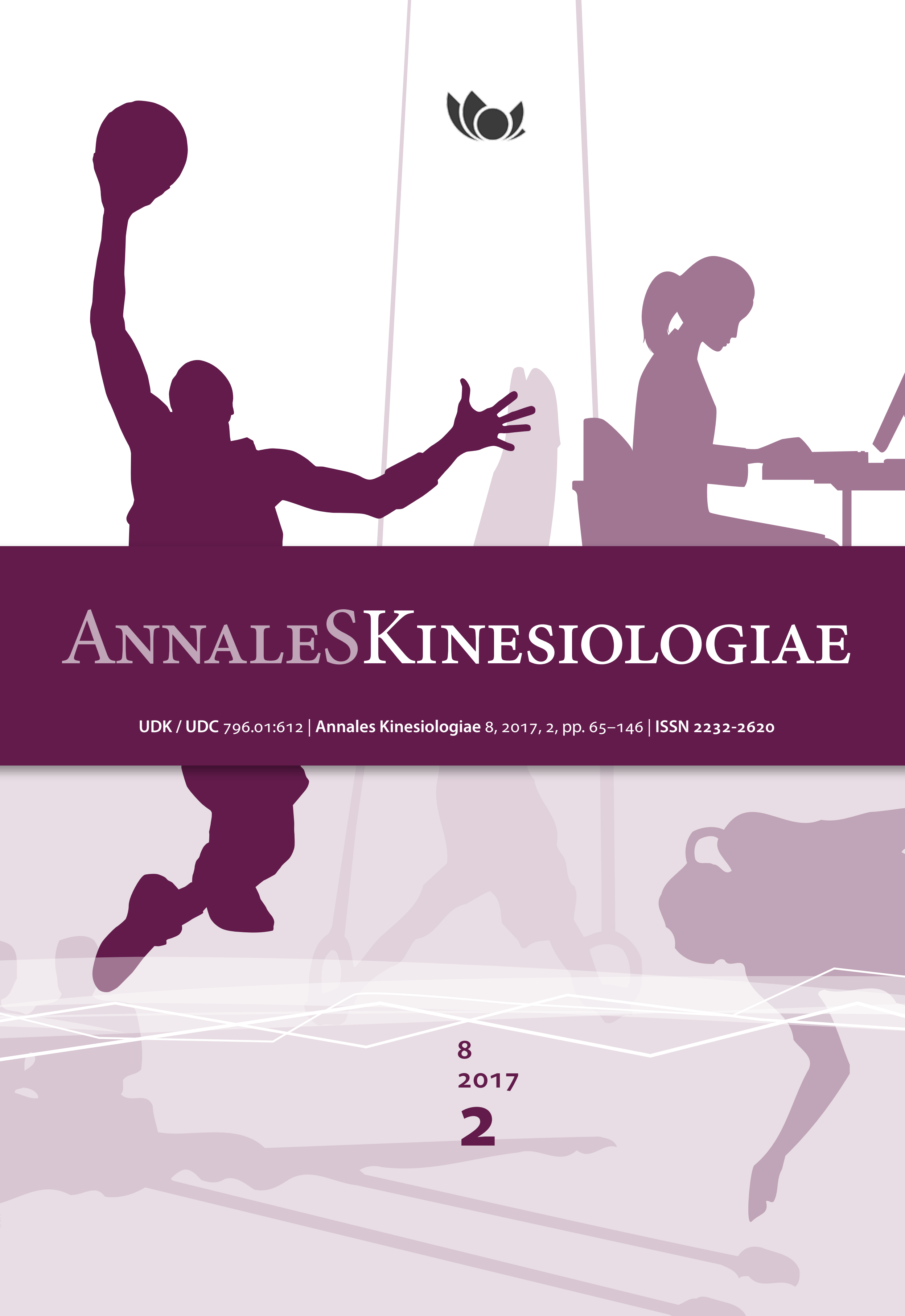

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EDITORIAL

Dear reader of the second issue of the scientific journal Annales Kinesiologiae Volume 8. We are pleased to present five new articles that address the broad interdisciplinary field of the anthropological branch of kinesiology, as well as the importance of human movement, as the main study subject of this scientific discipline**;** all of them throughout different life periods and in the light of promoting healthy development and active lifestyle in both childhood and third age period.

The introductory article focuses on the study of the female athlete triad and the associated negative energy balance, ovulation disorder, and osteopenia or even osteoporosis. Doyle-Baker, Mclean and Fung thus present the positive and negative effects of the menstrual cycle on sports performance and, through an empirical study, they determine the presence of the female athlete triad among elite female ice hockey players. This phenomenon is probable among athletes who are subject to high energy demands, although the authors find it difficult to confirm. Let's mention at this point that the abstract of this study was presented at the 8th International Scientific Conference on Kinesiology in Opatija, Croatia, and now we are glad to publish it as a full text in the present issue of the Annales Kinesiologiae journal.

Further on, there are three review articles from the field of physical activity and exercise in the third age that have been prepared under the framework of the PANGeA project – Physical Activity and Nutrition for Quality Aging funded by the Interreg cross-border cooperation program Slovenia – Italy 2007–2013. They are rounded up so that Capatti, Dalla Nora and Passaro first present the problems with age-related changes in body composition and metabolism, including sarcopenia, sarcopenic obesity and bone loss with a progressive decline in aerobic capacity, muscle mass and strength as the main risk factors for reducing the mobility of older people, especially when combined with associated chronic diseases. Then, through extensive literature review, they summarize some recommendations for physical activity, exercise and nutrition, which enable older adults to maintain the heart and lungs function, to increase cardiovascular fitness and endurance, as well as preserving muscle mass and strength, bone density and reducing the increment of fat. It turns out that in older adults, especially with a combination of strength and endurance training, and appropriate protein intake, we can act both preventively and curatively against sarcopenia, sarcopenic obesity, and metabolic syndrome. This is followed by a review by Rejc, Del Torto and Lazzer, which present exhaustively the effects of aging on maximal aerobic power, the effects of aerobic exercise in older adults. At the end the authors summarize current recommendations for aerobic exercise during the third age. In doing so, authors are also critical of the valid recommendations, as they draw attention to their too generic nature and give suggestions for improvements. The third part of the review papers then deals with the aspect of the impact of exercise on activation and the increase in the number (proliferation) of the skeletal muscles stem (satellite) cells, which facilitate the recovery of the "damaged" muscle tissue under various psycho-physiological loads and stimuli. Jurdana thus updates the view on the role of muscular satellite cells in regulating muscle mass in conjunction with the effectiveness of various training interventions in order to reduce the decline in muscle mass that may be present in all stages of life, and especially in old age. She notes that it is possible to stimulate the activation and proliferation of skeletal muscle satellite cells, through an appropriate diet, resistance and endurance exercise, thus improving skeletal muscle function and successfully combating against muscular atrophy and age-related sarcopenia. She emphasizes that more studies will be needed for further understanding of the role and impact of training variables on the activation of satellite cells in order to set optimal training stimuli for this purpose.

The report from the opening of the Mediterranean Health Centre (MHC) of the Science and Research Centre Koper rounds this issue of the journal. The MHC represents an important link between kinesiological science and practice as well as the socio-cultural environment and geographical location in which it was established, since the healthy Mediterranean lifestyle (in particular its typical diet, simplicity of living, social inclusion and interactions of people in this area) is thus enhanced by a healthy active lifestyle, exercise and training, kinesiological diagnostics and therapies based on modern scientific knowledge. All this has now become accessible to all of us, irrespective of age, gender, health status or other differences that can occur among people. This is also one of the most effective ways of implementing the mission to man-oriented kinesiology, which has an increasingly important role and responsibility in modern society in ensuring a healthy and balanced development of an individual and the society from childhood to the late adulthood.

Mitja Gerževič, PhD
Guest Editor

UVODNIK

Spoštovani bralec druge številke osmega letnika znanstvene revije Annales Kinesiologiae. Z veseljem vam predstavljamo novih pet prispevkov, ki široko interdisciplinarno področje antropološke veje kineziologije in pomen gibanja človeka, kot glavnega predmeta preučevanja te znanstvene discipline, obravnavajo preko različnih življenjskih obdobij v luči spodbujanja zdravega razvoja in aktivnega življenjskega sloga, tako v otroštvu kot v tretjem življenjskem obdobju.

Uvodni prispevek je nekoliko ožje usmerjen, in sicer v proučevanje ženske športne triade ter s tem povezanim pomanjkanjem energije oz. negativno energijsko bilanco, motnjo ovulacije in osteopenijo ali celo osteoporozo. Doyle-Baker, Mclean in Fung tako predstavljajo pozitivne in negativne vplive menstrualnega cikla na športno uspešnost ter preko empirične študije ugotavljajo pojavnost ženske športne triade med vrhunskimi igralkami hokeja na ledu. Pojav je med športnicami, ki so podvržene visokim energijskim zahtevam sicer možen, vendar avtorji ugotavljajo, da ga je težko potrditi. Naj omenimo, da je bil izvleček te študije predstavljen na 8. Mednarodni znanstveni konferenci o kineziologiji v Opatiji na Hrvaškem in ga sedaj z veseljem v celoti objavljamo v pričujoči številki revije Annales Kinesiologiae.

Sledijo trije pregledni članki s področja telesne oz. gibalne/športne aktivnosti in vadbe v tretjem življenjskem obdobju, ki so bili pripravljeni v okviru projekta PANGeA – Telesna aktivnost in prehrana za kakovostno staranje. Projekt je potekal v okviru programa čezmejnega sodelovanja Interreg Slovenija–Italija 2007–2013. Članki so zaokroženi tako, da Capatti, Dalla Nora in Passaro najprej predstavijo problematiko s staranjem povezanih sprememb v telesni sestavi in metabolizmu, vključujoč sarkopenijo, sarkopenično debelost in upad kostne mase ter trend postopnega upada aerobnih funkcij, mišične mase in moči kot glavnih dejavnikov tveganja za zmanjšanje mobilnosti starejših oseb, še posebej v povezavi s pridruženimi kroničnimi obolenji. Nato preko obširnega pregleda literature podajo priporočila za telesno aktivnost, vadbo in prehrano, ki starejšim osebam omogočajo ohranjanje ustreznega delovanja srčne in dihalne funkcije, izboljšanje srčno-žilne pripravljenosti in vzdržljivosti, kakor tudi ohranjanje mišične mase in moči, kostne gostote ter zmanjšanje pridobivanja maščobne mase. Izkaže se, da lahko pri starejših osebah, predvsem s kombinacijo vadbe za moč in vzdržljivost ter ustreznim vnosom beljakovin, delujemo tako preventivno kot kurativno proti sarkopeniji, sarkopenični debelosti in metabolnemu sindromu. Temu sledi pregled Rejca, Del Torta in Lazzerja, ki izčrpno predstavi vplive staranja na največjo aerobno moč, učinke aerobne vadbe pri starejših osebah in povzame trenutno veljavna priporočila za aerobno vadbo za to življenjsko obdobje. Pri tem se avtorji kritično opredelijo do veljavnih priporočil, saj opozorijo na njihovo preveliko generičnost in podajo predloge za izboljšanje. Tretji izmed preglednih prispevkov obravnava vidik vpliva vadbe na aktivacijo in povečanje števila (proliferacijo) matičnih (satelitskih) celic skeletnih mišic, ki olajšajo obnovo »poškodovanega« mišičnega tkiva po različnih psiho-fizičnih obremenitvah. Jurdana tako posodobi pogled na vlogo mišičnih satelitskih celic pri uravnavanju mišične mase v povezavi z učinkovitostjo različnih vadbenih intervencij za zmanjšanje upada mišične mase, ki je lahko prisotno v vseh življenjskih obdobjih, predvsem pa v starosti. Ugotavlja, da je tudi v starosti, preko ustrezne prehrane in vadbe proti uporu (za moč in silovitost) ter vadbe za vzdržljivost, možno spodbuditi aktivacijo in proliferacijo satelitskih celic skeletnih mišic, s tem izboljšati mišično funkcijo ter se tako uspešno boriti proti mišični atrofiji in starostno pogojeni sarkopeniji. Pri tem pa poudarja, da bo potrebnih več študij, ki bodo bolje osvetlile in pojasnile vlogo in vpliv vadbenih spremenljivk na aktivacijo satelitskih celic ter podale za ta namen najoptimalnejše vadbene dražljaje.

S poročilom z otvoritve Mediteranskega centra zdravja (MCZ) Znanstveno-raziskovalnega središča Koper pa zaokrožujemo celoto tokratnega izvoda revije. MCZ predstavlja namreč pomembno vez med kineziološko znanostjo in prakso ter družbeno-kulturnim okoljem in geografsko lego, v kateri deluje, saj zdrav sredozemski življenjski slog (predvsem zanj značilna prehrana, enostavnost bivanja ter socialna vključenost in interakcija ljudi na tem območju) nadgrajuje z zdravim aktivnim življenjskim slogom, vadbo in treningom ter kineziološko diagnostiko in terapijami, ki temeljijo na sodobnih znanstvenih dognanjih. Vse našteto je s tem centrom postalo dosegljivo prav vsem, ne glede na starost, spol, zdravstveno stanje ali druge razlike, ki se med ljudmi lahko pojavljajo. Prav to je eden od najučinkovitejših načinov udejanjanja poslanstva k človeku usmerjene kineziologije, ki ima v sodobni družbi vse pomembnejšo vlogo in odgovornost pri zagotavljanju zdravega in uravnoteženega razvoja posameznika in družbe od otroštva do starosti.

dr. Mitja Gerževič,
gostujoči urednik

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FEMALE ATHLETE TRIAD − PROBABLE BUT DIFFICULT TO CONFIRM IN FEMALE ICE HOCKEY PLAYERS

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ABSTRACT

Female hockey players have high energy expenditure and may enter a negative energy balance (EB) without noticeable body composition changes. Menstrual cycle (MC) and luteal phase (LP) length, EB, and bone mineral density (BMD) were tracked over nine months (mean, SD (±) in 12 ice hockey players (HP; age 21.1 ± 3.4 yrs; height (HT) 165.9 ± 4.6 cm; weight (WT) 64.7 ± 8.1 kg; body fat percent (BF %) 2.8 ± 3.8 %) and 12 non-athlete controls (C; age 21.4 ± 2.8 yrs; HT 169.5 ± 5.5 cm; WT 65.4 ± 5.4 kg; BF % 20.0 ± 3.1 %). HP MC (35.8 ± 11.2 days) was longer than C (29.8 ± 4.3 days) and HP LP (10.1 ± 2.1 days) was also longer than C (9.6 ± 2.8 days). Anovulation occurred in 50.0 % of HP versus 39.2 % of C. No group BMD differences were observed in lumbar spine ($p = 0.9$), hip ($p = 0.5$), and radial ($p = 0.7$) sites. A negative EB was identified (HP = -1026.52 ± 450.1; C = -780.00 ± 310.19 kcal / day), yet no significant within-group differences in WT (HP $p = 0.7$; C $p = 0.8$), BF % (HP $p = 0.97$; C $p = 0.6$), or fat free mass (HP $p = 0.6$; C $p = 0.98$) were found over the study duration. Rigorous hockey schedule likely contributed to 28 % completion of the Basal Body Temperature and MC recordings in HP compared to 70 % in C. Both groups entered a state of negative EB, but did not exhibit a BF % change associated with the Female Athlete Triad.

Keywords: *menstrual cycle, female ice hockey players, bone mineral density.*

ŽENSKA ŠPORTNA TRIADA – VERJETNA, A JO JE PRI HOKEJISTKAH TEŽKO DOKAZATI

IZVLEČEK

Za hokejistke je značilna visoka poraba energije, ki lahko vodi do negativnega energijskega ravnovesja (EB) brez opaznih sprememb v telesni sestavi. V obdobju 9 mesecev smo spremljali menstrualni ciklus (MC) in dolžino lutealne faze (LP), EB in mineralno gostoto kosti (BMD) pri 12 hokejistkah (HP; starost 21,1 ± 3,4 let; telesna višina (HT) 165,9 ± 4,6 cm; telesna masa (WT) 64,7± 8,1 kg; %telesnih maščob (BF %) 22,8 ± 3,8 %) in pri 12 kontrolnih ne-športnikih (C; starost 21,4 ± 2,8 let; HT 169,5 ± 5,5 cm; WT 65,4 ± 5,4 kg, BF % 20,0 ± 3,1 %). Pri skupini HP je bil zabeležen daljši MC (35,8 ± 11,2 dni) kot pri C (29,8 ± 4,3 dni) in pri HP je bil LP prav tako daljši (10,1 ± 2,1 dni) kot pri C (9,6 ± 2,8 dni). Pri 50,0 % HP je prišlo do anovulacije, medtem ko pri C le pri 39,2 %. Nismo ugotovili razlik v BMD na lumbarnih vretencih (p=0,9), medenici (p=0,5) in koželjnici (p=0,5). V času izvajanja študije smo zaznali negativno EB (HP =-1026,52 ± 450,1 kcal / dan; C = -780,00 ± 310,19 kcal / dan), ne pa tudi pomembnih razlik znotraj skupine pri WT (HP p = 0,7; C p = 0,8), BF % (HP p = 0,97; C p = 0,6), ali pusti masi (HP p = 0,6; C p = 0,98). Strog urnik hokejistk je najbrž razlog za zgolj 28 % popolnost njihovih zapisov bazalne temperature in MC, za razliko od 70 % poročanja pri C. Obe skupini sta dosegli negativno EB, ni pa bila vidna sprememba BF % v povezavi z žensko športno triado.

***Ključne besede**: menstruacijski ciklus, hokejistke, mineralna gostota kosti*

INTRODUCTION

There has been a dramatic increase in female athletic participation and intense physical activity (PA) in the past 30 years (Chen & Bryzyski, 1999; Highet, 1989; Nattiv, Agostini, Drinkwater, & Yeager, 1994; Prior, Vigna, & McKay, 1992). A growing concern among health care clinicians and practitioners is the potential detrimental effect of long term training on bone mineral density (BMD), which may be preceded by low energy availability (EA) (Olympic Charter, 2011). EA is defined as energy obtained through oral nutrition minus energy expended during exercise. It is reasonable to conclude that low energy may result because of increased energy expenditure (EE), decreased oral intake (either intentional or unintentional), or both (Kishner, 2016). Subsequently, musculoskeletal and reproductive dysfunction from this prolonged energy deficit may occur, inducing the amenorrhoeic condition (International Olympic Committee, 2005; Mountjoy et al., 2014; Nattiv et al., 1994; Otis, Drinkwater, Johnson, & Wilmore, 1997).

BMD decreases with the number of missed menstrual cycles (MC) accumulated over months and years. More specifically, a regular MC with either anovulation (absence of ovulation) or short luteal phase length (SLP) may pose also a risk for bone remodeling, imbalance and bone loss (Seifert-Klauss & Prior 2010). The Female Athlete Triad (Triad) and/or its individual components (decreased EA, menstrual dysfunction, and low BMD) have previously been identified in athletic women (Egan, Reilly, Whyte, Giacomoni, & Cable, 2003; Koehler, Achtzehn, Braun, Mester, & Schaenzer, 2013; Reed, De Souza, & Williams, 2013). All female athletes in any sport regardless of the competition level are at potential risk to develop the Triad. However, those females participating in endurance sports, such as track and field, swimming, and rowing, or in those events requiring subjective judging, such as gymnastics and figure skating, are most at risk (Martinsen & Sundgot-Borgen, 2013). The current risk profile of the Triad includes very little information on female athletes having 'average body weights and lean body mass' (LBM), who participate in predominantly team oriented, weight-bearing sports (e.g. ice hockey, field hockey, soccer, basketball, and volleyball). Female athletes in weight bearing sports may also be energy deficit without knowing it and have ovulatory disturbances leading to future bone loss (osteoporosis) or bone weakening (osteopenia).

The sport of ice hockey requires a long-term commitment to skill development and physical fitness which may impact a player's EE levels. It is possible that over time players may enter a state of negative EB without noticeable body weight change. The prevalence of inadvertent low EA is unknown in female ice hockey players. Therefore, the primary purpose of this study was to describe menstrual disturbances using basal body temperature (BBT) analysis to determine luteal phase (LP) length, and occurrence of anovulatory cycles in a group of female ice hockey players (HP) (17-25 years of age) participating in the Olympic Oval High Performance Training Program (HPTP). Our control (C) group were non-athlete students recruited from the University of Calgary Community. We hypothesized that HP would: (Ho_1) exhibit longer MC, shortened LP (<10 days), with a greater number of anovulatory cycles as indicated by the MC Diary and BBT measurements when compared with C; (Ho_2) be in a state of negative EB in the absence of any change in body composition; and (Ho_3) have greater baseline BMD values at the spine, hip, and radial site compared to C and the population reference standards.

METHODS

Our study took place over 9-months and encompassed the fall and winter university semesters (October to June). Recruitment occurred at the start of the fall semester (September 1999) and the data collection period had a staggered study start; October for HP and November for C. The staggered start was necessary because of scheduling issues and conflicts with booking the Dual Energy X-ray Absorptiometry (DXA) machine.

Volunteers were invited to participate:1) if HP had a five-year history of hockey specific training and if they committed to four or more 75 minute training sessions per week at the Olympic Oval in Calgary, Alberta, 2) following completion of the

Baecke Questionnaire of Habitual Physical Activity (Baecke, Burema & Frijters, 1982), used to evaluate their physical activity level, and 3) following completion of the Eating Disorder Inventory (EDI-2) questionnaire and screening tool (EDI-SC) to identify predisposition towards disordered eating (Garner, 1991).

Volunteers were excluded if they: (1) had used oral contraceptives during the preceding six months, (2) were smokers, (3) showed predispositions to disordered eating tendencies based on the EDI-2 and EDI-SC results, or (4) were involved in shift work (e.g. night shifts) as this would interfere with the accuracy of BBT methods (Prior, Vigna, Schulzer, Hall, & Bonen, 1990). Of the 38 volunteers, 4 HP and 5 C were excluded due to their use of oral contraceptives, and one HP scored positively for disordered eating tendencies and was referred to counseling.

Data Collection Procedures and Measurement

Participants attended a 2-hour information / education session during their study start week where demographic information, menstrual history and current menstrual status data were collected. A registered dietician provided education on how to correctly complete the 7-day dietary record including accurate recording of dietary intake, serving sizes, calories per serving, and how to read food labels and recall techniques. The participants were given similar detailed information on how to correctly complete the 7-day Activity Record, MC diary, and BBT measurements. During the study, multiple reminders (e.g. follow-up telephone contacts and emails) and assurances of confidentiality were incorporated into the data collection methods to increase response rate and decrease non-sampling errors (Ransdell, 1996).

Menstrual Cycle Length

MC was tracked on a calendar and participants were asked to identify the first day with a phone call to the research coordinator (RC). MC start was defined as the first day of menstrual flow and the final day was defined as the day before the onset of the next menstrual flow. MC length was calculated as the difference between the day before the onset of menstrual flow and the first day of the previous cycle and was calculated as the mean length of each recorded cycle from month 1 through to month 6 of the data collection period.

Basal Body Temperature

Participants were instructed to measure their oral BBT immediately upon waking and before standing using a low-reading digital thermometer read to the nearest 0.05°C. These measurements were recorded in the MC diary and in addition participants recorded their subjective observations about late rising, illness, amount of menstrual flow, emotions, and disturbed sleep. Participants also commented on subjective markers of ovulation such as mucous secretion and breast tenderness (Prior et al., 1990; Prior, 1996).

Luteal Phase Length

We chose the mean temperature method (MTM) to predict the onset of LP using pre-established criteria from Vollman (1977). According to Prior et al., (1990) the LP length determined by MTM is comparable to directly measured serum mid-cycle luteinizing hormone (LH) on the peak day ($r = 0.891$).

The temperatures of a given MC were averaged and a corresponding mean line drawn across the graph of the data. The start of the LP was defined as the first temperature to rise above the mean line and remain above for three consecutive days. The end of the LP was defined as the day before the onset of the next menstrual flow (Prior et al., 1990). Temperatures of more than 0.5°C above the mean were discarded as febrile values (Vollman, 1977). SLP was defined as those cycles having LP length < 10 days within a cycle of normal length (Prior et al., 1990).

Anovulation can be determined by a lack of thermal shift in BBT (Vollman, 1977). We designated anovulatory MCs as those cycles having irregular temperature patterns and the absence of a definite LP when a normal MC length of 21 to 36 days was maintained (Personal Communication, J. C. Prior, October 1999). The number of anovulatory cycles was expressed as a percentage of the total number of eligible cycles recorded for the study period.

Total Energy Expenditure (TEE)

EE includes the components of resting energy expenditure (REE), the thermic effect of food (TEF) and the energy expended through PA (EPA). REE is the largest single source of EE and accounts for approximately 50 to 75 % of an individual's TEE (Mahan & Escott-Stump, 1996; Van Zant, 1992). TEE is defined as the energy expended due to resting physiological functions (e.g. ventilation, cardiovascular activity, protein, glycogen, and triglyceride synthesis, and electrical activity within the cells) (Thompson & Manore, 1996).

We employed the Cunningham equation (1980) because of the ability to include fat free mass (FFM) in the calculation for estimated REE: (kcal / day) = 500 + 22 (FFM) (McArdle, Katch, & Katch, 1991). Thompson and Manore (1996) compared this equation against directly measured REE and to several predictive REE equations. The REE estimate derived from the Cunningham equation was the only estimate that was not significantly different than the directly measured REE in a study of 24 male and 13 female endurance athletes.

TEE was determined using estimated REE plus EPA and TEF (7 % of REE + EAP) (Personal Communication, K.A. Carter-Erdman, January 2000; Mahan & Escott-Stump, 1996). An estimate of the EPA was derived using the participants' 7-day Activity Records from three time points over the study collection period (October to June). Time 1 for the 7-day Activity Record started on the first day of menstrual flow during the month of October for HP and November for C. Records two and three were collected in similar fashion at two-month intervals. Participants were given instruction on recording daily activity into four 6-hour periods which included: (1) a general description of the activity (e.g. reclining, sitting, standing, walking, running, skating etc.); (2) an estimation of the effort involved (e.g. light, moderate, or vigorous effort); (3) a specific description of the activity performed (e.g. sitting-reading, standing-talking, walking to school etc.); (4) duration in minutes performing each activity; and (5) a check mark designating those activities that were sport-specific. Sport-specific activity was defined in this study as the performance of hockey related training.

Verbal instructions with working examples were given regarding the accurate measurement and recording of activity type, intensity, duration and exercising radial pulse counts. The mean heart rate (HR) during exercise was calculated using HR taken at time 1, 2, and 3 as indicated on the Activity Records. Participants were also asked to record their weight each Monday during the study period. Each Activity Record was collected and evaluated at the end of the 7-day period and illegible and/or questionable entries were confirmed via telephone contact or personal interview.

The accuracy of using Activity Records to estimate TEE is variable, with errors of various methods ranging from 6 to 30 % of actual energy need (Campbell, 1999). To establish the accuracy of several commonly employed methods of determining activity level, Miller, Freedson, and Kline (1994) tested five recording questionnaires against direct measurement of PA using a Caltrac accelerometer. The 7-day PA recall and the Caltrac were the only method that resulted in a significant Spearman rank order correlation coefficient ($r = 0.79$).

EPA was also estimated based on each groups' habitual physical activity patterns and calculated as a percentage of REE: EPA = 75 % of REE for HP and 45 % of REE for C (Heyward, 1997; Mahalko & Johnson, 1980). This method is based on the factorial approach to calculating energy requirements of individuals (World Health Organization, 1985).

Energy Intake

EI was collected at the onset of the first day of menstrual flow from the 7-day dietary record. Diet records, three in total, were also collected at two-month intervals simultaneously with the collection of the 7-day Activity Records. To avoid recall bias, participants were instructed to record dietary intake within 30 minutes of ingestion and they were contacted via phone (RC) to fact-check their intake. A nutrition intern categorized data for entry into "Nutritionist 5.0 – version 1.6" software package (First

Data Bank, San Bruno, CA). Data were analysed to yield daily caloric intake, daily percentages for individual nutrients, and macro and micronutrient intakes.

Energy Balance

EB was calculated as the difference between the mean EI from the 7-day diet record and mean TEE calculated using REE, TEF, and EPA estimated by Heyward's (1997) approach, as stated above.

Bone Mineral Density

Baseline BMD testing of the lumbar spine (L1-L4), femoral neck, and distal radius occurred in week 1 for each group. DXA measurements were completed by a nuclear medicine technician (Hologic QDR2000– rectilinear scanner; Hologic, Inc.). Scan time was approximately 2 to 4 minutes and the coefficient of variation (CV) was better than 1.0 % for spinal measurements and approximately 2 to 3 % for femoral neck measurements (Hologic Manufacturers Inc., [On-line] August 1999; Personal Communication, Dr. R. Kloiber, April 2000). Individual BMD measurements in g/cm^2 were compared to Hologic QDR 2000 reference standards of mean young adult BMD (T-score: standard deviation from the peak bone mass or young normal values of a female reference population) as well as across the groups (Kanis, Melton, Christiansen, Johnston & Khaltaev, 1994; Maggi, 1993). All reporting of the BMD values (BMD three decimal places; T-scores and Z-scores one decimal place) follow the Recommendations for Bone Mineral Density Reporting in Canada (Siminoski, et al., 2005).

Body Composition

A sport anthropometrist measured participant's height (HT), weight (WT), girths (10 sites), limb dimensions (8 sites), and skinfold thickness (15 sites) at start and end of the study, using a medical scale, Harpenden skinfold caliper, an anthropometer, and steel tape. The equations of Parizkova (1978) and Matiegka (1921) were used to estimate BF %, FFM (kg), and muscle mass percent (LMM %). Body composition in this study included: WT, BF, and FFM.

DATA ANALYSIS

The main outcome was the identification of menstrual disturbances related to MC length (> 36 days oligomenorrhea vs < 21 days polymenorrhea), and SLP (< 10 days), as indicated by the MC diary and BBT measurements. The independent variables were: EI, TEE, EB, body composition, and BMD.

Sample size

Sample size was calculated from the results of a one-year prospective study investigating the proportion of menstrual disturbances in runners, as no available literature on female ice hockey players was found. Thirteen subjects (N = 66) were identified as experiencing menstrual disturbances based on their SLP (≤ 10 days) (Prior et al. 1990). Therefore, a conservative estimate of 20 participants was required to achieve a power of 80 % and an alpha of 0.05 for this study (Brant sample size calculator [On-line], August 1998).

Statistics

Statistical analyses were not performed because of the descriptive nature of the study; partly due to low compliance from HP. We report means (SD (±)) and box plots with median values, 25th and 75th percentiles, ranges, and outliers for percent change in WT, BF, FFM, BMD, T-Scores, Z-Scores, EI, TEE, EB, MC length and LP length. Percent change in WT, BF, and FFM were calculated to describe the pre (T1)-post (T2) body composition change (Percent change = [(Time 2 – Time 1)/ Time 2]X100).

RESULTS

Three HPs were excluded because they did not provide MC diary data and one HP was excluded from the BMD analysis due to multiple missed appointments. Twelve hockey players (mean, SD (±): age = 21.1 ± 3.4 years; HT = 165.9 ± 4.6 cm; WT = 64.7 ± 8.1 kg; BF % 22.8 ± 3.8) and 12 non-athletes (age = 21.4 ± 2.8 years; HT = 169.5 ± 5.5 cm; WT = 65.4 ± 6.4 kg; BF % 20.0 ± 3.1) were enrolled in all testing sessions (n = 24). Anthropometric characteristics and exercise patterns for HP and C are listed in Table 1. No differences were observed between groups in anthropometric characteristics at the study start. The HP participated in l0.3 ± 4.1 as compared to the C 4.4 ± 1.8 number of exercise sessions per week. The calculated mean HR per exercise session was 151.1 ± 17.5 bpm and 137.2 ± 9.7 bpm in the HP and C, respectively. (see Table 1).

Table 1: Mean and standard deviations (SD (±)) of Hockey Player and Control group's anthropometric and exercise characteristics.

	Mean, SD	**Min**	**Max**
HP (n = 12)			
Fat Free Mass (kg)	49.80 ± 5. 10	41.30	57.40
Muscle Mass (kg)	23.70 ± 4.30	16.70	31.80
Fat Mass (kg)	14.90 ± 3.90	10.50	21.60
Body Fat (%)	22.80 ± 3.80	17.00	29.20
Volume of thigh (cm^3)	9217.90 ± 1336.70	7477.80	12130.20
Number of exercise / week	10.30 ± 4.10	5.00	21.00
Exercise duration / session (min.)	73.80 ± 14.90	60.00	90.00
Heart Rate / Session (bpm)	151.10 ± 17.50	125.00	175.00
C (n = 12)			
Fat Free Mass (kg)	52.20 ± 4.00	47.00	59.20
Muscle Mass (kg)	25.60 ± 3.10	21.40	32.90
Fat Mass (kg)	13.20 ± 3.10	8.50	18.80
Body Fat (%)	20.00 ± 3.10	14.90	25.40
Volume of thigh (cm3)	9185.90 ± 1011.20	7281.80	10392.80
Number of exercise / week	4.40 ± 1.80	2.00	9.00
Exercise duration / session (min.)	56.70 ± 27.80	30.00	120.00
Heart Rate / Session (bpm)	137.18 ± 9.70	123.00	154.00

The median (25th and 75th percentile) WT percent change over the study duration for the HP and C was: -1.43 % (-2.45 % and 6.38 %) and 0.30 % (0.13 % and 2.53 %), respectively. The median (25th and 75th percentile) BF % percent change for the HP and C was: -4.42 % (-9.18 % and 0.34 %) and 4.73 % (-3.44 % and 9.28 %), respectively. No significant within group mean differences were observed for WT (HP p = 0.7; C p = 0.8), BF % (HP p = 0.97; C p = 0.6), and FFM (HP p = 0.6; C p = 0.98).

Bone Mineral Density

Median values, 25th and 75th percentiles, ranges of the baseline BMD values (g / cm^2) T-scores and Z-scores at the lumbar spine (L1-L4), total hip, and distal radius are presented in Table 2. Median lumbar spine and hip BMD were greater in the C (1.059 and 1.07 g / cm^2) than in the HP (1.047 and 1.064 g / cm^2). HP had one BMD measurement at the hip (1.357 g / cm^2) that presented as an outlier (see Figure 1). There were no significant differences in mean lumbar spine, hip, or radial BMD values between the HP and C. However, the maximum value for all BMD measurement sites was greater in the HP.

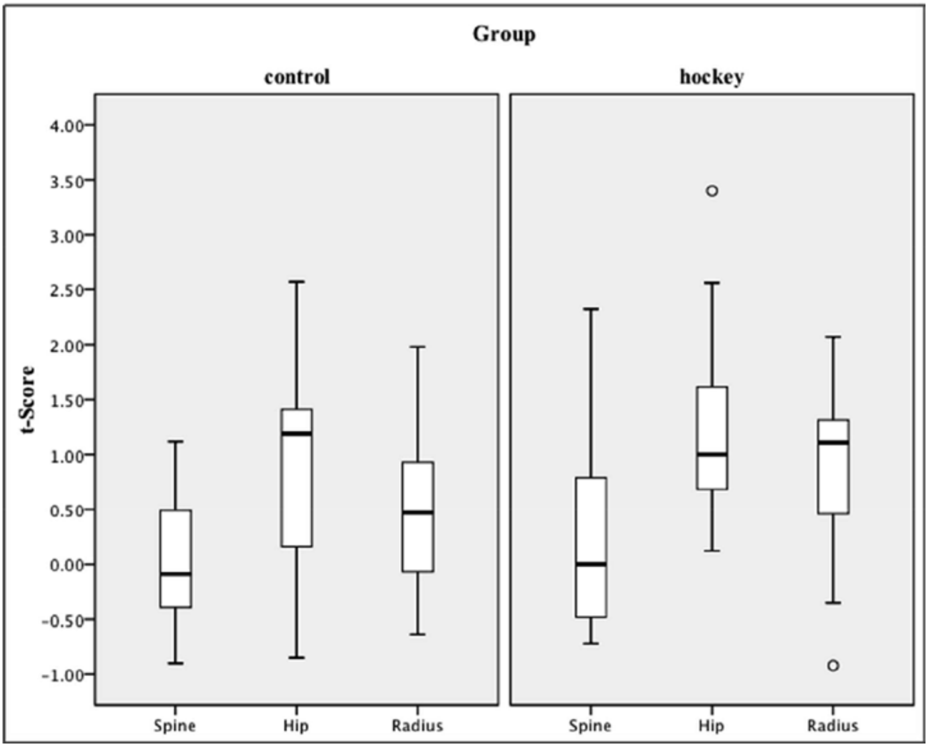
Median lumbar spine and hip T-scores were similar for HP and C. However median distal radius T-score was much larger for the HP (1.1) than C (0.5). The hip T-scores in the HP ranged from 0.1 to 3.4 as compared to a range of –0.9 to 2.6 in the C. The minimum lumbar spine (-0.7) and radial T-score (-0.9) in the HP and minimum T-score values at all sites in the C approached the WHO definition of osteopenia (-1.0 to -2.5). Figure 1 shows a comparison of HP and C T-scores values.

Table 2: Bone mineral baseline values by site and group

	Median	Percentile		Range	Median	Percentile		Range
		25th	75th			25th	75th	
Hockey (n = 11)						Control (n = 12)		
Lumbar spine L1-L4								
BMD g/cm^2	1.047	0.993	1.149	0.968 - 1.273	1.059	1.001	1.112	0.948 - 1.251
T-Score	0.0	-0.5	0.1	-0.7 - 2.3	0.1	-0.4	0.7	-0.9 - 1.9
Z-score	0.2	-0.4	1.1	-0.5 - 2.4	0.2	-0.2	0.9	-0.9 - 2.1
Total Hip								
BMD g/cm^2	1.064	1.007	1.116	0.956 - 1.357	1.07	0.982	1.12	0.838 - 1.257
T-Score	1.0	0.5	1.8	0.1 - 3.4	1.0	0.3	1.5	-0.9 - 2.6
Z-score	1.1	0.5	2.6	0.1 - 3.4	1.2	0.5	1.4	-0.9 - 2.6
Distal radius								
BMD g/cm^2	0.639	0.602	0.651	0.529 - 0.691	0.606	0.602	0.651	0.545 - 0.686
T-Score	1.1	0.4	1.3	-0.9 - 2.0	0.5	0.5	1.3	-0.6 - 1.9
Z-score	1.3	0.7	1.4	0.5 - 2.0	0.3	0.7	1.4	-0.5 - 1.9

Note: Outlier Total Hip HP 1.357 g/cm^2

Figure 1: Baseline BMD T-Scores at three sites for Control and Hockey Players.



Energy Expenditure

Table 3 lists the mean group values of EPA, REE, TEF and TEE for the HP and C. The mean HP-TEE was greater than C-TEE due to the varying contributions of EPA, TEF, and REE. EPA and TEF were greater in the HP than the C. The mean REE was greater for C. Values of EPA, REE, and TEF for both HP and C were verified to be within recommended theoretical percentages of TEE.

The median EI (25th and 75th percentile) values for the HP and C were: 1880.3 (1714.6 and 2080.85) and 1799.2 (1509.1 and 2038.5) kcal / day, respectively (see Table 4). One HP had an average daily energy intake of 1065.6 kcal / day and was represented as an outlier. The median (25th and 75th percentile) TEE for the HP and C groups were 2854.2 (2802.8 and 3080.28) and 2544.01 (2440.08 and 2659.55) kcal / day, respectively. The frequency distribution for HP-TEE is left skewed. Daily mean caloric intakes for the HP and C were 11 % (HP -1026.52 ± 450.1 kcal / day) and 15

Table 3: Components of mean (SD (±)) Total Daily Energy Expenditure by group

	HP n = 12			**C n = 12**		
kcal / day	**Mean, SD**	**Min**	**Max**	**Mean, SD**	**Min**	**Max**
EPA	1116.48 ± 79.30	985.71	1234.42	741.75 ± 39.50	690.10	810.68
REE	1594.98 ± 113.30	1408.16	1763.46	1648.33 ± 87.80	1533.56	1801.52
TEF	189.80 ± 13.50	167.57	209.85	167.30 ± 8.90	155.66	182.85
TEE	2901.26 ± 206.10	2561.44	3207.73	2557.38 ± 136.1	2379.32	2795.06

Note: Estimated Energy of Physical Activity (EPA), Resting Energy Expenditure (REE), Thermic Effect of Food (TEF) and derived Total Daily Energy Expenditure (TEE)

Table 4. HP and C Mean daily Energy Balance (calculated)

	Mean ± SD	
kcal / day	**Hockey n = 12**	**Control n = 12**
Daily Energy Intake	1882.89 ± 366.37	1777.31 ± 328.62
Range	1065 to 2419	1212 to 2219
Total Energy Expenditure	2901.26 ±206.1	2557.38 ± 136.06
Mean Energy Balance	-1018.37 ± 464.9	-780.07 ± 310.62

Note: Mean EB per day calculated as the difference between mean daily caloric intake (EI) and estimated total daily energy expenditure (TEE).

% (C -780.00 ± 310.19 kcal / day) less than that recommended by Health and Welfare Canada (1990) for females aged 16 to 49 years (2100 kcal / day).

There was a small difference of 105.5 kcal / day in daily EI between the HP and C but the number of exercise sessions per week and calculated mean HR per exercise session were also different. Thus, TEE between the HP and C was different and may have contributed to the difference in EB between the groups. Both groups maintained a state of negative EB for the study period as indicated by the mean TEE which exceeded daily caloric intake for both HP and C (see Table 4). The frequency distribution of HP-EB was symmetrical about the median and the median (25th and 75th percentile) was - 984.71 (- 1223.8 and - 767.7) kcal / day. The frequency distribution for C-EB was right skewed and the median (25th and 75th percentile) was - 681.72 (- 1046.57 and - 595.25) kcal / day. Energy balance remained in a negative state despite corrections for errors (self-report corrected factor ~30 %) in estimating daily EI such that mean corrected EB for the HP and C was –641.8 and –424.6 kcal / day, respectively.

Menstrual Cycle Characteristics

Out of a possible 78 cycles for the HP and 72 cycles for the C, only 22 HP and 51 C cycles were included in the analysis due to incomplete data. The HP and C median (25th and 75th percentile) LP lengths were 9.0 (9 and 11) and 9.4 (8.3 and 10.5) days, respectively. Mean LP length for the C (9.4 ± l.9) was slightly less than the HP (9.9 ± 1 .7) days. Both groups were classified as having SLP (<l0 days).

Table 5 indicates that three HP and three C had less than 25 % of eligible cycles defined as anovulatory. Five C and two HP had 25 to 50 % of eligible cycles designated as anovulatory. Two HP and two C had 51 to 75 % of eligible cycles considered anovu-

Table 5. Anovulatory cycle comparison between Hockey Players and Controls

HP ID	Cycle Tally	Cycles	% Anovulatory	C ID	Cycle Tally	Cycles	% Anovulatory
1	1	1	100	13	0	5	0
2	2	5	40	14	3	6	50
3	3	4	75	15	0	5	0
4	2	3	67	16	1	3	33
5	missing	missing	missing	17	4	6	67
6	missing	missing	missing	18	1	3	33
7	0	2	0	19	4	6	67
8	1	3	33	20	2	4	50
9	0	2	0	21	3	3	100
10	missing	missing	missing	22	1	1	100
11	1	1	0	23	1	5	25
12	2	2	100	24	0	5	0
Total	**11**	**22**	**50%**	**Total**	**22**	**51**	**43.1%**

Note: ID = subject number; cycle tally = number of anovulatory cycles out of total eligible cycles per subject; Cycles = Total number of eligible cycles; Anovulatory = Total anovulatory cycles; Percent = Total percent of anovulatory cycles per group.

latory. Finally, 75 to 100 % of eligible cycles were defined as anovulatory in two C and two HP. Thus, 11 out of 22 or 50 % of the HP eligible cycles were defined as anovulatory. Similarly, 43.1 % or 22 out of 51 C eligible cycles were defined as anovulatory.

DISCUSSION

In this paper, we describe menstrual disturbances across two groups: athletes (HP) and young university aged women (C) through the determination of MC and LP length, and occurrence of anovulatory cycles. Baseline BMD measurements were collected for comparison by group and with population based diagnostic reference standards (T-Scores). Descriptive data on EI, TEE, EB and body composition percent change over a 9-month study period were calculated and summarized.

Bone Mineral Density Measurements

BMD is an important component of bone strength and is considered the best method for diagnosis of osteopenia and osteoporosis. The maximum value for all BMD sites measured was greater in the HP, however, the median lumbar spine and hip values were slighter greater in the C. The median hip T-score may have been larger for the C because of the wider range in hip T-Scores (0.1 to 3.4) in HP when compared to C (–0.9 to 2.6). Surprisingly, the minimum values in HP's lumbar spine and radial T-score and C's lumbar spine, hip and radial T-score all approached the WHO definition of osteopenia. These results contrast with our hypothesis stating baseline BMD values at the spine, hip, and radial site in the HP would be greater than BMD values for population reference standards.

These BMD results are supported by Henderson, Price, Cole, Gutheridge, & Bhagat (1995) who reported normal values for mean BMD at the spine and proximal femur (Caucasian women, 20 to 50 years of age) to be 1.03 ± 0.113 g / cm^2 and 1.00 ± 0.114 g / cm^2, respectively. Mean BMD at the radius (0.69 ± 0.13 g / cm^2) reported by Faulkner et al. (1996) and Warren et al. (1991) is higher than mean radial BMD value for the HP and C in the current study. In studies of BMD in female runners, Drinkwater et al. (1984) and Rencken, Chesnut, and Drinkwater (1996) observed a significant difference in BMD at the lumbar spine for amenorrheic athletes (1.12 g / cm^2) when compared with matched eumenorrheic controls (1.30 g / cm^2). HP and C BMD at the lumbar spine were less than those reported in the Drinkwater et al. (1984) and Rencken et al. (1996) studies (HP = 1.066 ± 0.098 g/cm^2; C = 1.069 ± 0.086 g / cm^2). These comparisons are made with caution as measurement techniques may have been different and there was an mean age difference between our group (HP, 21.1 ± 3.4; C, 21.4 ± 2.8 years) and Drinkwater et al. (1984) (amenorrheic athletes 24.9 ± 1.3 years; eumenorrheic athletes 25.5 ± 1.4 years). It is possible that our HP and C may not be at their peak BMD for the lumbar spine due to their younger age (Recker et al., 1992).

Increased BMD in female athletes is reported to be dependent on the pattern of weight bearing activity and this effect is specific to the skeletal region stimulated by the activity. Specifically, increased BMD at the lumbar spine and lower body has been reported in male and female runners and weight lifters whereas increases at the distal radius have been reported for gymnasts and tennis players when compared to sedentary controls (Dalsky, 1990; Kannus, Haapasalo, & Sankelo, 1995; Petit, Prior, & Barr, 1999). Ice hockey players are assumed to experience constant skeletal loading at the hip during the motion of skating and loading of the upper body through the hands and wrists during stick handling. Thus, the research of Dalsky (1990), Kannus, Haapasalo, & Sankelo (1995), and Petit et al. (1999) into the activity specific accrual of bone mineral content would support increased mean BMD at these sites when compared to C. In this study, weight bearing sites specific to ice hockey did not manifest bone mass accrual specific to the activity pattern in the HP group when compared with the C. However, HP_1 (25.2 years old) and HP_9 (20.3 years old) did have T-scores at the hip that were greater than 2.5 SD above the mean peak bone mass or young normal values for population reference standards. HP_9 also had a radial T-score greater than 2 SD above the peak bone mass or young normal values for population reference standards.

Evidence of low bone mass (negative T-score values) was present in both groups: HP (5 lumbar spine -0.4 to -0.7 SD; 2 radial -0.4 and –0.9 SD); and C (6 lumbar spine -0.1 to -1.0; 2 hip -0.2 and –0.9 SD; and 3 radial -0.1 to –0.6 SD). These results raise specific concerns as Johnston and Slemenda (1991) estimate that the risk of fracture increases by 50-100 % for each decrease of 1 SD in BMD. In addition, Riggs et al. (1981) report the 90th percentile for vertebral BMD for patients with nontraumatic vertebral fractures to be 0.965 g / cm^2. HP_3 (18.1 yrs old) and HP_4 (20.9 yrs old) had mean lumbar spine BMD values of 0.968 g / cm^2 and 0.972 g / cm^2, respectively. C_{23} (29.6 yrs old) had a mean spinal BMD of 0.948 g / cm^2 which is less than the threshold below which the risk for non-traumatic vertebral fractures increases. Thus, each of these individuals with spinal BMD values nearing the fracture threshold and those having T-score values approaching –1 SD may be at risk for future fracture, indicating the potential need for monitoring and preventative education.

Several contributing variables have been emphasized in studies investigating osteoporosis or low BMD in female athletes. Henderson et al. (1995) suggest that BMD in young women is associated with their weight and muscle strength but not dietary intakes. Rencken et al. (1996) suggests amenorrhea and weight have a significant impact on BMD at the lumbar spine, trochanter, intertrochanteric region, and tibial shaft in amenorrheic, aerobically trained athletes. Further, these researchers recognize age of menarche as a significant predictor of lumbar spine BMD, suggesting that the later the age of menarche, the lower the lumbar spine BMD. Low calcium intakes (< 1000 mg / day), prolonged hypoestrogenism, occurrence of menstrual disturbances (specifically amenorrhea, SLP, and anovulatory cycles), have been recognized to differentially impact skeletal sites leading to low BMD values in female athletes (Drinkwater et al., 1984; Drinkwater, Bruemner, & Chesnut, 1990; Heaney, 1982; Prior et al., 1990; Warren et al., 1991). Although the current study neither attempted to measure circulating estrogen levels nor determine age of menarche, mean reported calcium levels were less than 1000 mg/day in both the HP (870.53 ± 250.99 mg / day) and C (785.33 ± 217.15 mg / day) and may partially contribute to the lack of bone-trophic effect for weight bearing activity (Heaney, 1982).

Nutritional Status, Energy Balance and Body Composition Changes

The identification of caloric restriction or lack of nutrition is important in all athletes because of the effect, both positive and negative, this may have on sport performance and subsequent health. The general perception is that female athletes involved in aesthetic or weight dependent sports, restrict caloric intake and become energy deficient. Although female hockey players do not fit the typical risk profile for the Triad with respect to body composition (e.g. low body weight and low fat mass), they are involved in intense physical training.

Median EI values for the HP and C were 1882.89 ± 366.37and 1777.31 ± 328.62 kcal / day, respectively. These values are less than our previous EI research with varsity soccer (1850.70 ± 417.40 kcal/day), basketball (2639.50 ± 586.21 kcal/day) and volleyball players (2134.80 ± 286.00 kcal/day) (Doyle-Baker, MacDonald, Hewitt, & Harris, 2000). They are also less than the 2100 kcal / day RNI (Required Nutrient Intake) by Health and Welfare Canada (1990). Jones, Martin, Wanfang, and Boyd (1997) suggest that Health Canada's RNI values for women aged 25 to 49 years (2100 kcal / day) are substantially below the true requirement level of the population age based subgroup. Thus, if the reported daily EI for the HP and C are below the current recommendations by 11 % and 15 %, respectively, this may be the first indication of the inadequacy of dietary caloric intake occurring.

Despite the indications of low caloric intake, it is important to consider that underreporting due to day to day variation in energy status, the disturbance of normal dietary practices, food recall problems, and errors related to analysing EI or calculating EE using activity records or generalized equations, contributes to a decrease in the accuracy of determining the energy requirement for a population. Specifically, when 7- day diet records were compared to the "gold standard" DLW method of determining EI and EE, errors in estimating EI of up to ± 30 % of actual need have been reported (deVries, Zock, Mensink, & Katan, 1994; Mahalko & Johnson, 1980; Sawaya, Tucker, & Tsay, 1996; Todd, Herdes, & Calloway, 1983). We calculated an error estimate of 20 % of actual energy need reported to determine a closer approximation of the daily caloric intake considering all possible sources of error. The mean corrected daily caloric intake (HP 2259.47; C 2132.77 kcal / day) would increase their totals to the current age specific RNI.

Energy Balance

Long term negative EB (EE exceeds EI) leads to a decrease in utilizable metabolic fuels, disrupted LH pulsatility, and subsequent alterations to thyroid metabolism and basal metabolic rate (Loucks & Heath, 1994; Loucks & Verdun, 1998; Loucks, Verdun, & Heath et al., 1998; Wade, Schneider, & Li, 1996). The "energy availability hypothesis" suggests that the GnRH pulse generator is disrupted by an unidentified signal that dietary intake is inadequate for the energy cost of reproduction and locomotion (Loucks et al., 1998).

On average, HP and C maintained a state of negative EB as indicated by their mean TEE exceeded their daily caloric intake. These results are comparable to those reported by Drinkwater et al. (1984), who reported that EB in a group of amenorrheic athletes was significantly lower than in an eumenorrheic group. In the Drinkwater, et al. (1984) study, EI were not different, but training was significantly more rigorous in the amenorrheic group. In our study, the HP and C were divided by exercise and not based on their menstrual status. There was a small difference of 105.5 kcal / day in daily EI between the groups and the HP had more rigorous training based on the number of exercise sessions per week and calculated mean HRs per exercise session which were different. Thus, TEE between HP and C was different and should have contributed to higher EI in HP and a difference in EB. However, both groups experienced a negative state despite corrections for errors in estimating daily EI; mean corrected EB for the HP and C was –641.80 and –424.60 kcal / day, respectively.

The EB findings are practically important in consideration of the research of Loucks (1996) who suggests that exercise contributes to menstrual disturbances only when the EE is not adequately replenished. This is supported in our study as although the exercise sessions per week and exercise intensity differed between HP and C both experienced similar MC aberrations, possibly because of negative EB. In a series of well controlled studies Loucks and coworkers (Loucks & Heath, 1994; Loucks & Verdun, 1998; Loucks, Verdun & Heath, 1998), suggest energy deficits are associated with significant suppression of triiodothyronine (a regulator of metabolic rate) and disruption of LH pulsatile secretion (a strong indicator of reproductive performance). Further, they suggest that the degree of disruption in LH pulse amplitude and frequency is in direct relation to the duration of energy depletion. Since successful reproductive function requires appropriate hormonal stimulation, individuals experiencing disruptions in LH pulsatility may experience a range of menstrual disturbances including amenorrhea, oligoamenorrhea, SLP, and anovulation. Our results may be suggestive of this phenomena since a state of negative EB was consistently maintained throughout the study and menstrual disturbances in the form of SLP, oligoamenorrhea, and anovulation did occur in both the HP and C.

Body Composition

Even though both the HP and C maintained a state of negative EB for the study period, percent changes in WT, BF, and FFM were very small (HP –1.43 %, –4.42 %, 0.82 %; C 0.30 %; 4.73 %; –0.15 %) respectively. This finding supports the hypothesis that HP would be in a state of negative energy balance in the absence of any change in body composition.

Menstrual disturbances can occur with or without weight loss (Yen, 1998). The general perception that changes in weight or body composition, specifically BF %, are an impetus to the onset of menstrual disturbances in female athletes has been challenged by several studies that have failed to correlate reproductive function and body composition (DeCree, 1998; I'Anson, Foster, & Foxcroft, 1991; Sanborn, Albrecht, & Wagner, 1987; Sinning & Little, 1987). Specifically, reproductive function has been linked to general availability of metabolic fuels (Loucks & Callister, 1993; Loucks & Heath, 1994; Loucks, Verdun, & Heath, 1998; Wade & Schneider, 1992). The results of the current study may lend support to the work of several researchers investigating the role that EA plays in regulating metabolism and producing a "hypometabolic state" in which available energy is stored or partitioned to only essential functions (Laughlin & Yen, 1996).

DeCree (1998) documented alterations to the thyroid axis in individuals experiencing energy deficits linked to menstrual disturbances. Such alterations lead to reduced BBT and reduced metabolic rate. The athletes in this current study did experience asymptomatic menstrual disturbances in the absence of significant changes in WT, FFM, and BF %. Although, thyroid levels were not measured, these results potentially lend support to future research directed at determining the possible mechanism behind menstrual disturbances, specifically SLP, oligo amenorrhea, and anovulation, and a reduction in metabolic rate leading to maintenance of body composition. Further, the lack of body composition change in this study supports the need to expand the present Triad model (its association with amenorrhea, eating disorders, and loss of weight) to adopt the understanding that menstrual disturbances do occur in active individuals who do not experience the symptomatic changes in body composition associated with amenorrhea in anorexia nervosa.

Menstrual Disturbances

HP and C mean menstrual cycle lengths were similar and were within the defined normal menstrual cycle length of 21-36 days. Maximum MC lengths are suggestive of the occurrence of oligomenorrhea (cycles greater than 36 days) in both groups (Abraham, 1978). Similarly, the minimum MC length are indicative of polymenorrhea (cycles length of less than 21 days) occurring in HP (Abraham, 1978). Mean LP length in days for the C was slightly less than that of the HP, yet both groups are classified as having SLP (< 10 days). These results support the hypothesis that the HP group would exhibit a longer MC length when compared with C. However, the data do not support the hypotheses that the HP group would exhibit SLP (< 10 days) and a greater number of anovulatory cycles when compared with C.

Luteal function was disturbed in 55 of 150 eligible cycles (sum of all SLP and anovulatory cycles) or in 36.70 % of recorded cycles in the HP and C combined. Specifically, anovulation occurred in 50.00 % of HP and 43.10 % of C cycles. Similar to the study by Prior et al., (1990) disruptions to the LP occurred in both highly active HP (10.30 ± 4.10 sessions / week) and in the recreationally active C (4.40 ± 1.80 sessions / week). Although, Prior et al (1990) did not mention energy status, the HP and C in this study were in a state of negative EB and this may be impacting GnRH responsiveness and LH pulsatility possibly leading to altered menstrual function. The results of this study may support the work of Loucks et al. (1998) who concluded that low EA, not stress of exercise or athletic involvement, alters LH pulsatility in exercising women. Thus, there is a potential that these “apparently healthy” HP and C may be exposed to the detrimental effects of low reproductive hormones without their knowledge.

Limitations

We realize that this study had a large participant burden as our original recruitment at the outset of the study exceeded the sample size calculation. However, the BBT method coupled with keeping a MC diary was time intensive for the HP with only a 28 % completion rate versus 70 % with the C. In addition, differences may be present but obscured due to errors inherent in the estimation of EB and / or because the C were university students and thus not a true “sedentary” control group. While EA did change, the effects on MC may not occur for months, and an effect on BMD may not occur for years (Drinkwater, Nilson, Ott, & Chesnut 1986; Hanley, 1996).

CONCLUSIONS

In conclusion, the prospective, descriptive nature of our study was designed to increase awareness regarding the presence of menstrual disturbances in a select population of female ice hockey players and to potentially highlight the need for intervention in this sub-population of female athletes related to future BMD loss. Despite the errors inherent in indirect measurement of energy status and menstrual cyclicity, the results of our study do add to the literature which now identifies a strong relationship between reproductive endocrinology, EB and BMD. We hypothesized that neither intensity of exercise nor body composition independently would disrupt menstrual function; rather, inadequate dietary replenishing of expended energy leading to long term negative EB would explain the occurrence of menstrual disturbances. Of course, this is well known today, but we did due diligence at the time of the study and contacted several researchers to ensure that we were identifying a plausible trend (Personal Communication, D. A., Haney; A. B. Loucks; V. Harber; and J. C. Prior, October 2000).

In 2014, the IOC published a consensus statement: beyond the Female Athlete Triad and introduced a more comprehensive term that employs a multi-disciplinary approach across all athletes. This reframing resulted in a new model renamed, Relative Energy Deficiency in Sport (RED-S), which is caused by energy deficiency relative to the balance between dietary EI and EE (Mountjoy et al., 2014).

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PHYSICAL ACTIVITY AND EXERCISE AS A KEY FACTOR IN SUCCESSFUL AGING

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ABSTRACT

Aging is associated with several changes in body composition and metabolism, including sarcopenia, sarcopenic obesity and decrease in bone mass; aerobic capacity, muscle mass and strength tend to decline progressively. These changes have considerable impact on the ability to perform daily activities, especially when associated with chronic diseases, such as type 2 diabetes, cardio-vascular disease and dyslipidemia, as well as geriatric syndromes, like mobility impairment, falls and frailty. Scientific research has shown that physical activity and exercise can slow the physiological aging clock. Particularly, active elderly people seem to age "successfully" compared to sedentary ones. The aim of our work is to review evidence-based recommendations for physical activity, exercise and diet that would help to preserve muscle mass and strength, and to reduce the gain of fat mass in older adults. Increasing levels of physical activity, in particular resistance training mixed with aerobic exercise, and adequate protein nutrition intake should be an integral component in the prevention and treatment of sarcopenia, sarcopenic obesity and metabolic syndrome in elderly subjects.

***Key words**: sarcopenia, physical activity, body composition, energy consumption, nutrition, aging.*

GIBALNA AKTIVNOST IN VADBA KOT KLJUČNA FAKTORJA PRI USPEŠNEM STARANJU

IZVLEČEK

Staranje je povezano s številnimi spremembami v telesni zgradbi in metabolizmu kot so npr. sarkopenija, sarkopenična debelost in zmanjševanje kostne gostote. Posledično se progresivno zmanjšujejo aerobna kapaciteta ter mišična masa in moč. Te spremembe odločilno vplivajo na sposobnost opravljanja vsakdanjih aktivnosti, še zlasti v povezavi z morebitnimi kroničnimi boleznimi kot so npr. sladkorna bolezen tipa 2, kardio-vaskularne bolezni in dislipidemija pa tudi raznimi geriatričnimi sindromi kot so motnje v gibanju, padci in krhkost. Znanstvene raziskave so pokazale, da gibalna aktivnost in vadba lahko upočasnita fiziološko uro staranja. Predvsem se zdi, da se aktivni starostniki starajo »bolj uspešno«, kot pa sedentarni. Namen članka je pregled priporočil, pridobljenih na osnovi raziskav in dokazov, za gibalno aktivnost, vadbo in prehrano, ki bi pomagali starejšim odraslim ohraniti mišično moč in maso ter hkrati zmanjševati maščobno maso. Povečevanje stopnje gibalne aktivnosti, predvsem vadbe za moč v povezavi z aerobno vadbo, ter ustrezna prehrana bogata s proteini, bi morale biti integralne komponente pri preprečevanju in obravnavi primerov sarkopenije, sarkopenične debelosti ter metaboličnih sindromov pri starejših osebah.

Ključne besede: *sarkopenija, gibalna aktivnost, telesna zgradba, poraba energije, prehrana, staranje*

INTRODUCTION

Aging is associated with major changes in body composition, including an increase and redistribution of adipose tissue and a decrease in skeletal muscle mass (sarcopenia) and bone mass, events that begin generally around the fourth decade of life (Fiatarone Singh, 2002).

Recent population trends (Swinburn et al., 2011) indicate an alarming rise in the prevalence of obesity among older adults, potentially adding a complementary condition that compounds the risk of poor health outcomes. The interplay between sarcopenia and rising trends in obesity in aging population emerged more than 20 years ago as an important public health concern in geriatrics (Evans & Campbell, 1993). The prevalence of sarcopenia and sarcopenic obesity increases with age, thus, muscle mass and strength lead to a progressive decline (Zamboni, Mazzali, Fantin, Rossi, & Di Francesco, 2008). These changes have considerable impact on the ability of performing daily activities (Batsis, Mackenzie, Lopez-Jimenez, & Bartels, 2015; Baumgartner et al., 2004) and have also significant consequences in health and functioning of the individual, because of their association with chronic disease's expression and severity, as well as with mobility impairment, risk of falls and frailty.

Changes in muscle, fat and bone surely relate to an excess / incorrect energy consumption, decreased energy expenditure in physical activity, or both factors in combination. However, in elderly subjects, other mechanisms are involved, such as changes in hormones regulating metabolism, like growth and sex hormones (Sakuma & Yamaguchi, 2012).

This declining need for energy due to a reduction in the amount of lean body mass and a more sedentary lifestyle, together with an inappropriate dietary intake, is the first step in the development of malnutrition in the elderly.

The main age-related changes in body composition and physiologic function which appear to influence nutrient requirements in older adults are listed in Table 1 (Blumberg, 1997). Dietary intervention has to be considered a key part of the demographic challenge of an aging population and should be a fundamental part of the public health policy necessary to reduce chronic diseases and compress morbidity (Bendich & Deck-

Table 1. Examples of Age-Related Changes in Body Composition and Physiologic Function that Influence Nutrient Requirements (modified from Blumberg, 1997).

Changes in body composition or physiological function	**Impact on nutrient requirements**
Decreased muscle mass	Decreased need for energy
Decreased bone density	Increased need for calcium and vitamin D
Decreased immune function	Increased need for vitamin B6, vitamin E and zinc
Increased gastric pH and decreased gastric motility	Increased need for vitamin B6, folic acid, calcium, iron, zinc, fiber and water
Decreased skin capacity for cholecalciferol synthesis	Increased need for vitamin D
Increased wintertime parathyroid hormone production	Increased need for vitamin D
Decrease calcium bioavailability	Increased need for calcium and vitamin D
Decreased efficiency in metabolic use of vitamin B6	Increased need for vitamin B
Increased oxidative stress and homocysteine levels	Increased need for beta-carotene, vitamin C, vitamin E, folate, vitamin B12 and B6
Decreased vitamin absorption	Increased need for food choices with high nutrient density

elbaum, 2015), as well as physical activity and exercise, capable of slowing down the physiological aging clock (Taylor et al., 2004).

METHODS

Relevant medical literature was identified from searches of PubMed and references cited in appropriate articles identified. Search terms used included sarcopenia, aging, physical activity, elderly, exercise, body composition, aging metabolism, nutrition. More detailed search terms were used following the identification of relevant mechanisms and to identify epidemiological studies. Selection of articles was based on peer review, journal and relevance.

AGE-RELATED CHANGES IN BODY COMPOSITION AND BODY METABOLISM

Sarcopenia, commonly associated with fat infiltration into muscles (sarcopenic obesity), is very common, with a prevalence of ~5 % in persons aged 65 years and as high as 50 % over the age of 80 (Janssen, 2010). It leads to a decline in muscle strength and power, supported also by altered muscle energetics, changes in tendon insertion, altered muscle coordination and decreased blood flow in the capillary bed of the muscle. Fat infiltration into muscle (myosteatosis) is associated with decreased strength and an increase in the prevalence of disability (Rolland et al., 2009).

As an adequate nutrient intake is essential to maintain muscle mass, the decline in food intake with aging plays a role in the development of sarcopenia. In particular, maintenance of muscle mass requires adequate protein intake; it is postulated that older persons require at least 1.2 g / kg of protein a day (Morley et al., 2010).

From a metabolic point of view, the most important consequence of sarcopenia is the decrease in energy expenditure (in particular for physical activity) and basal metabolic decline; moreover, the ability to increase or decrease energy expenditure to counterbalance overeating or undereating is impaired with an increased susceptibility to energy imbalance (both positive and negative) (Roberts & Rosenberg, 2006). A decline in basal metabolic rate (BMR) with aging is well recognized (Poehlman, 1992), associated to a loss of fat free mass (FFM) and a gain of fat (FM), a less metabolically active tissue.

A longitudinal study by Keys, Taylor, and Grande (1973) documented a decline in BMR with age of 1 – 2 % per decade. Other studies examining the role of aging on resting metabolic rate (RMR) and substrate oxidation (Frisard et al., 2007; Krems, Lührmann, Straßburg, Hartmann, & Neuhäuser-Berthold, 2005) indicate a reduction in RMR with age greater than what would be predicted from the observed modification of FM and FFM, suggesting that the lower RMR of older adults may be due in part to slowed organ metabolic rates (St-Onge & Gallagher, 2010).

The lowering of FFM and the increase of FM, in particular of visceral adipose tissue (VAT), is related to an increased risk of cardiovascular disease, type 2 diabetes, hyperlipidemia, hypertension and malignancy (Donohoe, Doyle, & Reynolds, 2011).

The association of central adiposity to poor health is related to VAT accumulation and associated with hyperinsulinemia and insulin resistance. VAT secretes a number of adipokines and inflammatory cytokines (TNF- α, IL-6, IL-1β) that can up-regulate nuclear factor-κB (NFκB), which leads to an increase in nitric oxide (NO), a substrate for reactive oxygen species (ROS) (Sonnenberg, Krakower, & Kissebah, 2004).

Moreover, excess adiposity is associated with a state of low-grade chronic inflammation, which interfere with adipose cell differentiation and adipokines pattern secretion, resulting in dysfunctional adipose tissue (Paniagua, 2016). Increased VAT is associated with elevated free fatty acids, impaired hepatic insulin clearance, resulting in hyperinsulinemia, increased gluconeogenesis, and elevation of very-low-density lipoprotein secretion (Matsuzawa et al., 1995).

In this state the subject presents a pro-inflammatory, pro-coagulant and insulin resistant state typical of the metabolic syndrome (Despres & Lemieux, 2006).

PHYSICAL ACTIVITY AND AEROBIC / ANAEROBIC EXERCISE

Regular physical activity is one of the most important protective factors against the development of chronic diseases (Harridge & Lazarus, 2017): sedentary individuals show an higher incidence of cardiovascular disease, diabetes mellitus, obesity as well as different malignancy. On the contrary, physical activity is inversely related to all-cause mortality in older adults (Brown et al., 2012).

Besides being active or un-active, also the total amount and intensity of physical activity is important. The physiologic adaptations to aerobic and resistance exercise are different: aerobic exercise improves cardiovascular function, that increase peak oxygen consumption without significantly changing strength, whereas resistance exercise improves neuromuscular adaptations, leading to an increase in strength, without significantly changing peak oxygen consumption. Despite lower baseline values, the available data suggest that older individuals have the same relative improvement in maximal strength and maximal aerobic capacity to resistance training and aerobic training regimens, respectively (Lambert & Evans, 2005). These physiologic adaptations may integrate with each other when the two types of training are performed together (Wilson et al., 2012). In a recent clinical trial (Villareal et al., 2017), the effectiveness of aerobic exercise has been compared with resistance exercise and combined exercise in reversing frailty and preserving muscle and bone mass, during weight loss in obese older adults. The authors show that combined aerobic and resistance training seems to provide the greatest benefits with respect to physical function (PF) and relative preservation of lean mass. In particular, the most effective exercise protocol for frail older adults seem to be a multi-component training, performed three times per week, with shorter-duration sessions (30 – 45 min), in order to prevent adverse health consequences (Theou et al., 2011).

PHYSICAL EXERCISE AND CHANGES IN BODY COMPOSITION

Human body is composed of water, protein, minerals, and fat. The total amount of body fat consists of essential fat (detectable in bone marrow, heart, lungs, liver, spleen, kidneys, muscles, and central nervous system) and storage fat, that accumulates in adipose tissue. Lean body mass (LBM), comprehensive of muscles, bones, ligaments, tendons, and internal organs, differences from FFM for the content of a small percentage of essential fat (bone marrow and organs).

Regular physical activity has an overall positive effect on body composition, modifying both FFM and FM, muscle volume, muscle strength, and physical mobility in older people, including overweight and obese individuals (Liao et al., 2017). Many studies describe a non-significant change, reliable to exercise, in FFM (Toth, Beckett, & Poehlman, 1999), even if exercise leads to an increase in skeletal muscle mass, especially if anaerobic one, with a related increase in strength.

According to different studies on elderly people, an aerobic exercise of moderate intensity (VO_{2max}> 60 %), is generally associated to a lowering in total body fat (FM), even in the absence of changes in dietary regime, proportional to the amount of training sessions. In particular, aerobic exercise can induce significant results on the loss of adipose tissue in the abdominal region (VAT) (Kay & Fiatarone Singh, 2006).

In order to estimate VO_{2max} and fitness index, a two-km walking test was developed by the UKK Institute in Finland (Laukkanen, Oja, Ojala, Pasanen, & Vuori, 1992). This test, relatively simple to administer, is a feasible and accurate alternative for determination of cardio-respiratory fitness in adults with both normal body weight as well as in overweight individuals. In Ferrara's population of PANGeA study (a mass population study we conducted with Slovenian colleagues, aiming at identifying the main elements involved in successful aging), applying the UKK-test, we found that fitness index is inversely correlated to waist circumference. In this population, applying a linear regression model, fitness index, independently of gender and age, predicts waist circumference, explaining the 32 % of its variability (R^2 square 0,322, standardized β-coefficient -0,477, $p < 0,001$) (Figure 1, data not published).

PHYSICAL EXERCISE AND CHANGES IN BODY METABOLISM

Even if not associated with a specific dietary regime, aerobic exercise and resistance training may be responsible for different and positive changes in body metabolism:

- improvement in glycemic control (Sigal et al., 2007), due to increases in muscle GLUT4 number and function (Holten et al., 2004),
- improvements in insulin sensitivity (Winnick et al., 2008),
- stimulation of lipid oxidation,
- improvement in lipid profile with increased clearance of atherogenic lipids, specially triglycerides (Katsanos, 2006), reduced levels of total cholesterol and apolipoprotein B (Holme, Høstmark, & Anderssen, 2007), changes in LDL par-

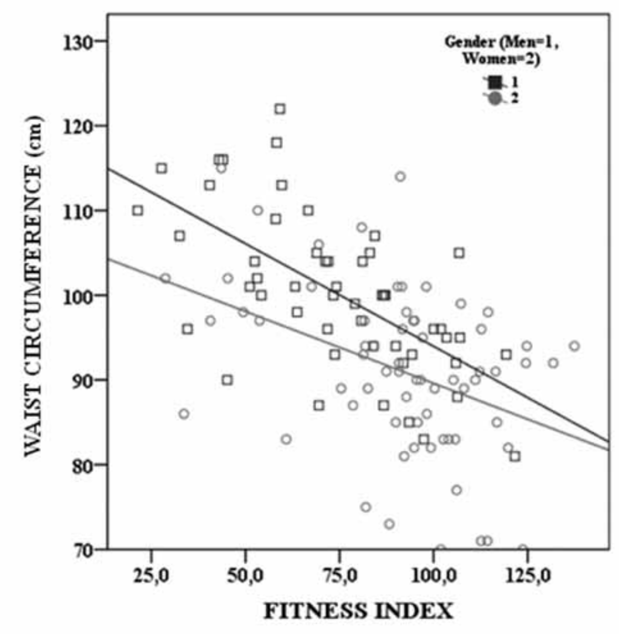

Figure 1. Pearson correlation between fitness index and waist circumference (PAN-GeA's Ferrara population): Men r=-0,62, p<0.001; Women r=-0,401, p=0,001.

ticle size (more than lower levels) and higher HDL concentration (Kraus et al., 2002),
- decrease of VAT in relation to an increased sympathetic tonus and consequential increased lipolysis (Ismail, Keating, Baker, & Johnson, 2012),
- decrease of VAT and pro-inflammatory state may contribute to improve glucose uptake (Fisher et al., 2011),
- decrease in biomarkers of inflammation like C-reactive protein (CRP) (Strasser, Arvandi, & Siebert, 2012),
- improvement in adiponectin and leptin profile (Simpson & Singh, 2008).

The impact of aerobic exercise on body metabolism is better and more significant if it is characterized by high intensity; strength training can provide up to a 15 % increase in metabolic rate, which is very helpful in terms of weight loss and long-term weight control.

A review published by Strasser provides strong support for the recommendation that physical activity, in particular resistance training mixed with aerobic exercise, should be an integral component in the prevention and treatment of obesity and metabolic syndrome risk factors (Strasser, 2013). Resistance training is an effective way to increase energy requirements, decrease body FM, and maintain metabolically active tissue mass. A consequent improvement in insulin sensitivity and in the lipid profile could reduce the risk of metabolic syndrome and type 2 diabetes and attenuate the development of cardiovascular disease in an elderly population (Ferrara, Goldberg, Ortmeyer, & Ryan, 2006).

PHYSICAL ACTIVITY AND RELATED BENEFITS

It is well established that with increasing age, individuals are more likely to experience functional declines, mobility limitations, and physical disability (Holmes, Powell-Griner, Lethbridge-Cejku, & Heyman, 2009). A large body of literature has supported the interrelationships among various factors affecting physical function (PF) in older adults (Villareal et al., 2011), like physical activity, body composition (fat mass and skeletal muscle mass), muscle capacity (leg strength and leg power), and muscle quality, whose aging-related changes tend to promote a decline in maximal aerobic power and skeletal muscle force production. Although the likelihood of physical limitations and disability increases with age, multiple studies have demonstrated that exercise is an effective intervention strategy for improving PF in older adults (Brady, Straight, & Evans, 2014).

Several intervention trials have reported improvements in PF after a resistance training program in relatively healthy older (Avila, Gutierres, Sheehy, Lofgren, & Delmonico, 2010; Henwood & Taaffe, 2005), as well as older adults with chronic health conditions (Yang, Wang, Lin, Chu, & Chan, 2006). In addition, aerobic training, often a cornerstone of an exercise program, has also been found to be beneficial at improving PF in older adults (Davidson et al., 2009).

The evidence suggests how the pillars of an effective exercise program should be both aerobic and resistance exercise (Chodzko-Zajko et al., 2009) and it is well known that both endurance exercise and resistance training can substantially improve physical fitness and health-related factors in older individuals (Conceição et al., 2014). This helps to maintain and increase skeletal muscle mass and respiratory fitness, with increase in resting metabolic rate and enhanced capacity for lipid oxidation during rest and exercise. Endurance training in particular is purported to be more effective for decreasing FM, resting heart rate and blood pressure, while resistance training has been shown to be more effective for increasing basal metabolism, bone mineral density (BMD) and muscle strength and power (Romero-Arenas, Martínez-Pascual, et al. 2013).

Regular physical activity can have a positive effect on disorders and diseases that affect muscles and bones (such as osteoarthritis, back pain and osteoporosis). Walking provides a modest increase in the loads on the skeleton above gravity and, therefore, this type of exercise has proved to be less effective in osteoporosis prevention. Strength exercises instead, seems to be a powerful stimulus to improve and maintain bone mass during the aging process (Gomez-Cabello, Ara, González-Agüero, Casajús, & Vicente-Rodríguez, 2012).

Many evidences show that physical activity programs aimed at strengthening muscles help the elderly to maintain balance, which decrease the likelihood and severity of falls and fractures, one of major health concerns for many older adults (Howe, Rochester, Neil, Skelton, & Ballinger, 2011).

Compared to sedentary people, older athletes enjoy a wide range of physiological benefits on health:

- a better profile in body composition including a lower accumulation of total and especially abdominal fat; greater volume muscle mass in upper and lower limbs,
- higher bone mineral density (BMD), especially in case of strength training with high-load low repetitions (Romero-Arenas, Blazevich, et al., 2013),
- articulation muscle more resistant to oxidative processes and fatigue,
- a better cardiac output during maximum exercise and improved cardiovascular fitness (Gibala, Little, MacDonald, & Hawley, 2012),
- less cardiovascular and metabolic stress during sub-maximal exercise (Lanza et al., 2008),
- significantly reduced coronary risk profile in relation to lowering of blood pressure (Whelton, Chin, Xin, & He, 2002), improvement in endothelium function (Maiorana, O'Driscoll, Taylor, & Green, 2003); low systemic inflammatory index; improved insulin sensitivity and glucose homeostasis; better lipid profile and lower waist circumference,
- slowed development of disability in old age.

Twenty to forty minutes a day of aerobic training leads to a lower probability to develop metabolic and cardiovascular diseases. Moreover, several studies have shown the beneficial effects of circuit weight training in individuals with CHD. Volaklis et al. combined resistance circuit and aerobic exercise program in patients with coronary artery disease. Subjects improved cardiovascular fitness (VO_2 peak 15.4 %) and muscular strength significantly in all exercises by an average of 28 % (Volaklis, Douda, Kokkinos, & Tokmakidis, 2006).

A Cochrane review of 121 randomized controlled trials of progressive resistance training (PRT) in older people showed that doing PRT 2–3 times per week improved physical function, gait speed, timed get-up-and-go, climbing stairs, and balance, and, more importantly, had a significant effect on muscle strength, especially in the high-intensity training groups (Crocker et al., 2013).

In order to optimize body composition, muscle strength gains and to develop cardiovascular function, Romero-Arenas, Martínez-Pascual, et al. (2013) recommended a circuit weight training with a minimum frequency of 2 sessions per week (with a volume ranging from 30 to 50 minutes) that could be implemented with endurance training.

DIET COMBINED WITH PHYSICAL ACTIVITY AND BODY COMPOSITION

Aging is related with the loss of skeletal muscle and bone mass along with progressive increase of adipose tissue. Recent investigations have attempted to modify these processes with various combinations of dietary and exercise intervention (Iglay, Thyfault, Apolzan, & Campbell, 2007; Kukuljan, Nowson, Sanders, & Daly, 2009).

Increasing the quantity and quality (essential amino acids, specifically leucine) of dietary protein stimulates muscle protein synthesis in the elderly (Børsheim et al., 2008), while protein supplementation at twice the Recommended Dietary Allowance (RDA) does not improve skeletal muscle function or increase muscle mass in healthy elderly weight lifters compared to those on a normal diet (Campbell & Leidy, 2007). Therefore, regular resistance exercises and the habitual ingestion of adequate amounts of dietary protein from high-quality sources are two important ways to slow the progression and treat sarcopenia. Assuming three meals are consumed each day, a relative protein dose of 0.4 – 0.5 g / kg / meal is consistent with recent expert opinions concerning the optimal daily protein intake (1.2 – 1.5 g / kg / day) for healthy older adults (Deutz et al., 2014).

This amount of protein markedly exceeds the RDA for protein (at present set at 0.8 g / kg ideal body mass / day for healthy adults, regardless of sex and age), but it is supported by several larger-scale longitudinal studies (Bartali et al., 2012; Gray-Donald et al., 2014). Several studies have also reported a positive relationship between protein intake and peak bone mass in older adults (Hannan et al., 2000; Sahni et al., 2014).

Increased intake of vitamin D stimulates gene expression and boosts muscle protein synthesis, facilitates neuromuscular function and enhances strength and balance (Muir & Montero-Odasso, 2011). In a recent clinical trial, Rondanelli et al. found a significant beneficial effect of supplementation with whey protein, essential amino acids, and vitamin D compared with placebo in elderly sarcopenic adults participating in controlled resistance training, with a gain of 1.7 kg in FFM. Supplementation attenuated the inflammatory state, as seen by the significant drops in CRP concentrations and leads to a reduced prevalence of malnutrition, assessed with the Mini Nutritional Assessment (MNA) (Rondanelli et al., 2016).

In all individuals older than 70 years of age, vitamin D intakes of at least 600 IU per day (up to 1000 IU / day) are recommended, in addition to the calcium requirement of 1200 mg per day (American Geriatrics Society Workgroup on Vitamin D Supplementation for Older Adults, 2014). For those individuals in whom there is inadequate calcium and vitamin D intake from diet, supplements and/or multivitamins can be used.

A recent review suggests that calcium and vitamin D supplementation, with or without osteoporosis therapy, may decrease the risk of fractures (Tricco et al., 2017). Anyway, the U.S. Preventive Services Task Force (USPSTF) concluded that the current evidence is insufficient to assess the balance of benefits and harms of combined vitamin D and calcium supplementation to prevent bone fractures in premenopausal women or in men (Moyer & U.S. Preventive Services Task Force*, 2013).

CONCLUSIONS

An inevitable consequence of advancing age is the gradual loss of muscle mass and strength, termed sarcopenia, frequently associated with a parallel increase in fat mass. This geriatric condition has known negative impacts on metabolic health, and in later life, the ability to perform everyday activities (Witard, McGlory, Hamilton, & Phillips, 2016).

This review highlights the major benefits of physical activity in the elderly in terms of body composition and metabolism. Active elderly subjects show a slower "aging clock" and a lighter burden of chronic morbidity. Aerobic exercises help to raise heart and lung efficiency and to increase cardiovascular fitness and endurance, while resistance training promotes an increase in muscle mass and bone density. Achieving these goals represents the first step of a realistic strategy for maintaining functional status and independence.

International guideline recommendations suggest that older people should perform at least 150 minutes of moderate physical activity per week and should be less sedentary in order to achieve health benefits (World Health Organization, 2010). Active elderly in particular, seems to develop a "successful aging" compared to sedentary ones.

Developing simple lifestyle interventions and safe, effective and sustainable ways to promote physical activity, aimed to preserve muscle mass and strength with advancing age, is crucial for the care of patients in mid-life and beyond.

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BENEFITS OF AEROBIC EXERCISE TRAINING WITH RECOMMENDATIONS FOR HEALTHY AGING

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ABSTRACT

The purpose of this articlet is to provide an overview of the importance of aerobic exercise and its characteristics for healthy aging. The first section briefly reviews the effects of aging on maximal aerobic power; Section 2 considers the effects of aerobic exercise training, and Section 3 summarizes the recommendations and some limitations of the current guidelines for aerobic exercise training. Physical activity cannot stop the biological processes; however, there is evidence that regular aerobic exercise can minimize the physiological effects of an otherwise sedentary lifestyle and increase active life expectancy by limiting the development and progression of chronic disease and disability conditions. The use of moderately standardized guidelines for exercise prescription resulted in safe and effective impact on health-related outcomes.

Keywords: *aerobic exercise; physical activity; training.*

POZITIVNI UČINKI AEROBNE VADBE S PRIPOROČILI ZA ZDRAVO STARANJE

IZVLEČEK

Namen tega članka je podati pregled razpoložljivih informacij o pomenu aerobne vadbe in njenih lastnosti za zdravo staranje. V prvem delu so na kratko predstavljeni učinki staranja na maksimalno aerobno moč. Drugi del obravnava učinke aerobne vadbe, medtem ko so v tretjem delu povzeta priporočila ter nekatere omejitve trenutno veljavnih smernic za aerobno vadbo. Telesna aktivnost ne more ustaviti bioloških procesov. Kljub temu pa je na voljo precej dokazov, da redna aerobna vadba zmanjšuje fiziološke učinke sicer sedentarnega življenjskega sloga ter obenem podaljšuje pričakovano življenjsko dobo s tem, ko omejuje nastanek in razvoj kroničnih bolezni ter pogojev invalidnosti. Uporaba zmernih standardiziranih smernic pri predpisovanju telesne vadbe, se odraža neposredno v varnih in učinkovitih vplivih na zdravje in z zdravjem povezana pričakovanja.

***Ključne besede**: aerobna vadba, telesna aktivnost, vadba.*

INTRODUCTION

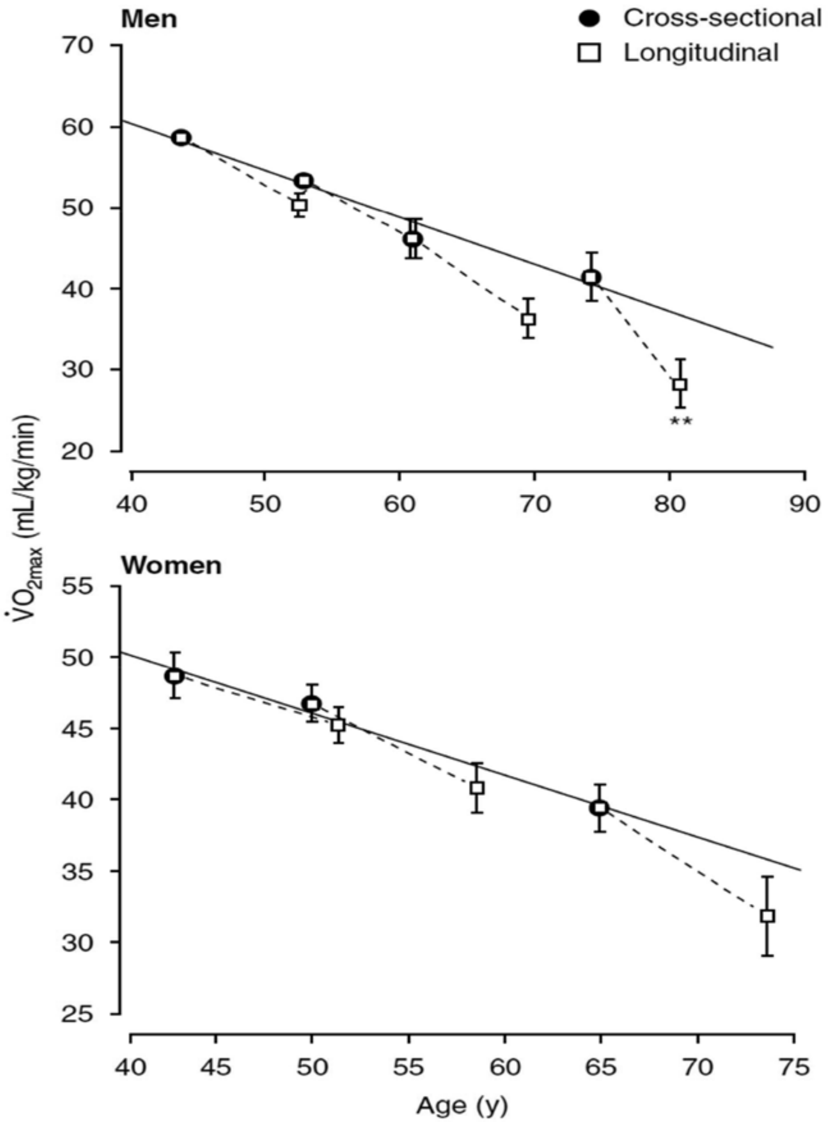
The most widely used terms of maximal aerobic functional power are peak and maximum oxygen uptake (V’O_2peak and V’O_2max). Both terms are often used as though they are synonymous, but there are important distinctions to be made between them (Whipp, Davis, Torres, & Wasserman, 1981). While the V’O_2peak is easier to define and determine, its relevance to physiological and patho-physiological functioning is less secure. It is, simply, the highest value of V’O_2 attained on the particular test, most commonly an incremental or other high-intensity test designed to bring the subject to the limit of tolerance – neglecting considerations of what time, or breath-number, frame of reference is chosen for the determination. Unfortunately, it is the highest value achieved regardless of the subject’s effort. And so while it defines the highest V’O_2 that was attained during the test it does not necessarily define the highest value attainable by the subject. This value is the V’O_2max: a term introduced by Hill and Lupton in 1923 (Hill & Lupton, 1923) as “the oxygen intake during an exercise intensity at which actual oxygen intake reaches a maximum beyond which no increase in effort can raise it”; its rigorous determination depends on a particular criterion having been met. Considering this, the demonstration V’O_2 does not continue to increase, or only to increase by a trivially-small amount, despite further increases in work rate “involving a large proportion of muscle mass” i.e., a V’O_2 “plateau” shows results when V’O_2 is plotted as a function of work rate.

Cross-sectional studies show that V'O_2peak typically declines 6 – 10 % per age decade in healthy men and women (Talbot, Metter, & Fleg, 2000; Wilson & Tanaka, 2000; Aspenes et al., 2011). However, the older individuals included in these studies conceivably presented favourable genetics as well as lifestyle differences, thus limiting the generalization of findings for their age peers. In a longitudinal study, a decline in peak V'O_2 of 20 – 25 % per decade was shown in 70+ healthy elderly subjects (Fig. 1) (Fleg et al., 2005). Hawkins et al. (Hawkins, Marcell, Victoria Jaque, & Wiswell, 2001) also subdivided the sample of master athletes by age group and found that loss rates in maximal oxygen uptake (V'O_2max) increased with age, from rates similar to those reported for sedentary subjects in the younger master athletes to rates four times greater than sedentary subjects in the older master athletes (Figure 1). The declines of 20 – 30 % per decade most likely reflect the periods of rapid decline associated with reductions in physical activity and exercise. These findings suggest that either accelerates the reduction in V'O_2max or increases the difficulty of maintaining training.

Decline in V'O_2max can be attributed to age-related reductions in both maximal cardiac output (Q'$_{max}$) and maximal arteriovenous oxygen (a-v O_2) difference in sedentary subjects, while in older endurance-trained individuals only the reduction of Q'max might explain the age-induced decline in maximal aerobic power (Ogawa et al., 1992). Maximal heart rate decreases about six to ten beats per minute per decade, and is responsible for much of the age-associated decrease in Q'max (Hawkins et al., 2001). However, a reduction in stroke volume during maximal exercise in older adults also contributes to the decline in cardiac output (Hagberg et al., 1985). In addition, left ventricular ejection fraction appears to be reduced in older adults during maximal exercise compared to young adults (Taylor & Groeller, 2008; Thomas, Paterson, Cunningham, McLellan, & Kostuk, 1993). Decreases in vascular capacity and local blood flow regulation, along with a decline in muscle oxidative capacity contribute to the overall reduction in maximal a-v O_2 difference observed with age (Sagiv, Goldhammer, Ben-Sira, & Amir, 2010; Toda, 2012). Coupled with poor oxygen delivery mechanisms, mitochondrial alterations also lead to a reduction in maximal capacity to utilize oxygen at the level of active skeletal muscle. For a submaximal exercise bout, cardiac output is lower in older adults, while a-v O2 difference may tend to increase as a compensatory response to maintain V'O_2. A reduction in stroke volume appears to be the major factor responsible for the lower cardiac output observed during submaximal exercise (Ogawa et al., 1992). Blood pressure is also higher in absolute as well as relative work rates in older adults as compared to younger adults. In addition, total peripheral resistance is generally higher in older adults for a given exercise intensity (Ogawa et al., 1992).

Although reduced physical activity with age contributes to decrease the maximal aerobic power, similar rates of decline are observed with age even among highly active individuals. However, the V'O_2peak of such athletic persons is substantially higher than that of their age peers (Fleg et al., 1994). Moreover, the cardiovascular (CV) system remains fully adaptable to training at any age (Kohrt et al., 1991) with relative increases in V'O_2max in adults of any age equivalent to those seen in young individuals. Given the effect of cardio vascular exercise training and greater fitness on CV disease

Figure 1: Cross-sectional versus longitudinal comparison of loss rates in maximal oxygen consumption (V'O2max) [mL/kg/min] in men and women master athletes (adapted from Hawkins & Wiswell, 2003).



**: Significantly different rate of loss compared with cross-sectional.

risk factors (Kelley & Sharpe Kelley, 2001), mortality, and all-cause mortality (Myers et al., 2002), recommending aerobic activity to adults of all ages would seem prudent (Balady, 2002).

EFFECTS OF AEROBIC EXERCISE TRAINING

The ability to maintain high aerobic power is a major determinant of an older adult's functional independence. Several observational studies have demonstrated that endurance athletes, even those in their 60s and beyond, maintain a V'O_2peak considerably higher than the one of less active age peers. For example, the V'O_2peak in distance runners aged 60 − 80 years was 30 − 40 % higher than active non-trained age peers in the Baltimore Longitudinal Study of (BLSA). In fact, their aerobic capacity was similar to that of BLSA participants 2 or 3 decades younger (Fleg et al., 1994).

As well, data from the Heritage Family Study suggest that genetics explains 47 % of the V'O_2peak response to 20 weeks of aerobic exercise training after adjustment for age, sex, baseline V'O_2peak, and baseline body mass and composition (Bouchard, 2012). Additionally, multiple studies have documented training-induced increases of 10 − 25 % in V'O_2peak among adults in their 60s to 80s, and these increases are similar to those in younger adults (Vaitkevicius et al., 2002). A meta-analysis of training studies in persons aged 60 and older found a mean increase in V'O_2peak of 16% (Huang, Gibson, Tran, & Osness, 2005). In general, higher intensity training and longer exercise duration elicited greater improvement.

In addition to its beneficial effects on aerobic capacity, exercise training produces multiple benefits that reduce risk factors for CV disease.

Hypertension

Hypertension is defined by a systolic blood pressure (BP) ≥140 mmHg and / or diastolic BP ≥90 mmHg, and it represents the leading risk factor for global burden of disease and mortality (Roger et al., 2012). High blood pressure contributes to 7.0 % of disability-adjusted life-years and 9.4 million deaths. Also, the estimated number of adults with hypertension will be increased to 1.56 billion by 2025 (Hu et al., 2016). The prevalence of hypertension increases with age and it represents a risk factor for the most common causes of morbidity and mortality in older age such as stroke, ischemic heart disease, heart failure and coronary events (Lloyd-Sherlock, Beard, Minicuci, Ebrahim, & Chatterji, 2014).

In both younger and older persons with hypertension, regular aerobic exercise reduces BP. Important mechanisms contributing to exercise-related BP reduction include a decrease in aortic stiffness and enhanced flow-mediated arterial dilation due to increased nitric oxide release from endothelial cells lining these blood vessels (DeSouza et al., 2000). The reductions in BP from aerobic exercise are often similar to those induced by a single antihypertensive drug. Mean BP reduction in a large meta-analysis averaged 3.8-2.6 mmHg (Whelton, Chin, Xin, & He, 2002). It is important to point out that lower intensity exercise equivalent to brisk walking demonstrated BP reductions similar to that of more intensive training in older hypertensive adults (Hagberg, Montain, Martin, & Ehsani, 1989).

Dyslipidemia

Abnormal blood lipids are powerful risk factors for CV events in older adults. Aerobic exercise training has beneficial effects on these abnormal lipid levels, irrespective of age. In a meta-analysis of aerobic exercise training trials in older adults, significant increases in high density lipoprotein (HDL) or "good" cholesterol averaged 2.5 mg/dl, and reduced total cholesterol / HDL cholesterol ratio were observed, independent of changes in body composition; improvements in blood lipids correlated with increases in V'O_2peak (Kelley, Kelley, & Tran, 2005). Weight loss, which is often observed during prolonged training programs, may further improve lipid profile (Katzel et al., 1995).

Glucose Tolerance

Aging is accompanied by reduced insulin sensitivity, which impairs glucose tolerance. This adaptation often results in type 2 diabetes mellitus, which itself is a potent risk factor for atherosclerotic CV disease. Both age-associated increase in body fat and reduced physical activity appear to contribute to the impairment of insulin sensitivity and glucose tolerance in older adults. Thus, it is not surprising that both weight reduction and aerobic exercise training ameliorate these impairments. A 9-month aerobic exercise intervention in 71 obese older men (61 ± 1 years, BMI 30.4 ± 0.4 [mean ± SD]) increased V'O_2peak by a mean of 17 % ($P < 0.001$) and reduced area (under the glycaemic curve) of an oral glucose tolerance test by an equal amount (Katzel et al., 1995).

Bone Density

Reduction in bone density associated with aging occurs in both sexes but accelerates in women after menopause, increasing risk for osteoporotic-related fractures, thus, worsening the quality of life. Bonaiuti et al. (2002) reported an increase in bone mineral density of hips and spine by 1.3 % and 0.9 %, respectively ($P = 0.055$ and $P = 0.011$, respectively), following walking activity. (Hatori et al., 1993) also showed an increase of bone mineral density of 1.1 ± 2.9 % ($P < 0.05$) in postmenopausal healthy women (45 to 67 years) following a 7-month training protocol that consisted of 30 minutes of high-intensity walking performed three times per week. Although estrogen replacement therapy reduces post-menopausal bone loss, the negative cardio vascular effects of estrogen have markedly curtailed its use. It seems worth noting that reduced bone mineral density can represent a potential threat for the health status of an individual. In fact, as mentioned above, reduced bone mineral density can increase the risk of fractures, which affects quality of life and leads to physical inactivity. Fortunately, weight bearing aerobic as well as resistance exercise can increase bone density in older adults by increasing the loading force on bone and stimulating osteoblast activity.

Depression

In a case-control study, the INERHEART study, it was found that psychological factors (e.g. depression, perceived stress and life events) were strong risk factors for myocardial infarction. Also, depression was officially recognized as a CV risk factor following the 2010 Global Burden of Disease Study and other studies (Yusuf et al., 2004; Elderon & Whooley, 2013). While it seems important to reiterate that modifiable health behaviour (e.g. physical inactivity, poor diet, smoking, dyslipidemia and medication non-adherence) are conceivably the most critical mediators for CV disease, the role of depression on CV diseases and physical (in)activity should be considered (Pan, Sun, Okereke, Rexrode, & Hu, 2011; Hamer, 2012). In fact, some studies reported associations between physical inactivity, depression and CV mortality also in the aged population (Win et al., 2011). In particular, the primary finding of Win and colleagues (2011) was that physical inactivity accounted for approximately 25 % of the increased risk of CV mortality due to depression in community-dwelling aged adults. Furthermore, Whooley et al. (2008) showed that, in a batch of outpatients with stable coronary heart disease, physical inactivity explained almost half of the association between depressive symptoms and CV events. Interestingly, in addition to the beneficial effects of cognitive behavioural therapy and antidepressant medication, regular exercise has been shown to reduce depressive symptoms. For example, Lavie and Milani (1995) have shown lower depression scores, reduced anxiety, and improved total quality of life (QOL) after cardiac rehabilitation (CR) in 85+ coronary patients. The program lasted 12 weeks in which 36 exercise and educational sessions were performed. Each session included: i) around 10 minutes of warm-up stretching and calisthenics; ii) 30 to 40 minutes of continuous upright aerobic and dynamic exercise (various combinations of walking, bicycling, jogging, rowing, etc.), along with light isometric exercises (e.g. hand weights); iii) around 10-minute cool-down period of stretching. The exercise intensity was prescribed with the aim of making patients attain approximately 75 % to 85 % of their maximal heart rate, or 10 to 15 beats per minute below the level of any exercise induced myocardial ischemia. The exercise prescriptions were periodically adjusted to guarantee a gradual increase in exercise performance. Moreover, all patients were oftentimes supported by physicians, dieticians, nurses, and exercise physiologists to comply with the exercise program (Lavie & Milani, 1995).

LONG-TERM BENEFITS OF EXERCISE IN OLDER ADULTS

Despite the study results on physiological benefits of exercise in older adults reviewed above, elderly people are more concerned about their functional independence and QOL than laboratory measurements. As noted earlier, the ability to perform daily activities generally requires an aerobic power > 20 mL/kg/min (Cress & Meyer, 2003). Ehsani et al. (2003) reported a $V'O_2$peak of 15.6 ± 2.7 ml/kg/min in frail octogenarians women; similarly, (Ades, Ballor, Ashikaga, Utton, & Nair, 1996) found a peak aerobic power equal to 21.5 ± 1.1 ml / kg / min in old women (70.4±4 years). These findings highlight the fact that older individuals are extremely close to the threshold for loss of independence (Cress & Meyer, 2003). In this population, regular aerobic training may prevent or significantly delay the crossing of "independence threshold"; furthermore ongoing clinical trials are rigorously examining the effects of regular exercise in preserving independence and reducing morbidity and mortality (Mazzeo & Tanaka, 2001).

Older adults are conceivably more concerned about their QOL than their longevity *per se*. Thus, improving and maintaining high QOL assumes great importance in the aged population. Because QOL is adversely impacted by illness and disability, improved physical function might be expected to cause parallel increases in QOL. In HF-ACTION, a trial of supervised aerobic exercise training followed by home exercise in adults with moderate-to-severe CHF, QOL improved significantly with training (Flynn et al., 2009). Similarly Austin, Williams, Ross, Moseley, & Hutchison (2005) observed improved QOL in 200 patients 60 − 89 years old (mean 72 years) after a 24-week program of aerobic exercise plus low-resistance strength training.

Aging is accompanied by an accelerating reduction of functional capacity, best quantified by $V'O_2$max and / or $V'O_2$peak; the degradation of maximal aerobic power is also induced by many comorbidities common to the older individuals (Huggett, Connelly, & Overend, 2005). However, numerous observational and interventional studies have demonstrated the beneficial effects of exercise training in older adults, both in healthy and diseased individuals (Mazzeo & Tanaka, 2001). A major challenge confronting the medical community and society is to increase significantly the participation of the aged population in such activities.

RECOMMENDATIONS FOR AEROBIC EXERCISE TRAINING

Current recommendations for improving CV fitness and reducing disease risks are certainly effective (Chodzko-Zajko et al., 2009), even though further research is needed to identify the minimum effective dosage for intensity and volume to improve $V'O_{2max}$ and reduce disease and mortality outcomes. These recommendations call for 15 – 60 minutes of aerobic activities that include large muscles, rhythmic movement, 3 – 5 days per week, at an intensity equivalent to 40 – 85 % of $V'O_2$max (55-90% HRmax). This means that practicing exercise at high intensity and volume, just like athletes would, is not a prerequisite for significant improvements in CV performance and health status in general.

The practice of light- to moderate-intensity physical activity on a more frequent basis, on the other hand, is identified as a requirement to optimize health by the recent guidelines issued by the Centers for Disease Control and Prevention / American College of Sports Medicine (ACSM) (Pate et al., 1995). This should be the main focus of aerobic exercise prescription for adults of all ages (Balady, 2002). Therefore, the ACSM guidelines should provide the basis for exercise prescription for most adults (Chodzko-Zajko et al., 2009). The current consensus recommendations of the ACSM and American Heart Association (AHA) with respect to the frequency, intensity, and duration of exercise and physical activity for older adults are summarized below.

The ACSM / AHA Physical Activity Recommendations are generally consistent with the 2008 Physical Activity Guidelines for Americans by the Department of Health and Human Services (DHHS) (Physical Activities Guidelines Advisory Committee, 2008), which also recommend 150 minutes / week of physical activity for health benefits. However, the DHHS Guidelines note that additional benefits occur as the amount of physical activity increases through higher intensity, greater frequency, and / or longer duration. The DHHS Physical Activity Guidelines point out that if older adults cannot perform 150 min of moderate-intensity aerobic activity per week because of chronic conditions, they should be as physically active as their abilities and conditions allow.

Main recommendations for aerobic exercise training

Frequency: For moderate-intensity activities, accumulate at least 30 or up to 60 (for greater benefit) minutes / day in bouts of at least 10 min each to total 150 – 300 minutes / week, at least 20 – 30 minutes / day or more of vigorous-intensity activities to total 75 – 150 minutes / week, an equivalent combination of moderate and vigorous activity.

Intensity: On a scale of 0 to 10 for level of physical exertion, 5 to 6 for moderate-intensity and 7 to 8 for vigorous intensity.

Duration: For moderate-intensity activities, accumulate at least 30 minutes / day in bouts of at least 10 min each or at least 20 minutes / day of continuous activity for vigorous-intensity activities.

Type: Any modality that does not impose excessive orthopaedic stress; walking is the most common type of activity. Aquatic exercise and stationary cycle exercise may be advantageous for those with limited tolerance for weight bearing activity.

Despite the extremely favourable health outcomes promoted by the above described physical activity, it may be argued that the proposed exercise guidelines are too generic. The adoption of a common prescription approach may not allow to achieve the full therapeutic potential of physical exercise treatment. In fact, these guidelines do not take into account some crucial aspects of exercise prescription, such as the recovery period among each training session (especially between the ones carried out at high intensity and / or high volume) as well as training periodization and the individualization of training variables (volume, intensity and frequency) based on the individual needs of the older individuals. In particular, it is known that periodized training promotes better physical improvements compared to non-periodized training programs in the healthy population, and this may conceivably be the case also for non-healthy individuals (Issurin, 2010). Another positive aspect of periodization is the reduced risk of overtraining and its side effects (Fry, Morton, & Keast, 1992). ACSM guidelines also reported that exercise and physical activity progression for older adults should be individualized, also using exercise tolerance as an additional criteria (Chodzko-Zajko et al., 2009).

CONCLUSIONS

A body of evidence clearly indicates that involvement in exercise programs induces several benefits for older individuals. These favourable ameliorations involve noteworthy health-related issues including CV disease, metabolic syndrome, diabetes mellitus and osteoporosis. However, not only the cardio-metabolic profile is improved in response to exercise program, indeed, it is reported that physical activity positively affects also more functional benefits that allow for continued independence and the ability to perform daily life activities and reduced cognitive symptoms (e.g. depression and anxiety). As a matter of fact, all these exercise-induced benefits are vanished if the aged population is not involved in a regular physical activity regime. Nowadays, extreme importance is placed on the development of new strategies and educational programs with the aim to inform the older population about the meaningful benefits of regular exercise and increase their involvement, adherence and compliance to such programs.

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EXERCISE EFFECTS ON MUSCLE STEM CELLS

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ABSTRACT

Satellite cells are skeletal muscle stem cells that facilitate muscle repair and regeneration after "damage" which occurs after physiological stimuli: exercise, post-training micro-injuries and electrical stimulation. Exercise stimuli lead to activation and proliferation of these cells from their quiescent state, therefore, increasing cell numbers having the potential to provide additional myonuclei to their parent muscle fibre or return to a quiescent state. Different exercise modalities are the focus of numerous studies on satellite cells activation. An increase in muscle activity augments satellite cells proliferation as well as skeletal muscle mass and function, both in young and elderly.

This review provides an updated view of the contribution of skeletal muscle satellite cells in regulating skeletal muscle mass and the efficiency of the exercise intervention to attenuate the decline in muscle mass.

***Keywords**: Satellite cells, acute and chronic exercises, micro damage, muscle regeneration.*

UČINEK TELESNE AKTIVNOSTI NA SKLETNO-MIŠIČNE STAMINALNE CELICE

IZVLEČEK

Satelitske celice so skeletno-mišične staminalne celice, ki omogočajo popravilo in regeneracijo mišičnih vlaken po mikropoškodbah, kot je telesna aktivnost, trening ali *električna stimulacija. Telesna aktivnost sproži prehod satelitskih celic iz stanja mirovanja v aktivacijo in proliferacijo, kar jim omogoča, da se namnožijo in povečajo svoje število. Novonastale satelitske celice se lahko vežejo na matično mišično vlakno ali preidejo v stanje mirovanja. Veliko študij preučuje, kako različne oblike vadbe vplivajo na aktivacijo satelitskih celic ter njihovo število.*

Znano je, da mišična aktivnost povečuje proliferacijo satelitskih celic, posledično tudi mišično maso in funkcijo, tako pri mladih kakor tudi pri starejših osebah. Pregled literature v tem članku prikazuje posodobljen pogled na vlogo satelitskih celic pri vzdrževanju mišične mase in pomembnost vloge telesno/gibalne aktivnosti v preventivi pred izgubo le-te.

Ključne besede*: Satelitske celice, akutna in kronična vadba, mikro poškodbe, mišična regeneracija*

INTRODUCTION

Skeletal Muscle Regeneration

Skeletal muscle regeneration is a highly integrated process involving the activation of various cellular and molecular responses; skeletal muscle stem cells play a pivotal role in this process.

Adult skeletal muscle is a stable post-mitotic tissue, small daily injury can be repaired without causing inflammatory responses and cell death. Muscle injury such as extensive physical activity is related to myofibre necrosis, inflammatory responses, and activation, differentiation, and fusion of satellite cells. Muscle regeneration includes the above mentioned processes with a new myofibre formation. Mammalian skeletal muscles consist of different multinucleated myofibers, grouped in slow (type 1) and fast types (2A, 2X, and 2B) with different myosin heavy chain (MyHC) composition. Myofibres also differ in their metabolic profile, extending from slow /oxidative to fast / glycolytic (Schiaffino & Reggiani, 2011).

The maintenance of skeletal muscle mass depends on mono nucleated muscle precursors or muscle satellite cells. In addition, pericytes, resident in small vessels of skeletal muscle, contribute to its growth and regeneration during postnatal life (Dellavalle et al., 2011). Satellite cells were identified over 50 years ago through electron microscopy by Mauro in 1961. The satellite cell population varies by age, muscle type, and activity, and are used during muscle regeneration and repair due to their special self-renewal and multi-differentiation capabilities. At birth, satellite cells account for 15 % of the entire myofibre nuclei population (Thornell, Lindström, Renault, Mouly, & Butler-Browne, 2003). That proportion latter decreases to between 1 % and 6 % of total myonuclear content in mature muscle fibres (Roth et al., 2000). The major function of satellite cells is contribution to the maintenance of muscle mass, regeneration, and hypertrophy by differentiating into myocytes during human's lifespan. Satellite cells are normally non-proliferative, mitotically quiescent and they become activated in response to stimuli such as myotrauma upon injury or muscle growth (Bischoff & Heintz, 1994), or when skeletal muscle tissue is heavily used during physical activities such as weight lifting or running.

When skeletal muscle is injured, damaged or exercised, satellite cells are activated from their quiescent state, proliferated and fused into existing fibres to provide new myonuclei or return to quiescence (Dhawan & Rando, 2005). Activation of those cells is not restricted to the site of muscle damage. Satellite cells are activated, migrate and proliferate from different parts of myofibre. However, the number of satellite cells appears to increase in the end part of myofibres, where longitudinal elongation of the skeletal muscle occurs (Yin, Price, & Rudnicki, 2013).

Importantly, satellite cells have a limited capacity of division entering a state of irreversible growth arrest after a finite number of cell division (Chargé & Rudnicki, 2004). The self-renewing proliferation of satellite cells maintains the stem cell population and provides abundant myogenic cells which proliferate, differentiate, and fuse to generate new myofibre formation (Yin et al., 2013). With these additional nuclei, muscle fibres can synthesize more proteins and create more contractile myofilaments (actin and myosin) in skeletal muscle cells (Chargé & Rudnicki, 2004). It is interesting to note that high numbers of satellite cells are found associated to slow-twitch muscle fibres as compared to fast-twitch muscle fibres within the same muscle, as they are regularly going through cell maintenance repair from daily activities (Martin & Lewis 2012). As satellite cells are constantly replenished during lifetime and are essential for muscle fibre maintenance, a decline in number and reduced proliferative capacity of satellite cells and / or their inability to become activated and proliferate upon stimuli might contribute to muscle fibre atrophy observed in the elderly (Bischoff, 1994; Seale & Rudnicki, 2000).

Many researchers are interested in new training programs and exercise for developing skeletal muscle mass. However, the mechanism(s) by which exercise induces skeletal muscle hypertrophy remain poorly understood. Through exercise, the muscular work done against progressively challenging overloads leads to increases in muscle mass.

Individual satellite cells respond to exercise and they are influenced by such factors as training status, age, nutrition, and the intensity and volume of the exercise. Duration, frequency and intensity of exercise are important contributing factors in satellite cells activation. Passive stretching of contracted muscles may cause multiple micro damages, disruption of contractile elements or necrosis. For many of us, this happens after changes in the locomotor behaviour by severe onset of exercise. Also after 20 minutes of stepping up and down induces a remarkable increase in muscle-derived proteins in plasma, which reflects some muscle damage (Wernig, 2003). It has been shown that the intensity of the exercise is an important factor in satellite cells activation and muscle regeneration (Martin & Lewis, 2012). Similarly, other researchers have demonstrated that an increase in satellite cell content depends on the intensity rather than the duration of exercise (Bazgir, Fathi, Rezazadeh Valojerdi, Mozdziak, & Asgari, 2017). Intensive exercise such as resistance training bout induces damage to the muscle fibres, causing activation and proliferation of satellite cells. This biological effort often leads to increasing in muscle fibre cross-section area or hypertrophy (Bischoff, 1994). On the other hand, various muscle groups and types react differently to intensity and volume of exercise.

During muscle hypertrophy, muscle fibre size appears to be related to the size of the myonuclear domain, defined as the amount of cytoplasm within a muscle fibre controlled by single myonuclei (Hall & Ralston, 1989). Protein synthesis of single myonuclei is confined in myonuclear domain. The amount of cytoplasm controlled by each myonuclei in adult muscle fibre is relatively constant, thus supporting the theory that satellite cells are required for muscle hypertrophy in order to keep the myonuclear domain constant. Considering the presence of other stem cells in skeletal muscle, their contribution in muscle regeneration and hypertrophic growth is possible (Blaauw & Reggiani, 2014).

METHODS

Electronic databases MEDLINE, PubMed, and Science Direct including the articles published up to 2017 were used to search literature sources. Different keywords were used: satellite cells (SC), SC during exercise, skeletal muscle regeneration and hypertrophy, SC during ageing. Based on the keywords and review articles, satellite cells activation and proliferation during exercise was described.

RESULTS

Satellite Cell during Ageing

It is well known that impairments in satellite cell function during aging result in an impaired muscle fibre regenerative response (Sousa & Muñoz-Cánoves, 2016; Snijders & Parise, 2017) leading to the gradual loss of muscle mass and function (sarcopenia) which diminishes muscle recovery after injury in elderly individuals. In many cases this leads to disability and the subsequent loss of independence. A lower number of satellite cell pool and the exhausted proliferative capacity of aged satellite cells may contribute to accelerated loss of skeletal muscle mass during ageing (Renault, Thorne, Eriksson, Butler-Browne, & Mouly, 2002; Sajko et al., 2004; Joanisse, Nederveen, Snijders, McKay, & Parise, 2017). Other possible reasons for impaired muscle recovery in elderly individuals relate to the reduced production of growth factors or affect the e–c (excitation–contraction) coupling mechanism (muscle-contracting mechanism), (Delbono, O'Rourke, & Ettinger, 1995). It has also been demonstrated that inadequate muscle fibre vascularization occurring during ageing process may be an important cause of impaired regulation of satellite cells in older adults (Snijders & Parise, 2017).

A certain number of satellite cells seems to be necessary for muscle regeneration during the ageing process. Bengal's recent review (Bengal, Perdiguero, Serrano, & Muñoz-Cánoves, 2017) has identified the network of cell-intrinsic and cell-extrinsic factors and processes contributing to satellite cells decline during ageing. Most of the studies suggest that the mentioned decline is caused by age-associated extrinsic (environmental changes) and intrinsic mechanisms (DNA damage, oxidative stress). Both mechanisms contribute to muscle stem cell dysfunction. Based on this idea, many studies propose to rejuvenate aged satellite cells to improving muscle repair in the elderly. (Rando & Chang, 2012; Bengal et al., 2017). Other studies proposed that inadequate activation of Notch signalling, necessary for cell proliferation and cell fate determination, contributes to the loss of regenerative proprieties of aged skeletal muscle (Conboy, Conboy, Smythe, & Rando, 2003; Bjornson et al., 2012).

Satellite Cells Activation and Proliferation after Exercise

Resistance and endurance types of exercise training improve muscle mass and strength, and increase the performance capacity in young and elderly. Satellite cells are involved in muscle maturation during postnatal development, regeneration after injury, hypertrophy, hyperplasia and atrophic post-muscle recovery (Dhawan & Rando, 2005; Chargé & Rudnicki, 2004). The increase of satellite cell proliferation and activation takes place after short-term muscle activity (Darr & Schultz, 1987), but the increases in satellite cell numbers only occur after a long term resistance or endurance training (Martin & Lewis, 2012).

Many studies reported the result of a number of satellite cells following exercise in human (Table 1). Satellite cells get activated from their quiescent state and are involved in muscle regeneration after micro-injuries that follow exercise. Satellite cell content and activity after endurance training can be correlated with time and intensity, duration and frequency of exercise. It has been observed that satellite cell content increased after 30 to 155 minutes of moderate to high-intensity endurance exercises (Parise, McKinnell, & Rudnicki, 2008; Van de Vyver & Myburgh, 2012; Bazgir et al., 2017), while no such increase was associated after 30 minutes of low--intensity exercise (Smith, Maxwell, Rodgers, McKee, & Plyley, 2001). These data confirm the role of the intensity of exercise in satellite cell activation and their role in regeneration and muscle repair.

The acute satellite cell response to exercise has been examined in humans using maximal eccentric contractions of the vastus lateralis muscle by isokinetic dynamometry, as eccentric exercise is considered to induce maximal levels of muscle damage (Gibala, MacDougall, Tarnopolsky, Stauber, & Elorriaga, 1995). Indeed, it appears that the satellite cell response to acute exercise in humans occurs during the first 24 hours after eccentric exercise, however, there was a significant increase in satellite cells

Table 1: Summarized studies on human satellite cell numbers following different exercise type (Martin & Lewis, 2012).

Acute/chronic Training	Exercise type	Muscle Analysed	Satellite Cell number	References
Acute exercise training	RT	VL	increase	Crameri et al. 2004
	RT	VL	increase	Dreyer et al. 2006
	RT	VL	increase	O'Reilly et al. 2008
	RT	VL	increase	McKay et al. 2009
	ES	VL	increase	Mackey et al. 2012
			increase	Mackey et al. 2009
Chronic training	RT	VL	increase	Roth et al. 2001
	RT	VL	# increase	Petrella et al. 2008
	RT	VL	increase	Mackey et al. 2010
	RT	VL	increase	Kadi et al. 2004
	RT	VL	increase	Mackey et al., 2007
	RT	VL	increase	Verdijk et al. 2009
	RT	TR	increase	Kadi and Thornell, 2000
	RT, ET	EDL, VL	increase	Verney et al. 2008
	ET	Pl	* increase	Kurosaka et al. 2011
	ET	VL	increase	Charifi et al. 2003
	ET	VL	increase	Shefer et al. 2010

Abbreviations:
RT= Resistance training, ET= Endurance training, EDL= Extensor digitorum longus, ES= Electrical stimulation, VL=Vastus lateralis, Pl= Plantaris, Tr= Trapezius,
* increase of SC number only with high intensity training.
# increase of SC number only seen in individuals who responded most robustly to RT.

over pre-exercise values even at later times (O’Reilly et al., 2008). Therefore, satellite cells get activated and proliferated in 24 hours after exercise, and increase considerably between 72 and 96 hours, thereafter they decline in number. (McKay et al., 2009; O’Reilly et al. 2008).

The research data on humans suggest that resistance and endurance training can increase satellite cell content and activation in response to exercise periods from 9 to 16 weeks. (Kadi, Charifi, Denis, & Lexell, 2004; Petrella, Kim, Mayhew, Cross, & Bamman, 2008; Shefer, Rauner, Yablonka-Reuveni, & Benayahu, 2010). However, it should be noted that the resistance type of training leads to an expansion in satellite cell pool with myonuclear addition (Petrella et al., 2008). Muscle fatigue seems to be a stimulus for activation, proliferation and differentiation of satellite cells. Unfortunately, there is little research available that compares satellite cells quantity in response to endurance exercise and resistance exercise. Verney et al. (2008) observed an increase in satellite cell content in the deltoid (resistance-trained) and VL (endurance-trained) muscles after 14 weeks in elderly individuals. Interestingly, a failure in increasing the satellite cell pool after a training intervention was observed in obese, diabetic population (Snijders, Verdijk, Hansen, Dendale, & van Loon, 2011).

As to animal models, a study on rat soleus muscle showed no increase in satellite cell content after one week of running for 30 minutes per day on treadmill (Smith et al., 2001). While a similar study on mice anterior tibialis muscle reported a significant increase in the satellite cell number and activation (Parise et al., 2008), due to the different composition of muscle fibre type. The study of Smith and Merry (2012) described a six-week resistance type or endurance type of exercise in rats and found no difference between the proportional gains in satellite cell number of the same muscles between groups. These data suggest that endurance and resistance training enhance the satellite cell pool to a similar extent (Marin & Lewis 2012). In addition, a differential response in fibre-type expansion of satellite cells in response to exercise was observed. Fibre-type classification demonstrates that satellite cells are not equally distributed among the various fibre types and muscles. In human studies, no difference has been detected in satellite cell numbers between fibre types in vastus lateralis untrained muscle of young healthy individuals (Kadi et al., 2006; Verdijk et al., 2007; Snijders et al., 2012), while other studies revealed a greater number of satellite cells in type I fibres and, consequently, minor adaptive potential (Martin & Lewis, 2012; Bazgir et al., 2017). It was demonstrated that untrained rodent muscle type I fibres contain a greater number of satellite cells in respect to type II fibres. In response to training, the number of satellite cells in type II fibre was increased, while the same increase was not observed in type I (Verdijk et al., 2009; Smith & Merry, 2012). This process seems sensible as type II fibres give a greater contribution to muscle mass (hypertrophy) and show a higher responsiveness to resistance training (Martin & Lewis 2012; Bazgir et al., 2017). All these data underline the need of future investigation because the mechanisms of exercise-induced satellite cells activation are not completely understood.

CONCLUSION

The maintenance of skeletal muscle mass and regenerative capacity depends on a functional pool of muscle satellite cells. A loss of skeletal muscle satellite cells and defects in their activity are associated with a variety of neuromuscular and other disorders which lead to muscle atrophy.

Exercise training has been successfully applied to augment satellite cells muscle mass and improve muscle function also in elderly. It has been demonstrated that satellite cell activation after exercise together with adequate nutrition are the most effective countermeasures for ageing sarcopenia.

Modalities of exercise, intensity, duration and frequency are correlated with satellite cell content. They are activated and proliferate after acute exercise training, and are increased in number after resistance and endurance training. There are still some discrepancies between the role of volume and the intensity of exercise on satellite cells activation. To clarify the optimal exercise stimuli for satellite cell activation and differentiation, further research is required.

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MEDITERRANEAN HEALTH CENTRE

Institute for Kinesiology Research of the Science and Research Centre Koper
Arena Bonifika, Koper, Slovenia; December 13th, 2017

On December 13th, 2017, Institute for Kinesiology Research of the Science and Research Centre Koper held an opening ceremony of its new Mediterranean Health Centre. The Centre was established with a strong support of the Municipality of Koper and will significantly add to the development of sports activities locally as well as in the wider region. With the cooperation of the team of experts from the nearby Izola General Hospital, the main purpose of the Centre is to give support in rehabilitation after sports injuries.

A solemn opening ceremony was introduced with a speech held by the Head of the Institute for Kinesiology Research of the ZRS Koper, Assoc. Prof. Boštjan Šimunič, Ph.D., who summarized the past achievements of the Institute. Prof. Rado Pišot, Ph.D., Director of ZRS Koper, further explained the planned purposes of the new Mediterranean Health Centre premises. He pointed out the Center's general support of active healthy living and of the widely acknowledged Mediterranean lifestyle. Its main focus is going to be on functional diagnostics, development and planning of modern and holistic exercise programs, rehabilitation programs designed to the specific individual needs, thus, providing safe and quick return of active sportsmen and women to their activities and designing healthy ergonomic working conditions.

The Municipality of Koper has been a strong supporter of the Centre since the beginning. As the Mayor, Mr. Boris Popovič put it, it represents an important step towards the realization of a long-time ambition of the Municipality of Koper to become an important destination of sports tourism. The support given to the opening of the Medi-


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terranean Health Centre was also expressed by the President of the Slovenian Olympic Committee Mr. Bogdan Gabrovec, and the Head of the Institute of Sports Medicine of the Faculty of Medicine of the University of Maribor, Assoc. Prof. Matjaž Vogrin, M.D., Ph.D.

With this even, the Institute for Kinesiology Research of the ZRS Koper rounded up the year 2017 that can be marked as a successful one. It was the year in which the Institute obtained two important and worldwide acknowledged accreditations, namely a FIFA Medical Centre of Excellence (in collaboration with the Institute of Sports Medicine of the Faculty of Medicine of the University of Maribor, the Brdo National Football Centre and Maribor University Medical Centre) and an Olympic Sports Medicine Centre (in collaboration with Izola General Hospital), the latter providing sportsmen with fast and efficient injury diagnostics and first aid.

The Centre opening earned special acknowledgement by the President of UEFA, Mr. Aleksander Čeferin, who sent his congratulations to the entire team of the Institute for Kinesiology Research of the ZRS Koper.

Matej Kleva

MEDITERANSKI CENTER ZDRAVJA

Inštitut za kineziološke raziskave, Znanstveno-raziskovalno središče Koper
Arena Bonifika, Koper, 13. December 2017

V koprski Areni Bonifika je 13. decembra 2017 Inštitut za kineziološke raziskave Znanstveno-raziskovalnega središča Koper (ZRS) slovesno odprl prostore Mediteranskega centra zdravja. Center, ki je zaživel ob podpori Mestne občine Koper, bo pomembno prispeval k razvoju športa v lokalnem okolju in širše, v prvi vrsti pa bo ob sodelovanju strokovnjakov Splošne bolnišnice Izola (SBI) namenjen rehabilitaciji posameznikov po športnih poškodbah.

Svečana otvoritev se je pričela z uvodnim pozdravom predstojnika Inštituta za kineziološke raziskave, ZRS Koper, dr. Boštjana Šimuniča, ki je povzel vse dosedanje dosežke Inštituta, ki ga vodi. Direktor ZRS Koper, prof. Rado Pišot, je v nadaljevanju pojasnil, da bodo novi prostori Mediteranskega centra zdravja namenjeni vsesplošni podpori zdravju, ki bo slonela na mediteranskem življenjskem slogu s svetovno priznanim znakom odličnosti. Aktivnosti centra bodo osredotočene na funkcionalno diagnostiko, razvoj in načrtovanje sodobnih in celostnih programov vadbe in rehabilitacije po meri posameznikov, z zagotavljanjem varnega vračanja po poškodbah pa tudi načrtovanju zdravega delovnega okolja.

Odprtje centra je omogočila Mestna občina Koper, saj sodeč po besedah župana, gospoda Borisa Popoviča, Mediteranski center zdravja predstavlja pomemben korak k uresničitvi dolgoletnih prizadevanj občine, da bi Koper postal tudi športno-turistični center. Podporo odprtju Mediteranskega centra zdravja sta izrazila tudi predsednik Olimpijskega komiteja Slovenije, gospod Bogdan Gabrovec in predstojnik Inštituta za športno medicino Medicinske fakultete Univerze v Mariboru, dr. Matjaž Vogrin.


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Inštitut za kineziološke raziskave ZRS Koper, je s tem dogodkom zaključil uspešno leto 2017, v katerem je pridobil dve pomembni mednarodni akreditaciji – FIFA medicinski center odličnosti (v sodelovanju z Inštitutom za športno medicino Medicinske fakultete v Mariboru, Centrom NZS Brdo in UKC Maribor) in Olimpijski referenčni športno-medicinski center (v sodelovanju s Splošno bolnišnico Izola), v katerem bodo športniki po diagnostiki dobili hitro pomoč. Obe akreditaciji sta svoj sedež dobili v Mediteranskem centru zdravja. Hkrati se s tem imenovanjem zaokrožuje slovenska mreža referenčnih medicinskih centrov.

Ob odprtju Mediteranskega centra zdravja je čestitke Inštitutu za kineziološke raziskave ZRS Koper poslal tudi predsednik UEFE, gospod Aleksander Čeferin.

Matej Kleva

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Conference proceeding contributions

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