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The Tromsø Study: *Fit Futures*: a study of Norwegian adolescents' lifestyle and bone health.

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Abstract

Summary Bone mass achievement predicts later fracture risk. This population-based study describes bone mineral density levels (BMD) and associated factors in Norwegian adolescents. Compared with international reference ranges, BMD levels appear higher and physical activity levels are positively associated with BMD.

Purpose Norway has one of the highest reported incidences of osteoporotic fractures. Maximization of peak bone mass may prevent later fractures. This population-based study compared BMD levels of Norwegian adolescents with international reference ranges and explored associated factors.

Methods All first year upper secondary school students, aged 15-19 years in the Tromsø region were invited to the *Fit Futures* study in 2010-2011. Over 90% of the invited participants attended, 508 girls and 530 boys. BMD was measured at total hip, femoral neck and total body by dual x-ray absorptiometry. Lifestyle variables were collected by self-administered questionnaires and interviews. All analyses were performed sex stratified, using linear regression models.

Results In girls mean BMD (SD) was 1.060 (0.124), 1.066 (0.123) and 1.142 (0.077) g/cm² at the total hip, femoral neck and total body respectively. In boys corresponding values were 1.116 (0.147), 1.103 (0.150) and 1.182 (0.097), with significant higher values than the Lunar pediatric reference at 16 years of age In girls, height and self-reported intensive physical activity of more than four hours a week and early sexual maturation were positively associated with BMD at both femoral sites ($p<0.047$). Among boys age, height, body mass index, physical activity and alcohol intake were positively ($p<0.038$), whereas early stages of sexual maturation and smoking was negatively ($p<0.047$) related to BMD.

Conclusions Despite the heavy fracture burden, Norwegian adolescents´ BMD levels are higher than age-matched Caucasians. Physical activity is associated with 1 SD increased BMD levels in those involved in competition or hard training.

Keywords Population-based study – Adolescents – DXA – Sexual maturation – Physical activity – Bone mineral density

Introduction

Osteoporotic fractures constitute a major economic burden for the health care sector and communities in western societies [1-3]. Hip fractures are regarded as the most serious osteoporotic fracture and Norway has one of the highest reported incidences of hip fractures in the world [4]. Bone mineral density (BMD) is a strong predictor of fracture risk [5]. Research on fracture risk has primarily focused on bone mass in the elderly. However, there is a growing awareness of the importance of bone mass during growth, as a compensation for the later inevitable bone loss and prevention of fractures in the elderly [6]. Studies on the achievement of peak bone mass and factors that may influence it are scarce, and few population-based studies have been published. In the framework of the Tromsø Study in Norway we conducted a population-based youth survey, *Fit Futures* (2010-2011) including more than one thousand Norwegians representing both urban and rural district adolescents. The objectives of the present study were to 1) describe BMD levels in Norwegian adolescents; 2) compare these levels with Lunar paediatric reference values, 3) identify predictors which may influence the acquisition of peak bone mass at the femoral sites.

Materials and methods

Study population and design: *Fit Futures*

In 2010-2011 all first year upper secondary school students in the two neighbouring municipalities Tromsø and Balsfjord were invited to participate in the cross sectional study *Fit Futures,* which is an expansion of the Tromsø study [7]. The invited cohort included 1117 participants mainly aged 15 – 19 years, and 1038 adolescents (508 girls and 530 boys) attended the survey providing an attendance rate of 92.9 % (Fig.1). All first year students wishing to participate were accepted, even though they were too old to be ordinary first year students. Since older adolescents were considered to diverge substantially in biology and behaviour from their younger peers, the main analyses are based on data from 469 girls and 492 boys 15-18 years. For the comparison with the Lunar reference, we also included the 18 year old participants, similar to the reference. All examinations were performed during the school day in a well-established research unit

run by dedicated research technicians at the University Hospital of North Norway (UNN). The Norwegian Data Protection Authority and The Regional Committee of Medical and Health Research Ethics (reference number 2009/1282 and 2011/1702/REK nord) approved the study in July 2010 and October 2011, respectively. All participants gave written informed consent; participants aged 16 years and above signed at the study site. Younger participants brought written permission from their guardian.

Measurements

BMD at the total hip was the main outcome variable in the present study. In all participants BMD was measured as g/cm² at the total hip, the femoral neck and total body by dual x-ray absorptiometry (DXA) (GE Lunar prodigy, Lunar Corporation, Madison, WI, USA). Due to quality control and unexpected circumstances 10 scans were excluded. In vivo the densitometer coefficient of variation in percentage (CV%= [SD/mean] x 100) has been estimated to 1.17 % and 1.72 % at the total hip and femoral neck respectively [8], while the CV for total body BMD measurements has not been previously calculated in our lab. For the assessment of BMD we used the Lunar enCORE pediatric reference data [9]. As described in its reference list, this compilation of bone mass values collected with different LUNAR devises, by many researchers, without any intentional standardization, has been described in eight studies published during 1990–2007. The reference data include mean BMD values and SD, for healthy children and adolescents aged 5 to 19 years, living in the USA, Australia and Northern part of Europe, neither suffering from chronic diseases nor using medications known to influence bone mass. The reference values are age adjusted and stratified for sex and ethnicity [9]. The Fit Futures adolescents Z-score was obtained using the Lunar DXA pediatric application, version 1.34.

Height and weight were measured in all participants wearing light clothing without shoes. BMI was calculated as weight (kg) divided by the squared height (m^2), and categorised in accordance with Cole et al´s BMI cut-off points for adolescents and children [10, 11] and the WHO index for those older than 18 years [12].

Questionnaires

Information about past medical history and use of medication, including contraceptives, were obtained through a clinical interview. At the DXA-lab technicians registered ethnicity and excluded participants with possible pregnancy. Information on lifestyle was collected by self-administered electronic questionnaires. Pubertal status in girls was determined through the following question: *"If you have started menstruating, how old were you when you had your first menstruation?"* Age at menarche was categorised into "Early" (<12,5 years at menarche), "Intermediate" (12,5 – 13,9 years) or "Late" (≥ 14 years) sexual maturation. Pubertal status in boys was classified according to Pubertal Developmental Scale (PDS). The boys rated secondary sexual characteristics as growth spurt, pubic hair growth, and changes in voice and facial hair growth on a scale from 1 (have not begun) to 4 (completed). Total score of the four items was summarized and divided in 4, in order to maintain the original range from 1-4. The answers were categorised into "Have not begun" (<2) "Barely started" (2- 2,9), "Underway" (3 – 3.9) or "Completed" (4). Reliability of self-reported menarche age is well established [13], whereas the PDS originally was developed by Petersen et al, and has later been validated [13, 14]. The PDS-questions were included after the survey had started and implied considerable missing values.

The questions on tobacco use comprised smoking and snuffing; each with three alternative answers; daily, sometimes or never. Alcohol consumption included drinking frequency rated from 1 (never) to 5 (four or more times per week). The answers were categorised into never, up to once pr. month, or more than twice a month. The questions on physical activity comprised type and duration of exercise and physical exertion in leisure time in an average week during the last year following the Gothenburg instrument [15]. The alternatives were four; sedentary activities, moderate activity (walking, cycling or exercises at least 4 hours a week), participation in recreational sports at least 4 hours a week or participation in hard training or sports competitions several times each week.

We dichotomized past medical history, use of medications and contraceptives, into disease and medication known for their influence on bone mass, and those not, in concordance with the Lunar reference manual. The contraceptives were categorized into no hormonal contraceptive, oestrogens and progestin, or progestin-only.

Statistical analyses

All statistical analyses were performed sex stratified. Mean and SD were calculated for continuous variables, and categorical variables were described in terms of number and percentage. Sex differences were explored by Student´s *t*-tests and Pearson´s chi-squared test (Table 1). For comparisons of the study cohort's BMD levels with the Lunar pediatric reference at different age groups, we used the sample size independent "One sample *t*-tests for a single group" (Table 2). Due to lack of numbers of observations in the Lunar age groups, calculations of 95% CIs were impossible, therefore only p-values are reported. ANOVA with the Bonferroni correction for multiple comparisons were used to assess differences in mean BMD according to ethnicity, pubertal state, physical activity, other life style variables like smoking, snuffing and alcohol use, chronic diseases, use of medication and hormonal contraceptives. We then performed simple linear regression for BMD levels at the hip, crude and age adjusted (Table 3). Variables contributing at a 10% significance level ($p<0.1$), like age, height, BMI, sexual maturation, physical activity, smoking and alcohol consumption (in boys), together with hormonal contraceptives (in girls), diseases and medication known to affect bone, were used for multiple regressions analyses, modelling to the highest adjusted R^2 (Table 4). Multiple imputations were performed for predictors and outcome variables included in the multivariate models, as these variables were regarded adequate for prediction of missing puberty values. Puberty status in girls and all other variables had less than 3% missing. We considered the main reason for 23% missing in the boys puberty data to be related to the late introduction of the PDS-questions, and we therefore assumed missing at random (MAR). Automatic methods were used to create 5 imputed datasets with pooled parameter estimates reported. Confounding and interaction were explored, and adjustment for the interaction term between BMI and sexual maturation in girls, was included in the femoral neck model. In girls, we also performed analyses stratified on sexual maturation. Residual analyses were used to assess linearity; normal distribution, variance homogeneity and outliers, and no assumptions were harmed. All analyses were performed using the Statistical Package of Social Sciences software (SPSS v. 22) and all values of $p<0.05$ were considered significant.

Results

The study population included 508 girls and 530 boys, the majority aged 15 to 19 years; their characteristics are shown in Table 1. There was no difference in the BMI distribution between the sexes, more than 20 % were classified as overweight or obese for age. Mean age at menarche was 12.95 years (SD 1.19). For most of the boys, sexual maturation was categorized as "Underway". A total of 98 % of the participants answered the questions on lifestyle (smoking, snuffing, alcohol, physical activity). All lifestyle variables, except smoking, were significantly differently distributed between the sexes. The reported mean participation in sports outside school was 4.28 (SD 2.10) hours a week (data not shown).

The mean BMD (SD) was 1.060 (0.124) g/cm² at the total hip, 1.066 (0.123) g/cm² at the femoral neck and 1.142 (0.077) g/cm² at the total body in girls; BMD (SD) levels in boys were 1.116 (0.147), 1.103 (0.150) and 1.182 (0.097) g/cm² at the total hip, femoral neck and total body, respectively. The crude BMD values were significantly higher ($p<0.001$) at all sites in boys versus girls.

Mean Z-score at the total hip, femoral neck and total body were positive at nearly all sites (Fig. 2). For the 16 years old, CI for the difference did not include 0 at any of the three sites in girls, or at the total hip and total body in boys (Fig. 2), which indicates higher BMD levels in the FF1 population. Age stratified BMD levels compared to Lunar paediatric reference values, after excluding participants with comorbidities, use of medication or hormonal contraceptives known to influence bone mass acquisition, are shown in Table 2. Mean BMD values were 1.7 -7.1% higher than the reference, significantly at all three sites in girls and boys 16 years of age ($p<0.001$), at femoral neck in girls aged 17 years ($p= 0.014$), at total hip and total body in boys aged 15 years ($p= 0.005$ and $p<0.001$ respectively), and at total body in boys 17 years of age ($p=0.017$).

The univariate analyses including participants <18 years illustrated in Table 3, showed a positive correlation between age and BMD in boys ($p< 0.001$), which was not present in girls. In both sexes height, weight, BMI, sexual maturation, smoking and physical activity levels were significantly correlated with BMD. Alcohol use was in addition positively correlated with BMD at the femoral sites in boys. Adjusting for age only made minor changes.

In multivariate analyses; the determinants were height, BMI, sexual maturation, physical activity, smoking and alcohol intake, with differently explained variation seen among girls and boys. After imputation of missing values, age became significant in boys ($p<0.038$) on behalf of the association of sexual maturation. Pooled parameter estimates for the imputed models, controlled for comorbidity, medication and hormonal contraceptives known to influence BMD, with the highest adjusted R^2, are displayed in Table 4. In boys; age, height, BMI, together with recreational sport, hard training and alcohol use were positively ($p<0.038$) associated with BMD, whereas smoking and an initial stage of sexual maturation ($p< 0.047$) were associated with lower BMD. Hard training boys had 13 % higher BMD levels at total hip and femoral neck (0.144 and 0.141 g/cm^2 respectively, $p<0001$) compared to their sedentary peers, which equals a BMD difference of almost 1SD. In girls too, height, BMI and higher levels of physical activity were positively ($p<0.047$) associated with BMD, while late puberty indicated lower BMD levels at both femoral sites. There was a significant interaction between BMI and sexual maturation at the femoral neck. Stratified analyses showed that BMI was more strongly associated with femoral neck BMD levels in late sexually maturated girls. For girls in early menarche, BMD increased by 0.007 g/cm^2 ($p= 0.003$) for one unit increase of BMI, for girls in intermediate menarche by 0.009 g/cm^2 ($p<0.001$) and late menarche by 0.014 g/cm^2 ($p< 0.001$) (data not shown). Among hard training girls BMD levels were 11% higher at both sites, i.e. approximately 1 SD (0.117 and 0.123 g/cm^2, $p<0.001$), respectively. Among girls participating in recreational sports, BMD levels at the femoral neck were significantly, but only modestly higher compared to those classified as sedentary.

Discussion

Despite the heavy burden of osteoporotic fractures in elderly Norwegians [4, 16, 17], BMD levels appeared higher in Norwegian adolescents compared to the Lunar pediatric reference data. Height, participation in intensive physical activity of more than four hours a week and early sexual maturation were significantly associated with higher BMD levels at both femoral sites in girls, whereas BMI only at the total hip. The corresponding variables in boys were age, height, BMI, higher physical activity levels and alcohol intake, whereas early stage of sexual maturation and smoking were negatively related to BMD. The highest

physical activity levels were associated with more than 1 SD higher BMD levels; i.e. 11 % increase in girls and 13 % in boys involved in sports competition or hard training. According to Rizzoli et al, a 10 % increase in peak bone mass corresponds to a gain of 1 SD BMD in adulthood, and a fracture risk reduction of up to 50% [6].

The differences observed in mean BMD values between the Lunar population and the *Fit Futures* adolescents could be due to calibration error, different hard-ware devices, operator technique, selection bias, anthropometric differences of the population (secular trends) or true population differences. Our Lunar Prodigy was calibrated daily according to the manufactures quality assurance manual. Different research technicians operated the densitometer, no intra-/inter reliability tests were made, but the technicians were well trained, and had operated the device daily for a long period, which should reduce the risk of systematic error. A former study at the same research unit estimated the densitometer coefficient of variation at the total hip measured in vivo [8]. The differences reported were small and clinically irrelevant, as reported by others [18].

The Lunar reference data were collected in different studies, mainly using old pencil beam scanners (DPX-devises). Previous studies have evaluated relationships between DPX devises and the newer narrow fan bean densitometers (Prodigy). In general, small differences between the two types of absorptiometry were found, and by use of cross-calibration equations and factors, results from the two different devices were considered compatible [19, 20].

Cross-sectional studies are vulnerable to certain biases like non-participation or survivors-bias. In *Fit Futures* approximately 14 % of the eligible population were not invited due to chronic illness, dropouts from school, or because the study team were unable to contact them, another 7% of the invited did not attend the study for unknown reasons. Dropout from school may be associated with an unhealthy life style, and individuals suffering from chronic illness, may also have lower BMD levels [21]. However, a participation rate of 93% is high, and should contribute to robust estimates.

In comparison with the Lunar reference our mean BMD values in girls and boys were significantly higher (1.7-7.1%) at nearly all sites at the age of 16 and 17 years. This is in concordance with Kolle et al, who in a study of 145 Norwegian females aged 13-39 years, showed mean BMD values to be 3.4-5.1% higher than the US/European reference ($p<0.005$) for this age range [22]. DXA- technique tends to underestimate the

true density value for smaller bones and overestimates for larger bones, something that could explain our high BMD levels. Only two studies of the Lunar compilation [23, 24] reported the participants' height and weight according to age groups, so the opportunities for comparison of anthropometric measurements were limited. A study of Norwegian growth curves from 2009 shows that height and BMI have increased during the last 30 years [25]. The mean height of our participants was just below the 50% percentile of the Norwegian growth curves, and the weight somewhat higher. Even though the *Fit Futures* population may be slightly higher and heavier than the Lunar reference, we consider the potential for overestimation due to height to be small, and the difference could more likely be related to weight.

Most studies report that late menarche is associated with lower BMD [26]. In the Lunar reference, the puberty data, if collected, were collected in different ways. It was therefore challenging to explore the influence of puberty in this comparison. This was also the case for confounding factors like diet (calcium and vitamin D), physical activity and unhealthy lifestyle (smoking and alcohol use), which were sparsely described.

There are limited newer population-based studies describing BMD values for our respective age groups. Except for the 17 years old boys and girls in the ALSPAC cohort, reporting higher BMD levels at the hip [27] , most studies report lower BMD values for total hip and total body than *Fit Futures* do [28-32]. After converting Hologic measurements to GE-Lunar [33] mostly all our mean BMD values remained higher, which support our conclusions; the overall tendency of higher *Fit Futures* than Lunar values, may be explained by true population differences.

Participants reporting diseases or medication known to influence bone were few, and when excluded from the analyses, hardly any changes were seen. Even though one third of the girls reported use of hormonal contraceptives, this did not influence bone mass. In our data 4 % were classified as non-white. Exclusion of these participants did not change the results, and these individuals were included in the final analysis. Classification of Sami ethnicity was self-reported based on cultural belonging, and we were therefore unable to report any genetic influence from Sami heritage.

Body weight is the largest single determinant of adult bone mass [34]. In our cohort more than 20% were classified as overweight or obese for age according to Cole et al´s definition of overweight and obesity in childhood [11]. The importance of BMI and weight in adolescence is not clearly understood. Some studies

suggest a negative effect of fat mass during childhood and adolescence; obese children are more prone to falls and to fractures due to lower bone mass relative to body size [35]. Other studies report a positive association between fat mass and bone growth before puberty [36-38]. In girls it seems as puberty attenuate and reverse this effect between fat and bone [36]. This supports our finding; BMI was positively associated with BMD with higher beta values in late sexually matured girls.

Most studies report that late commencement of puberty is associated with lower BMD in both sexes [26, 28, 39]. For girls with intermediate and late sexual maturation, we found a negative association with BMD levels at the femoral neck as described by Heaney 2000; who reported that late menarche is a skeletal disadvantage, and that there is an association between age at menarche and fracture risk. Boys in the beginning of puberty had considerably lower BMD levels at the total hip compared to those with completed sexual maturation, as indicated by Arabi et al [28].

A review from Bieleman et al [40] indicated that more studies have found positive associations between physical activity and bone mass in males than in females at weight bearing anatomical sites, and that low participation in peak strain activities may explain the weaker associations found in females. In our study both boys and girls that reported hard training had more than 1 SD higher BMD at the femoral sites, compared to those who were sedentary. A former Norwegian population-based study reported participation in sports outside school for 16-years-old youth to a mean of 3.72 (SD 2.37) hours a week [41]. Compared to these, our adolescents reported slightly higher physical activity levels (0.56 hours, CI [0.325-0.795]). Despite the lack of evidence of effect of physical activity during adolescence on later fracture risk, it is important to take into account its influence of physical activity practice in later life [40].

Tobacco use has been associated with lower BMD levels in adolescence in some, but not all studies [34]. The effect of smoking is also influenced by dose and duration [42], which can explain diverging study results. Longitudinal studies of adolescent boys and girls in early adulthood have shown adverse effect of smoking [21, 43]. In our study, smoking was negatively associated with BMD at the hip only in boys. Among the *Fit Futures* girls, approximately 5 % reported daily smoking, compared to 15 % in Dorn et al´s study, which can explain the lack of significance in the multivariate model.

In our study there were no significant associations of alcohol on BMD levels in girls, supported also by Dorn et al. On the contrary; in boys, alcohol consuming was significantly and positively associated to BMD

at the hip sites, supported by findings in adults where moderate alcohol consumption appears to be beneficial to men´s bone health [44, 45]. This effect has been suggested by Wosje and Kalkwarf to be linked to the promotion of endogenous estrogen synthesis, and thereby increasing BMD in individuals with low circulating estrogen concentrations [44]. Anyhow; this result must be interpreted carefully, as the questions about alcohol consumption has not been validated for this age group.

The strengths of this study lie in its population-based design, high attendance rate of both sexes, and the representation of urban and rural adolescents, which makes the results highly representative for Norwegian adolescents. The main limitation is the cross-sectional design, which limits causal inferences. The nature of our self-reported questions of physical activity level and sexual maturation (non-differential measurement errors) may weaken the associations. Also the considerable male puberty missing values may cause bias. A comparison of age, weight, height, BMI and BMD measurements, showed that participants with no puberty data were significantly younger (0.3yrs, $p<0.001$), with lower BMD femoral neck (0.034 g/cm^2 $p=0.038$) and total body (0.028 g/cm^2, $p=0.007$) values. Since boys in the beginning of the survey were not offered this question, it is likely that they were younger, and therefore with lower age related BMD values, which supports our assumption of randomly missing puberty data. Information on dietary calcium intake and vitamin D levels would also have strengthened the result.

This study is the first to report BMD levels in Norwegian adolescents. Despite the heavy fracture risk in the Norwegian elderly, there is no reason to believe that mean BMD levels of todays Norwegian adolescents are lower compared to other European populations. Among modifiable factors, BMI and physical activity levels are associated with higher BMD levels in both sexes. The interaction between these two factors should be elaborated further, and the most optimal levels identified. The negative effect of early smoking on male BMD is noted, and the effect of snuffing will be followed carefully. Given the positive association between modest alcohol intake and BMD in males, longitudinal data will be valuable. Intermediate and late sexual maturation were associated with substantial lower BMD levels compared to menarche age <12.5 years. One SD reduction in BMD values can lead to more than a doubled fracture risk in the elderly [6]. Therefore, longitudinal data will be highly important for following the possible adverse effect of late sexual maturity – will it level out?

In conclusion; BMD values from the *Fit Futures* will be a valuable supplement to other population-based reference values for BMD levels at the hip and total body for adolescents 16 -17 years of age. Furthermore, peak bone mass seems to be modifiable by life style factors; their longitudinal effects will be followed in further studies.

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Conflicts of interest

None of the authors had any conflicts of interest to declare with respect to this paper.

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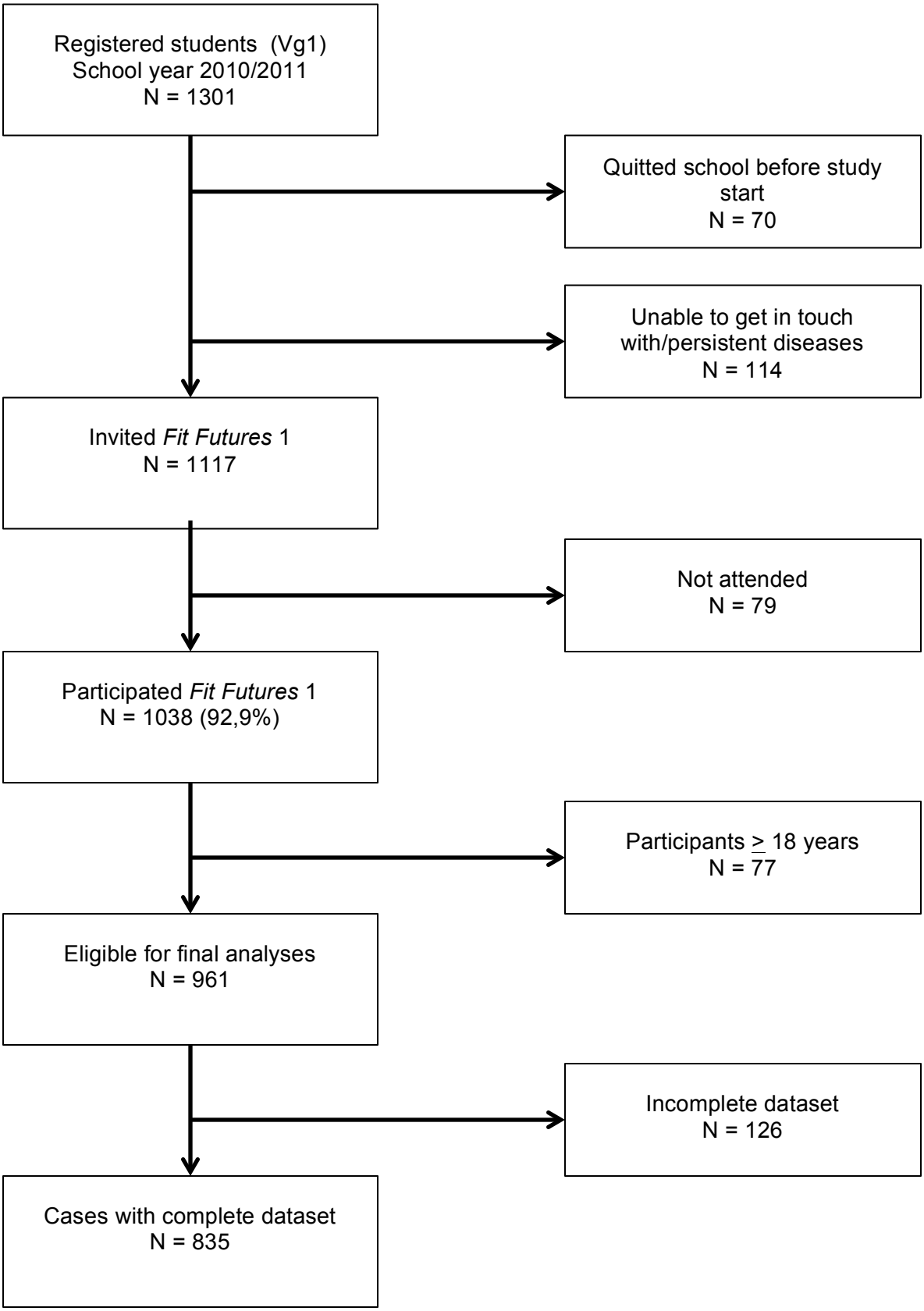
Figure Captions

Fig. 1 Participation in the Tromsø Study: *Fit Futures* 2010-2011

Fig. 2 BMD Z-score for age groups at total hip, femoral neck and total body, in girls and boys, the Tromsø Study: *Fit Futures.* Observations in each age group included, and statistically significant values displayed with filled symbols


Registered students (Vg1)
School year 2010/2011
N = 1301
Quitted school before study start
N = 70
Unable to get in touch with/persistent diseases
N = 114
Invited *Fit Futures* 1
N = 1117
Not attended
N = 79
Participated *Fit Futures* 1
N = 1038 (92,9%)
Participants ≥ 18 years
N = 77
Eligible for final analyses
N = 961
Incomplete dataset
N = 126
Cases with complete dataset
N = 835

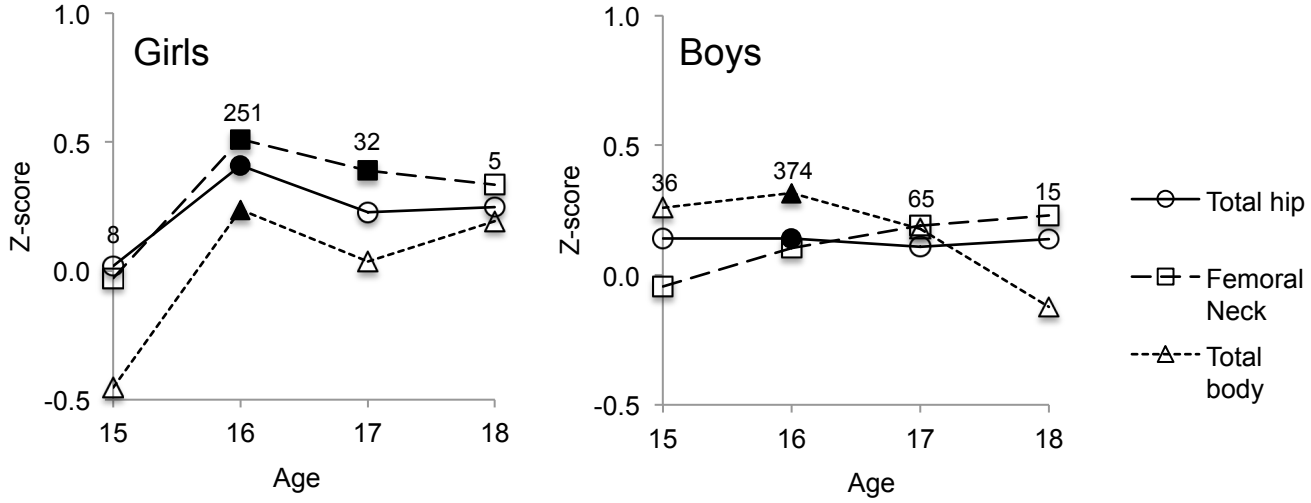


Fig2_090514.xlsx

Table 1 Characteristics of the study participants in The Tromsø Study: *Fit Futures* 2010-2011, n=1038. Mean (SD) of continuous variables and percentage (number of observations) of categorical variables are given for girls and boys respectively.

Characterics	n	GIRLS	n	BOYS	$P_{Equality}$
Age, years	508	16.94 (1.36)	530	16.82 (1.04)	0.111
Height, m	505	1.65 (0.07)	529	1.77 (0.07)	<0.001
Weight, kg	505	61.4 (12.2)	529	70.6 (14.6)	<0.001
BMI, kg/m^2	505	22.6 (4.2)	529	22.5 (4.2)	0.676
BMI class [a]	505		529		0.422
Underweight for age		5.3 (27)		7.5 (40)	
Normal weight for age		71.7 (364)		68.1 (361)	
Overweight for age		15.4 (78)		16.4 (87)	
Obese for age		7.1 (36)		7.7 (41)	
BMD total hip, g/cm^2	502	1.060 (0.124)	526	1.116 (0.147)	<0.001
BMD femur neck, g/cm^2	503	1.066 (0.123)	526	1.103 (0.150)	<0.001
BMD total body, g/cm^2	502	1.142 (0.077)	526	1.182 (0.097)	<0.001
Ethnicity	503		526		0.882
White		94.9 (482)		95.3 (505)	
Other		4.2 (21)		4.0 (21)	
Puberty girls					
Age at menarche	492	12.95 (1.19)			
Sexual maturation	497				
Early (<12,5 yrs)		31.3 (159)			
Intermediate (12.5 – 13.9)		43.7 (222)			
Late (≥ 14)		21.9 (111)			
No menarche yet		1.0 (5)			
Puberty boys					
PDS-status			409		
Completed				7.7 (41)	
Underway				56.2 (298)	
Barely started				13.2 (70)	
Not begun				0 (0)	
Smoking	498		520		0.107
Daily		5.5 (28)		3.8 (20)	
Sometimes		15.9 (81)		20.2 (107)	
No, never		76.6 (389)		74.2 (393)	
Snuffing	499		519		0.002
Daily		18.9 (96)		28.1 (149)	
Sometimes		13.2 (67)		12.1 (64)	
No, never		66.1 (336)		57.7 (306)	
Alcohol	500		518		0.011
Never		23.4 (119)		30.4 (161)	
Up to once pr. month		44.7 (227)		36.4 (193)	
Twice or more pr. month		30.3 (154)		30.9 (164)	

Physical activity	500		520	<0.001
Sedentary		14.8 (75)	29.1 (154)	
Moderate		40.6 (206)	24.9 (132)	
Sports		27.4 (139)	22.6 (120)	
Hard training		15.7 (80)	21.5 (114)	
Chronic or persistent disease	506		525	0.016
Yes		32.5 (165)	27.5 (146)	
1 disease		18.3 (93)	15.7 (83)	
2 diseases		3.9 (20)	3.6 (19)	
≥ 3 diseases		2.8 (14)	0.8 (4)	
Medication daily	506		527	<0.001
Yes		37.0 (188)	26.8 (142)	
1 medication		16.1 (82)	13.8 (73)	
2 medications		12.4 (63)	5.1 (27)	
≥ 3 medications		3.7 (19)	2.3 (12)	
Hormonal contraceptives				
Yes		34.6 (176)		
oestrogen and progestogens		29.3 (149)		
progestogens only		4.3 (22)		

[a] BMI class: Age < 18 years; Coles index according to sex and age, Age ≥ 18 years; WHOs index

Table 2 The Tromsø Study: *Fit Futures* BMD values (g/cm^2) excluded participants using hormonal contraceptives, with diseases[a], or medication[b] known to affect BMD, compared to Lunar Pediatric reference[c], described as mean (SD) at different age groups, sex and sites. Differences displayed in percentage, significant result (p<0.05) in **bold**

	Total hip, mean (SD)				Femoral neck, mean (SD)				Total body, mean (SD)			
GIRLS Age	*Fit Futures*	Lunar	Diff. %	p-value	*Fit Futures*	Lunar	Diff. %	p-value	*Fit Futures*	Lunar	Diff. %	p-value
15	1.012	1.010	0.2	0.963	0.996	1.000	- 0.4	0.920	1.085	1.085	0	1.000
n = 8	(0.120)	(0.130)			(0.109)	(0.135)			(0.080)	(0.084)		
16	1.065	1.010	5.4	**<0.001**	1.071	1.000	7.1	**<0.001**	1.144	1.125	1.7	**<0.001**
n = 251	(0.120)	(0.134)			(0.120)	(0.139)			(0.077)	(0.081)		
17	1.038	1.010	2.8	0.131	1.052	1.000	5.2	**0.014**	1.120	1.125	- 0.4	0.651
n = 32	(0.102)	(0.134)			(0.114)	(0.139)			(0.061)	(0.076)		
18	1.042	1.010	3.2	0.471	1.044	1.000	4.4	0.233	1.147	1.125	2.0	0.444
n =5	(0.090)	(0.131)			(0.070)	(0.135)			(0.058)	(0.070)		
BOYS Age												
15	1.078	1.010	6.7	**0.005**	1.049	1.010	3.9	0.076	1.139	1.070	6.4	**<0.001**
n = 36	(0.135)	(0.131)			(0.128)	(0.124)			(0.086)	(0.089)		
16	1.115	1.065	4.7	**<0.001**	1.101	1.060	3.9	**<0.001**	1.178	1.120	5.2	**<0.001**
n = 374	(0.145)	(0.135)			(0.150)	(0.128)			(0.095)	(0.093)		
17	1.136	1.120	1.4	0.437	1.136	1.110	2.3	0.189	1.200	1.170	2.6	**0.017**
n = 65	(0.165)	(0.137)			(0.158)	(0.131)			(0.099)	(0.096)		
18	1.131	1.120	1.0	0.750	1.133	1.110	2.1	0.520	1.208	1.220	1.0	0.653
n = 15	(0.135)	(0.137)			(0.135)	(0.132)			(0.101)	(0.097)		

[a] Diseases (ICD10) known to affect bone:
E03 Hypothyroidism, E10 Diabetes type I, F50.9 Eating disorders, K90.0 Celiac disease, M13 Arthritis

[b] Medication (ATC) known to affect bone:
D07A Plain corticosteroids, H03A Thyroid preparations, N03A Antiepileptic,
R01AD Corticosteroids, R03BA Glucocorticoids (inhalants), H02A Corticosteroid for systemic use

[c] Lunar enCore, Supplement til pediatrisk referansedata. 1. revision ed: GE Healthcare; 2010-Nov

Table 3 Association between basic characteristics, lifestyle variables and BMD (g/cm^2) at total hip and femoral neck in girls and boys <18 years (univariate linear regression analyses), The Tromsø Study: *Fit Futures.* Statistically significant results at 10% level displayed in **bold.**

	Total hip				Femoral neck			
	Crude		Age adjusted		Crude		Age adjusted	
GIRLS n = 469	Beta	p-value	Beta	p-value	Beta	p-value	Beta	p-value
Age (years)	0.010	0.461			0.014	0.310		
Height (m)	0.002	**0.027**	0.002	**0.030**	0.003	**< 0.001**	0.003	**< 0.001**
Weight (kg)	0.004	**< 0.001**	0.004	**< 0.001**	0.004	**< 0.001**	0.004	**< 0.001**
BMI (kg/m^2)	0.010	**< 0.001**	0.010	**< 0.001**	0.009	**< 0.001**	0.009	**< 0.001**
Ethnicity (White/Other)	0.008	0.840	0.006	0.879	-0.007	0.857	-0.009	0.803
Sexual maturation	*Reference: Menarche age < 12.5 years*							
Intermediate	-0.027	**0.036**	-0.027	**0.038**	-0.027	**0.040**	-0.026	**0.042**
Late	-0.053	**0.001**	-0.054	**0.001**	-0.055	**< 0.001**	-0.055	**< 0.001**
Smoking (No/Yes)	-0.025	**0.077**	-0.026	**0.074**	-0.030	**0.033**	-0.031	**0.031**
Snuffing (No/Yes)	-0.003	0.821	-0.004	0.764	-0.003	0.786	-0.004	0.712
Alcohol use (No/Yes)	-0.013	0.320	-0.014	0.294	-0.012	0.369	-0.013	0.330
Physical Activity	*Reference: Sedentary*							
Moderate	0.012	0.473	0.013	0.461	0.016	0.356	0.016	0.340
Sports	0.031	**0.087**	0.031	**0.086**	0.037	**0.037**	0.037	**0.036**
Hard training	0.113	**< 0.001**	0.112	**< 0.001**	0.118	**< 0.001**	0.118	**< 0.001**
Comorbidity (No/Yes)	0.006	0.616	0.005	0.662	0.006	0.636	0.005	0.699
Diseases known to affect bone [a] (No/Yes)	-0.019	0.726	-0.019	0.733	-0.041	0.454	-0.041	0.461
Medication daily (No/Yes)	0.003	0.820	0.003	0.824	0.001	0.928	0.001	0.935
Medication known to affect bone [b] (No/Yes)	-0.041	0.232	-0.042	0.225	-0.036	0.292	-0.037	0.281
Hormonal contraceptives	*Reference: No use of hormonal contraceptives*							

Oestrogens & progestin	-0.001	0.959	-0.002	0.861	0.004	0.753	0.002	0.881
Progestin-only	0.009	0.759	0.006	0.824	0.016	0.570	0.013	0.353

	Total hip				Femoral neck			
	Crude		Age adjusted		Crude		Age adjusted	
BOYS n = 492	Beta	p-value	Beta	p-value	Beta	p-value	Beta	p-value
Age (years)	0.058	**<0.001**			0.072	**< 0.001**		
Height (m)	0.004	**< 0.001**	0.004	**< 0.001**	0.006	**< 0.001**	0.006	**< 0.001**
Weight (kg)	0.004	**<0.001**	0.004	**< 0.001**	0.004	**< 0.001**	0.004	**< 0.001**
BMI (kg/m^2)	0.011	**< 0.001**	0.011	**<0.001**	0.012	**< 0.001**	0.012	**< 0.001**
Ethnicity (White/Other)	0.072	0.122	0.058	0.217	0.076	0.115	0.057	0.227
Sexual maturation	*Reference: Completed (PDS 4)*							
Underway	-0.042	0.123	-0.036	0.189	-0.057	**0.036**	-0.050	**0.066**
Just started	-0.074	**0.020**	-0.066	**0.037**	-0.112	**< 0.001**	-0.103	**0.001**
Smoking (No/Yes)	-0.035	**0.027**	-0.039	**0.012**	-0.020	0.223	-0.025	0.116
Snuffing (No/Yes)	-0.001	0.964	-0.002	0.891	0.012	0.374	0.011	0.428
Alcohol use (No/Yes)	0.028	**0.050**	0.027	**0.057**	0.031	**0.032**	0.030	**0.037**
Physical Activity	*Reference: Sedentary*							
Moderate	0.012	0.453	0.011	0.492	0.025	0.143	0.023	0.168
Sports	0.092	**< 0.001**	0.089	**< 0.001**	0.108	**< 0.001**	0.104	**< 0.001**
Hard training	0.154	**< 0.001**	0.148	**< 0.001**	0.155	**< 0.001**	0.147	**< 0.001**
Comorbidity (No/Yes)	-0.017	0.262	-0.020	0.168	-0.008	0.585	-0.013	0.394
Diseases known to affect bone[a] (No/Yes)	-0.057	0.346	-0.060	0.318	-0.063	0.309	-0.066	0.276
Medication daily (No/Yes)	-0.007	0.637	-0.010	0.508	-0.006	0.702	-0.009	0.542
Medication known to affect bone[b] (No/Yes)	-0.044	0.377	0.050	0.309	-0.036	0.475	-0.044	0.381

[a] Diseases (ICD10) known to affect bone:
E03 Hypothyroidism, E10 Diabetes type I, F50.9 Eating disorders, K90.0 Celiac disease, M13 Arthritis
[b] Medication (ATC) known to affect bone:
D07A Plain corticosteroids, H03A Thyroid preparations, N03A Antiepileptics,
R01AD Corticosteroids, R03BA Glucocorticoids (inhalants), H02A Corticosteroid for systemic use

Table 4 Pooled parameter estimates after multiple imputations [a] in the multivariate linear regression models [b] with the highest adjusted R^2, explaining associations between basic characteristics, lifestyle variables and BMD (g/cm^2) at total hip and femoral neck in girls and boys <18 years, adjusted for possible confounders [c,d]. Statistically significant result (p<0.05) in **bold.** The Tromsø Study: *Fit Futures*

	FEMALES n = 469				MALES n = 492			
	Total hip		Femoral neck		Total hip		Femoral neck	
	Beta	p-value	Beta	p-value	Beta	p-value	Beta	p-value
Age (years)	0.017	0.197	0.017	0.177	0.030	**0.038**	0.035	**0.013**
BMI (kg/cm^2)	0.011	**<0.001**	0.002	0.533	0.012	**<0.001**	0.012	**< 0.001**
Height (cm)	0.002	**0.011**	0.003	**<0.001**	0.002	**0.014**	0.004	**< 0.001**
Sexual maturation:	*Reference: Menarche age < 12,5 years*				*Reference: Completed*			
Intermediate /Underway	-0.028	**0.019**	- 0.124	**0.002**	-0.020	0.388	-0.027	0.241
Late /Just started	-0.051	**<0.001**	- 0.236	**0.002**	-0.035	0.198	-0.054	**0.047**
Physical activity:	*Reference: Sedentary*							
Moderate	0.004	0.810	0.009	0.582	0.007	0.640	0.016	0.272
Sports	0.023	0.170	0.033	**0.047**	0.077	**< 0.001**	0.091	**< 0.001**
Hard training	0.117	**<0.001**	0.123	**<0.001**	0.144	**< 0.001**	0.141	**< 0.001**
Alcohol (No/Yes)					0.035	**0.006**	0.028	**0.021**
Smoke (No/Yes)					-0.029	**0.037**		
BMI*Sexual maturation			0.004	**0.017**				
Diseases known to affect bone[c] (No/Yes)	-0.013	0.793	-0.032	0.512	-0.023	0.648	-0.018	0.718
Medication known to affect bone[d] (No/Yes)	-0.036	0.245	-0.036	0.237	-0.074	0.078	-0.065	0.124
Hormonal contraceptives	*Reference: No hormonal contraceptives*							
Oestrogens & progestin	-0.006	0.563	0.000	0.987				
Progestin-only	-0.006	0.800	0.005	0.839				
	Adjusted R^2: 22.9%		Adjusted R^2: 24.3%		Adjusted R^2: 32%		Adjusted R^2: 34.7%	

[a] Variables included in the imputation procedure:
Sex, age, height, weight, BMI, alcohol, smoke, physical activity, menarche age, PDS-score, hormonal contraceptives, medication and diagnosis known to affect bone, BMD total hip and femoral neck

[b] Variables with p-values<0.1 in univariate analyses included

[c] Diseases (ICD10) known to affect bone:
E03 Hypothyroidism, E10 Diabetes type I, F50.9 Eating disorders, K90.0 Celiac disease, M13 Arthritis
[d] Medication (ATC) known to affect bone:
D07A Plain corticosteroids, H03A Thyroid preparations, N03A Antiepileptics, R01AD Corticosteroids, R03BA Glucocorticoids (inhalants), H02A Corticosteroid for systemic use