of Health, 2008). It is estimated that 60% of these individuals will fully recover, 20% will partially recover and 20% will never recover (Ministry of Health, 2008). About 40% of those who develop anorexia nervosa will later develop bulimia nervosa (Ministry of Health, 2008). Eating disorders are debilitating and cause significant distress not only to the individual but also to their families and may have long-term adverse health outcomes (Gilbert et al., 2000). Eating disorders, in particular bulimia nervosa can cause tooth decay, oesophageal ulceration, electrolyte imbalanced, cardiac arrythmia and suppressed thyroid functioning (Fisher, 1992). Anorexia nervosa can result in amenorrhea and osteoporosis, as well as hypoglycaemia and in extreme cases, can result in organ failure

(O'Brien et al., 2017). Treatment is often long term and expensive, and in some instances may require in-patient hospital care costing the health care system (Hay et al., 2014).

2.9.1 What we know in female athletes

Female athletes are at a higher risk than male athletes for developing disordered eating behaviours and eating disorders (Van-Niekerk & Card, 2018; Sundgot-Borgen & Torstveit, 2004). There appears to be higher rates of disordered eating behaviours among athletes from aesthetic sports (e.g., gymnastics, diving), with 46.6% presenting with disordered eating (Whitehead et al., 2020), compared to females in endurance sports (24%) (Logue et al., 2019). Female athletes in weight-sensitive sports are thought to be more susceptible to disordered eating due to the increased pressure to meet the perceived athletic body ideals (Ismailova & Landowska, 2016; Reed et al., 2013). Although LEA can occur with and without clinical eating disorders, investigating eating behaviours in active females can provide insight into their risk of developing LEA as their athletic careers progress.

2.9.2 What we know in general population females

The research shows the lifetime prevalence of clinical eating disorders in adolescents is around 13%, of which 1.7% and 2.6% meet criteria for anorexia nervosa and bulimia nervosa, respectively (Mairs & Nicholls, 2016). While there seems to be a high prevalence of disordered eating among female athletes' other studies suggest a greater proportion of non-athletic females are at an increased risk of eating disorders (Van-Niekerk & Card, 2018; Whitehead et al., 2020; Wollenberg et al., 2015; Martinsen et al., 2009) and compulsive exercise behaviours (Fewell, 2018). This may be due to individuals with frequent dieting behaviours having greater emotional attachment to and pre-occupation with exercise frequency (Ackard et al., 2002).

For athletes and non-athletes there are similar factors that may impact on the risk of disordered eating and eating disorders. Nearly half of female (mean age 21.1 years) athletes (51%) and non-athletes (48%) reported negative thoughts and feelings around self-esteem, body image and hurtful peer modelling as a significant contributor for disordered eating and risk of developing an eating disorder (Arthur et al., 2018). In addition, 19% of athletes and 13% of non-athletes reported that being around others with an eating disorder or being surrounded by people who frequently engaged in weight loss talk contributed to their eating disorder risk (Arthur et al., 2018). These findings suggest that non-athletes and athletes with eating disorders tend to have similar levels of impairment. However, for athletes, sporting pressures have a more significant impact on disordered eating, whereas for non-athletes, lack of support (e.g., people not willing to understand or be sensitive regarding an individual's relationship with food) appears to be a more significant influencer on disordered eating behaviours (Arthur et al., 2018).

2.9.3 Body dissatisfaction - what we know in female athletes and general population females

Research into body dissatisfaction among athletes and non-athletes remains inconsistent, with a meta-analysis review finding some research reporting higher prevalence in athletes, whereas other research has found greater body dissatisfaction among non-athletes, (Hausenblas & Downs, 2001; Swami et al., 2009). The overall effect size from the meta-analysis review by Hausenblas & Downs (2001) showed that athletes appear to have more positive body image perspective compared to controls. Self-esteem and females' perceptions about their body weight can be an important determinant of the onset of eating disorders (Ackard & Peterson, 2001). Among females, dieting frequency is positively associated with eating disorder behaviours and its characteristics, body dissatisfaction and drive for thinness (Ackard et al., 2002; Blair et al., 2017 and Ponorac et al., 2018).

Body dissatisfaction can lead to the use of weight loss products including laxatives, diuretics and appetite suppressants (Ponoarc et al., 2018; Whitehead et al., 2020). Some research reports non-athletic females have a higher use of laxatives compared to female athletes (5.7% vs 1.4%), suggesting greater body dissatisfaction among non-athletes (Martinsen et al., 2009). In a study by Blair et al. (2017), non-athletic females were found to be at higher risk for disordered eating and were significantly more dissatisfied with their body shape than athletic females (Blair et al., 2017). The use of weight loss products and disordered eating is a concern as it may mean females who partake in these unhealthy weight loss strategies are less likely to be achieving adequate nutritional intake to support normal physiological functions. Body dissatisfaction may lead to restrictive dieting, which can impact an individual's overall nutritional intake and result in a state of clinical LEA for prolonged periods of time.

2.10 Menstrual dysfunction

Reproduction is closely related to EA, with a healthy reproductive cycle requiring sufficient energy to maintain the proliferation of the endometrium in the luteal phase of the menstrual cycle (Mountjoy et al., 2018). In a state of LEA, there is insufficient energy available to maintain high rates of reproduction, growth and development resulting in downregulated reproductive processes (Elliot-Sale et al., 2018). In a state of LEA, the downregulation of menstruation results in suppression of luteinising hormone pulsatility and subsequently there appears to be a decline in the primary reproductive hormones estrogen and progesterone, which is outlined in the work by Loucks et al. (1998) and Koltun et al. (2019).

Among healthy menstruating females (mean age 21 years) LEA caused by excessive EEE reduced luteinising hormone pulse frequency to a greater extent over a 24-hour period compared to LEA induced by dietary restriction (Loucks et al., 1998). Koltun et al. (2019) further found that among females (mean age 20 years) EA predicted luteinising hormone pulse frequency, in which every single unit decrease in EA, resulted in a 0.017 pulse per hour decrease in luteinising hormone pulse frequency (Koltun et al., 2019). The consequences of the decreases in luteinising hormone pulsatility lead to a downregulation oestrogen and progesterone, subsequently leading to the suppression of the hypothalamic pituitary ovarian axis and the inhibition of gonadotropin releasing hormone (Loucks & Thuma, 2003 & Iwasa et al., 2018). The suppression of gonadotropin releasing hormone will likely result in delayed puberty and/or functional hypothalamic amenorrhea (Iwasa et al., 2018). The results of the research by Loucks et al. (1998) showed that prolonged exercise itself does not have a negative impact on the hypothalamic pituitary ovarian axis, rather it appears that the energy cost of exercise and the effect on EA has the profound impact on menstrual function in regards to luteinising hormone pulsatility (Loucks et al., 1998). Menstrual disorders may be preventable or reversed by increasing EI to compensate for the increased EEE.

Menstrual disorders exist on a continuum from normal (eumenorrhea) to dysfunctional (oligomenorrhea) and to no menses at all (amenorrhea) (Brown, 2011). Amenorrhea is the clinical endpoint of menstrual dysfunction which may be due to a wide variety of underlying causes. Primary amenorrhea is usually diagnosed when an individual has no onset of menses by age 15 and is typically related to dysfunction of processes controlled by the hypothalamus

(Rebar, 2018). In contrast, secondary amenorrhea or functional hypothalamic amenorrhea is when there is no menstruation for ≥3 months (Rebar, 2018). Functional hypothalamic amenorrhea is responsible for 20 to 35% of secondary amenorrhea and is primarily related to weight loss, disordered eating, stress and exercise (Bedford et al., 2010; Schneider & Warren, 2006; Meczekalski et al., 2014). Among active females, Black at el. (2018) found that those who had previously or were currently exhibiting secondary amenorrhea, 73.3% had LEA further highlighting the relationship between LEA and menstruation (Black et al., 2018).

2.10.1 Menstrual dysfunction related to dietary intake and exercise

Insufficient EI either from unintentional or intentional food restriction is one of the primary contributors of exercise related menstrual disorders in active females (Mountjoy et al., 2014). Research has demonstrated that active females with menstrual disorders consumed 12% less overall energy compared to those without menstrual disorders, emphasising the quantity and quality of diet and energy required to maintain healthy reproductive functioning (Hand et al., 2016).

Physical activity has shown to be advantageous to help relieve some menstrual pain and symptoms (Dusek, 2001; Bavil et al., 2018) indicating that physical activity is not always detrimental to the female menstrual cycle. The prevalence of dysmenorrhea has been reported to be higher in sedentary females (57.3%) as compared to athletes (26.7%) (Dusek et al., 2001), which could be explained by higher physical activity levels associated with a lower incidence of dysmenorrhea (Bavil et al., 2018).

Current research may suggest a dose response of physical activity that supports menstrual cycle function, in which large increases in physical training place a female at increased risk of

LEA and subsequently results in the downregulation of menstrual functioning (Williams et al., 2001). However, if EA is maintained in the presence of physical activity, this may serve to reduce negative outcomes on the hypothalamic-pituitary-ovarian axis and pre-menstrual symptoms. Thus, maintaining quality nutritional intake while exercising and having a good relationship with food may support healthy reproductive functioning in females.

2.11 Knowledge of LEA and its consequences among health care professionals and in the general population

Individuals with health concerns often seek help from health professionals, so it is important that these health professionals are aware of the risk factors and physiological outcomes associated with a LEA. A recent study in New Zealand investigated knowledge of LEA and the female athlete triad among health care professionals from a variety of fields including dietitians, physiotherapists, nutritionist, physicians and personal trainers (Ashby, 2019). Overall, 55% of health care professionals understood the female athlete triad and 23% knew of LEA, leading the authors to conclude that health care professionals were generally unaware of these terms (Ashby, 2019).

Athletes themselves may also not be aware of the health implications of excessive training and under-eating, with 60% of female athletes reporting that they had never heard of the female athlete triad, 24% could name at least two of the components and only 8% could name all components of the female athlete triad (Day et al., 2015). These results in comparison to a more recent study where knowledge of the female athlete triad among active females in New Zealand suggested that 35% had heard of at least one of the three female athlete triad components (Winter et al., 2019). Furthermore, 13.5% had good knowledge of the two components and 2.9% had good knowledge of all three components of the female athlete triad (Winter et al., 2019).

As expected, there are some knowledge gaps and misconceptions around LEA amongst females as shown in the research by Day et al. (2015). Prior to any form of nutrition education, 64% of females thought skipping a period was normal, 60% did not think skipping a period was related to training intensity and 40% believed there was a set body fat percentage for optimal performance (Day et al., 2015). This implies that there are females who are unaware of some of these risk factors of LEA which could potentially lead to a downward spiral and increase their risk of LEA if these behaviours continued. Although some knowledge gaps were evident, 85% of females indicated a desire to learn more about female health which emphasises the need for nutrition education interventions for this group of exercising females (Winter et al., 2019). Other studies have shown that athletes have the nutrition knowledge necessary for correcting eating patterns although they may choose not to practise that knowledge for various reasons (Day et al., 2015). The recent study by Day et al. (2015) found that despite an increase in nutrition knowledge after the nutrition education intervention there were no changes in daily EI or eating behaviour.

Although education interventions can help with some knowledge deficits there also needs to be a focus on opportunities to practise dietary skills and understanding barriers to making dietary change (Day et al., 2015; Winter et al., 2019). Nutrition interventions may also play an important role to help prevent and detect signs and symptoms of LEA especially before negative health and performance consequences occur.

2.12 Conclusion

The impact of LEA in females and males has been demonstrated across a variety studies. To date, the literature has focused on female athletes, however, it is clear that LEA is also prevalent among other population groups such as recreational females and male athletes and non-athletes (Slater et al., 2016; Black et al., 2018; Viner et al., 2015; Koehler et al., 2016; Wasserfurth et al., 2020; Mountjoy et al., 2018).

Low energy availability can occur with and without clinical eating disorders and thus using disordered eating questionnaires may not always be appropriate to determine LEA. To enable accurate assessment of LEA risk among females, the use of a validated questionnaire such as the LEAF-Q will help to determine an individual's risk of LEA (Logue et al., 2019). Determining if an individual is at risk of LEA may aid in the early detection of a clinical LEA state and should be used as a screening tool for detection. Early detection is especially important given that physiological and psychological health consequences can be observed among those who remain in LEA even for a short duration (days), although more severe consequences are observed the longer an individual is in a state of LEA (months/years) (Mountjoy et al., 2018). The current research suggests that inadequate knowledge among exercising individuals is one of the main contributors to LEA (Ashby, 2019; Day et al., 2015).

It is important to raise awareness around the implications of LEA and the related health consequences not only in exercising individuals but also among coaches and health professionals (Day et al., 2015). Understanding the associated risk factors of LEA such as body dissatisfaction, disordered eating behaviours and exercise behaviours may be helpful in recognising if a female is at risk of LEA prior to presenting with physiological symptoms.

Chapter 3: Research study

Abstract

Background: Low energy availability (LEA) has primarily been studied in athletic females with few studies examining the risk of LEA among non-athletic females. Low energy availability may result from dietary manipulations or changes to exercise behaviours which can lead to long term negative health implications. Therefore, understanding some of the risk factors of LEA in non-athletic females may allow for early identification of habits that contribute to LEA. The overall aim of the present study is to investigate the associations of eating and exercise behaviours on LEA risk in non-athletic females in New Zealand. As well as investigating the relationships between physical activity, menstrual function and eating behaviours in non-athletic females in New Zealand through an online questionnaire. We hypothesise that non-athletic females with high and/or frequent exercise behaviours will be classified as at risk of LEA and will have negative feelings towards exercise and eating compared to those not at risk of LEA.

Method: The risk of LEA was determined using a validated screening tool; LEAF-Q which was completed through an online survey. Additional risk factors of LEA were further identified using the New Zealand Physical Activity Questionnaire - short form, CIA Questionnaire and Eating Disorder Questionnaire. The population group consisted of females aged 18–30 years who did not classify themselves as athletes. Participants were proficient in English, not currently breast-feeding and not currently suffering or had previously suffered from an eating disorder in the last 12 months.

Results: The results showed 51% (n=77) of participants were classified as at risk of LEA. The study demonstrated 26.5% of participants were classified as having low physical activity levels, 42.4% as moderate physical activity levels and 31.1% as reaching high levels of physical

activity. There was a significant relationship between number of days of vigorous activity and LEAF-Q score ($p<0.001$). Individuals with a high level of impairment on the CIA had significantly higher LEAF-Q scores across all three CIA components; eating habits ($p<0.001$), exercise habits ($p=0.002$) and body shape ($p=0.002$).

Conclusion: This cross-sectional study highlights the multi-directional relationship between exercise and eating behaviours and LEA risk. Non-athletic females in this current study presented with poor perceptions towards eating and body weight based on the CIA questionnaire which may place them at risk of LEA.

3.1 Introduction

Reduced EI can be intentional or unintentional. Intentional reductions in EI may be a way of controlling body weight and may be achieved through restrictive dieting and exclusion of certain food groups. These dietary behaviours may be related to disordered eating, body dissatisfaction and societal pressures around body weight (Mountjoy et al., 2014). There may also be unintentional reductions in EI through increased energy expenditure or inadequate knowledge about nutritional practices. These reductions in EI, both intentional and unintentional, and in combination with an increase in energy expenditure through exercise may result in a state of LEA (Mountjoy et al., 2018).

Low energy availability can lead to long-term health implications which were originally thought to be limited to the Female Athlete Triad, a syndrome compromised of disordered eating, amenorrhea and osteoporosis (Thein-Nissenbaum, 2013). Disordered eating behaviours may cause LEA and down regulate the reproductive and skeletal systems (Warren & Stiehl, 1999). Females who have an elevated dietary restraint have a significantly greater incidence of low bone mineral density than female adolescent runners without dietary restraint (Barrack et al., 2008). A decrease in lean body mass, BMI, low levels of insulin-like growth factor and malnutrition are all contributing factors to low bone mass. The most important factor to counteract bone loss, especially in young females with disordered eating behaviour is the normalisation of body weight (Misra, 2008). Unfortunately bone mineral density may not improve with weight gain and restoration of menses, which places these females at risk of other health complications later in life such as osteoporosis (Thein-Nissenbaum, 2013) and stresses the importance of early identification. It is now recognised that the consequences of LEA are multi-factorial which has led to the introduction of RED-S, that identifies the multiple physiological and psychological systems affected by LEA in both males and females (Mountjoy et al., 2014).

Low energy availability has been primarily investigated in an athletic setting, as it was thought to be due to high training volumes and sporting pressures. Hence, until recently LEA had typically been considered an athlete problem. Research regarding body image and eating disorder concerns in athletes has found 25.3% of female athletes from various sporting backgrounds to have severe or moderate body image dissatisfaction, which is concerning given that body image concern is a strong predictor in the development of eating disorders (Neves et al, 2017; Smith & Petrie, 2008). Of female athletes from a range of sports (mean age 19.3 years), 23.5% perceive themselves as being overweight (Haase, 2011), and reported more disordered eating compared to athletes who perceived themselves as normal weight (Haase, 2011). Such thoughts and behaviours may place these individuals at risk of LEA.

The implications of similar behaviours (eating and exercise) and perceptions of body weight and body satisfaction in recreationally active or sedentary females and association with LEA risk is currently unknown. A study by Slater et al. (2016) is the only study within New Zealand looking at LEA risk among recreationally active females, of which 45.0% were at risk of LEA. A recent study by Logue et al. (2019) was conducted in Ireland and found that 23.3% of recreationally active females were at risk of LEA, which is lower than that reported by Slater et al. (2016). This discrepancy may be attributed to differences in study design, as Slater et al. (2016) include participants who may or may not be meeting the physical activity guidelines, whereas Logue et al. (2019) included females who were physically active which was defined by adhering to the physical activity guidelines.

Given that recent research has shown LEA occurs not only in female athletes but appears to be highly prevalent in recreationally active females, research into the risk factors inclusive of behaviours towards eating and exercise and perceptions of body satisfaction is required. Findings from further investigations may aid in identifying early risk behaviours that contribute to chronic LEA and associated health outcomes.

3.2 Methods

3.2.1 Participants

The study was a cross-sectional design in which 531 female participants residing in New Zealand were recruited. Participants were recruited through university advertising and email lists, word of mouth and social media platforms (e.g., Facebook, Instagram, Twitter) between the period of September 2019 and January 2020. Participants completed an online pre-participation screening questionnaire (Appendix A) to determine eligibility for the study. Inclusion criteria were being female, aged between 18-30 years, proficient in English, not currently breast-feeding and not suffering from an eating disorder or previously suffered from an eating disorder in the last 12 months (Appendix B). Participants that met the inclusion criteria were required to read an information sheet (Appendix C) on the study and complete an online consent form before proceeding to the full questionnaire. Recruitment, screening and final numbers for the research study are presented in Figure 1. Ethical approval was received from the Massey University Ethics Committee (SOA 19/35) prior to recruiting participants.

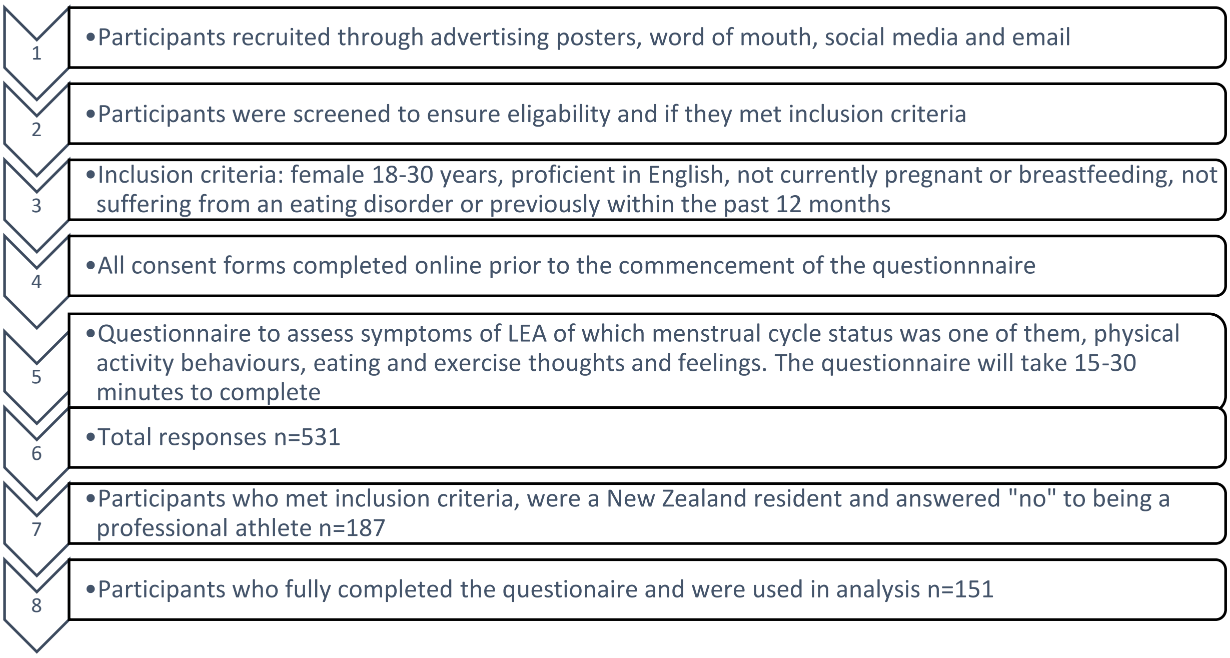

Figure 1. Flow diagram of inclusion and exclusion criteria for participant recruitment and data analysis

3.2.2 Procedure

Data were collected through an online questionnaire which consisted of an amalgamation of previously validated questionnaires, using Qualtrics^{XM} Survey Software (Qualtrics, Provo, UT).

The online questionnaire contained 64 items and took approximately 15-30 minutes for participants to complete. Demographic information was collected, including self-reported weight and height to enable estimation of BMI and whether participants classified themselves as professional athletes. Non-athletes were defined in this study as participants who responded “no” to the screening question asking if they considered themselves a professional athlete. Defining whether someone is an ‘athlete’ can be challenging due to the different terminology and ambiguity as there may be some individuals who perform at a competitive level and excel in their respective sporting fields but might not necessarily consider themselves an athlete. Any participant who answered “yes” to being a professional athlete was excluded from the current data set, to ensure only non-professional recreationally active or sedentary females were included in the analysis.

The online questionnaire incorporated four validated questionnaires. The New Zealand Physical Activity Questionnaire (short form; NZPAQ-SF) was used to assessed physical activity (Maddison et al., 2007), the Eating Disorder Questionnaire (EDE-Q) (Mond et al., 2004; Gideon et al., 2018) and Clinical Impairment Assessment (CIA) Questionnaire (Bohn & Fairburn, 2008) were used to assess participants’ perceptions towards exercise and eating habits, body shape and weight and LEAF-Q was used to assess LEA risk (Melin et al., 2014).

3.2.3 Physical activity

The NZPAQ-SF was used to assess the three dimensions of physical activity; frequency, duration and intensity of physical activity over a 7-day period (Maddison et al., 2007). The NZPAQ-SF strongly correlates to the International Physical Activity Questionnaire-long form which has been shown to have acceptable reliability (0.79 between 0 and 8 days and 0.74

between days 8 and 15) when assessing physical activity ($p<0.001$) (Madison et al., 2007). Individuals were asked to recall their physical activity over the past 7 days, including the frequency and time duration of brisk walking, moderate and vigorous activity. The time duration and number of days per week of physical activity was used for the analysis in this study. For the purpose of the current study, individuals who were well exceeding the physical activity guidelines (see below) were classified as high physical activity, individuals who were meeting the guidelines are moderate physical activity and individuals who were not currently meeting the guidelines are classified as low physical activity. We have defined exercise levels based on the following criteria:

<u>High physical activity:</u> ≥10 days of moderate and vigorous activity

<u>Moderate physical activity:</u> ≥5 and <10 of moderate and vigorous activity

<u>Low physical activity:</u> <5 days of moderate and vigorous activity

3.2.4 Clinical Impairment (CIA) Questionnaire

The purpose of the CIA is to measure overall impairment and severity related to eating disorder features. The CIA has 3 components which are categorised into exercise habits, eating habits and body weight perception. Each component of the CIA has 16 items that are rated on a Likert scale, with 'not at all' scoring 0, 'a little' scoring 1, 'quite a bit' scoring 2 and 'a lot' scoring 3. For each component the participant can score between 0 to 48 on the 16 item questions. For each component (exercise habits, eating habits and body weight perception) a score of >16 indicates a high level of impairment and a score ≤16 indicates a low level of impairment. A combined score across all 3 categories of >48 indicates a high level of impairment and a combined score ≤48 indicates a low level of impairment (Bohn et al., 2008).

3.2.5 Eating Disorder Questionnaire (EDE-Q)

The EDE-Q is considered the gold standard for assessment of eating disorders and has been shown to have an acceptable reliability and has been validated among clinical, adult and adolescent population groups (Luce & Crowther, 1999; Mond et al., 2006). The CIA and EDE-Q are designed to be completed immediately after each other to ensure that patients have their eating disorder features in mind when filling in the CIA. The EDE-Q was not used in this analysis as the focus was on CIA and association with LEAF-Q.

3.2.6 Low energy availability in females Questionnaire (LEAF-Q)

The LEAF-Q is a validated questionnaire used to assess participants' risk of LEA based on the presentation and frequency of symptoms commonly associated with LEA (Melin et al., 2014). The LEAF-Q is a 25-item questionnaire which focuses on the physiological symptoms of LEA including current and previous injuries, gastrointestinal and reproductive function (Melin et al., 2014). Each question in the respective category is graded to give the overall score depending on the frequency and severity of their symptoms, which would give an overall score between 0 and 25. The variable score producing the highest sensitivity and specificity for the corresponding Triad end point was used to determine the cut-off for each category (Melin et al., 2014). A score of ≥2 for gastrointestinal symptoms, ≥2 for injuries and ≥4 for menstrual function, gives an overall score of ≥8, which indicates the individual is at risk of LEA

(Melin et al., 2014). A score of <8 was an indicator the individual is not at risk of LEA (Melin et al., 2014).

3.2.7 Menstrual status

Normal and irregular menstruation was determined using the information gathered from the LEAF-Q. Information on an individual's menstrual status was provided through questions on the duration of the menstrual cycle, when their last period was and how many periods they have had in the last year. Individuals who responded "yes" to the question "do you have normal menstruation" were reported as having normal menstruation.

3.2.8 Data analysis

For this cross-sectional study a sample size of 265 participants was needed to have the least amount of Type 1 and Type 2 errors, and for the results to be statistically significant. By selecting the maximum chance of type 1 errors at 0.5% and type 2 errors at 25% and the smallest beneficial correlation as 0.1, the quality of the hypothesis test can be increased (Hopkins, 2006). The sample size number was determined by using outcomes from previous published studies using similar population and methods (Slater et al., 2016).

3.2.9 Statistical analysis

Data were collected using the online survey software, Qualtrics), and exported to MS Excel. All statistical analyses were performed using IBM SPSS statistics version 27. Means and standard deviations (SD) were calculated. Spearman, r correlation co-efficient were used to assess the degree of linear association between variables. In order to examine the mean differences between the groups, t-tests were implemented. All variables were checked for normality using Kolmogorov-Smirnov test. Effect sizes were calculated by determining the difference between the means divided by the standard deviation. Based on the effect size calculation, a value of 0.2 was considered small, 0.5 medium and 0.8 was a large effect size. Regression analysis was conducted to explore the relationships between the dependent variable (LEAF-Q score) and independent variables (BMI, age, overall physical activity score, eating behaviours, menstrual function). A value of $p<0.05$ was considered significant between all groups.

3.3 Results

3.3.1 Characteristics of participants

A total of 531 participants took part in the online questionnaire, of which 187 participants met the inclusion criteria outlined for this study. Of these 187, a total of 151 participants fully completed the questionnaire and were therefore included in this analysis. The demographic characteristics of participants are presented in Table 2. Participants were predominantly New Zealand European (78.4%), either currently or had previously been enrolled in tertiary education (95.3%) and were non-smokers (94.7%). The mean age of participants was 23.0 ± 3.2 years, with age group analysis showing that 29.1% were 18-20 years, 46.4% were 21-25 years and 24.5% were 26-30 years of age. There was no significant relationship between LEAF-Q score and age ($p=0.574$). Participants average self-reported height and weight and BMI are shown in Table 2. According to the BMI cut-off values, majority of the participants (68.4%)

met the classification of a healthy weight (18.5-24.9 kg·m^{-2}). There was a non-significant (p=0.188) negative correlation -0.122 between LEAF-Q score and BMI.

Table 2. Demographic characteristics of participants

Characteristics (n=151)	
Variable	Mean ± SD
Age (years)	23 ± 3.2
Weight (kg)	65.7 ± 12.7
Height (m)	1.7 ± 0.1
BMI (kg·m^2)	23.8 ± 4.6
Age categories n (%)	
18 – 20 years	44 (29.1%)
21 – 25 years	70 (46.4%)
26 – 30 years	37 (24.5%)
Tertiary education (n=150) n (%)	
Yes	143 (95.3%)
No	7 (4.7%)
Smoking status n (%)	
Yes	8 (5.3%)
No	143 (94.7%)
Ethnicity[1] (n=171) n (%)	
New Zealand European	134 (78.4%)
Māori	10 (5.6%)
Cook Island Māori	1 (0.6%)
Chinese	5 (2.9%)
Indian	3 (1.8%)
Other[2]	18 (10.5%)
BMI category (n=117) n (%)	
Underweight (<18.5 kg·m^2)	7 (6.0%)
Healthy weight (18.5-24.9 kg·m^2)	80 (68.4%)
Overweight (25-29.9 kg·m^2)	19 (16.2%)
Obese (≥30 kg·m^2)	11 (9.4%)

[1]Multiple ethnicities were able to be selected by participants
[2]Other: Japanese, Singaporean, French, New Zealand Asian, Middle eastern, Filipino, Pakistani, Hispanic, South African, British, Dutch, Australian
SD=standard deviation; BMI=body mass index

3.3.2 Oral contraceptives and menstruation

In this current study, 52.3% of participants used oral contraceptive pills; of those not currently using oral contraceptive pills, 33.1% had used them previously. Only 14.6% of participants reported having never used oral contraceptive pills. Other forms of contraceptive use are reported in Table 3. Over half of the participants (64.2%) reported having normal menstruation, 6.6% were unsure if their menstruation was normal and 29.1% reported they did not have normal menstruation (Table 4).

Table 3. Contraceptive use among females

Oral contraceptive pill use	**n (%)**
Currently using	79 (52.3%)
Previously used	50 (33.1%)
Never used	22 (14.6%)
Other contraceptives	23 (15.2%)
Hormonal coil	4 (2.6%)
Hormonal implant	8 (5.3%)
Depo Provera injection	9 (6.6%)
Mirena	1 (0.3%)
Pill[1]	1 (0.3%)

[1] One participant answered they used pill as a form of another contraceptive

Table 4. Previous and current menstrual dysfunction in recreational/non-athletic females

Menstrual disorders	n (%)
Irregular menstruation	44 (29.1%)
Unsure if their menstruation is normal	10 (6.6%)
Normal menstruation	97 (64.2%)
Regular periods	85 (87.6%)
Irregular periods	11 (11.3%)
History of heavy bleeding	0 (0%)
Amenorrhea	62 (41.1%)
Currently have amenorrhea	19 (12.6%)
Previous amenorrhea	43 (28.5%)
Gaseous or bloated abdomen[1]	112 (74.2%)
Cramps or stomachache[1]	71 (47.0%)

[1] Determined by those who reported having these symptoms once or twice a week, several times a week or several times a day.

No participants reported that they were currently experiencing heavy menstrual bleeding (Table 4). However, when further analysed, 47.7% of participants reported previously experiencing changes in menstruation, of this, 32.9% of these participants reported that one of the changes was that they bled for more days. A greater proportion (81.0%) of those who were not at risk of LEA compared to those at risk of LEA had never experienced amenorrhea, whereas 22.0% of participants who were at risk of LEA reported that they had not had a period during the previous 3 months and as such would be currently classified as amenorrhoeic.

3.3.3 LEAF-Q score

The average LEAF-Q score was 8.3 ± 4.5, with 51% (n= ___) classified as at risk of LEA (score ≥8) and 49% (n= ___) classified as not at risk of LEA (score <8). Of the 6.2% of participants who identified as Māori or Cook Island Māori (n=11), 5 were classified as not at risk of LEA and 6 (55%) were classified as at risk of LEA.

3.3.4 Physical activity habits

In this study, 47.7% of participants reported they did >30 minutes of moderate activity on >5 days a week and 25.8% reported they did >15 minutes of vigorous activity on >5 days a week. Additional analysis demonstrated that 26.5% of participants were classified as having low physical activity levels, 42.4% as moderate physical activity levels and 31.1% as reaching high levels of physical activity. Of participants at risk of LEA (n=77), 20.8% had low, 42.9% had moderate, and 36.4% had high physical activity levels. Of participants not at risk of LEA (n=74), 32.4% had low, 41.9% had moderate and 25.7% had high physical activity levels. The mean LEAF-Q score for the low, moderate and high physical activity group was 6.7, 8.5 and 9.2, respectively.

3.3.5 Physical activity and LEA risk

Pearson correlation showed there was no significant difference between days of moderate activity and LEAF-Q score ($p=0.057$). Although there was a significant positive relationship between days of vigorous activity and LEAF-Q score ($p=0.038$). One-way ANOVA between physical activity groups (low, moderate, high) and LEAF-Q score, showed there was a statistical difference ($F(2)=3.797$, $p=0.025$). A paired samples t-test showed that there was a significant difference between days of moderate activity and LEA risk ($p<0.001$) and between days of vigorous activity and LEA risk ($p<0.001$). A chi-squared test, between physical activity groups and LEA risk (not at risk/at risk) showed no significant relationship ($r(2)=3.328$, $p=0.189$).

3.3.6 Clinical Impairment Assessment (CIA)

The average score for the eating habits component of the CIA was 11.9, with 70.2% of participants classified as low level of impairment and 29.8% as a high level of impairment. For the second component, exercise habits, participants scored a mean 8.5, with 86.1% classified as a low level of impairment and 13.9% as a high level of impairment. For the third component, body weight and shape, participants scored a mean of 10.5, with 77.5% having a low level of impairment and 22.5% reporting a high level of impairment (Table 5). Cumulatively, when considering all three CIA categories, 77.5% of participants were classified with a low level of impairment and 22.5% had a high level of impairment indicated by an overall global score ≥48. On average, the global CIA score was 10.3 ± 10.4.

Table 5. Level of clinical impairment in each CIA subcomponent and average mean score between groups for LEAF-Q score

	Mean ± SD	Low level of impairment on CIA[1] (n=117)	High level of impairment on CIA (n=34)
		Mean ± SD	**Mean ± SD**
LEAF-Q score[2]	8.3 ± 4.5	7.6 ± 4.3	10.4 ± 4.3
CIA variable[3]	**Mean ± SD**	**Low level of impairment n (%)**	**High level of impairment n (%)**
Exercise habits	8.5 ± 9.4	130 (86.1%)	21 (13.9%)
Eating habits	11.9 ± 10.8	106 (70.2%)	45 (29.8%)
Body weight and shape	10.5 ± 11.2	117 (77.5%)	34 (22.5%)

[1]Determined by the combined CIA score across all 3 CIA variables (>48 indicates a high level of impairment and ≤48 indicates a low level of impairment)

[2]LEAF-Q score of ≥8 is classified as at risk of LEA and a LEAF-Q score of <8 is classified as not at risk of LEA

[3] For each CIA variable a score >16 indicates a high level of impairment and a score ≤16 indicates a low level of impairment

Pearson correlation showed that participants who reported irregular menstruation had higher scores across all CIA components (eating, exercise behaviour and body weight perception) compared to those with normal menstruation (Table 6), though the relationship was not significant, exercise ($p=0.128$), eating habits ($p=0.350$) and body shape ($p=0.927$).

Table 6. CIA score among those with normal and irregular menstruation

CIA variable[1]	Mean ± SD		P-value
	Normal menstruation (n=97)	**Irregular menstruation (n=44)**	
Exercise habits	8.1 ± 8.4	10.1 ± 11.8	0.128
Eating habits	11.3 ± 9.7	14.3 ± 13.3	0.350
Body shape and weight	9.7 ± 10.3	13.1 ± 13.4	0.927

[1] For each CIA variable a score >16 indicates a high level of impairment and a score ≤16 indicates a low level of impairment

One-way ANOVA between groups using the Brown-Forsythe test, revealed a significant difference between LEA risk and CIA exercise habits (df (1) =7.878, p=0.006), eating habits (df (1) =16.158, p<0.001) and CIA body shape df (1) =17.875, p<0.001). The results indicated a significant difference in LEAF-Q scores between those with a low level of impairment compared to high level impairment on the global CIA. Individuals classified as having high impairment had significantly higher LEAF-Q scores across all three CIA components; eating habits (t (149) =3.865, p<0.001), exercise habits (t (149) =3.192, p=0.002) and body shape (t (149) =3.217, p=0.002) compared to those with low impairment as shown in Table 7.

One-way ANOVA between groups showed there was a statistically significant difference between physical activity (low/moderate/high) and CIA exercise habits (F (2) =4.688, p=0.011), CIA eating habits (F (2) =3.509, p=0.032) and CIA body shape and weight (F (2) =3.771, p=0.025). There was a positive relationship observed between physical activity and the CIA components.

Table 7. Average LEAF-Q scores for low- and high-level impairment on CIA

CIA variable	LEAF-Q score[1] (Mean ± SD)		P-value
	Low level of impairment on each CIA variable[2]	High level of impairment on each CIA variable	
Exercise habits	7.4 ± 4.4	11.1 ± 4	0.002**
Eating habits	7.4 ± 4.1	10.3 ± 4.7	<0.001***
Body shape and weight	7.7 ± 4.3	10.4 ± 4.2	0.002**

**p<0.05, **p<0.01, ***p<0.001*

[1]LEAF-Q score of ≥8 is classified as at risk of LEA and a LEAF-Q score of <8 is classified as not at risk of LEA

[2] For each CIA variable a score >16 indicates a high level of impairment and a score ≤16 indicates a low level of impairment

3.4 Discussion

This cross-sectional study adds to the current literature providing insight on eating and exercise behaviours and the subsequent associations to LEA risk in healthy but non-athletic females in New Zealand. This research has identified several key areas of concern for females in relation to their risk of LEA. The main finding from this study was that 51.0% of females were at risk of LEA based on the LEAF-Q scoring. This result is similar to a previous study in New Zealand in which the risk of LEA among recreationally active females was explored using the LEAF-Q (Black et al., 2018). In that study, 63.2% of participants were at risk of LEA (Black et al., 2018); this percentage is higher than that reported by Slater et al., (2016) in which 44.9% of females were identified as at risk of LEA. Among active females in Ireland, 39.7% were at risk of LEA, based on LEAF-Q scoring (Logue et al., 2019). Furthermore, Logue et al. (2019) showed that there was a higher risk of LEA among active females who participated in competitive sport (76.7%) compared to females who were recreationally active (23.3%).

Some of these differences are likely to be influenced by the eligibility criteria for participants in the various studies. Participants in the Black et al. (2018) study were currently meeting or exceeding the physical activity guidelines in New Zealand, while Slater et al., (2016) focused on females in New Zealand who might engage in regular physical activity but did not necessarily meet the physical activity guidelines. Based on the inclusion criteria from this current study, there was no requirement for participants to be meeting the physical activity guidelines, which could explain why the percentage of participants at risk of LEA is lower than that seen in the Black et al. (2018) study. A higher percentage of those at risk of LEA in this current study compared to Slater et al., (2016) could be explained by the larger number of participants recruited in this study. Despite differences in participant recruitment there is still a need to focus on healthy exercising females, as the risk of LEA remains a persistent issue among females which needs to be acknowledged.

It would be expected that individuals who are at risk of LEA would have a lower BMI, however this current study found no significant relationship between LEA risk and BMI. Ackerman et al. (2018) reported an interesting finding in which participants with LEA had a significantly higher BMI than those with adequate EA, indicating that LEA might not necessarily be visible and even those with a normal BMI (18.5-24.9 $kg{\cdot}m^2$) may have a low body fat percentage and be at risk of LEA. This emphasises the importance of appropriate screening tools and not solely relying on body composition changes to determine LEA risk. Given that most of the current research to date on LEA has been carried out among Europeans, there is a need to possibly warrant research into other ethnic groups given that 55% (n=6) of Māori or Cook Island Māori participants in this study were at risk of LEA

Typically, prevalence of LEA is thought to be high among athletic females (Melin et al., 2019) however inference from the current research findings would suggest that this may not be correct. To date, research on LEA has focused on athletic individuals, however our research and that of others (Slater et al., 2016; Black et al., 2018; Logue et al., 2019) has clearly highlighted the high prevalence of LEA in non-athletic females. This current study investigated individuals' overall menstruation patterns and examined the presence or absence of menses in females. Information regarding menstrual cycles was gathered using the LEAF-Q, however there was no specific time frame over which menstrual data or duration of absence of menses was assessed, making it difficult to establish the length of time an individual has been experiencing these symptoms. This timeframe is important to acknowledge, as the related health consequences of LEA, such as irregular menstruation, often result after chronic LEA (i.e., months). The subsequent health declines and treatment durations are typically related to the severity and duration of LEA with both short-term and long-term LEA having detrimental consequences on physiological health (Wasserfurth et al., 2020). The consequences of LEA and the severe health implications such as endocrine and metabolic consequences for example decreased anabolic hormones and decreased basal metabolic rate can be observed after only 5 days in a state of LEA (<30 $kcal \cdot kgFFM^{-1} \cdot day^{-1}$) among healthy young females (Wasserfurth et al., 2020), indicating that female reproductive health is highly sensitive to short term LEA.

Within this current study, 64.2% (n=97) of participants reported normal menstruation and 29.1% (n=44) had irregular menstruation. This finding is similar to previous research among healthy female students (18-25 years) of which 27.0% to 33.3% were found to have irregular menstruation (Rafigue & Al-Sheikh, 2018; Fujiwara & Nakata, 2007). Menstrual function is strongly influenced by nutritional status and energy availability (Stickler et al., 2019), indicating that inadequate EA may be one of the root causes of irregular menstruation, especially when there are no other underlying medical conditions. Physical activity duration (hours per week) and intensity has been shown to be have a significant positive association on the presence of menstruation disorders among active females of reproductive age (Peinado-Molina et al., 2020). It is then not surprising that athletes have reported greater prevalence of menstrual disorders in comparison to non-athletes given that the training schedules of athletes are typically more physically demanding than those of individuals involved in recreational sports (Melin et al., 2014; Melin et al., 2015). This difference was outlined in the literature as early as 1983 where 45.0% of athletes experienced irregular menstruation while a smaller percentage of non-athletic controls (33.1%) reported irregular menstruation (Carlberg et al., 1983). However, it is possible that there is a subset of individuals who are not classified as athletes, yet participate in exercise of similar duration, volume and intensity to athletes. Exercising females, who were classified as exercising if they participated in more than 2 hours of purposeful physical activity over a 7-day week, 52.1% experienced abnormal cycles which included luteal phase deficiency (either short or inadequate) or anovulatory cycles (De Souza et al., 2009). The prevalence of abnormal cycles was significantly greater in the exercising group of females compared to the sedentary females (De Souza et al., 2009). The findings of this current research align with the literature on the prevalence of menstrual disorders, in particular irregular menstruation, among non-athletic females (Rafigue and Al-Sheikh, 2018; Fujiwara & Nakata, 2007).

In this current study there was no significant relationship observed between normal or irregular menstruation and LEAF-Q score. Given that menstrual function was a component of the overall LEAF-Q score it is unlikely that selecting one variable of the LEAF-Q would result in a significant relationship with the global LEAF-Q score. This inconclusive relationship is not supported by the current literature, as research suggests that individuals with LEA tend be at greater risk of menstrual disorders (Ackerman et al., 2018). Ackerman et al. (2018) found that athletes who were classified as having LEA were at increased risk of menstrual dysfunction (55.8%) compared to those with adequate EA (45.73%) (Ackerman et al., 2018). Further, research on the effects of menstrual function specifically related to EA suggests that menstrual disorders increase linearly by 9% as EA decreases (Lieberman et al., 2018). There was no significant relationship between normal and irregular menstruation and LEAF-Q score in this study. Although, as menstrual disorders are associated with deviations from the physiological norm, they are further related to health consequences such as increased cardiovascular disease, osteoporosis, depression and anovulation (Berga, 2020), indicating that 29.1% of females in this cohort may be at risk of further complications of LEA outlined on the RED-S syndrome list.

The CIA questionnaire explored individuals' eating, exercise, and body shape and weight thoughts and feelings over the previous 28 days. The CIA is designed to assess the severity of psychosocial impairment resulting from an eating disorder (Bohn et al., 2008). The underlying cause of LEA may range from unintentional undereating to severe eating disorders, which highlights the appropriateness of using the CIA in this research. A high drive for thinness is seen in those with eating disorders and was linked in other research to LEA as female athletes who had a high drive for thinness score on the eating disorder inventory have been associated with having reduced resting energy expenditure (De Souza et al., 2007).

In this current study the average global score across all three CIA components was 10.3 ± 10.4. In a previous study the CIA global scores were highest among females with clinical eating disorders (17.7 ± 10.7), followed by high-risk females (10.6 ± 8.5) and low risk controls (3.0 ± 3.3) (Vannucci et al., 2012). A more recent study in Italy among healthy, female (98%) and male (2%) controls compared to females with diagnosed eating disorders, found the CIA global score was 2.6 ± 4.2 (Calugi et al., 2018), similar to that reported in the study by Vannucci et al., (2012). Comparing the average CIA scores from Vannucci et al., (2012) and the current study would indicate that females in the current study were at high risk of an eating disorder or disordered eating behaviours. The mean global CIA scores in prior studies among healthy females in Sweden and Norway with no clinical eating disorder and in adolescents from Fiji ranged between 6 and 9 (Becker et al., 2010; Reas et al., 2010; Welch et al., 2011). The global CIA score in this current study is higher than previously reported in other studies among healthy females (Becker et al., 2010; Reas et al., 2010; Welch et al., 2011) which is concerning as it suggests that a large proportion of females in this current study are at high risk of an eating disorder or disordered eating patterns. Menstrual dysfunction, a symptom of LEA and is often associated with restricted eating patterns such as cognitive dietary restraint, which reflects the intentional effort of restricting EI in an attempt to achieve a certain body weight (Bedford et al., 2010). Females with more frequent subclinical ovulatory disturbances reported higher cognitive dietary restraint (Bedford et al., 2010). The participants in the research by Bedford et al. (2010) were healthy females (aged 19-35 years), similar to the participants in this current cohort indicating that cognitive dietary restraint may be common and more prevalent than we think among this group of females.

The average CIA score was highest for eating habits, followed by body shape and weight and lastly, exercise habits. An individual's eating habits were not further explored in this study, other than to assess their thoughts and feelings around their current eating habits. Given the overwhelming amount of conflicting information that is available to females regarding the types of foods they should or should not be eating, it is not surprising that the eating habits impairment category has the highest score. The current research suggests that LEA through reduction in EI tends to be the prominent way, given that when EI is increased to make up for the increase in energy expenditure, less severe consequences of LEA are observed (Bedford et al., 2010). It would be expected that if an individual has more negative thoughts and feelings around eating and exercise, they may be at an increased risk of LEA. In this current study individuals who were classified as at risk of LEA had significantly higher scores across all three CIA components; exercise, eating, body shape and weight. A higher score on the CIA indicates a greater level of impairment, which supports our hypothesis that those females who are at risk of LEA will have more negative feelings towards exercise and eating compared to those not at risk of LEA.

Low energy availability can be created either through suboptimal nutritional intake or by increasing EEE without upregulating EI. Therefore, we aimed to explore the duration and intensity of physical activity among healthy females. The current recommendations for physical activity in New Zealand are 150 minutes of moderate or 75 minutes of vigorous activity (or combined equivalent) across the week (Ministry of Health, 2020). This level of activity was achieved by 73.5% of participants, with 31.1% of the total participants classified as having high physical activity levels due to them reportedly exceeding these levels.

Of those classified as having high physical activity levels, 59.6% were at risk of LEA. In contrast, LEA risk among those with low levels of physical activity was only 20.8%. Despite substantial evidence for the positive health effects of meeting physical activity guidelines, without proper awareness of energy requirements, many non-athletic females may be putting themselves at risk of LEA, either intentionally or unintentionally. Further supporting these findings was the significant difference found between days of vigorous, but not moderate activity and LEAF-Q score. High intensity workouts often require a higher carbohydrate intake to support the use of glycolytic energy pathways (Casazza et al., 2018). If an individual is not aware of these increased energy requirements or purposely restricts their EI, this can lead to an inadequate energy supply to meet the energy demands for exercise and normal physiological functioning. Inadequate energy supply in such circumstance may increase the risk of LEA and an individual may present with a high LEAF-Q score. Overall, the finding that those with high physical activity habits are more likely to be at risk of LEA somewhat supports our hypothesis that non-athletic females with high and/or frequent physical activity behaviours will be classified as at risk of LEA. Therefore, proper awareness and education can be a starting point to ensure that exercising females are aware of the long-term health implications of high physical activity and low EI. Coaches and other health care professionals all need to be fully informed about these health implications to help ensure that LEA is detected early on.

Low energy availability results not only from increased energy expenditure but from a decoupling of EI to current energy expenditure. This study collected information on participants average exercise duration and intensity across the week, looking at intentional physical activity, thus did not take into account non-exercise associated thermogenesis. Non-exercise associated thermogenesis includes physical activity that is not planned such as chewing food or household chores or walking around shops, and thus the exclusion of these activities can lead to an underestimation of an individual's overall energy expenditure (Burke et al., 2000). There is the possibility that individuals at risk of LEA may have done more incidental physical activity and were not increasing EI to match expenditure. Given that the CIA was highest for the eating habits component, it could be suggested that the decoupling of EI and energy expenditure in the current cohort may come from inadequate nutritional knowledge to compensate for their physical activity levels. It would be expected that eating impairment based on CIA has been ongoing for at least the past 28 days, given the CIA questionnaire is based on individuals' thoughts and feelings over the past 28 days. A more detailed assessment of physical activity may further help to provide further insight into these associations.

Non-athletic females in this current study presented with poor perceptions towards eating and body weight which may place them at risk of LEA. The findings from this study lead to the conclusion that individuals with more negative thoughts and feelings around their eating, exercise and body shape have significantly higher physical activity levels. Subsequently, non-athletic females may present with symptoms associated with chronic LEA. It is important to raise awareness around the implications of LEA and the related health consequences. Understanding the associated risk factors of LEA which this study has outlined, such as body dissatisfaction, disordered eating behaviours and exercise behaviours, may be helpful in recognising if a female is at risk of LEA prior to presenting with physiological symptoms and enabling corrective action to be taken early.

Chapter 4: Research conclusions

4.1 Conclusion

Understanding the implications of exercise and eating behaviours to reduce the risk of LEA among all females is crucial to ensuring their long-term health. The current study has demonstrated that LEA is not restricted to elite athletes as has traditionally been thought but is also prevalent among general female populations. The research has demonstrated that individuals participating in recreational sports might present with a greater eating disorder risk than athletic females (Levitt, 2008). It is possible that individuals who are involved in organised sports such as those with multiple team trainings across the week often have the support of coaches, health care professionals, physicians, dietitians and physiologists who can provide performance, health and nutritional information compared to more recreational sports or activities. Those involved in recreational sports may have less support and may be more likely to misinterpret information and apply suboptimal nutritional strategies. On the other hand, if trainers are not educated on the consequences of over-training and under-eating, specifically in females, the consequences could be just as detrimental. It is crucial that exercising females have an understanding of the female menstrual cycle for optimal performance and health benefits to ensure that this is used to guide tracking of overall training status and overall health. There appears to be a need for a massive shift in the mindset that 'thinner is better' in terms of performance and overall health. Optimal body size for performance is not about eating less and training more, it is about fuelling for the work required. In combination, with the support from either coaches or from other supports such as family then performance, physical and psychological benefits from physical activity can be achieved. Through possible screening tools, compulsive exercise behaviours can be identified early on, this may also raise awareness of the psychological factors underpinning LEA such as poor mental health (Logue et al., 2020).

Many different factors can cause irregular periods, including other underlying menstruation disorders such as polycystic ovary syndrome, luteal phase deficiencies, hypothalamic anovulation, endometriosis and premenstrual syndrome (Brown, 2011). However, in the absence of medical disorders that may alter menstrual patterns, irregular menstrual cycles may be a result of over-exercising accompanied with undereating, stress, and birth control. It is therefore important to identify the primary cause of the menstrual dysfunction, especially if the individual is looking for effective treatment (Godari et al., 2020). We can determine from this study that a proportion of females reported to have normal menstrual cycles although there were 29.1% who had irregular menstruation, which could be due to a range of underlying issues.

When considering menstrual function, most studies do not account for the various disordered menstrual patterns, and may only consider eumenorrheic or amenorrhoeic states (Brown et al., 2017). Therefore, the impact of a chronic state of LEA on changes to reproductive hormones and alterations to menstrual cycles (anovulation, luteal deficiency or short luteal phase) are still largely unknown. This current research does not take into consideration other possible menstrual hormonal patterns which is important to acknowledge, given that luteal phase defects are highly prevalent among active females (De Souza & Nulsen, 1992). Luteal phase defects account for 57% of total menstrual disorders among females exercising for less than one hour a week (Lieberman et al., 2018). A reduction in luteinising hormone pulse frequency can be seen when EA is purposely reduced to <30 $kcal \cdot kgFFM^{-1} \cdot day^{-1}$ leading to an increased likelihood of developing menstrual disturbances such as luteal phase defects (Koltun et al., 2019). Given that 29.1% of participants in this study experience irregular menstruation, they may also have luteal phase defects or anovulation yet be unaware of this.

Amenorrhea is the clinical end-point of menstruation which may be due to a wide variety of underlying causes. Primary amenorrhea is usually diagnosed when an individual has no onset of menses by age 15 years and is typically related to a dysfunction of processes controlled by the hypothalamus (Brown, 2011). Alternatively, secondary amenorrhea is when there is an interruption to menstruation for ≥3 months (Brown, 2011). Functional hypothalamic amenorrhea is responsible for 20-35% of secondary amenorrhea and is primarily related to weight loss, stress and exercise (Meczekalski et al., 2014). The differences in aetiology between primary and secondary amenorrhea is important to recognise if treatment of amenorrhea is sought. In this present study, 12.6% of females stated that they had not had a period for at least the previous three months, and 28.5% of females reported that their period had stopped for three months at some stage in the past. The proportion of participants that had experienced amenorrhea in this current study is higher in comparison to research by Black et al., (2018) in which a lower percentage of recreationally active females reported previously having amenorrhea (18.4%) and a higher percentage currently with amenorrhea (21.1%) (Black et al., 2018).

For most females exercise has a positive impact on health, however if EEE increases more than EI a variety of clinical manifestations can occur resulting in LEA (Wasserfurth, 2020). This may occur when females unintentionally enter LEA during periods of intense training or during a structured diet phase. Previous studies have shown secondary amenorrhea, absence of menstruation ≥ 3 months to be prevalent in 3-5% of females (Meczekalski et al., 2014) and a higher prevalence varying from 4.8% to 35% among athletic females, with differences in prevalence often dependent on the type of sport (Hoch et al., 2011; Melin et al., 2014; Barrack et al., 2014; Dadgostar et al., 2009). Prior studies in recreationally active females have found the prevalence of secondary amenorrhea to be 13.6% among recreational dancers (Witkos & Wrobel, 2019) and 18.4% in recreationally active females (Black et al., 2018) which is similar to this current research. These findings highlight the need for further investigation into recreationally active females given that weight loss and exercise are the main causes of functional hypothalamic amenorrhea.

Negative thoughts and feelings around eating, exercise and body shape and weight, may drive individuals to carry out problematic behaviours and engage in more disordered eating behaviours (Dahlgren et al., 2017). Concerns can be raised for individuals who have a high drive to be thin and who continue to restrict their EI as initial weight loss begins to plateau (Trexler et al., 2014; Wasserfurth et al., 2020), increasing their risk of LEA through insufficient EI. When an individual is in a state of chronic LEA metabolic consequences such as a decline in basal metabolic rate may result in an unfavourable body composition. Negative perceptions around weight and body image that were prevalent among this cohort and may cause females to adopt more aggressive weight loss methods such as further decreasing their EI, creating a very negative spiral (Wasserfurth et al., 2020; Areta et al., 2021).

High physical activity behaviours, exceeding the current physical activity recommendations may also be a contributing factor to LEA given that in this study 33.1% had high physical activity behaviours and of those with high physical behaviours, over half (59.6%) were at risk of LEA. It is suggested that participation in regular physical activity may also play a casual role in the development of excessive weight and diet concerns and that excessive physical activity may be a precursor for eating disorders (Davis et al., 1990). This warrants further investigation, given that research has shown individuals who have frequent dieting behaviours have a greater emotional attachment to and pre-occupation with exercise (Ackard et al., 2002). Among self-identified regular exercising females those who were classified as being chronic dieters engaged in much more frequent and intense physical activity than non-dieters (Davis et al., 1990).

The overall theme evident from this present study was the significant multi-directional relationship that was observed between eating and exercise behaviours and the risk of LEA among non-athletic females within New Zealand. Non-athletic females in this current study presented with poor perceptions towards eating and body weight which placed them at risk of LEA. Subsequently, non-athletic females may present with symptoms associated with chronic LEA. Non-athletic females in this study who presented with high physical activity behaviours, above the current physical activity recommendations in New Zealand, were at greater risk of LEA than those not meeting the physical activity guidelines. This research accentuates the multi-directional relationship between physical activity, eating behaviours and LEA risk among non-athletic females and the need for future awareness and education.

4.2 Strengths and limitations

This study provides valuable insights into the impact of LEA on females and some of the associated risk factors. In order for statistical significance to be observed a sample size of 265 participants was needed. We were unable to reach this number due to time constraints of data collection and the inclusion of only participants who had fully completed the questionnaire. Although slightly lower than desired participant numbers mean that the study may not have been sufficiently powered to detect some significant differences, a sample number of 151 is still considered a large population sample for this given research field.

All information was collected through an online questionnaire in which participants self-reported their weight and height. This self-report may have introduced inaccuracies, as females tend to overestimate their height and underestimate their weight (Engstrom, 2003). There are also many inaccuracies with using BMI as an indicator of health, as BMI does not take into consideration age, sex, bone structure, fat or muscle distribution (Rothman, 2008).

Individuals' physical activity levels were estimated based on achieving a threshold of physical activity on any number of days, thus physical activity may be under-estimated among some participants in the study. Due to the self-reporting nature of the survey, there may be inaccuracies in the amount of physical activity as seen in the Dystad et al. (2014). Females and more educated participants in general are often more thorough in their estimation of their physical activity level compared to men and less educated people (Dyrstad et al., 2014). There is still likely to be some over-estimation of self-reported physical activity from social desirability response bias (Dyrstad et al., 2014). There are also likely to be individual differences among participants' definitions of moderate and vigorous activity.

The strength of this current study is the use of validated screening tools such as the CIA, LEAF-Q and NZPAQ- short form to assess eating, exercise, body shape and weight, thoughts and feelings, risk of LEA and physical activity behaviours. The entire questionnaire was conducted online which allowed participants to complete it in their own time. It also allowed for participants' answers to be anonymous and a larger group of participants to be reached. The CIA focuses on an individual's thoughts and feelings over the past 28 days which provided an understanding of overall long-term trends in behaviour which are more likely to have an impact on the individual and their risk of LEA. The questionnaires used in this cross-sectional study are based largely on self-reports. Therefore, information on exercise duration, classification as a non-athletes and heaviness of menstrual bleeding are based on the individual's perspectives of themselves. These questions are cross checked by our analysis i.e. frequency and duration of exercise to ensure there are no individuals with very high physical activity levels that may indicate the individual is an athlete.

4.3 Future recommendations

The LEAF-Q can be used to aid in identifying individuals who may be at risk of LEA, however it is important that the LEAF-Q is used in conjunction with other measures of EA. The use of EA calculations through calculating EI and EEE can further support health care professionals in determining an individual's risk of LEA. It may also be valuable to gain more information regarding individuals' physical activity habits to determine other incidental activity across the day to get a more accurate representation of their overall energy expenditure. Based on the findings from the CIA, it would indicate a portion of recreationally active females are dissatisfied with their body and that these negative thoughts and feelings may currently be driving their exercise behaviours and place them at risk of LEA. Education around the consequences of excessive exercise and inadequate EI, and the benefits of exercise other than as a way to control weight, will help early identification of an individual's risk of LEA to prevent negative health implications occurring.

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Appendices

Appendix A. Pre-participation online questionnaire

Questionnaire for BELieF in Females study

Demographics:

1) How old are you?
Years:

2) What is your height:
cm:

3) What is your current weight:
kg:

4) Do you smoke:	
Yes	No

5) Are you currently taking any forms of medication other than an oral contraceptive pill?
Please specify:

6) Are you currently enrolled in Tertiary Education	
Yes	No

If No

6a) Have you previously been enrolled in Tertiary education?	
Yes	No

7) Which Ethnic group do you belong to? Please tick which ever applies to you (you may tick more than one box)
• New Zealand European • Maori • Samoan • Cook Island Maori • Tongan • Niuean • Chinese • Indian • Other: Please specify

Short form Physical Activity - Full Questionnaire
LEAF-Q menstrual questionnaire-Questions 2 (Gastrointestinal issues through to menstrual issues will be completed as part of this survey)
Eating Disorder Questionnaire
Clinical Impairment Questionnaire

Appendix B: Inclusion Criteria

MASSEY UNIVERSITY
COLLEGE OF HEALTH
TE KURA HAUORA TANGATA

BELieF in Females study

Please complete the following questions prior to signing the consent form

Screening Questionnaire		
Are you proficient in English?	Yes	No
Are you pregnant or breastfeeding?	Yes	No
Have you suffered from an eating disorder	Yes	No

Appendix C: Information sheet

BELieF in Females

Information Sheet

Researchers Introduction

We would like to invite you to participate in this study investigating low energy availability in females. The purpose of this study is to investigate behavioural risk factors associated with low energy availability in females living in Auckland New Zealand. This study is being conducted by a group of researchers at Massey University, Auckland New Zealand.

Please read the Information Sheet carefully before deciding whether or not to participate

The details of the lead researcher for this study are:

Dr Claire Badenhorst
Lecturer
School of Sport, Exercise and Nutrition
College of Health
Massey University
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Project Description and invitation

Energy availability is difference between the energy we get from food and the energy we expend during exercise. This remaining energy is utilised by the body to maintain normal physiological functioning. Lack of energy availability in female athletes' results in a number of negative health consequences including declines in bone health and irregular menstrual cycles. While behaviours and risk factors associated with low energy availability have been investigated in an athletic setting, the implications of similar behaviours in recreationally active or sedentary females is currently unknown. Therefore the aim of the study is to investigate behaviour risk factors for low energy availability in females aged 18-30 years.

Participant Identification and Recruitment

Who are we looking for?

We are looking for 265 females to participate in this study. To take part in the study you should:

- Be between 18-30 years of age
- Be proficient in English
- Not be pregnant or currently breastfeeding.
- Have no known eating disorders currently or in the last 12 months

Project procedures

What is going to happen?

If you decide to take part in this study after you have read and had time to consider the information contained in this information sheet, you will be asked to complete a short screening questionnaire to ensure that you meet the study criteria. If you meet the inclusion criteria you will be invited to take part in the study. If you are willing to participate, you will be invited to complete a series of online questionnaires. The information that will be collected will include: age, body composition (height and weight), demographics, physical activity level, eating behaviours and menstrual cycle function.

Data Management

The data will be used only for the purposes of this project and no individual will be identified. Only the investigators and administrators of the study will have access to personal information and this will be kept secure and strictly confidential. Participants will be identified only by a study identification number. Results of this project may be published or presented at conferences or seminars and no individual will be able to be identified. At the end of this study the list of participants and their study identification number will be disposed of. Any raw data on which the results of the project depend will be retained in secure storage for 10 years, after which it will be destroyed.

A summary of the project findings will be available to all study participants. All participants will be sent this information via email or a personal letter.

Participants Rights:

You are under no obligation to accept this invitation. Should you choose to participate, you have the right to:

- Decline to answer any particular question
- Withdraw from the study at any time, even after signing a consent form (if you choose to withdraw you cannot withdraw your data from the analysis after the data collection has been completed)
- Ask any questions about the study at any time during participation
- Provide information on the understanding that your name will not be used unless you give permission to the researcher
- Be given access to a summary of the project findings when it is concluded

What are the benefits and risks of taking part in this study?

There will be no cost to you for any of the procedures or assessments taken as part of this study.

The principle benefit of taking part in this study is that you will contribute to a novel area of research and assist us in increasing our understanding of factors that influence female health in New Zealand

There are no personal risks to your health as a result of completing this research.

Project Contacts:

If you have any questions regarding this study, please do not hesitate to contact either of the following people for assistance:

Principal Researcher:

Dr Claire Badenhorst (School of Sport, Exercise and Nutrition)

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Associated Researchers:

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Ethics Committee Approval Statement

Thank you for considering participation in this study