

Relationship Of Menstrual Cycle Disorder With Bone Mineral Density In Adolescent Female Athlete

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Abstract

Purpose: The purpose of this study is to analyze the relationship of menstrual cycle disorder with bone mineral density in adolescent female athlete

Methodology: The study was conducted on May-July 2019 at BPPLP in Central Java Province, Salatiga Athletics Club, Athletics and Swimming Club Semarang State University. This study used cross sectional design, 70 subjects aged 13-21 years were selected by purposive sampling method. The data included height, weight, body fat percentage measured by microtoise and Bioelectrical Impedance Analyzer, menstrual cycle history and food intake obtained through questionnaire interview and Semi Quantitative Food Frequency Questionnaire, physical activity (current bone physical activity/cBPA, past bone physical activity/pBPA, total bone physical activity/tBPA) measured by Bone-specific Physical Activity Questionnaire and bone mineral density measured by Quantitative Ultrasound method. Data were analyzed by chi-square and binnary logistic regression test.

Results: All of subjects had normal bone mineral density, 21.4% of subjects had menstrual cycle disorder. Menstrual cycle disorder, body fat percentage, cBPA, and nutrition intakes (energy, protein, calcium, vitamin D, magnesium, potassium, sodium) had no relation with bone mineral density, but intake phosphorus, pBPA and tBPA had relation with bone mineral density. Regression analysis showed that pBPA had the strongest influence with bone mineral density (p=0.032).

Applications/Originality/Value: This study contributes to solving nutrition problem in adolescent female athlete and as a basis for intervention in the problem of female athlete triad.

INTRODUCTION

Participation of female athlete in recent years has increased steadily. Statistic of The International Olympic Committee shows that as many as 22 female athletes (2,2%), since Paris Olympic in 1900, were allowed to participate in Olympic Championship for the first time. The participation has increased steadily up to 5059 female athletes (45%) participating in the Rio de Janeiro Olympic 2016.(International Olympic Committee, 2018) The female athlete participation increase that is also occurred in adolescent athlete is in the Summer Youth Olympic Games. Since the first championship that was held in 2010 the participation of 1682 athletes (47,2%) increase up to 1851 (49,3%) in the 2014 championship. Moreover, there were equal participation of male and female athletes in 2018 championship which are 2000 adolescenst male athletes (50%) and 2000 adolescenst female athletes (50%) who participated in the championship.(International Olympic Committee, 2018)(International Olympic Committee, 2014)(International Olympic Committee, 2010)

Female athlete who participates actively in sport event has risk suffering various health problems such as malnutrition, anemia, menstrual cycle disorder and decrease bone mineral density. According to several studies the prevalence of adolescent female athlete with osteopenia are arround 0-39,8% and osteoporosis are arround 0-15,4%. As many as 21,8% of 170 adolescent female

athletes have low bone mineral density. Another study conducted on 80 adolescent female athletes show that 13% athletes have Z-score of bone mineral density -1 to -1,9 and 3% of athletes have Z-score of bone mineral density <-2.(J. Thein-nissenbaum & Hammer, 2017)(Gibbs, Williams, & Souza, 2013)(Hoch et al., 2010)(Nichols, Rauh, Lawson, Ji, & Barkai, 2006)

The factor that can affect to bone mineral density is hypoestrogen.(Nattiv et al., 2007) Hypoestrogen can occur due to low energy availability. Athlete with a long duration and high intensity of exercise has risk suffering low energy availability.(Mielgo-ayuso, Luzardo-socorro, & Palacios, 2015) Research shows that 4 of 13 adolescent female sprint athletes have low energy availability and 331 of 833 female athletes in Ireland also have risk suffering low energy availability.(Sygo, Clinic, Coates, Mountjoy, & Burr, 2018)(Logue et al., 2019) Low energy availability can inhibit the secretion of Gonadotropin Releasing Hormone (GnRH) which causes inhibition of Luteizing Hormone (LH) and Folicle Stimulating Hormone (FSH) secretion, that can inhibit ovarian stimulation and decrease estrogen and progesteron production. Prolonged follicular phase due to absence of LH and estrogen causes menstrual cycle disorder.(Mosavat, Mohamed, & Mirsanjari, 2013) Menstrual cycle disorder occur in 2-5% of women population and 79% of them occur in athlete.(Katherine A Beals; Melinda M Manore, 2007) A study of 249 adolescent female athletes show that 20,1% have oligomenorrhea, 7,2% have secondary amenorrhea and 0,8% have primary amenorrhea.(J. M. Thein-nissenbaum, Rauh, Carr, Loud, & Mcguine, 2012)

Athlete with estrogen deficiency hormone has risk suffering low bone mineral density. Estrogen hormone can inhibit bone remodelling that is able to increase bone formation. Bone resorption increase and bone formation decrease can be influenced by several hormones such as cortisol and leptin.(Mosavat et al., 2013)(Matzkin, Curry, & Whitlock, 2010)(Nattiv et al., 2007) Low body weight is related with low leptin level which is leptin secreted by body fat tissue.(Subarjati & Nuryanto, 2015)(Nattiv et al., 2007) Body fat percentage <17% can cause menarche disorder and <22% can cause menstrual disorder.(Osman, Nahas, El-bana, Hamada, & Saab, 2017) Commonly, athlete has lower body fat percentage than non athlete.(Bradbury, Guo, Cairns, & Armstrong, 2017)

Adequacy of several nutritions play a role in maintaining bone mineral density. Calcium intake is useful to maintain the structure and composition of bone mineral density.(Sanders et al., 2009) Absorption of calcium in the body is aided by vitamin D because without vitamin D the body absorbs only 10-15% of calcium intake. Phosphorus intake is important for bone mineralization. Adequate potassium and magnesium intake also play a role in maintaining bone mineral density. Excessive protein and sodium intake can cause urinary calcium, but adequate protein intake also contributes in calcium absorption of intestine.(Fink & Mikesky, 2015)(Whitney E; Rolfes SR, 2011)(Gallagher, 2008)

Athlete with low bone mineral density has risk suffering stress fractur and injury 3,6-4,5 times compared to athlete with normal bone mineral density.(Rauh, Facsm, Nichols, & Barrack, 2010) Short menstrual cycle disorder is related with fertility rate decrease, meanwhile long menstrual cycle disorder is related with anovulation, infertility and miscarriage occurrence.(Gudmundsdottir, Flanders, & Augestad, 2011) Based on this background, the aim of this study is to determine the relationship of menstrual cycle disorder with bone mineral density in adolescent female athlete.

METHODS

This study was an observasional research with cross sectional design and conducted on may-july 2019 at the BPPLP Central Java, Salatiga Athletics Club, Athletics and Swimming Club

Semarang State University The sample were estimated using population proportion formula for calculating sample size, with 66 people minimum sample size. The subjects came from various sport such as karate, taekwondo, swimming, weighlifting, table tennis, pencak silat, volleyball, sepak takraw, judo, fencing, boxing, rowing, beach volleyball and athletics athlete.

The subjects was selected by screening process that obtained 87 subjects after that, subjects were selected again using the purposive sampling method based on inclusion and exclusion criteria as many as 70 subjects were involved in this study. The inclusion criteria were young female athlete BPPLP Central Java, Salatiga Athletics Club, Athletics and Swimming Club Semarang State University aged 13-21 years, had menstruated for at least one year, not have history of osteoporosis and other bone disorders, not consuming alcohol, not taking supplement and drug that can affect to bone mineral density (such as glucocorticoid, anticonvulsant, anticoagulant, etc) routinely for more than 1 month, not consuming caffeine sources (tea, coffee, soft drink) excessively (>3 medium cups/day). The exclusion criteria was subject resigned during this study.

Independent variable in this study is menstrual cycle disorder. Data of menstrual cycle disorder was obtained through history of menstrual cycle recall. Menstrual cycle disorder defined as menstrual disorder that has been experience during 12 past months and characterized by length of distance between the first day of menstrual cycle and the first day of the next menstrual cycle less than 21 days or more than 35 days. Menstrual cycle disorder is divided into three: polymenorrhea (<21 days), oligomenorrhea (>35 days), amenorrhea (>3 months). Dependent variable in this study is bone mineral density. Bone mineral density value was obtained through measurement by densitometer Osteosys Sonost 3000 with Quantitative Ultrasound method. Bone mineral density was categorized as normal if the T-score $1 \leq SD < 2.5$, osteopenia if $-2.5 \leq SD < -1$ and osteoporosis if < -2.5 . Confounding variable in this study are energy, protein, calcium, vitamin D, phosphorus, magnesium, sodium, potassium intake, physical activity and body fat percentage.

Nutrition intake data was obtained through food intake history interview during past month with semi quantitative food frequency questionnaire method and recorded in household size, and then analyzed by nutrisurvey 2007 software. Energy requirement was obtained from Basal Metabolic Rate (BMR) calculation summed with Specific Dynamic Action (SDA), physical activity factor, sport activity factor and growth factor. Protein requirement is 1,2 g/kg BB for endurance sport and 1,4 g/kg BB for strength sport, while the requirement of calcium, vitamin D, phosphorus, potassium, magnesium, and sodium were based on Nutritional Adequacy Rate 2019. Nutrition requirement was categorized to be adequate if the intake 80-100% of needs, excessive if the intake >100% of needs and inadequate if the intake <80% of needs.

Body fat percentage and body weight were measured by Bioelectrical Impedance Analysis (BIA) Tanita DC 360, height was measured by microtoise GEA before measuring body fat percentage, then height data was inputted on the BIA to obtained body fat percentage. Body fat percentage categorized as normal if >22% and low if <22%. Physical activity data obtained through interview by Bone-specific Physical Activity Questionnaire (BPAQ). Physical activity consists of three components are Current Bone-specific Physical Activity (cBPA), Past Bone-specific Physical Activity (pBPA), and total Bone-specific Physical Activity (tBPA). cBPA is data of physical activity that has been routinely carried out for 12 last month. pBPA is data physical activity that has been routinely carried out in the past since the age of one year, while tBPA is the mean of the cBPA and pBPA. Physical activity data was processed and analyzed by BPAQ calculator. The categorization of physical activity was based on the upper and lower quartile value from the result of the physical activity score data.

Univariate data analysis was used to describe the characteristic of variables in this study. Bivariate data analyzed by chi-square test to determine the relationship between variables in this study. Multivariate data analyzed by binary logistic regression test. This study was approved by Health Research Ethic Committee Faculty of Medicine, Diponegoro University/Central General Hospital, dr. Kariadi No.291/EC/KEPK/FK UNDIP/VII/2019.

RESULTS

Subjects Characteristic

Subject characteristic table shows that age range of subject is 13 to 21 years with median age 16,00±2,06 years. Mean of body fat percentage of subject is 26,68±6,44. Minimum value of bone mineral density -0,5 SD while the maximum was 2,8 SD with mean of bone mineral density 1,12±0,89 SD.

Table 1. Subject characteristic

Variable	Min	Max	Mean/Median±SD
Age (year)	13	21	16,00±2,06 ^b
Weight (kg)	41,4	87,6	54,15±8,99 ^b
Height (cm)	150	172	160,30±5,97 ^a
Body Fat Percentage (%)	16	49,8	26,68±6,44 ^a
Bone Mineral Density (SD)	-0,5	2,8	1,12±0,89 ^a
Menarche (year)	10	16	13,00±1,22 ^b
Nutrition Intake			
Energy (kcal)	1086	4428	2232±876 ^a
Protein (g)	32,5	251,4	74,50±40,58 ^b
Calcium (mg)	93	1834,1	437,75±338,24 ^b
Vitamin D (µg)	0	17,9	2,30±3,12 ^b
Phosphorus (mg)	360,5	2725,2	870,60±441,71 ^b
Magnesium (mg)	108,1	819,5	223,85±149,36 ^b
Potassium (mg)	673,4	8418	1961,65±1665,74 ^b
Sodium (mg)	194,8	3471,1	1139,79±653,81 ^a
Physical Activity			
cBPA (score)	0,7656	34,3134	11,02±6,21 ^a
pBPA (score)	0,154	159,62	22,34±26,88 ^b
tBPA (score)	1,372	86,3361	16,95±15,21 ^b

^a= mean, ^b= median

The results of this study indicate all of subjects in this study had normal bone mineral density based on T-score category. Table 2 shows that almost all of subjects (91,4%) had positive T-score and there were 8,6% of subjects that had negative T-score. As many as 21,4% of subjects suffered menstrual cycle disorder. The types of menstrual cycle disorder suffered by subject were polymenorrhea (10%), oligomenorrhea (10%), and secondary amenorrhea (1,4%). Most of subjects (72,9%) had normal body fat percentage. Physical activity includes component of cBPA,

pBPA, tBPA and most of subjects (74,3%) included in normal category, while 25,7% of subjects included low category.

Table 2. Description of bone mineral density, menstrual cycle, body fat percentage and physical activity

Characteristic	n	%
Bone Mineral Density (T-Score)		
T-score negative	6	8,6
T-score positive	64	91,4
Menstrual Cycle		
Menstrual cycle disorder	15	21,4
Normal	55	78,6
Type of Menstrual Cycle		
Polymenorrhea	7	10
Eumenorrhea	55	78,6
Oligomenorrhea	7	10
Amenorrhea	1	1,4
Body Fat Percentage		
Low	19	27,1
Normal	51	72,9
Physical Activity		
cBPA		
Low	18	25,7
Normal	52	74,3
pBPA		
Low	18	25,7
Normal	52	74,3
tBPA		
Low	18	25,7
Normal	52	74,3

Table 3 shows that 65,7% of subjects suffered lack energy intake. Protein intake of most subjects (70%) included in adequate category and the other subjects included in inadequate category. Calcium intake in almost all of subjects (92,9,%) were inadequate intake. Moreover, 94,3% of subjects suffered vitamin D deficiency. Phosphorus intake more than half of subjects (58,6%) included in inadequate category. Magnesium intake which is 55,7% of subjects were adequate. Most of subjects potassium intake (87,1%) were inadequate and 69,2% of subjects also suffered sodium deficiency.

Table 3. Description of nutrition intake

Characteristic	n	%
Energi Intake		
Inadequate	46	65,7
Adequate	24	34,3
Protein Intake		
Inadequate	21	30
Adequate	49	70
Calcium Intake		
Inadequate	65	92,9
Adequate	5	7,1
Vitamin D Intake		
Inadequate	66	94,3
Adequate	4	5,7
Phosphorus Intake		
Inadequate	41	58,6
Adequate	29	44,1
Magnesium Intake		
Inadequate	31	44,3
Adequate	39	55,7
Potassium Intake		
Inadequate	61	87,1
Adequate	9	12,5
Sodium Intake		
Inadequate	44	62,9
Adequate	26	37,1

Relationship of Menstrual Cycle Disorder, Body Fat Percentage, Physical Activity and Nutrition Intake with Bone Mineral Density in Adolescent Female Athlete

Bivariate analysis of this study used chi-square test. Each variable were grouped into two categories for the purposes of this analysis. Table 4 shows that there was no significant relationship between menstrual cycle disorder and bone mineral density ($p=0,619$), beside there were no significant relationship between body fat percentage, current physical activity/cBPA, nutrition intake (energy, protein, calcium, vitamin D, magnesium, potassium and sodium) with bone mineral density ($p>0,05$). Table 4 also shows that there were significant relationship between phosphorus intake, past physical activity/pBPA and total bone physical activity/tBPA with bone mineral density.

Table 4. Relationship of menstrual cycle disorder, body fat percentage, Physical activity and nutrition intake with bone mineral density

		Bone Mineral Density				p
		Negative		Positive		
		n	%	n	%	
Menstrual Cycle	Menstrual cycle disorder	1	6,7	14	93,3	0,619
	Normal	5	9,1	50	90,9	
Body Fat Percentage	Low	2	10,5	17	89,5	0,522
	Normal	4	7,8	47	92,8	
Physical Activity						
cBPA	Low	3	16,7	15	83,3	0,172
	Normal	3	5,8	49	94,2	
pBPA	Low	4	22,2	14	77,8	0,034*
	Normal	2	3,8	50	96,2	
tBPAQ	Low	4	22,2	14	77,8	0,034*
	Normal	2	3,8	50	96,2	
Nutrition Intake						
Energy	Inadequate	3	6,5	43	93,5	0,334
	Adequate	3	12,5	21	87,5	
Protein	Inadequate	1	4,8	20	95,2	0,412
	Adequate	5	10,2	64	91,4	
Calcium	Inadequate	5	7,7	60	92,3	0,370
	Adequate	1	20	4	80	
Vitamin D	Inadequate	5	7,6	61	92,4	0,307
	Adequate	1	25	3	75	
Phosphorus	Inadequate	1	2,4	40	97,6	0,041*
	Adequate	5	17,2	24	82,8	
Magnesium	Inadequate	2	6,5	29	93,5	0,453
	Adequate	4	10,3	35	89,7	
Potassium	Inadequate	4	6,6	57	93,4	0,168
	Adequate	2	22,2	7	77,8	
Sodium	Inadequate	2	4,5	42	95,5	0,132
	Adequate	4	15,4	22	84,6	

Fisher Exact Test *Significan (p<0,05)

Relationship of Menstrual Cycle Disorder with Bone Mineral Density in Adolescent Female Athlete after Controlling Confounding Variable

The result of the bivariate analysis shows that variable of cBPA, pBPA, tBPA, phosphorus, potassium and sodium intake were variables that are included to criteria for multivariate test (p<0,25). Table 5 shows that low pBPA was a factor that can cause low bone mineral density (p<0,05). The influence of pBPA on the bone mineral density was 7,143 times. Occurrence probability calculation used equation formula $p = 1 / (1 + \exp (-y))$ and showed that chance of low bone mineral density

in subject with low pBPA score is 22,2%, while the chance of low bone mineral density in subject with normal pBPA is 3,8%.

Table 5. The most significantly correlated factor to bone mineral density

Variable	Coefisien	p	RP	IK 95%	
				Min	Max
pBPA	1,966	0,032	7,143	1,183	43,120

Binnary logistic regression with Backward selection method

DISCUSSION

Subject in this study were 70 adolescent female athletes consist of karate, taekwondo, swimming, weighlifting, table tennis, pencak silat, volleyball, sepak takraw, judo, fencing, boxing, rowing, beach volleyball and athletics athlete. Subject is 13-21 years old with median age $16 \pm 2,06$ years. The result shows that all of subjects had normal bone mineral density based on T-score, with mean of T-score $1,12 \pm 0,89$ SD. In general athlete has higher bone mineral density than ordinary people. Strong emphasis given during sport or physical activity can affect to athlete bone mass. A study on 318 adolescent athlete groups consist of swimming, soccer, karate, judo, kung fu, basket and 111 adoloscent non athlete groups shows that athlete group had higher mean of bone mineral density of lower limbs and spine than non athlete, whereas at upper spine and whole body density only karate athletes have lower bone mineral desity compared to non athlete group. (Maillane-vanegas et al., 2018)

Most of subject (78,6%) had normal menstrual cycle, while 21,4% subjects suffered menstrual cycle disorder. The types of menstrual cylce disorder suffered by subject consist of polymenorrhea (10%), oligomenorrhea (10%), and secondary amenorrhea (1,4%). Irregular menstrual cycle becomes main symptom of anovulation accompanied by the decrease of ovarian steroid secretion and production. The main causes are hypothalamic amenorrhea function related to the decrease of gonadotropin releasing hormone (GnRH) and dysregulation of hypothalamic ptuitary hipofisis adrenal (HPA). (Bae, Park, & Kwon, 2018) There is GnRH production increase by hypothamalic during puberty that causes *Follicle Stimulating Hormone* (FSH) and *Luteinizing Hormone* (LH) production increase by ptuitary gland and steroid hormone increase by gonad. Luteal phase disorder due to too high exercise in athlete are characterized by low FSH level, the decrease of estrogen-progesteron between luteal and follicular phase, and the last one are short luteal occurrence, no luteal phase or long follicular phase. Abnormal hormone profile in athlete with hypothalamic amenorrhea is related to high physical exercise. This causes low level of GnRH and estrogen. Excessive physical exercise causes hypothalamic function decrease and LH secretion, it can cause menstrual cycle disorder. (Wodarska et al., 2013)

72,9% of subjects had body fat percentage that were included in normal category with mean of percent 26,68%. Body fat percentage category that is used is fat percentage category which can affect regularity of menstrual cycle, because this study aims to determine the menstrual cycle disorder relationship with bone mineral density. Physical activity was measured by *Bone-specific Phisycal Activity Questionnaire* (BPAQ). BPAQ is a questionnaire designed to look at the history of physical activity that is able to predict bone strength for lifetime. All components of physical activity (cBPA, pBPA, dan tBPA) show that the most subject (74,3%) had normal physical activity. Physical activity or exercise can increase bone mechanical strength, as a result there will be bone formation and bone mass increase. (Ooi & Sahrir, 2018)

Protein intake of most subjects (70%) were included adequate whereas energy intake almost all subjects (84,5%) were included inadequate. Micronutrient intake most of subject also included inadequate such as calcium (92,9%), vitamin D (94,3%), phosphorus (58,6%), magnesium (44,3%) potassium (87,1%) and sodium (62,9%). This result was consistent with the study conducted in 40 female athletes aged 18-22 years in America which show that mean of energy, magnesium, calcium, potassium and sodium intake during the beginning season until the end season still below from nutrition requirement recommendation. Athlete's nutrition intake which is less than requirement can be due to lack of nutritional knowledge in choosing and implementing appropriate diet.(Nepocaty, Balilionis, & O'Neal, 2017) Weight limitation program that is implemented in several sports also one of the factors of athlete's intake nutrition.

The result of this study shows that there was no relationship between menstrual cycle disorder and bone mineral density ($p=0,619$). This can be caused T-score bone mineral density of all athletes in this study included as normal. The result in 76 athletes aged 13-29 years in India also show the same result as this study, this is caused bone mineral density also can be influenced by sexual hormone (estrogen, progesterone).(Kaushal & Multani, 2017) Estrogen hormone has important role in the physiology of bone mineral density and bone formation with inhibiting bone resorption and increasing bone formation. Chronic estrogen hormone deficiency can cause the decrease of bone mineral density.(Matzkin et al., 2010) The inhibition of GnRH secretion causes inhibition of FSH and LH from pituitary gland, thus inhibiting ovarian stimulation then decreases estrogen and progesterone production.(Mosavat et al., 2013) This result study shows that 93,33% subjects who suffer menstrual cycle disorder have normal bone mineral density with positive T-score. This can occur because menstrual cycle disorder accumulation that is suffered by subject has not reached chronic hypoestrogen.

This result study shows that body fat percentage did not have significant relationship with bone mineral density. this can be due to the strength of bone particularly determined by dynamic load from muscle pressure not static for example fat mass.(Maspaitella & Dieny, 2012) A Research about the risk factors that are related to bone mineral density of female athlete in Malaysia also shows that body fat percentage did not has significant relationship with bone mineral density, whereas the most influential factors with athlete's bone mineral density in this study were energy expenditure and physical activity. Athlete has intensity and duration of high physical activity/exercise, which is able to provide high mechanical loading in order to result in osteogenic effect that stimulates bone formation.(Hamzah, Hamid, & Ahmad, 2018)

The result cBPA in this study shows that there was no significant relationship with bone mineral density. A research in Korea shows that current activity was related significantly with bone mineral density.(Kim, Jung, Hong, Park, & Choi, 2013) The difference of this result study was caused by the difference of physical activity instrument usage. The study used global physical activity questionnaire instrument developed by WHO, while this study used BPAQ instrument calculating current physical activity for past year.(Kim et al., 2013)(Weeks & Beck, 2008) The result of this study shows that there was significant relationship between the component of past physical activity/pBPA and total physical activity/tBPA with bone mineral density ($p=0,034$). A research of early adult male subject in Korea shows that past activity had significant relationship with bone mineral density and became the most influential factor with bone mineral density. The past physical activity of subject in the study was physical activity in adolescence. Physical activity during adolescence, proven, is important physical activity that can affect bone mineral density of male early adult. Physical exercise in early puberty also give big impact on bone strength in the future.(Kim et al., 2013) Most of subjects in this study had participated to be an athlete

since childhood in order to give a big impact on their bone mineral density. The previous study of female athlete also shows that tBPA had significant relationship with bone mineral density. (Hamzah et al., 2018) tBPA is a mean result of physical activity that had been done. (Weeks & Beck, 2008) Physical activity causes bone having mechanical strength that is given by muscle contraction and gravity loading, so osteocyte has cell deformation that causes bone remodelling. Weight bearing activity 3-5 times/week and resistance exercise 2-3 times/week can maintain bone health. (Goolsby & Boniquit, 2017)

The result of this study shows that only phosphorus intake that was related significantly with bone mineral density ($p=0,041$). The result of female adolescent research in Padang also shows that the result had the same positive correlation. Phosphorus and calcium are in bone and teeth in the form of calcium phosphate that is part of hydroxyapatite crystal which is insoluble and gives bone strength. Phosphorus has a function to support bone growth and replacement. Unbalance ratio between phosphorus and calcium can inhibit calcium absorption. (Noprisanti, Masrul, & Defrin, 2018) Energy, calcium, protein, vitamin D, magnesium, potassium and sodium intake were unrelated with bone mineral density. Another study also shows that there was no significant relationship between vitamin and mineral intake with women calcaneal bone mineral density. This was caused by another excessive mineral and vitamin that have a role in bone matrix formation. (Ramayulis, Pramantara, & Pangastuti, 2011) In contrast to the previous theory, most of nutritional intake of subject are less than requirement but all of subjects had normal bone mineral density. This can be caused by puberty in adolescent getting increase more than 50% of total bone mass. Growth hormone increase, insulin-like growth hormone factor I (IGF I) and estrogen also maximize bone mineral weight. IGF I hormone can increase bone mineral density by increasing work of osteoblast through stimulation of collagen synthesis. Estrogen hormone give positive influence on bone strength. The hormone maintains the balance of reorganization and formation bone by giving effect osteocyte and osteoblast antiapoptotic and effect osteoclast apoptotic. (Goolsby & Boniquit, 2017) Bone mineral density is also highly correlated with heredity/genetic. Genetic factor contributes 50% of total bone density variation in entire population included adolescent. The peak of adolescent bone density 80% is influenced by genetic factor. (Schtscherbyna, Ribeiro, & Farias, 2013) Childhood and adolescence physical activity is also the key in maintain bone mass. (Kim et al., 2013)

pBPA was the most influential factor on bone mineral density. The result of this study shows that most of athletes who had high pBPA score were athletes who had participated in sport for long time. A study shows that ex-female athlete with high physical activity history in the past having significant effect with higher bone mineral density than non athlete. (Andreoli, Celi, Volpe, & Sorge, 2011) Physical activity can affect bone mineral density because physical activity gives mechanical loading of bone. High mechanical loading can increase the process of bone formation, so the bone mineral density increases. Osteocyte can detect and respond to mechanical loading of bone. Osteocyte is the key of bone remodelling process from mechanical loading that occurs by maintaining bone homeostasis through osteoblast and osteoclast. Bone loading increases bone formation and decreases bone resorption by decreasing osteoclast activity. Frequency, level and large amount sufficient mechanical loading are needed to give osteogenesis effect and osteocyte response that needs mechanical loading with high and repetitive intensity. (Hong & Kim, 2018) Most of subjects who had high pBPA score come from team sport games and martial art sport, while athlete who had low pBPA score come from athletics (running) and swimming. This can be caused by team sport games and martial art that have exercise/physical activity combination that have many variation. On the other hand team sport games and martial art it self also have high Ground

Reaction Force (GRF) score. High ground reaction force activity gives positive impact on bone density and can reduce the risk of fracture. (Kelley, Kelley, & Kohrt, 2012)

CONCLUSION

All of subject had normal bone mineral density based on T-score. As many as 21,4% of subjects suffering menstrual cycle disorder. Menstrual cycle disorder, body fat percentage, energy intake, protein, calcium, vitamin D, magnesium, potassium, sodium intake and cBPA had no relationship with bone mineral density in adolescent female athlete, but phosphorus intake, pBPA, tBPA had relationship with bone mineral density in adolescent female athlete. The result of multivariate analysis showed that pBPA is the most influential factor of bone mineral density in adolescent female athlete.

RESEARCH LIMITATION

This study can not directly describe the role of hormonal factor that influence menstrual cycle disorder and bone mineral density because there was no laboratory test.

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REFERENCES

- Andreoli, A., Celi, M., Volpe, S. L., & Sorge, R. (2011). Long-term effect of exercise on bone mineral density and body composition in post-menopausal ex-elite athletes : A retrospective study. *European Journal of Clinical Nutrition*, 66(1), 69–74. <https://doi.org/10.1038/ejcn.2011.104>
- Bae, J., Park, S., & Kwon, J. (2018). Factors associated with menstrual cycle irregularity and menopause. *BMC Women's Health*, 18(36), 1–11.
- Bradbury, K. E., Guo, W., Cairns, B. J., & Armstrong, M. E. G. (2017). Association between physical activity and body fat percentage, with adjustment for BMI: a large cross-sectional analysis of UK Biobank. *BMJ Open*, 7, 1–9. <https://doi.org/10.1136/bmjopen-2016-011843>
- Fink, H. H., & Mikesky, A. E. (2015). *Practical Applications in Sport Nutrition*. (R. Dearborn, Ed.) (4th ed.). USA: Jones & Bartlett Learning.
- Gallagher, M. L. (2008). The Nutrients and Their Metabolism. In L. K. Mahan & S. E. Stump (Eds.), *Krause's Food and Nutrition Therapy* (12th ed., pp. 39–135). Kanada: Elsevier.
- Gibbs, J. C., Williams, N. I., & Souza, M. J. D. E. (2013). Prevalence of Individual and Combined Components of the Female Athlete Triad. *Journal of American College of Sport Medicine*, 985–996. <https://doi.org/10.1249/MSS.0b013e31827e1bdc>
- Goolsby, M. A., & Boniquit, N. (2017). Bone Health in Athletes : The Role of Exercise, Nutrition, and Hormones. *Sport Health*, 9(2), 108–117. <https://doi.org/10.1177/1941738116677732>
- Gudmundsdottir, S. L., Flanders, W. D., & Augestad, L. B. (2011). A longitudinal study of

- physical activity and menstrual cycle characteristics in healthy Norwegian women – The Nord-Trøndelag Health Study. *Norsk Epidemiologi*, 20(2), 163–171.
- Hamzah, S., Hamid, M. S. A., & Ahmad, N. S. (2018). Bone Mineral Density and Associated Risk Factors among Female Athletes : A Cross-2 Sectional Study Bone Mineral Density and Associated Risk Factors among Female Athletes : A Cross-Sectional Study. *Sains Malaysiana*, 47(1), 123–129. <https://doi.org/10.17576/jsm-2018-4701-15>
- Hoch, A. Z., Pajewski, N. M., Moraski, L., Guillermo, F., Wilson, C. R., Hoffmann, R. G., ... Gutterman, D. D. (2010). Prevalence of the Female Athlete Triad in High School Athletes and Sedentary Students. *Clinical Journal of Sport Medicine*, 19(5), 421–428. <https://doi.org/10.1097/JSM.0b013e3181b8c136>.PREVALENCE
- Hong, A. R., & Kim, S. W. (2018). Effect of Resistance Exercise on Bone Health. *Endocrinology and Metabolism*, 33, 435–444.
- International Olympic Committee. (2010). Singapore 2010 Youth Olympic Games Official Report. Retrieved January 28, 2019, from <https://www.olympic.org/singapore-2010>
- International Olympic Committee. (2014). Official Report of the 2nd Summer Youth Olympic Games — Nanjing 2014. Retrieved January 28, 2019, from <https://www.olympic.org/nanjing-2014>
- International Olympic Committee. (2018). Factsheet Women in the Olympic Movement Update–October 2018. Retrieved January 28, 2019, from <https://www.olympic.org/women-in-sport>
- Katherine A Beals; Melinda M Manore. (2007). Nutritional concerns of Female Athletes. In Neil Spuraway; Don Maclaren (Ed.), *Advances in Sport and Exercise Science Series Nutrition and Sport* (pp. 187–206). China: Elsevier.
- Kaushal, H., & Multani, N. K. (2017). Association of Menstrual Function With Bone Mineral Denisty Amongst Punjabi University Female Athletes. *International Journal Physiotherapy*, 4(4), 212–216.
- Kelley, G. A., Kelley, K. S., & Kohrt, W. M. (2012). Effects of Ground and Joint Reaction Force Exercise on Lumbar Spine and Femoral Neck Bone Mineral Density in Postmenopausal Women : A Meta-analysis of Randomized Controlled Trials. *BMC Musculoskeletal Disorders*, 13(1), 1–19. <https://doi.org/10.1186/1471-2474-13-177>
- Kim, J., Jung, M., Hong, Y., Park, J., & Choi, B. (2013). Physical Activity in Adolescence Has a Positive Effect on Bone Mineral Density in Young Men. *Journal of Preventive Medicine and Public Health*, 46, 89–95.
- Logue, D. M., Madigan, S. M., Heinen, M., Delahunt, E., Corish, C. A., Logue, D. M., ... Corish, C. A. (2019). Screening for risk of low energy availability in athletic and recreationally active females in Ireland active females in Ireland. *European Journal of Sport Science*, 19(1), 112–122. <https://doi.org/10.1080/17461391.2018.1526973>
- Maillane-vanegas, S., Agostinete, R. R., Lynch, K. R., Ito, I. H., Luiz-de-marco, R., Rodrigues-junior, M. A., & Turi-lynch, B. C. (2018). Bone Mineral Density and Sports Participation. *Journal of Clinical Densitometry*. <https://doi.org/10.1016/j.jocd.2018.05.041>
- Maspaitella, M. L., & Diény, F. F. (2012). Relationship of Calsium and Phosphorus, Body Mass Index, Body Fat Percentage, Sport Habits, Age of Menarche with Bone Minereal Density in Female Adolescent. *Journal of Nutrition College*, 1(1), 229–240.
- Matzkin, E., Curry, E. J., & Whitlock, K. (2010). Female Athlete Triad : Past , Present and Future. *Journal of the American Academy of Orthopaedic Surgeons*, 23, 424–432.

- Mielgo-ayuso, J., Luzardo-socorro, R., & Palacios, G. (2015). Evaluation of nutritional status and energy expenditure in athletes. *Nutricion Hospitalaria*, *31*, 227–236. <https://doi.org/10.3305/nh.2015.31.sup3.8770>
- Mosavat, M., Mohamed, M., & Mirsanjari, M. O. (2013). Effect of Exercise on Reproductive Hormones in Female Athletes. *International Journal of Sport and Exercise Science*, *5*, 7–12.
- Nattiv, A., Loucks, A. B., Manore, M., Sanborn, C. F., Borgen, J. S., & Warren, M. P. (2007). The Female Athlete Triad. *Journal of the American College of Sport Medicine*, 1867–1882. <https://doi.org/10.1249/mss.0b013e318149f111>
- Nepocaty, S., Balilionis, G., & O’Neal, E. K. (2017). Analysis of Dietary Intake and Body Composition of Female Athletes over a Competitive Season. *Journal of Sport Medicine Science*, *6*(2), 57–65.
- Nichols, J. F., Rauh, M. J., Lawson, M. J., Ji, M., & Barkai, H. (2006). Prevalence of the Female Athlete Triad Syndrome Among High School Athletes. *Journal Archives of Pediatrics & Adolescent Medicine*, *160*, 137–142.
- Noprisanti, Masrul, & Defrin. (2018). Relationship of Protein, Calcium, Phosphorus and Magnesium Intake with Bone Mineral Density in Female Adolescent of SMP NEGERI 5 Padang. *Journal Kesehatan Andalas*, *7*, 29–36.
- Ooi, F. K., & Sahrir, N. A. (2018). Physical Activity , Bone Remodelling and Bone Metabolism Markers. *Journal of Exercise, Sports & Orthopedics*, *5*(2), 3–6.
- Osman, D. A., Nahas, E. M. El, El-bana, R. A., Hamada, H. A., & Saab, I. M. (2017). Bone mineral density and body composition according to menstrual status in female gymnasts : An observational study . *Biomedical Research*, *28*(19), 8390–8396.
- Ramayulis, R., Pramantara, I. D., & Pangastuti, R. (2011). Relationship of Vitamin, Mineral Intake, Ratio of Calcium and Phosphorus with Women Calcaneus Bone Mineral Density. *Jurnal Gizi Klinik Indonesia*, *7*(3), 115122.
- Rauh, M. J., Facsm, À., Nichols, J. F., & Barrack, M. T. (2010). Relationships Among Injury and Disordered Eating, Menstrual Dysfunction, and Low Bone Mineral Density in High School Athletes: A Prospective Study. *Journal of Athlete Training*, *45*(3), 243–252.
- Sanders, K. M., Nowson, C. A., Kotowicz, M. A., Briffa, K., Devine, A., & Reid, I. R. (2009). Calcium and bone health: position statement for the Australian and New Zealand Bone and Mineral Society, Osteoporosis Australia and the Endocrine Society of Australia. *Medical Journal of Australia*, *190*(6), 316–320.
- Schtscherbyna, A., Ribeiro, B. G., & Farias, M. L. F. de. (2013). Bone Health, Bone Mineral Density and Sports Performance. In D. Bagchi, S. NAIR, & C. K. SEN (Eds.), *Nutrition and Enhanced Sports Performences* (pp. 75–80). San Diego: Elsevier.
- Subarjati, A., & Nuryanto. (2015). Relationship Body Mass Index with Leptin Level and adiponectin. *Journal of Nutrition College*, *4*(2), 428–434.
- Sygo, J., Clinic, C., Coates, A. M., Mountjoy, M., & Burr, J. F. (2018). Prevalence of Indicators of Low Energy Availability in Elite Female Sprinters. *International Journal of Sport Nutrition and Exercise Metabolism*, 1–23. <https://doi.org/10.1123/ijnsnem.2017-0397>
- Thein-nissenbaum, J., & Hammer, E. (2017). Treatment strategies for the female athlete triad in the adolescent athlete : current perspectives. *Journal of Sport Medicine*, *8*, 85–95.
- Thein-nissenbaum, J. M., Rauh, M. J., Carr, K. E., Loud, K. J., & Mcguine, T. A. (2012). Menstrual

- Irregularity and Musculoskeletal Injury in Female High School Athletes. *Journal of Athletic Training*, 47(1), 74–82.
- Weeks, B. K., & Beck, B. R. (2008). The BPAQ : A bone-specific physical activity assessment instrument. *Osteoporosis International*, 19, 1567–1577. <https://doi.org/10.1007/s00198-008-0606-2>
- Whitney E; Rolfes SR. (2011). The Fat Soluble Vitamins: A, D, E and K. In Williams P (Ed.), *Understanding Nutrition* (12th ed., pp. 354–380). USA: Wadsworth Cengage Learning.
- Wodarska, M., Witkos, J., Drosdzol-Cop, A., J.Dabrowska, Dabrowska-Galas, M., Hartman, M., ... Skrzypulec-Plinta, V. (2013). Menstrual cycle disorders in female volleyball players. *Journal of Obstetrics and Gynaecology*, 33, 484–488. <https://doi.org/10.3109/01443615.2013.790885>